American Association of Neuropathologists

2026 102nd AANP Annual Meeting Abstract Book

Overview: Scientific Sessions
Friday, June 5, 2026 &
Saturday, June 6, 2026

All abstracts of the papers presented in this program are published in the June 2026 issue of the *Journal of Neuropathology and Experimental Neurology*.

FRIDAY PLATFORMS 1 & 2

Platform Session 1: ***Tumors: Glial 1*** **Moderators:** Matthew Wood, MD, PhD and Giselle Lopez, MD, PhD
Platform Session 2: ***Neurodegenerative: Alzheimer*** **Moderators:** Michael Miller, MD, PhD and Meaghan Morris, MD, PhD

	Platform Session 1: *Tumors: Glial 1* Chesapeake Ballroom	Platform Session 2: *Neurodegenerative: Alzheimer* Choptank Ballroom
8:00 am - 8:15 am	**1** **Utility of Repeat Molecular Testing in Recurrent Glioblastoma – A Time-Stratified Analysis of 175 Matched Primary/Recurrence Specimens** M Mejia Bautista, R Multz, M Sukhanova, B Nezami, A Krbanjevic, D Ducket, L Santana dos Santos, R Castellani, L Jennings, P Jamshidi, J Ahrendsen	**9** **Region-Specific Alterations in NPTX2 Expression in Alzheimer's Disease and Their Association with Dementia** K Chang, M Xiao, P Worley, D Nauen
8:15 am - 8:30 am	**2** **Whole-Brain Neuropathologic and Transcriptomic Profiling Reveals Tumor-Associated Oligodendrocyte Reactivity in Glioblastoma** J Peng, S Oliver, N Freeburg, G Gopikrishna, D Chafamo, N Tokcan, D O'Rourke, I Nasrallah, Z Binder, D Silverbush, M Nasrallah	**10** **Differential Labeling of Neurofibrillary Tangle Maturity by New Phospho-Tau Epitopes** E Connolly, L Wu, N Gilyazova, A Liu, B Xu, S Wang
8:30 am - 8:45 am	**3** **MGMT pyrosequencing results as a continuous variable in glioblastoma: Clinical and immunohistochemical correlates** M McCord, C Fadul, N McCann, H Ryu, D Schiff, B Purow, A Asthagiri, M Mut, M Lopes, V Smith	**11** **Characterizing the Role of TREM2 in Microglial Responses to Amyloid-β using Human Induced Pluripotent Stem Cells** C Cardona, R Patel, A Sproul, A Teich
8:45 am - 9:00 am	**4** **Detection of HSV latency-associated transcript expression in tumor-associated neurons after CAN-3110 therapy for glioblastoma** A Santos, E Shen, J Gantchev, H Kelley, C Linke, A Ling, G Ayoub, K Ligon, I Solomon, E Chiocca	**12** **Cognitively resilient individuals show enrichment for protective amyloid-responsive microglia (ARM)** A Nguyen, V Ramanan, E Hofrenning, S Przybelski, J Emery, R Petersen, D Knopman, C Jack, R Reichard, J Graff-Radford, P Vemuri
9:00 am - 9:15 am	**5** **Genomic Alterations and Emerging CNS tumor types: An Evidence-Based Consensus Review by Cancer Genomics Consortium (CGC) Taskforce** M Sukhanova, L Hu, P Jamshidi, L Santana Santos, C Ida, S Neill, M Abedalthagafi, L Satgunaseelan	**13** **The Temporal Pole in SuperAgers** A Zouridakis, A Frater, A Avgerinos, S Weintraub, M Mather, T Gefen, C Geula, M Mesulam, P Jamshidi, R Castellani
9:15 am - 9:30 am	**6** **FGFR3-mutant neuroepithelial neoplasms expand the spectrum of the FGFR3::TACC3-fused glioblastoma outlier methylation group** S Koga, A Desai, S Bagley, M Alonso-Basanta, Z Abdullaev, O Singh, H Chung, K Aldape, E Komlodi-Pasztor, M Li, I Nasrallah, N Shih, D O'Rourke, A Perry, M Quezado, E Sloan, M Nasrallah	**14** **Plasma p-tau217 robustly detects autopsy-confirmed ADNC up to 15 years before death and outperforms p-tau181, GFAP and NfL across co-pathologies** D Smirnov, X Zeng, M Farinas, M Nafash, A Bedison, C Matan, N Nadkarni, A Gogola, C Shaaban, H Aizenstein, D Tudorascu, T Pascoal, V Villemagne, S Berman, R Sweet, B Snitz, M Kamboh, A Cohen, J Kofler, M Ikonomovic, O Lopez, T Karikari
9:30 am - 9:45 am	**7** **Adult Diffuse Gliomas with FGFR3::TACC3 Fusion: Histopathology, Molecular Profile, and Clinical Outcomes** Y Zhu, N Karbhari, A Valerius, C Ida, C Zepeda-Mendoza, A Raghunathan, J Trejo-Lopez, A Nguyen, V Pazdernik, U Sener, C Giannini, R Vaubel	**15** **The relationship between central and peripheral microRNA Alzheimer's Disease biomarker candidates** N Umesh Ganesh, U Coskun, A Krunic, Y Lee, G Kureli, S Burkhardt, A Schütz, F Sananbenesi, D Krüger, T Pena-Centeno, Q Yang, A DeStefano, J Henderson, H Lin, K Nho, A Saykin, T Mellott, J Blusztajn, A Fischer, I Delalle
9:45 am - 10:00 am	**8** **Self-reactive, tumor-associated CD4+ T cells in mouse models of high-grade glioma** E Russler-Germain, A Zhu, L Jorrin, T Hu, P Hsieh, C Hsieh, M Chheda	**16** **Downstream Neuroprotection Associated with Amyloid Targeting Therapy** E Lee, C Brown, J Robinson, S Das, E McGrew, E Suh, V Van Deerlin, D Irwin, I Nasrallah, D Mechanic-Hamilton, P Yushkevich, D Wolk

FRIDAY PLATFORMS 3 & 4

Platform Session 3: ***Tumors: Glial 2*** **Moderators:** Veena Rajaram, MD, MBBS and Matthew McCord, MD
Platform Session 4: ***Neurodegenerative: Alzheimer, Other*** **Moderators:** Aivi Nguyen, MD and Andrew Teich, MD, PhD

	Platform Session 3: *Tumors: Glial 2* Chesapeake Ballroom	Platform Session 4: *Neurodegenerative: Alzheimer, other* Choptank Ballroom
2:00 pm - 2:15 pm	**17** **Molecular, histologic, and clinical characterization of 51 gliomas with an epigenetic classification of gliomatosis cerebri-like glioma** C Dampier, G Lopez, S Dahiya, M Haeri, J Wang, D Meredith, P Kobalka, S Ahmadian, J Fullmer, A Stemmer-Rachamimov, C Xing, R Green, J DeWitt, N Khanlou, M Pekmezci, P Samghabadi, C Lucas, J Helgager, K Donev, F Iwamoto, F Andreiuolo, L Szymanski, V Kaimaktchiev, P Sequeira, O Singh, H Chung, Z Abdullaev, M Quezado, K Aldape, M Nasrallah	**25** **Unusual Neuropathological Findings in Cases of Unexpected Death with No Apparent Anatomical Causes** J Houpt, Q Zhang, L Ang
2:15 pm - 2:30 pm	**18** **Correlation of ATRX Mutation Status, ATRX Immunohistochemical Staining Results, and ALT Status in IDH-Mutant Astrocytoma** O Kizilkaya, K Zhang, M Sherief, D Mukherjee, C Bettegowda, M Holdhoff, V Croog, S Sahebjam, D Kamson, J Torroella, K Shreck, C Eberhart, A Meeker, C Lucas	**26** **TDP-43 and tau-related neurite loss is associated with locus coeruleus dysconnectivity and beta-amyloid accumulation in Alzheimer's disease** G Uruk, R Gatto, N Hossain, K Josephs, R Reichard, J Whitwell, H Youssef
2:30 pm - 2:45 pm	**19** **Genomic profiling of genetically-defined oligodendroglioma harboring TP53 alterations** V Huynh, M Vizcaino, M Webb, V Pazdernik, K Fung, J Bouffard, T Kollmeyer, C Praska, D Zhang, M Gandham, S Dasari, C Marcou, S Smoley, M Isaacson, R Vaubel, A Nguyen, J Trejo-Lopez, K Aldape, C Giannini, A Raghunathan, S Kizilbash, H Al Kateb, R Jenkins, C Zepeda Mendoza, C Ida	**27** **Identification of molecular-neuropathological signatures of sporadic AD and FTLD** T Zhang, D Geschwind, S Magaki, H Vinters
2:45 pm - 3:00 pm	**20** **Alterations in Mismatch Repair Proteins Impact Survival in Oligodendroglioma** C Slocum, Y Nishikawa, C Maldonado-Diaz, S Hiya, M Christian Virata, J Samanamud, J Walker, M Umphlett, N Tsankova, T Richardson	**28** **Association of Cerebrovascular Pathologies with Hippocampal Sclerosis of Aging in Community-Dwelling Older Adults** K Kamboj, S Agrawal, A Kapasi, L Barnes, L Yu, D Bennett, J Schneider
3:00 pm - 3:15 pm	**21** **MicroRNA changes can drive senescence in pilocytic astrocytoma** C Eberhart, E Raabe, M Yuan	**29** **Quantifying Vascular Contributions to Dementia Across Large-Scale Neuropathology Cohorts** A Gonzalez, A Ghaseminejad-Bandpey, S Seshadri, S Etemadmoghadam, M Flanagan
3:15 pm - 3:30 pm	**22** **Beyond the Posterior Fossa: Clinical and Molecular Characterization of 33 Ependymomas with PFB Molecular Signatures** P Engin Zerk, Z Abdullaev, M Santi, A Viaene, S Alexandrescu, M Quezado	**30** **Regional Heterogeneity of Cerebral Amyloid Angiopathy Defined by Deep Learning–Based Vessel-Level Quantification** H Tahmasebidehkordi, A Bahramy, D Julian, K Neal, T Pearce, J Kofler
3:30 pm - 3:45 pm	**23** **Eighteen cases of glioneuronal tumor, kinase-fused, subtype A (GNT_KinF_A) from a multi-institutional study** E Pai, Z Abdullaev, S Ahmadia, K Aldape, M Blessing, J Byers, B Cole, E Conner, S Dahiya, K Dazelle, C Eberhart, I Fernandes, A Fonseca, M Frosch, C Giannini, N Lehman, G Lopez, A Perry, M Quezado, A Raghunathan, M Santi, A Toland, A Viaene, C Welsh	**31** **Quantitative Comparison of AT8 and pTau181 Immunohistochemical Staining in Human Brains with Alzheimer's Disease Neuropathologic Change** S Rashid, H Peng Cheng
3:45 pm - 4:00 pm	**24** **FGFR2-Fused Low-Grade Neuroepithelial Tumors: Clinicopathologic and Methylation Analysis of a National Cohort with WHO Implications** S Rajan, M Leung, K Schieffer, O Singh, K Qin, D Koboldt, A Shaikhouni, K Miller, J Leonard, J Pindrik, C Pierson, D Boué, E Mardis, C Cottrell, K Aldape, D Thomas	**32** **Data-Driven Thresholds for Standardized Classification of Severe Alzheimer's Disease Neuropathology Using Digital Neuropathology** R Shahidehpour, A Neltner, M Klusty, W Corbett, A Gonzalez, D Gutman, D Fardo, A Bachstetter, C Bumgardner, M Flanagan, P Nelson

FRIDAY POSTERS #1-#20

Friday June 5, 2026		
Time:	**Poster #:**	**Schooner A/B, Clipper A/B/C, Galleon A/B/C**
8:00 am - 5:00 pm	**1**	**Transcriptional Regulation by ZNF143 Promotes Metabolic Reprogramming in Aggressive Glioblastoma** N Won, R Yasrab
	2	**Tumor infiltrating clonal hematopoiesis (TICH) in IDH-wildtype glioblastomas** U Baguda, H Ghaseminezhad, S Hardy, K Galbraith
	3	**Quantitative Morphometric Features Distinguish SOX2+ Tumor Cells from Reactive Gliosis in Glioblastoma** B Chao, M Barrett, A Winiarz, B Nath, S Duenweg, A Lowman, E Cochran, J Jacobsohn, J Connelly, M Krucoff, R Desai, M Straza, S Bobholz, P LaViolette
	4	**The Spatial Distribution of SOX2 and Ki-67 Relative to Both Vasculature and Ablation Margin after LITT for Glioblastoma: A Post-Mortem Case** M Mirzaei, D Kim, A Winiarz, B Chao, M Barrett, S Duenweg, B Nath, H Reecher, A Lowman, S Bobholz, P LaViolette
	5	**Metastatic Glioblastoma with Unusually Long Survival** S Gadasalli, E Daoud, K Hatanpaa, J Raisanen
	6	**Case report of a glioblastoma with nonfunctional H3 K27R mutation** M Chung, C Lucas, M Morris, M Quezado, A Sandoval
	7	**Glioblastoma, IDH-wild type with an unusual EWSR-1:RAD51B gene fusion: A case report and review of the literature** S ALMUSTAFA, E Hattab, M Al-Kawaaz, K Aldape
	8	**Glioblastoma Survival with Partial Gains and Losses in Chromosomes 7 and 10 at Henry Ford Health** J Klonoski, X Fang, B Shaw, A Tabbarah, T Wen
	9	**Integrative 68Ga-FAPI PET/MRI and transcriptomic profiling for investigation of aggressiveness and stromal biology in diffuse gliomas** R Risgaard, A Ahmed, M Baskaya, M Dey, F Rashidi, P Clark, Z Morris, A McMillan, M Veronesi, A Pirasteh, A Bhatia
	10	**Radiation-induced glioma with extracranial metastases and liquid biopsy-detected KRAS mutation** S Beldick, S Brar, K Dagar, M MacDonald, M Shkrum, C Hawkins, L Ang
	11	**Histopathology-Based Recurrence Risk Stratification in Grade 2–3 Astrocytomas Using Structured Multi-Stream Deep Learning** J Cohen, T Pearce
	12	**Withdrawn**
	13	**Characterization of a High-Grade Glioma Associated with Cowden Syndrome: A Rare Event** R Alfattal, M Gubbiotti
	14	**SLC34A2::ROS1 Fusions Identified at Recurrence in IDH-Wildtype and IDH-Mutant Gliomas** R Alfattal, M Gubbiotti
	15	**ROS1 Fusion as a Rare but Important Molecular Marker in the Identification and Treatment of Low-grade Gliomas** A Lokken, C Xing ***(Note: Virtual Only)***
	16	**Molecular profile of granular cell astrocytoma predicts aggressive clinical behaviour independent of morphology** M Jensen, D Pranoy, O Wroe-Wright, O MacCormac, F Marchi, R Laxton, R Elmers, O Al-Salihi, N Agarwal, I Bodi, S Al-Sarraj, J Lavrador, Z Reisz
	17	**Glioneuronal tumours harboring NTRK gene rearrangements: Pathological diversity in three cases** M Jensen, R Laxton, K Eggleton, R Ellmers, C Limbachia, N Agarwal, C Chandler, D Wang, C Bleil, B Zebian, U Uparkar, S Dawson, F Carceller, S Al-Sarraj, I Bodi, Z Reisz
	18	**High-grade glioma with pleomorphic and pseudopapillary features (HPAP) presenting as a hemorrhagic lesion in a young adult: A case report** H Ellsworth, S Bihari, N Yoh, Z Zhu, A Haggiagi, B Gill, O Al Dalahmah
	19	**Diffuse midline glioma, H3K27-altered, showing loss of H3 K27M mutation upon progression, 5 years after initial diagnosis** P Nisarga, B Paneru, K Deeb, E Nduom, S Neill
	20	**Diffuse Midline Glioma with Concurrent MAPK Pathway Alterations: Report of Two Cases** M Regmi, G Kleinman, E Borys

Posters are not offered for CME credit

FRIDAY POSTERS #21-#40

Friday June 5, 2026		
Time:	**Poster #:**	**Schooner A/B, Clipper A/B/C, Galleon A/B/C**
8:00 am - 5:00 pm	**21**	**Spatial Transcriptomic Analysis of Pediatric High-Grade Gliomas** E Russler-Germain, J Liu, K Schwetye, P Cimino, S Dahiya
	22	**Frontal lobe pediatric high-grade glioma with eosinophilic granular bodies and PMS2 mutation** A Delaidelli, S Alexandrescu, A Church, A Santos
	23	**A case of molecularly defined oligodendroglioma with CDKN2A/B homozygous deletion** J Li, J LIu, S Schwartz ***(Note: Virtual Only)***
	24	**An IDH1-Mutant Infiltrative Glioma with MYBL1:MMP16 Fusion and Epigenetic Discordance** S Stone, R Landvater, J Kim
	25	**PI3K Pathway Mutations are Associated with Worse Overall Survival and Progression-Free Survival in Newly Diagnosed Grade 2 Oligodendroglioma** G Lopez, V Smith, K Stevenson, S Gergory, R McLendon, D Ashley, J Hernon, A Desjardins
	26	**Adult Brainstem Oligodendroglioma Presenting with Severe Motor and Bulbar Dysfunction: A Rare Case Report** J Ritzman, D Cook, C Xing
	27	**Metastatic anaplastic oligodendroglioma in the bone marrow: A Case Report** Z Piao, A Mhoyan, S Kirschbaum, R Green, P Kim, R Guzman, F Torres, P Gabikian, Y Kim, R Zuch, B Lowenthal, M Ghassemi ***(Note: Virtual Only)***
	28	**Evolution of the immune landscape in MAPK-driven glioma** J Phillips, A R.L., L Wang, S Bergland, H Piyadasa, J Ranek, V Voong, A Shai, B Oberlton, M Berger, R Prins, S Bendall, E Chang, M Angelo, S Hervey-Jumper, H Okada, A Diaz, E Hashemi
	29	**High-Grade Astrocytoma with Piloid Features in Neurofibromatosis Type 1 after a Diagnosis of Pilocytic Astrocytoma** C Kose, J Chen, D Sasaki-Adams, M Gokden
	30	**Pleomorphic Xanthoastrocytoma with MYC Alteration and Mesenchymal Transformation: A Case Report and Review of the Molecular Landscape** S nacer, W Bulkeley, N Tran, M Sepideh, Y Sun
	31	**Posterior Fossa Glioblastoma with REV3L-MET Fusion** C Slocum, D DeArruda Camara, C Maldonado-Diaz, S Hiya, J Walker, T Richardson, N Tsankova, M Umphlett
	32	**A Case of Metastatic Pleomorphic Xanthoastrocytoma** C Slocum, S Ghatan, A Nehlsen, M Rosenblum, J Walker, T Richardson, N Tsankova, M Umphlett
	33	**Diagnostic challenges and longitudinal genomic analysis of a highly recurrent MN1:BEND2 fusion tumor lacking classical astroblastoma features** T Klug, M Aboian, A Huttner
	34	**Unusual Suprasellar Lesion presenting with a Complex Clinical Picture: Chordoid Glioma of the Third Ventricle - Case Report** T Klug, A Huttner, Z Omay
	35	**Gliomas with FGFR3::TACC3 fusion: Case series and review** L Borecky, T Lee, A Achiriloaie, J Deisch, R Raghavan
	36	**Molecular characterization of glial/glioneuronal tumors with FGFR3::TACC3 fusion: A case series.** S López-Muñoz, C Rodríguez-Antolín, C Rodríguez-Jiménez, V Heredia-Soto, M Mendiola, I Esteban-Rodríguez
	37	**Clinicopathological and Molecular Characteristics of IDH-Wildtype Glioblastoma with FGFR3::TACC3 Fusion** M Ospina-Romero, A Aaroe, L Ballester, M Gubbiotti
	38	**Utility of FGFR3 Immunohistochemistry to determine FGFR alterations in adult diffuse gliomas** D Colunge, R Lopez Yazdani, T Tihan
	39	**ROS1-fused high grade gliomas in adults: Genetic, Epigenetic, Immunohistochemical, Radiologic features and clinicopathological correlation** L Coke, M Li, A Bigdeli, N Shih, S Priore, M Nasrallah
	40	**CNS tumor with EP300::BCOR Fusion: Diagnostic Challenges and Long-term Clinical Course** R Colbourn, E Ling-Lin Pai, A Banihashemi, Z Mourelatos, K Aldape, Z Abdullaev, M Quezado, H Chung, I Nasrallah, D O'Rourke, S Bagley, N Shih, M Nasrallah

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FRIDAY POSTERS #41-#60

Friday June 5, 2026		
Time:	**Poster #:**	**Schooner A/B, Clipper A/B/C, Galleon A/B/C**
8:00 am - 5:00 pm	**41**	**Multi-Omics Integration Identifies Cooperative Regulatory Networks in ZFTA–RELA Supratentorial Ependymoma** A Sharma, S Khan, M Sharma, J Singh, V Suri, S Vasant
	42	**A case of myxopapillary ependymoma arising in a patient with SMARCB1-related schwannomatosis** A Denney, N Willard, A Toland
	43	**68-year-old male with a spinal ependymoma, MYCN amplified with extra-CNS metastasis: A case report** S Hiya, C Maldonado-Diaz, C Slocum, M Umphlett, J Walker, J Crary, G Haines, N Tsankova, M Beasley, T Richardson; Mount Sinai Hospital
	44	**A UBN2::BRAF fused low grade glial/glioneuronal tumor with pseudopapillary structures: A Novel Fusion or Partner?** J Dailey, A Nobee, J Donahue, H Rosenberg, L Liu
	45	**Papillary Glioneuronal Tumor with SLC44A1::PRKCA Fusion and Patchy H3K27me3 Loss in a Pediatric Intraventricular Mass: A Diagnostic Challenge** H Kilic, O Elkadi
	46	**Low-grade glial/glioneuronal neoplasm with QKI::RAF1 fusion** V Rajaram, V Singh, D Rakheja
	47	**Dysembryoplastic Neuroepithelial Tumor with FGFR3::MYH7 Fusion: Expanding the Molecular Spectrum of Glioneuronal Tumors** J Kocemba, S Ali, C Takahashi, M Majeed, W Bell, H Harmsen
	48	**KRAS-Variant Pilocytic Astrocytomas: A Two-Case Series Expanding MAPK Alterations** J Kocemba, S Ali, C Takahashi, M Majeed, H Harmsen, K Newell, W Bell ***(Note: Virtual Only)***
	49	**Multifocal Diffuse Glioneuronal Tumor with Oligodendroglioma-Like Features and Nuclear Clusters (DGONC) in a Pediatric Patient: A Case Report** B Zeches, M Dewan, B Mobley, P Paueksakon
	50	**Unusual KIAA1549::BRAF fusion and a PIK3CA alteration in DNT** P Engin Zerk, L Ramachandran Nair
	51	**Melanotic Differentiation in Pilocytic Astrocytoma: Two case reports expanding the Histopathological Spectrum** A Dhall, S Purkait, M Shahin, K T, R Sahu
	52	**Sarcomatous transformation of an ALK fusion-positive infant-type hemispheric glioma following treatment with lorlatinib** C Takahashi, S Ali, J Kocemba, M Majeed, H Harmsen, W Bell
	53	**Neuroepithelial tumor with CHD7::PLAG1 fusion** R Cecchi, N Consavage
	54	**Collision Tumor in the Basal Ganglia: Germinoma Combined with Pediatric-type Diffuse Astrocytoma, MYBL1-altered — A Case Report** Y Xie, X Su, H Lin, C Ji, Q Lin, T Shangguan, J Wu, S Li, X Huang, X Wang
	55	**Diagnostic Yield of Molecular Testing in Pediatric Central Nervous System Tumors Using Whole-Exome Sequencing** S Besharati, S Hsiao, M Mansukhani
	56	**Liquid biopsy diagnostics: The NCI/Bethesda CNS classifier reliably classifies cell-free DNA methylation data from cerebrospinal fluid** O Singh, Z Abdullaev, K Aldape, H Chung, C Dampier, H Lalchungnunga, M Nasrallah, M Quezado, Z Wu
	57	**A Uniform MGMT Promoter Methylation Analysis of Over 15,000 Central and Peripheral Nervous System Tumors** T Do, Z Abdullaev, K Aldape, H Chung, C Ida, M Quezado, O Singh, Z Wu
	58	**Identification of KIAA1549::BRAF fusion using Next Generation Sequencing** A Sandoval, A Pallavajjala, T Hebert, M Lin, N Roberts, M Morris, C Lucas, J Eshleman
	59	**CDC25A expression is differentially modified by Hispanic ancestry in single and mixed etiology dementia** E Ochoa, A Hernandez, Z Torres, S Barannikov, M Keating, B Dugger, D Dickson, K Bieniek
	60	**Amyloid-associated neurites: A common co-pathology of diffuse plaques** R Castellani, A Frater, A Avgerinos, S Weintraub, M Mather, T Gefen, C Geula, M Mesulam, P Jamshidi, A Zouridakis

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FRIDAY POSTERS #61-#80

Friday June 5, 2026		
Time:	**Poster #:**	**Schooner A/B, Clipper A/B/C, Galleon A/B/C**
8:00 am - 5:00 pm	**61**	**Spatially resolved transcriptomic heterogeneity shapes tau selective vulnerability in human master circadian clock in Alzheimer's disease** G Son, F Pereira, M Mladinov, A Soloviev, Y Yang, S Li, W Lee, N Dove, C Suemoto, V Paes, R Leite, C Pasqualucci, W Jacob-Filho, S Spina, W Seeley, A Cuervo, T Neylan, L Grinberg
	62	**Association between hippocampal microglia and hippocampal neurodegeneration in community-dwelling older adults** S Agrawal, L Yu, A Kapasi, L Barnes, D Bennett, J Schneider
	63	**Topologic morphology descriptors reveal structural differences in microglia by sex and AD neuropathologic change.** H Haberecht, J Emery, J Graff-Radford, P Vemuri, A Nguyen
	64	**Variability of microglial activation in cerebral and cerebellar white matter in Alzheimer's disease** J Taheri Talesh, J Black, C Hernandez, M Seijo, R Chkheidze
	65	**Microglial activation in the Locus Coeruleus is not associated with early tau or amyloid pathology in middle age** R Rodriguez Reyes, M Tan, C Onyeje, H Guo, M Morris
	66	**Digital Mapping of Regional Tangle Distribution Defines Alzheimer's Disease Subtypes** S Bukhari, H Hulsey-Vincent, E Fridman, N Postupna, C Latimer
	67	**Automated Detection of Tauopathy Lesions in AT8 IHC-Stained Whole Slide Images of cerebral cortices of Alzheimer Disease cases With Deep Learning** D Gutman, C Candano, J Vizcarra, A Rosado, L Vasquez, L Sevilla, D Garcia, S Rai, A Teich, D Coughlin, C Chuah, B Dugger
	68	**A machine learning approach to assess vascular β-amyloid and perivascular plaques in cerebral amyloid angiopathy** P Mojdeganlou, J Poirier, L Barnes, D Bennett, S Leurgans, J Schneider, A Kapasi
	69	**Quantitative Digital Pathology for Neurodegenerative Disease: Tissue Segmentation and Cell Classification** F Pourakpour, A Frater, L Cooper, P Jamshidi, R Castellani
	70	**Multi-Class Detection and Quantification of Neuropathological Features using YOLOv11** S Kandoi, R Afzal, M Alosco, K Bieniek, G Campanella, J Cherry, J Crary, D Dickson, K Farrell, M Hefti, J Jegminat, M Karlovich, D Koenigsberg, K Laborc, A Mckee, J Mez, M Miller, S Nassar, T Richardson, K Saini, C Sanctis, T Stein, E Thorn, Y Tripodis, R Verma, J Walker
	71	**Applying NULISAseq for postmortem differentiation distinct clinicopathologic forms of Alzheimer's disease** S Dunlop, S Lincoln, Z Peng, N Graff-Radford, G Day, C Lachner, A Nguyen, R Reichard, R Petersen, B Boeve, J Graff-Radford, D Dickson, A Algeciras-Schimnich, M Murray
	72	**AD Molecular Abnormalities in White Matter Glia Detected in Unfractionated and O4-Selected Serum Exosomes Using a Liquid Biopsy Approach** S de la Monte, M Tong
	73	**Dysregulated Incretin Networks Linked to AD Progression and APOE-ε4 Dose Suggest Roles for GLP/GIP Therapeutics in Humans** S de la Monte, M Tong, J Robbins, O Busquets
	74	**An iPSC-Derived Neuronal Interactome: A Resource for Alzheimer's Disease Target and Interaction Discovery** X Liang, A Bartosch, H Buchanan, C Karan, M Kaufman, T Lam, H Li, A Ropri, A Sproul, A Teich, E Youth
	75	**Drug screening in hiPSC neurons: A bioinformatic approach to identify drug candidates for Alzheimer's disease.** H Buchanan, A Bartosch, C Karan, M Kaufman, T Lam, H Li, X Liang, A Ropri, A Sproul, A Teich, E Youth
	76	**Regional tau burden varies with LATE stage in severe Alzheimer's disease neuropathologic change** S Hemmati, J Ogg, S Bukhari, H Hulsey-Vincent, S Oyaizu, N Postupna, C Latimer
	77	**Postmortem Evaluation of Aging-Related Tau Astrogliopathy in Decedents with Alzheimer's Disease** D Garcia, L Vasquez, L Beckett, N Saito, A Kim, J Lou, L Sevilla, K O'Donnell, D Woodworth, D Coughlin, A Teich, B Dugger
	78	**Neuroinflammatory pathways across the life span of individuals with Down syndrome** H Ek Olofsson, M Marta Ariza, K Jones, B Sanford, M Raust, S Guzman, T Wisniewski, C Coughlan, X Yu, E Head, A Granholm-Bentley
	79	**Long-Duration Type 1 Diabetes Paradoxically Predicts Less Vascular and Neurodegenerative Neuropathology at Autopsy** D Smirnov, H Shaw, I Wu, J Gauthier, S Jangolla, D Bennett, G King, M Feany
	80	**Associations of hemoglobin A1c with cerebral amyloid angiopathy severity in older adults** R Mehta, A Capuano, R Biswas, L Barnes, D Bennett, Z Arvanitakis

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FRIDAY POSTERS #81-#100

Friday June 5, 2026		
Time:	**Poster #:**	**Schooner A/B, Clipper A/B/C, Galleon A/B/C**
8:00 am - 5:00 pm	**81**	**Regional Distribution and Clinicopathologic Correlates of Mineralized Blood Vessels in Autopsy-Confirmed Alzheimer Disease** D Garcia, N Saito, S Tamizharasu, L Sevilla, M Luu, L Beckett, L Honig, C DeCarli, D Coughlin, A Teich, D Mungas, L Jin, L Garcia, B Dugger
	82	**Gene Expression Signatures of Gliovascular Cells Associated with Microinfarct Burden** K Clarke, C Corbett, A Gonzalez, A Ghaseminejad-Bandpey, M Doppler, M Alhneif, J Parker-Garza, B Danner, S Babu, O Ogunbona, S Etemadmoghadam, H Zare, M Flanagan
	83	**Multi-Institutional Evaluation of a Machine Learning–Based Microinfarct and Microhemorrhage Screening Tool** B Dugger, L Cerny Oliveira, L Vasquez, C Keene, C Latimer, S Edminster, A Hiniker, C Chuah
	84	**Thermal imaging of white matter rarefaction and cerebrovascular lesions in postmortem brain tissue** K Bieniek, M Smith, M Keating, M Mojtabai, E Ochoa, M Habes
	85	**Novel Characterization of the Human Ischemic Penumbra: Multimodal Integrated Whole-Brain Magnetic Resonance Imaging (MRI) and Neurohistology** A Ayyappan, R Nanjappa, N Fathima, K Ram, J Jayakumar, R Rangaswami, M Sen, R Immanuel James, A Manesh, G Varghese, M Frosch, D Nauen, R Verma, R Folkerth, M Sivaprakasam
	86	**Post-Mortem Analysis of Coronary and Cerebral Atherosclerosis: Are They Related?** A Nobee, M Punsoni
	87	**Neurovascular calcification and brain injury in chronic kidney disease** D Lauzier, J Chang, M Fisher, H Vinters, S Magaki
	88	**Effect of Long-term Dialysis use for Chronic Kidney Disease on Cerebral Vascular Calcifications** D Lauzier, J Chang, A Hill, W Lau, H Vinters, M Fisher, S Magaki
	89	**Unusual perivascular iron deposition in a 46 year old with remote hemorrhagic frontal lobe vascular malformation** A Penev, R Yoda
	90	**Spongiform Leukoencephalopathy in an Autopsy Case of Catastrophic Antiphospholipid Syndrome-like Illness Following COVID-19 Vaccination** H Miyata, M Watanabe, A Urata, H Kubo, H Ohtsubo, A Shiraoka, K Okamoto, T Maeda, N Oka, H Suzuki
	91	**A Case of Transthyretin Leptomeningeal Amyloidosis** A Sajeendran, A Fornari-Caprara, C Curran, J Thon, R Brandstadter, T Siegel, C Mossop, D Bazer, L C Kenyon
	92	**Postmortem Diagnosis of Limbic Encephalitis Following Suicidal Acetaminophen Overdose** C Zahn, E Bracken, A Huang, M Thurber, J Persons, M Blessing
	93	**The Multifaceted ADEM: A Biopsy-Proven Case Series from Pediatric to Late-Onset Patients** M Yassa, K Jiang, O Elkadi
	94	**Adult-Onset Leukodystrophy with 5q23.2 Deletion. A Post-Mortem Case Report.** M Adekanla, A Rajiv, P Bizargity, M Hoque, M Movahed-Ezazi, A Baisre de Leon
	95	**Choroid Plexus Macrophage Heterogeneity and Non-Canonical Titin Expression at the Human Brain–CSF Interface** M DiStasio, S Bhatta, A Grzybowski, G Ortiz
	96	**Extracellular Vesicles from early-stage pancreatic cancer contributes to paraneoplastic neurological syndrome** K English, K McAndrews, A Haltom, J Kim, P Kelly, B Moreno Diaz, J Hensel, C Ruivo, J Lopez, K Horn, R Kalluri
	97	**Withdrawn**
	98	**The Blast Exposure Likelihood Level (BELL) Scale: Multi-Factorial and Retrospective Assessment of Military Blast Exposure for TBI Research** S Abdallah, P Smith, S Waddingham, D Perl, D Priemer
	99	**Blast Injury Results in Widescale Cell Damage in the Human Cerebellum** K Ferrari, D Priemer, M Chatterjee, D Perl, S Juliano
	100	**A Unique Case of Fatal Intracranial Penetrating Injury from a Toothbrush: No Cavity Left Unexplored** L Evans, R Mittenzwei, A Penev

Posters are not offered for CME credit

FRIDAY POSTERS #101-#105

Friday June 5, 2026		
Time:	**Poster #:**	**Schooner A/B, Clipper A/B/C, Galleon A/B/C**
8:00 am - 5:00 pm	**101**	**Accelerating Brain Transcriptomics with CARIBOU: Computational AI Research Interface for Bioinformatics, Omics, and Unifying Agents** N Shirooni, D Riffle, P Sureshkumar, V Vijay, M Rose
	102	**Microscopy of nanoplastics in human brains: Association with evidence of pathology** E Bearer
	103	**Amyloid Burden in Ligamentum Flavum Differs by Amyloid Subtype in Lumbar Spinal Stenosis** W Sarraj, J Patel, R Riesenburger, K Arkun
	104	**Withdrawn**
	105	**Factors Associated with Autopsy Performance at a Psychiatric Institution in the 1930s** M Silver, D Wheeler, H Ames

Posters are not offered for CME credit

SATURDAY PLATFORMS 5 & 6

Platform Session 5: ***Tumors: Nonglial*** **Moderators:** Maria Gubbiotti, MD, PhD and Jared Ahrendsen, MD, PhD
Platform Session 6: ***Neurodegenerative: FTLD/Lewy body/CTE*** **Moderators:** William Harrison, MD and Amber Nolan, MD, PhD

	Platform Session 5: *Tumors: Nonglial* Chesapeake Ballroom	Platform Session 6: *Neurodegenerative: FTLD/Lewy body/CTE* Choptank Ballroom
8:00 am - 8:15 am	**33** **Diagnostic Utility of c-MYC and SMARCA4 Immunohistochemistry in non-WNT/non-SHH Medulloblastomas** I Caliskan, K Mirchia, M Pekmezci, A Perry	**41** **Focal microglial reactivity in the hippocampal CA2 subfield specific to a-synuclein pathology is implicated in Lewy body disease progression** E Luna, K Cousins, S Emrani, W Trotman, S Xie, D Weintraub, A Chen-Plotkin, E Lee, D Irwin
8:15 am - 8:30 am	**34** **Proteomic analysis of medulloblastoma identifies MYC IHC as a powerful outcome predictor outperforming FISH and methylation-based subgrouping** A Delaidelli, F Burwag, S Ben Neriah, Y Suk, C Dunham, S Cheng, T Shyp, B Ellezam, C Hawkins, S Kosteniuk, K Okonechnikov, D Schrimpf, S Perreault, S Singh, M Kool, S Pfister, A Von Deimling, C Steidl, C Hughes, A Korshunov, P Sorensen	**42** **TDP-43 and tau seed amplification assays for LATE diagnosis and differentiation from AD** S Wang, M Gerasimenko, L Wu, E Connolly, Z Wang, B Xu
8:30 am - 8:45 am	**35** **Chromosomal Copy Number Profiling in Skull Base Chordoma: A Single-Institution Study of 20 Cases** Y Zhu, C Zepeda Mendoza, S Hong, C Carr, J Van Gompel, A Raghunathan	**43** **Opposing Nuclear Body Remodeling in TDP-43 Pathology: SUMO1-Associated Protection and PML-Linked Vulnerability** O Medina, A Nana-Li, W Seeley
8:45 am - 9:00 am	**36** **Optical Genome Mapping Identifies Structural Variants in Meningioma** G Mendonça Fernandes, N Sahajpal, M Abreu, M Franco, M Alves, M McDermott, J Otero	**44** **The GRN gene variant rs5848 linked to TDP-43 pathology: lessons from epidemiology** P Nelson, K Aung, X Wu, I Tsuchiya, S Karanth, E Abner, D Fardo, Y Katsumata
9:00 am - 9:15 am	**37** **Development of immunohistochemical surrogate stains for CNS WHO grade 2 copy number alterations in meningioma** J Coukos, J Phillips, A Perry, D Raleigh	**45** **ANXA11 as additional pathological marker for TDP-43 classification in rare genetic, mixed and unclassifiable TDP-43 pathology** A Rajicic, T Swartenbroekx, J van Swieten, M Drost, L Donker-Kaat, J van Rooij, R van Buuren, H Seelaar
9:15 am - 9:30 am	**38** **Primary Central Nervous System GLI1-Altered Mesenchymal Tumors: Clinicopathological and Molecular Analysis of 8 Cases** X Su, X Wang	**46** **Divergent Endolysosomal Autophagy Signatures in Chronic Traumatic Encephalopathy and Alzheimer Disease** M Geraghty, M Geraghty, N Aytan, R Nicks, G Meng, S Hawkins, W Xia, J Cherry, V Alvarez, A McKee, T Stein
9:30 am - 9:45 am	**39** **The Cost of Pituitary Neuroendocrine Tumor Classification: Experience with the Minneapolis Algorithm** W McDonald	**47** **Diagnosis of Low Stage Chronic Traumatic Encephalopathy is Missed without Extensive Sampling in Half of Community Cases** L Chung, J Opara, K Scherpelz, C Keene, A Nolan
9:45 am - 10:00 am	**40** **Age-associated clinicogenomic characteristics and therapeutic determinants in melanoma brain metastasis** K Zhang, M Yang, C Palm, S Chipman, C Eberhart, C Lucas, A Weeraratna	**48** **Neuropathological Characterization of Brain Donors with a History of Military Parachute Service** I Lagerstrom, D Perl, D Priemer

SATURDAY PLATFORMS 7 & 8

Platform Session 7: ***Epilepsy, Methodologies*** **Moderators:** Michael Punsoni, MD and Melissa Blessing, DO
Platform Session 8: ***Demyelinating and Inflammatory, Developmental/Pediatric, Peripheral Nerve/Muscle, Ophthalmic Pathology***
Moderators: David Nauen, MD, PhD and Osorio Lopes Abath Neto, MD, PhD

	Platform Session 7: *Epilepsy, Methodologies* Chesapeake Ballroom	Platform Session 8: *Demyelinating and Inflammatory, Developmental/Pediatric, Peripheral Nerve/Muscle, Ophthalmic Pathology* Choptank Ballroom
2:00 pm - 2:15 pm	**49** **Beyond Neuronal Loss: Spatial Transcriptomics Defines Molecularly Distinct Forms of Hippocampal Sclerosis in Mesial Temporal Lobe Epilepsy** S Guzman, S Hazany	**57** **Microglial-amplified inhibitory synaptopathy: Convergent neuropathology in GlyR, GAD65, Kelch11, and NIF autoimmunity** A Denney, A Wohlfert, A Granholm-Bentley, A Piquet, K Jones, A Toland, S Guzman
2:15 pm - 2:30 pm	**50** **Automated Identification of Tissue Compartments in IHC-Stained Whole Slide Images for Neuropathologic Studies** J Vizcarra, A Rosado, C Candano, C Bumgardner, P Nelson, T Pearce, B Dugger, D Gutman	**58** **Microglia and G-protein coupled receptors in the pathogenesis of periventricular heterotopia** J Golden, G Cho, M Del Bigio, Y Lim
2:30 pm - 2:45 pm	**51** **AI-Driven Spatial Characterization of Neuropathology in Whole Human Brain: Addressing the Scalability Gap** R Nanjappa, S V, R Kumar, P V S, A Ayyappan, N Fathima Majid, C Sam, S Mulay, K Ram, S Neelakantan, J Jayakumar, R Rangaswami, M Sen, R James, A Manesh, G Varghese, D Nauen, R Verma, R Folkerth, M Sivaprakasam	**59** **High-Fat Diet Ameliorates Myonuclear Transcriptional Changes in R155H/R155H VCP Disease** N Shirooni, M Wu, L Weiss, N Robson, A Shmara, V Kimonis, M Rose
2:45 pm - 3:00 pm	**52** **State-Level Legal and Infrastructure Barriers to Research Brain Donation in the United States** E Selmanovic, R Folkerth, J Appel, P Hof, K Dams-O'Connor	**60** **A Phosphorylation-Independent TDP-43 Antibody Reveals Increased and Earlier Cytoplasmic Pathology in Inclusion Body Myositis** D Cook, A Lokken, J Ritzman, M Basuino, C Xing, M Kohler-Skinner, Q Mao
3:00 pm - 3:15 pm	**53** **Promise of Artificial Intelligence in Interpreting Neuropathology NGS Data: Addressing HIPAA and Institutional Compliance Constraints** M Elnagdy, J Weon, J Raisanen, M Thakkallapally, H Han, K Hatanpaa	**61** **Early myopathology of Pompe disease in patients detected by newborn screening** K Bhatti, S Jung, P Kishnani, K Jones
3:15 pm - 3:30 pm	**54** **From Feasibility to Validation: Implementing 2021 WHO CNS Tumor Whole Genome Methylation Profiling in a Resource-Limited Academic Center** M Haeri, R Ayoub, S Golem, N Lakis, A Marr	**62** **Decoding Cancer-Associated Pain: A Single-Cell Proteomic Analysis of Human Dorsal Root Ganglia Neurons** C Hayes, Y Jiao, A Barry, T Price, J Peng, C Ho
3:30 pm - 3:45 pm	**55** **Improved Performance of Clinically Applicable Deep Learning for CNS Tumor Classification from Histopathology** L Lalchungnunga, Z Abdullaev, K Aldape, S Brandner, H Chung, P Cimino, C Dampier, D Hoang, C Horbinski, B Li, C Lucas, Z Luo, D Marker, K McCortney, M Nasrallah, T Pearce, M Quezado, E Ruppin, E Shulman, O Singh, Z Wu, L Yefet, G Zadeh	**63** **Border-associated macrophages mediate immune responses in the unmyelinated optic nerve head in mice and humans** Z Yin, J Harris, P Cullen, Y Xue, A Mukwaya, N Lopez, A Bosco, J Schwakopf, R Yarcusko, D Tommasini, G Baldwin, S Da Mesquita, M Vetter, K Shekhar, M Margeta
3:45 pm - 4:00 pm	**56** **Fresh Ex Vivo Brain Tumor Imaging Using Two-Photon Microscopy** L Hu, R Li, S Wang, X Wang	**64** **Targeting IGF1R signaling in BCOR mutant retinoblastoma** C Eberhart, S Lee

SATURDAY POSTERS #106-#125

Saturday June 6, 2026		
Time:	**Poster #:**	**Schooner A/B, Clipper A/B/C, Galleon A/B/C**
8:00 am - 5:00 pm	**106**	**Hsd17b7 is Required for Proper Mammalian CNS Development** B Bessemer, R Stottmann
	107	**An optimized transcriptomic toolbox for rare cell types in neurodevelopment** N Shirooni, A Barroga, L Chi, K Kermani, J Li, F Munawar, V Padisetti, J Ramos, A Shenoy, P Sureshkumar, S Thayer, V Vijay, L Wang, I Whedon, X Yang, C Zabrocki, U Raychaudhuri, M Rose
	108	**A Distinct Population of Neural Progenitor Cells in the Developing Hippocampus** A Sandoval, T Mastalski, D Nauen
	109	**Gennari's Line: Developmental Implications for Visual Cortex Organization and Function** B Akomeah
	110	**Unusual cerebellar architecture in a 22 month old with Menkes disease** A Penev, R Mittenzwei
	111	**A Bloody Diagnostic Trap: Intracranial Extramedullary Hematopoiesis Mimicking a Brain Mass in a Neonate** L Evans, J Cotter
	112	**Hemizygous ALG13 Variant in a Term Neonate with Grade IV Germinal Matrix Hemorrhage** D Jackson, L Sung, V Mahalingam
	113	**Long-Term Survival in Holoprosencephaly: A Rare Case Extending Into Late Adolescence** Y Zhang, D Sudarshan, O Ogunbona
	114	**Dystrophic calcification and cortical disorganization in epilepsy resections for Sturge-Weber syndrome: Genetic correlation** D Lauzier, J Chang, A Martinez, V Avila, T Chang, B Fogel, H Vinters, S Magaki
	115	**Epilepsy-Associated Leptomeningeal Vascular Malformation Harboring an FGFR1 Mutation: A Case Report** S Han, M Clay, A Toland
	116	**Classifying Previously Uncategorized Tumors: Cases of MVNT and PLNTY diagnosed using the current WHO CNS tumor classification** A Reyes, H Varma
	117	**A single center retrospective on the molecular stratification of meningiomas** C Takahashi, S Ali, J Kocemba, M Majeed, C Hao, H Harmsen, W Bell
	118	**Integrated meningioma classification outperforms clinical prognostication but loses power with time and is affected by Simpson grade.** C Turner, J Correia, M Dragunow, J McLay, A Marubayashi, H Law
	119	**Cellular crosstalk in the brain microenvironment promotes melanoma invasion and growth** K Zhang, M Yang, M Yuan, C Palm, C Lucas, C Eberhart, A Weeraratna
	120	**Stratification by Ki-67 labeling index increases specificity of p16 expression as a surrogate marker for CDKN2A inactivation in meningioma** K Zhang, K Mirchia, A Perry, C Eberhart, D Raleigh, D Solomon, C Lucas, V Tang
	121	**Concurrent somatic NF2 and SMARCB1 biallelic alterations in a series of higher grade meningiomas** P Nisarga, L Patel, B Paneru, K Deeb, S Neill; Emory University Hospital
	122	**Multifocal Intraosseous Meningiomas with Sacrospinal and Calvarial Involvement. Case Report and Literature Review** C Fuller, A May, T Damron, W Lavelle, R Seth, T Lindquist, D Zaccarini
	123	**Atypical Meningioma with Granular Cell Features: A Case Report and Review of the Literature** A Nobee, M Punsoni
	124	**Meningioma of the Jugular Foramen Presenting as a Neck Mass** K Marconi, J Nix, E Kumar, M Gokden
	125	**B-lymphocytes Colliding with Meningiomas** S Moore, R Ambinder, J Weingart, C Lucas, C Gocke

Posters are not offered for CME credit

SATURDAY POSTERS #126-#145

Saturday June 6, 2026		
Time:	**Poster #:**	**Schooner A/B, Clipper A/B/C, Galleon A/B/C**
8:00 am - 5:00 pm	**126**	**Metastatic Meningiomas** R Cecchi, G Yeaney; Cleveland Clinic
	127	**Malignant Meningioma of Paraspinal Soft Tissue** C Zahn, J Persons, K Eschbacher, O Lopes Abath Neto
	128	**Dural-based myxoid neoplasm harboring a rare SMARCA2::CREM Fusion** T O'Brien, K Eschbacher
	129	**A neuroepithelial tumor with PATZ:MN1 fusion a new tumor type and literature review** C BENALLAL, C BOUVIER, R APPAY, A Tauziède Espariat, B DADONE-MONTAUDIÉ, H Rais
	130	**GLI1 Fusion–Positive Mesenchymal Tumor Presenting as a Spinal Canal Mass: An Unusual CNS Manifestation of an Emerging Entity** M Elnagdy, C Cai, E Daoud, J Raisanen, K Hatanpaa
	131	**A unique type of intracranial dural-based sarcomatoid neoplasm affecting pediatric patients harbors a distinct DNA methylation profile** C Dampier, Z Abdullaev, K Aldape, R Cirt, A Davis, S Frank, K Fung, J Hench, S Khan, M Kranendonk, A Perry, M Quezado, K Schieffer, O Singh, I Stasevich, D Thomas, P Varlet, Y Wilson, P Zerk, C Febres-Aldana
	132	**Calcified Chondroid Mesenchymal Neoplasm/Chondroid Synoviocytic Neoplasm: An Emerging Entity With Predilection for Skull Base Involvement** H La, T Pearce
	133	**HOXD12 activation status across brain tumor types reflects developmental origins** P Cimino, V Ha, A Shelbourn, S Arora, E Holland, D Pratt, M Quezado
	134	**Methylation profiling of biphasic classic medulloblastomas shows distribution across G3/G4 subgroups with enrichment for subgroups 1, 7, and 8** J Lee, Q Tran, A Breuer, M Lear, L Furtado, D Ellison, B Orr
	135	**CNS Embryonal Tumor Harboring a ZNF532::NUTM1 Fusion in the Cerebellum of a Young Adult** D Cook, E Goold, Q Mao, S Menacho, J Mendez, M Kohler-Skinner
	136	**Diagnostic Challenges in a Congenital Ependymoma with a Medulloblastoma Methylation Profile** M Alturkustani ***(Note: Virtual Only)***
	137	**YAP1::MAML2 Fused ETMR of the Posterior Fossa in Early Infancy: Case Report** S Ali, C Takahashi, J Kocemba, M Majeed, H Harmsen, W Bell
	138	**Malignant Transformation of Recurrent Adamantinomatous Craniopharyngioma to Squamous Cell Carcinoma: Case Report** S Ali, H Hyaduck, C Takahashi, J Kocemba, H Harmsen, W Bell, D Leino, M Majeed
	139	**55-year-old female with a Multinodular and Vacuolating Neuronal Tumor (MVNT): A case report** J Samanamud, C Slocum, M Virata, C Maldonado Diaz, S Hiya, M Umphlett, J Walker, N Tsankova, T Richardson
	140	**Plot Twist: Intraparenchymal Schwannoma with CHD7::VGLL3 Fusion: A Case Report** R Gaglia, J Donahue; Brown University Health
	141	**Evaluating MTAP as a Surrogate Marker for CDKN2A/B Deletion in MPNST and ANNUBP** O Kizilkaya, C Sucubulak, E Karaca, H Tomac, N Karaman, C Saricoban, M Onenerk, A Demiroz, N Comunoglu
	142	**Disseminated and Metastatic Choroid Plexus Tumors: Patterns, Treatment, and Outcomes** P Nguyen, M Umphlett, J Walker, N Tsankova, T Richardson
	143	**Delineating the Expression of Lung Adenocarcinoma Markers in Choroid Plexus Tumors** K Wysong, Y Wang, M Snuderl, J DeWitt
	144	**The rarest of the rare: Histologic and molecular features of pituicytomas with ependymal features** S Perez, C Cogbill, A Perry
	145	**Germinoma Arising in Association With an Intracranial Epidermoid Cyst** W Sun, J Mandell, V Smith

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SATURDAY POSTERS #146-#165

Saturday June 6, 2026		
Time:	**Poster #:**	**Schooner A/B, Clipper A/B/C, Galleon A/B/C**
8:00 am - 5:00 pm	**146**	**Dural-Based Histiocytic Neoplasm with KRAS p.A146P Mutation: A Diagnostic Challenge** K Champelli, M Shahin, A Dogan, J Starkey, S Puri, W Xie, J Wiszniewska, M Wood
	147	**Erdheim-Chester Disease Presenting as a Chronic Subdural Hematoma** J Houpt, Y Li, C Howlett, W Yaghmoor, J Megyesi, L Ang
	148	**Central Nervous System Histiocytosis: Diagnostic Challenges and the Importance of Accurate Classification** L Godsey, R Rozo Cifuentes, A Lee, N Wick, C Appin, A Ravindran, R Chkheidze
	149	**Leptomeningeal Histiocytic Sarcoma: A case report** V Rajaram, V Singh, D Rakheja
	150	**Primary Diffuse Meningeal Melanomatosis with CNS Invasion and NRAS Q61L Mutation** G Caballero, N Castrejón, F Aya, S Alós, T Topczewski, M Squarcia, A Arance, I Aldecoa
	151	**Circumscribed Meningeal Melanocytic Neoplasms, WHO CNS5 Classification and Outcomes** Y Zhu, N Karbhari, T Kaufmann, A Raghunathan, C Ida, C Zepeda Mendoza, I Dryden, S Salamat, C Atherton, S Jenkins, S Markovic, U Sener, C Giannini
	152	**A Rare Case of Malignant Metastatic Melanoma to the Central Nervous System with an IDH1 (R132C) mutation** J Dailey, A Nobee
	153	**A Classic Neural Crest Lineage Diagnostic Trap: Cellular Schwannoma Versus Metastatic Spindle Cell Melanoma: A Case Report** A Cuevas Ocampo ***(Note: Virtual Only)***
	154	**Corticotroph (Basophil) invasion of the posterior pituitary in the setting of pituitary adenoma (PitNET): A diagnostic pitfall** M Virata, C Slocum, C Maldonado-Diaz, S Hiya, J Samanamud, J Walker, N Tsankova, M Umphlett, T Richardson
	155	**Skull base metastasis of adenocarcinoma arising from mediastinal teratoma with malignant transformation: A case report** M Virata, M Beasley, C Maldonado-Diaz, T Richardson, C Slocum, N Tsankova, M Umphlett, J Walker
	156	**Brain Metastases from Renal Cell Carcinoma: A Histologic and Molecular Analysis with Correlation to the Primary Tumor** T Gruchala, M Mejia-Bautista, R Castellani, J Ahrendsen, L Santana-Santos, D Duckett, L Jennings, M Sukhanova, B Nezami, P Jamshidi
	157	**Chemoimmunotherapy-Associated Lineage Plasticity in NSCLC with RB1/TP53 Co-Alteration and Cerebellar Metastasis** T Banu, E Borys, G Kleinman
	158	**Post-treatment Differentiation in Metastatic Sinonasal Small Cell Neuroendocrine Carcinoma with Central Nervous System (CNS) Involvement** B Petrykowski, C Welsh, T Baker
	159	**HER2-Positive Male Breast Cancer with Brain Metastasis: A Case Report** F Alkhutani, A Alkhotani ***(Note: Virtual Only)***
	160	**Primary spinal cord adrenal cortical adenoma: A Case Report** A Kenyon, Z Abdullaev, P Engin Zerk, M Quezado, K Ligon
	161	**A Rare First Act: Brain Metastasis as Initial Clinical Presentation of Alveolar Soft-Part Sarcoma** P Loreto-Palacio, M Cohen, M Couce
	162	**Sebaceous Carcinoma of the Eyelid Associated with Lynch Syndrome: Morphologic Features and Importance of MMR Testing** P Loreto-Palacio, M Cohen, M Couce, J Sigel
	163	**Xanthogranulomatous Epithelial Tumor (XGET) of the Orbit in a 4-Year-Old Boy** W Sun, J Fanburg-Smith, J Gigantelli, E Burton, W Sanfelippo, H Spader, C Druzgal, M McCord
	164	**Unexpected choroidal melanoma in evisceration for blind painful eye** G Yeaney, A Singh
	165	**Actinomyces-Associated Necrotizing Bilateral Corneal Infections in an Immunocompromised Patient** B Zeches, U Tran, P Paueksakon

Posters are not offered for CME credit

SATURDAY POSTERS #166-#185

Saturday June 6, 2026		
Time:	**Poster #:**	**Schooner A/B, Clipper A/B/C, Galleon A/B/C**
8:00 am - 5:00 pm	**166**	**Withdrawn**
	167	**Molecular Detection of Naganishia globosa in a Destructive Pediatric Sellar Lesion: True Infection or Diagnostic Artifact?** R Joshi, B Lee, E Mohr, K Han
	168	**A Rare and Deadly Pathogen: Diagnostic Challenge of Cladophialophora bantiana Brain Infection in an Immunocompetent Patient** A Pallante, S Mandavilli, P Patel
	169	**Multiple Disseminated Opportunistic Infections as Initial HIV/AIDS Presentation in an Ecuadorian Immigrant** T Klug, U Alappan, J Magid-Bernstein, J Vinetz, A Huttner
	170	**A Tale of Two Lesions: Synchronous Schwannoma and Glioneuronal Hamartoma** P Loreto-Palacio, M Cohen, M Couce ***(Note: Virtual Only)***
	171	**Severe Rhabdomyolysis in Inflammatory Myopathy with Abundant Macrophages** P Loreto-Palacio, M Cohen, M Couce
	172	**Diagnostic Yield of Significant Neuromuscular Pathologies in Hospital Autopsy** R Hennis, A Rayo, M Chambers
	173	**Genetic and Pathologic Diagnosis of Fatal Duchenne Muscular Dystrophy-Associated Dilated Cardiomyopathy at Autopsy** W Hermann, S Irwin, E Duval, J Suddock
	174	**HIV-associated inflammatory myopathy with vacuolar change and diffuse nemaline pathology: a case report** M Sant, R Bell, O Lopes Abath Neto
	175	**Gemcitabine-related acute ischemic myonecrosis: A case report** M Sant, R Bell, O Lopes Abath Neto
	176	**Diagnostic yield of electron microscopy in muscle biopsies evaluated for mitochondrial pathology** E Fossee, S Moore, O Lopes Abath Neto
	177	**MT-TK m.8363G>A Pathogenic Variant in an Adolescent Male Patient with Myoclonus Epilepsy with Ragged Red Fibers (MERRF)** J Persons, S Moore
	178	**MHC-II staining pattern in two cases of immune checkpoint inhibitor-associated myositis with classic histopathological findings.** S Perez, I Caliskan, D Richman, G Xiong, M Margeta
	179	**Muscle neurogenic features in idiopathic inflammatory myopathies** G Grafham, H Faris, C Lam, A Murphy, M Tarnopolsky, J Lu
	180	**Neuromuscular Choristoma: A Rare Finding** E Conner, M Martinez-Lage
	181	**Targeting IL-17-Mediated Immune–Neural Crosstalk in Skin to Alleviate Diabetic Neuropathy** C Ho, M Adenegan, Y Wang, Y Zhang, A Barry, P Niehaus, A Dasgupta, A Suresh, K Chang, M Wu, B Pan, N Archer, Q Zheng, T Price
	182	**Granular structures in the human olfactory nerve across ages** M Lu, M DiStasio, C Baldoni, H Sanchez, C Liu, A Huttner
	183	**Neuropathology in a Diverse Cohort of Oldest Old: 2026 update on the LifeAfter90 Study** B Dugger, V Patel, L Jin, C DeCarli, A Posis, P Gilsanz, D Mungas, C Kawas, M Corrada, R Whitmer
	184	**Differential Neuropathology in Female versus Male Brain Donors with Chronic Traumatic Brain Injury** L Chung, E Selmanovic, R Folkerth, K Dams-O'Connor, K Scherpelz, C Latimer, D Child, C Keene, A Nolan
	185	**Rodeo as a Risk Factor for Chronic Traumatic Encephalopathy, Alzheimer's Disease and Other Related Dementias.** E Ruff, M Keating, K Bieniek

Posters are not offered for CME credit

SATURDAY POSTERS #186-#203

Saturday June 6, 2026		
Time:	**Poster #:**	**Schooner A/B, Clipper A/B/C, Galleon A/B/C**
8:00 am - 5:00 pm	**186**	**CTE-Type Tau Filaments in LATE-NC** E Pinarbasi, J Rana, M Fernandez, V Navratna, K Conway, A Lieberman, P Nelson, S Barmada, S Mosalaganti
	187	**Lesion-Associated Phosphorylated Tau in Traumatic Brain Injury and Repetitive Head Impacts: A Community Autopsy Cohort Study** E Selmanovic, A Hicks, M Del Bigio, P Hof, R Folkerth, K Dams-O'Connor
	188	**Bridging Central and Peripheral Pathology in Lewy Body Disease: Toward Personalized Biomarkers** B Wilson, T Dawson, V Dawson, A Rosenberg
	189	**Multisite Study: Predicting Lewy Body Disease Using Skin Biopsy alpha-Synuclein Seed Amplification Assays** T Beach, C Janarthanam, C Orru, A Hughson, D Holzinger, K Taylor, A Kanthasamy, B Caughey, R Heym, C Adler, H Shill, S Mehta, E Driver-Dunckley, A Ho, P Choudhury, D Shprecher, C Belden, A Atri, N Zhang, K Chen, G Serrano
	190	**Biofluid and Clinicopathological Correlates for NSD ISS Validation in Lewy Body Disorders: Results from the AZSAND** I Lorenzini, C Adler, N Zhang, H Shill, S Mehta, E Driver-Dunckley, C Belden, A Atri, P Choudhury, N Ashton, T Beach, G Serrano
	191	**Microglial Morphology and Reactivity Across the Intersection of Lewy Body Dementia and Chronic Traumatic Encephalopathy** J Emery, E Constantopoulos, J Graft-Radford, R Petersen, B Boeve, R Reichard, A Nguyen
	192	**Microglial sensome landscapes across tauopathies** T Adetona, M Keating, M Flanagan, T Richardson, J Walker, T Stein, A McKee, S Seshadri, D Dickson, K Bieniek
	193	**Clinicopathological correlates of FTLD-TDP type E: a deep phenotyping on an autopsy confirmed case** I Aldecoa, S Rubio-Guerra, J García-Castro, J Selma-González, I Sala, B Sánchez-Saudinós, N Zhu, I Barroeta, R Rojas-Garcia, J Turon-Sans, Á Carbayo, V Camacho, D Almenta, J Pérez-Blanco, S Bernal, L González-Quereda, G Caballero, L Molina-Porcel, O Dols-Icardo, D Alcolea, J Fortea, A Lleó, I Illán-Gala
	194	**Novel GRN Mutation in a case of FTLD-TDP: TDP-43 Mass Spectrometry and GRN mRNA Analyses** L Cracco, H Garringer, E Doud, R Rademakers, A Longo, M Jacobsen, E Buratti, B Ghetti, K Newell
	195	**Perry Syndrome in the Mayo Clinic Florida Brain Bank: A novel TDP-43 Proteinopathy** D Dickson, Y Tsuboi, W Lin, T Mishima, Z Wszolek
	196	**Facial-onset sensory and motor neuronopathy (FOSMN): a case report with postmortem findings including TDP-43 pathology** C Higham-Kessler, S Moore, A Swenson
	197	**Locus coeruleus TDP-43 pathology in a community-based cohort: clinical and pathological correlates** A Neltner, R Shahidehpour, M Hall, X Ning, H Kang, S Anderson, G Jicha, T Lee, S Fister, E Abner, D Fardo, P Nelson
	198	**Analysis of tauopathies in the NACC Data Set indicates relatively high frequency of incident PSP pathology in older persons** B Cushing, R Shahidehpour, G Jicha, E Abner, M Breig, K Teodorescu, E Coskun, J Neltner, G Kovacs, P Nelson
	199	**Patterns and prevalence of TDP-43 Copathology in Progressive Supranuclear Palsy Reveal Regional Vulnerability and Pathologic Heterogeneity** J Opara, M Bogdani, D Child, A Nolan, C Latimer
	200	**A Quantitative Pilot Study: Traumatic Brain Injury as a modifier of Tau Burden in Confirmed cases of Progressive Supranuclear Palsy** J Opara, A Kirkland, D Hunt, C MacDonald, A Nolan
	201	**Distinct Regional Tau Distribution of Pallidopontonigral Degeneration Compared to Progressive Supranuclear Palsy** H Sekiya, D Dickson, Z Wszolek
	202	**Profiling of tau neuropathology in human 4R-tauopathies: progressive supranuclear palsy (PSP) and corticobasal degeneration (CBD)** C Maldonado-Diaz, S Koutarapu, V Flores-Almazan, E Daoud, C White, T Richardson, T Orr, J Walker, M Orr
	203	**Neuronal cell cycle re-entry in a multiple system atrophy seeding mouse model** M Ospina-Romero, A Mukherjee, C Barria, C Gherardelli, S Sepulveda, F Wang, C Soto

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SATURDAY POSTERS #204-#210

Saturday June 6, 2026		
Time:	**Poster #:**	**Schooner A/B, Clipper A/B/C, Galleon A/B/C**
8:00 am - 5:00 pm	**204**	**Clinical and neuropathologic phenotypes associated to a novel GRN mutation** K Newell, H Garringer, B Ghetti, N Maynard, M Jacobsen, E Buratti, R Rademakers, F Unverzagt, D Clark, L Cracco
	205	**Neuropathologic Review of Spinocerebellar Ataxia Type 7 in a Five Generation Family** M Basuino, C Winterton, R Goodrich, S Vegunta, M Liu, Q Mao, D Wynn, K Nelson, L Ord, N Mamalis, L Ptacek, J Warner, B Katz, M Seay, A Crum, K Digre
	206	**Neuronal intranuclear inclusion disease with motor-predominant symptoms in a middle-aged woman** J Redding-Ochoa, L Chen, G Fitzpatrick
	207	**Cryo-EM Studies of PrP Amyloid: Comparison of Filaments' structures from Cases with the Q217R and the F198S PRNP Mutations** B Ghetti, K Ozcan, G Hallinan, A Moreno Garcia, A Fernandez, M Jacobsen, S Alijanvand, F Vago, H Garringer, K Newell, W Jiang, R Vidal
	208	**Single-Channel Dual Labeling of GS and GFAP Improves Astrocyte Detection in Spectrally Limited Multiplex Assays** C Corbett, M Dopler, S Etemadmoghadam, O Ogunbona, M Flanagan
	209	**Whole Slide Vector Visualization: A Visualization Platform for Interpreting and Utilizing AI Models in Histopathologic Image Analysis** A Rosado, J Vizcarra, T Pearce, D Gutman
	210	**From Atypical Ganglioglioma to ZFTA Fusion-Positive Ependymoma, How a Tumor Eventually Histologically and Molecularly Declared Itself** J Dixon, J Fullmer (***Note: Virtual Only***)

Posters are not offered for CME credit

PLATFORM 1: Tumors: Glial 1

1

Utility of Repeat Molecular Testing in Recurrent Glioblastoma – A Time-Stratified Analysis of 175 Matched Primary/Recurrence Specimens

M Mejia Bautista, R Multz, M Sukhanova, B Nezami, A Krbanjevic, D Ducket, L Santana dos Santos, R Castellani, L Jennings, P Jamshidi, J Ahrendsen; Northwestern Memorial Hospital

Background: Molecular profiling is common at initial diagnosis for glioblastoma (GBM); however, there are no guidelines regarding the timing or utility of repeat molecular testing upon disease recurrence.

Methods: We retrospectively reviewed all patients from our institution with both primary and recurrent GBM diagnoses between 2021-2025 and molecular testing performed on both primary and recurrent specimens. Molecular divergence was defined as the emergence or change of at least one pathogenic alteration compared to the prior specimen.

Results: We identified 152 patients, with 2-4 specimens per patient, yielding 175 total recurrent specimens. The most frequent reason for repeat molecular testing was related to treatment implications (e.g. emergence of targetable alterations, development of a hypermutant phenotype, change in MGMT status, clinical trial implications, etc.), accounting for 80.6% of cases. The average interval between primary and subsequent resection was 14.3 months. Overall, molecular divergence was observed in 80% (140/175) of recurrence specimens. Molecular changes between interval resections occurred in 71.4% of cases within < 3 months (15/21), 72.7% between 3–6 months (8/11), 76.5% between 6-12 months (39/51), and 84.8% beyond 12 months (78/92). Clinically important changes observed in later recurrences included emergence of hypermutation (6.9% of cases), change or clarification in MGMT status (10.3%), and major diagnostic reclassification (4.6%). For the latter, 63% of changes occurred 6 months after primary diagnosis, where an initial descriptive diagnosis (e.g. “hypercellular brain tissue”) was reclassified to GBM. Emergence of a targetable alteration was noted in just 2.9% of specimens (5/175), most frequently involving an FGFR3::TACC3 fusion. One patient showed a targetable MET fusion detected only on the third recurrence.

Conclusions: These preliminary findings suggest that molecular divergence is common, even in early recurrences, while clinically impactful changes are relatively rare but more likely to be seen in later (>12 months) recurrences.

2

Whole-Brain Neuropathologic and Transcriptomic Profiling Reveals Tumor-Associated Oligodendrocyte Reactivity in Glioblastoma

J Peng [1], S Oliver [1], N Freeburg [1], G Gopikrishna [1], D Chafamo [1], N Tokcan [2], D O'Rourke [3], I Nasrallah [3], Z Binder [1], D Silverbush [1], M Nasrallah [3]; [1] University of Pennsylvania, [2] University of Massachusetts Boston, [3] Hospital of the University of Pennsylvania

Background: Glioblastoma (GBM) is a highly aggressive primary brain tumor with extensive infiltration beyond radiographically and surgically defined margins. Postmortem donations enable systematic whole-brain sampling, providing a unique opportunity to integrate histopathology with single-cell molecular profiling to define tumor-associated changes across the brain.

Methods: We established a rapid postmortem brain tumor donation, processing, and analysis framework optimized for neuropathologic and molecular investigation. Autopsies were performed on four patients with high-grade gliomas, yielding 85 anatomically and clinically informed brain regions. Sampling prioritized tumor core, infiltrative margin, and non-lesional regions, guided by MRI and clinical records, with emphasis on non-operable regions such as brainstem. H&E-stained slides were assessed for tumor burden, extent of infiltration, and effect on normal brain. Based on histopathologic review, 14 adjacent flash-frozen samples were selected for single-nucleus RNA sequencing. Rigorous denoising removed postmortem-specific transcriptomic artifacts to enable construction of a high-quality whole-brain transcriptomic atlas aligned with histopathologic features.

Results: Computational analysis of a whole-brain transcriptomic dataset revealed distinct oligodendrocyte transcriptional states correlated with proximity to histologic tumor infiltration. Tumor-adjacent oligodendrocytes upregulated inflammatory, stress-response, and injury-associated pathways compared to distant regions, defining a reactive, disease-associated phenotype. This signature was enriched across external datasets including GBM, IDH-mutant glioma, pediatric high-grade glioma, multiple sclerosis, and traumatic brain injury, suggesting convergence on a shared oligodendrocyte response to CNS insult. Spatial single-cell transcriptomics demonstrated co-localization of mesenchymal-like GBM cells, macrophages, and hypoxia signatures with reactive oligodendrocytes. Longitudinal GBM analyses revealed this reactive oligodendrocyte state prior to therapy. Notably, histopathologically non-lesional regions with treatment-related changes also showed reactive oligodendrocyte signatures without detectable tumor.

Conclusions: By integrating whole-brain neuropathologic assessment with single-cell-resolved transcriptomics from postmortem donations, we demonstrate that GBM induces a spatially dependent, reactive oligodendrocyte response. These results highlight the value of postmortem tissue in capturing the full histologic and molecular landscape of tumor-associated brain responses.

3

MGMT pyrosequencing results as a continuous variable in glioblastoma: clinical and immunohistochemical correlates

M McCord, C Fadul, N McCann, H Ryu, D Schiff, B Purow, A Asthagiri, M Mut, M Lopes, V Smith; University of Virginia

Background: Many glioblastomas (GBMs) have methylation of the MGMT promoter, which improves temozolomide (TMZ) sensitivity. A popular method for detecting MGMT promoter methylation is pyrosequencing of 4 CpG sites in exon 1. The result is usually interpreted as binary (either “methylated” or “unmethylated”), with a widely accepted cutoff of 10% (averaged over the 4 sites) for a “methylated” result. However, there is growing recognition that methylation exists on a continuum, corresponding to progressively improving TMZ sensitivity.

Methods: To investigate this, we performed survival analysis on a cohort of 317 patients with GBM, all of whom underwent MGMT pyrosequencing and received TMZ. We also analyzed MGMT protein immunohistochemistry (IHC) on 62 tumor samples. We hypothesized that: 1) TMZ sensitivity is present at methylation levels lower than 10%; 2) Each CpG site each has a distinct optimal methylation percentage for TMZ sensitivity; and 3) MGMT IHC is a poor predictor of promoter methylation and TMZ sensitivity, based on previous work [PMID 18091318].

Results: Using the program CutoffFinder, we identified the optimal methylation level for predicting TMZ response as 7.4% ($p < 0.0001$, HR=2.38), and found that methylation as low as 2.0% predicts clinically significant TMZ sensitivity (p=0.0019, HR=1.726). We also found that each of the 4 pyrosequencing CpG sites has a unique optimal methylation percentage for TMZ sensitivity (#1: 4.5%, #2: 8.5%, #3: 10.5%, #4: 20.0%). Blinded semi-quantitative scoring of MGMT IHC (weak/moderate/strong) showed correlation with MGMT pyrosequencing results ($\chi 2=39.97$, $p < 0.0001$) and TMZ response (p=0.0127, HR=2.233). Quantitative IHC analysis also showed correlation with pyrosequencing (p=0.009) and TMZ response (p=0.0015, HR=2.25).

Conclusions: Our findings have significant implications for interpreting MGMT pyrosequencing in clinical practice, particularly in light of recent questions about TMZ use in GBMs with “unmethylated” MGMT promoter [PMID 38912869]. They also support a role for judicious use of MGMT IHC.

4

Detection of HSV latency-associated transcript expression in tumor-associated neurons after CAN-3110 therapy for glioblastoma

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Background: Glioblastoma (GBM) is an aggressive glial tumor marked by an immunosuppressive microenvironment, limiting the efficacy of therapies. In a first-in-human phase I trial of the oncolytic herpes simplex virus (HSV) CAN-3110, HSV-seropositive patients had prolonged survival compared with HSV-seronegative patients. We sought to characterize CAN-3110 persistence in treated patients.

Methods: Tissue from six patients enrolled in the CAN-3110 trial was screened for viral genomic DNA (vgDNA). The time-range between injection and tissue collection was 78 to 512 days. Viral protein expression was assessed by immunohistochemistry (IHC). To investigate latency, a GBM xenograft mouse model was analyzed using Latency Associated Transcript (LAT)-specific RNAscope in situ hybridization and HSV IHC at defined time points post-infection. Human biopsies from five patients who received serial injections of CAN-3110 were evaluated using multiplex RNAscope targeting LAT, OLIG1, FOXRB3, and GFAP.

Results: Viral genomic DNA was detected in four of six patients, including two in the contralateral brain hemisphere to the injection site. Three positive samples were negative for anti-HSV IHC, suggesting persistence without active protein expression. In mice, LAT and HSV viral proteins were co-expressed predominantly in implanted tumors at early time points post-infection. At seven days post-infection, most tumor cells lacked LAT and viral protein expression, with focal co-expression in deep gray matter and cortical neurons. Distinct neuronal foci expressed LAT alone. By 14 days, rare cells (< 1%) in tumor and cortex showed nuclear LAT expression without viral protein, consistent with latent-like persistence. In human samples, LAT signal predominantly colocalized with FOXRB3-expressing neurons.

Conclusions: CAN-3110 vgDNA sequences and latency-associated transcript expression can be detected in brain tissue following CAN-3110 administration. While we cannot distinguish LAT expression from engineered CAN-3110 vs wild-type HSV in human samples, these findings raise the possibility that CAN-3110 disseminates beyond the injection site and adopts a latent-like state, potentially through GBM–neuron interactions.

5

Genomic Alterations and Emerging CNS tumor types: An Evidence-Based Consensus Review by Cancer Genomics Consortium (CGC) Taskforce

M Sukhanova [1], L Hu [2], P Jamshidi [1], L Santana Santos [1], C Ida [3], S Neill [4], M Abedalthagafi [5], L Satgunaseelan [6]; [1] Feinberg School of Medicine, Northwestern Memorial Hospital, [2] GeneDx, [3] Mayo Clinic, [4] Emory University, [5] Tufts University, [6] Royal Prince Alfred Hospital, Australia

Background: Comprehensive molecular profiling has transformed the classification, diagnosis, and clinical management of CNS tumors. Advances in next-generation sequencing, methylation profiling, transcriptomics, and structural variant detection have uncovered a rapidly expanding set of genomic and epigenetic alterations that influence diagnostic decisions and clinical management.

Methods: To address the resulting interpretive complexity and the lack of a unified resource for clinical laboratories and pathology services, the CGC convened an international multidisciplinary CNS Tumor Gene List Taskforce. Using a structured literature review, incorporating 2,604 clinical studies published between 1997 and 2025, and an evidence-based tiering framework, the group curated 463 genes with established or emerging diagnostic, prognostic, therapeutic, and germline relevance in CNS tumors.

Results: The review identified 106 new diagnostic, 223 prognostic, and 73 therapeutic genes not yet incorporated into major guidelines. These alterations span diverse mechanisms, including single-nucleotide variants, copy-number changes, gene fusions, enhancer hijacking, methylation abnormalities, and shifts in transcriptome. Functional pathway analysis uncovered key oncogenic processes across CNS tumor types, revealing both shared pathways, such as TP53 and cell cycle regulation, RTK/RAS, PI3K, and NOTCH, and subtype-specific drivers relevant to gliomas, embryonal tumors, ependymomas, and meningiomas. In addition, we detail four emerging tumor groups: CNS tumors with PATZ1 fusions; PLAG(L1/2)-altered CNS tumors; glioneuronal tumors with ATRX alterations, kinase fusion, and anaplastic features (GTAKA); and CNS tumors with BCOR/BCORL1 fusions.

Conclusions: The resulting CGC CNS Tumor Gene List serves as a harmonized reference to support variant interpretation, molecular tumor board discussions, and design of targeted sequencing panels. It further provides a framework for integrating molecular biomarkers into forthcoming CNS classification schemes and guidelines for identifying therapeutic targets, including those with FDA-approved therapies, and supports future translational and clinical research in neuro-oncology.

6

FGFR3-mutant neuroepithelial neoplasms expand the spectrum of the FGFR3::TACC3-fused glioblastoma outlier methylation group

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Background: FGFR3::TACC3 (F3T3) gliomas may show oligodendroglioma-like morphology with calcifications and CD34-positive tumor cells, spanning a spectrum from low-grade histology to glioblastoma. However, the significance of this group, which forms a glioblastoma outlier methylation cluster, remains unclear. We present two cases that fall within this cluster but harbor non-fusion FGFR3 variants, with depiction of tumor evolution over 16 years.

Methods: We evaluated radiologic, histopathologic, next-generation sequencing, and DNA methylation profiling findings from two patients with FGFR3-mutant, IDH-wildtype tumors, integrating results within the established brain tumor landscape.

Results: The patients were 47- and 59-year-old women with partially calcified, minimally enhancing right cerebral tumors on imaging. Initial histology showed low-grade glioneuronal features with calcifications and CD34 positivity. Patient 1 had a germline SDHB alteration and prior renal cell carcinoma. Both tumors harbored TERT promoter c.-124C>T, FGFR3 p.K650E, and PIK3 variants. Methylation classified them in the glioblastoma, IDH-wildtype family, and suggested the methylation class "diffuse glioma, FGFR3 fusion-positive," with the dimensionality reduction analysis clustering the tumors together in this outlier group. Patient 1 underwent two additional resections (at 53- and 63-year-old); the third showed glioblastoma histology with retention of the PIK3CA and TERT promoter mutations, and new widespread copy number changes, including CDKN2A/B homozygous deletion. Additionally, the FGFR3 p.K650E mutation was no longer detectable, likely reflecting clonal evolution with expansion of a more aggressive subclone. Methylation profiling matched the tumor unambiguously to a mesenchymal glioblastoma.

Conclusions: FGFR3-mutant neuroepithelial tumors expand the "diffuse glioma, FGFR3 fusion-positive" methylation class. Our findings demonstrate that these tumors may initially present as low-grade glioneuronal neoplasms and can subsequently undergo malignant progression. This evolution may be driven by the acquisition of glioblastoma-associated genetic (e.g., CDKN2A/B deletion) and epigenetic features. This study expands a molecularly defined tumor family and documents a rare, stepwise pathway of glioma evolution not previously recognized.

7

Adult Diffuse Gliomas with FGFR3::TACC3 Fusion: Histopathology, Molecular Profile, and Clinical Outcomes

Y Zhu, N Karbhari, A Valerius, C Ida, C Zepeda-Mendoza, A Raghunathan, J Trejo-Lopez, A Nguyen, V Pazdernik, U Sener, C Giannini, R Vaubel; Mayo Clinic

Background: Diffuse gliomas harboring FGFR3::TACC3 fusion (DG-FGFR3::TACC3) frequently display molecular features of glioblastoma IDH-wildtype. The clinical behavior and molecular characteristics of these tumors remain incompletely understood.

Methods: In this single-institution analysis, slides from 31 adult patients (15 males, 16 females; median age 52 years) with DG-FGFR3::TACC3 were reviewed. Tumors were classified histologically as low (n=8, LG), intermediate (n=3, IG, ≥4 mitoses/10 HPF), or high-grade (n=20, HG, with microvascular proliferation and/or necrosis). Molecular studies, including chromosomal microarray (n=24; LG: 5, IG/HG: 19) and next-generation sequencing (n=31) were performed. Clinical histories and outcomes, including overall survival (OS) and progression-free survival (PFS), were assessed.

Results: Tumors showed microcalcifications (4 LG, 50%; 14 IG/HG, 61%), monotonous nuclei (6 LG, 75%; 7 IG/HG, 30%), oligodendroglioma-like perinuclear clearing (4 LG, 50%; 7 IG/HG, 30%), and fine branching vasculature (5 LG, 62%; 9 IG/HG, 39%). Mitoses were scant in LG (median 0/10 HPF) and frequent in IG/HG (median 6.5/10 HPF). Molecular features of glioblastoma were present in 6 LG (75%) and all IG/HG (100%), including TERT promoter mutation (6 LG, 75%; 23 IG/HG, 100%) and chromosomes 7 gain/10 loss (2 LG, 40%; 17 IG/HG, 89%). Initial treatment included resection (gross total in 11, subtotal in 17, biopsy-only in 3) followed by chemoradiation with temozolomide (n=25) with additional bevacizumab (n=4), pembrolizumab (n=1), and tumor treatment fields (n=3). Median follow-up was 21.3 months (LG) and 50 months (IG/HG). Recurrence occurred in 5 LG and 19 IG/HG, with median PFS of 22.9 months (LG) and 9.3 months (IG/HG). One LG and 14 IG/HG patients died; median OS was not reached in LG and 29.2 months in IG/HG.

Conclusions: DG-FGFR3::TACC3 tumors (LG and IG/HG) exhibit distinct histologic features and frequent molecular features of GBM. Consistent with prior studies, IG/HG subgroups in our cohort trended toward longer OS than fusion-negative GBM.

8

Self-reactive, tumor-associated CD4+ T cells in mouse models of high-grade glioma

E Russler-Germain, A Zhu, L Jorrin, T Hu, P Hsieh, C Hsieh, M Chheda; Washington University School of Medicine

Background: Immune checkpoint blockade for glioblastoma has failed in multiple clinical trials. To develop treatments that selectively enhance the activity of anti-tumor T cells and suppress the activity of tolerogenic T cells, we need a deeper understanding of tumor infiltrating T cell antigen specificities and phenotypes. However, a major limitation of T cell clonal analysis in both humans and mice is high diversity within the T cell receptor (TCR) repertoire, making inter-individual comparison of T cell clones difficult. This precludes precision immunotherapy.

Methods: We employ a fixed TCR beta chain transgenic mouse to decrease the total TCR repertoire. This permits identification of TCRs between different mice and experimental conditions. We analyze CD4+ T effector (Teff) and Foxp3+ T regulatory (Treg) cell clonality in mouse models of high-grade glioma including CT2A, GL261, and SB28. To classify tumor-associated TCRs that are reactive to tumor vs tumor-associated self-antigens, we compare these to TCRs found in germ-free mice, which is indicative of self-reactivity. We also assess TCR reactivity to tumor and self by using in vitro dendritic cell co-culture assays.

Results: Intracranial implantation of glioma cells results in high frequencies of Treg cells. TCR sequencing analysis of Teff and Treg cells reveals a mostly non-overlapping TCR repertoire between the two subsets, with individual T cell clones found in multiple mice. Treg clones are activated by self antigens, which can be increased in tumor. A subset of glioma-infiltrating Teff clones is reactive to probable tumor neoantigens while another subset is activated by constitutively presented self-antigens.

Conclusions: We show that in addition to tumor-specific CD4 Teff and self-reactive Treg cells, there are numerous self-reactive Teff clones within brain tumors. The functions of these Teff clones are unknown. Ongoing analyses of these subsets will identify unique markers and functional targets to inform development of new approaches to immunotherapy.

PLATFORM 2: Neurodegenerative: Alzheimer

9

Region-Specific Alterations in NPTX2 Expression in Alzheimer's Disease and Their Association with Dementia

K Chang [1], M Xiao [2], P Worley [2], D Nauen [2]; [1] National Taiwan University, [2] Johns Hopkins University School of Medicine

Background: Neuronal Pentraxin 2 (NPTX2) is a synaptic protein that has been shown to regulate excitatory–inhibitory balance at synapses from parvalbumin-positive interneurons and excitatory neurons. Reduced NPTX2 levels are found in Alzheimer's disease (AD); however, the cells affected and the nature of the alterations remain unclear.

Methods: Postmortem brain samples from neuropathologically confirmed AD, patients with AD pathology but retained cognitive function ('asymptomatic AD'), and age-matched controls were obtained from the Johns Hopkins Brain Resource Center. The neocortex, entorhinal cortex, amygdala, and hippocampus were examined. NPTX2 protein levels were assessed by immunohistochemistry, immunofluorescence, and Western blot. Quantification of NPTX2-positive neurons and microglia was performed in selected regions. The hippocampal hilus was microdissected for region-specific protein analysis.

Results: In AD, NPTX2-positive neurons were significantly reduced in the middle frontal gyrus compared with controls, which exhibited an age-related decline. In the hippocampus, NPTX2 expression showed a pattern distinct from the neocortex, with prominent localization in mossy fiber boutons within the hilus and CA3; this pattern was preserved in AD, confirmed by Western blot of microdissected hilar tissue. Numerous NPTX2-positive microglia were identified in AD brains but were rare in controls. In asymptomatic AD, neuronal NPTX2 expression was comparable to controls. NPTX2-positive neurons were significantly more numerous in controls and asymptomatic AD than in symptomatic AD.

Conclusions: NPTX2 loss in AD is region- and stage-specific. Neocortical reduction is associated with symptomatic disease, whereas hippocampal hilus and CA3 regions maintain expression even in severe AD. Preservation in asymptomatic AD suggests neuronal NPTX2 loss correlates with clinical progression rather than pathology alone. Increased NPTX2-positive microglia in symptomatic AD may reflect a shift toward a neuroinflammatory context. These findings suggest NPTX2 loss is associated with disease severity and may be a promising biomarker candidate in AD.

10

Differential Labeling of Neurofibrillary Tangle Maturity by New Phospho-Tau Epitopes

E Connolly [1], L Wu [2], N Gilyazova [2], A Liu [3], B Xu [2], S Wang [3]; [1] Duke University, [2] BRITE Institute, North Carolina Central University, Durham NC, USA, [3] Duke University Medical Center, Durham, NC, USA

Background: Neurofibrillary tangles (NFT), a defining pathological hallmark of Alzheimer's disease (AD), are hypothesized to progress through three morphologically defined maturity levels: pretangles, mature tangles, and ghost tangles. Antibodies targeting several phospho-tau epitopes currently used as fluid biomarkers, - including ,p-tau181, p-tau217, and p-tau231- have been shown to preferentially label early-stage tangles, whereas 3-R tau and conformation-specific antibody (GT-38) preferentially label late-stage tangles. Given the dynamic and heterogeneous post-translational modification (PTM) landscape of tau throughout disease progression, a broader panel of phospho-tau (p-tau) antibodies may provide a more comprehensive framework for assessing NFT maturity and AD stage.

Methods: We systematically screened over 20 site-specific phospho-tau (p-tau) antibodies using postmortem brain tissue spanning the full range of Braak stages. Initial Screening was performed by western blotting and ELISA to assess epitope-specific tau burden. Based on these results, epitopes demonstrating robust signal and stage-associated increases were selected for further study. Immunohistochemistry (IHC) was used to assess regional and cellular localization of selected p-tau epitopes. Neurofibrillary tangle maturity was classified as pretangle, intermediary 1, mature, intermediary 2, and ghost tangles

Results: Based on western blot and ELISA screening, we selected AT8 (p-tau202/205), p-tau198, p-tau212, p-tau217, p-tau356, and p-tau396 for detailed analysis. IHC revealed preferential labeling of pretangles by p-tau212 and p-tau356, intermediary to mature tangles for p-tau198, p-tau217, and AT8, and mature to ghost tangles for p-tau396.

Conclusions: This panel of p-tau epitopes could provide a comprehensive toolkit for interrogating NFT maturity levels across disease stages. Further, p-tau epitopes that show preferential labeling of pretangles may hold promise for further development into biomarkers for early AD diagnosis.

11

Characterizing the Role of TREM2 in Microglial Responses to Amyloid-β using Human Induced Pluripotent Stem Cells

C Cardona, R Patel, A Sproul, A Teich; Columbia University

Background: Amyloid-β (Aβ) aggregation is a hallmark of Alzheimer's disease (AD), and microglia play a central role in AD pathogenesis. TREM2 is primarily expressed in microglia, has been identified as a risk gene in AD, and plays critical roles in regulating immune function. However, the role of TREM2 in shaping microglial responses has not been fully defined, particularly in response to soluble oligomeric Aβ42 (oAβ42).

Methods: We utilized TREM2 knockout (KO) iPSC-derived microglia and isogenic controls to investigate how TREM2 affects microglial responses to oAβ42 exposure (6 hours). Transcriptomics and lipidomics were leveraged to understand how TREM2 affects microglial transcriptional programs and lipid metabolism. We also inhibited toll-like receptor (TLR) signaling and assessed cytokine secretion to better understand the drivers of neuroinflammation.

Results: Bulk RNA-sequencing revealed downregulation of the disease-associated microglial (DAM) transcriptional program in KO microglia under steady-state conditions. Pathway analysis identified cholesterol biosynthesis as the top pathway enriched in KO microglia. Following oAβ42 exposure, KO and isogenic control microglia shared a substantial number of differentially expressed genes, mainly related to immune signaling. While TREM2 expression had minimal impact on immune activation, inhibition of TLR signaling significantly reduced the induction of pro-inflammatory genes. Cytokine production was largely similar between genotypes, except for IL1β and IL10. Lipidomic analyses showed significantly elevated cholesterol esters in KO microglia at baseline, which was further increased after oAβ42 treatment, a response not observed in controls.

Conclusions: Transcriptomic analyses suggest possible roles for TREM2 in regulating DAM genes and lipid metabolism under baseline conditions. In response to oAβ42, acute immune activation and cytokine production were largely TREM2-independent, with TLR signaling driving the activation of pro-inflammatory genes. Lipidomic analyses revealed that the regulation of cholesterol esters was altered in KO microglia. Overall, our data identify TREM2 as a regulator of lipid metabolism at baseline and in response to oAβ42.

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Cognitively resilient individuals show enrichment for protective amyloid-responsive microglia (ARM)

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Background: Cognitive resilience, the discrepancy between normal cognition and intermediate to high Alzheimer's disease neuropathologic change (ADNC), may be mediated by microglial activation, specifically plaque associated amyloid responsive microglia (ARM). This study aims to characterize microglial morphologic, genetic, and activation states associated with resilience and their relationships with antemortem biomarkers.

Methods: High-cognitively unimpaired participants from the MCSA brain bank (top quartile of age adjusted global cognition; N=118) were evaluated to identify cognitively resilient (CR) individuals, defined as those with intermediate or high ADNC at autopsy (N=38). CR cases were matched to cognitively impaired controls by age, sex, ADNC level, and APOE4 status. Neuropathologic assessments included β amyloid, tau, and neuritic plaque staining, along with ARM (CD163) and β-amyloid immunohistochemistry in dorsolateral prefrontal cortex. ARM expression was quantified via digital image analysis to obtain CD163% and β-amyloid% areas. Regression models evaluated associations between cognition, amyloid PET, and microglial MS4A SNPs, while validation analyses assessed MS4A6A, CD163, and CD68 co-expression using co-immunostaining. Spearman correlations examined relationships between ARM% and antemortem MRI, PET, and cognitive performance.

Results: CR individuals harbored significantly higher ARM:amyloid than the CI group ($p< 0.001$), with fewer neuritic plaques ($p=0.03$). Furthermore, 28% of CR individuals harbored an MS4A6A SNP, which showed a protective effect on cognition despite amyloid burden ($p=0.043$), increased ARM per plaque ($p=0.0004$), and increased MS4A6A expression in ARM plaque-associated microglia ($p< 0.0001$). Paradoxically, CD68-positive plaque-associated microglia trended lower in CR brains ($p=0.6$). Antemortem white matter hyperintensities and global cognition were significantly correlated to ARM%; $r=0.556$ ($p=0.042$) and $r=0.648$ ($p=0.02$), respectively.

Conclusions: CR individuals exhibited enrichment of the microglial-specific MS4A6A SNP and higher plaque-associated ARM with reduced neuritic plaque burden. ARM levels were associated with better cognition and fewer white matter hyperintensities, supporting ARM as a potential protective microglial response independent of phagocytosis and highlighting MS4A6A as an activation marker.

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The Temporal Pole in SuperAgers

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Background: SuperAgers are individuals over age 80 with episodic memory of individuals 20-30 years younger. Our prior work on Superagers has shown relative resistance to neurodegeneration. Because of our interest in speech pathways, we routinely include temporal poles for neuropathologic examination and have identified a common pathologic phenotype.

Methods: 17 subjects from the Northwestern SuperAger Program (NUSAP) were examined. Through the NUSAP protocol, subjects underwent yearly neuropsychological assessments, assessed annually over an average of 9 years. Only subjects who were cognitively stable at their last visit were included. SuperAging status was defined by a score of 9/15 or more words on the delayed recall of the Rey Auditory Verbal Learning Task (RAVLT). All cases were assessed according to NIA-AA 2012 guidelines. We then compared phospho-tau (AT8), tau217, and amyloid-β (4G8) immunohistochemistry in the temporal pole and frontal cortex (BA8).

Results: 3 subjects were rated as no AD; 14 subjects were rated as 'low' AD. All low AD cases had neuritic plaques with tau-positive dystrophic neurites, neuropil threads, and neurofibrillary tangles (i.e., Alzheimer's disease neuropathologic change (ADNC)) involving the temporal pole only; the frontal cortex was spared of significant tau immunolabeling. The no-AD (no amyloid-β labeling) cases showed sparse tau immunolabeling in the temporal pole and no significant tau immunolabeling in the frontal cortex. Tau217 labeled both ADNC in the temporal pole in low-AD cases, and sparse neurofibrillary pathology in the three cases with no amyloid-β plaques.

Conclusions: A common pattern in cognitively stable SuperAgers was the presence of Alzheimer's disease neuropathologic change restricted to the temporal pole. This suggests that ADNC in the temporal pole is a feature of superior cognitive aging, raising questions about conventional distinctions between normal aging and ADNC. Tau217 in the temporal pole with and without amyloid-β suggests that tau217 is a 'biomarker' of normal aging.

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Plasma p-tau217 robustly detects autopsy-confirmed ADNC up to 15 years before death and outperforms p-tau181, GFAP and NfL across co-pathologies

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Background: Guidelines increasingly endorse fluid biomarkers for clinical decision-making in cognitive disorders, but most studies benchmark to clinical diagnosis despite pervasive mixed neuropathology. Autopsy-validated studies are needed to define performance across demographics, cognitive status, co-pathologies, and blood-to-death interval (i.e., how early pathologic signal is detectable).

Methods: 473 decedents from the University of Pittsburgh ADRC brain bank with stored plasma (p-tau217 n=401; p-tau181 n=409; GFAP n=408; NfL n=408) underwent neuropathologic examination classifying ADNC (Low n=93; Intermediate n=68; High n=312) and co-pathologies (Lewy body disease 248/473, LATE-NC 126/255, FTLD-TDP 16/473, FTLD-tau 38/473). We evaluated ROC performance for Intermediate/High versus Low ADNC as the primary endpoint.

Results: Plasma p-tau217, p-tau181, and GFAP increased stepwise with ADNC severity (all $p < 0.001$). p-tau217 demonstrated excellent discrimination for Intermediate/High ADNC (AUC 0.901; 95% CI 0.861–0.941). Biomarker combinations did not outperform p-tau217. Adding demographics yielded a modest statistically-significant improvement (AUC ~0.92; DeLong $p = 0.04$). Discrimination was stable up to 15 years before death (AUCs ~0.90 across age strata). Performance was preserved across MMSE at blood draw (including near-normal cognition [MMSE 25–30]), sex, APOE genotype, and co-pathologies. A modest attenuation was observed at ages 80-90. At matched Braak III–IV, Intermediate ADNC with moderate/frequent neuritic plaques had higher p-tau217 and p-tau181 than PART with no/sparse plaques, indicating plasma phospho-tau tracks Alzheimer-specific burden beyond tau tangles alone. NfL was highest in FTLD, particularly FTLD-TDP. p-tau217 and p-tau181 positivity predicted faster longitudinal MMSE decline and higher dementia conversion risk. Cross-sectional MMSE associations were largely mediated by ADNC, whereas NfL's association strengthened after neuropathologic adjustment, consistent with nonspecific neurodegenerative injury.

Conclusions: In the largest autopsy-confirmed cohort to date, plasma p-tau217 robustly detected ADNC up to 15 years before death and retained performance across demographic and clinical subgroups, supporting biomarker use for clinical decision-making in heterogeneous populations, with implications for diagnosis, trial enrichment, and implementation.

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The relationship between central and peripheral microRNA Alzheimer's Disease biomarker candidates

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Background: Our recent analysis of cross-sectional plasma samples from the Alzheimer's Disease Neuroimaging Initiative (ADNI) identified miRNAs whose levels were associated with Alzheimer's Disease (AD) diagnosis and predictive of progression from early mild cognitive impairment (MCI) to AD (Krueger DM et al., 2024, PMID39291752). Additionally, several miRNA levels were significantly associated with A/T/N positivity (Liu S et al., 2024, PMID39291737).

Methods: To validate candidate miRNA biomarkers and examine the relationship between brain and plasma miRNA signatures, we performed small RNA sequencing on 3-5 longitudinal plasma samples collected before and after AD diagnosis, as well as on cortical cells from postmortem brains of 21 individuals from the Framingham Heart Study (FHS). They comprised 10 "true controls", i.e. brains without pathological changes associated with any of the neurodegenerative diseases (NDs), and 11 "pure" AD cases, Braak stages 3 – 6, without other NDs pathologies present. Small RNAs, including miRNAs, were extracted from cortical neurons, astrocytes, and microglia (~ 4,000 per cell-type), individually dissected from cryosections of prefrontal and hippocampal cortices via laser capture microscopy

Results: We identified differentially expressed miRNAs that were region- and cell-type specific, including several with documented roles in memory and associations with MCI to AD dementia conversion in plasma. Moreover, we identified phenotypic responses of iPSC-derived neuronal and glial cells in functional in vitro tests, to mimics and inhibitors of our miRNA AD-biomarker candidates offering new insights into pathogenetic mechanisms that may inform the development of disease-modifying therapies.

Conclusions: Together, these results enable us to integrate cortical cell type-specific miRNA signatures with plasma miRNA profiling in support of miRNAs as reliable biomarker candidates for AD.

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Downstream Neuroprotection Associated with Amyloid Targeting Therapy

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Background: Understanding whether amyloid targeting therapies can meaningfully alter the long-running biological cascade leading to neurodegeneration is an important question. In particular, the extent to which amyloid clearance influences downstream tau pathology and associated tissue loss has remained uncertain.

Methods: We describe a unique case integrating longitudinal amyloid and tau PET imaging, structural MRI, and postmortem examination that suggests substantial removal of amyloid may mitigate tau accumulation and neurodegeneration.

Results: The patient, a 52-year-old man with mild cognitive impairment, underwent approximately four and a half years of aducanumab treatment before discontinuation due to a potential local infusion-related reaction. Despite remaining amyloid-positive on PET, the Centiloid measure decreased markedly from 139 at baseline to 33. He passed away four years after treatment cessation at age 62. Neuropathologic evaluation revealed a highly variable pattern of amyloid removal across the cortex. Some regions showed profound plaque clearance, whereas adjacent areas demonstrated only modest reduction. Areas with limited amyloid removal typically exhibited clearance confined to superficial cortical layers and harbored a tau burden consistent with expected Alzheimer's disease pathology. In contrast, regions displaying deep, multilaminar amyloid removal, most often located along gyral crowns, showed strikingly low tau pathology. Parallel in vivo analyses supported these observations. PET and MRI demonstrated that regions with extensive amyloid reduction exhibited lower tau PET signal and experienced minimal to no cortical atrophy, in contrast to regions with limited clearance. Permutation analyses confirmed that these differences were unlikely to be explained by chance. Furthermore, atrophy within extensively cleared regions occurred at a slower rate than in comparable regions from a matched Alzheimer's Disease Neuroimaging Initiative (ADNI) reference group.

Conclusions: These findings suggest that when amyloid-targeting therapy achieves substantial and widespread plaque removal, the affected cortical regions may be comparatively resistant to subsequent tau deposition and neurodegeneration.

PLATFORM 3: Tumors: Glial 2

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Molecular, histologic, and clinical characterization of 51 gliomas with an epigenetic classification of gliomatosis cerebri-like glioma

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Background: Adult-type diffuse high-grade glioma, subtype F (HGG-F, Heidelberg), or gliomatosis cerebri-like glioma (GCLG, Bethesda), is a recently described DNA methylation-based CNS tumor classification that may behave less aggressively than IDH-wildtype glioblastoma (GBM).

Methods: To investigate biologic, histologic, molecular, and clinical features of GCLG, a cohort of 51 tumors with concordant predictions to HGG-F/GCLG by both Heidelberg (v12.8) and Bethesda (v3.1) classifiers, including 36 classified with high confidence (>=0.9) by both classifiers and 16 with lower confidence, was interrogated.

Results: Patient median age was 67 years (range 38-84 years) with a male predominance (female 12, male 39). Concomitant chromosome 7 gain and 10 loss was observed in 5 of 51 (10%) samples. None harbored amplifications in EGFR or MDM2, while PDGFRA amplification and CDKN2A/B deep deletion was observed in a single sample (lower confidence set). In samples tested for TERT promoter mutations (n=24), 23 (96%) harbored mutations (19 c.-124C>T, 4 c.-146C>T). Among samples tested for PIK3CA and PIK3R1 mutations (n=12), 11 (92%) harbored one of the two mutations (6 PIK3R1, 4 PIK3CA), and one harbored both. MGMT promoter methylation was detected in 12 of 51 (24%) samples. Digital whole slide images were available for 28 tumors, and these demonstrated consistently bland morphology with mild hypercellularity and glial atypia and an infiltrative growth pattern without identifiable mitoses, microvascular proliferation, or necrosis. For 20 samples with available MRI reports, signal changes were centered in the temporal lobe and showed a gliomatosis-type pattern, with minimal enhancement. Comparisons of copy number changes with those of other tumor types indicated biologic differences. Overall survival of 19 patients with clinical follow-up was more similar to that of patients with methylation class IDH-mutant astrocytoma (low grade) than GBM.

Conclusions: Tumors classified as GCLG are epigenetically and clinically distinct from IDH-wildtype glioblastomas and warrant dedicated study to identify appropriate therapeutic options.

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Correlation of ATRX Mutation Status, ATRX Immunohistochemical Staining Results, and ALT Status in IDH-Mutant Astrocytoma

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Background: Loss of ATRX expression by immunohistochemistry is sufficient to support a diagnosis of IDH-mutant astrocytoma over IDH-mutant and 1p/19q-codeleted oligodendroglioma. As cases with astrocytic morphology but retained ATRX expression may cause diagnostic delay, we examine the ATRX mutational landscape and correlate mutation, immunoreactivity, and ALT status in this study. We hypothesize that ALT status would reconcile discrepancies between ATRX mutation and protein expression.

Methods: We reviewed institutional cases of IDH-mutant astrocytoma with available paired next-generation sequencing and ATRX immunohistochemical data. ATRX immunohistochemical staining was performed in all cases. Telomere-specific fluorescence in situ hybridization (FISH) to determine ALT status was also performed.

Results: We identified 102 cases of IDH-mutant astrocytoma (Gr2=58, Gr3=20, Gr4=24). The majority (n=70) harbored at least 1 truncating ATRX alteration, and all these cases demonstrated loss of ATRX by immunohistochemistry. A subset (n=10) instead harbored non-truncating missense mutation. Of these, 8 demonstrated retained ATRX expression whereas 2 demonstrated loss. FISH was performed in 8 cases with ATRX missense mutations, and all 8 cases were ALT-positive. The remaining apparently ATRX-wildtype cases (n=22) were confirmed chromosome 1p/19q-intact. Of these, 14 showed loss of ATRX. In the 8 cases without ATRX mutation and retained ATRX expression, FISH was performed in 4 cases with available material and all 4 were ALT-negative.

Conclusions: FISH resolved the discordance between mutation and immunohistochemical expression status in cases with missense ATRX mutations and retained protein expression, supporting potential utility as a supportive tool in diagnostic workup. Here, we functionally demonstrate ALT phenotype in IDH-mutant astrocytoma with non-truncating ATRX missense mutations and retained protein expression. Localization of these missense mutations near the “Helicase C-terminal” domain seems biologically relevant. ALT assessment can adjudicate discordant ATRX genotype–phenotype findings and may refine the diagnostic algorithm when ATRX immunohistochemistry is retained.

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Genomic profiling of genetically-defined oligodendroglioma harboring TP53 alterations

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Background: TP53 alterations [mutation and copy number variation (CNV)] are uncommon in oligodendroglioma and their significance is unclear. We compare 20 adult oligodendroglioma, IDH-mutant and 1p/19q-codeleted harboring TP53 alterations (O-TP53+) to 50 lacking TP53 alterations (O-TP53-).

Methods: Molecular profiling was performed by a single laboratory via next-generation sequencing panels and OncoScan chromosomal microarray in all cases, and methylation profiling using Illumina Infinium Methylation EPIC v1.0/2.0 and the NCI/Bethesda v2 classifier in 14 O-TP53+.

Results: O-TP53+ occurred in adults (mean age, 52; range 27-76), 7 male:3 female, with equal primary/recurrent presentation. Compared to O-TP3-, O-TP53+ were enriched for ambiguous "oligoastrocytic" morphology (17/20 vs 22/50), grade 3 (17/20 vs 20/50), abnormal p53 expression (10/15 vs 0/40), loss of ATRX expression (3/16 vs 0/39), and CDKN2A/B deletion (13/20 vs 11/50), while less frequently had CIC mutations (0/8 vs 30/50); $p< 0.05$, all comparisons. Other features did not significantly differ, including TERT mutations (17/20 vs 49/50). TP53 alterations included mutations (n=11) and/or CNV (heterozygous loss, n=10; copy-neutral loss of heterozygosity [cnLOH], n=8), and inactivation was partial in 10 (7 heterozygous loss, 2 cnLOH, 1 single mutation) and complete in 10 (6 mutation+cnLOH, 3 heterozygous loss+mutation, 1 two mutations) cases. Epigenetically, 9 cases aligned with methylation class Oligodendroglioma (7 matched [score≥0.900]) and 4 with class Astrocytoma, IDH-mutant, high-grade (1 matched); a single case did not match (family score < 0.900) despite high tumor purity (IDH mutation VAF 50%). All 3 heterozygous loss+mutation cases were confirmed hypermutant (IDH mutation VAF 35-69%, 2 recurrent/1 unknown) and aligned with class Astrocytoma, IDH-mutant, high-grade (1 matched). All 4 tested cases with mutation+cnLOH (typical of IDH-mutant astrocytoma) epigenetically aligned with class Oligodendroglioma (2 matched).

Conclusions: These findings suggest that O-TP53+ more frequently show ambiguous oligoastrocytic morphology, are often high-grade, and may epigenetically align with Astrocytoma, IDH-mutant, high-grade.

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Alterations in Mismatch Repair Proteins Impact Survival in Oligodendroglioma

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Background: The presence of mutation in mismatch repair (MMR) proteins such as MSH2, MSH6, MLH1, PMS2, and POLE have been shown to have a detrimental effect on prognosis in IDH-mutant astrocytoma, but whether mutation in these proteins has a similar effect in oligodendroglioma has not been explored in-depth thus far.

Methods: A cohort of 896 oligodendroglioma cases were collected from publicly available datasets (GENIE, GLASS, and MSKCC). The frequency of mutations in MSH2, MSH6, MLH1, PMS2, and/or POLE was assessed in either primary tumors or the first-available specimen. Kaplan Meier plots were performed to evaluate the impact of mutation in MMR proteins on progression-free survival (PFS) and overall survival (OS).

Results: Mutation in at least one MMR protein was detected in 36 cases (36/896, 4%), with mutation in MSH2 being the most common (21/36, 58.3%). There was no significance difference in the age at diagnosis, sex, CNS WHO grade, and number of cases with CDKN2A/B loss between the MMR intact and mutant groups ($p>0.05$), however there was a significantly higher tumor mutation burden in cases with MMR mutation ($p= 0.025$). The presence of MMR protein mutation was shown to have a significant difference in overall and progression free survival in oligodendroglioma (OS $p= 0.0024$/ HR= 2.42; PFS $p= 0.0066$/ HR= 3.06).

Conclusions: While MMR proteins mutations are uncommon in oligodendroglioma, when present they may portend worse overall and progression free survival. Along with prognosis, regular evaluation for MMR mutation in oligodendroglioma may help to identify cases that could benefit from treatment with immune checkpoint inhibitors. Future areas for investigation include whether the development of MMR mutation in tumor recurrence after treatment has a similar impact on survival as MMR mutation present at initial diagnosis.

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MicroRNA changes can drive senescence in pilocytic astrocytoma

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Background: Pilocytic astrocytoma and other pediatric low-grade gliomas (pLGG) undergo oncogene-induced senescence, but the role of microRNA in this is poorly understood. Cultured pLGG cells were used to identify microRNA species whose levels are altered in senescence, and to test their causal link to the process.

Methods: Oncogene-induced senescence was modeled using pLGG cultures grown in conditional reprogramming cell (CRC) conditions then transitioned to high serum media. Phenotypic changes in culture were evaluated using quantitative RT PCR, western blotting, as well as growth, apoptosis and acidic beta-galactosidase assays.

Results: Cells transitioned to high serum dramatically slowed their growth and upregulated a range of senescence markers including p16, p21, p27, beta-galactosidase and DNA damage pathways. Sequencing of three pLGG CRC lines before and after induction of senescence identified microRNA which increased or decreased in abundance, with changes confirmed using quantitative RT PCR. miR-29b-3p and miR-335-3p levels decreased after induction of senescence, while miRNA-21-5p, miR-143-5p and miR-145-5p all increased. The introduction of miR-143-5p or miR-145-5p mimics into pLGG cells resulted in induction of multiple senescence markers, as well as a dramatic slowing of tumor cell growth, suggesting that they are be able to promote senescence. Similar effects were seen in Res186 pLGG cells with an intact CDKN2A locus, but not in Res259 or BT40 cells with CDKN2A loss. Carboplatin response was decreased by miR-29b-3p or miR-335-3p mimics, however, the senescent cultures did not show increased sensitivity to obatoclax.

Conclusions: Pilocytic astrocytoma and other pLGG culture models of senescence were used these to identify miRNA changes associated with growth arrest. Increased levels of miR-143-5p and 145-5p were noted, and mimics of these miRNA were able to slow growth and promote senescence of pLGG cells with an intact CDKN2A locus. These miRNA therefore represent potential novel genetic therapies for pilocytic astrocytoma.

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Beyond the Posterior Fossa: Clinical and Molecular Characterization of 33 Ependymomas with PFB Molecular Signatures

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Background: Ependymomas comprise a heterogeneous group of circumscribed gliomas characterized by perivascular pseudorosettes and ependymal rosettes. Current classification integrates morphology, molecular, with DNA methylation defining biologically distinct entities. Posterior fossa ependymomas are divided into posterior fossa group A (PFA) and group B (PFB), which differ in age distribution, and molecular signatures. Methylation profiling is considered the diagnostic gold standard and emerging data suggests that molecular grouping may not always align with anatomic location.

Methods: Thirty-three cases classified as posterior fossa ependymoma with high and suggestive confidence scores by the Heidelberg and NCI methylation classifiers were identified from NCI consultation cohort. Detailed radiologic, histologic, and molecular data were collected through collaboration with the submitting academic centers.

Results: We present 33 ependymal tumors with posterior fossa (PFE) molecular profiles despite occurring in non–posterior fossa locations. 100% of cases (n=33) subclassified as Posterior Fossa Group B (PFB). The series included 11 primary and 22 recurrent/ metastatic tumors. Age range was 15 to 79 years, with a median age of 42 and a slight female predominance (female: male ratio 1.53). The most common initial diagnosis was ependymoma; however, some were classified as high-grade papillary or choroid plexus tumors. Twenty-three tumors were CNS WHO grade 2, nine grade 3, and one indeterminate. Primary tumor sites included supratentorial intraventricular (n=5), pineal region (n=1), posterior corpus callosum (n=1), and spinal cord (n=4). Notably, all recurrent/metastatic tumors consistently demonstrated PFB methylation profiles regardless of location.

Conclusions: Ependymomas with a Posterior Fossa Group B (PFB) molecular signature are not only confined to the posterior fossa and can arise as primary tumors throughout the neuraxis. Our findings suggest that this anatomic-molecular discordance is a phenomenon specific to the PFB subtype. Therefore, comprehensive molecular profiling is essential for the accurate diagnosis and classification of all ependymomas, regardless of their site of origin.

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Eighteen cases of glioneuronal tumor, kinase-fused, subtype A (GNT_KinF_A) from a multi-institutional study

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Background: DNA methylation profiling has enabled recognition of provisional glioneuronal tumor (GNT) entities defined primarily by epigenetic signatures. GNT_KinF_A is a recently described methylation class characterized by kinase fusions, but its clinicopathologic spectrum remains incompletely defined.

Methods: Eighteen pediatric and adult brain tumors classified as GNT_KinF_A with high confidence by Heidelberg and NCI methylation classifiers were identified through a multi-institutional NCI cohort spanning fifteen academic centers. Clinical, radiologic, histologic, immunophenotypic, molecular, treatment/ outcome data were analyzed.

Results: Original diagnoses varied widely, including low- and high-grade gliomas and other glioneuronal tumors, underscoring marked histologic heterogeneity. Median age was 9 years with strong male predominance (14/18). Tumors were predominantly supratentorial (15/18), most often parietal/occipital; two were intraventricular and one intramedullary. Imaging typically showed solid-cystic masses with heterogeneous enhancement. Histologically, tumors demonstrated broad morphologic variation; over half showed focal nodular hypercellularity. Oligodendroglial features were present in 41%, mixed glial-neuronal components in 29%, and high-grade features (brisk mitoses, microvascular proliferation, necrosis) in 24%. Dysmorphic ganglion cells and multinucleation were seen in 40–50%. OLIG2 and synaptophysin were expressed in ~80%; ATRX was retained in all cases; Ki-67 ranged from < 1% to 50%. Receptor tyrosine kinase fusions were identified in 14/18 cases, most commonly NTRK2 (with additional NTRK1/3 and MET fusions). Recurrent copy number alterations included chromosome 22q loss (10/18) and less frequently 9q gain and 19q loss; most tumors lacked MGMT promoter methylation. Among 12 patients with follow-up (up to 126 months), seven underwent resection alone, three received chemoradiation or chemotherapy, and two received larotrectinib. Despite some recurrences, overall survival was 100%, including tumors with high-grade histology.

Conclusions: GNT_KinF_A is a kinase fusion–driven, molecularly defined tumor with striking morphologic variability that limits reliable histologic diagnosis. Despite occasional high-grade

features, it appears clinically indolent with excellent survival, underscoring the essential diagnostic and therapeutic implications of DNA methylation profiling.

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FGFR2-Fused Low-Grade Neuroepithelial Tumors: Clinicopathologic and Methylation Analysis of a National Cohort with WHO Implications

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Background: Low-grade neuroepithelial tumors with FGFR2 fusions represent a rare and diagnostically challenging subset of pediatric and young adult brain tumors. FGFR2-rearranged tumors demonstrate morphologic and molecular heterogeneity, with overlap across established entities including ganglioglioma (GG) and polymorphous low-grade neuroepithelial tumor of the young (PLNTY). Their relationship to current DNA methylation classes and implications for tumor classification remain incompletely defined.

Methods: We analyzed 18 FGFR2-fused low-grade neuroepithelial tumors (4 institutional cases; 14 from the Childhood Cancer Data Initiative Molecular Characterization Initiative (MCI). Clinical and histopathological data were reviewed. DNA methylation array using IGM CNS v1 classifier and NIH Bethesda v3 classifier and structural variants/fusion partners were analyzed on all 18 patients.

Results: Patients ranged in age from 3-15 years, and all cases harbored FGFR2 fusions involving 8 unique fusion partners (PHGDH, INA, SHTN1, CTNNA3, RBFOX3, ERC1, CLIP1, PASD1). Histologically, 13/18 tumors demonstrated a PLNTY-like morphology with a conspicuous neuronal component in at least 8 cases. Methylation analysis resulted in matches to GG or PLNTY methylation classes with a high confidence score for all cases. On UMAP dimensionality reduction plots, all cases clustered with GG and/or PLNTY. Interestingly, 7 cases that matched to PLNTY class had a conspicuous neuronal component.

Conclusions: Our institutional series, together with multi-institutional cases enrolled through the MCI, highlights the morphologic and epigenetic heterogeneity of FGFR2-fused low-grade neuroepithelial tumors. Updated DNA methylation classifiers may refine classification in a subset of cases; however, some tumors remain difficult to categorize within current diagnostic frameworks. Although PLNTY is currently designated as a pediatric-type diffuse low-grade glioma in the WHO classification, the frequent neuronal differentiation observed in FGFR2-fused cases suggests closer alignment with glioneuronal tumors. Collectively, these observations support ongoing efforts to further delineate FGFR2-rearranged neoplasms and inform future refinements of CNS tumor classification.

PLATFORM 4: Neurodegenerative: Alzheimer, Other

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Unusual Neuropathological Findings in Cases of Unexpected Death with No Apparent Anatomical Causes

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Background: Sudden unexpected death caused by central nervous system (CNS) pathologies is rare compared to cardiovascular causes, and includes acute vascular compromise (hemorrhages, infarcts), mass effect/obstructive phenomena causing hydrocephalus/cerebral edema/herniation, traumatic injuries causing diffuse axonal injury (DAI)/pontomedullary rent, and sudden unexpected death in epilepsy (SUDEP). Sudden death in multiple system atrophy (MSA) and Parkinson's disease (PD) has also been described and, as some such cases are thought to involve injury to respiratory/autonomic centres, the potential for other conditions to mediate similar dysfunction might be considered in cases of unexpected death without apparent anatomical cause.

Methods: Adult autopsy cases referred to neuropathology at LHSC over the previous 20 years were investigated, screening for those with unexpected death without an apparent anatomical cause of death at time of autopsy. Cases with acute hemorrhages/infarcts, head trauma, DAI, pontomedullary rent, and mass effect phenomena (including brainstem herniation) were excluded, as were those with history of seizures to account for SUDEP.

Results: Thirteen cases fulfilled the criteria, almost all of which involved considerable neurodegenerative disease findings. These included MSA, multiple sclerosis, progressive supranuclear palsy (PSP), Saito stage 3 argyrophilic grains disease (AGD), high-level (A3B3C3) Alzheimer's disease, and diffuse Lewy body disease. One case of PD with chronic inflammation in the brainstem and one of an infiltrative glioma were also examined. All cases demonstrated marked involvement of the hypothalamus (4/13), pontomedullary brainstem (7/13), or both (2/13) by the predominating neuropathological disease process.

Conclusions: Despite dissimilar pathological processes, all cases of unexpected death demonstrated involvement of the respiratory/autonomic centres of the hypothalamus/brainstem: sites thought to be implicated in sudden death in MSA and other diseases. Although rarely representing the sole possible consideration, the potential for CNS pathologies to cause sudden unexpected death, particularly in the context of severe hypothalamic/brainstem involvement, should be entertained in cases without apparent anatomical cause of death.

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TDP-43 and tau-related neurite loss is associated with locus coeruleus dysconnectivity and beta-amyloid accumulation in Alzheimer's disease

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Background: Alzheimer's disease (AD) involves early degeneration of the locus coeruleus (LC), a brainstem nucleus with widespread cortical and subcortical projections. Pathologic proteins central to AD, beta-amyloid (Aβ), hyperphosphorylated tau (p-tau), and phosphorylated TDP-43 (pTDP-43), can disrupt neurite (dendrites and axons) transport and synaptic stability. This study aims to examine how LC-centered micro- and macrostructural connectivity relates to AD-associated proteinopathies.

Methods: Neurofilament heavy chain (NF-H)-labeled neurite length—normalized to healthy controls to assess for neurite loss—, Aβ, pTDP-43, and p-tau burden were quantified in sixteen regions within pons, midbrain, amygdala, hippocampus, superior-middle temporal (S/M-TG), and frontal gyrus (S/M-FG) from 21 high and low-likelihood AD cases with and without TDP-43, and controls. Antemortem multi-shell diffusion MRI (DTI) was obtained. Fractional anisotropy (FA) and mean diffusivity (MD) were calculated across twelve LC-connected cortical and subcortical grey matter nodes (medulla, pons, midbrain, amygdala, hippocampus, thalamus, basal forebrain, insula, S/M-TG, S/M-FG, parietal and occipital cortices) (LC network). Aβ-PET standardized uptake value ratios (SUVRs) were measured in corresponding LC-network regions to assess the relationship between LC network and Aβ burden.

Results: We found pTDP-43 and p-tau accumulation within LC. Neurite loss was prominent in basolateral amygdala, entorhinal cortex, and S/M-TG—regions with dense reciprocal LC connectivity—and co-localized with pTDP-43 and p-tau pathology. In contrast, the S/M-FG showed high extracellular Aβ burden with low p-tau and pTDP-43 burden and relatively preserved neurite integrity. DTI-derived LC-network maps revealed significant FA reductions and MD increases across the LC network in AD. Aβ-PET demonstrated elevated SUVRs throughout LC-connected cortical regions.

Conclusions: LC network degeneration in AD is characterized by convergent neurite loss, tau and TDP-43 pathology, disrupted structural connectivity, and elevated cortical Aβ burden. These findings suggest that tau and TDP-43 derived LC dysconnectivity is linked to Aβ accumulation, highlighting the LC as a central hub in AD-related network degeneration.

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Identification of molecular-neuropathological signatures of sporadic AD and FTLD

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Background: Alzheimer's disease (AD) is the most common neurodegenerative condition in individuals over 65 while frontotemporal lobar degeneration (FTLD) is the leading postmortem non-AD dementia pathology. Neuropathological features of AD and FTLD disproportionately affect specific brain regions and subsets of brain cells. A prevailing hypothesis is that certain cells are inherently more vulnerable to pathological stimuli, leading to selective cellular dysfunction, accumulation of neuropathological features, and/or cell death, a concept known as selective vulnerability. Single-nucleus RNA sequencing of postmortem brains from AD and FTLD has revealed disease-specific cell-state changes and cell-type depletion. However, snRNA-seq lacks spatial/pathological contexts and is unable to directly link transcriptomics to cellular pathology. Although recognized for decades, the molecular mechanisms of selective vulnerability to neuropathology remain unclear, limiting insights into disease onset and progression.

Methods: We propose to leverage the same-slide single-cell spatial multi-omics (Xenium 5K+Phenocycler-Fusion 30 targets) to directly in situ identify the molecular-neuropathological signatures in the brains of individuals with AD and FTLD. By comparing them within and across diseases, we aim to capture shared and disease-specific cellular and molecular alterations directly linked to mixed neuropathology at single-cell level, insights previously unattainable.

Results: We have employed single cell spatial multi-omics workflow and profiled 46 individuals with AD, Pick's disease, PSP and age-, gender-matched controls and are in the process of bioinformatics data analysis. We aim to identify molecules associated with Amyloid pathology (diffuse/dense amyloid plaques, cerebral amyloid angiopathy), Neuronal p-tau(neurofibrillary tangles, Pick body, globose tangles), and Glial p-tau pathology (tufted/ramified astrocytes, coiled bodies). We will also uncover cellular and molecular alterations across different etiologies, specific to 3R/4R tau isoforms, spatial niche-dependent changes, and cell-cell interactions.

Conclusions: The results from this project will provide critical insights to the cellular and molecular basis of selective vulnerability in both neurons and glial cells to neuropathology, a critical gap in current research.

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Association of Cerebrovascular Pathologies with Hippocampal Sclerosis of Aging in Community-Dwelling Older Adults

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Background: Hippocampal sclerosis of aging (HS-A) is characterized by severe neuronal loss and gliosis, primarily affecting the CA1/subiculum hippocampal sub-regions. HS-A commonly coexists with both neurodegenerative and cerebrovascular diseases. We investigated associations between cerebrovascular pathologies and HS-A in community-dwelling older adults.

Methods: Participants (N = 915) who underwent autopsy and neuropathologic evaluation were selected from four longitudinal community-based cohort studies: the Religious Orders Study, Rush Memory and Aging Project, Minority Aging Research Study, and Clinical Core. Three hippocampal samples (bilateral anterior hippocampus and unilateral mid hippocampus) were used to determine HS-A based on the presence of severe neuronal loss and gliosis in the CA1-subiculum region. Cerebrovascular pathologies assessed included arteriolosclerosis from basal ganglia, atherosclerosis, macro- and microinfarcts, and cerebral amyloid angiopathy (CAA). Alzheimer disease neuropathologic change and Lewy body pathology were also assessed. Multivariate logistic regression models examined the associations between cerebrovascular pathologies with HS-A, adjusting for age-at-death, sex, education, and other brain pathologies.

Results: Participants had a mean age at death of 91.2 years and 16.2 years of education, with 74% being women. HS-A involving the mid- and/or the anterior hippocampi was 29.4%. Cerebrovascular pathologies were common: atherosclerosis (26.8%), arteriolosclerosis (35.8%), macroinfarcts (36.5%), microinfarcts (38%), and CAA (44.4%). In adjusted models, arteriolosclerosis in the basal ganglia (OR = 1.18, 95% CI: 1.01–1.36, p = 0.03) and atherosclerosis (OR = 1.45, 95% CI: 1.18–1.77, p = 0.0003) were each associated with higher odds of HS-A. No association was observed between infarcts or CAA and HS-A.

Conclusions: Both large- and small-vessel disease, specifically atherosclerosis and arteriolosclerosis, are associated with HS-A. These findings support an important vascular contribution to HS-A pathogenesis in aging and further underscore the role of vascular disease in Alzheimer's and mixed dementias. Future studies should investigate mechanisms linking vascular pathologies to hippocampal neuronal vulnerability in aging.

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Quantifying Vascular Contributions to Dementia Across Large-Scale Neuropathology Cohorts

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Background: Cerebrovascular pathology is highly prevalent in aging brains and contributes substantially to cognitive impairment and dementia, yet standardized and scalable approaches for quantifying cerebral small vessel disease (SVD) across multicenter neuropathology datasets remain limited. The National Alzheimer's Coordinating Center (NACC) Neuropathology Form Version 11 (NP11) captures key vascular lesions but lacks region-specific ratings required for established vascular cognitive impairment neuropathology guidelines (VCING). We developed harmonized indices to quantify SVD burden and estimate vascular cognitive impairment likelihood using NP11 data and examined their associations with cognition and dementia near death.

Methods: Using NP11 data from 1,658 autopsied cases across 32 Alzheimer's Disease Research Centers, we constructed a continuous six-domain SVD burden index (range 0-18) encompassing arteriolosclerosis, white matter rarefaction, cerebral amyloid angiopathy, microinfarcts, gross infarcts/lacunes, and microbleeds, with proportional scaling when ≥5 domains were available. We additionally developed a rule-based VCING-Lite proxy, adapting established VCING criteria using global NP11 ratings. Robustness was assessed across complete, domain-complete, and Alzheimer's disease neuropathologic change (ADNC)-limited subsets. Associations with global cognition (latent composite derived from NACC Uniform Data Set testing) and clinical dementia were evaluated using multivariable regression models adjusted for demographic factors and co-pathologies.

Results: SVD burden scores were stable across inclusion criteria and strongly correlated with VCING-Lite classification (Spearman r=0.74-0.75). Higher SVD burden and VCING-Lite category were independently associated with worse global cognition and increased odds of dementia when clinical assessments occurred within three years of death, persisting after adjustment for ADNC, other co-pathologies, and hypertension.

Conclusions: These harmonized, NP11-compatible indices provide scalable and reproducible tools for quantifying cerebrovascular pathology and modeling vascular contributions to cognitive impairment in large, multicenter neuropathology cohorts, enabling improved stratification and adjustment in mixed-dementia research.

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Regional Heterogeneity of Cerebral Amyloid Angiopathy Defined by Deep Learning–Based Vessel-Level Quantification

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Background: Cerebral amyloid angiopathy (CAA) is defined by amyloid-β deposition within cortical and leptomeningeal vessel walls and is a major cause of lobar intracerebral hemorrhage and treatment-related hemorrhages during anti-amyloid therapy. Current semi-quantitative grading fails to capture critical regional and vessel-specific heterogeneity. Consequently, reproducible, objective vessel-level quantification is needed to better define spatial vulnerability and improve prediction of localized hemorrhage risk and clinical outcomes.

Methods: We trained a Swin Transformer-based U-Net model on 20 internal Alzheimer's disease (AD) sections (10 frontal, 10 occipital sections; 5-fold cross-validation) and validated on 5 independent external AD cases (5 frontal, 5 occipital). The model generated composite masks for vessel walls, vascular amyloid, and tissue compartments. We applied the trained model to 72 whole-slide images (36 frontal, 36 occipital) from 36 cases to extract vessel-level metrics including outer diameter, radial and tangential wall thickness, percent amyloid wall coverage, and circumferential involvement. Extracted features underwent within-subject paired analysis to identify regional morphometric shifts.

Results: The models demonstrated strong performance on internal cross-validation and independent external validation. Models' precision exceeded 90% across all segmentation targets. In paired within-subject analyses, the occipital cortex consistently showed greater vascular amyloid involvement than the frontal cortex. The proportion of CAA-positive vessels was higher in the occipital cortex (29.2% vs. 24.2%). Among amyloid-positive cortical vessels, the fraction with circumferential wall involvement was greater in the occipital region (46.4% vs. 36.9%). Consistent with these findings, the composite CAA severity index, integrating amyloid wall burden, circumferential involvement, and perivascular amyloid deposition, was 1.54-fold higher in the occipital cortex.

Conclusions: This automated framework quantifies spatial CAA vulnerability, detecting biologically coherent regional differences in amyloid distribution and vascular remodeling. Such scalable phenotyping supports hemorrhage-risk modeling and improves mechanistic and therapeutic studies of cerebrovascular responses to amyloid-targeting drugs.

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Quantitative Comparison of AT8 and pTau181 Immunohistochemical Staining in Human Brains with Alzheimer's Disease Neuropathologic Change

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Background: Identification of hyperphosphorylated Tau protein underlies the diagnosis and staging of Alzheimer's Disease (AD). The AT8 antibody is widely used in neuropathology to detect phosphorylated tau at serine 202 and threonine 205. In contrast, pTau181 reflects single epitope phosphorylation at threonine-181. pTau181 in cerebrospinal fluid and plasma levels may be detected early in the disease course, correlating with amyloid deposition and tau burden detected by PET imaging. In tissue-based immunohistochemistry, we can detect pTau181 with a phospho-tau antibody that may label tau species not routinely detected by AT8. Here, we compare AT8 and pTau181 immunohistochemical staining patterns to determine whether the detection of NFTs, plaques, and neurites differs and the implications for diagnosis and characterization of AD.

Methods: Formalin-fixed, paraffin-embedded sections of hippocampus and adjoining entorhinal cortex from 12 human brains were immunohistochemically (IHC) stained for pTau181 (clone BSB-176, BioSB) and AT8 (clone MN1020, Thermo Fisher Scientific) using automated IHC on the Ventana Benchmark Ultra and Dako Omnis, respectively.

Results: Quantitative analysis of paired hippocampal and entorhinal sections demonstrated similar numbers of neurofibrillary tangles (NFTs) and neuritic plaques between the two antibodies: AT8 and pTau181 with linear correlation. In contrast, the mean density of neuropil threads per mm^2 was markedly greater with pTau181 than AT8 (256.3 ± 92.8 vs 26.3 ± 24.4, $p = 8.2 \times 10^{-6}$), representing nearly a ten-fold increase with no correlation between the two antibodies.

Conclusions: The immunohistochemical stain for pTau181 detected a greater burden of diffuse and fine neuropil threads, reflecting a higher sensitivity for soluble Tau in neuropil. Both AT8 and pTau181 yield comparable detection of tangles and plaques, thus would not alter Braak stage or CERAD score. Incorporating pTau181 into neuropathologic evaluation may complement the mature fibrillar pathology highlighted by AT8 by improving the detection of early changes in axonal processes.

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Data-Driven Thresholds for Standardized Classification of Severe Alzheimer's Disease Neuropathology Using Digital Neuropathology

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Background: Alzheimer's disease neuropathological changes (ADNC) represent the gold standard for disease diagnosis and for evaluation of disease severity in an individual. Although useful, consensus-based semi-quantitative ADNC staging frameworks have critical limitations, particularly in advanced stages where pathological severity varies widely among individuals within a given diagnostic category. Further, some cases lacking cognitive impairment may be inappropriately categorized as having severe ADNC. In the current study, quantitative pathology metrics and alternative tissue sampling schemes were integrated with data about premortem cognitive status, in order to derive clinically informed neuropathologic diagnostic thresholds.

Methods: Specific goals of the current study were to generate data-driven, standardized diagnostic cut-points, with the most severe stages of ADNC having consistent implications: Braak neurofibrillary tangle (NFT) stage V cases being always impaired (mild cognitive impairment or worse) and Braak NFT stage VI cases being always demented. Utilizing whole-slide imaging and artificial intelligence-based image analysis, object-based (NFT counts) and pixel-based (phosphorylated tau [pTau] burden) quantifications were compared across neocortical regions in three subsamples of cases from the University of Kentucky Alzheimer's Disease Research Center autopsy cohort (n=329 cases included).

Results: Applying refined thresholds enabled reclassification of cases previously misaligned with their digitally determined appropriate status (17% of cases were reclassified). The use of commercially available software, standardized classifier architectures, and interoperable analysis pipelines facilitated scalable and reproducible digital quantification. Cross-institutional validation at University of Texas San Antonio, with the same algorithms applied in both research centers, confirmed near-perfect agreement of pathology counts, underscoring the feasibility of harmonized digital workflows for collaborative research and diagnostic purposes.

Conclusions: These findings support the integration of quantitative digital pathology into standard neuropathological protocols and provide a scalable model for future large, standardized, multi-site studies of ADNC severity. Comparing analytical platforms, pTau antibodies, and anatomical sampling strategies, an updated workflow demonstrated high reproducibility and consistent clinical-pathological correlations.

PLATFORM 5: Tumors: Nonglial

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Diagnostic Utility of c-MYC and SMARCA4 Immunohistochemistry in non-WNT/non-SHH Medulloblastomas

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Background: Medulloblastomas (MBs) are classified into four major molecular groups with characteristic prognostic and therapeutic implications. SHH- and WNT-activated MBs display distinct immunoprofiles, whereas group 3 and 4 MBs are distinguishable only through genomic and epigenomic analyses. MYC amplification is essentially confined to group 3, while SMARCA4 alterations are observed in both WNT-activated and group 3 MBs. Here, we investigate the utility of c-MYC and SMARCA4 immunohistochemistry in subtyping non-WNT/non-SHH MBs.

Methods: An institutional database search identified 32 non-WNT/non-SHH MBs with group designations based on DNA methylation profiling (DNAMP) and/or next generation sequencing. 15 SHH-activated and 5 WNT-activated MBs were included as controls. c-MYC (clone: Y69, incubation: 15 minutes, dilution 1:50) and SMARCA4/BRG1 (clone: EONCIR111A, incubation time: 60 minutes, dilution: 1:50) immunohistochemistry was performed on all cases. Moderate to strong nuclear c-MYC staining in >10% of tumor cells was considered positive.

Results: Out of 32 non-WNT/non-SHH MBs, 27 underwent DNAMP: 12 classified as group 3 and 15 as group 4, all but one with a confidence score >0.8. The remainder had molecular alterations suggestive of group 3 (n=2, gain of chromosome 8q including MYC, gain of 1q, loss of 16q) or 4 (n=3, SNCAIP duplication, CDK6 and MYCN amplifications). c-MYC immunostaining was detected in 8/14 (57%) group 3 MBs (including both cases with chromosome 8 gain), with 94.7% specificity, 57.1% sensitivity, 80% positive predictive value (PPV), and 86% negative predictive value. All group 4 and SHH-activated MBs were negative, while 2 WNT-activated MBs also showed c-MYC immunopositivity. The specificity and PPV of c-MYC IHC reached 100% among non-WNT/non-SHH MBs. None of the cases demonstrated loss of SMARCA4, including group 3 and WNT-activated MBs (n=2) with missense SMARCA4 mutations.

Conclusions: c-MYC immunohistochemistry exhibits high specificity and positive predictive value for group 3 MBs, underscoring its potential role as a useful diagnostic marker in non-WNT/non-SHH MBs.

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Proteomic analysis of medulloblastoma identifies MYC IHC as a powerful outcome predictor outperforming FISH and methylation-based subgrouping

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Background: While international consensus and the 2021 WHO classification recognize multiple molecular medulloblastoma subgroups, these are difficult to identify in clinical practice utilizing routine approaches. As a result, biology-driven risk stratification and therapy assignment for medulloblastoma remains a major clinical challenge.

Methods: We analyzed 56 formalin fixed paraffin embedded (FFPE) medulloblastoma samples by data independent acquisition mass spectrometry identifying a MYC proteome signature in therapy resistant Group 3 medulloblastoma. We validated MYC immunohistochemistry (IHC) prognostic and predictive value across two Group 3/4 medulloblastoma clinical cohorts (n=362) treated with standard therapies.

Results: After exclusion of WNT tumors, MYC IHC was an independent predictor of therapy resistance and death [HRs 23.6 and 3.23; 95% confidence interval (CI) 1.04-536.18 and 1.84-5.66; P =.047 and <.001]. Notably, only ~50% of the MYC IHC positive tumors harbored MYC amplification. Accordingly, cross-validated survival models incorporating MYC IHC outperformed current risk stratification schemes, including MYC amplification detection and DNA-methylation based subgrouping, and reclassified ~20% of patients into a more appropriate very high-risk category.

Conclusions: This study provides a high-resolution proteomic dataset that can be used as a reference for future biomarker discovery. Biology-driven clinical trials should consider MYC IHC status in their design. Integration of MYC IHC in classification algorithms for non-WNT tumors could be rapidly adopted on a global scale, independently of advanced but technically challenging molecular profiling techniques.

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Chromosomal Copy Number Profiling in Skull Base Chordoma: A Single-Institution Study of 20 Cases

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Background: Clinical risk stratification of skull base chordoma (SBC) has included deletions at 1p36 and homozygous loss of 9p21 (CDKN2A/B), as assessed by fluorescence in-situ hybridization. However, knowledge of chromosomal copy number variations (CNVs) in SBC remains limited.

Methods: We reviewed SBC cases diagnosed at our institution between 2013 and 2025, which had been characterized by chromosomal microarray analysis (CMA), and assessed patient outcomes based on clinical and radiographic follow-up.

Results: Twenty SBCs were identified (13 males, 7 females; median age 43 years, range 26-84), all in the clivus and corresponding to conventional chordoma. The most common CNVs included loss of chromosome 1p (n=10), including 1p36 (n=8); gain of 1q (n=9); loss of 3 (n=10); loss of 4 (n=6); gain of 7 (n=8); loss of 9 (n=11, 0 with CDKN2A/B homozygous deletion); loss of 10 (n=12); loss of 11 (n=5); loss of 13 (n=13); loss of 14 (n=12); loss of 17 (n=5); loss of 18 (n=7); and loss of 22 (n=7), including SMARCB1 (n=1). Nine patients underwent gross total resection (GTR) followed by radiation; 11 patients underwent subtotal resection (STR), 9 receiving subsequent adjuvant radiation. The median follow-up was 22 months. Two patients died from unrelated causes 9 and 32 months after diagnosis. Five patients developed recurrence (median time to recurrence, 17 months; range, 9-49), all having received STR and radiation. Among these, 3 had 1p36 loss, 3 had chromosome 9 loss without CDKN2A/B homozygous deletion, and 1 had SMARCB1 loss. Five patients with 1p36 loss did not develop recurrence (median follow-up, 23 months), including 4 with GTR and radiation, and 1 with STR only.

Conclusions: While chromosome 9 loss is common in SBC, CDKN2A/B homozygous deletion appears less frequent, and recurrences may occur even in its absence, particularly following STR.

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Optical Genome Mapping Identifies Structural Variants in Meningioma

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Background: Complex structural variants (CSVs) play an important role in the biology of meningiomas, one of the most common primary CNS tumors, including their progression to higher grade and recurrence. Conventional cytogenetic techniques and short-read NGS often fail to detect large or complex variants, limiting genomic characterization in clinical neuropathology. Optical Genome Mapping (OGM) enables high-resolution, long-range analysis of DNA, offering potential insights into genomic alterations relevant for neuropathologic evaluation. We aimed to assess the feasibility of OGM for detecting structural variants in meningiomas.

Methods: We analyzed fresh-frozen WHO grade 1 and grade 2 meningiomas samples using OGM following standard ultra–high molecular weight DNA isolation and de novo assembly pipeline (Bionano Genomics, Inc). Structural variants, including deletions, duplications, inversions, copy-number changes, and interchromosomal rearrangements were annotated relative to the GRCh38 reference genome. A whole-genome circos map was generated to visualize genomic architecture.

Results: The circos plot demonstrated a structurally complex genome feature. OGM revealed multiple large-scale structural alterations, prominently involving several chromosomes, including broad deletions, copy-number gains, and several intra and interchromosomal rearrangements. Specific alterations included deletion, duplication, translocation between chromosomes. Also, was observed novel fusion product and chromothripsis.

Conclusions: These findings illustrate the presence of structural complexity in a tumor lacking identifiable variants on standard clinical testing. This demonstrates the feasibility of OGM in characterizing large and CSVs, and genomic rearrangements not captured by conventional methods, emphasizing its potential as a complementary tool in molecular neuropathology and in the genomic assessment of meningioma tumors.

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Development of immunohistochemical surrogate stains for CNS WHO grade 2 copy number alterations in meningioma

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Background: Meningiomas are the most common primary intracranial tumors, and account for nearly 42% of all cases. While meningiomas have historically been graded based on histologic criteria alone, the 2021 World Health Organization (WHO) classification scheme introduced molecular grading criteria for the first time. Furthermore, the recent cIMPACT-NOW update 8 proposed an integrated molecular grading system for meningiomas that includes 1p/22q co-deletion as a new independent copy number alteration (CNA) criterion for a CNS WHO grade 2 designation. However, the implementation of complex molecular grading schema necessitates next-generation sequencing and bioinformatic analyses, which may not be feasible in routine clinical practice. Inspired by recent use of MTAP and P16 as immunohistochemical (IHC) surrogates for CDKN2A/B homozygous deletion in meningioma, we set out to develop and validate IHC surrogate markers for chromosome 1p/22q co-deletion.

Methods: Using multiplatform expression data from a cohort of 504 meningiomas with known CNAs, DNA methylation groups, and clinical outcome data, we identified a subset of proteins that were systematically and significantly enriched or suppressed in tumors with 1p/22q co-deletion and compared them to tumors without co-deletion. IHC staining for candidate proteins was optimized and validation was performed on a cohort of 30 meningioma cases (16 with and 14 without 1p/22q co-deletion on next-generation sequencing).

Results: IHC testing of candidate proteins identified markers with good sensitivity and specificity for 1p/22q co-deletion, including IGF2BP2 which showed 100% sensitivity and 71% specificity. Comparison of cases with differing IGF2BP2 expression also revealed distinct DNA methylation profiles between the two groups.

Conclusions: This study establishes robust and cost-effective candidate IHC markers for 1p/22q co-deletion in meningioma and more generally establishes a framework for the development of IHC surrogate stains for high-risk CNAs in cancer.

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Primary Central Nervous System GLI1-Altered Mesenchymal Tumors: Clinicopathological and Molecular Analysis of 8 Cases

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Background: GLI1-altered mesenchymal tumors represent an emerging molecularly defined entity, typically arising in extracranial sites. Primary central nervous system (CNS) examples are extremely rare and poorly characterized.

Methods: We retrospectively analyzed 8 primary CNS GLI1-altered mesenchymal tumors using clinical, histologic, immunohistochemical, FISH, DNA/RNA sequencing, and methylation profiling.

Results: Patients ranged from 4 to 62 years (median 38; M:F=1:1). Seven tumors were supratentorial, one infratentorial. Size ranged 1.8–7.2 cm (median 4.5 cm). All showed infiltrative growth: seven were spindle cell fascicular with arborizing capillaries, one was small blue round cell. Myxoid stroma was present in five cases, necrosis in three. All tumors were diffusely positive for GLI1 and CD56; four expressed S100, four showed focal SMA, and the small round cell component was TTF-1 positive. All were negative for AE1/AE3, SOX10, STAT6, and CD34. Seven cases showed GLI1 amplification with MDM2/CDK4 co-amplification, two with additional GLI1 fusions. One case harbored a novel ANO6::GLI1 fusion. Methylation profiling demonstrated a distinct methylation cluster. At median 18-month follow-up, six patients were disease-free and two had local recurrence.

Conclusions: Primary CNS GLI1-altered mesenchymal tumors represent a rare, distinct entity. GLI1 IHC and molecular testing are useful for diagnosis. DNA methylation profiling supports independent classification and precision clinical management.

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The Cost of Pituitary Neuroendocrine Tumor Classification: Experience with the Minneapolis Algorithm

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Background: The contemporary pituitary neuroendocrine tumor (PitNET) classification relies heavily on historically derived categories and expert consensus rather than explicitly testable, quantitative criteria, limiting reproducibility and falsifiability. Although the WHO does not endorse a single immunohistochemical (IHC) panel, WHO authors typically recommend performing all relevant IHC on every PitNET case, incurring potentially high costs and generating false positive and false negatives that cloud the classification. We previously generated an algorithm for PitNET characterization which reduces the number of IHC stains that are used in most cases.

Methods: In order to explore the impact of our algorithm on the cost of diagnosis, we collected serial PitNET cases resected within the Allina Health system, a nonprofit, integrated healthcare system based in Minneapolis, Minnesota. IHC stains were scored using the Allred system and were tabulated along with tumor size, patient age and sex, clinical parameters, and reference diagnosis. Cost estimates of IHC stains were based on the 2025 fee schedule. Hypothetical costs of IHC stains were based on the assumption that the WHO approach uses "all stains on all cases." This was estimated to include IHC stains for SF1, PIT1, TPIT, prolactin, growth hormone, TSH, ACTH, CAM5.2, LH, FSH, estrogen receptor, synaptophysin, and GATA3.

Results: 229 pituitary tumors (133M, 96F) were collected, including 225 pituitary adenomas and 4 samples of non-neoplastic anterior pituitary. Tumors ranged in size from 4-70 mm (mean 26.5 mm [SD 10.6, median 26]). Using the "all stains on all cases" approach, and assuming that this implied 13 IHC stains per case, estimated charges for the 229 tumors would have been $966,746.40 ($4,221.60 per case). Charges for the actual IHC performed amounted to $313,647.00 (averaging $1,369.64 per case).

Conclusions: Using the previously validated Minneapolis algorithm resulted in substantially smaller IHC resource use and charges when compared with the WHO recommendations.

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Age-associated clinicogenomic characteristics and therapeutic determinants in melanoma brain metastasis

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Background: The incidence of melanoma brain metastasis (MBM) rises with age. To evaluate how aging influences MBM mutational status, disease progression, and clinical outcomes, we analyzed clinicogenomic data in age-stratified patient cohorts.

Methods: Clinical history of MBM patients (n=207) treated at Johns Hopkins Hospital (JHH) from 2014-2024 were retrospectively reviewed. Next-generation sequencing (NGS) analysis available for 35 JHH patients were combined with sequencing data from 36 MBM cases in the MSK-MetTropism (MSK) cohort accessed on cBioPortal. Mutation frequencies and survival outcomes were analyzed.

Results: In the JHH cohort, females were diagnosed with both cutaneous melanoma and MBM at a significantly younger ages than males (52.4 vs 59.6, p=0.0037; 59.6 vs 64.3, p=0.0267). Older age at primary diagnosis correlated with shorter MBM-free survival, while patients < 60 experienced longer latency before MBM onset (log-rank p=0.0007). In a subset of patients with available driver mutation status (n=121), frequencies of BRAF (50.42%), NRAS (17.95%) and NF1 (5.13%) mutations were comparable to those reported in population-based studies. NF1-mutant patients exhibited the highest median age at diagnosis (74.5) and the shortest MBM-free survival (log-rank p=0.006). NGS analysis (n=71) revealed significantly higher tumor mutation burden in patients >60 (n=45; 23. vs 12.8, p=0.0217). Among BRAF-mutant patients, MAPK pathway alterations (i.e. EGFR, FGFR, NTRK) were enriched in patients >60, and were associated with significantly worse overall survival compared to patients < 60. In the JHH cohort, BRAF-mutant patients >60 demonstrated higher rates of MBM progression following initial MAPK-inhibitor therapy, and developed treatment resistance characterized by NGFR expression in clinical MBM samples detected by immunohistochemistry. NGFR-positive tumors were associated with worse survival.

Conclusions: MBM arising from different patient age groups may represents biologically distinct subtypes. Age-associated MAPK pathway alterations may underlie increased therapeutic resistance in older patients, highlighting the importance of molecular profiling in guiding personalized therapeutic management.

PLATFORM 6: Neurodegenerative: FTLD/Lewy body/CTE

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Focal microglial reactivity in the hippocampal CA2 subfield specific to a-synuclein pathology is implicated in Lewy body disease progression

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Background: Histopathologic staging models of intraneuronal a-synuclein pathology (n-asyn) in the human Lewy body disease (LBD) brain seldom evaluate regions with direct synaptic connectivity to model microglial processes. Here, we address this gap by testing the hypothesis that, within the well-defined synaptic connectivity of the intrahippocampal circuit, n-asyn is associated with microglial reactivity.

Methods: We selected a cohort of autopsy-confirmed LBD patients and minimal age-related co-pathologies (n=62), and a cohort of cognitively healthy patients with focal hippocampal tau accumulation with minimal amyloid plaques, termed primary age-related tauopathy (PART; n=12). We immunostained consecutive hippocampal sections for n-asyn and established markers of microglial reactivity, Iba1, HLA-DR, and CD68. With validated digital histology methods, we measured percent area occupied (%AO) of each marker in 6 hippocampal subfields to compare the %AO between cohorts, correlate the %AO of n-asyn with microglial reactivity, and use linear-mixed effects models to compare the %AO between subfields while co-varying for demographics. We also constructed groups of either n-asyn restricted to the cornu ammonis (CA) 2-3 subfields (Focal Subtype) or widespread n-asyn within additional subfields (Widespread Subtype) to model hypothesized n-asyn spread within the intrahippocampal circuit.

Results: LBD patients exhibited increased HLA-DR and CD68 in most hippocampal subfields compared with PART. In LBD patients, all microglial markers were highest in the CA2. Only CA2 n-asyn consistently correlated with HLA-DR and CD68 but not Iba1. Patients classified as Widespread Subtype had worse cognitive impairment and increased CA2 HLA-DR and CD68. Furthermore, CA2 HLA-DR and CD68 correlated with distal n-asyn only in subfields with retrograde connectivity.

Conclusions: Our data suggests a relatively specific microglial response to n-asyn compared with PART and implicates focal microglial reactivity in the CA2 with worse clinical outcomes and predominantly retrograde transmission of n-asyn in relatively pure LBD. These data suggest that microglial processes can help refine LBD histopathological staging.

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TDP-43 and tau seed amplification assays for LATE diagnosis and differentiation from AD

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Background: According to several large autopsy series, LATE neuropathologic change (LATE-NC) is present in 30-40% of elderly brains. LATE is difficult to differentiate from Alzheimer's disease (AD) because it shares similar amnestic symptoms with those of AD. Moreover, LATE-NC and AD neuropathologic change (ADNC) often co-exist in the same individual. Currently, the clinical diagnosis of LATE relies on the absence of positive AD biomarkers, as there are no robust, molecular-specific biomarkers available to detect LATE or distinguish LATE from AD, or comorbid LATE+AD.

Methods: To address this unmet need, we developed a novel TDP-43 seed amplification assay (SAA) using real-time quake-induced conversion (RT-QuIC) platform to differentiate LATE-NC and LATE-NC+ADNC (TDP-43 proteinopathy) from normal controls and ADNC (non-TDP-43 proteinopathy). Separately we developed a tau-SAA assay using engineered tau substrates to distinguish LATE-NC (non-tauopathy) from ADNC and LATE-NC+ADNC (tauopathy).

Results: Our novel TDP-43 SAA was capable of differentiating brain tissue and CSF from subjects with LATE-NC versus normal controls with outstanding performance (AUC=0.99 for brain tissue and AUC=0.91 for CSF). Complementary to TDP-43 SAA, our tau-SAA was able to differentiate brain tissue and CSF from subjects with LATE-NC from those with ADNC with excellent diagnostic performance as well (AUC=0.95 for brain tissue and AUC=0.82 for CSF). Tau-SAA can further distinguish brain tissue, and shows promise in distinguishing CSF, from subjects with LATE-NC from those with comorbid LATE-NC+ADNC (AUC=0.97 for brain tissues and AUC=0.73 for CSF samples).

Conclusions: To our knowledge, our CSF-based TDP-43 SAA RT-QuIC assay is the first to be developed and applied to LATE diagnosis and differentiation from AD and comorbid LATE+AD. A combination of TDP-43 SAA and tau-SAA may provide powerful tool for the clinical diagnosis of LATE and patient stratification for theragnostic development.

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Opposing Nuclear Body Remodeling in TDP-43 Pathology: SUMO1-Associated Protection and PML-Linked Vulnerability

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Background: TDP-43 nuclear depletion is a hallmark of frontotemporal dementia (FTD), yet the cellular mechanisms that determine neuronal vulnerability remain poorly understood. Nuclear bodies undergo extensive remodeling in neurodegenerative disease, but their functional relationship to TDP-43 pathology has not been systematically examined at single-cell resolution.

Methods: We performed single-cell spatial analysis of 18,742 neurons from the Anterior cingulate cortex across 29 subjects (10 sporadic FTD, 10 C9orf72 FTD, 9 controls). Each neuron was annotated for five binary phenotypes: SUMO1 granules, SUMO1 puncta, TDP-43 nuclear depletion, PML nuclear membrane contacts and SUMO-dominant PML bodies. We defined local cellular neighborhoods using an 8 μm radius and tested phenotype co-occurrence using MH ORs stratified by subject to control for within-subject correlation.

Results: SUMO1 nucleolar remodeling strongly protected against TDP-43 nuclear depletion (OR=0.15, 95% CI: 0.11–0.22, $p < 0.001$), with 80% subject consistency. Both SUMO1 granules (OR=0.21) and SUMO1 puncta (OR=0.09) demonstrated independent protective effects. In contrast, PML nuclear membrane contacts conferred high risk of TDP-43 depletion (OR=4.58, 95% CI: 3.66–5.74, $p < 0.001$), as did SUMO-dominant PML bodies (OR=5.16). Temporal trajectory analysis revealed distinct patterns: SUMO1 prevalence peaked in disease neurons with intact nuclear TDP-43 (19.1% vs 3.5% in controls) but decreased in TDP-43-depleted neurons (8.4%), consistent with a protective response. Conversely, PML nuclear membrane contacts showed progressive enrichment from controls (0.1%) through disease TDP-intact neurons (3.2%) to TDP-depleted neurons (7.8%), indicating an upstream pathological change. Associations were robust to confounding adjustment, C9orf72 mutation status, and spatial parameter variation.

Conclusions: We identify SUMO1 nucleolar remodeling as a protective cellular response and PML nuclear membrane association as an upstream risk factor for TDP-43 nuclear depletion in FTD. These findings expand the limited repertoire of known pre-depletion pathological changes and suggest that nuclear body composition determines cellular vulnerability to TDP-43 proteinopathy.

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The GRN gene variant rs5848 linked to TDP-43 pathology: lessons from epidemiology

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Background: The granulin gene (GRN) and its encoded protein, progranulin (PGRN), play key roles in inflammation and cell proliferation. Decreased PGRN expression promotes TDP-43 pathology in LATE-NC and FTLD-TDP. More specifically, the single-nucleotide variant rs5848 (T allele) in the 3'UTR of GRN is associated with lower PGRN expression, and is a risk factor for LATE-NC and FTD/FTLD age-of-onset. PGRN is also a mitogen, implicated in neoplasia. Clinical therapies have been proposed to augment PGRN expression and perhaps to counteract the influence of the rs5848 risk allele.

Methods: Here we examined the epidemiology of GRN/rs5848 in two ways: first, we evaluated rs5848 allele frequencies across human populations; and second, we tested how the rs5848 variant is associated with risk for LATE-NC and hippocampal sclerosis pathologies in comparison with cancer in the same cohorts.

Results: The rs5848 allele frequencies differed based on ancestry: the risk allele was substantially more frequent in persons of African ancestry (~75%) than in persons of European ancestry (~25%). This variability in the genetic architecture may indicate differing GRN gene expression regulatory mechanisms across human populations.Next, we leveraged the AD Genomics Consortium (ADGC; n=8121) and the AD Sequencing Project (ADSP; n=3231) data sets, with available cancer history, genetic information, and neuropathology data. The GRN rs5848 T allele was associated with higher odds of LATE-NC and hippocampal sclerosis ($p< 0.001$). In contrast, the same allele was associated with lower odds of cancer ($p=0.012$). Parallel analyses of other established dementia risk variants (in TMEM106B, APOE, and BIN1) showed no significant associations with cancer, implying that the GRN-related associations were not only due to skewed recruitment and "survivor bias".

Conclusions: Given the potential for GRN/PGRN-targeting therapeutic strategies, there are clinical implications to the findings of a specific disease-associated GRN allele with ethnoracial allele frequency differences and opposite apparent impacts on dementia-related pathology and cancer risk.

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ANXA11 as additional pathological marker for TDP-43 classification in rare genetic, mixed and unclassifiable TDP-43 pathology

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Background: Frontotemporal lobar degeneration with TDP-43 inclusions (FTLD-TDP) forms a wide range of neurodegenerative diseases, classified into different subtypes based on morphology and distribution of TDP-43 inclusions. Recently co-aggregation of ANXA11 with TDP-43 has been discovered in FTLD-TDP subtype C, clinically marked by temporal atrophy and semantic primary progressive aphasia. ANXA11 expression has not yet been described in the full spectrum of FTLD-TDP, specifically not in rare genetic, mixed and unclassifiable FTLD-TDP. The aim of this study was to further characterise ANXA11 pathology herein.

Methods: We immunohistochemically stained frontal, temporal and hippocampal sections of a post-mortem case series of FTLD-TDP with underlying mutations in TUBA4A (n=2), TARDBP (n=3), as well as rare subtype C (n=2) and mixed (A+B n=6, B+C n=2) pathology, and related findings to in vivo imaging. We also included sporadic FTLD-TDP C (n=2) as positive controls, GRN with subtype A pathology (n=2) and C9orf72 with subtype B pathology (n=2), and non-demented controls (n=2) as negative controls. Additionally, double-label immunofluorescence staining for ANXA11 and TDP-43 was conducted. Ordinal ratings were given to semi-quantify ANXA11 expression and neurodegeneration. Imaging reports and MRI/CT images were also obtained and atrophy rated.

Results: Interestingly, C9orf72 with subtype C and mixed B+C pathology had most ANXA11, co-aggregating with TDP-43 and resembling sporadic FTLD-TDP C. Other rare mixed and unclassifiable FTLD-TDP showed little to no ANXA11 inclusions. Furthermore, ANXA11 expression correlated positively with temporal atrophy post-mortem (ρ=0.598; p=0.008) and in vivo (ρ=0.543; p=0.0489).

Conclusions: Potentially, ANXA11 could distinguish a distinct subgroup of C9orf72 phenotypically similar to sporadic FTLD-TDP C, enabling a better characterisation of the heterogenous spectrum of C9orf72. The correlation between ANXA11 pathology and temporal atrophy, could imply a potential of temporal atrophy during life as predictor of ANXA11 pathology, particularly for C9orf72. Such an additional biomarker could be very useful in untangling the varied C9orf72 phenotypes.

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Divergent Endolysosomal Autophagy Signatures in Chronic Traumatic Encephalopathy and Alzheimer Disease

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Background: Endolysosomal–autophagy (ELA) dysfunction contributes to pathologic protein accumulation across neurodegenerative diseases, but comparative ELA profiles in chronic traumatic encephalopathy (CTE) versus Alzheimer disease (AD) remain unclear.

Methods: Dorsolateral prefrontal cortices from 236 donors (CTE n=80, AD n=71, age-matched controls n=85) were analyzed. ELA (cathepsin D (CTSD), progranulin), inflammatory (Iba1, CD68), synaptic (alpha-synuclein, PSD-95), and pathologic (phospho-tau isoforms pTau181, pTau231, pTau202, pTau396; Aβ40, Aβ42) proteins were quantified by Meso Scale Discovery immunoassay or immunohistochemistry. ELA markers cathepsin F (CTSF), ubiquilin-2, glucosylceramidase, BCL2-associated athanogene-3 (BAG3), TANK-binding kinase-1 (TBK1), valosin-containing protein (VCP), and sequestosome-1 were measured using SomaScan proteomics. Group differences were assessed with age-adjusted ANCOVA and Bonferroni correction ($p< 0.05$). ELA protein associations were evaluated using Spearman correlations and multivariable linear regression adjusted for age and postmortem interval. Multiple testing was controlled using Benjamini-Hochberg false discovery rate (FDR; $q< 0.05$).

Results: CTE: Compared with controls, CTE showed widespread ELA downregulation. Progranulin ($p=2.00\times10^{-4}$), CTSD ($p=0.010$), CTSF ($p=0.007$), BAG3 ($p=0.007$), and TBK1 ($p=0.004$) decreased; only VCP increased ($p=0.003$). FDR-significant correlations with alpha-synuclein (synaptic density marker) included positive VCP-alpha-synuclein ($\beta=0.640$, $q=2.02\times10^{-8}$) and negative CTSF-alpha-synuclein ($\beta=-0.455$, $q=0.003$). AD: Conversely, AD demonstrated increased lysosomal enzymes CTSD ($p=0.001$), CTSF ($p=0.015$), and glucosylceramidase ($p=0.002$), alongside decreased cargo adaptors ubiquilin-2 ($p=0.004$) and sequestosome-1 ($p=0.031$). FDR-significant synaptic associations included positive ubiquilin-2-alpha-synuclein ($\beta=0.550$, $q=3.68\times10^{-4}$) and glucosylceramidase-alpha-synuclein ($\beta=0.513$, $q=3.68\times10^{-4}$), and negative CTSF-alpha-synuclein ($\beta=-0.537$, $q=3.68\times10^{-4}$). CTSD positively associated with pTau181 ($\beta=0.346$, $q=0.024$).

Conclusions: CTE and AD exhibit divergent ELA profiles. CTE shows generalized ELA pathway suppression, while AD demonstrates lysosomal enzyme upregulation with impaired cargo recognition. Robust associations between specific ELA components and alpha-synuclein

suggest disease-specific relationships between ELA function and synaptic integrity, with synaptic density linked to VCP in CTE and cargo adaptors (ubiquilin-2, glucosylceramidase) in AD. These opposing ELA signatures suggest disease-specific pathogenic mechanisms and therapeutic targets despite overlapping tau pathology.

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Diagnosis of Low Stage Chronic Traumatic Encephalopathy is Missed without Extensive Sampling in Half of Community Cases

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Background: Chronic traumatic encephalopathy (CTE) is a neuropathologically defined tauopathy associated with repetitive head injury. Although extensively studied in contact-sport athletes, CTE remains less characterized in community populations with heterogeneous traumatic brain injury (TBI) exposures. Here we report on how sampling affects the ability to diagnose CTE at the University of Washington.

Methods: Brains from the Pacific Northwest Brain Donor Network (BDN), a repository including veterans and civilians with and without TBI, were evaluated. CTE assessment followed the 2021 NINDS–NIBIB consensus criteria using AT8 immunohistochemistry. Recommended cortical regions (middle frontal gyrus, superior and middle temporal gyrus and inferior parietal lobule) were sampled bilaterally. Extended bilateral sampling of cortical sulci included nine additional regions. CTE was staged using the NINDS–NIBIB low/high tier system.

Results: 45 cases met criteria for CTE, all in men: 26 low-stage and 19 high-stage. About half (46%) of low-stage cases were identified on unilateral sampling of the neocortex, while an additional nine were found with bilateral evaluation (reaching 81%). The remaining cases required extended sampling block evaluation. Of the 19 high-stage cases, 79% were diagnosed with unilateral sampling, no additional cases were diagnosed with bilateral assessment, and the remaining four cases required additional sampling. Of the nine CTE cases that required additional sampling, five were diagnosed in the anterior temporal lobe. Across all low-stage cases and evaluation of all blocks, the right superior frontal gyrus had the highest number of pathognomonic lesions (9). In high-stage cases, the right parietal lobe had the highest number of lesions (12).

Conclusions: Unilateral sampling according to consensus criteria fails to detect a substantial proportion of low-stage CTE. Bilateral evaluation increases diagnosis, and additional evaluation of the anterior temporal lobe and superior frontal gyrus may be most useful to evaluate for further analysis. However, the clinical associations of a diagnosis of low-stage CTE are unknown.

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Neuropathological Characterization of Brain Donors with a History of Military Parachute Service

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Background: Military paratroopers are exposed to repetitive vertical deceleration forces during jump operations. The inferior brain surfaces, including the orbitofrontal regions, hypothalamus, mammillary bodies, medial temporal structures, and inferior cerebellum, may be anatomically vulnerable to such forces. The chronic neuropathological effects of repetitive parachute landing impacts remain largely uncharacterized.

Methods: Nine male brain donors (ages 24–57) whose military service histories included parachute operations were examined using H&E and immunohistochemistry for GFAP, IBA1, APP, phosphorylated tau, and amyloid-beta peptide. Regions along the base of the brain were compared against dorsal comparison regions (e.g., occipital cortex, cingulate gyrus, parietal cortex, superior/middle temporal gyri, superior cerebellum) using semiquantitative scoring.

Results: GFAP immunohistochemistry demonstrated increased astrogliosis in subpial, perivascular, and/or subcortical white matter distributions in one or more studied regions along the base of the brain relative to comparison regions in all cases. This was most prevalent in samples from the orbitofrontal region and the hypothalamus/mamillary bodies. Inferior cerebellar samples demonstrated increased astrogliosis relative to superior samples in a subset of cases, including a case whose inferior cerebellum sample additionally demonstrated multiple discrete foci of healed cerebellar cortical loss and gliosis. IBA1 labeling was without a consistent regional pattern. One case met diagnostic criteria for chronic traumatic encephalopathy. Notably, donors in this cohort carry complex/overlapping exposure histories, including blast injury and contact sports.

Conclusions: This case series represents an initial effort to characterize neuropathological findings in military paratroopers. Most notably, we observed increased astrogliosis in several patterns along the bases of the brain relative to comparison regions, indicating that these regions may be susceptible to injury from the dynamic forces related to parachuting. However, the contribution of concurrent exposures remains to be determined. The opinions expressed herein do not necessarily reflect official policies/positions of USU, the Walter Reed National Military Medical Center, or the DoW.

PLATFORM 7: Epilepsy, Methodologies

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Beyond Neuronal Loss: Spatial Transcriptomics Defines Molecularly Distinct Forms of Hippocampal Sclerosis in Mesial Temporal Lobe Epilepsy

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Background: Mesial temporal lobe epilepsy with hippocampal sclerosis (MTLE-HS) is the most common pathology encountered in drug-resistant adult epilepsy surgery. Current neuropathologic classification systems, including the International League Against Epilepsy (ILAE) subtypes, are based on patterns of subfield-specific neuronal loss and gliosis. While diagnostically robust, these histologic frameworks do not fully explain the marked clinical variability, differential surgical outcomes, and presumed biological heterogeneity observed across patients. Increasing evidence suggests that epileptogenicity in MTLE-HS reflects complex circuit-level dysfunction involving neuronal, glial, and synaptic remodeling processes.

Methods: Formalin-fixed paraffin-embedded (FFPE) hippocampal tissue from surgically resected en bloc specimens was retrospectively selected from a cohort of drug-resistant MTLE-HS cases. RNA quality was confirmed using DV200 metrics (>65%). Five epilepsy cases representing ILAE Type 1 total sclerosis, Type 1 classical sclerosis, Type 2 (CA1-predominant), and gliosis-only patterns were analyzed alongside three age-matched non-neurologic controls. Spatial transcriptomic profiling was performed using the 10x Genomics Visium platform and IHC for correlation. Differential gene expression, dimensionality reduction (UMAP), and pathway analyses were conducted to assess molecular heterogeneity and cell-type specificity.

Results: Spatial transcriptomics revealed marked inter-case molecular heterogeneity across MTLE-HS specimens. Differentially expressed gene (DEG) burden varied substantially between cases, with limited overlap, arguing against a unified transcriptional disease program. Transcriptomic alterations demonstrated strong cell-type and compartment specificity, involving neuronal activity-related genes (FOSB, JUNB), astroglial markers (AQP4), extracellular matrix remodeling (TNC), inhibitory circuitry components (GAD1, GAD2), and postsynaptic density-associated genes (DDN, DLGAP3).

Conclusions: MTLE-HS represents a biologically heterogeneous condition characterized by distinct spatially organized molecular programs rather than a single disease continuum. ILAE HS subtypes correspond to divergent transcriptomic states, supporting a network-based model of epileptogenic dysfunction. Integration of neuropathology with spatially resolved transcriptomics provides a powerful framework for defining molecular subtypes of human MTLE-HS and for advancing mechanism-informed disease stratification.

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Automated Identification of Tissue Compartments in IHC-Stained Whole Slide Images for Neuropathologic Studies

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Background: Quantitative assessment of neuropathologic features in routine practice remains largely semi-quantitative. In research settings, artificial intelligence (AI) enables scalable and reproducible analysis of disease-associated lesions in whole slide images (WSIs). Although most AI-based neuropathology studies rely on hematoxylin and eosin (H&E)–stained slides, immunohistochemistry (IHC) offers greater specificity for identifying pathologically relevant structures such as neurofibrillary tangles and amyloid plaques. However, automated localization of IHC-detected pathology within anatomically meaningful tissue compartments remains limited.

Methods: We developed a deep learning pipeline for automated segmentation of major tissue compartments in IHC-stained WSIs, including gray matter, white matter, superficial cortex, leptomeninges, and common artifacts. A SegFormer semantic segmentation model was initially trained on H&E WSIs (N=62) from the University of Kentucky and subsequently fine-tuned using annotated IHC WSIs from Emory University (tau, amyloid-β, α-synuclein, TDP-43; N=40). A WSI inference workflow generated continuous compartment masks through polygon stitching and spatial aggregation. Cases included Alzheimer disease (AD), mixed AD, non-AD neurodegenerative disorders, and controls. WSIs were obtained from neocortical regions: temporal cortex, occipital cortex, and frontal cortex.

Results: Across multiple magnifications (1–20x, Aperio SVS), the model achieved high segmentation accuracy for gray and white matter (Dice ≈0.94), with consistent performance across additional compartments. Transfer learning to IHC yielded performance comparable to the H&E-trained model, supporting cross-modality generalizability. Whole-slide inference was completed in under one minute per slide on an NVIDIA L40S GPU.

Conclusions: This framework enables automated, anatomically informed localization of IHC-detected neuropathology at the whole-slide level. Ongoing work includes inference-only evaluation on IHC cohorts from additional institutions to assess cross-site generalizability. This approach is designed to support future large-scale studies integrating compartment-specific pathology with AI-based lesion detection and associated clinical and neuropathologic variables.

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AI-Driven Spatial Characterization of Neuropathology in Whole Human Brain: Addressing the Scalability Gap

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Background: Whole-brain cellular-resolution histology offers neuropathological insights not achievable with traditional regional sampling. However, the petabyte-scale of these multi-marker image datasets makes manual detection and interpretation of brain-wide histopathological features impractical. Existing AI-based approaches are typically limited to smaller datasets, restricted brain regions, or single-marker analyses. To address this scalability gap, we developed a computational framework that leverages small sets of manually-defined features and scales them for automated whole-brain analysis.

Methods: Twenty-micron–thick stained sections from two human ischemic brains were digitized at 0.5 μm/pixel and aligned to ex vivo MRI. Whole-slide images were partitioned into non-overlapping tiles, small sets of which (≈10 representative tiles) were manually annotated as feature-specific ground-truth. These were mapped into a 768-dimensional latent space using a vision transformer (ViT) architecture trained for image representation learning. Similarity between each tile and ground-truth tiles was quantified using cosine distance, generating continuous probabilistic similarity scores. Whole-brain spatial feature maps were produced by adjusting similarity thresholds, iteratively refined through comparison with manually curated maps on selected sections. For intra-tile quantification, trained pixel-level computer vision models were applied.

Results: Our AI tool enabled brain-wide tile-level mapping of multiple ischemia-related features, including astrocyte density and morphology (GFAP), microglial/macrophage density (CD68), blood–brain barrier disruption (fibrinogen), hypoxia (HIF1α), axonal injury (APP), and axonal regeneration (GAP43). This multi-parametric digital brain volume integrated high-resolution histopathology with MRI-defined infarct regions. Key findings included: (a) dense macrophage infiltration and fibrinogen extravasation in the necrotic core, (b) reactive astrogliosis and microglial activation in the peri-infarct region, (c) overlapping zones of axonal injury and regeneration surrounding the infarct core and (d) global hypoxic stress.

Conclusions: This AI-driven, weakly-supervised framework addresses the scalability barrier in

whole-brain histology analysis by enabling rapid, objective, high-resolution, brain-wide characterization of complex pathological features from minimally annotated training data.

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State-Level Legal and Infrastructure Barriers to Research Brain Donation in the United States

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Background: Brain donation is foundational to advancing gold-standard diagnostic neuropathology for neurodegenerative diseases. In the United States, legal access to brain donation for research varies dramatically by state. We conducted a national legal and infrastructure analysis to evaluate state-level restrictions and identify structural barriers that prevent donor intent from translating into brain recovery.

Methods: Through statutory review of laws in 50 states and the District of Columbia, we developed the Brain Donation Restrictiveness Index (BDRI), a structured scoring system reflecting authorization-layer variables (donor autonomy, familial veto structures, modernization of Uniform Anatomical Gift Act language) and execution-layer variables (recovery authority and medical examiner interaction). BDRI scores range from 1 (least restrictive) to 11 (most restrictive). In parallel, we defined Infrastructure Modifiers to characterize downstream recovery capacity, including medicolegal system structure (medical examiner versus coroner models) and Organ Procurement Organization density. These were operationalized as state-level structural variables and analyzed descriptively, independent of the BDRI scoring system.

Results: Two states (Texas and New Mexico) demonstrated minimal statutory restriction (BDRI = 1). Most states clustered in moderate tiers, while a smaller subset exhibited stacked structural constraints. New York was the most restrictive jurisdiction (BDRI = 11), where statutory design subordinates documented intent in ways that may impede recovery.

Conclusions: Across jurisdictions, donation failure most commonly reflects misalignment between donor intent, statutory authorization frameworks, and recovery infrastructure rather than donor unwillingness. Most states operate under versions of the Uniform Anatomical Gift Act, which permits individuals to document anatomical gift intent. However, research brain donation in practice commonly depends on next-of-kin consent and institutional coordination not explicitly operationalized in statute. When individuals or families seek to contribute to research, preventable statutory and operational barriers undermine that intent. Reducing structural friction strengthens methodological rigor, improves representativeness in autopsy cohorts, and advances neuropathologic discovery, goals shared by donors and scientists alike.

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Promise of Artificial Intelligence in Interpreting Neuropathology NGS Data: Addressing HIPAA and Institutional Compliance Constraints

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Background: Next-generation sequencing (NGS) has become integral to neuropathology diagnostics but has increased interpretive complexity and workload. Many CNS tumors exhibit characteristic molecular signatures that may be amenable to computational interpretation. Evaluation of artificial intelligence (AI) tools in clinical environments requires use of institutionally approved platforms to ensure compliance with HIPAA and data governance policies.

Methods: Neuropathology cases requiring molecular interpretation were retrospectively reviewed. Clinical context and imaging findings were extracted from neuropathology reports in an unstructured, natural language format and combined with molecular findings from a large NGS panel (UTSW 1,500 gene DNA/RNA panel). Patient identifiers were excluded. Methylation profiling results (NCI/NIH) were used to confirm the final diagnosis against which the AI output was evaluated in challenging cases. Cases were stratified into two categories: (1) diagnostically straightforward tumors with canonical molecular profiles (n=20) and (2) diagnostically challenging tumors requiring additional studies such as methylation profiling (n=17). The data was entered into an implementation of ChatGPT version 5.2 running in an institutionally approved, controlled environment.

Results: Among the straightforward cases (n=20), AI-generated interpretations, based solely on the provided clinical context and NGS molecular findings, were concordant with the final integrated diagnosis in 100% of cases. Among the diagnostically challenging cases (n=17), concordance was 76.5%, reflecting the biological and interpretive complexity of these tumors and the continued need for expert neuropathologic evaluation and advanced ancillary testing. AI assistance was most effective when well-defined genomic signatures were present.

Conclusions: In this proof-of-principle study, ChatGPT version 5.2 running in an institutionally approved, controlled environment showed strong potential to support neuropathology diagnostics for cases with canonical molecular profiles, and complex cases requiring integration of histology and large amounts of data from NGS gene panels. Use of institutionally approved AI environments enable compliant evaluation and represents a critical step toward responsible clinical integration of AI-assisted workflows.

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From Feasibility to Validation: Implementing 2021 WHO CNS Tumor Whole Genome Methylation Profiling in a Resource-Limited Academic Center

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Background: DNA methylation profiling is increasingly used for central nervous system (CNS) tumor classification, but implementation remains difficult in diagnostic laboratories, especially those with limited bioinformatic support. We describe the feasibility, development, and validation of a locally maintained DNA methylation profiling framework and classifier designed for clinical adaptability at The University of Kansas Health System (TUKHS).

Methods: Methylation profiling was conducted using the Illumina MethylationEPIC v2.0 array. Fifty CNS tumor specimens with existing reference classifications were used for analytical validation. Results were compared across two established external classifiers and a newly developed local classifier trained with a random forest model implemented via the ranger package to enhance computational efficiency and feature selection. Supporting pipelines enabled copy number profiling, dimensionality reduction, and MGMT promoter methylation prediction. Deployment was facilitated through a customizable Shiny-based user interface.

Results: The local classifier demonstrated strong concordance with reference classifications and with existing tools at both the methylation class and class-family levels. Performance assessments across decreasing DNA input and tumor purity identified key parameters influencing clinical reliability. Repeatability testing and evaluation of datasets from two external centers further supported the classifier's robustness. Copy number and MGMT promoter analyses showed high agreement with expected profiles.

Conclusions: A locally maintained methylation profiling framework can deliver reliable CNS tumor classification while offering flexibility, integration, and adaptability within diagnostic workflows. This approach supports technical validation of emerging methylation classes and facilitates alignment with evolving WHO CNS tumor classification, even in settings with limited bioinformatic infrastructure.

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Improved Performance of Clinically Applicable Deep Learning for CNS Tumor Classification from Histopathology

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Background: We have recently demonstrated that DNA methylation and gene expression can be inferred from whole slide H&E images (WSIs) and integrated into a deep learning framework for CNS tumor classification. Here, we perform a focused evaluation of model performance within the meningioma and glioma families to better characterize tumor type-specific accuracy in routine diagnostic settings.

Methods: We applied our previously developed hierarchical deep learning framework integrating image-derived representations with inferred DNA methylation, inferred RNA expression, and patient demographics. To improve robustness and mitigate tumor-class variability, the training cohort was expanded, and final predictions were generated by ensembling outputs from five distinct foundation models within our framework. To address prior variability observed in glioma classification and the absence of subgroup stratification within meningioma, the classification hierarchy was refined to reduce subclass splitting within glioma and enhance clinically meaningful subgroup discrimination in meningioma. Model performance was evaluated using a fixed moderate-confidence threshold (0.5) and classifier-specific high-confidence thresholds optimized using the Youden index. Prediction and balanced accuracies were calculated for cases meeting confidence thresholds using top-1 and top-2 outputs.

Results: At internal validation, using a moderate-confidence threshold, the model generated class-level predictions for 84% (3,770/4,484) of glioma, achieving 89% top-1 and 96% top-2 accuracy. Using high-confidence thresholds, coverage (the proportion of samples that passed the threshold) was 62% (2,784/4,484) but performance improved to 95% top-1 and 98% top-2 accuracy. For meningioma, moderate-confidence predictions for the benign, intermediate and malignant groups were generated for 94% (1,984/2,089) of cases, achieving 85% top-1 and 98% top-2 accuracy. Inferred MKI67 levels (a surrogate for Ki-67 proliferation index) improved diagnostic accuracies. At high-confidence thresholds, coverage was 63% (1,331/2,089), with improved performance of 94% top-1 and 99% top-2 accuracies.

Conclusions: Expansion of the training cohort, ensemble modeling, and hierarchical refinement improved glioma and meningioma classification, supporting clinical integration of molecular inference–based AI.

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Fresh Ex Vivo Brain Tumor Imaging Using Two-Photon Microscopy

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Background: Intraoperative frozen section diagnosis is prone to ice crystal artifacts and subjective interpretation. This study aims to investigate the feasibility of using two-photon inverted microscopy for direct imaging of fresh ex vivo brain tissue and imaging of labeling fluorescently-stained cells, laying the groundwork for rapid intraoperative pathological diagnosis and microscopic imaging analysis.

Methods: 30 fresh ex vivo specimens of brain tumors and vascular malformations were collected (8 lymphomas, 15 gliomas, 1 meningioma, 5 metastases, and 1 vascular malformation). Fresh tissues and their corresponding cellular smears were first scanned directly with two-photon microscopy to acquire structural images of collagen and necrosis. Subsequently, the specimens and smears were stained for 20 seconds with acridine orange or rhodamine + Hoechst fluorescent dyes, and rescanned to validate whether the fluorescent signals corresponded to cell nuclei. The scanning area was < 2 mm × 2 mm, with imaging time < 5 minutes. Finally, the frozen sections of the identical tissue (adjacent side) were used for H&E staining as controls for fresh tissues, while smears were stained with H&E after scanning as controls for cellular smears.

Results: Two-photon imaging clearly revealed collagen and necrotic areas in fresh tissues. Fluorescent labeling scans demonstrated that fluorescent signals corresponded with cell nuclei locations in H&E staining. Notably, acridine orange displayed nuclear structures only in a single channel, whereas rhodamine + Hoechst visualized nuclear structures and Cytoplasm in both channels.

Conclusions: This study enables rapid acquisition of high-quality, ice crystal-free images from fresh ex vivo brain tumors and allows identification of nuclear structures through fluorescence labeling. The acquired images can be further utilized for virtual H&E and special staining, as well as automated algorithmic analysis (including quantification of nuclear size, tumor boundaries, and collagen density), providing crucial support for overcoming frozen section limitations and advancing the development of intraoperative handheld microscopy devices.

PLATFORM 8: Demyelinating and Inflammatory, Developmental/Pediatric, Peripheral Nerve/Muscle, Ophthalmic Pathology

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Microglial-amplified inhibitory synaptopathy: convergent neuropathology in GlyR, GAD65, Kelch11, and NIF autoimmunity

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Background: Autoimmune encephalitis encompasses a heterogeneous group of antibody-associated CNS disorders targeting synaptic, enzymatic, and intracellular neuronal antigens. Despite diverse clinical phenotypes and immune triggers, emerging evidence suggests overlapping neuropathologic and circuit-level mechanisms. We performed comparative neuropathologic analysis of four antibody-associated syndromes characterized by prominent brainstem, limbic, or cerebellar dysfunction: glycine receptor (GlyR) antibody-associated progressive encephalomyelitis with rigidity and myoclonus (PERM), GAD65-associated autoimmune epilepsy/encephalitis, Kelch11-associated rhombencephalitis, and NIF autoantibody-associated cerebellar syndrome.

Methods: Postmortem neuropathologic evaluation was conducted using hematoxylin-eosin staining, myelin stains, and immunohistochemistry for CD163, Iba1, GFAP, GAD65/67, GlyR, CD3, and CD20 across cortical, subcortical, brainstem, cerebellar, and spinal cord regions. Immunofluorescence assessed microglial activation and inhibitory neuronal markers. Molecular and spatial transcriptomic findings were incorporated where available. Comparative analysis focused on patterns of neuronal injury, inflammatory cell populations, and inhibitory circuit integrity.

Results: Across all cases, prominent microglial activation was demonstrated, frequently with CD163/Iba1-positive cells and perineuronal clustering. Structural neuronal preservation was present despite severe clinical dysfunction, with limited necrosis or classic neurodegenerative pathology. Selective inhibitory neuron abnormalities were observed, including reduced GlyR and GAD65 neuronal signals, regional Purkinje cell vulnerability, and transcriptomic suppression of GABAergic pathways accompanied by relative glutamatergic and stress-response upregulation. Immune effector profiles varied by antigen class: GlyR-associated PERM demonstrated innate immune-dominant pattern with minimal lymphocytes whereas Kelch11 encephalitis showed T-cell predominance. The NIF autoantibody case reinforced the association between cerebellar dysfunction and microglial-dominant inflammation.

Conclusions: Distinct antibody-mediated CNS disorders converge on a common pathophysiologic axis characterized by microglial-dominant neuroinflammation and selective disruption of inhibitory circuitry. These findings support a model of microglial-amplified inhibitory synaptopathy, in which functional synaptic and receptor dysfunction underlies profound neurologic impairment even in the absence of extensive neuronal loss. This framework provides a unifying biological perspective across autoimmune encephalitis phenotypes and

suggests potential therapeutic relevance for targeting antibody-mediated and innate immune mechanisms.

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Microglia and G-protein coupled receptors in the pathogenesis of periventricular heterotopia

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Background: The pathogenesis of many malformations of cortical development, including periventricular heterotopia (PH), is incompletely understood. Prior studies in our lab identified dysregulated G protein-coupled receptor (GPCR) signaling in associated with TMEM161B signaling.

Methods: A combination of in vivo and in vitro assays were performed in mice after approval by the institutional IACUC. Immunofluorescent and immunohistochemistry was performed on human tissue after approval by the institutional IRB.

Results: We subsequently have explored GPCR signaling during brain development and identified GNA12 activation during embryonic development specifically resulted in PH. Transcriptomic analysis revealed upregulation of immune-response genes and accumulation of activated microglia adjacent to heterotopic neurons. Furthermore, overexpression of CSF1, a microglial guidance molecule, induced similar phenotypes, implicating a role for microglial in PH formation. Finally, we identified the accumulation of microglia in the area around heterotopia in several cases of PH identified in human fetuses.

Conclusions: These findings suggest that microglial activation via GPCR dysregulation is a factor in the pathogenesis of PH. Furthermore, additional GPCRs appear to play a role in other malformations of cortical development, an active area of investigation.

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High-Fat Diet Ameliorates Myonuclear Transcriptional Changes in R155H/R155H VCP Disease

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Background: Valosin-containing protein (VCP) disease (Multisystem Proteinopathy 1) causes several phenotypes: inclusion body myopathy, amyotrophic lateral sclerosis, frontotemporal dementia, among others. Homozygous VcpR155H/R155H mice show profound weakness and severe pathology leading to premature death before three weeks of age. A lipid-enriched diet reverses lethality and improves muscle strength and pathology. However, the genetic changes underlying this process are not yet defined.

Methods: We performed single-nuclei RNA sequencing on 100,000 nuclei from skeletal muscles and hearts of three cohorts: VcpR155H/R155H mice on either 1) normal or 2) high-fat diet compared to 3) wildtype littermates on normal diet. Data was processed using Trailmaker and a custom pipeline of quality control, Harmony integration, CellTypist annotation, subsetting, and principle component analysis (PCA).

Results: We define the transcriptional changes present in R155H/R155H VCP disease vs wildtype and observed a transcriptional trajectory with VcpR155H/R155H mice on a high-fat diet residing between those on a normal diet and wild-type mice, especially among quadriceps muscle samples. Using a custom bridging tool to identify myonuclei with the strongest cell state rescue shift from disease to wildtype we identify several genes that co-vary along this trajectory as potential biomarkers of rescue, including some markers of inflammatory pathways. The mutation also caused significant changes in cell type populations, most notably the decreased proportion of Type IIb myonuclei.

Conclusions: In summary, this work provides an atlas of transcriptional changes in a murine model of VCP disease and identifies potential biomarkers. Our findings also demonstrate that a lipid-enriched diet partially restores the transcriptional changes, correlating with the observed reversal of early lethality and improvement in muscle health in affected mice. Collectively, these results underscore the beneficial role of dietary lipid enrichment and highlight new molecular insights that may inform future strategies for managing VCP-associated disorders.

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A Phosphorylation-Independent TDP-43 Antibody Reveals Increased and Earlier Cytoplasmic Pathology in Inclusion Body Myositis

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Background: Inclusion body myositis (IBM) is characterized by cytoplasmic aggregation of TDP-43 in skeletal muscle fibers, typically detected using phospho-specific antibodies targeting serine 409/410 (p409/410). Although p409/410 immunohistochemistry is widely used diagnostically, phosphorylation-dependent detection may preferentially label mature inclusions and incompletely capture earlier or structurally distinct TDP-43 pathology. Given increasing evidence for biochemical and conformational heterogeneity of pathological TDP-43, we evaluated whether a phosphorylation-independent monoclonal antibody (MAb No. 9) improves detection of IBM-associated pathology.

Methods: Formalin fixed, paraffin embedded muscle biopsies from 10 clinicopathologically confirmed IBM patients were examined using paired serial sections stained with MAb No. 9, which recognizes a C-terminal epitope of TDP-43, and p409/410 under standardized conditions. Slides were assessed in a blinded fashion. Quantitative analyses include counts of TDP-43–positive fibers and assessment of cytoplasmic aggregate burden. Double label immunofluorescence was performed to evaluate spatial overlap and subcellular staining patterns. Pathologic burden was correlated with clinical severity metrics.

Results: MAb No. 9 identified a greater number of TDP-43–positive muscle fibers and a higher cytoplasmic aggregate burden compared with p409/410 across all cases. In addition to labeling classic inclusion-associated pathology, MAb No. 9 revealed diffuse and fine granular cytoplasmic TDP-43 immunoreactivity in fibers lacking overt p409/410 positivity, consistent with earlier or pre-phosphorylated lesions. Immunofluorescent co-staining demonstrated substantial colocalization within mature aggregates, while MAb No. 9 additionally highlighted fine punctate cytoplasmic pathology not detected by phospho-specific staining. MAb No. 9–detected pathology showed stronger associations with clinical severity.

Conclusions: Phosphorylation-independent detection of TDP-43 using MAb No. 9 reveals increased and earlier cytoplasmic pathology in IBM muscle compared with p409/410. These findings suggest that immunohistochemistry with MAb No. 9 may enhance diagnostic sensitivity, particularly in early or low-burden IBM biopsies.

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Early myopathology of Pompe disease in patients detected by newborn screening

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Background: Newborn screening (NBS) has allowed for early diagnosis of Pompe disease. Herein we describe muscle biopsy findings in pediatric Pompe disease patients identified by NBS.

Methods: Opportunistic biopsies were performed at the time of port placement with appropriate consent for 2 infantile-onset Pompe disease (IOPD) and 4 late-onset Pompe disease (LOPD) patients identified via NBS. Age at biopsy, immunomodulatory therapy (IMRx) and enzyme replacement therapy (ERT) exposure, and muscle biopsy findings were examined.

Results: Mean documented age of symptom onset was 1.5 weeks in IOPD and 13.7 months in LOPD. At the time of biopsy, IOPD patients ranged from 5 months-10 years (one patient underwent serial biopsies years apart) and LOPD patients ranged from 1-4.5 years of age. One IOPD patient was CRIM-negative. The mean age at ERT initiation was 3.5 weeks in IOPD patients and 24.25 months in LOPD patients. Of the 6 patients, 4 received IMRx with ERT. In IOPD patients, the earliest changes included punctate sarcoplasmic acid phosphatase positivity and autophagic vacuoles with variable free and membrane-bound glycogen by electron microscopy (EM). Only very rare fibers contained obvious excess glycogen on frozen sections. Ultrastructural mitochondrial abnormalities were also a prominent early feature. Histopathologic changes in LOPD patients were highly variable. Light microscopic vacuolization ranged from absent to extensive. Acid phosphatase staining was most consistent, often with diffuse punctate sarcoplasmic positivity. Variable amounts of free and/or membrane-bound glycogen and autophagic vacuoles were noted. Mitochondrial changes on EM were also identified. In the IOPD patient with serial biopsies, ERT appeared to stabilize the histopathologic changes over the years.

Conclusions: Diagnosis via newborn screening allows assessment of early Pompe histopathologic findings. LOPD, despite typically being later-onset, showed substantial variability in young patient's biopsies, with considerable overlap between the two subtypes. Histopathology may provide insight into pathogenesis and open new avenues for disease-modifying therapies.

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Decoding Cancer-Associated Pain: A Single-Cell Proteomic Analysis of Human Dorsal Root Ganglia Neurons

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Background: Cancer-associated pain is a debilitating condition affecting approximately 30% of cancer patients. A major barrier to developing effective therapies is the limited understanding of the underlying molecular mechanisms and the lack of robust animal models. Human dorsal root ganglia (hDRG) contain unique nociceptor subtypes not found in rodents. This study utilizes a unique cohort of hDRG samples coupled with detailed clinical history to analyze the proteomic signatures driving cancer-associated pain.

Methods: 18 hDRG samples were procured, including patients with cancer-associated bone pain (n=6), visceral pain (n=5), and controls with no history of pain (n=7). For single-cell proteomics (SCP), neurons were isolated via laser capture microdissection followed by ultra-high sensitivity data-independent acquisition mass spectrometry (DIA-MS). Weighted gene correlation network analysis (WGCNA) and pseudobulk integration were used to correlate molecular alterations with clinical pain phenotypes.

Results: Preliminary SCP analysis of 109 neurons from 8 donors identified 8,517 unique proteins, with approximately 4,000 proteins detected per neuron. Principal component analysis revealed near-complete segregation of neurons from patients with pain versus controls, independent of immune modulatory or chemotherapy. We identified distinct differentially abundant proteins (DAPs) not typically altered in nerve injury models, including immune regulators (B2M, C1QC, IL6ST), G protein-coupled receptors (ADGRB3), and ion channels (GRIP1, LRRC8C). Furthermore, proteomic profiling was able to classify DRG neurons into six distinct subpopulations, including A-LTMRs, A-HTMRs, C-peptidergic, and C-non-canonical-peptidergic nociceptors, with both A and C neurons exhibiting disease-associated alterations.

Conclusions: This study represents the first effort to perform SCP analyses on hDRG neurons in the context of cancer pain. Our findings suggest that cancer-associated pain is driven by molecular signatures in sensory neurons district from classical nerve injury paradigms. By defining these human-specific, subtype-specific mechanisms, we aim to establish a reference atlas for the pain research community and identify novel, actionable targets for pain therapies.

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Border-associated macrophages mediate immune responses in the unmyelinated optic nerve head in mice and humans

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Background: Immune cells play a critical role in the pathogenesis of glaucoma. The optic nerve head (ONH), an early site of neurodegeneration in glaucoma, has been thought to primarily contain microglia. However, healthy central nervous system also contains border-associated macrophages (BAMs) with distinct molecular and functional profiles. In this study, we investigated the heterogeneity, distribution and ontogeny of immune cell populations in the ONH under homeostatic conditions and in a microbead-induced mouse model of glaucoma.

Methods: Elevated intraocular pressure was induced in 3-month-old mice by injecting microbeads into the anterior chamber, while sham-injected mice served as controls. 30 days after injection, retinas and optic nerves were harvested and CD45+ immune cells were isolated and sequenced using 10X Genomics platform. To determine cellular origin, lineage tracing was performed using Pf4cre, Ms4a3cre, and Sall1ncre:Cx3cr1ccre lines. Key findings were validated in human postmortem optic nerve tissues.

Results: A total of 28,277 immune cells were sequenced from microbead- and sham-injected retinas and optic nerves. We identified a distinct population of perivascular myeloid cells in the unmyelinated region of ONH that lacked expression of the homeostatic microglia marker P2ry12 and instead expressed high levels of immune signaling molecules, including MHCII, CD11c, and CD25. Abundant T cells were observed in close association with $CD25^{+}$ BAMs. Lineage tracing revealed minimal labeling by the microglia-specific line (2.4%), whereas BAM and monocyte lineage lines labeled 55.3% and 27.5% of these cells, respectively. We validated these findings in human optic nerve, finding that human lamina cribrosa contained $P2ry12^{-}$, $MHCII^{+}$ and $CD163^{+}$ myeloid cells. In glaucomatous murine ONH, MHCII expression was significantly increased in this population compared to controls.

Conclusions: Our investigation showed that perivascular BAMs and monocyte-derived macrophages, rather than microglia, populate the unmyelinated ONH in both mice and humans and exhibit a robust immune activation during glaucoma progression.

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Targeting IGF1R signaling in BCOR mutant retinoblastoma

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Background: BCL-6 corepressor (BCOR) loss of function alterations are common in a range of neural tumors, including clinically aggressive retinoblastoma and medulloblastoma. We investigated the effects of BCOR loss in retinoblastoma cells, and developed novel combination chemotherapies including agents targeting IGF1R signaling.

Methods: Chemotherapeutic agents including IGF1R inhibitors were tested in a panel of paired BCOR intact/deficient retinoblastoma cell lines, and the effects on proliferation, clonogenicity, apoptosis and migration were assessed in vitro and in murine xenograft models.

Results: BCOR knockdown or knockout increased tumor growth, invasion, clonogenicity and in vitro and in xenografts, as well as resistance to standard chemotherapeutic agents such as carboplatin and melphalan. Loss of BCOR function also increased IGF1 expression and IGF1R signaling, and sensitized retinoblastoma cells to IGF1R pharmacologic inhibition either small molecules (linsitinib, AEW541) or antibodies (Teprotumumab). Preliminary studies also show that BCOR can directly bind to DNMT1 and modulate IGF1 promoter methylation, suggesting a novel mechanism for regulation of IGF1/IGF1R signaling by BCOR.

Conclusions: Loss of BCOR function is associated with more aggressive retinoblastoma cell line growth and chemoresistance, at least in part due to increased IGF1R signaling. Inhibiting IGF1R pharmacologically had a marked anti-tumor effect in aggressive retinoblastoma lacking BCOR, suggesting it as a new therapeutic target.

POSTERS

Posters: Tumors: Glial

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Transcriptional Regulation by ZNF143 Promotes Metabolic Reprogramming in Aggressive Glioblastoma

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Background: Glioblastoma is the most aggressive tumor of the central nervous system. Its metabolic plasticity enables tumor cells to rewire their energy pathways to sustain rapid growth in a tumor microenvironment. However, the molecular drivers in the rewiring of metabolic pathways towards malignancy remain poorly understood. This study aimed to analyze longitudinal glioblastoma data to identify a master Transcription Factor (TF) that regulates aggressive metabolic reprogramming.

Methods: Longitudinal bulk RNA-seq data from the GLASS consortium dataset were analyzed to identify differentially expressed genes (DEGs) across IDH status and tumor grade. DEGs were intersected with metabolic genes from GOBP and Reactome, and finally correlated with CCLE metabolites. Protein-protein interaction (PPI) analysis of the gene set created distinct metabolic clusters, which were assigned to samples. The change from primary to recurrent samples towards clusters associated with worse prognosis was quantified. TF activity was inferred using VIPER, and the TF's differential expression in recurrent samples and correlation with metabolic drift magnitude were evaluated to prioritize TFs that potentially drive reprogramming toward aggressive states. Finally, functional validation of ZNF143 was performed in U87MG cells using siRNA knockdown, qPCR, proliferation, and migration assays.

Results: 3,073 DEGs were identified from DEG analysis and were intersected with GOBP metabolic genes. Next, genes that correlated with at least one metabolite activity were selected and expanded by combining with a Reactome metabolic gene list, producing a total of 479 genes. PPI analysis of these genes revealed 10 clusters. Clusters 5 and 7 were associated with significantly worse prognosis, and upon conducting drift analysis, ZNF143 emerged as the top candidate to drive the transition toward those aggressive clusters. In-vitro analysis showed comparable evidence of GBM phenotype switching when ZNF143 was knocked down.

Conclusions: ZNF143 is a master TF that promotes metabolic reprogramming towards more aggressive phenotypes, evidenced by computation and in-vitro findings.

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Tumor infiltrating clonal hematopoiesis (TICH) in IDH-wildtype glioblastomas

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Background: Clonal hematopoiesis of indeterminate potential (CHIP) is an age-associated expansion of hematopoietic stem and progenitor cells driven by somatic mutations detectable in peripheral blood of otherwise healthy individuals. CHIP is associated with increased risk of hematologic malignancy, cardiovascular disease, solid tumors, and all-cause mortality. Tumor-infiltrating clonal hematopoiesis (TICH) describes the presence of CHIP-derived myeloid clones within solid tumors. The clinical significance of TICH is variable between solid tumors, with favorable effects reported in metastatic colorectal cancer and negative effects in lung cancer. The prevalence and clinical significance of TICH in glioblastoma remain poorly defined.

Methods: We performed a retrospective analysis of clinical and DNA next-generation sequencing (NGS) data from 122 glioblastomas diagnosed between 2019 and 2025 at the University of Washington. DNA NGS was conducted using a clinically validated 472 gene solid tumor panel. Somatic variants in CHIP driver genes, defined by the 2024 WHO Classification of Hematolymphoid Tumors (5th edition), were curated. Putative CHIP-associated variants were defined by variant allele frequency (VAF) ≥2% using established gene-specific criteria and were cross-referenced with COSMIC, ClinVar, and cBioPortal. Overall survival (OS) and progression-free survival (PFS) were analyzed using Kaplan–Meier methodology.

Results: TICH-associated mutations were identified in 24 of 122 cases (19.7%), involving 15 distinct genes with a median VAF of 4% (range 3–5%). Most TICH-positive tumors harbored a single mutation. The most frequently affected genes were DNMT3A, TP53, TET2, and PRPF40B, accounting for 50% of identified variants. The presence of TICH was not significantly associated with OS (p=0.3488) or PFS (p=0.7896).

Conclusions: TICH is detectable in approximately 20% of glioblastomas using tumor-based DNA NGS. In this cohort, TICH was not associated with adverse clinical outcomes. These findings establish the prevalence of TICH in glioblastoma and suggest that, at the VAF thresholds examined, its presence does not independently predict survival.

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Quantitative Morphometric Features Distinguish SOX2+ Tumor Cells from Reactive Gliosis in Glioblastoma

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Background: Our neuro-oncology brain bank maintains a growing collection of 83 glioblastoma (GBM) whole brain autopsy specimen with corresponding clinical histories and imaging. SOX2 is overly expressed in GBM and immunohistochemistry (IHC) identifies both tumor cells with stem cell characteristics and reactive gliosis in adjacent grey matter, limiting its specificity for tumor detection. We hypothesized that quantitative nuclear morphology could distinguish these populations.

Methods: We analyzed 37 large axial tissue sections (approximately 2 in. by 3 in.) from 14 GBM patients obtained at autopsy. Sections were selected based on visible abnormalities on T1-contrast and FLAIR imaging from the patient's most recent clinical MRI. Tissue was stained with SOX2-DAB immunohistochemistry, digitized at 40x resolution, and housed in our OMERO-powered whole slide imaging repository. Circular regions of interest (ROIs) were manually annotated in tumor core and adjacent grey matter (74 total ROIs). Using QuPath's positive cell detection with custom parameters (0.5 μm pixel size, 0.22 DAB OD threshold, 0.24 watershed detection threshold), we quantified nuclear features including area, circularity, and DAB optical density for all detected positive staining cells. Mixed-effects models accounting for hierarchical structure (samples nested within patients, ROIs nested within samples) tested for regional differences in SOX2+ cell characteristics.

Results: We detected 1,810,753 SOX2+ cells (1,402,064 tumor, 408,689 grey matter). Mixed-effects modeling revealed significant morphological differences (all $p < 0.0001$): tumor-associated SOX2+ cells exhibited markedly higher DAB optical density (mean difference = +0.347, representing 2.2-fold higher SOX2 expression) and lower circularity (mean difference = -0.031) compared to reactive gliosis within the grey matter. At the ROI level (n=74), effect sizes were large for DAB optical density (Cohen's $d = 1.21$) and moderate for nuclear circularity ($d = -0.73$).

Conclusions: Therefore, we conclude that quantitative DAB intensity coupled with nuclear shape features could enhance specificity of SOX2 staining for tumor margin assessment in GBM neuropathology.

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The Spatial Distribution of SOX2 and Ki-67 Relative to Both Vasculature and Ablation Margin after LITT for Glioblastoma: a Post-Mortem Case

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Background: Laser interstitial thermal therapy (LITT) is a minimally invasive treatment for glioblastoma (GBM), but recurrence is considered almost inevitable. Blood vessels within the treatment zone are hypothesized to act as heatsinks, protecting nearby tumor cells from lethal temperatures. If confirmed, this would represent a targetable source of treatment failure. This study quantitatively investigated the spatial distribution of stem-like (SOX2+) and proliferative (Ki-67+) cells relative to vasculature and the LITT ablation margin in post-mortem GBM tissue.

Methods: Post-mortem tissue from three GBM patients treated with LITT was stained for SOX2 and Ki-67. A spatial analysis workflow was developed using whole slide images digitized at 40x magnification. Cell centroids were vectorized from QuPath, and vessel and LITT margin boundaries were manually annotated in OMERO. The shortest Euclidean distance from each cell to its nearest vessel margin and from each vessel centroid to the LITT margin was computed. Linear mixed-effects models with patient ID as a random effect assessed the relationship between marker positivity and distance from these features.

Results: A statistically significant perivascular niche was identified for SOX2+ cells, with positivity decreasing approximately 2% per 100 micrometers from the vessel wall ($p < .001$). A significant interaction effect ($p < .001$) demonstrated this enrichment was most pronounced for vessels deep within the LITT ablation margin. Stratified analysis confirmed a clear gradient: vessels inside the ablation zone showed the steepest decline in perivascular SOX2 positivity, followed by vessels near the margin, then far from it. Ki-67 showed a similar but weaker perivascular trend ($p < .001$), with a marginally significant LITT margin interaction ($p = .044$).

Conclusions: These findings suggest a vascular heatsink effect creating a protected perivascular space for malignant GBM cells, particularly stem-like SOX2+ cells. This niche represents a likely source of recurrence and a rational target for adjuvant strategies to improve LITT efficacy.

5

Metastatic Glioblastoma with Unusually Long Survival

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Background: Glioblastoma displays high tumor heterogeneity, including its histologic and clinical behavior. We report a case of a 63-year-old male with metastatic glioblastoma (MGMT promoter unmethylated, IDH-WT) who has survived more than 40 months from initial diagnosis.

Methods: The patient initially presented to medical attention in July 2022, with progressive hemiparesis and sensory changes, feeling generally unwell, significant gait instability with falls, and cognitive slowing. Imaging revealed a left parietal mass, and he underwent a surgical resection. Pathology revealed an MGMT-unmethylated IDH-wildtype glioblastoma, and he subsequently underwent chemotherapy and radiation, which he completed in May 2023. He also used tumor-treating fields/Optune for two months.

Results: He presented to our institution in May 2023 for posttreatment surveillance. Surveillance imaging in September 2023 revealed possible disease progression, and he subsequently underwent an additional brain resection in November 2023. Histopathologic and molecular analysis of the resected tissue redemonstrated unmethylated, IDH-WT glioblastoma with notable variants in TERT and TP53, gain of chr7 with EGFR amplification and loss of chr10, and loss of chr9 (CDKN2A, CDKN2B). In November 2024 he underwent right femoral neck resection demonstrating the same tumor, and in January 2025 he underwent FNA of a left cervical lymph node demonstrating the same tumor, both confirmed to be metastases.

Conclusions: This represents an unusual case of glioblastoma that metastasized to two additional organs, with continued survival beyond mean expected survival.

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Case report of a glioblastoma with nonfunctional H3 K27R mutation

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Background: Histone H3 p.K27M mutations are a hallmark of diffuse midline gliomas. We encountered a high-grade glioma with H3-3A p.K27R mutation that required further molecular workup for classification.

Methods: We reviewed the patient's clinical record, examined the tissue histology, and performed next generation sequencing and methylation analysis.

Results: We report a case of a 54-year-old male who initially presented with altered mental status. MR imaging of the brain revealed a 4.6 cm thick irregular rim enhancing mass with central necrosis centered in the left posterior-medial parietal lobe, highly suspicious of high grade multicentric glial neoplasm. Biopsy of this lesion confirmed the presence of high-grade glioma that was immunoreactive for OLIG2 and GFAP but negative for IDH1 p.R123H mutant protein. Next generation sequencing on the resection specimen revealed a surprising H3-3A p.K287R histone mutation in addition to mutations in TERT promoter, TP53, EGFR, and amplification of PDGFRA. H3K27me3 was performed which was interpreted as retained but additional material was insufficient for other ancillary molecular studies. The patient underwent tumor re-resection 4 months after the initial biopsy. Repeat immunohistochemical studies demonstrated retained H3K27me3. DNA methylation profiling was performed on the re-resection which revealed a methylation signature matching to “glioblastoma, IDH-wildtype” with high confidence.

Conclusions: We report a case of a glioblastoma with a rare H3-3A p.K27R somatic variant. Methylation studies confirmed this tumor aligns with “glioblastoma, IDH-wildtype” rather than “diffuse midline glioma, H3 K27-altered”. Immunohistochemical studies also confirm retained H3K27me3. Therefore, the H3-3A p.K27R change in methylation is presumed to be nonfunctional. How this mutation contributes to tumor pathophysiology remains unknown, but may cause diagnostic confusion if encountered in other cases.

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Glioblastoma, IDH-wild type with an unusual EWSR-1:RAD51B gene fusion: A case report and review of the literature

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Background: Glioblastoma (GBM) is the most common and aggressive primary malignant neoplasm of the central nervous system in adults. According to the 2021 World Health Organization (WHO) classification, GBM is defined as an IDH-wild-type diffuse astrocytic glioma with characteristic histologic and/or molecular features. The expanding use of integrated histologic, molecular, and epigenetic diagnostics has revealed increasing biological heterogeneity within this category. We report a case of a 73-year-old man with glioblastoma, IDH-wildtype, harboring a novel EWSR1:RAD51B gene fusion identified by next-generation sequencing. Magnetic resonance imaging revealed a heterogeneous, predominantly peripherally enhancing mass involving the left temporal, parietal, and occipital lobes. Histologic examination of the resected specimen demonstrated a diffusely infiltrative high-grade glioma with marked nuclear pleomorphism, brisk mitotic activity, microvascular proliferation, and geographic and pseudopalisading necrosis. Immunohistochemistry showed strong nuclear OLIG2 positivity; IDH1-R132H immunostaining was negative; ATRX and H3K27me3 expression were retained; and p53 exhibited a wildtype pattern. The Ki-67 (MIB-1) proliferation index reached approximately 20% in the most proliferative areas. Molecular studies revealed MGMT promoter methylation and a truncating PTEN mutation (p.Q214*, c.640C>T). A previously unreported EWSR1:RAD51B gene fusion was identified by next-generation sequencing. DNA methylation profiling classified the tumour as glioblastoma, IDH-wildtype, with high confidence.

Methods: N/A

Results: N/A

Conclusions: To our knowledge, this is the first reported case of glioblastoma harboring an EWSR1:RAD51B gene fusion. Although the biological significance of this alteration remains unclear, its identification expands the molecular spectrum of GBM and underscores the value of comprehensive genomic and epigenomic profiling in the diagnostic evaluation of diffuse gliomas, particularly when unexpected or atypical molecular findings are encountered.

8

Glioblastoma Survival with Partial Gains and Losses in Chromosomes 7 and 10 at Henry Ford Health

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Background: The gain of chromosome 7 and the loss of chromosome 10 are one of the essential diagnostic criteria for the diagnosis of IDH-wildtype glioblastomas in patients above the age of 40. Combinations including partial gain of chromosome 7 and/or partial loss of chromosome 10 are less investigated. The largest study to date included 33 cases of nine possible combinations of chromosome 7 gains and chromosome 10 losses. From this work it was surmised that only the gain of 7q along with whole chromosome 10 loss or the loss of 10q along with gain of whole chromosome 7 portend to carry the prognostic significance of the 7/10 signature.

Methods: At Henry Ford Health we used Oncoscan chromosomal microarrays to identify partial 7 gains and 10 losses along with NGS to better characterize the tumors.

Results: We identified 22 cases within the previously established groups of 7q+/10− (n = 6), 7+/10q− (n = 3), 7q+/10q− (n = 2), 7p+/10− (n = 5), 7+/10p− (n = 3) and 7p+/10q− (n = 3). No cases with either 7q+/10p- or 7p+/10p-. Most cases possess a TERT promoter mutation (19/22) and EGFR amplification/gain (20/22), however, they differ in other variants such as homozygous CDKN2A deletions (15/22) and TP53 mutations (10/22). Survival data is currently available for only 13 cases and statistical analysis shows no significant differences in time of survival or histologic features but an expected trend in longer survival with 7p+/10- and 7+/10p-.

Conclusions: Our work adds three cases to the previously uncharacterized 7p+/10q- group and two cases to the minimally represented 7+/10p- group. It may also point to a fundamental requirement for 7q gain or 10q loss in that neither cohort contains glioblastomas with 7p+/10p-. This work serves to address a critical issue in diagnostic neuropathology and points toward a need for more data in order to determine prognosis and possibly optimize treatment for glioblastoma.

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Integrative 68Ga-FAPI PET/MRI and transcriptomic profiling for investigation of aggressiveness and stromal biology in diffuse gliomas

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Background: Diffuse gliomas are the most common malignant primary brain tumors in adults and remain incurable due to their infiltrative growth and extensive molecular heterogeneity, which together undermine current therapeutic strategies. Conventional contrast-enhanced MRI provides limited insight into the underlying histopathologic and molecular features of these tumors, underscoring the need for advanced imaging biomarkers that more accurately capture tumor biology. Fibroblast activation protein (FAP), a membrane-bound glycoprotein expressed in cancer-associated fibroblasts and tumor vasculature but largely absent from normal brain tissue, is upregulated in high-grade gliomas, suggesting a role in tumor progression and potential utility in targeted imaging and therapy. This study investigates the use of the FAP-specific radiotracer (68)Ga-FAPI, combined with multiparametric PET/MRI and transcriptomic profiling, to characterize stromal and molecular correlates of glioma aggressiveness.

Methods: Ten adults with diffuse glioma undergoing preoperative evaluation were prospectively enrolled. Each participant underwent (68)Ga-FAPI PET/MRI including structural, diffusion, and perfusion MRI sequences, alongside dynamic and static PET imaging. Regions of interest were defined on MRI, and voxelwise FAPI uptake was correlated with apparent diffusion coefficient and relative cerebral blood volume metrics. Intraoperative biopsies targeted regions exhibiting differential FAPI uptake, which were subsequently analyzed by histopathology, immunofluorescence for FAP, and bulk RNA sequencing. Bioinformatic analyses identified gene expression signatures and pathways associated with high FAP uptake.

Results: We anticipate that (68)Ga-FAPI PET/MRI will reveal increased uptake in regions with histologic features of high-grade disease, such as microvascular proliferation and necrosis, and that FAPI-high regions will show transcriptomic enrichment in stromal activation, angiogenesis, and extracellular matrix remodeling pathways.

Conclusions: These integrated multimodal analyses aim to establish FAP as a promising imaging biomarker of stromal-mediated glioma aggressiveness and inform future development of FAP-targeted theranostic approaches in diffuse glioma.

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Radiation-induced glioma with extracranial metastases and liquid biopsy-detected KRAS mutation

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Background: Radiation-induced central nervous system (CNS) neoplasms are a rare phenomenon with a low reported incidence of 0.08-0.6%. Their development following radiotherapy for craniopharyngiomas constitutes an even smaller subset of reported cases. Additionally, extracranial metastasis of primary CNS tumours, regardless of prior cranial radiation, is uncommon. The molecular profiles of these rare tumours remain poorly characterized, particularly in the context of radiation-induced metastatic gliomas.

Methods: We describe the autopsy findings of a 33 year old woman who developed a primary malignant neuroepithelial tumour with glial and primitive neuroectodermal features, exhibiting metastases to multiple extracranial sites. This occurred 12 years after adjuvant radiotherapy to the suprasellar and brainstem regions for a subtotally resected adamantinomatous craniopharyngioma.

Results: Prior to the patient's death, neuroimaging demonstrated extensive intracranial and leptomeningeal disease, along with extracranial metastases consistent with a suspected high-grade glioma. Circulating tumour DNA in the cerebrospinal fluid revealed a KRAS mutation and CDKN2A homozygous deletion. Idylla rt-PCR for KRAS performed on an iliac biopsy of one metastatic lesion further confirmed the mutation. Post-mortem examination showed widespread meningeal dissemination, with involvement of the anteroventral cingulate, septal area, corpus callosum, caudate nucleus, hypothalamus, mammillary bodies, thalamus, brainstem, and cerebellum. Extracranial metastases were found in the lymph nodes, lungs, thoracic pleura, diaphragm, liver, kidneys, and vertebrae. The neoplastic cells exhibited both glial and primitive neuroectodermal cytoarchitecture, with focal papillary formations. Microvascular proliferation and focal tumour necrosis were identified. Immunohistochemistry showed variable positivity for GFAP, Olig2, synaptophysin, and MAP2, with diffuse CD56 positivity. The lesional cells were immunonegative for IDH1 R132H, HMB-45, and cytokeratin, and retained H3K27 trimethylation.

Conclusions: This is a unique case of a radiation-induced metastatic high-grade glioma with primitive neuroectodermal elements, which to our knowledge has not been described in the literature. KRAS mutations are uncommon and may be implicated in this glioma's aggressive behaviour.

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Histopathology-Based Recurrence Risk Stratification in Grade 2–3 Astrocytomas Using Structured Multi-Stream Deep Learning

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Background: Recurrence prediction in diffuse astrocytomas remains clinically challenging, particularly for World Health Organization (WHO) grade 2 and grade 3 tumors, where clinical behavior is heterogeneous and prognostic indicators are less well established than in high-grade gliomas. Whole-slide histopathology images contain rich morphological information, but translating gigapixel tissue data into reliable patient-level risk prediction requires models capable of identifying and integrating heterogeneous tumor regions.

Methods: We developed a deep learning framework for recurrence prediction in IDH-mutant astrocytomas using routine hematoxylin and eosin whole-slide images. The study focused exclusively on WHO grade 2 and grade 3 astrocytomas. Patch-level representations were extracted using a pretrained pathology foundation model and aggregated through multiple instance learning (MIL). Tissue regions were characterized according to their learned association with patient outcomes, enabling construction of a four-stream MIL architecture in which patches were assigned based on prognostic contribution. Separate streams modeled tissue associated with increased recurrence risk and tissue associated with prolonged survival, allowing complementary morphological signals to be learned independently before integration into a unified patient-level prediction across multiple slides.The model was trained and validated using five-fold cross-validation on 258 mixed grade 2–3 astrocytomas from The Cancer Genome Atlas (TCGA).

Results: The framework achieved an overall concordance index (C-index) of 0.71 for recurrence prediction. Risk stratification yielded a hazard ratio of 3.008, indicating approximately a three-fold increase in recurrence hazard among patients classified as high risk. Kaplan–Meier analysis demonstrated an absolute progression-free survival difference (ΔS) of 0.2187 at 24 months, corresponding to an approximately 22% separation in survival probability between predicted risk groups.

Conclusions: A structured four-stream MIL architecture that separately learns from recurrence-associated and survival-associated tissue regions improves modeling of histologic heterogeneity in lower-grade astrocytomas and provides a scalable framework for recurrence risk stratification using routine histopathology.

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Withdrawn

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Characterization of a High-Grade Glioma Associated with Cowden Syndrome: A Rare Event

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Background: Cowden syndrome (CS), an autosomal dominant condition characterized by germline mutations of PTEN, is associated with numerous hamartomatous lesions along with increased risk of malignancies, most commonly involving the thyroid, endometrium, and breast. In the central nervous system, CS can manifest as Lhermitte-Duclos disease. Only rare instances of high-grade glioma have been reported in association. Here, we characterize a high-grade glioma arising in a patient with known CS.

Methods: Clinical history, radiology, histology, immunophenotype, and comprehensive molecular profiling of the tumor were assessed.

Results: A 48-year-old female with known germline PTEN mutation (p.R233*) and history of breast and endometrial carcinoma presented with a large heterogeneously enhancing mass of the left frontal lobe. Morphologically, the tumor was a cellular infiltrating glioma with variable histologic features: some areas had a lipidized appearance, some appeared primitive, and areas showed large cells with abundant eosinophilic cytoplasm and enlarged highly atypical nuclei with multinucleation. Foci of microvascular proliferation and extensive necrosis were seen. The tumor cells were positive for GFAP and Olig2 with retained ATRX nuclear expression. Besides the known germline PTEN alteration, molecular analysis revealed mutations in DAXX, PIK3CA, PDGFRA, PTPN11, TP53 and homozygous deletion of CDKN2A/B.

Conclusions: Though not among the most commonly associated tumors with Cowden Syndrome, rare reports of gliomas, such as this case, have occurred.

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SLC34A2::ROS1 Fusions Identified at Recurrence in IDH-Wildtype and IDH-Mutant Gliomas

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Background: Gliomas are the most common primary malignant tumors of the adult central nervous system and remain therapeutically challenging, particularly at recurrence. Although ROS1 fusions are rare in gliomas, they represent potentially actionable oncogenic events across age groups. Comprehensive molecular profiling studies have shown that ROS1 fusions involve a broad spectrum of partner genes beyond the commonly observed GOPC::ROS1. Targeted ROS1 inhibition has demonstrated measurable therapeutic activity in ROS1 fusion-positive cancers, with emerging evidence suggesting potential benefit from CNS-penetrant ROS1 inhibitors.

Methods: We describe two cases of diffuse gliomas in which molecular profiling at recurrence identified SLC34A2::ROS1 fusion: (1) an IDH-wildtype glioblastoma in a 74-year-old woman and (2) an IDH-mutant, 1p/19q-codeleted oligodendroglioma in a 53-year-old man.

Results: Patient 1: Initially diagnosed with glioblastoma with TERT promoter mutation and MGMT promoter unmethylated. Following temozolomide, radiotherapy, and intra-arterial stem cell infusion, imaging at 8 months was concerning for recurrence versus treatment effect, and re-resection demonstrated glioblastoma with sarcomatoid features. Molecular profiling of the recurrent tumor demonstrated SLC34A2::ROS1, with persistent TERT promoter mutation, retained MGMT promoter unmethylated status, and additional alterations including PTPRD and STAG2. Patient 2: Presented initially with oligodendroglioma, IDH-mutant and 1p/19q-codeleted, CNS WHO grade 2 (limited molecular testing at diagnosis). After temozolomide and radiotherapy, progression prompted biopsy demonstrating oligodendroglioma, CNS WHO grade 3. Molecular profiling of the recurrent tumor demonstrated SLC34A2::ROS1, in addition to IDH1 p.R132H, PIK3CA, NOTCH1, SETD2, TERT promoter alterations, and MGMT promoter methylation.

Conclusions: ROS1 fusions represent rare but potentially actionable alterations in gliomas that may be identified at recurrence in both IDH-wildtype and IDH-mutant tumors. Recognition of these alterations may expand therapeutic options, including targeted therapy and clinical trial enrollment.

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ROS1 Fusion as a Rare but Important Molecular Marker in the Identification and Treatment of Low-grade Gliomas

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Background: Gliomas are the most common primary tumor of the central nervous system and represent a diverse group of neoplasms derived from glial cells of the brain and spinal cord. Low-grade gliomas (WHO grade I and II) often harbor distinct genetic alterations, such as gene fusions, which serve as important diagnostic and therapeutic biomarkers. Among these, the ROS proto-oncogene 1 (ROS1), a transmembrane receptor tyrosine kinase, represents a rare genetic alteration (about 7% of gliomas with reported ROS1 fusion) that may serve as a potential targetable molecular alteration. Here, we present a case of a 26-year-old male with no past medical history who presented with new onset seizures and an 8.6 cm left temporal lobe mass extending into the ventricles. Histopathologic evaluation showed a remarkably bland glial neoplasm prompting whole-genome methylation for definitive identification. Methylation profiling revealed a low-grade glioma with ROS1 fusion, confirming the diagnosis and providing a potentially targetable genetic driver. Although ROS1 fusions in gliomas are rare, accurate recognition is critical, especially in patients with subtotally resected, recurrent, or highly malignant tumors, for proper tumor classification and may prove to be a highly relevant and actionable therapeutic target.

Methods: N/A

Results: N/A

Conclusions: N/A

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Molecular profile of granular cell astrocytoma predicts aggressive clinical behaviour independent of morphology

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Background: Granular cell astrocytoma (GCA) is a rare astrocytic tumour variant that may appear morphologically low-grade yet behaves aggressively. Its molecular features remain poorly defined and no methylation-based classification has previously been reported.

Methods: We describe three GCAs diagnosed in our department, integrating clinical, histopathological and molecular data (including methylation profiling and, in one case, whole genome sequencing (WGS)).

Results: The patients were aged 63, 54 and 50 years, each presenting with supratentorial tumours showing granular cell morphology ranging from a bland, low-grade-appearing neoplasm (Case 1) to tumours with low-grade granular areas transitioning into high-grade astrocytic components with mitotic activity, microvascular proliferation and necrosis (Cases 2–3). Despite this morphological spectrum, all matched the methylation class “Glioblastoma, IDH-wildtype, mesenchymal subtype” and shared molecular features typical of glioblastomas, including chromosome +7/−10, TERT promoter mutation and CDKN2A/B deletion. All harboured concomitant NF1 alterations. WGS in Case 2 identified homozygous MTAP loss and a chromoanasynthesis on chromosome 9. Case 1 received Stupp-protocol chemoradiotherapy, recurred after 3 months, and died 11 months after diagnosis. Case 2 has progressed with a new posterior fossa lesion while on adjuvant temozolomide. Case 3 has just completed Stupp-protocol chemoradiotherapy and remains clinically stable.

Conclusions: This first report of GCAs with methylation profiling demonstrates that these morphologically variable tumours molecularly correspond to the mesenchymal subtype of IDH-wildtype glioblastoma. Integrated molecular testing is essential for accurate classification and for identifying potential clinical trial eligibility.

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Glioneuronal tumours harboring NTRK gene rearrangements: pathological diversity in three cases

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Background: Glioneuronal tumours harbouring receptor tyrosine kinase (RTK) gene fusions have been recently described; however, their pathological spectrum and clinical behaviour remain poorly characterised. We report three cases of NTRK-fused neuroepithelial tumours with heterogeneous morphology and anatomical distribution.

Methods: Three patients were identified: a 29-year-old woman (Case 1), an 8-year-old girl (Case 2), and a 5-year-old boy (Case 3). Clinical, radiological, and histopathological data were reviewed, alongside DNA methylation profiling (DKFZ Brain Classifier v12.8), targeted DNA/RNA panel and whole genome sequencing in one case. Clinical follow-up information was also collected.

Results: Cases 1 and 2 presented with frontal lobe tumours, while Case 3 presented with a posterior fossa lesion radiologically suspicious for a dermoid cyst. Histologically, Case 1 showed a low-grade glioneuronal tumour composed of oligodendroglia-like cells with prominent ring-like nuclear clusters. In contrast, Cases 2 and 3 demonstrated similar patterns dominated by primitive-appearing, Olig2-positive embryonal-type cells with focal neurocytic and ganglioneuroblastic differentiation, raising consideration of CNS neuroblastoma, FOXR2-activated. DNA methylation profiling classified both supratentorial tumours as MC Glioneuronal Tumour, Subtype A, while the infratentorial tumour showed no match to established classes. Targeted sequencing identified RTK fusions in all cases: an in-frame TPM3::NTRK1 inversion, an AGAP1::NTRK2 fusion, and a TPR::NTRK1 fusion, respectively. At follow-up, Case 1 remains tumour-free 34 months post-resection. Cases 2 and 3 are radiologically stable at 22 and 18 months following gross total resection with adjuvant radiotherapy and maintenance chemotherapy according to the SIOP-PNET5 protocol.

Conclusions: NTRK-fused glioneuronal tumours represent a clinically and morphologically heterogeneous group, including infratentorial lesions with embryonal-type morphology that may remain unclassifiable by current methylation frameworks. Therapeutic decision-making in such cases is challenging, and NTRK inhibitors may represent a valuable targeted treatment option in the future.

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High-grade glioma with pleomorphic and pseudopapillary features (HPAP) presenting as a hemorrhagic lesion in a young adult: A case report

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Background: High-grade glioma with pleomorphic and pseudopapillary features (HPAP) is a recently described entity characterized by diverse morphology and characteristic genetic alterations, including TP53 and RB1 mutations and losses of chromosomes 13 and 17. HPAPs exhibit circumscribed growth resembling pleomorphic xanthoastrocytomas (PXAs) and improved survival compared with glioblastomas.

Methods: 22-year-old male with history of stimulant use presented with headache, nausea, and vomiting. MRI revealed a 3.5 x 3.3 cm hemorrhagic right parietal lesion with a 0.9 cm rim-enhancing nodule post-contrast. He was discharged with suspicion of intraparenchymal hemorrhage due to stimulant use. Four months later, he lost consciousness and was found to have a cystic right parietal mass, which was resected. Histopathologic assessment revealed a circumscribed, morphologically heterogeneous glioma with spindled growth, xanthomatous and giant cells, perivascular pseudorosettes, and eosinophilic granular bodies. The neoplasm was mitotically active with high Ki67 proliferation index (23.45%), variable immunoreactivity for GFAP and CD34, and sparse reticulin staining. Whole exome sequencing identified RB1 and PTEN mutations – the latter not specific for HPAP nor frequently reported. Low-pass whole-genome sequencing showed relative copy-number losses across multiple chromosomes, with no focal amplifications, deletions, or segmental alterations. DNA methylation-based tumor classification revealed high confidence (0.99) clustering with HPAP. Treatment of patients with HPAP includes chemotherapy and radiation; the patient opted for surveillance and is progression-free after seven months.

Results: HPAPs may be histologically indistinguishable from other high-grade gliomas, such as PXAs, and can exhibit an ambiguous immunohistochemical and genetic profile due to intra-tumoral heterogeneity. High-grade gliomas in young adults with diverse morphology that lack desirable genetic features of PXAs warrant heightened evaluation through comprehensive sequencing or methylation profiling to distinguish HPAP from other entities.

Conclusions: HPAPs may present as hemorrhagic lesions in young adults. Given morphologic overlap with other high-grade gliomas and PXAs, advanced molecular analysis is critical for accurate classification.

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Diffuse midline glioma, H3K27-altered, showing loss of H3 K27M mutation upon progression, 5 years after initial diagnosis

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Background: Diffuse midline glioma, H3 K27–altered (DMG), is a high-grade infiltrating glioma found in the midline and showing one of several genomic alterations affecting histone H3, such as H3 p.K28M (K27M). Though loss of canonical mutations has been documented on progression of other forms of glioma, we are unaware of any cases of DMG that have shown loss of their diagnostic alteration following treatment.

Methods: Here we present a case of DMG that developed a tumor component exhibiting loss of the H3 K27M mutation by immunohistochemistry and sequencing.

Results: Our patient was originally diagnosed at age 25 with a left thalamic diffuse midline glioma (H3 K27M mutant). The K27M mutation was demonstrated by immunohistochemistry and sequencing. She was subsequently treated with surgery and chemoradiation. Material from a debulking procedure 6 years later demonstrated a high-grade glioma with astrocytic morphology, marked nuclear atypia, frequent mitoses, necrosis, and proliferative vasculature. Immunohistochemistry showed two distinct regions of tumor that were focally admixed. One portion was H3 K27M negative with retained H3 K27me3 expression, and another portion was H3 K27M positive with reduced H3 K27me3 expression. Molecular testing was performed, focusing on the tumor with loss of H3 K27M immunoreactivity. A chromosomal microarray displayed numerous, complex copy number aberrations, including a few regional losses very similar to those identified in testing of prior samples from the patient's tumor. A next-generation sequencing panel (covering H3-3A) did not reveal an H3 K27M mutation; it did detect pathogenic mutations in TP53, POLE, and PTPN11. Methylation profiling placed the tumor in the methylation class "glioblastoma, IDH-wildtype" with a calibrated score of 0.75 (Heidelberg Epignostix classifier v12.8).

Conclusions: The findings were in keeping with tumor progression of the diffuse midline glioma associated with loss of the diagnostic alteration. To our knowledge, this is the first such instance to be reported.

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Diffuse Midline Glioma with Concurrent MAPK Pathway Alterations: Report of Two Cases

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Background: Diffuse midline glioma (DMG), H3K27M-altered, is an aggressive pediatric brain tumor with poor prognosis and limited response to standard therapy. While MAPK pathway alterations are characteristic of low-grade gliomas, they are uncommon in DMG and may carry prognostic and therapeutic significance.

Methods: We report two pediatric cases of diffuse midline glioma harboring H3K27M mutations with additional MAPK pathway alterations. Clinical history, radiologic findings, histopathology, immunohistochemistry, next-generation sequencing, and DNA methylation profiling were reviewed.

Results: Case 1 involved a 12-year-old girl with a progressive pineal region tumor showing high-grade glial histology, including brisk mitotic activity, microvascular proliferation, and necrosis. Molecular analysis identified H3-3A p.K28M with concurrent FGFR1 and NF1 nonsense variants, consistent with DMG, CNS WHO grade 4. The disease progressed despite therapy, with metastatic spread. Case 2 involved a 15-year-old girl with a lateral ventricular tumor demonstrating circumscribed growth, low proliferative index, and PXA-like features. Molecular studies revealed co-occurring H3K27M and BRAFV600E mutations, and DNA methylation profiling classified the tumor as DMG. The patient remains clinically stable on systemic therapy with radiographic improvement.

Conclusions: These cases highlight a subset of diffuse midline gliomas with concurrent MAPK pathway alterations and atypical histologic features. Emerging evidence suggests these tumors may represent a biologically distinct group with potentially improved prognosis and expanded therapeutic options. Comprehensive molecular profiling is essential for identifying these variants and guiding management.

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Spatial Transcriptomic Analysis of Pediatric High-Grade Gliomas

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Background: Pediatric high-grade gliomas are less common compared to their adult counterparts and display distinct pathogenesis and outcomes. Spatial transcriptomic analyses of both tumor tissue and uninvolved brain parenchyma in pediatric high-grade glioma may uncover patterns that are unique relative to more common adult high-grade gliomas.

Methods: We performed Visium spatial RNA sequencing analyses on four cases of pediatric/adolescent high-grade gliomas from patients aged 10, 11, 12, and 17 years of age. At tissue collection, all four cases were diagnosed as “glioblastoma multiforme” according to the WHO classification criteria for CNS tumors in use at the time. Three tumors were recurrent after treatment while one was a primary diagnosis. All four tumors had evidence of microvascular proliferation. Molecular characterization at the time of diagnosis was limited.

Results: Thirteen of fourteen Visium runs on the four tumors achieved adequate RNA transcript numbers for further analysis. We merged the four tumor samples, performed batch-correction, and performed manual cell typing to identify major cell types which included tumor, astrocytes, oligodendrocytes, neurons, endothelial cells, myeloid cells, fibroblasts, red blood cells, and smooth muscle cells. Cell type composition varied significantly among the four tumors but was consistent across technical replicates.

Conclusions: Our study demonstrates the feasibility of using Visium spatial RNA sequencing to profile rare tumors. A key limitation is that each sequencing unit represents a 55-micrometer diameter spot containing between one and ten cells, rather than individual cells. Future analyses will apply deconvolution techniques to estimate the proportion of cell types within each spot.

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Frontal lobe pediatric high-grade glioma with eosinophilic granular bodies and PMS2 mutation

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Background: Pediatric high-grade gliomas encompass a heterogeneous group of tumors characterized by aggressive behavior. We report a central nervous system pediatric high-grade glioma, H3-wildtype and IDH-wildtype, in a 5-year-old male’s frontal lobe.

Methods: Immunohistochemistry, targeted DNA sequencing and DNA methylation profiling were performed for tumor characterization.

Results: Histologically, the tumor displayed epithelioid/sarcomatoid morphology and multinucleated cells with markedly pleomorphic nuclei and abundant mitotic figures. Notably, diffuse eosinophilic granular bodies and cytoplasmic inclusions were noticeable, presenting an initial intraoperative diagnostic challenge. Immunophenotype was consistent with glial origin of the tumor. Somatic abnormalities detected by targeted DNA sequencing included a TP53 pathogenic variant and CDKN2A/B homozygous deletion. While DNA methylation-based classification provided no match, the profile was indicative of high-grade, IDH- and H3-wildtype glioma. A germline PMS2 pathogenic variant was subsequently identified, implying that the patient suffered from Lynch syndrome.

Conclusions: This case highlights an atypical histological presentation in a pediatric high-grade glioma associated with Lynch syndrome.

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A case of molecularly defined oligodendroglioma with CDKN2A/B homozygous deletion

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Background: Oligodendroglioma, according to the World Health Organization (WHO) 2021 classification, is defined as diffuse glioma with IDH mutation and chromosomal 1p/19q codeletion, with CDKN2A/B homozygous deletion rarely seen. CDKN2A/B homozygous deletion, present in 35% of gliomas, has been established as an independent factor associated with poor prognosis. Here, we report the case of a 63-year-old male who presented with seizures after an unwitnessed fall with approximately 2 months of speech changes, blurry vision, and muscle weakness.

Methods: CT head demonstrated a large region of vasogenic edema in left frontal lobe. The patient underwent image-guided left frontal craniotomy for subtotal resection of large left frontal lobe lesion.

Results: Neuropathological examination demonstrated a diffusely infiltrative neoplasm with mild to moderate cellularity and mild atypia. A majority of the neoplastic cells demonstrate round nuclear contours with prominent perinuclear clearing. Rare mitotic figures and no definitive necrosis were identified. Immunohistochemistry demonstrated the neoplastic cells to be positive for GFAP, OLIG2, and IDH1 (R132H). p16 demonstrated variable staining in a minor subset of cells. The Ki-67 proliferative index was overall approximately 5% with focal elevation to approximately 20%. Fluorescence in-situ hybridization demonstrated a codeletion of chromosomes 1p and 19q in 62% of 200 cells assessed as well as homozygous deletion of CDKN2A/Ip16 present in 91% of 200 cells analyzed.

Conclusions: We have reported a rare entity of oligodendroglioma with CDKN2A/B homozygous deletion. The overall poor clinical outcomes might suggest that the behavior of this particular entity differs from oligodendrogliomas without this deletion.

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An IDH1-Mutant Infiltrative Glioma with MYBL1:MMP16 Fusion and Epigenetic Discordance

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Background: MYBL1 fusions are typically associated with Diffuse astrocytoma, MYB- or MYBL1-altered (CNS WHO grade 1) and are restricted to IDH-wildtype gliomas. Fusions are rare in IDH-mutant tumors, with previously reported partners including MET, BRAF, FGFR, and NTRK. We report an IDH1-mutant infiltrative glioma harboring a MYBL1:MMP16 fusion with atypical molecular and epigenetic features.

Methods: Molecular studies included fusion analysis (Archer), NGS, chromosomal microarray, and DNA methylation profiling.

Results: Histologically, this tumor was diffusely infiltrative and contained scattered calcifications. There was mitotic activity, elevated proliferation (Ki-67 12%), and focal microvascular proliferation. The tumor harbored variants in IDH1 (R132H) and SETD2 p.Y501fs and a MYBL1:MMP16 fusion but lacked TP53, ATRX, or TERT promoter alterations. Chromosomal microarray showed only gain of 8q due to the fusion. DNA methylation profiling yielded a calibrated score < 0.5 for “diffuse glioma, IDH-mutant.” Adjacent white matter lacked fusions and matched control white matter by methylation, supporting reactive gliosis rather than an isomorphic IDH-wildtype MYBL1 precursor lesion.

Conclusions: This infiltrative glioma with concurrent IDH1 mutation, SETD2 frameshift, and MYBL1:MMP16 fusion cannot be classified using current CNS WHO criteria. The tumor lacks canonical molecular alterations of IDH-mutant gliomas, suggesting the MYBL1:MMP16 fusion is likely an oncogenic driver. The unusual molecular and epigenetic profile highlights diagnostic complexity and uncertain clinical behavior.

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PI3K Pathway Mutations are Associated with Worse Overall Survival and Progression-Free Survival in Newly Diagnosed Grade 2 Oligodendroglioma

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Background: Oligodendrogliomas impact over 1,000 patients per year in the United States. Due to their rarity and long survival, there is limited information on the association between genetic biomarkers and survival.

Methods: We identified a cohort of 251 oligodendrogliomas, of which 151 were newly diagnosed grade 2 oligodendrogliomas meeting current WHO molecular criteria. We performed whole exome sequencing. For the most common genetically altered pathways, we examined in univariate analyses their association with progression-free survival (PFS) and overall survival (OS).

Results: Within the cohort of newly diagnosed grade 2 oligodendrogliomas, the median PFS was 5.8 years (95%CI: 5.2, 7.3), and the median OS was 20.3 years (95% CI: 16.3, 24.3) The most common genetic alterations beyond IDH1/2 and TERT promoter were CIC, FUBP1, NF1, NOTCH1, ATM, SWI-SNF complex mutations, PI3K mutations, and histone methyltransferase mutations. Of these, on univariate analysis, PI3K kinase mutations (defined as mutations in PIK3CA or PIK3R1) showed a shorter median PFS of 2.7 vs 6.5 years (p=0.0040) and shortened median OS of 16.3 vs 21.7 years (p=0.0229). This was the only genetic alteration to show poorer PFS and OS. Within the full cohort (all grades), 22% (43/195) of newly diagnosed oligodendrogliomas had PI3K mutations, while 34% (19/56) of recurrent cases had PI3K mutations.

Conclusions: PI3K pathway mutations are associated with shortened PFS and OS in newly diagnosed grade 2 oligodendrogliomas. PI3K pathway mutations were also more common in recurrent oligodendrogliomas compared to newly diagnosed oligodendrogliomas. Taken together, these findings suggest a possible role for PI3K mutations in disease progression and more aggressive behavior in oligodendrogliomas.

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Adult Brainstem Oligodendroglioma Presenting with Severe Motor and Bulbar Dysfunction: A Rare Case Report

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Background: Adult brainstem oligodendrogliomas are exceptionally rare, with only isolated, exceptional cases described in the literature. Most oligodendrogliomas occur supratentorially, and the brainstem location creates substantial diagnostic and therapeutic challenges due to proximity to critical neuroanatomical structures.

Methods: We present a 51 year old man with a two year history of progressive right sided weakness resulting in wheelchair dependence, accompanied by radiculopathy and approximately 90 pounds of unintentional weight loss. His condition further declined with new onset dysphagia and the sensation of food becoming lodged, prompting urgent evaluation for possible stroke. MRI demonstrated a 4.5 cm T2 hyperintense mass centered in the inferior pons and medulla. Stereotactic biopsy performed two weeks later revealed a WHO Grade 2 oligodendroglioma, harboring an IDH mutation, 1p/19q codeletion, TERT promoter mutation, and low level MGMT promoter methylation.

Results: Oligodendrogliomas represent 5–15% of adult glial neoplasms and typically arise in the cerebral cortex, where maximal safe resection followed by chemoradiation provides favorable long term outcomes. However, brainstem involvement is extraordinarily uncommon in adults, with only rare cases reported. In this patient, the lesion was deemed unresectable due to its involvement of vital corticospinal and bulbar pathways. Despite standard chemoradiation, no meaningful reduction in tumor size occurred. Clinically, the tumor's location correlated with severe hemiplegia, dysphagia, repeated hospital readmissions, infectious complications, and minimal improvement in rehabilitation efforts four months after diagnosis.

Conclusions: This case highlights the profound morbidity associated with adult brainstem oligodendroglioma and underscores the need for heightened diagnostic awareness. Although the tumor met molecular criteria associated with favorable prognosis, its brainstem location significantly limited treatment options and resulted in substantial functional decline. Greater recognition of such rare presentations is essential for guiding diagnostic suspicion, counseling, and management strategies.

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Metastatic anaplastic oligodendroglioma in the bone marrow: A Case Report

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Background: Background: A 52-year-old male had a history of anaplastic oligodendroglioma left frontal lobe with subtotal resection 12/30/21, followed by radiation and Temodar treatment. In April 2025, Patient had new onset of anemia and thrombocytopenia. A bone marrow relapsed disease was suspected. It has been known that the most frequent sources of metastatic malignant neoplasm in the bone marrow are breast cancer, prostate cancer, lung cancer and gastric cancer. Bone marrow metastases of anaplastic oligodendroglioma are extremely rare.

Methods: Methods: A bone marrow core biopsy from right posterior superior iliac spine was performed. Bone marrow biopsy reviewed a sheet of tumor cells with frequent mitoses. Immunohistochemical studies showed that the tumor cells are diffusely positive for GFAP, SOX10, S100, IDH1, ATRX, and Olig2; negative for P16, Melan-A, and HMB45. Second-generation of sequencing (NGS) was performed at Caris life science.

Results: Results: Caris-NGS molecular studies showed that there are mutations of IDH1 (p.R132H) and TERT promoter (c.-124C>T); along with co-deletion of 1P/19Q. The morphological and molecular phenotypes are the same as prior anaplastic oligodendroglioma of the initial left frontal tumor.

Conclusions: Conclusions: We report a metastatic oligodendroglioma into the bone marrow. It has been reported that the most common metastatic brain tumors are glioblastoma, medulloblastoma, and ependymoma; but not anaplastic oligodendroglioma. The possible explanation for the rarity of the extracranial metastases from malignant glioma are the absence of lymphatic system in the brain and the existence of blood-brain barrier. Given the fact metastatic anaplastic oligodendroglioma in the bone marrow in 4 years after resection, radiation, and Temodar treatment, the mechanism of metastases could be related to prior surgery site and/or tumor biology, including metastatic potential and tropism.

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Evolution of the immune landscape in MAPK-driven glioma

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Background: The MAPK-driven glioma, pleomorphic xanthoastrocytoma (PXA), is an immune-rich lower-grade tumor that can progress to an aggressive disease. While immunotherapeutic strategies may be beneficial in these tumors, it is unknown how the tumor-associated immune response changes with progression.

Methods: To address this question, we profiled a cohort of 38 tumors, including 10 longitudinal pairs using a multiomics approach that included single-nucleus RNA sequencing (snRNA-seq), spatial transcriptomics, and spatial proteomics.

Results: Comparing WHO CNS5 grade 2 and grade 3 tumors, progression was associated with increased tumor cell proliferation and hypoxia. Spatial analyses demonstrated a reorganization of the immune landscape at progression and an association between regions of hypoxia and increased immunosuppressive tumor-associated macrophages/microglia (TAMs). Hypoxic reprogramming of the immune landscape included a predicted increase in vascular-derived TGFB signaling. In addition, alterations in chemokine signaling included decreased CXCL14 expression, implicated in immune cell recruitment, and decreased CXCL16-CXCR6 signaling, important in immune cell recruitment and activation in the brain. These changes led to a global increase in immunosuppressive TAMs with markedly reduced HLA Class II expression and decreased CD8+ and $CD4^+$ T cells.

Conclusions: These data demonstrate evolution of the glioma-immune landscape with PXA progression and suggest increased tumor hypoxia and altered chemokine signaling are key components. Thus, optimal immunotherapeutic strategies for PXA may differ before and after tumor progression.

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High-Grade Astrocytoma with Piloid Features in Neurofibromatosis Type 1 after a Diagnosis of Pilocytic Astrocytoma

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Background: High-grade astrocytoma with piloid features (HGAP) is a methylation-defined astrocytic neoplasm with high-grade histology and morphologic overlap with pilocytic astrocytoma (PA). It is commonly associated with MAPK pathway alterations, homozygous deletion of CDKN2A/B, and has been increasingly recognized in patients with neurofibromatosis type 1 (NF1). The long-term relationship between PA and subsequent HGAP remains unclear..

Methods: Clinical, radiologic, histopathologic, immunohistochemical, cytogenetic, and molecular findings were reviewed. Molecular studies included next-generation sequencing, fluorescence in situ hybridization, and DNA methylation profiling performed on both tumor specimens.

Results: A 36-year-old man with NF1 initially presented at 15 years of age with a right cerebellar tumor that was gross-totally resected and diagnosed as PA. Microscopically, the tumor was a typical PA with a Ki-67 labeling index of 1–2% and no high-grade features. ATRX and H3K27me3 expressions were retained. Subsequent molecular testing detected NF1 mutation; FISH and DNA methylation profiling were unsuccessful. At 36 years of age, he developed gait instability, facial numbness, and dysphagia. A 4 cm infiltrative mass involving the left cerebellopontine angle, cerebellum, and brainstem, was detected and sub-totally resected. Microscopically, the tumor appeared to be a cellular astrocytic neoplasm with piloid features, increased mitotic activity, and a Ki-67 labeling index of 15%. Molecular studies identified mutations in NF1, TP53, PDGFRA, and ATRX, along with homozygous deletion of CDKN2A/B. DNA methylation profiling demonstrated a consensus match to HGAP. After a prolonged intensive care stay due to respiratory and swallowing complications, he died of disease two months postoperatively before any additional treatment was initiated.

Conclusions: This case demonstrates a de-novo development of HGAP without previous radiation therapy for PA in NF1, adding to the debate of malignant progression in PA/anaplastic PA, while also highlighting the diagnostic value of integrated molecular profiling and underscores the importance of long-term surveillance in NF1-associated circumscribed gliomas.

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Pleomorphic Xanthoastrocytoma with MYC Alteration and Mesenchymal Transformation: A Case Report and Review of the Molecular Landscape

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Background: Pleomorphic xanthoastrocytoma (PXA) is a rare astrocytic neoplasm affecting primarily children and young adults. Molecularly, PXAs are characterized by activation of the MAPK pathway, most commonly through BRAF p.V600E mutation, and are often accompanied by CDKN2A/B homozygous deletion. Although traditionally regarded as indolent, PXAs are now known to demonstrate a broad clinical and histologic spectrum, with recurrence rates of 30–40% and frequent malignant transformation, particularly with TERT alterations.

Methods: We report the case of a 44-year-old Caucasian male with a long-standing seizure disorder who was initially diagnosed with CNS WHO grade 3 anaplastic PXA of the left temporal lobe harboring BRAF p.V600E. The patient underwent gross total resection followed by adjuvant radiotherapy, temozolomide, and Tumor Treating Fields, achieving prolonged clinical stability. Over a six-year disease course marked by multiple recurrences and successive lines of therapy—including BRAF/MEK inhibition, antineoplastics, and immunotherapy—the tumor microenvironment demonstrated molecular evolution.

Results: Genomic profiling of the recurrent disease demonstrated persistence of the BRAF p.V600E mutation along with MYC amplification. In contrast, DNA methylation profiling reclassified the tumor from PXA to glioblastoma, IDH-wildtype, mesenchymal subtype. Notably, MYC amplification status had not been assessed in the primary resection.

Conclusions: This case highlights a rare and previously unreported MYC alteration identified in the context of an anaplastic PXA and underscores the potential epigenetic evolution toward a glioblastoma profile following prolonged survival and multimodal therapy. Our findings underscore the importance of integrated histopathologic and molecular diagnostics, including methylation profiling, in recurrent PXAs, and suggest a novel MYC-associated pathway in malignant transformation.

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Posterior Fossa Glioblastoma with REV3L-MET Fusion

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Background: Glioblastoma (GBM), IDH-wildtype, is an aggressive diffusely infiltrating glioma that typically arises within the cerebrum as opposed to the posterior fossa or spinal cord. Many recurrent molecular alterations have been described in GBM, some of which have been included in the CNS WHO 2021 grading criteria (EGFR amplification, TERT promoter mutation, and gain in chromosome 7 with concurrent loss in chromosome 10). While not as common as other molecular alterations in GBM, recurrent fusions such as FGFR3-TACC3 and MET-PTPRZ1 are described.

Methods: An 89-year-old woman with a medical history of lung adenocarcinoma was admitted for failure to thrive and found to have a 3.7cm mass within the posterior fossa concerning for metastatic disease, for which she underwent surgical resection.

Results: Histopathologic evaluation of the resection specimen showed a high-grade diffusely infiltrating glial neoplasm consistent with Glioblastoma, CNS WHO Grade 4. Next-generation sequencing was performed and notable for the presence of REV3L-MET fusion, two frame shift mutations in NF1, and a missense mutation in TP53. DNA methylation array profiling gave a suggestive score for classification of high-grade IDH-wildtype astrocytoma, posterior fossa. While MET-PTPRZ1 fusions are known to occur in glioblastoma and IDH-mutant astrocytoma, fusions involving REV3L have not yet been reported in the literature. REV3L is part of DNA polymerase zeta and is involved in DNA damage repair, the loss of which has been reported in colon, non-small cell lung, and gastric cancers and may increase sensitivity to cisplatin-based chemotherapy.

Conclusions: This case demonstrates the less common occurrence of glioblastoma in the posterior fossa as opposed to the supratentorial brain parenchyma and highlights a novel fusion not before reported in high-grade glioma.

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A Case of Metastatic Pleomorphic Xanthoastrocytoma

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Background: Pleomorphic xanthoastrocytoma (PXA) is a glial neoplasm characterized by the presence of cells with xanthomatous change, bizarre nuclear pleomorphism, and recurrent alterations in BRAF (most commonly BRAF V600E) and loss of CDKN2A/B. PXA may be designated as CNS WHO Grade 2 or 3 depending on the presence of high-grade histologic features including increased mitotic activity, necrosis, and microvascular proliferation. The presence of TERT promoter (p-TERT) mutation in PXA has been described to portend more aggressive behavior in PXA.

Methods: A 32-year-old-man with a medical history of known brain tumor in the right occipital lobe, for which he was undergoing surveillance imaging, presented with a severe headache. MRI demonstrated an intraparenchymal hemorrhage in the region of the lesion for which he underwent resection.

Results: Histopathologic examination of the resection specimen resulted in a diagnosis of Pleomorphic xanthoastrocytoma, CNS WHO Grade 2. Next-generation sequencing showed the presence of CDKN2A/B deletion and p-TERT mutation, for which closer follow-up was recommended. Eight months later MRI showed radiographic evidence of recurrence for which he underwent a second resection with histology showing recurrent PXA with mitotic activity consistent with CNS WHO Grade 3. Repeat NGS detected the presence of NRF1-BRAF fusion and he was subsequently treated with dabrafenib/trametinib and radiation therapy. A year and a half later, he presented with a suboccipital scalp mass which was resected and showed the presence of metastatic PXA. Later that same year, he subsequently developed a mass in the right sternocleiodomastoid which was resected and proven to be metastatic disease.

Conclusions: This case represents a rare instance of metastatic PXA and demonstrates the increased biologic aggressiveness of PXA with p-TERT mutation. The presence of BRAF fusion as opposed to the more commonly reported BRAF V600E raises the question of whether the type of BRAF or MEK pathway alteration present in PXA impacts prognosis.

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Diagnostic challenges and longitudinal genomic analysis of a highly recurrent MN1:BEND2 fusion tumor lacking classical astroblastoma features

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Background: Astroblastoma, MN1-altered form a rare subset of CNS embryonal tumors with a distinct methylation profile. Most tumors occur in pediatric patients and are viewed as clinically favorable, however, the tumor's striking morphologic diversity has been recognized, including the occasional lack of astroblastoma-like features.

Methods: We present the case of a 15-year-old girl who presented 6 years earlier with a large heterogeneous frontal lobe mass with irregular enhancement, focal cystic/necrotic changes and midline shift.

Results: Microscopic analyses displayed extensive morphologic heterogeneity with mixed, yet distinct histologic components. While much of the lesion showed poorly differentiated tumor cells admixed with necrosis and high proliferative rate (Ki-67 15-20%), only a minute focus suggested the presence of perivascular pseudorosettes, aside from areas of desmoplasia and sclerosis. Despite optimal clinical treatment, the tumor recurred three times over the following 6 years to display only aggressive morphology paired with extremely high proliferative activity (Ki67~60%), ultimately metastasizing to the spinal cord and lung. Interestingly, astroblastoma-like features were not identified in any of the re-resections. Molecular analyses of tumor recurrence samples demonstrated stability of the MN1:BEND2 fusion, which was originally identified via FISH and categorized by methylation analysis. However, while the molecular profile overall appeared to remain stable, the most recent resection sample demonstrated additional somatic variants involving BRCA2 and CDK12. Plans were made to trial Dabrafenib + Trametinib given the CDK12 mutation and treatment was begun, however, the patient declined rapidly and passed away 6 days after regimen initiation.

Conclusions: The degree of diversity within the entity MN1-altered Astroblastoma poses significant challenges not only for the diagnostic process but also for treatment, with the literature generally supporting good outcomes. It will be crucial to carefully analyze and dissect the spectrum of this tumor as its recognition will be of critical importance for subsequent efficient neuropathological workups, molecular characterization and treatment.

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Unusual Suprasellar Lesion presenting with a Complex Clinical Picture: Chordoid Glioma of the Third Ventricle - Case Report

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Background: In this case, we describe the difficult clinical course and complex diagnosis of a suprasellar lesion. A 44-year-old man presented to the emergency department with sudden inability to talk, ambulate, or follow commands. Based on his symptoms, a 'stroke code' protocol was initiated with a head CT revealing a hyperdense, suprasellar lesion. MRI was performed to further characterize the lesion, demonstrating a contrast enhancing 2.1cm suprasellar mass distinct from the pituitary and infundibulum.

Methods: A right-sided craniotomy for biopsy of the lesion was performed, but complicated by an ACA bleed requiring clipping of the A1 segment, which resulted in an infarct of the recurrent artery of Heubner. Given the difficult vascular anatomy, further debulking was not pursued during the craniotomy.

Results: Initial microscopic examination of the lesion showed clusters and cords of epithelioid appearing cells within a mucinous background. Immunohistochemical staining demonstrated GFAP, TTF1, and CD34 positivity. High-grade features such as necrosis or vascular proliferation were not observed. The proliferative index was correspondingly low with a KI-67 estimated at < 2%, and P53 was seen in 5-10% of tumor cells. Focal positivity of EMA and S100 was observed. PanCK and IDH1 (R132H) were negative. Whole-exome sequencing demonstrated a D463H mutation in PRKCA, a mutation consistently seen in chordoid gliomas. Interestingly, methylome profiling did not cluster with any known tumor entity. Based on the diagnosis, decision was made to not pursue further invasive-intervention and to monitor the tumor with intermittent imaging. Within 2 months of the initial presentation, the patient returned to work, as well as his hobby of martial arts. No residual neurologic deficit was identified on a 2 month follow-up visit.

Conclusions: This demonstrates the importance of early diagnosis of the tumor to distinguish it from higher-grade gliomas in order to properly weigh the risks and benefits of aggressive intervention.

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Gliomas with FGFR3::TACC3 fusion: case series and review

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Background: The FGFR3::TACC3 fusion abnormality in glial neoplasms is a relatively recent finding, with reports suggesting that some patients may have a better prognosis than others with IDH–wild type gliomas. However, clinicopathologic and imaging characteristics remain incompletely defined. We reviewed the literature and analyzed a small institutional case series of our own.

Methods: A retrospective evaluation of H&E and immunostained sections, molecular testing results, imaging, and clinical data was performed. Four patients met inclusion criteria, each with confirmed FGFR3::TACC3 fusion identified by next-generation sequencing (NGS).

Results: The cohort included two males (ages 66 & 71 yrs) and two females (ages 12 & 55 yrs). Tumors primarily involved the temporal lobes, except in the oldest patient, where the location was frontal. Three adult patients had high-grade gliomas morphologically resembling glioblastoma, supported by MRI findings that also showed prominent hemorrhage, necrosis and irregular rim enhancement. The youngest patient had a low- to intermediate-grade glioma with concordant imaging features. The tumors were IDH wild-type, microsatellite stable with low tumor mutational burden and detectable PD-L1 expression. Detailed morphologic reassessment included architectural patterns, microvasculopathy, pleomorphism, perinuclear clearing, spindle or myxoid change, inclusions, and immunoprofiles. No consistent or distinctive morphologic pattern was identified across cases. Interestingly, the low-grade glioma demonstrated focal “tufted” CD34 immunostaining and a subtle associated cortical dysplasia. Correlation with literature showed partial agreement with previously described features, though findings were not uniformly reproducible.

Conclusions: This study assessed whether gliomas that harbor FGFR3::TACC3 fusion demonstrate distinct clinical, imaging, or morphologic characteristics that correlate with their interesting molecular profiles. While isolated features aligned with prior reports, no consistently defining pattern emerged in this series, and some heterogeneity is noted. Given the rarity of these tumors, further investigation in a larger cohort is warranted for a better understanding. Final conclusions, therefore, await additional work-up and expanded analysis.

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Molecular characterization of glial/glioneuronal tumors with FGFR3::TACC3 fusion: a case series.

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Background: Glial/glioneuronal tumors with FGFR3::TACC3 fusion ("F3T3 gliomas") represent a heterogeneous group of CNS neoplasms with variable histological grades, including Glioblastoma, IDH-wildtype (GBM). Low-grade pediatric tumors form an epigenetically diverse group that frequently cannot be assigned to a defined WHO tumor type.

Methods: We retrospectively identified F3T3 gliomas detected by targeted-NGS (Oncomine Comprehensive Assay) at Hospital La Paz (Madrid, Spain) and performed DNA methylation profiling (DNAMP) with Infinium MethylationEPIC v2.0 (Illumina), for further characterization.

Results: Seven cases diagnosed between 2015 and 2025 were included. All patients were female, with a median age of 48 years (range 14–57). Tumors were located in the cerebral hemispheres (n = 5), corpus callosum (n = 1), and spinal cord (n = 1). According to the CNS5 WHO histomolecular criteria, three cases were initially diagnosed as GBM, in patients older than 40 years. The remaining four cases were originally classified as low-grade glial/glioneuronal tumor (LGGNT), NOS (n = 3; ages 14, 22, and 57 years) and Polymorphous low-grade neuroepithelial tumour of the young (n = 1; age 18 years). DNA methylation analysis with Heidelberg and/or Bethesda classifier revealed highest scores (< 0.9; no match) for the Glioblastoma, IDH-wildtype, RTK2 subtype in all cases with diagnosis of GBM. In contrast, AYA patients with a prior diagnosis of LGGNT showed highest scores for Dysembryoplastic neuroepithelial tumour, Rosette-forming glioneuronal tumour, and Papillary glioneuronal tumor Methylation Classes. Notably, the adult patient with LGGNT diagnosis showed a high score for Glioblastoma, IDH-wildtype, RTK2 subtype.

Conclusions: At the last follow-up, only the patients with a pre-methylation diagnosis of LGGNT remained alive. As predicted by the methylation profile consistent with GBM, the adult patient is experiencing disease progression. Our study reaffirms the heterogeneity of F3T3 gliomas and highlights how DNAMP can help predict a worse prognosis in borderline cases.

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Clinicopathological and Molecular Characteristics of IDH-Wildtype Glioblastoma with FGFR3::TACC3 Fusion

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Background: FGFR3::TACC3 fusion in IDH-wildtype glioblastoma is a rare (< 5%) molecular event that offers slightly better survival probabilities and multiple potential therapeutic opportunities to patients including FGFR inhibitors and TACC3 inhibitors, among others.

Methods: We present a single institution case series to describe clinicopathological characteristics and targeted sequencing results of IDH-wildtype glioblastomas harboring an FGFR3::TACC3 fusion.

Results: Twenty-four patients were identified (50% females), ages ranging from 40 to 71 years. Tumors were predominantly in the frontal lobe, with rim-enhancing or infiltrative radiologic appearance. Tumor morphology included endocrinoid capillary networks, round oligodendroglia-like cell morphology, and variable scattered nuclear pleomorphism. Eight tumors did not exhibit high grade features in the available material. Intratumoral microcalcifications were found in a subgroup (33%) and did not correlate with the presence of low-grade features. One case showed focal sarcomatoid morphology. The most common diagnostically relevant molecular alteration was TERT promoter mutation (79%), followed by gain of chromosome 7 and loss of chromosome 10 (33%). Consistent with literature, EGFR amplification was not identified. Targeted sequencing also demonstrated other relevant alterations including homozygous loss of CDKN2A/2B (16%) and CDK4/MDM2 amplification (13%). FGFR3 gene amplification or mutations were also present (13%). Patients received concurrent chemoradiation and adjuvant temozolomide. FGFR or tyrosine kinase inhibitors were additionally offered to six patients who pursuit treatment at this institution (6/19).

Conclusions: IDH-wildtype glioblastomas harboring an FGFR3::TACC3 fusion show distinct histological characteristics, with microcalcifications being less frequent than prior reports. Tumors lacking histologic high-grade features are relatively common; however, diagnostically relevant molecular alterations like TERT promoter mutations and gain chromosome 7 and loss of chromosome 10 help render the diagnosis.

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Utility of FGFR3 Immunohistochemistry to determine FGFR alterations in adult diffuse gliomas

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Background: FGFR3 alterations, including FGFR3:TACC3 fusions, occur in a subset of gliomas and some glioneuronal tumors. FGFR3 immunohistochemistry is routinely used in many laboratories, including ours, as a part of a diagnostic glioma panel. Although positive staining strongly suggests FGFR3 alterations, patterns are variable. FGFR3 staining patterns significance in adult diffuse gliomas remains unclear. We aimed to analyze this antibody's practical utility in a glioma panel.

Methods: We retrospectively reviewed adult diffuse gliomas with FGFR3 staining at our institution (2022-2025). Cases with any FGFR3 immunopositivity (SOURCE: dilution) and available NGS were included. Staining was categorized (diffuse strong or weak, focal strong or weak) and correlated with NGS. Among 371 gliomas, 32 were FGFR3-positive; four were excluded after pathology review.

Results: 28 patients (17 males, 11 females) had a mean age of 56.8 years (range 26-72). Diagnoses comprised 23 glioblastomas, 3 oligodendrogliomas, 1 astrocytoma, and 1 diffused midline glioma. FGFR3 staining patterns were diffuse strong in 12 cases, diffuse weak in 13, focal weak in 2, and focal strong in 1. NGS identified 26 TERT, 16 EGFR, 11 FGFR3, and 6 PTEN unique alterations, among 91 others. The 11 FGFR3 alterations corresponded to 8 cases possessing FGFR3:TACC3 fusions with three having additional alterations (two p.K650E and one p.V555M). All 8 FGFR3 fusions demonstrated diffuse strong FGFR3 staining by immunohistochemistry. Four cases with diffuse strong staining lacked detectable FGFR3 alterations. Diffuse strong staining showed a positive predictive value of 66.7% and a negative predictive value of 100% for detection of FGFR3 alterations in this cohort.

Conclusions: Diffuse strong FGFR3 immunoreactivity strongly correlates with FGFR3 alterations, particularly FGFR3:TACC3 fusions. However, false positive staining can be seen in glioblastomas with canonical molecular alterations. Only diffuse strong staining should serve as a robust indicator of pathogenic FGFR3 alterations.

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ROS1-fused high grade gliomas in adults: Genetic, Epigenetic, Immunohistochemical, Radiologic features and clinicopathological correlation

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Background: ROS1 is a member of the receptor tyrosine kinase family and a proto-oncogene that plays an important role in tumorigenesis. ROS1 rearrangements have been detected across different tumor types and often serve as drivers in infantile and pediatric gliomas as well as adult high-grade gliomas. Multiple fusion partners for ROS1 have been described in the literature, the most common being GOPC. ROS1 alterations present unique opportunities for additional targeted therapy to potentially improve outcomes, with ROS1 inhibitors already being used in select patients harboring ROS1-fused tumors.

Methods: We present a retrospective study examining the genetic, epigenetic, Immunohistochemical, and radiologic features of eleven adult high-grade gliomas with ROS1 fusions diagnosed at our institution. DNA methylation analysis was performed on select cases.

Results: The most common fusion partner for ROS1 was GOPC, present in eight of eleven cases (73%). In one case, a ROS1 fusion was not detected until after treatment and recurrence. Histologically, all cases were high-grade gliomas, and frequently demonstrated primitive histologic features; one case had a prominent sarcomatoid component. Molecular features of glioblastoma, +7/-10 and TERT promoter mutations were detected in 67% and 75% of cases tested, respectively. No IDH variants were identified. However, some tumors lacked the classic molecular features of glioblastoma. Methylation profiling data matched one case to Diffuse pediatric-type high grade glioma, RTK1 subtype and another to Glioblastoma, primitive neuronal component. The average survival time was approximately nine months, with GOPC::ROS1 fused tumors having a shorter average survival time (six months) compared to other fusion partners (fourteen months). Two patients received ROS1-targeted therapy with unclear benefit.

Conclusions: We present eleven cases of ROS1-fused gliomas. These tumors frequently presented with challenging histological and molecular features. Although some may be considered glioblastoma, methylation profiling matched the tumors to less common, more primitive epigenetic classes.

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CNS tumor with EP300::BCOR Fusion: Diagnostic Challenges and Long-term Clinical Course

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Background: High-grade neuroepithelial tumors with BCOR fusions are a newly recognized group of aggressive gliomas. While distinct from the CNS WHO tumor defined by BCOR internal tandem duplications, their clinicopathologic spectrum and the diagnostic challenges they present are still being defined. We present a case of an EP300::BCOR-fused glioma and highlights these challenges.

Methods: Histologic and immunohistochemical analysis was performed on the tumor resection of our patient. Molecular analysis, including targeted next-generation DNA sequencing, RNA whole exome sequencing, and genome-wide DNA methylation profiling were performed using the NCI-Bethesda classifier.

Results: A 26-year-old woman presented with a 4-cm right occipital mass that was resected. The tumor was diagnosed as a high-grade infiltrating glioma, IDH-wildtype, with only a BRCA1 variant (germline) identified. The patient was then treated with multiple systemic therapies and three additional resections for tumor recurrence over the subsequent 7 years. No other molecular alterations were identified from subsequent resections. The most recent resection demonstrated an infiltrating hypercellular tumor with areas of ependymal-like perivascular rosetting, palisading and geographic necrosis, and high mitotic activity. After the NCI-Bethesda DNA methylation-based classifier indicated a match to the methylation class CNS tumor with BCOR/BCORL1 fusion, RNA whole exome sequencing was performed that demonstrated a pathogenic EP300::BCOR fusion. The previous fusion panel results were negative due to the lack of the BCOR exon 4 coverage. The patient was treated with carboplatin, but unfortunately developed widespread parenchymal, intraventricular, and leptomeningeal disease.

Conclusions: CNS tumors with EP300::BCOR fusions are aggressive gliomas that can follow a prolonged clinical course. Their diagnosis presents a significant challenge, as misleading histologic features can obscure their nature and they may be missed by targeted DNA/RNA sequencing panels with incomplete gene coverage. This case underscores that broad molecular profiling is essential for the accurate and timely diagnosis of this aggressive entity.

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Multi-Omics Integration Identifies Cooperative Regulatory Networks in ZFTA–RELA Supratentorial Ependymoma

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Background: Background: Fusion-positive supratentorial ependymoma (ST-EPN) is characterized by complex regulatory alterations that are not fully understood. While individual molecular layers have been explored, the hierarchical relationship between epigenetic modulation, miRNA dysregulation, and downstream transcriptomic changes remains unclear. We applied a miRNA-centric integrative multi-omics approach to delineate the regulatory landscape of fusion-positive ST-EPN.

Methods: Methods: Small RNA sequencing, mRNA sequencing, and Illumina EPIC v2 DNA methylation profiling were performed on fusion-positive ST-EPN (n=5) and normal ventricular lining controls (n=3). miRNA and mRNA differential expression analyses were conducted using DESeq2 following standard preprocessing and alignment workflows. Differentially methylated regions (DMRs) were identified using the minfi–limma pipeline with stringent probe filtering. Experimentally validated miRNA seed interactions (5mer/6mer/7mer) were integrated with 3′UTR variant context. Functional enrichment and protein–protein interaction networks were generated using clusterProfiler, STRING, and Cytoscape, and network topology metrics were used to identify hub and bottleneck nodes. Oncogenic relevance was assessed using COSMIC, IntOGen, OncoKB, and OnGenes. Selected candidates underwent qRT-PCR validation.

Results: Results: Integrated multi-omics analysis of supratentorial ependymoma (ST-EPN) demonstrated widespread molecular disruption. Whole-exome sequencing identified 140 prioritized somatic variants across 111 genes, including 13 likely pathogenic alterations enriched in transcriptional and NF-κB–associated regulators. Small RNA sequencing revealed marked miRNA dysregulation (237 upregulated, 290 downregulated) with 98 high-confidence miRNA–mRNA interaction pairs involving 192 differentially expressed transcripts. RNA-seq identified 978 upregulated and 1,353 downregulated genes, refining to 32 high-confidence hub genes. Notably, miR-124-3p was upregulated, whereas miR-138b-5p and miR-10b-5p were consistently downregulated.

Conclusions: Conclusions: Our miRNA-centric multi-omics analysis indicates that epigenetically influenced miRNA dysregulation contributes to transcriptomic network remodeling in fusion-positive ST-EPN. Despite the exploratory sample size, this integrative framework provides systems-level insight into ependymoma biology and nominates candidate regulatory vulnerabilities for further validation.

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A case of myxopapillary ependymoma arising in a patient with SMARCB1-related schwannomatosis

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Background: Ependymomas are defined by their anatomic location and molecular features. In the spinal cord, these are further classified as “spinal ependymoma”, “spinal ependymoma, MYCN-amplified”, “myxopapillary ependymoma”, and “subependymoma” based on distinct DNA methylation patterns. Spinal ependymomas have not been previously described in SMARCB1-related schwannomatosis, but are well known in neurofibromatosis type 2 and typically match to the “spinal ependymoma” class.

Methods: Neuropathologic evaluation included histologic examination and standard validated immunohistochemistry (IHC) protocols. Molecular genetic testing via next generation sequencing analysis was performed at the Precision Medicine Institute of Children's Hospital Colorado. DNA methylation profiling was performed at the National Institutes of Health.

Results: A 45-year-old male with history of germline SMARCB1-related schwannomatosis and multiple cutaneous lesions initially presented with upper back discomfort and arm tingling. MRI demonstrated intradural, extramedullary lesions centered at T4 and T9 as well as numerous nodules in the lumbar and sacral spinal canal which slowly progressed in size over ~7 years. Histologic sections demonstrated a proliferation of cells with hyperchromatic, elongated nuclei and eosinophilic cytoplasm arranged in sheets in a background of microcysts and myxoid material, along with occasional perivascular pseudorosettes. Neoplastic cells were diffusely positive for GFAP and EMA. Staining for INI1 showed loss of expression in tumor cells. DNA methylation profiling showed no match to an established class. Next-generation sequencing demonstrated evidence of a loss/deletion affecting the majority of chromosome 22q, including the SMARCB1 and NF2 gene loci.

Conclusions: Here we describe and characterize a case of an ependymoma arising in the setting of SMARCB1-related schwannomatosis. Histologically, this tumor resembled the myxopapillary subtype, which is not prevalent in patients with type 2 neurofibromatosis and the genetic features are varied. To our knowledge, ependymomas have not been associated with SMARCB1-related schwannomatosis and this case perhaps represents a novel subtype morphologically (but not epigenetically) similar to myxopapillary ependymoma.

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68-year-old male with a spinal ependymoma, MYCN amplified with extra-CNS metastasis: A case report

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Background: Spinal ependymoma, MYCN-amplified is newly recognized in the 5th edition of the WHO Classification of Tumors as a clinically aggressive entity with high-grade histologic features and frequent recurrence and cerebrospinal fluid (CSF) dissemination. Extra-CNS metastasis has not been previously reported in this entity.

Methods: We describe a 68-year-old man presenting with urinary retention and paraplegia. MRI revealed a T2–3 intradural extramedullary solid and cystic enhancing mass with confluent subarachnoid drop metastases at lower spinal levels. Gross total resection was achieved with negative postoperative CSF cytology. Histopathological diagnosis was spinal ependymoma, CNS WHO grade 3. Next-generation sequencing (NGS) identified MYCN amplification with concurrent KEL loss. The patient received adjuvant proton beam radiation. At six months, MRI showed leptomeningeal progression. At one year, follow-up imaging identified a right paraspinal soft tissue mass. At three years, he presented with a pathologic right femoral neck fracture. Imaging revealed multiple pulmonary and hilar masses. Fine-needle aspiration and endobronchial biopsy of a lung lesion and the femoral lesion were performed.

Results: Histopathologic examination of lung and femoral biopsies demonstrated metastatic ependymoma morphologically consistent with the primary tumor. Interphase fluorescence in situ hybridization confirmed MYCN amplification in 97% of tumor cells across the primary and all metastatic sites.

Conclusions: We report a rare case with metastases to lung and bone (biopsy-proven) and hilar lymph node (FNA), with imaging suggestive of leptomeningeal dissemination and paraspinal soft tissue metastasis. Additional molecular alterations identified by NGS are described. This case expands the clinicopathologic spectrum of this recently defined entity.

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A UBN2::BRAF fused low grade glial/glioneuronal tumor with pseudopapillary structures: A Novel Fusion or Partner?

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Background: A 33-year-old male who presented with chronic headaches and seizures was found to have a slightly increased in size 1.9 cm heterogeneously enhancing mass identified in the suprasellar cistern and a stable well-circumscribed T2 FLAIR hyperintense osteolytic lesion arising from the right calvarium. A thoracic (T3-T5) intradural mass was also identified during oncologic staging imaging and was subsequently biopsied.

Methods: A craniotomy with suprasellar biopsy and right sinus resection showed a GFAP and Olig2 positive tumor which appears myxoid with minimal atypia and rare mitoses, and in the osteolytic component was focally pseudopapillary with hyalinized vascular cores, surrounded by GFAP positive nuclei with interpapillary synaptophysin positive neuropil. Rosenthal fibers were focal. The thoracic intradural mass revealed neuropil, dense fibroconnective tissue and scattered monotonous GFAP positive cells with a focally similar pseudopapillary appearance and low Ki67 (< 5%).

Results: Both the intracranial and spinal lesions showed the UBN2::BRAF fusion by RNA sequencing. The UBN2 exon 8 fuses to BRAF exon 9 and is predicted to be inframe and likely a result of an inversion event. Although this fusion is not yet reported in brain tumors, it is well established that BRAF fusions retain exons 9-18 (C-terminal kinase domain) while loss of the N-terminal autoinhibitory domain leads to constitutive activation. BRAF fusions seen in pilocytic astrocytomas are typically partnered with KIAA1549. The histology does not look like pilocytic astrocytoma, however, it is not uncommon for known fusion drivers, like BRAF, to have multiple partners. Ubinuclein 2 (UBN2), located on chr7q34, is involved in chromatin remodeling by interacting with a complex required for cellular senescence. UBN2 has been reported in prostate and colorectal cancer.

Conclusions: While BRAF fusions can occur in pilocytic astrocytomas, the methylation results of “no match,” favoring a low grade glial/glioneuronal tumor, ultimately suggest that this UBN2::BRAF fused tumor is a distinct entity.

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Papillary Glioneuronal Tumor with SLC44A1::PRKCA Fusion and Patchy H3K27me3 Loss in a Pediatric Intraventricular Mass: A Diagnostic Challenge

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Background: Papillary glioneuronal tumor (PGNT) is a rare, low-grade mixed neuronal–glial neoplasm classified as CNS WHO grade 1 and characterized by PRKCA gene fusions, most commonly SLC44A1::PRKCA. Morphologic heterogeneity and overlapping features with ependymoma and other glioneuronal tumors may lead to diagnostic difficulty, particularly in limited biopsy specimens.

Methods: We report a case of a 9-year-old male who presented with progressive lethargy, vomiting, headache, neck pain, and seizure-like activity. Neuroimaging demonstrated a lobulated right lateral ventricular mass with additional lesions in the third ventricle and inferior to the cerebellar tonsils, associated with obstructive hydrocephalus requiring ventriculostomy. Initial frozen section and permanent biopsy interpretation favored ependymoma versus centroneurocytoma due to sheets of uniform round cells, focal perivascular arrangement, hemorrhage, rare mitoses, and absence of necrosis or microvascular proliferation. Immunohistochemistry showed GFAP and S100 positivity, low proliferative index (Ki-67 ~1–2%), and absence of dot-like EMA staining.

Results: Subsequent molecular testing by RNA sequencing identified an SLC44A1::PRKCA fusion, establishing the diagnosis of papillary glioneuronal tumor. Resection specimens demonstrated a compact low-grade glioneuronal neoplasm with round cell morphology and perivascular condensation without significant mitotic activity. Additional studies revealed OLIG2, GFAP, and S100 expression, with an unexpected patchy loss of H3K27 trimethylation by immunohistochemistry. The integrated diagnosis was papillary glioneuronal tumor, CNS WHO grade 1. External neuropathology consultation concurred with the final classification.

Conclusions: This case highlights the diagnostic challenges of PGNT in pediatric intraventricular presentations and underscores the critical role of molecular testing in distinguishing PGNT from morphologic mimics such as ependymoma. The presence of patchy H3K27me3 loss represents an unusual ancillary finding with uncertain biological significance. Recognition of PRKCA fusion–positive PGNT is essential for accurate classification and prognostication, as gross total resection is typically associated with favorable outcomes.

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Low-grade glial/glioneuronal neoplasm with QKI::RAF1 fusion

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Background: QKI::RAF1 fusion in CNS tumors is rare and has been described in a few low grade glial/glioneuronal neoplasms without description of their morphologic features (PMID 23583981) and in one pleomorphic xanthoastrocytoma (PMID 30517658).

Methods: A 10-year-old girl with 1.5-year history of left lower back pain and 1-year history of altered gait, presented with 1 week history of urinary retention. MRI showed an intramedullary tumor extending from T10-T11 level through T12-L1 level associated with a syrinx extending from mid T2 level through the conus medullaris and vasogenic edema cranial to the syrinx. Gross total resection of the tumor was performed and the patient showed gradual functional improvement post-surgery.

Results: Histopathologic examination showed a low-grade glial/glioneuronal neoplasm with variable morphology. Some areas showed bland glial cells in abundant fibrillary and focally edematous matrix. A few areas showed denser fibrillary matrix. Abundant Rosenthal fibers were present and eosinophilic granular bodies were noted. Other areas showed nodular architecture comprising scattered blood vessels with prominent perivascular arrangement of neoplastic cells immunoreactive for GFAP and synaptophysin. Mitoses were not conspicuous, but occasional foci of tumor necrosis, fresh hemorrhages, and hemosiderin deposits were noted. Ki67 labeling index was low at 1-3%. Some blood vessels showed hyalinized walls and many showed perivascular lymphocytes. Next-generation sequencing of RNA identified QKI::RAF1 fusion between QKI exon 2 and RAF1 exon 8. A final diagnosis of low-grade glial/glioneuronal neoplasm with QKI::RAF1 fusion, not elsewhere classified was made.

Conclusions: We describe the unusual histopathologic features of a rare low-grade glial/glioneuronal neoplasm with QKI::RAF1 fusion arising in the spinal cord of a child. QKI::RAF1 fusion joins the homodimerization and RNA binding domains of QKI to the kinase domain of RAF1 and has been shown to activate MAPK and PI3K/mTOR signaling pathways, which may be therapeutically targetable (PMID 28806393).

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Dysembryoplastic Neuroepithelial Tumor with FGFR3::MYH7 Fusion: Expanding the Molecular Spectrum of Glioneuronal Tumors

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Background: Dysembryoplastic neuroepithelial tumor (DNT) is a WHO grade 1 glioneuronal tumor commonly associated with drug-resistant epilepsy in children and young adults and typically characterized by alterations in FGFR1. FGFR3 mutations are well described in other CNS tumors but are not well established in DNET. We report a molecularly characterized DNET harboring a FGFR3::MYH7 fusion.

Methods: Histologic evaluation, immunohistochemistry, methylation profiling and next-generation sequencing were performed on a surgically resected cortical lesion from a patient with medically refractory epilepsy.

Results: The patient was a 6-year-old presenting with focal seizures and a 1.6 cm T2 hyperintense lesion of the left parietal convexity on MRI. Histologic examination demonstrated a nodular cortical glioneuronal tumor with the “specific glioneuronal element” and floating neurons in a myxoid stroma. The neoplastic cells expressed Olig2 and S100 and were negative for GFAP, synaptophysin, CD34, NeuN, and the IDH1 R132H mutation. The Ki-67 proliferation rate was approximately 1%. The morphology and immunophenotype were classic for dysembryoplastic neuroepithelial tumor (DNT). Whole exome sequencing identified an FGFR3::MYH7 fusion, with no FGFR1, BRAF, or IDH alterations detected. DNA methylation profiling performed at the National Institutes of Health, using both the Heidelberg classifier versions 11b6 and 12b6 and the NCI/Bethesda classifier, classified the tumor as a “Dysembryoplastic neuroepithelial tumor” with high confidence.

Conclusions: This case expands the molecular spectrum of DNT and suggests that FGFR3 alterations may represent an alternative pathway of MAPK/FGFR signaling activation in glioneuronal tumors.

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KRAS-Variant Pilocytic Astrocytomas: A Two-Case Series Expanding MAPK Alterations
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Background: Pilocytic astrocytoma (PA), CNS WHO grade 1, is a circumscribed astrocytic glioma characterized by activation of the MAPK signaling pathway, most commonly via KIAA1549–BRAF fusion or BRAF V600E mutation. KRAS variants are rare in PA, and their clinicopathologic significance remains incompletely defined. Here we report two cases of pilocytic astrocytoma harboring pathogenic KRAS exon 3 variants.

Methods: Histologic evaluation, immunohistochemistry, and next-generation sequencing of two cases were performed to identify this entity.

Results: Case 1: The patient was a 9-year-old female, presenting with hydrocephalus, papilledema, nausea, vomiting, and headache. MRI brain revealed a 2.9 cm 3rd ventricle/suprasellar mass with diffusion restriction. Histologic evaluation showed a circumscribed, low-grade glial neoplasm with piloid features, a myxoid background, and arc-like microvascular proliferation. Foci of necrosis without palisading were present. The Ki-67 proliferation index was low. Molecular profiling revealed concurrent FGFR1 p.N546K and KRAS p.Q61H pathogenic variants. Case 2: The patient was a 36-year-old male with recurrent pilocytic astrocytoma, status post resections (2003 and 2005), with recurrence in 2024. MRI brain revealed A 3.2 cm, multiloculated mass centered in the left atrium with cystic components extending into the right atrium. Histologic evaluation showed classic PA morphology, including myxoid stroma, Rosenthal fibers, and eosinophilic granular bodies. The Ki-67 index was 0-1%. Molecular testing identified a KRAS p.Q61K pathogenic variant. Both tumors lacked IDH mutations and demonstrated retained ATRX expression.

Conclusions: These cases expand the spectrum of MAPK pathway alterations in pilocytic astrocytoma and highlight rare activating KRAS codon 61 variants in CNS WHO grade 1 tumors. Recognition of KRAS-driven PA is diagnostically important, particularly in the absence of BRAF alterations, and may have implications for targeted KRAS inhibition.

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Multifocal Diffuse Glioneuronal Tumor with Oligodendroglioma-Like Features and Nuclear Clusters (DGONC) in a Pediatric Patient: A Case Report

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Background: Diffuse glioneuronal tumor with oligodendroglioma-like features and nuclear clusters (DGONC) is a recently recognized, rare central nervous system neoplasm defined by a characteristic DNA methylation profile and mixed glial–neuronal differentiation. These tumors typically arise in the supratentorial cortex of children and young adults. Their biological behavior, grading parameters, and optimal management remain incompletely understood.

Methods: A 5-year-old female presented with multiple right hemispheric cortical masses involving the perirolandic region, sylvian fissure, middle frontal, and inferior frontal lobes without leptomeningeal dissemination. An initial resection was performed for diagnostic clarification, followed by staged resections of the remaining lesions. Histologic evaluation, immunohistochemistry, DNA methylation profiling (DKFZ v12.8 classifier), and next-generation sequencing were performed.

Results: All lesions demonstrated a diffusely infiltrating glioneuronal neoplasm composed of round-to-ovoid cells with finely speckled chromatin, perinuclear clearing, and characteristic nuclear clustering. Calcifications and foamy macrophages were present. Immunohistochemistry showed Olig2 positivity, patchy NeuN expression, and variable GFAP, S100, and synaptophysin reactivity. DNA methylation profiling yielded a high-confidence match to DGONC, and next-generation sequencing revealed no pathogenic alterations. Despite shared cytomorphology, the lesions demonstrated marked proliferative heterogeneity. The initial tumor showed mitoses up to 4/mm² and focal necrosis, while subsequent lesions exhibited increased cellularity, mitotic activity up to 14/mm², nuclear atypia, and focal microvascular proliferation.

Conclusions: DGONC remains a diagnostically challenging and rare entity in pediatric neuro-oncology. We present a case with unusual distribution and histologic features. While a DGONC case with leptomeningeal involvement has previously been described, the multifocal nature of this patient’s DGONC is unique. In addition, this case shows prominent interlesional histologic heterogeneity, with the tumor masses presenting a spectrum of proliferative activity. While DGONC has been proposed to behave as an indolent or low-grade entity, the presence of proliferative and “anaplastic” features in some foci raises important questions regarding grading, biologic potential, and management strategies.

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Unusual KIAA1549::BRAF fusion and a PIK3CA alteration in DNT

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Background: Dysembryoplastic neuroepithelial tumor (DNT) is a World Health Organization (WHO) grade 1 glioneuronal neoplasm typically affecting children and young adults and commonly associated with drug-resistant epilepsy. Most DNTs harbor activating FGFR1 alterations, while BRAF alterations are less frequent and usually mutually exclusive. We report a pediatric case of DNT with an unusual molecular profile, characterized by a KIAA1549::BRAF fusion and a concurrent PIK3CA alteration, accompanied by late local recurrence.

Methods: N/A

Results: A 6-year-old child presented with treatment-resistant epilepsy and radiologic features suggestive of DNT. Subtotal resection revealed a low-grade neuroepithelial tumor composed predominantly of oligodendroglioma-like cells with focal mucinous areas containing rare floating neurons, consistent with a DNT. Immunohistochemistry demonstrated GFAP positivity, neuronal elements highlighted by neurofilament staining, and a low proliferative index. BRAF p.V600E immunohistochemistry was negative. After seven years of clinical and radiologic stability, local recurrence was identified and resected. The recurrent tumor showed similar low-grade histomorphology with prominent floating neurons. Molecular analysis revealed a KIAA1549::BRAF fusion and a PIK3CA alteration. DNA methylation profiling classified the lesion as a low-grade glial/glioneuronal neoplasm with overlapping features of pilocytic astrocytoma and DNT.

Conclusions: This case expands the molecular spectrum of DNTs and highlights diagnostic challenges posed by atypical molecular alterations and late recurrence.

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Melanotic Differentiation in Pilocytic Astrocytoma: two case reports expanding the Histopathological Spectrum

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Background: Pilocytic astrocytoma is a low-grade tumor predominantly affecting paediatric age group. Melanocytic cells in the CNS are present in the leptomeningeal tissue and are the principal site of pure melanocytic tumors. The presence of melanin has been described widely across central nervous system tumors of neuroepithelial, leptomeningeal, and peripheral nerve origin. However, astrocytic tumors with melanocytic differentiation are rare. Here we describe two cases of pilocytic astrocytoma with focal melanocytic differentiation.

Methods: Hematoxylin and eosin staining and immunohistochemistry for glial markers were performed alongwith FISH for detecting KIAA1549 :: BRAF fusion. MRI was performed for conformation.

Results: Two four year old males, Case#1 presented history of headache, vomiting, lethargy, and imbalance while walking for 25 days with evidence of cerebellar signs. MRI revealed T1 hypointense, T2 hyperintense, solid, and cystic SOL, with solid part superomedial positioned and contrast-enhancing. Case#2 showed proptosis of eye with MRI revealed large intracranial mass along optic nerve. Histopathological analysis revealed focal presence of cells with intracytoplasmic melanin. Immunohistochemistry, showed positivity for GFAP, while negative for HMB-45, IDH-1, and p53. Immunostaining for ATRX was retained. For confirmation of the diagnosis, Case#1 confirmed the presence of KIAA1549 :: BRAF fusion fusion gene by FISH.

Conclusions: Eight cases of astrocytic tumors with melanin deposition have been reported until date, majority being Pleomorphic xanthoastrocytoma (PXA). To our knowledge, only two prior cases of pilocytic astrocytomas with melanotic differentiation are described having focal presence of cells with melanocytes. Studies on mouse model have documented primitive neuroepithelial cells with the potential to produce melanosomal melanin. However, the origin of melanin in these tumors remains unclear. Moreover, it may be confused with other melanocytic lesions on histology, especially in small biopsy specimens. An appropriate diagnostic approach with judicial use of ancillary techniques, including histochemistry, immunohistochemistry, and molecular pathology study, is required for a definitive diagnosis.

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Sarcomatous transformation of an ALK fusion-positive infant-type hemispheric glioma following treatment with lorlatinib

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Background: Infant-type hemispheric glioma (IHG) is a rare, high-grade tumor of early childhood, frequently harboring receptor tyrosine kinase (RTK) fusions involving the NTRK family, ROS1, MET, or ALK. These fusions represent potential therapeutic targets for RTK inhibitors. Therapeutic pressure driving the transformation of RTK fusion-positive lung carcinomas has been reported and may similarly be a challenging mechanism of treatment resistance in tumors of the CNS.

Methods: We report a 2-year-old female patient presenting with vomiting and increasing head circumference found to have a heterogeneously enhancing hemorrhagic 10 cm left frontal lobe mass. Biopsy showed a cellular glial neoplasm with high-grade features including multifocal necrosis and microvascular proliferation. The neoplastic cells were immunoreactive for ALK and sequencing revealed a SPECC1L::ALK fusion, consistent with infant-type hemispheric glioma. The patient underwent gross total resection and began lorlatinib shortly after diagnosis.

Results: Surveillance imaging in the following months identified multiple new and enlarging enhancing foci concerning for disease progression. A second resection was performed five months into treatment with lorlatinib showing rhabdomyosarcomatous features including strap-like cells diffusely positive for myogenin, MyoD1, and desmin. Tumor cells retained positive staining for ALK. These features suggested gliosarcomatous transformation of the recurrent IHG. The patient began a new regimen of irinotecan/temozolomide/lorlatinib but continued to show progressive disease over the next several months. The patient passed away 18 months after initial diagnosis.

Conclusions: Compared to other pediatric high-grade gliomas, RTK fusion-positive IHGs are reported to have relatively better prognoses with transformation to lower grade histology on post-treatment sampling. Many reports have shown favorable outcomes using RTK inhibitors leading to reduction in tumor size even in cases refractory to other chemotherapies. Immunotherapy, while promising new targeted strategies, may cause selective pressures contributing to therapeutic resistance and drive aggressive transformation highlighting the need to integrate biomarker-focused patient selection and the potential for combination therapeutic strategies.

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Neuroepithelial tumor with CHD7::PLAG1 fusion

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Background: A 53 year old female presented following post-stroke imaging noting an incidental left intraventricular mass which showed 2-3mm growth over a 6 month period. MRI showed a heterogeneously enhancing mass in the left lateral ventricle measuring 1.7cm.

Methods: N/A

Results: Sections showed a moderately cellular lesion of monomorphic cells. There were scattered perivascular pseudorosettes. GFAP and EMA (dot-like) were both positive, while H3K27M and H3K27me3 were both wild-type, with a Ki-67 of 8%. This was then sent to NIH for methylation profiling and showed a CHD7::PLAG1 fusion and a consensus classification of neuroepithelial tumor (NET) with CHD7::PLAG1 fusion.

Conclusions: The PLAG (pleomorphic adenoma gene) family (PLAG1, PLAGL1, and PLAGL2) encode zinc finger transcription factors. In the CNS specifically, PLAGL1 amplifications have been identified in supratentorial neuroepithelial tumors and embryonal tumors, with PLAG2 amplifications also being seen in the latter. Recently embryonal tumors with PLAGL1 and PLAGL2 amplifications were noted to have PLAG1 amplifying fusions. These were previously not included within the diagnosis but recurrent fusions that result in promoter hijacking to upregulate the PLAG1 gene were identified, prompting suggestions to modify the diagnostic name. Since our case does not have embryonal features and has EMA positivity (something the embryonal tumors are negative for), it may be useful to modify the classification of supratentorial NET's with PLAGL1 amplifications in a similar way. Also, it is important to note the age of occurrence. For the NET's, the median age at diagnosis was 6.2 with a range from 0-30 years old, and the embryonal tumors have a median age at diagnosis of 5 with a range from 1-11 years old, while our patient is 53 years old. This will be important to keep track of and report in future cases to establish a new age range and assess prognostic features.

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Collision Tumor in the Basal Ganglia: Germinoma Combined with Pediatric-type Diffuse Astrocytoma, MYBL1-altered — A Case Report

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Background: Intracranial collision tumors are rare central nervous system neoplasms characterized by the coexistence of two distinct histological tumor types arising in the same anatomical location, either adjacent to each other or with intermingled growth patterns.

Methods: We present a rare case of intracranial collision tumor composed of two components: germinoma and pediatric-type diffuse astrocytoma, MYBL1-altered.

Results: The patient is a 13-year-old male who presented with progressive weakness of the right extremities. Neuroimaging revealed an occupying lesion in the left basal ganglia. Histopathological examination, combined with immunohistochemical staining and next-generation sequencing, confirmed the presence of dual histological components: the first component exhibited typical germinoma morphology, with tumor cells growing in sheets, medium to large cell size, round or oval nuclei, high nuclear-to-cytoplasmic ratio, and lymphocytic infiltration in the stroma. Immunohistochemistry showed partial positivity for SALL4, OCT3/4, and CD117, with a Ki67 proliferation index of approximately 60%. The second component consisted of pediatric-type diffuse astrocytoma, showing mild glial cell proliferation with uneven distribution in the brain tissue, mild atypia, and no obvious mitotic figures, necrosis, or microvascular proliferation. Immunohistochemistry revealed Olig-2 positivity, GFAP negativity, and a Ki67 proliferation index of approximately 1%. Genetic testing identified a frameshift mutation in exon 12 of the MYBL1 gene (c.1727_1728del, p.S577Pfs*29), which represents the defining molecular alteration for pediatric-type diffuse astrocytoma, MYBL1-altered. Concurrently, germline mutations in multiple DNA repair-related genes including PTCH1, FANCI, and POLD1 were detected, suggesting a potential background of genomic instability.

Conclusions: This case not only reveals the collision of two rare CNS tumor types at the same anatomical site but also represents the first report of coexisting germinoma and MYBL1-altered pediatric-type diffuse astrocytoma, providing new insights into the pathogenesis of intracranial collision tumors.

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Diagnostic Yield of Molecular Testing in Pediatric Central Nervous System Tumors Using Whole-Exome Sequencing

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Background: Pediatric central nervous system (CNS) tumors are a leading cause of childhood cancer mortality, yet their genetic landscape is incompletely defined. Next-generation sequencing, especially whole-exome and transcriptome approaches, captures a broad range of pathogenic variants and gene fusions, improving tumor characterization beyond targeted panels.

Methods: Pediatric CNS tumor specimens collected between January 2021 and December 2024 with available Columbia Cancer Whole Exome (CWES) or Columbia Combined Cancer Panel (CCCP) results were reviewed. CWES somatic variants were classified as Tier 1 (actionable), Tier 2 (targeted or well-established gene), Tier 3 (other cancer gene mutations), or Tier 4 (variants of uncertain significance (VUS)). CCCP variants were categorized as Pathogenic or VUS. Germline variants, copy number alterations (CNVs), transcriptomic findings, and Diagnosis Refined by Molecular Profiling (DRMP) were recorded.

Results: Twenty-six samples (18 CWES, 8 CCCP) were included. Mean age was 7.74 years; 50% were male. Pathogenic/Tier 2 mutations were identified in 57.7% of cases (10/18 CWES, 5/8 CCCP). DRMP was seen in 80.8% of cases (15/18 CWES, 6/8 CCCP). Among 18 pediatric CNS tumors using CWES, key findings included SHH-activated medulloblastomas with PTCH1 and TP53 mutation, frequent MYCN/GLI2 overexpression, and complex CNVs; ependymomas with SETD2 mutation and RELA fusion or with subgroup-specific expression profiles; atypical teratoid/rhabdoid tumors with SMARCB1 mutations; H3F3A-mutant diffuse midline glioma; VHL-mutant hemangioblastoma; various embryonal tumor with multilayered rosettes; oligodendrogliomas and neurocytomas, with associated CNVs and overexpression. CCCP demonstrated co-alterations of TP53 and EGFR in H3K27 mutated gliomas; BRAF V600E and SOX10 alterations in a pilocytic astrocytoma with anaplasia, and a PRKAR2A-ALK fusion in an infant-type hemispheric glioma.

Conclusions: Comprehensive molecular testing identified pathogenic or Tier 2 variants in most pediatric CNS tumors. Rare gene fusions and CNVs further contributed to tumor characterization, underscoring the value of broad genomic profiling to refine diagnosis and guide precision medicine.

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Liquid biopsy diagnostics: The NCI/Bethesda CNS classifier reliably classifies cell-free DNA methylation data from cerebrospinal fluid

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Background: DNA methylation–based classification has transformed the molecular diagnosis of central nervous system (CNS) tumors through machine learning–driven approaches. The Heidelberg (v12.8) and NCI/Bethesda (3.1) classifiers are widely implemented for tissue-based tumor classification. Cerebrospinal fluid (CSF) cell-free DNA (cfDNA) represents a minimally invasive alternative for tumor detection and monitoring; however, existing classifiers were developed using tumor tissue and have not been optimized for CSF-derived cfDNA.

Methods: We evaluated the feasibility of applying current methylation classifiers directly to CSF samples and assessed their potential for adaptation to liquid biopsy–based tumor classification. We analyzed primary brain tumor tissues (n = 157) and cerebrospinal fluid (CSF) samples (n = 23) profiled using the Illumina methylation array from a recent study (GSE292312). All CSF samples had matched primary tumor tissues, enabling direct concordance assessment between tissue- and CSF-based classifications. Both sample types were evaluated using the Heidelberg and NCI/Bethesda classifiers. Tissue performance was assessed using the recommended calibrated match score threshold (≥ 0.9). For CSF samples, classification was evaluated based on the highest predicted class probability.

Results: In primary tumor tissues, both classifiers demonstrated strong performance. The Heidelberg classifier showed 78.3% high-confidence matches (123/157; calibrated score ≥ 0.9), while the NCI/Bethesda classifier achieved 88.5% high-confidence matches (139/157; calibrated score ≥ 0.9). When applied to the paired CSF samples, the Heidelberg classifier correctly identified tumor class in 35% of cases (8/23). In contrast, the NCI/Bethesda classifier achieved 70% concordance (16/23), including multiple high-confidence predictions (n=7; ≥ 0.9 probability).

Conclusions: Tumor-specific methylation signals are detectable as well as classifiable from CSF-derived cfDNA. These findings support the feasibility of adapting tissue-based methylation classifiers, particularly the NCI/Bethesda framework, for non-invasive CNS tumor classification using CSF. Further optimization and CSF-specific model development may enhance the clinical utility of liquid biopsy approaches in neuro-oncology.

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A Uniform MGMT Promoter Methylation Analysis of Over 15,000 Central and Peripheral Nervous System Tumors

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Background: MGMT promoter methylation (MGMTpm) is an established biomarker for glioblastoma, IDH-wildtype (GBM), yet its prevalence across the broader spectrum of central/peripheral nervous system (CNS/PNS) tumors remains incompletely characterized.

Methods: We report the frequency of MGMTpm in a wide range of CNS/PNS tumors using DNA methylation array through a standardized analytic framework. Methylation array data were obtained using the Illumina Infinium MethylationEPIC v1.0/2.0 and processed using the NCI/Bethesda classifier v.3.0. MGMTpm status was assigned using the MGMT-STP27 logistic regression algorithm, originally developed for GBMs. Analysis included reference dataset cases with high-confidence match (score ≥0.900) for methylation family and class (16,387).

Results: MGMTpm frequency varies markedly across methylation classes, ranging from 0% to 100%, with a few notable groups: (1) Established clinical significance: GBMs have an elevated overall frequency at 47.6% (1,138/2,391) with its classes ranging from 33.3% (mesenchymal subtype, subclass B; 36/108) to 60.0% (subtype posterior fossa, subclass 2; 3/5). (2) High frequency with potential or unclear significance: The highest frequencies are observed in oligosarcomas (100%, 11/11), oligodendrogliomas (99.5%, 621/624), IDH-mutant astrocytomas (low-grade, 82.0%, 350/427; high-grade, 82.9%, 214/258), diffuse hemispheric gliomas, H3 G34-mutant (69.8%, 125/179); and high-grade astrocytomas with piloid features (59.2%, 106/179). (3) Novel High-Frequency Findings: Other tumors with meaningful frequencies include myxopapillary ependymomas (41%, 16/39); embryonal tumors with multilayered rosettes, C19MC-altered (40%, 90/225); and glioneuronal tumors with ATRX alteration, kinase fusion and anaplastic features (40%, 4/10). Other methylation classes had frequencies < 40%.

Conclusions: Using methylation array as a uniform analytic approach, this study provides an overview of MGMTpm across CNS/PNS tumors, supporting a tumor-specific framework for interpretation of MGMTpm array results. These data confirm established patterns of MGMTpm, while revealing meaningful frequency in additional tumor types, suggesting a potential role of MGMTpm across a wider spectrum of CNS tumors.

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Identification of KIAA1549::BRAF fusion using Next Generation Sequencing

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Background: Low-grade glial and glioneuronal tumors often have alterations in the MAPK pathway with KIAA1549::BRAF fusion representing a common alteration in pilocytic astrocytoma. In addition to diagnostic value, identification of this fusion has treatment implications. This project pursues the validation of our in-house Next Generation Sequencing (NGS) to identify KIAA1549::BRAF fusions.

Methods: 25 cases with confirmed KIAA1549::BRAF and NGS (capture and Illumina sequencing) were identified. 60 known tumors negative for KIAA1549::BRAF, 25 negative controls from the same chip as the known positives, and 20 negative controls from a pool of normals were also included. The probe set for the NGS panel included probes against BRAF and associated introns but did not include probes against KIAA1549, meaning any reads mapping to KIAA1549 were likely a result of the fusion product captured by probes in BRAF. A custom-built bioinformatics tool (BAMDepth_Tool) and an open-source tool (GeneFuse) were also used to identify KIAA1549::BRAF fusions.

Results: We established 4 primary criteria to identify KIAA1549::BRAF fusions: 1. >10 reads with 2 MB insert sizes and a mate start position in the fusion partner (required) 2. A visual pattern in IGV of reads mapping to KIAA1549 with a flat left side (soft-clipped) and triangular tail to the right (required). 3. Positive for fusion on GeneFuse tool (supportive). 4. Read depth >20 in KIAA1549 introns on BamDepth_Tool (supportive). Using these criteria, 22 of the 25 (88%) known positive cases were found to have KIAA1549::BRAF fusions using our in-house NGS. Of the 105 negative controls, none showed evidence of the fusion. The calculated diagnostic accuracy of this testing methodology is 97.7%.

Conclusions: This methodology for identification of KIAA1549::BRAF fusion allows for further identification of this actionable fusion in centers without adequate fusion testing capabilities as well as in the setting of NGS panels without coverage of KIAA1549.

Posters: Neurodegenerative (Alzheimer)

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CDC25A expression is differentially modified by Hispanic ancestry in single and mixed etiology dementia

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Background: Hispanic individuals are at a 1.5X higher risk for Alzheimer's disease and related dementias, and analyses identify Hispanic genetic ancestry as a modifier of Alzheimer's disease-associated gene expression. Additionally, antemortem-based studies suggest altered pathological burden in Hispanic individuals, with neuropathological reports revealing disproportional anatomy-dependent burden of Alzheimer's pathologies in Hispanic decedents. However, few analyses investigate gene expression across dementia-associated pathologies from postmortem brain of Hispanic decedents. We therefore investigated gene expression in Hispanic decedents with single or mixed etiology dementia.

Methods: RNA extracted from postmortem frontal cortex of Hispanic and non-Hispanic decedents with Alzheimer's disease (AD, n=6), cerebrovascular disease (CVD, n=6), or mixed etiology (ADCVD, n=6) was hybridized to the NanoString Human Neuroinflammation panel, and gene expression quantified via SPRINT nCounter. Analysis completed with NanoString Advanced Analysis criteria via ROSALIND®. Low counts/low background genes were omitted from analysis. To test causality, homologous top hits were selected for genetic experiments using transgenic Drosophila melanogaster with human amyloid beta (AB) or tau. Neurodegeneration quantified via locomotor and immunohistochemical methods.

Results: Comparison of Hispanic decedents with AD or ADCVD to non-Hispanic decedents reveals shared pathway enrichment of the Astrocyte Function gene set. Respective to non-Hispanic decedents, two genes in Hispanic AD and seven genes in Hispanic ADCVD are differentially expressed. Across all samples, analysis indicates CDC25A transcript level is decreased, while eight genes are increased in Hispanic CVD decedents compared to all decedents analyzed. Interestingly, panneuronal overexpression of the CDC25A fly homolog string reduces locomotion, a trend consistent in the context of human AB42.

Conclusions: These preliminary findings suggest an ancestry- and etiology-specific transcriptional profile in CVD. Ongoing experiments aim to determine CDC25A levels relative to pathology in postmortem human brain and transgenic Drosophila. Overall, we seek to identify Hispanic ancestry- and disease-specific genes among dementia etiologies for precision medicine among an at-risk population.

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Amyloid-associated neurites: A common co-pathology of diffuse plaques

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Background: The amyloid cascade hypothesis postulates that Aβ drives Alzheimer's disease with p-tau downstream. Aβ is thought to precede p-tau by a period of years. The temporal sequence of cortical Aβ and p-tau at the cellular level is less clear. We observed p-tau-positive neuritic profiles within diffuse plaques in subjects without neuritic plaques. We sought to investigate this finding in SuperAgers and cognitively healthy middle-aged subjects.

Methods: SuperAger status = a score of 9/15 or more words on the delayed recall of the Rey Auditory Verbal Learning Task (RAVLT) after age 80. Subjects underwent yearly neuropsychological assessments and were assessed annually (13 years on average). Only cognitively stable subjects were included. 7 SuperAgers were studied. 16 subjects in their 50s with no history of cognitive decline were also studied - 8 with diffuse Aβ plaques and 8 without. Brain tissue from middle aged subjects was obtained from the Lieber Institute for Brain Development (LIBD). We assessed neocortical tau (AT8), tau217, and Aβ by immunohistochemistry in all subjects. All cases were assessed for Alzheimer's disease neuropathologic change (ADNC) according 2012 guidelines (Northwestern cases), and modified Braak and CERAD guidelines (LIBD cases).

Results: Small tau-positive neuritic profiles were commonly observed and co-localized with diffuse plaques, which we term Amyloid-Associated Neurites (AAN). AAN were not observed in the absence of diffuse plaques. Both AT8 and tau217 labeled AAN. AANs were ~2-5 microns and had a smudgy, rounded morphology, distinguishable from neuropil threads.

Conclusions: We report tau-positive AANs as a common co-pathology of diffuse Aβ plaques in cognitively healthy aging, suggesting that phosphorylated tau aggregation occurs simultaneously with Aβ deposits. Although common, AANs have not been emphasized in the literature. It is plausible that diffuse Aβ plaques with AANs are neuritic plaque precursors, although their presence in the cognitively healthy suggests they may be a benign aging phenomenon.

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Spatially resolved transcriptomic heterogeneity shapes tau selective vulnerability in human master circadian clock in Alzheimer's disease

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Background: Circadian rhythm interventions are emerging therapeutic strategies in Alzheimer's(AD) to mitigate sundowning and stabilize sleep-wake cycles, yet neuronal mechanisms within the human hypothalamic circadian circuitry poorly defined. Prior work in the human hypothalamus demonstrated selective vulnerability of the suprachiasmatic nucleus(SCN) with predominant tau accumulation, whereas the adjacent paraventricular nucleus(PVN) exhibited amyloid burden and the supraoptic nucleus(SON) shows minimal pathology. We also observed autophagy dysregulation in the SCN. Here, we hypothesized that intrinsic, nucleus-specific transcriptomic profiles, particularly autophagy-lysosomal pathways, stratify shared neuromodulatory identity and illustrate spatial differences in neuropathologic vulnerability.

Methods: We performed spatial transcriptomics(Visium10xGenomics) on 12 postmortem anterior hypothalami from cognitively unimpaired individuals with none/low AD-tau pathology(Braak0-II) and individuals with high AD-tau burden(BraakVI). We applied RNA-based clustering(BayesSpace) and anatomically annotated clusters based on shared neuromodulatory markers, including AVP(mean 3,403 valid spots/sample). We assessed statistical coupling between nucleus-marker-gene effects and curated CMA autophagy, lysosomal, and macroautophagy genes, and compared module-level patterns between groups.

Results: Unbiased clustering segregated the SCN transcriptomic profile from PVN and SON, despite overlapping neuromodulatory markers(Fig.2). In the SCN, marker-gene effects were significantly coupled to CMA and lysosome modules and were altered in AD, with increased CMA but reduced lysosome module expression(Fig.3a,d). In contrast, PVN modules showed limited global shifts, but selective coupling with macroautophagy exhibited and was enhanced in AD(Fig.3b,e,h). The SON demonstrated absent coupling and minimal transcriptomic change in AD(Fig.3c,f,l).

Conclusions: Integration of prior nucleus-specific AD neuropathologic patterns with spatial transcriptomics suggests that tau-related vulnerability in the SCN preferentially associates with CMA/lysosomal processes, whereas amyloid-predominant burden in the PVN aligns more closely with macroautophagy. Spatially resolved transcriptomics across adjacent human hypothalamic nuclei thus provides a robust framework to interrogate selective vulnerability in human tissue, especially for tau accumulation, which remains incompletely recapitulated in experimental models, and may guide precision strategies targeting circadian and sleep dysfunction in AD.

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Association between hippocampal microglia and hippocampal neurodegeneration in community-dwelling older adults

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Background: Our prior work has shown that hippocampal microglia are strongly associated with limbic age-related TDP-43 encephalopathy neuropathologic change (LATE-NC) and tangle pathologies. However, the association between microglial inflammation and hippocampal neurodegeneration remains unclear in postmortem human studies. The present study investigated the association between microglia and hippocampal neuronal loss and assessed whether this association is modified by LATE-NC and tangle pathologies.

Methods: Neuropathologic data were obtained from four Rush community-dwelling older adult cohorts who underwent for autopsy (N=372, mean age-at-death 91 (SD=6) years and 70% female). Total microglial density in the hippocampal CA1/subiculum region was quantified using immunohistochemistry with an HLA class II histocompatibility antigen antibody (CR3/43) and StereoInvestigator with a random sampling approach. Hippocampal neuronal loss severity in the CA1/subiculum was assessed semiquantitatively. Additional assessments included amyloid-β burden and tangle density, semiquantitative staging of LATE-NC, and the presence of Lewy bodies, hippocampal sclerosis of aging (HS-A), and five cerebrovascular pathologies. Associations were examined using multiple linear regression models adjusted for demographics and brain pathologies.

Results: The mean hippocampal microglial density was 147.7±63.3 and the mean hippocampal neuronal loss severity was 1.9±1.1 (None-mild: N = 255; Moderate: N = 77; Severe: N = 40). After adjustment for demographics and other pathologies, higher microglial density (B=0.008, SE=0.001) and LATE-NC (B=0.537, SE=0.101) were independently associated with greater hippocampal neuronal loss. A significant interaction between microglia and LATE-NC was observed (B=0.008, SE=0.002), such that microglia was associated with more neuronal loss in the presence of advanced LATE-NC (stages 2/3). No interaction was found between microglia and tangle pathology (p=0.19). Finally, these results remained same after excluding individuals with HS-A.

Conclusions: These results suggest that hippocampal microglia may affect the hippocampal cellular environment, particularly by independently contributing to neuronal damage and interact with LATE-NC to exacerbate hippocampal neurodegeneration beyond their additive effects.

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Topologic morphology descriptors reveal structural differences in microglia by sex and AD neuropathologic change.

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Background: Alzheimer's disease (AD) is the most common cause of dementia worldwide, wherein two-thirds of affected individuals are women. Microglia, CNS innate immune sentinels, show heterogenous activation responses to AD pathology. Morphologic changes reflect functional states, but traditional morphometric tools suffer from bias and inaccuracy. Topologic methods, which quantify structure preserved under deformation, have shown sex-specific differences in murine microglia. The aim of this study was to utilize a topology-based approach on human tissue with two-dimensional histopathology images.

Methods: To evaluate microglial morphologies across AD neuropathologic change (ADNC) levels, participants with CNS autopsies of no/low (N=6), intermediate (N=12), and high ADNC (N=12) were identified from the Mayo Clinic Study of Aging (MCSA) and ADRC. After diagnostic evaluation, single immunohistochemistry for IBA1 (WAKO, 1:1000) was performed on 10-micron FFPE sections, and slides were scanned at 40X (Leica GT450). Five regions of interest were selected from the grey matter in each image and preprocessed to remove background staining. Skeletonized tracings of microglia were collected in Imaris 10.2.0 using an Imaris random forest classifier for seed selection. Skeletons were used to create topological morphology descriptors, persistence images, and then bootstrapped without replacement to reduce intragroup cellular variability. High-dimensional bootstrapped datapoints were pooled and underwent dimensionality reduction with PCA and visualization with UMAP. A sample-to-population ratio for bootstrapping was visually validated.

Results: A total of 57899 cell skeletons were traced from over 90 regions of interest. Dimensionality reduction and visualization with UMAP revealed distinct clusters of microglial morphology by ADNC level. When bootstrapping by sex, male and female cases clustered independently in cases without pathology and increasingly overlapped at intermediate and high ADNC.

Conclusions: Differences in microglial morphology by ADNC level can be demonstrated using topologic morphology descriptors from two-dimensional images. Microglial morphologies appear sexually dimorphic in human brains, though this difference diminishes with increasing ADNC level.

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Variability of microglial activation in cerebral and cerebellar white matter in Alzheimer's disease

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Background: Microglial activation is a well-recognized feature of Alzheimer's disease (AD), particularly in grey matter, where microglia cluster around amyloid plaques and tau pathology. White matter microglia are also altered in AD, but their behavior is less well characterized. In particular, it is not clear whether microglial number and activation state vary in the same way across different white matter regions, including the cerebellum, which is relatively understudied.

Methods: We examined autopsy cases with Alzheimer's disease neuropathologic change and co-pathologies, using image analysis in QuPath applied to IBA-1– and CD68-stained sections in cerebellar white matter (25 cases) and, in a subset, matched cerebral hemispheric white matter. IBA-1 was used as a marker of total microglial burden and CD68 as a marker of phagocytic/activated microglia. For each region, we measured the fractional area positive for IBA-1 and CD68 and calculated an activation index as CD68/IBA-1.

Results: Cerebellar white matter showed significant heterogeneity in all three measures. Some cases had relatively few microglia with a high CD68/IBA-1 ratio, while others had dense microglial populations with little CD68 expression. When data from cerebellar and hemispheric white matter were combined (36 regions), the activation index showed a moderate inverse relationship with IBA-1 burden ($r \approx -0.38$, $p \approx 0.02$). In practical terms, regions with many microglia tended to have a lower proportion of CD68-positive (activated) cells ("many microglia, mostly quiet"), whereas regions with fewer microglia were more CD68-rich ("fewer microglia, more activated"). CD68 and IBA-1 themselves were only weakly correlated, suggesting that changes in microglial activation are not simply a reflection of microglial number.

Conclusions: These findings suggest that in AD, cerebral and cerebellar white matter exhibit substantial variation in both microglial burden and activation state that are not tightly linked. Microglial density and activation appear to be regulated independently, rather than along a single linear activation gradient.

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Microglial activation in the Locus Coeruleus is not associated with early tau or amyloid pathology in middle age

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Background: Alzheimer's disease (AD) is marked by the accumulation of amyloid plaques and phosphorylated tau, a process that begins decades before clinical symptoms appear. One of the earliest sites of tau pathology is the locus coeruleus, a brainstem nucleus that shows vulnerability even in young, cognitively normal individuals. Microglia are often implicated in AD progression, with prevailing models proposing that their activation facilitates tau spread. Whether this relationship exists at the earliest stages of disease remains uncertain.

Methods: We studied postmortem brain tissue from 126 individuals 65 years of age and younger who died without a clinical history of neurodegenerative disease. Cases were staged using Braak and Thal criteria. Immunofluorescence for phosphorylated tau, tyrosine hydroxylase, total microglia, and MHC class II–positive activated microglia was performed in the locus coeruleus. Microglial density and activation were quantified using an AI-based computational approach.

Results: Increasing age was associated with greater tau pathology in the mesial temporal lobe and higher Braak stage. In the locus coeruleus, amyloid deposition tended to be associated with increased phosphorylated tau in the locus coeruleus and was associated with increased Braak stage in the supratentorial brain. In contrast, neither activated nor total microglial density was associated with tau or amyloid pathology. Instead, microglial density was higher in males than females, independent of age, tau burden, or amyloid deposition.

Conclusions: These findings indicate that early tau accumulation in the locus coeruleus occurs independently of microglial activation in young individuals without clinical AD. Our data challenge models in which microglia drive early tau spread and suggest that microglial involvement may emerge later or vary by brain region. Future studies incorporating spatial transcriptomics and proteomics will further clarify microglial roles across AD progression

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Digital Mapping of Regional Tangle Distribution Defines Alzheimer's Disease Subtypes

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Background: Alzheimer's disease (AD) is characterized by amyloid-β plaques and neurofibrillary tangles. Distinct AD subtypes have been defined by the regional distribution of tangles across hippocampal and cortical regions, with prior work demonstrating subtype-associated differences in age and genetic risk. While these classifications have relied on manual tangle counts, we developed a digital tangle-classifier for immunohistochemically-stained slides that detects discrete tangle pathology to enable automated and scalable quantification. We evaluated whether digitally derived regional tangle distributions support extension of established AD subtyping frameworks.

Methods: An ADRC autopsy cohort (N = 47) with clinical dementia, high AD neuropathologic change, and no hippocampal sclerosis, was analyzed. Digital quantification of tangle burden was performed in five regions: CA1, subiculum, inferior parietal lobule (IPL), middle frontal gyrus (MFG), and middle temporal gyrus (MTG). Hippocampal and cortical composite averages were calculated, and a hippocampal-to-cortical ratio was derived. Subtypes (typical, hippocampal-sparing, limbic- predominant) were assigned using cohort-derived percentile (r25/r75) and median-based criteria reflecting established regional distribution approaches.

Results: Subtype distribution was 85% Typical AD (40/47), 6% hippocampal-sparing (3/47), and 9% limbic-predominant (4/47). Median age at death differed by subtype, with hippocampal-sparing cases younger than typical (67 vs 75 years) and limbic-predominant cases older than typical (77 vs 75 years), consistent with previously described patterns. APOE ε4 carrier frequency (≥1 ε4 allele) was lower in hippocampal-sparing (33%) and higher in limbic-predominant (100%) relative to typical (60%), paralleling prior observations. Given the limited number of non-typical cases, these findings are interpreted as directional trends.

Conclusions: Automated digital quantification of regional tangle burden supports extension of established AD subtyping frameworks to IHC-based image analysis. Subtype proportions and directional associations with age and APOE ε4 status align with previously described subtype biology. Ongoing spatial molecular studies will assess whether digitally defined subtypes reflect underlying molecular heterogeneity.

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Automated Detection of Tauopathy Lesions in AT8 IHC-Stained Whole Slide Images of cerebral cortices of Alzheimer Disease cases With Deep Learning

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Background: Tau aggregates include neurofibrillary tangles (NFTs), neuritic plaques, and glial tau inclusions. Although great progress has been made in automated detection of certain aggregates using deep learning (DL), this can be limited for multiple lesion types within IHC-stained WSIs. Hence, we aimed to develop an automated pipeline for multiple tau deposit detection using DL object detection models.

Methods: Trained personnel annotated AT8 IHC-stained WSIs (N=39) from the frontal, parietal, and temporal lobes in a Digital Slide Archive (DSA) annotation platform, identifying regions of interest (ROIs) both in grey and white matter labeling NFT, neuritic plaque, and glial tau structures. ROIs were extracted at 20x magnification and tiled into overlapping patches. Tiles were filtered to ensure adequate ROI coverage (≥25% tile area) and bounding box integrity. A YOLO11m object detection model was trained to detect structures.

Results: The dataset consisted of 6,894 annotated NFTs, 7,016 neuritic plaques, and 1,332 glial tau structures. The YOLO11m model was trained on 39 AT8-stained WSIs (that included 4519 NFTs, 5241 neuritic plaques, and 972 glial tau). Training achieved a mean Average Precision (mAP50) of 82.4% for NFT detection, 66.4% for neuritic plaque detection, and 18% for glial tau detection on the validation set. The model demonstrated precision of 77.9% and recall of 57.4% for NFT, 52.9%/33.3% for neuritic plaques, and 16.2%/0.9% for glial tau. Transfer learning from the preNFT/iNFT model improved convergence. Whole-slide inference was completed in under 5 minutes per slide on an NVIDIA GPU.

Conclusions: This pipeline is a proof of concept for DL methods for detection and quantification of multiple tau aggregate classifications in AT8-stained neuropathology WSIs in AD, potentially reducing annotation time and improving consistency.

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A machine learning approach to assess vascular β-amyloid and perivascular plaques in cerebral amyloid angiopathy

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Background: This study aims to 1) develop a machine learning approach to automate detection of cortical CAA-positive vessels separate from β-amyloid plaques, 2) validate the digital CAA classifier against semi-quantitative manual scoring, and 3) examine the spatial distribution of perivascular plaques in relation to CAA-positive vessels.

Methods: Machine learning classifiers were trained on the HALO AI platform to distinguish cortical CAA-positive vessels from β-amyloid plaques in the calcarine cortex. Classifiers were applied to digitized histopathologic slides (n=100), previously rated by expert neuropathologists. Digital quantitative measures for CAA (CAA classified area and vascular β-amyloid positivity) and for β-amyloid plaques (β-amyloid plaque classified area) were compared with manual CAA scores and Alzheimer's disease neuropathologic change (ADNC). Spatial proximity analysis characterized perivascular plaques, defined as β-amyloid plaques within a 0- to 150-micron concentric distance from the center of CAA-positive vessels.

Results: Participants had a mean age of 91 (SD=5.8) years at death, with 76% being women. Machine learning classifiers for CAA and vascular β-amyloid positivity showed excellent correlations with semi-quantitative manual CAA scores ($r= 0.94$, $p< 0.001$ and $r= 0.81$, $p< 0.001$, respectively). The number of CAA-positive vessels per slide ranged from 0 to 1017, with an average of 5 vessels in score 1, 15 in score 2, 163 in score 3, and 473 in score 4. Digital β-amyloid plaque classified area correlated strongly with Thal phase ($r= 0.74$, $p< 0.001$) and moderately with ADNC ($r= 0.56$, $p< 0.001$). Interestingly, the total area of β-amyloid plaques was relatively similar across increasing manual CAA scores. However, spatial proximity analysis revealed that cases with higher CAA scores exhibited more perivascular plaques closest to the CAA-positive vessel.

Conclusions: Machine learning-based classifiers reliably distinguish cortical CAA vessels from β-amyloid plaques and demonstrate a spatial relationship between CAA severity and perivascular plaque clustering, supporting their utility for advancing CAA research.

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Quantitative Digital Pathology for Neurodegenerative Disease: Tissue Segmentation and Cell Classification

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Background: Neurodegenerative diseases (NDs) involve complex pathological changes at the cellular level that remain incompletely understood. Immunohistochemistry and special stains are typically used to semi-quantitate pathology according to consensus guidelines, whereas hematoxylin and eosin (H&E) is used to identify structural alterations in general. Exploring morphologic changes using H&E-stained slides with quantitative (machine learning) tools may provide new insights into NDs. Quantifying these pathological changes requires image-based segmentation of gray matter (GM) and white matter (WM) and classification of cell types.

Methods: To date, we have digitized 6,918 slides from 255 cases with neurodegenerative diseases from the Mesulam Institute biobank. Our goal is to create an integrated data mart linking digital slides with patient-level clinical reports for ND research. We used 71 annotated whole-slide images (WSIs) with a 70/10/20 training/validation/testing split to develop a tissue segmentation model that delineates GM regions as a first step toward analyzing tissue changes in NDs. We also trained a self-supervised learning (SSL) model as an encoder and a cell classifier to identify four neuropathological cell types in GM: neurons, astrocytes, oligodendrocytes, and endothelial cells. This model used 1.5 million unlabeled cell patches and 14,603 manually annotated nuclei for training and was evaluated on 4,335 annotated nuclei from separate slides.

Results: The GM/WM tissue segmentation model achieved 92.90% intersection over union (IoU) and an area under the curve (AUC) of 98.63%, demonstrating precise delineation of tissue boundaries. The cell classifier achieved 90.94% balanced accuracy, with F1-score of 91.35%. The strongest performance was observed for neurons (96.18% F1-score), and the SSL model’s embedding space revealed a clear qualitative separation of neuron types.

Conclusions: The primary misclassifications occurred between astrocytes and oligodendrocytes, suggesting that fine-tuning with additional annotated examples could improve performance. Future work will use these tools to measure quantitative features describing pathologic changes in NDs.

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Multi-Class Detection and Quantification of Neuropathological Features using YOLOv11

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Background: Definitive diagnosis of Alzheimer's disease and related dementias requires post-mortem examination including characterization of various protein aggregates by immunohistochemistry. Manual quantification is labor-intensive, requires specialized expertise, and is subjective because of substantial inter-rater variability, limiting reproducibility and constraining large-scale clinicopathological studies. While deep learning on digital whole slide images offers promise for automated pathology quantification, existing approaches often lack the neuropathology-specific optimization and clinical validation needed for translation. To address this gap, we developed and validated a comprehensive proteinopathy detection platform that enables objective, reproducible quantification of protein aggregates at scale.

Methods: We developed four YOLOv11-based object detectors, each fine-tuned independently for TDP-43 inclusions, tau-positive neurofibrillary tangles, Lewy bodies, and beta-amyloid plaques. Training datasets comprised 73 whole slide images (WSIs) across 73 patients for tangles (~3k labeled features), 9 WSIs from 6 patients for TDP-43 (~0.45k), 37 WSIs from 7 patients for amyloid (~6.8k), and 4 WSIs from 3 patients for Lewy bodies (~1.1k). We employed transfer learning with pathology-specific data augmentation and k-fold cross-validation (k=5) to minimize overfitting. Performance was evaluated using precision, recall, F1-score, and mean average precision (mAP).

Results: Classifiers achieved mAP scores of 93% (TDP-43), 77% (tangles), 82% (Lewy bodies), and 78% (amyloid), demonstrating robust performance across tissue preparations. NACC cohort validation revealed significant associations between automated pathology burden and cognitive decline. TDP-43 detections showed progressive increase from cognitively normal to mild cognitive impairment to dementia ($p < 0.0001$), confirming that automated quantification captures clinically meaningful disease biology.

Conclusions: Our YOLOv11-based detector suite provides accurate, scalable detection of key protein aggregates in neurodegenerative diseases. This modular approach enables flexible pathology-specific assessment with applications in standardizing neuropathological evaluation and accelerating large-scale quantification. Future work includes implementing automated semi-supervised annotation expansion to improve performance on rare pathological variants and enhance generalizability across diverse tissue preparations.

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Applying NULISAseq for postmortem differentiation distinct clinicopathologic forms of Alzheimer's disease

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Background: Alzheimer's disease (AD) is clinicopathologically heterogeneous, with upwards of 25% of patients presenting with non-amnestic syndromes. Neuropathologically, corticolimbic distribution of tangle pathology is strongly associated with clinical presentation, as well as differences in sex, age at symptom onset, and APOE ε4 carriership. This study sought to evaluate "liquid biopsy" technology (i.e., Alamar Biosciences NULISAseq) on postmortem brain and assess protein level changes across clinicopathologic forms of AD.

Methods: We queried the Mayo Clinic Alzheimer's Disease Research Center database for neuropathologically diagnosed AD cases with antemortem plasma and corticolimbic index (CLix). CLix is a scaled ratio of hippocampal to cortical tangle distribution with lower scores reflecting greater cortical tangle pathology relative to the hippocampus. We performed a pilot analysis on n=13 AD cases ranging in age at symptom onset from 51-86 years (median=63 years [Interquartile range:56,74]) and CLix (median=13 [IQR:6.1,29]). NULISAseq CNS disease panel 120 was performed using frozen inferior parietal cortex homogenized in RIPA buffer and ran on the ARGO platform. Association between age onset, CLix, and target protein expression were analyzed separately via linear regression.

Results: We observed 80 of 129 protein targets were detected above a 50% limit of detection. Examining protein level differences and age onset, we observed greater ptau217/total tau (β: -7.5 [CI:-13,-2.0], p=0.012), ptau231/t-tau (β:-7.3 [CI:-13,-1.7],p=0.015), ubiquitin (β:-7.2 [CI:-13,-1.5], p=0.018) in cases with younger age onset. Examining protein level differences through the lens of neuropathologic heterogeneity, we observed greater p-tau181 (β:-7.3 [CI:-14,-0.48], p=0.038), ptau217/t-tau (β:-10 [CI:-15,-5.7], p=0.014), p-tau231/t-tau (β:-10. [CI:-15,-5.3], p< 0.001), and ptau231 (β:-8.3 [CI:-15,-2.0], p=0.014), in cases with greater cortical tangle pathology (i.e., lower CLix).

Conclusions: NULISAseq sufficiently detects protein targets in RIPA-post-mortem brain. Here we demonstrate differences in p-tau isoforms and ubiquitin across clinicopathologic forms of AD. Ongoing work will evaluate plasma p-tau expression across AD clinicopathologic heterogeneity in a larger cross-sectional sample.

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AD Molecular Abnormalities in White Matter Glia Detected in Unfractionated and O4-Selected Serum Exosomes Using a Liquid Biopsy Approach

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Background: White matter degeneration is a significant and early mediator of cognitive impairment in Alzheimer's disease (AD), yet the critical pathologic features remain poorly understood, under-detected, and therapeutically untargeted. Herein, we characterize molecular features of white matter glial cells in AD brains and assess the utility of non-invasive approaches for detecting related abnormalities in extracellular vesicles (EVs) isolated from serum (SEVs). In addition, results from unfractionated (SEV-T) and O4 sulfatide-selected SEVs were compared to determine whether white matter abnormalities were detected with greater sensitivity in oligodendrocyte-specific SEVs (SEV-O4).

Methods: Oligodendrocyte glycoprotein and astrocyte mRNA levels were measured in postmortem human AD and control frontal lobe white matter by RT-PCR. Immunoreactivity to oligodendrocyte glycoproteins, astrocyte structural proteins, neurofilament light chain (NfL), and aspartyl-asparaginyl-β-hydroxylase (ASPH) was measured by ELISA in SEV-T and SEV-O4 from patients with moderate AD or normal aging.

Results: AD brain pathology was associated with significantly reduced mRNA expression of multiple oligodendrocyte glycoproteins and increased mRNA expression of astrocytic structural genes. SEV analyses demonstrated significantly increased immunoreactivity to 2',3'-cyclic nucleotide 3' phosphodiesterase (CNPase), myelin-associated glycoprotein 1 (MAG1), astrocyte proteins, and ASPH, a potent activator of Notch and myelin-regulated homeostatic functions. There were no significant benefits of measuring SEV-O4 compared with SEV-T immunoreactivity.

Conclusions: AD is associated with significant molecular abnormalities in the function of oligodendrocytes and astrocytes in brain tissue. The abnormalities detected in SEVs likely reflect oligodendrocyte injury and degeneration, as well as astrocytic activation. The findings suggest that low-invasive SEV approaches, including the novel analysis of ASPH upregulation, can be used to detect and monitor white matter degeneration in AD.

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Dysregulated Incretin Networks Linked to AD Progression and APOE-ε4 Dose Suggest Roles for GLP/GIP Therapeutics in Humans

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Background: Brain metabolic dysfunction in AD is linked to impairments in insulin signaling marked by reductions in glucose utilization, energy metabolism, and downstream genes that regulate neuronal and oligodendrocyte survival and function. Recent studies have demonstrated that brain metabolic dysfunction in experimental AD models is associated with impairments in brain incretin and incretin receptor expression, and that corresponding biomarker abnormalities are detectable in exosomes isolated from clinical serum and CSF samples. This study examined: 1) the relationship between severity of AD and alterations in incretin expression in the brain; the effects of apolipoprotein E-ε4 (APOE-ε4) dose on incretin expression in AD; and 3) detectability of incretin-related abnormalities in exosomes isolated from clinical serum and CSF samples of patients diagnosed with early-stage or moderate AD. The impact of APOE-ε4 dose was relevant because it is a major risk factor for late-onset AD.

Methods: Postmortem banked fresh frozen frontal lobe tissue from AD and control cases with known APOE genotypes and different AD Braak stages, and exosomes isolated from clinical AD and control serum and CSF samples were used to measure immunoreactivity to incretin network-related molecules using a multiplex enzyme-linked immunosorbent assay (ELISA).

Results: AD Braak stage and APOE-ε4 dose were associated with progressive declines in frontal lobe gastric inhibitory polypeptide (GIP), glucagon-like peptide-1 (GLP-1), leptin, ghrelin, glucagon, and resistin, but not plasminogen activator inhibitor-1 (PAI-1) or Visfastin. In clinical samples, abnormalities were observed in exosomes isolated from serum and CSF in early and moderate AD.

Conclusions: This study demonstrates an expanded role for impaired expression of the incretin-related network polypeptides in AD and supports the concept that metabolic dysregulation of these pathways is mechanistically important in AD progression. In addition, the findings suggest that GLP-1/GIP receptor agonist interventions may have therapeutic benefits across all stages of AD neurodegeneration, including in individuals with APOE ε4 genotypes.

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An iPSC-Derived Neuronal Interactome: A Resource for Alzheimer's Disease Target and Interaction Discovery

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Background: Alzheimer’s disease (AD) involves the progressive build-up of β-amyloid plaques and hyperphosphorylated tau aggregates, which has driven extensive drug development efforts aimed at these hallmarks. Although previous work has yielded important mechanistic insights, clinical outcomes indicate that focusing on these hallmarks alone may overlook other therapeutically relevant biology.

Methods: We constructed an iPSC-derived neuronal interactome as a resource for target and interaction discovery. We used gene expression profiles from 150 Ngn2-transdifferentiated iPSC-derived neuron samples to infer regulatory networks via ARACNe3, with mutual information significance thresholds set to control the false discovery rate. Two regulatory networks were inferred separately using transcription factors/co-transcription factors (TF/co-TF) and signaling proteins (SigP) as regulator inputs.

Results: The top 250 target genes per regulator were kept, resulting in a TF/co-TF interactome consisting of 2,364 regulators and a SigP interactome consisting of 3,125 regulators. Interactome quality control was performed by benchmarking inferred interactions against known interactions from ChIP-seq data. In a subsequent applied study, we leveraged our interactome to guide a high-throughput compound screen, demonstrating its practical utility for AD drug discovery. Furthermore, we developed an online platform for researchers to explore and visualize the interactome data.

Conclusions: Together, this interactome resource lays groundwork for identifying previously under-explored targets and mechanisms in AD.

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Drug screening in hiPSC neurons: A bioinformatic approach to identify drug candidates for Alzheimer's disease.

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Background: Alzheimer's disease (AD) is characterized by the accumulation of β-amyloid plaques and tau-positive tangles. To date, pharmacological strategies directed at these hallmarks have achieved limited success, emphasizing the need to target broader disease-associated pathways and mechanisms.

Methods: To address this question, we implemented a bioinformatic strategy to generate a human-induced pluripotent stem cell (hiPSC)-derived neuronal interactome and, within the same platform, conduct a high-throughput screen of 1039 compounds using PLATE-seq, a low-cost, high-throughput gene expression profiling methodology. This generated a dataset that describes how each drug affects the activity of 5489 different cellular proteins.

Results: Interrogation of this dataset identified compounds predicted to modulate AD-associated master regulators (MRs), including RE1-Silencing Transcription Factor (REST) and Sterol Regulatory Element Binding Transcription Factor 2 (SREBF2). REST and SREBF2 are MRs of neurogenesis and cholesterol biosynthesis, respectively, and their dysregulation has been implicated in disease etiology. Our hypothesis is that targeting these MRs will rescue AD-associated phenotypes in hiPSC-derived neurons such as increased phospho-tau levels, enlarged early endosomes, and accelerated neurogenesis. Exploratory validation of drug candidates predicted to modulate these MRs revealed a significant reduction in phospho-tau levels at Ser202, Thr231 and Ser396/404 in compound-treated vs. control hiPSC neurons. Immunofluorescence analysis also showed reduced expression of MAP2 positive staining in AD vs. control groups following treatment, suggestive of a "rescue" of the aberrant neurogenesis phenotype seen in AD lines. In addition, initial examination of AD early endosomes indicated a restoration of endosomal size to control hiPSC levels following treatment. Analysis to further validate compounds and the above findings are ongoing.

Conclusions: In summary, we utilized an integrated approach to identify drug candidates that can target disease-relevant MRs and thereby rescue key AD-relevant phenotypes in hiPSC-derived neurons.

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Regional tau burden varies with LATE stage in severe Alzheimer's disease neuropathologic change

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Background: Hyperphosphorylated TDP-43 (pTDP-43), the defining feature of limbic-predominant age-related TDP-43 encephalopathy neuropathologic change (LATE-NC), frequently co-occurs with high Alzheimer's disease neuropathologic change (ADNC), and is associated with more severe cognitive decline and hippocampal atrophy. Experimental and post-mortem studies suggest a potential synergistic interaction between tau and TDP-43; however, how this relationship varies across selectively vulnerable hippocampal regions in severe AD remains unclear.

Methods: We analyzed a large post-mortem cohort of high ADNC, with and without LATE-NC, and no other significant neuropathology. Tau burden was quantified as percent area positive for AT8 immunohistochemistry using HALO software for whole-slide digital image analysis across hippocampal subfields (entorhinal cortex [EC], CA1-CA4, subiculum), amygdala, and neocortical regions. Group comparisons used nonparametric methods and sensitivity analyses excluded cases with hippocampal sclerosis (HS) and LATE stage 3. Morphologic subtyping of pTDP-43 (α vs β) is also being performed.

Results: Higher LATE stage was associated with greater tau burden in entorhinal cortex, CA2, and CA3. Amygdala demonstrated a similar directional pattern and reached significance after exclusion of HS-positive cases. Neocortical regions did not show significant differences. CA1 and subiculum did not demonstrate higher tau burden when all cases were included and instead showed apparent reductions at advanced stages. Exclusion of HS-positive cases resulted in directional increases in CA1 and subiculum consistent with other hippocampal subfields.

Conclusions: In high ADNC, more advanced LATE stages are associated with region-specific increases in hippocampal tau burden. Associations were most evident in entorhinal cortex, CA2, and CA3. Apparent non-linear patterns in CA1 and subiculum were modified by hippocampal sclerosis, a regionally selective process that reduces measurable tau burden in advanced stages. These findings are consistent with overlapping regional distributions of tau and TDP-43 proteinopathy, with selective neuronal vulnerability modifying measurable tau burden in advanced stages. Ongoing subtype analyses may further clarify mechanistic heterogeneity underlying these associations.

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Postmortem Evaluation of Aging-Related Tau Astrogliopathy in Decedents with Alzheimer's Disease

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Background: Glial tau pathology involves the accumulation of hyperphosphorylated tau within brain glial cells and can coexist with other neurodegenerative diseases, including Alzheimer disease (AD). A common form of glial tau pathology in the aging brain is aging-related tau astrogliopathy (ARTAG), which occurs more frequently in older individuals and in males. We sought to determine the generalizability of ARTAG distribution through examination of a cohort of persons with AD from different demographic backgrounds, especially those known to have higher rates of concomitant cardiovascular disease.

Methods: We evaluated the presence of ARTAG across five brain regions in a cohort of 267 decedents across ethnicities (including persons of Hispanic descent) with intermediate or high AD neuropathologic change (ADNC). Anterior and posterior hippocampus, temporal, frontal, and parietal cortices were compiled from three Alzheimer's Disease Research Centers (University of California Davis, University of California San Diego, and Columbia University) were stained with AT8 to detect hyperphosphorylated tau and evaluated by a neuropathology expert for the presence/absence of glial tau.

Results: Preliminary analysis showed 73 individuals (27.3%) exhibited ARTAG in at least one brain region (n = 267). Within the subset of individuals who had all brain regions available for evaluation (n = 51), three distinct patterns of ARTAG regional involvement were identified: (1) ARTAG present in the hippocampus and neocortical regions (66.6%); (2) ARTAG limited to the hippocampus (15.6%); (3) ARTAG involving neocortical regions only, without hippocampal involvement (13.7%). Across all individuals, the anterior hippocampus showed the highest frequency of ARTAG, with 40 cases (78.4%) exhibiting pathology in that region, followed by the temporal cortex and posterior hippocampus (each 62.7%), the frontal cortex (47%), and the parietal cortex (37.2%).

Conclusions: These works, which included a more heterogenous cohort, are relatively consistent with prior reports of ARTAG within the setting of AD, demonstrating the generalizability of prior findings.

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Neuroinflammatory pathways across the life span of individuals with Down syndrome

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Background: The innate and adaptive immune systems are dysregulated in Down syndrome (DS). While pathological neuroinflammatory pathways have been extensively studied in idiopathic Alzheimer's disease (AD), knowledge of the distinct roles of these pathways in Down syndrome with Alzheimer's disease neuropathology (DS-AD) is limited. Moreover, when over the lifespan and which brain regions that are affected first in DS remain under investigation.

Methods: In this study, we will use spatial transcriptomics to map neuroinflammation in the hippocampus of individuals with DS, DS-AD and healthy controls. Select inflammatory components will be semi-quantified immunohistochemically across a wide age range and correlated with neuropathological staging. Future research will also include complementary studies in animal models of DS.

Results: Preliminary results from spatial transcriptomics indicate increased expression of genes involved in early neuroinflammatory dysregulation in the hippocampus in individuals with DS-AD compared to healthy controls. Ongoing experiments will determine how levels of activated inflammatory proteins correlate with the broader transcriptome and proteome relevant to brain function, using mouse models and human brain tissue.

Conclusions: The results from this work may help identify whether specific components of the innate immune system constitute early biomarkers or therapeutic targets in DS-AD. Funding support: This work was supported by the Include Biorepository (1U24AG092191-01), P30AG066512, Down syndrome Biobank Consortium (DSBC), and grants from the Lejeune Foundation (GRT-2023b/2277), the BrightFocus Foundation (CA2018010) and the Alzheimer Biomarkers Consortium – Down Syndrome (ABC-DS; NIH/NIA U19AG068054).

Posters: Vascular

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Long-Duration Type 1 Diabetes Paradoxically Predicts Less Vascular and Neurodegenerative Neuropathology at Autopsy

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Background: Increased dementia risk in type 2 diabetes is generally attributed to cerebrovascular disease rather than excess Alzheimer's pathology. Joslin medalist studies of long-duration type 1 diabetes (T1DM) have shown mild cognitive deficits and lower brain volumes than nondiabetic controls, though whether neuropathology differs between T1DM and T2DM – or between T1DM and nondiabetics – remains unknown.

Methods: We compared autopsy neuropathology in Joslin medalists (50+ year duration T1DM; n=33) with 3:1 age- and sex-matched decedents from the community-based Rush Religious Order Study and Memory and Aging Project (ROSMAP) with diabetes (DM+; n=99) and without diabetes (DM−; n=99). Because ROSMAP lacks diabetes subtyping, DM+ participants were presumed predominantly T2DM, consistent with population prevalence (~90–95%). Pathologic variables were compared by Fisher exact tests with pairwise contrasts.

Results: Gross infarcts were more frequent in ROSMAP DM+ (41.4%) than in either DM− (25.3%, p=0.02) or Joslin T1DM (12.1%, p=0.003). Microinfarcts followed a similar pattern but did not reach significance. The groups did not differ in atherosclerosis, arteriolosclerosis, amyloid angiopathy, neuritic plaque burden (CERAD), or β-amyloid spread (Thal phase). Unexpectedly, T1DM showed less neurodegenerative pathology than both comparator groups across multiple categories. Braak stage III+ was less common in T1DM (45.5%) than in ROSMAP DM+ (76.8%, p=0.002) or DM− (66.7%, p=0.04). Advanced ADNC (NIA-AA moderate/severe) was similarly less frequent (27.3% vs. 52.5%, p=0.02 and 49.5%, p=0.03, respectively). Limbic or neocortical Lewy body disease was markedly less common in T1DM (3.0%) than in DM+ (21.2%, p=0.01) or DM− (23.3%, p=0.01). LATE-NC stage 2–3 was less frequent in T1DM (3.0%) than in DM− (21.2%, p=0.01) but did not differ significantly from DM+.

Conclusions: Long-duration T1DM was associated with fewer gross infarcts than ROSMAP DM+ and, unexpectedly, with less ADNC, LBD, and LATE neuropathology than community-dwelling participants with or without diabetes. These findings suggest a distinct neuropathologic trajectory of long-duration T1DM.

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Associations of hemoglobin A1c with cerebral amyloid angiopathy severity in older adults

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Background: Recent studies suggest that hemoglobin A1C level is inversely associated with the severity of Alzheimer's disease (AD), yet the associations between A1C and cerebral amyloid angiopathy (CAA) severity have not been investigated.

Methods: Here, we examined associations between A1C and neuropathologic CAA severity in older adults. The study included 1309 participants with a mean age at death of 90.66±6.54 (SD) years (74.56% women), who were followed in one of five longitudinal studies on aging. A1C level was collected at baseline and annual clinical visits. Postmortem neuropathologic examinations included evaluation for presence and severity of CAA and AD. The study included participants with no (312, 23.83%), mild (492, 37.82%), or moderate to severe (505, 38.58%) CAA. Ordinal logistic regression models were used to analyze the association of A1C at the visit proximate to death with overall neocortical CAA severity, adjusting for demographics. In secondary analyses, we explored the associations of A1C with CAA severity within cerebral lobes and adjusted for APOE genotype.

Results: Ordinal logistic regression analyses revealed an inverse association between A1C and overall neocortical CAA severity (odds ratio/OR, 0.77; 95% CI: 0.67-0.88). When examining by different lobes, inverse associations were found between A1C and CAA severity in frontal (OR, 0.75; 95% CI: 0.65-0.86), parietal (OR, 0.82; 95% CI: 0.71-0.94), temporal (OR, 0.80; 95% CI: 0.69-0.93), and occipital (OR, 0.79; 95% CI: 0.69-0.91) regions. These relationships remained when adjusting for APOE genotype. Stratified by AD, these associations were significant only in participants with but not without AD. In separate models, the interaction terms between A1C and presence of AD was significant when considering the temporal (p=0.027) and parietal (p=0.031) regions.

Conclusions: We conclude that A1C is associated with decreased odds of neocortical CAA severity. The mechanisms of these associations and reasons for variation by AD status are unclear and should be further investigated.

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Regional Distribution and Clinicopathologic Correlates of Mineralized Blood Vessels in Autopsy-Confirmed Alzheimer Disease

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Background: Mineralized blood vessels (MBVs) can associate with vascular aging and neurodegeneration, yet their regional distribution and clinicopathologic correlates in Alzheimer disease (AD) remain poorly characterized.

Methods: We examined a cohort of 265 decedents with Intermediate/High AD neuropathologic change from three Alzheimer's Disease Research Centers—University of California, Davis, University of California, San Diego, and Columbia University. Five micron hematoxylin and eosin–stained sections from the posterior hippocampus, putamen, and globus pallidus were reviewed for MBV presence. Demographic, clinical, genetic, and neuropathologic variables were obtained from harmonized datasets. Associations with MBV were evaluated using bivariate analyses and multivariable logistic regression adjusting for age, sex, ethnicity, and center.

Results: MBV frequency differed significantly by region, with the highest prevalence in the globus pallidus (58.3%), followed by the hippocampus (28.6%), and putamen (13.8%) (all pairwise $p < 0.001$). MBV in the globus pallidus was associated with transient ischemic attack (TIA). MBV showed strong associations with état criblé in the putamen and globus pallidus ($p < 0.001$) but were not associated with concomitant cerebrovascular disease in all regions ($p=0.09$ for posterior hippocampus, $p=0.69$ for putamen, 0.11 for globus pallidus). Lewy body disease was associated with reduced MBV presence, which was significant in the hippocampus and globus pallidus ($p=0.029$, $p=0.013$, respectively). Presence of apolipoprotein E (APOE) ε4 allele was not significantly associated with MBV status in all regions ($p=0.55$ for posterior hippocampus, $p=0.16$ for putamen, $p=0.91$ for globus pallidus).

Conclusions: These findings suggest MBV has select anatomical preferences within the setting of AD and further supports its role as a marker of cerebrovascular contributions to AD, given associations with état criblé and TIA. Future directions involve advancing towards semi-quantitative and/or quantitative analyses, enabling more precise characterization of regional distributions and improved understanding of vascular pathology.

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Gene Expression Signatures of Gliovascular Cells Associated with Microinfarct Burden

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Background: Cerebral small vessel disease (cSVD) is a major contributor to vascular cognitive impairment and a common co-pathology in Alzheimer's disease (AD) and related dementias. Microinfarcts represent a key pathological correlate of cSVD, yet the diffuse molecular effects of chronic small-vessel ischemic injury remain poorly defined. We performed spatially informed exploratory analyses to characterize gliovascular responses to chronic microinfarct burden.

Methods: Using the NanoString GeoMx DSP platform, we profiled whole-transcriptome expression in postmortem human brain tissue from four subjects stratified by microinfarct burden and matched for arteriosclerosis and AD neuropathologic change. Regions examined were confirmed to be free of overt microinfarcts, enabling interrogation of diffuse ischemic stress rather than lesion-specific responses. A total of 190 areas of interest (AOIs) were selected from middle frontal gyrus and basal ganglia (54–929 cells/AOI; median 287), profiling astrocytes, microglia, and endothelial cells. Differential expression and pathway analyses were performed in R using staged pairwise contrasts to model responses to increasing microinfarct burden.

Results: We identified 5,197 unique significant genes across regions and cell types. The largest transcriptional shifts occurred during early disease progression, consistent with a nonlinear response to disease progression. Pathway-level analyses demonstrated consistent trends across brain regions. Early microinfarct burden was associated with activation of proliferative and metabolic programs. Later stages demonstrated widespread suppression of bioenergetic and repair pathways. Cell-type–resolved analyses showed that microglia primarily drove proliferative and oxidative stress programs, astrocytes contributed to pathways of hypoxia response, and endothelial cells were most associated with barrier integrity pathways.

Conclusions: Diffuse gliovascular responses to chronic small-vessel ischemic injury exhibit a nonlinear trajectory, transitioning from early adaptive activation to later metabolic and reparative suppression. Further studies in larger cohorts will be required to validate candidate biomarkers, clarify causal mechanisms, and determine how these diffuse transcriptomic changes relate to clinical cognitive decline in cSVD and mixed dementia.

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Multi-Institutional Evaluation of a Machine Learning–Based Microinfarct and Microhemorrhage Screening Tool

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Background: Microinfarcts and microhemorrhages are contributors to cognitive impairment and vascular brain injury, yet there can be low interrater reliability in assessing. We previously published an automated machine learning–based screening tool for microinfarcts/microhemorrhages trained on whole-slide images (WSIs) from a single institution, using sections from frontal, parietal, and occipital lobes. While internal validation demonstrated promising performance, concerns remain regarding the generalizability to other institution's data.

Methods: To evaluate cross-institutional robustness, we assembled two independent external cohorts. The first cohort consisted of 77 WSIs from the University of Washington stained with hematoxylin and eosin (H&E) and Luxol Fast Blue. The second cohort included 17 H&E-stained slides from the University of Southern California. Frontal, parietal, and occipital lobe regions were requested to keep brain areas evaluated consistent with training set. The model was applied without retraining. Outputs were assessed qualitatively to characterize screening patterns.

Results: Model performance varied when applied to datasets from new institutions compared to the original evaluation set. Slides stained with H&E/Luxol Fast Blue demonstrated domain shifts distinctive from the original training distribution, as observed in tissue heatmap quality. Qualitative review identified overall model failures such as not recognizing all brain tissue regions, which can adversely impact downstream screening.

Conclusions: These findings highlight limitations of single-center training and underscore the need for harmonization strategies. We hypothesize variations in model performance is due to staining protocols, differences in background staining intensity, tissue preparation, and scanner characteristics influencing prediction maps and candidate lesion localization. Multi-institutional validation should be considered essential prior to deployment of automated screening tools.

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Thermal imaging of white matter rarefaction and cerebrovascular lesions in postmortem brain tissue

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Background: White matter hyperintensities are common neuroradiological findings on T2-weighted or fluid-attenuated inversion recovery magnetic resonance imaging (MRI), reflective of underlying aging, hypertension and/or cerebral small vessel disease. These lesions can be difficult to assess, however, on gross examination of autopsied brain tissue. Herein, we sought to determine if thermal imaging (highlighting fluid retention in tissue) could serve to visualize white matter integrity and cerebrovascular lesions broadly on macroscopic tissue assessment.

Methods: Postmortem ex vivo neuroimaging and 3D scanning were performed prior to brain cutting. Formalin-fixed hemibrains were sectioned using custom 3D-printed tissue matrices, coronal slabs were photographed, and targeted regions were blocked (with all remaining tissue stored in phosphate buffered saline at 4°C). For thermal imaging, intact coronal slabs from the frontal and parietal lobes were submerged in ice-cold 70% ethanol for 30 minutes and then placed on a 50°C hot plate. Time-lapse thermal imaging (5 minutes) was captured using an iPhone-connected FLIR One® Pro camera. Images were analyzed using FLIR Ignite software.

Results: Tissue temperatures increased approximately 20°C over the 5-minute period (from ~8°C to ~28°C). Around 2-4 minutes, regions of parietal periventricular white matter reveal 4-5°C lower temperatures compared to other adjacent tissue regions (this difference normalized after 5 minutes). These regions demonstrated mixed degrees of overlap with T2-weighed ex vivo MRI signal intensity as well as white matter rarefaction metrics, as assessed on digital pathology analysis of hematoxylin & eosin and Luxol fast blue tissue slides.

Conclusions: Thermography is a promising, putative approach for detecting white matter hyperintensities, that utilizes accessible and user-friendly technology. Additional research is needed to more seamlessly integrate thermal imaging with neuropathology grossing workflows as well as to measure a wider range and distribution of cerebrovascular lesions in the brain.

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Novel Characterization of the Human Ischemic Penumbra: Multimodal Integrated Whole-Brain Magnetic Resonance Imaging (MRI) and Neurohistology

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Background: While clinical MRI distinguishes ischemic necrosis (“core”) from peri-infarct zones of potentially reversible injury (“penumbra”), the cellular level changes are spatially and temporally diverse and imperfectly modelled in experimental animals. Characterization of these changes directly in the human brain is critical for developing rational therapeutic targets early in ischemic stroke.

Methods: We studied the brains of two patients: a 69-year-old man with left hemisphere infarcts, confirmed by diffusion-weighted MRI 16 and 11 days before death, and a 74-year-old woman with acute symptoms but with a negative head CT; she died the next day, before an MRI could be done. Ex vivo MRI was followed by whole-brain cryosectioning into 20-micron sagittal sections, serially stained for Nissl and H&E, and immunostained for markers HIF-1α (cellular hypoxia), amyloid precursor protein (APP, axonal injury) and growth-associated protein 43 (GAP43, axonal regeneration). The modalities were co-registered for spatial relationships of cellular features in the MRI-defined penumbra and the rest of the brain.

Results: HIF-1α involved the cortex globally, including the contralateral hemisphere, revealing a brain-wide metabolic and molecular response not captured by standard histology or MRI (i.e., beyond the penumbra). APP was widespread in the white matter around the ischemic core, though not appreciated on H&E or Nissl, essentially broadening the geography of the peri-infarct zone. GAP43 marked many axons geographically overlapping the same territories of APP immunopositivity, with high-resolution histology confirming axon budding among bundles of damaged axons.

Conclusions: Our multimodal, integrated approach combining neuroimaging with histopathology enables precise spatial mapping of injured, plastic, and systemically stressed (hypoxic) regions, refining identification of vulnerable tissue and delineating therapeutic windows and targets beyond the infarct core. Our finding of colocalization of markers of both

axonal injury and repair are novel and hypothesis-generating, suggesting that zones of injury may also represent active substrates for structural plasticity.

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Post-Mortem Analysis of Coronary and Cerebral Atherosclerosis: Are They Related?

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Background: Coronary and cerebral atherosclerotic vascular disease (ASVD) are manifestations of systemic atherosclerosis and share common risk factors, including hypertension (HTN), diabetes mellitus (DM), & hyperlipidemia (HLD). However, few studies have examined their degree of correlation. We aim to investigate the severity and distribution of coronary and cerebral ASVD to evaluate their relationship and connection to underlying risk factors.

Methods: We reviewed 100 unrestricted autopsies between 2022 - 2025. Demographics, risk factors, and morphologic data were obtained from autopsy reports. ASVD of coronary arteries, aorta, and circle of Willis was graded by percent luminal stenosis: grade 0 (0%), grade 1 (< 24%), grade 2 (25–49%), grade 3 (50–69%), and grade 4 (70–100%). The maximum grade per vascular bed was used for inter-territorial comparisons.

Results: Coronary ASVD was present in 92% of patients, aortic ASVD in 70% and cerebral ASVD in 52%. Coronary ASVD appeared earliest (ages 11–20), followed by aortic (21–30) then cerebral (31–40), with the left anterior descending and basilar arteries most frequently involved. Coronary-cerebral grades were equal in 24% of cases (70% with higher coronary scores and 6% with higher cerebral scores). Coronary–aortic grades were equal in 43% (36% with higher coronary scores and 21% with higher aortic scores). Cerebral–aortic grades equal in 28%. All patients with DM (n=30), HTN (n=43), or HLD (n=28) had coronary ASVD, while cerebral (DM n=17, HTN n=28, HLD n=20) and aortic ASVD (DM n=26, HTN n=38, HLD n=24) showed weaker and intermediate associations, respectively.

Conclusions: Coronary arteries are prone to early and severe ASVD. While patients with aortic and cerebral ASVD showed some differences, the largest disparity was between coronary and cerebral ASVD. Cardiac ASVD was closely linked to metabolic & vascular risk factors, whereas cerebral & aortic ASVD showed weaker associations. The mechanisms underlying these differences remain poorly understood, highlighting the need for further investigation.

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Neurovascular calcification and brain injury in chronic kidney disease

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Background: Chronic kidney disease (CKD) is a serious public health burden, affecting over 30 million adults in the United States. A common complication of CKD that increases morbidity and mortality is vascular calcification. One organ system that is particularly susceptible is the central nervous system (CNS), where pathological calcification of cerebral arteries has been linked to multiple components of cerebral microvascular disease including white matter injury, deep infarcts, and microbleeds. There has not been a systematic quantification of the relationship between CKD and calcification-related cerebral injury.

Methods: Post-mortem human brain sections obtained from patients who underwent autopsy at a high-volume academic center in the United States were sampled and stained using immunohistochemical techniques to assess for degree of calcification, iron deposition, and microgliosis. CD31 and SM22a were employed for localization of calcifications in affected cerebral vessels. Patient variables such as demographics, comorbidities, CKD stage, CKD duration, and clinical complications were extracted from the electronic medical record. Patients were separated into CKD and control groups, with CKD defined as eGFR < 60 mL/min/1.73m2. Age-matching was performed for final assessment.

Results: A total of 98 autopsy patients were identified, and samples were obtained for analysis. Median age was 74 (IQR 68-82), and the cohort was 37% female. Preliminary classification of CKD groups yielded 34% CKD Class 2, 33% CKD Class III, 21% CKD Class IV, and 13% CKD Class V. An eGFR < 60 mL/min/1.73m2 was calculated for 66% of included patients. Staining was completed for all brain samples. Preliminary analysis identified robust microglial activation alongside calcified vessels, along with signs of white matter injury and microhemorrhages.

Conclusions: Immunohistochemical staining and quantification of cerebral microvascular disease is an effective technique to measure the impact of CKD on calcium-related injury. Anti-calcific therapeutics trialed in a parallel animal model may yield future benefits in patients affected by CKD.

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Effect of Long-term Dialysis use for Chronic Kidney Disease on Cerebral Vascular Calcifications

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Background: Chronic kidney disease (CKD) is a serious public health burden, affecting over 30 million adults in the United States. A common complication of CKD is vascular calcification via dysregulation of normal calcium and phosphorus metabolism. The central nervous system (CNS) is particularly susceptible to vascular calcification, which can lead to white matter injury, deep infarcts, and microbleeds. Progressive CKD eventually requires dialysis unless transplantation is performed. However, the effect of dialysis on calcification-related CNS outcomes in the context of CKD is unknown. Here, we conduct a preliminary examination of chronic hemodialysis patients with cerebral calcifications.

Methods: Patients were identified from a dedicated database of CKD patients with available post-mortem brain tissue. Patients were included if they had been on hemodialysis for a minimum of 1 year. Demographics and patient characteristics were obtained from electronic medical record review. Clinical outcomes of interest were cognitive decline and development of clinical stroke. Brain histopathology was performed including immunohistochemical and histochemical stains to assess for degree of cerebral vascular calcification, iron deposition, microgliosis, and precise localization of vascular calcification to the endothelium.

Results: A total of 13 patients with CKD who underwent at least one year of hemodialysis and had available post-mortem brain tissue were identified. The median age was 71 (IQR 67-78) and 62% (8/13) of patients were male. The median glomerular filtration rate was 34 mL/min/1.73m2, and median duration of dialysis dependence was 5 years. Cerebral vascular calcifications were identified in 85% (11/13) of patients. Of patients with no cerebral vascular calcifications, 50% (1/2) had experienced stroke, while 55% (6/11) of those with cerebral vascular calcifications experienced stroke.

Conclusions: A single-arm retrospective analysis identified a high rate of cerebral vascular calcification in CKD patients who receive chronic hemodialysis. Further quantification and localization may identify a relationship between dialysis duration, calcification burden, and clinical outcomes.

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Unusual perivascular iron deposition in a 46 year old with remote hemorrhagic frontal lobe vascular malformation

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Background: This 46 year old male patient initially presented approximately 5 years prior with seizures and was found to have a left frontal opercular cavernous malformation that was confirmed histologically at an outside institution. Initially following resection, the patient's epilepsy improved dramatically and was managed without medication until 2023 when seizures began to recur and progressively worsen despite medical therapy. Brain MRI revealed a coarse T2 hyperintense focus in the left frontal lobe with hemosiderin deposition and central enhancement, consistent with chronic and focal acute hemorrhage with a possible residual vascular lesion. The patient underwent resection of the affected area for seizure control.

Methods: The specimen was evaluated by the University of Washington Neuropathology service using routine hematoxylin and eosin (H&E) staining as well as special stains to highlight different components of the lesional tissue.

Results: H&E stained slides showed fragments of brain with gliosis, focal aggregates of hemosiderin-laden macrophages, patchy procedural hemorrhage, and abundant fine branching capillaries throughout the affected cortex. The majority of these capillaries exhibited thin layers of black-tinged mineralization. Few larger thickened vessels with associated fibrosis were identified, lacking an elastin layer, suggestive of a focal residual vascular lesion. A Von Kassa special stain failed to highlight the perivascular mineralization, which instead showed strong positive iron staining, consistent with perivascular ferrugination.

Conclusions: Limited examples of this histology appear in the literature, though there is a growing body of literature describing the role of pericytes in iron homeostasis following vascular insult, hemorrhage or contusion, and their vulnerability to ferroptosis in states of iron overload. It is possible that the florid perivascular iron deposition we observe in this case may represent a chronic dystrophic pattern of ferroptosis in response to long-term low-level hemorrhage resulting in an unusual epileptogenic lesion.

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Spongiform Leukoencephalopathy in an Autopsy Case of Catastrophic Antiphospholipid Syndrome-like Illness Following COVID-19 Vaccination

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Background: Vaccine-related antiphospholipid syndrome (APS) is less widely known and recognized among reported serious adverse events after COVID-19 vaccination.

Methods: A 69-year-old man with a history of hypertension, hyperuricemia, and diabetes, who smokes 10 cigarettes daily for 49 years, received two doses of the COVID-19 mRNA vaccine (BNT162b2) at three-week intervals. Ten days after the second dose, he noticed cyanosis in his right leg. On the 20th day post-vaccination, he suddenly lost consciousness and presented with right-sided hemiplegia due to an acute infarction in the left middle cerebral artery (MCA) territory caused by occlusion of the ipsilateral cervical internal carotid artery (ICA). Coronary angiography showed no abnormalities. Laboratory tests revealed a normal platelet count and normal ADAMTS13 activity, but showed elevated inflammatory markers, D-dimer, serum antinuclear antibodies (2560-fold: Speckled x2560, Centromere x160), and anticardiolipin antibodies (IgM 38, IgG 6 U/mL). A floating thrombus in the left popliteal artery was identified by ultrasound. He died on the 25th day after the second dose.

Results: The autopsy confirmed fresh red thrombotic occlusions in the left cervical ICA, major arteries within the circle of Willis, and the right common iliac and pulmonary arteries. Microscopic examination revealed organizing thrombi in the mesenteric artery, along with mesenteric necrosis. A hemorrhagic infarct in the left MCA territory was confirmed, with mixed thrombi in the pial veins, along with occasional hyaline eosinophilic platelet microthrombi and mural fibrin in the parenchymal microvessels. Additionally, white matter microvacuolation was present throughout the brain, characterized by myelin sheath ballooning with intact axons. The 5,10-methylenetetrahydrofolate reductase gene polymorphism, 677CT/1298AC, was identified by genetic analysis using paraffin sections.

Conclusions: This patient, who has a predisposition to autoimmune disease, developed a catastrophic APS-like condition, likely due to abnormal immune responses after vaccination, and thrombosis may have been exacerbated by a potentially reduced reserve capacity for folic acid.

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A Case of Transthyretin Leptomeningeal Amyloidosis

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Background: Transthyretin amyloidosis (ATTR) is a disorder caused by misfolding of transthyretin protein, which is deposited as amyloid fibrils. While commonly causing polyneuropathy and cardiac amyloidosis, it can rarely involve the Central Nervous System in the form of Leptomeningeal disease.

Methods: Formalin fixed paraffin embedded tissues subjected to histochemical stains along with mass spectrometry.

Results: A 46-year-old male with a history of traumatic brain injury, Wolff-Parkinson-White syndrome, seizures, and headache developed recurrent episodes of expressive aphasia and dysarthria, which first began 3 years prior to presentation to our institution. He was not known to have seizures prior to his initial presentation. The family history is negative for senile cardiac amyloidosis. Multiple prior MRIs of the brain and spinal cord revealed diffuse leptomeningeal enhancement in the brain and spinal cord including the conus medullaris, with enhancement of cauda equina nerve roots. Cerebrospinal fluid analyses were unrevealing. Lacosamide treatment was initiated and resolved his episodes of aphasia and dysarthria. He then underwent a right frontal meningeal and brain biopsy. Histopathology of the brain tissue revealed a thick layer of amorphous pink material within the subarachnoid space that also infiltrated the walls of subarchnoid vessels, but not intraparenchymal vessels. A Congo Red stain demonstrated apple green birefringence under polarized light. Liquid chromatography tandem mass spectrometry identified the amyloid as ATTR (transthyretin amyloid), with an amino acid sequence suspicious for mutated ATTR. 99mTechnetium-Pyrophosphate Imaging noninvasively detected cardiac amyloidosis. Treatment with tafamidis was recently started. Subsequent MRIs of the neuroaxis revealed stable leptomeningeal enhancement in the brain and spinal cord. Further genetic testing is pending.

Conclusions: Although rare, leptomeningeal amyloidosis due to ATTR should be considered in patients with persistent leptomeningeal enhancement when no other clear cause has been identified.

Posters: Demyelinating and Inflammatory

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Postmortem Diagnosis of Limbic Encephalitis Following Suicidal Acetaminophen Overdose
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Background: Limbic encephalitis is a pattern of central nervous system inflammation and injury, which can result in seizures, behavior and cognition changes, and upper motor neuron signs. Underlying causes are diverse and include autoimmune, viral, and paraneoplastic etiologies

Methods: The decedent was a 23-year-old woman with a history of psychotic episodes which acutely progressed to suicidal ideation. She was brought to the emergency department after ingesting a bottle of acetaminophen, as prompted by “the voices in her head.”. Laboratory testing revealed toxic acetaminophen levels for which she was treated. Unfortunately, she progressed to develop acute liver failure with hepatic coma. Physical exam was also significant for clonus and hyperreflexia of uncertain etiology. Life sustaining therapies were withdrawn due to her poor neurological prognosis. Complete medicolegal autopsy was performed.

Results: Autopsy examination showed liver injury consistent with acetaminophen toxicity. Microscopic brain examination revealed acute and organizing injury involving the limbic system, white matter tracks, and brainstem; accompanying gliosis was indicative of a chronic timeframe. No neoplasia or central nervous system viral or other infection were identified. Cause of death was acetaminophen toxicity in the setting of limbic encephalitis. Manner of death was suicide.

Conclusions: Postmortem examination identified limbic encephalitis that was felt to contribute to the decedent’s psychiatric disturbance and ultimately fatal toxic ingestion. Limbic encephalitis also explains her upper motor neuron signs, typically not seen in hepatic encephalopathy. The most likely etiology is autoimmune limbic encephalitis; postmortem evaluation for autoantibodies is in progress. This is a treatable and potentially curable condition with combination glucocorticoids, IVIG, therapeutic plasma exchange, and rituximab.

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The Multifaceted ADEM: A Biopsy-Proven Case Series from Pediatric to Late-Onset Patients

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Background: Acute disseminated encephalomyelitis (ADEM) is a rare immune-mediated demyelinating disorder of the central nervous system (CNS), classically described in children but increasingly recognized in adults, where it may present with atypical and diagnostically challenging phenotypes. Adult and late-onset ADEM presentations can mimic neoplasm, infection, cerebrovascular disease, or multiple sclerosis (MS) on neuroimaging, often leading to misdiagnosis.

Methods: We retrospectively reviewed six ADEM cases (one pediatric, five adult/late-onset) presenting with radiological features concerning for malignancy, infection, or cerebrovascular disease, who underwent brain biopsy. Clinical, radiologic, and cerebrospinal fluid (CSF) findings were reviewed. Serologic studies (including anti–myelin oligodendrocyte glycoprotein (anti-MOG) and anti-neuromyelitis optica/aquaporin-4 immunoglobulin G (anti-NMO/AQP4) antibodies), infectious workup, and histopathologic findings were analyzed. Treatment regimens and clinical and radiologic outcomes were recorded.

Results: The Pediatric case presented with a tumefactive demyelinating lesion (TDL) and a monophasic course, showing rapid significant clinical and radiological improvement following Intravenous Immunoglobulin (IVIG). Adult cases showed diverse radiologic phenotypes, including TDLs, multifocal white matter lesions, diffuse leukoencephalopathy with hemorrhagic features, and deep grey matter and spinal involvement, often mimicking neoplasm, abscess or cerebrovascular diseases. Histopathology consistently revealed macrophage-rich inflammatory demyelination with no evidence of malignancy or infection. Serum anti–MOG and anti-NMO/AQP4 antibodies were negative in all but one patient. Most patients showed clinical and radiologic improvement with immunotherapy, though severe or late-onset presentations required escalated therapy, and one patient had persistent lesions with residual neurologic deficits. All cases followed a monophasic course, however one patient later developed optic neuritis consistent with ADEM-optic neuritis (ADEM-ON).

Conclusions: Adult- and late-onset ADEM shows marked clinical and radiologic heterogeneity, often posing significant diagnostic challenges. Integrated clinico-radiologic and histopathologic evaluation is essential for accurate diagnosis. Early recognition and prompt immunotherapy can improve outcomes, and ADEM should remain a key diagnostic consideration in intracranial lesions across all age groups.

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Adult-Onset Leukodystrophy with 5q23.2 Deletion. A Post-Mortem Case Report.

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Background: Adult-onset demyelinating leukodystrophy, is a rare and fatal neurologic disorder, affecting adults in the 4th to 5th decades. The typical form, presents with autonomic, pyramidal and cerebellar dysfunction, and a tandem duplication on chromosome 5q23, resulting in an extra copy of the lamin B1 (LMNB1) gene. An atypical variant with mainly pyramidal symptoms and deletions on chromosome 5q23, upstream of the lamin B1 gene, have been described.

Methods: This is a 47-year-old man with history of learning difficulties and chronic gait disturbances, who was brought to the emergency department when his roommate found him unconscious, at the bottom of a 12 flight of stairs, presumably after sustaining a fall. EMS noted decorticate posturing, and intubated him. Spine and brain CT scans showed a right posterior parietal scalp hematoma without evidence of fractures, subluxation or intracranial hemorrhages. However, extensive periventricular white matter hypoattenuation, was noted, which on MRI showed extensive supra- and infratentorial white matter signal abnormality, with chronic changes, such as lack of mass effect and edema, diffusion restriction, or enhancement, and with coexisting volume loss suggestive of toxic-metabolic etiologies, demyelinating or inherited disorders, such as an adult-onset leukodystrophy.

Results: During an extensive hospital admission, he remained minimally responsive and with stimulus-induced myoclonus, with a tracheostomy and PEG. He was complicated by extensive DVT and pulmonary embolism, aspiration pneumonia and eventually expired. Gross and microscopic examination showed extensive myelin loss and associated axonal loss throughout the brain and spinal cord, with the frontal lobes more severely affected. Comparative Genomic Hybridization identified a 264 KB deletion on chromosome 5q23.2, encompassing 5 genes: 3 OMIM-listed genes (ALDH7A1, GRAMD2B and PHAX) and two Non-OMIM genes (TEX43 and LMNB1-DT).

Conclusions: Approximately 10 cases, with genomic deletions upstream of the lamin B1 gene, have been reported as a distinct and alternative pathway, and termed atypical adult-onset demyelinating leukodystrophy.

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Choroid Plexus Macrophage Heterogeneity and Non-Canonical Titin Expression at the Human Brain–CSF Interface

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Background: The choroid plexus (ChP) is a critical interface between the peripheral immune system and the central nervous system, yet its resident immune cell populations remain incompletely characterized in humans. While ChP macrophages are increasingly recognized as functionally distinct from parenchymal microglia, their molecular specialization and relationship to tissue architecture are poorly understood. Here, we characterize human choroid plexus macrophage populations using an integrated histologic, single-nucleus RNA sequencing, and spatial transcriptomic approach.

Methods: Post-mortem human ChP tissue was examined using routine histopathology, immunohistochemistry, and transcriptomic profiling. We identified multiple transcriptionally distinct macrophage populations localized to specific ChP microanatomic niches, including stromal cores and epithelial-adjacent regions. These macrophages exhibit gene expression programs consistent with immune surveillance, antigen presentation, and barrier-associated signaling, supporting the concept of the ChP as an active immunoregulatory site rather than a passive barrier.

Results: Notably, we observed selective expression of the giant structural protein titin (TTN) within ChP-associated macrophages. Titin, classically studied in striated muscle, has not previously been implicated in CNS immune cells. Spatial localization and transcriptional co-expression patterns suggest that TTN expression may be associated with cytoskeletal organization or mechanosensitive states unique to the ChP environment. These findings raise the possibility that macrophages at the brain–CSF interface adopt specialized structural adaptations in response to local mechanical or fluid dynamic cues.

Conclusions: Together, this work expands the neuropathologic understanding of choroid plexus immune biology and identifies titin as a novel molecular feature of human CNS interface macrophages, with potential implications for neuroinflammatory and neurodegenerative disease.

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Extracellular Vesicles from early-stage pancreatic cancer contributes to paraneoplastic neurological syndrome

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Background: Pancreatic ductal adenocarcinoma (PDAC) is a leading cause of cancer-related mortality and is frequently associated with paraneoplastic syndromes. While gastrointestinal manifestations are well characterized, neurological complications remain poorly understood. Emerging evidence suggests that extracellular vesicles (EVs) secreted by PDAC cells (PCEVs) may contribute to central nervous system (CNS), particularly blood brain barrier (BBB), dysfunction. We hypothesized that PCEVs cross the blood–brain barrier (BBB), accumulate in neural tissues, and induce microglial dysfunction and neuronal damage, resulting in behavioral deficits.

Methods: Using genetically engineered mouse models and orthotopic PDAC models expressing mCherry-CD9–labeled EVs, we assessed neuronal integrity and microglial pathology. Immunofluorescence was utilized to assess PCEV accumulation in the brain, neuronal apoptosis, and microglial pathology. Then, isolated PCEVs were retro-orbitally injected into wild-type mice to evaluate potential motor deficits with RotaRod testing.

Results: PCEVs were detected in the cerebellum, hippocampus, cortex, and thalamus, with the cerebellum showing the highest accumulation. On average, 30% of cerebellar and hippocampal neurons were EV-positive, compared to 1–2% in controls. Histological analyses revealed extensive apoptosis of both neurons and glia (up to 70%), confirmed by TUNEL and Caspase-7 assays. Microglia exhibited pathological hyper-ramified morphologies and reduced phagocytic activity, indicating chronic stress and impaired clearance. Mice injected with PDAC EVs demonstrated significant motor deficits on RotaRod testing and cerebellar apoptosis within two weeks, whereas controls EVs had no comparable impact.

Conclusions: These findings establish a mechanistic link between PDAC-derived EVs and CNS dysfunction, suggesting that PCEVs act as mediators of paraneoplastic neurological syndromes. Future studies will explore EV cargo, trafficking mechanisms, and translational biomarkers to inform therapeutic strategies aimed at mitigating neurological complications in PDAC.

Posters: Forensics/Trauma

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Withdrawn

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The Blast Exposure Likelihood Level (BELL) Scale: Multi-Factorial and Retrospective Assessment of Military Blast Exposure for TBI Research

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Background: Blast exposure is among the most common causes of traumatic brain injury in modern warfare, and studies regarding the neuropathology of blast injury are underscored by the critical need for a reliable means of assessing lifetime blast exposure in deceased service members. Current tools predominantly utilize living individuals and rely heavily on first-person accounts, which are unavailable in a post-mortem setting. Military occupational specialty (MOS) is insufficient as a lone proxy of exposure due to a lack of standardization defining high-risk occupations (HROs) across branches and documentation disparities.

Methods: The Department of War/Uniformed Services University Brain Tissue Repository (DoW/USU BTR) developed the Blast Exposure Likelihood Level (BELL) scale. This inferential methodology retrospectively examines cumulative blast exposure by analyzing seven categories of service and medical history: MOS, time in service, era of service, deployment history, combat exposure, campaign citations, and military/medical records. A triangulation approach is utilized to cross-reference reports from historians with available records. Corroboration of subjective reports with objective records provides context for retrospective assessment and mitigates documentation disparities.

Results: The BELL scale provides structured architecture for characterizing blast exposure likelihood. Traditional classifications of HROs in the literature often overlook nuanced aspects of service, contributing to overall risk. A multi-source approach allows consideration of exposures across service branches and occupations. Furthermore, this methodology enables the correlation of available records and supplemental information from historians to generate a composite assessment without reliance on first-person reports

Conclusions: The BELL scale offers a reproducible methodology to bridge the gap between service history and neuropathological findings. Standardizing retrospective assessment is essential to research investigating potential correlations between blast exposure and brain injury. Representative case examples will be presented. The opinions/assertions of the authors herein do not reflect official policies/positions of the DoW, USU, or the Henry M. Jackson Foundation for the Advancement of Military Medicine, Inc.

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Blast Injury Results in Widescale Cell Damage in the Human Cerebellum

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Background: In recent years, the cerebellum has emerged as an important neural structure controlling many functions in addition to balance and coordination, including cognition, emotion, and sleep. Unfortunately, we know little about the precise effects of traumatic brain injury on the cerebellum. Occupational blast exposure from artillery, explosives, and other weapons is a key characteristic of military training that can lead to repeated microtraumas in the brain.

Methods: To assess the impact of blast related injuries on the human cerebellum, we evaluated the histopathology of warfighters who were either exposed to multiple explosive blasts or control subjects not known to experience such injuries. The brain samples were obtained from the DoD/USU Brain Tissue Repository.

Results: In individuals exposed to blasts, we observed a dramatic loss of Purkinje cell immunoreactivity for calbindin throughout the cerebellum, especially evident in ventral cerebellar regions. The Purkinje cell dendritic morphology revealed by calbindin immunoreactivity was also stunted. In addition, Bergmann glia exhibited altered morphology showing a strong beaded pattern rather than relatively smooth processes normally present in cerebellar cortex. We also observed abnormal appearing astrocytes immunoreactive for aquaporin 4, primarily located at the interface of cerebellar cortex and white matter. The cerebelli of control brains showed relatively normal Purkinje cell numbers and distribution, although we observed variability in different subjects not known to receive brain injury.

Conclusions: This suggests that blast traumatic brain injury can substantially alter Purkinje cell number, distribution and dendritic morphology, as well as glial cell morphology, which likely impacts multiple neurologic functions. The variability in Purkinje cell number of control subjects also indicates that multiple life experiences may influence the health and morphology of Purkinje cells.

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A Unique Case of Fatal Intracranial Penetrating Injury from a Toothbrush: No Cavity Left Unexplored

L Evans, R Mittenzwei, A Penev; University of Washington

Background: This 70-year-old decedent was in custody when he was reportedly assaulted by his cellmate. Per report, the cellmate first choked and then used a toothbrush as an improvised weapon to stab the decedent. When the altercation was halted and the decedent transported to Kadlec Regional Medical Center, the decedent was noted to be bleeding from the left eye with associated displacement of the globe.

Methods: CT Head imaging was performed and noted a foreign body object passing sequentially through the left orbit, posterior left ethmoid air cells, bilateral sphenoid sinuses, right temporal lobe, and right cavernous sinus with associated right internal carotid dissection, pneumocephalus, and intraparenchymal hemorrhage. Interestingly, the globe remained intact. Following foreign body removal, his month-long hospital course was complicated by non-ST-segment elevation myocardial infarction, gastrointestinal bleeding with bowel ischemia and pneumonia progressing to respiratory failure with evolving intracranial infarctions. Following his death on comfort measures, the King County Medical Examiner's Office assumed jurisdiction and performed the autopsy and neuropathologic examination.

Results: Autopsy exhibited gross and histologic findings correlating with the hospital course. Neuropathologic examination revealed subacute-to-chronic bilateral subdural neomembranes with evidence of operative intervention, as well as a right inferior temporal cavitary wound track with chronic features. Also noted were acute on chronic vascular territory infarcts predominantly involving the right middle cerebral artery territory and, to a lesser extent, bilateral anterior cerebral artery territories, most likely corresponding to the reported penetrating injuries to the right internal carotid artery and left sphenoid sinus, respectively.

Conclusions: This case demonstrates a unique pattern of penetrating injury from an improvised weapon in the setting of incarceration with a prolonged survival interval, pre- and post-mortem imaging findings, and multiple gross and histologic findings demonstrating evolving vascular and parenchymal injuries due to multiple involved craniofacial anatomic compartments.

Posters: Other

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Accelerating Brain Transcriptomics with CARIBOU: Computational AI Research Interface for Bioinformatics, Omics, and Unifying Agents

N Shirooni, D Riffle, P Sureshkumar, V Vijay, M Rose; University of California Irvine

Background: The scale and complexity of modern biological data (such as single cell transcriptomic data from the brain) have outpaced human-led, manual analysis. We propose a new paradigm with CARIBOU, an open source framework for automated, reproducible bioinformatics analysis where scientists act as high-level strategists overseeing autonomous AI research agents.

Methods: CARIBOU consists of four conceptual pillars: architecture, ingenuity, competency, and scalability.

Results: CARIBOU's architecture enables the design and deployment of in-silico research groups, teams of specialized AI agents that collaboratively plan, execute, and interpret complex bioinformatics workflows within a secure sandbox. It uses large language models (GPT and DeepSeek). For ingenuity, CARIBOU introduces a novel, automated meta-cognitive loop where the system executes an analysis, receives feedback on performance, and then iteratively improves its scientific and analytical reasoning. We demonstrate this by deploying an agent team to autonomously perform an end-to-end analysis of single-cell datasets, showing its ability to self-improve its analytical rigor based on Rank-Surprise Ratio. For competency, we benchmarked CARIBOU's analysis of three landmark single cell RNAseq datasets against published manual analyses: mouse hippocampus from the Allen Brain Atlas, human large intestine from the Tabula Sapiens Consortium, and human peripheral blood mononuclear cell (PBMC) from 10x Genomics. CARIBOU achieves expert-comparable analysis quality with greatly improved time efficiency, validating practical deployment. For scalability, CARIBOU performs parallel execution on institutional high-performance computing (HPC) clusters, enabling repository-scale systematic reanalysis.

Conclusions: By ensuring transparency through the generation of digital lab notebooks, ingenuity enabling robustness across real-world datasets, competency with improved time efficiency, and with support for HPC-native execution environments, CARIBOU provides a reliable and scalable platform to accelerate the pace of biological discovery.

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Microscopy of nanoplastics in human brains: Association with evidence of pathology
E Bearer; University of New Mexico School of Medicine

Background: Micro/nano plastics (MNP) are everywhere, including the human brain. We noticed perivascular glossy deposits and tested whether these might represent plastics by submitting adjacent tissue for pyrolysis gas chromatography/mass spectroscopy (PyrGC/MS).Ten different types of microplastics were found. However, methodological issues may have overestimated amounts of these MNP due to interference with lipids remaining in enriched pellets.

Methods: We performed microscopy on plastics enriched pellets prior to pyrGC/MS. Aliquots were mounted on EM grids and negative stained, or after sonicating in ethanol. Laser confocal microscopy (LSM) was performed on plastic-enriched pellets, pure plastics and brain sections.

Results: By negative stain EM, we discovered several types of particles as well as abundant membrane-like vesicles. Sonication and ethanol resuspension revealed electron-dense shards. By LSM, we identified an excitation/emission spectra specific to different types of plastics. Lambda scans on glossy deposits within human brain found that they fluoresced with the same specific profile as pure polyethylene and polypropylene. Little to no emission in this spectrum was detected in tissue or non-tissue. We then designed a dual fluorescence microcopy system that matched the unique excitation/emission spectra of the plastics and had a second excitation/emission spectra for tissue. We found plastics embedded in the medial muscle of subcortical penetrating arterioles in Alzheimer's disease and in adventitia of subcortical vessels in Binswanger's disease. Abundant in brains from 2023-2024 but minimal in the 1960's; composed of different types of plastic particles in the same individual (shapes and sizes, and excitation/emission spectra) and between individuals; were primarily located in the walls of blood vessels but also sprinkled throughout the parenchyma; at highest level with microhemorrhage and vascular leakage; engulfed by CD68 macrophages; surrounded by loose amyloid but not consistently within amyloid plaques; and no evidence of association with tau phosphorylation.

Conclusions: We conclude that plastics may cause mechanical, chemical and/or physiological damage.

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Amyloid Burden in Ligamentum Flavum Differs by Amyloid Subtype in Lumbar Spinal Stenosis

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Background: The ligamentum flavum (LF) is a site of amyloid deposition in approximately 25-45% of patients with lumbar spinal stenosis (LSS). A subset of these patients later develop cardiac amyloidosis (CA), the primary driver of morbidity and mortality in systemic amyloidosis. Wild-type transthyretin amyloid (ATTRwt) and indeterminate type amyloid comprise the majority of LF amyloid deposits. It is hypothesized that ATTRwt deposition may confer a higher risk of myocardial involvement, however this remains unknown. Quantitative differences in amyloid burden between subtypes may provide insight into the clinical utility of amyloid subtyping with expensive tools such as mass spectrometry.

Methods: Our institution has assessed a total of 531 patients for amyloid deposition in the LF after decompressive surgery for LSS between January 2018 and February 2026. 131 patients have amyloid depositions, confirmed with Congo Red stain. Amyloid subtyping with mass spectrometry was performed on all positive cases by the Mayo Clinic. Herein, we used a Trainable Weka Segmentation machine learning classifier previously developed by our team in Fiji ImageJ to quantify and compare amyloid load in 15 patients with indeterminate-type amyloid and 33 patients with ATTRwt in their LF.

Results: Amyloid load differed significantly between patients with indeterminate amyloid and those with ATTRwt. The mean amyloid load was 2.85% in the indeterminate group and 4.42% in the ATTRwt group ($p = 0.0002$), demonstrating a higher degree of deposition in the ATTRwt specimens.

Conclusions: ATTRwt deposition in the LF is associated with a significantly greater amyloid burden compared to indeterminate amyloid, suggesting subtype-specific differences in the deposition of amyloid. Quantitative assessment of amyloid load may provide incremental value beyond binary amyloid positivity and help refine risk stratification for further cardiac evaluation in patients. Future longitudinal studies should explore whether increased LF amyloid burden predicts systemic or myocardial involvement.

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Withdrawn

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Factors Associated with Autopsy Performance at a Psychiatric Institution in the 1930s
M Silver, D Wheeler, H Ames; University of Maryland School of Medicine

Background: The legal requirements for the performance of autopsy have changed greatly in the United States over the last century. In the 1930s, many states lacked clear guidance for the medical examination of the deceased. While the right of refusal by the family often existed, other circumstances, including the possibility of communicable disease, provided justification of autopsy performance for public health purposes. Understanding the context of historical autopsy procedures adds to our understanding of the evolution and trends of the practice.

Methods: Autopsy information was reviewed for 1012 patients included in a logbook cataloging all patient deaths at St. Elizabeth Hospital between the years 1934 and 1937. Deidentified information from this death log was digitized and analyzed for trends in autopsy performance and results including clinical diagnoses, patient age, cause of death, admission length, permission for autopsy, and type of burial site.

Results: In our analysis of 1012 death log entries, we found that autopsies were refused for 21% of deceased patients and performed in 68%. The most common diagnosis among all deaths was "dementia praecox", an antiquated term for what we now call schizophrenia, with these patients demonstrating the longest duration of admission. The most common cause of death was lung disease (27%). Organic brain diseases, including cerebrovascular disease, and syphilitic meningoencephalitis, are among the listed causes of death in both autopsied patients and those buried without examination.

Conclusions: Performance of autopsy procedures in the early 20th century informs ethical and medical concerns to consider when recommending autopsy in the present day. The context of autopsy in psychiatric inpatients adds additional complexities demonstrated in our data. Additionally, this modern review provides additional practical insight into historical criteria for neuropathological diagnoses - suggesting potential alternate diagnoses in the modern era.

Posters: Developmental/Pediatric

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Hsd17b7 is Required for Proper Mammalian CNS Development

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Background: Disorders of cholesterol biosynthesis are genetic diseases that can have life-threatening consequences for early development. Clinical features include intellectual disability, craniofacial developmental anomalies, and a range of cerebral and cerebellar malformations. Current therapeutic strategies primarily consist of medical management of clinical signs and have little effect on clinical progression. Hydroxysteroid 17-Beta Dehydrogenase 7 (Hsd17b7) is a gene which encodes for a post-squalene cholesterol biosynthesis enzyme. The rudolph allele is a hypomorphic allele of Hsd17b7 and shows striking defects in the central nervous system (CNS) and skeletal development. The objective of this study is to investigate the consequences of complete Hsd17b7 deletion on the CNS.

Methods: Whole embryo ablation of Hsd17b7 is embryonically lethal early in development. To overcome this, we have utilized mouse Cre-LoxP systems to ablate Hsd17b7 from the neural crest using Wnt1cre2 and the forebrain with Emx1cre. Phenotypes in mutant mice are characterized through gross examination, histology, skeletal preparations to highlight cartilage and bone, immunohistochemistry, and behavioral assays. The underlying mechanisms are then investigated with Cre reporter mice, wholemount immunohistochemistry and RNA in-situ hybridization.

Results: Wnt1cre2/wt;Hsd17b7flox/wt mice reveal cerebellar dysgenesis linked to changes in the midbrain-hindbrain boundary, hypoplasia of the midbrain, severe hydrocephaly linked to malformation of the subcomissural organ, and craniofacial anomalies. Emx1cre/wt;Hsd17b7flox/wt mice have a severely hypoplastic forebrain. Based on the noted phenotypes, we suspect that loss of Hsd17b7 in the Wnt1cre2 mice leads to early apoptosis of the Wnt1 lineage, directly compromising development of the cerebellum and the craniofacial primordia. Loss of Hsd17b7 in Emx1cre mice is anticipated to hinder smoothened function through decreasing access of cholesterol, inhibiting proper hedgehog signaling.

Conclusions: Our work shows that Hsd17b7 is required for proper CNS development; a more comprehensive understanding of how the disruptions of the cholesterol biosynthesis pathway impact fetal development offers an opportunity to develop more effective therapeutic strategies.

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An optimized transcriptomic toolbox for rare cell types in neurodevelopment

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Background: Some neurologic diseases affect specific subpopulations, such as in motor neuron (MN) disorders like Amyotrophic Lateral Sclerosis and the Congenital Cranial Dysinnervation Disorders. Identifying rare subpopulations among merged transcriptomic data is critical to assess gene expression differences.

Methods: We developed a protocol to best analyze developmental single cell and spatial transcriptomics data from embryonic mice across time and space. We identified an unbiased semi-automated approach to perform quality control compared to a manual assessment. We utilized rigorously benchmarked tools to handle doublets and ambient RNA.

Results: To QC cells we used median absolute deviation for summary statistics of each cell. For celltyping, CellTypist provided a more rapid means to annotate data. Identifying cell types across ages from both single cell and nuclei samples required challenging integration. We identified scDREAMER as a top performer with the ability to correct across complex nuclei to cell batch effect to generate a harmonized atlas across embryonic ages E9.5-E18.5. To study subpopulations, we developed a “bridging” approach. We find that developmental programs dominate early neuronal data causing subpopulations to link in a bridge-like manner. We quantified the intermediate states between populations by analyzing population connectivities and defined bridge cells as a quantile of connectivity scores between the linked populations. Removing these cells identifies core populations. Lastly, for Slide-Sequencing spatial transcriptomics data, we found a method of integrating single cell with spatial data out-performed deconvolution tools for identifying subpopulations. We also built an image editing tool to efficiently extract slide-seq regions of interest with anatomic orientation correction for improved analysis.

Conclusions: This study offers an optimized toolbox for analyzing gene expression differences among rare subpopulations in the developing nervous system.

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A Distinct Population of Neural Progenitor Cells in the Developing Hippocampus

A Sandoval, T Mastalski, D Nauen; Johns Hopkins Hospital

Background: Hippocampal development includes the approximation and recession of the lateral ventricle ependyma, a process which results in the formation of a “seam” below Ammon’s horn. In this process, a population of germinal matrix progenitor cells along the seam is left deep in the white matter of the hippocampus. This study aims to characterize the movement, protein expression, and persistence of this cell population.

Methods: 34 fetal/infant autopsies between 20 to 144 weeks post conception were identified. The cases were evaluated for a population of morphologically distinct candidate progenitor cells along the subammonic seam. In a subset of the cases immunostaining was used to confirm the identity of the cells. The position of this cell population along the seam, staining pattern of the cells, and persistence over development were assessed.

Results: The cells express a combination of INSM1, NeuN, and calretinin throughout their lifespan, confirming their identity as neural progenitors. The cell population decreases in size over development. Additionally, the distance of the cells from the ventricle increases as the seam closes, the cells “migrate” up to 17 mm from the lateral ventricle in this process.

Conclusions: The neural progenitor cells persist along the seam for up to 16 weeks postnatally, migrating up to 17 mm from the lateral ventricle. The population stains for neural progenitor markers. These clusters are not visibly associated with vessels, indicating a distinct method of spread from the germinal matrix. The cells could contribute to axon pathfinding between entorhinal cortex and hippocampus, but further studies are required to determine their fate.

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Gennari's Line: Developmental Implications for Visual Cortex Organization and Function
B Akomeah; Jachie-Pramso Senior High School

Background: Gennari's Line, a distinctive myelinated band in primary visual cortex (V1), plays a critical role in visual processing. Its development and organization are key to understanding visual cortex maturation and potential vulnerabilities in neurodevelopmental disorders.

Methods: I reviewed histological and imaging data on Gennari's Line in human brains (ages 20-60) and compared with non-human primates. Immunohistochemistry (myelin, neuronal markers) and MRI tractography were used to assess structure and connectivity. Functional implications were explored via visual task performance correlations

Results: Gennari's Line showed age-related increases in myelination and was more pronounced in humans vs. non-human primates. Disruptions in Gennari's Line structure correlated with visual processing deficits in developmental disorders (e.g., amblyopia). Connectivity patterns suggested a role in integrating magnocellular and parvocellular visual pathways.

Conclusions: Gennari's Line's structure and development are crucial for understanding visual cortex organization. Alterations may contribute to visual processing disorders, highlighting its potential as a biomarker for neurodevelopmental studies. Integrated approaches combining structure, function, and connectivity are needed to elucidate its role in visual processing .

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Unusual cerebellar architecture in a 22 month old with Menkes disease

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Background: This 22-month-old male child was found prone and unresponsive in his crib. Gestational history included term birth, average birth weight and height. Following birth, he began to experience recurrent seizures and was evaluated at Seattle Children's Hospital where workup was consistent with Menkes disease, a rare congenital disorder of copper malabsorption. Intravenous copper supplementation was subsequently initiated, and despite appropriate therapy, his epilepsy progressively worsened, exhibiting breakthrough seizures while on multi-agent therapy.

Methods: The scene investigation and autopsy were conducted by the King County Medical Examiner's Office.

Results: Autopsy revealed multiple diagnostic features of Menkes disease including height and head circumference < 3rd percentile for age, low body weight (5-10th percentile), skin laxity, generalized bone under-mineralization with abnormal costochondral ossification, "kinked" hair-shafts, and microcephaly, among others. Multiple remote and subacute rib fractures were noted, without evidence of acute injury and without clinical concerns for traumatic etiologies, typical of Menkes disease and a mimic for abuse-pattern injury. Neuropathologic examination revealed multiple features of Menkes disease including generalized cortical atrophy with overall foreshortening and small brain size for age (678 g, < 5th percentile for 1 year). Focal chronic subdural hemorrhages were identified, another common Menkes disease finding that mimics abuse-pattern injuries. Interestingly, the cerebellum was markedly small (59 g, predicted weight 76.8 g), and cut sections revealed shrunken, atrophic, and firm folia. Microscopy revealed dysgenesis in the form of an attenuated external granule cell layer, usually as a thin ribbon throughout the superficial part of the molecular layer, but occasionally at the pial surface, more consistent with the cellularity typically seen at 1 month of age.

Conclusions: Descriptions of cerebellar architecture in Menkes disease are uncommon in recent literature and this case can both further our understanding of this rare disorder and encourage awareness to avoid misdiagnosis and medicolegal implications of suspected trauma.

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A Bloody Diagnostic Trap: Intracranial Extramedullary Hematopoiesis Mimicking a Brain Mass in a Neonate

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Background: A male patient was born to a G1P0 mother at 37 weeks and 5 days of a pregnancy complicated by maternal SARS-CoV-2 infection. Physical exam revealed diffuse petechiae and abnormal posturing and eye movements, concerning for seizure. Initial studies identified severe thrombocytopenia, with a platelet count of 11 microliters. An ultrasound revealed an intracranial hemorrhage (ICH); subsequent MRI demonstrated a 6.8 cm right extra-axial hemorrhage, suggestive of an epidural versus a subdural hematoma with mixed chronicity of blood products, midline shift, intraventricular hemorrhage, and trapped lateral ventricle. He underwent craniotomy for hematoma evacuation.

Methods: Friable tissue was noted within the hematoma; an intraoperative consultation showed a focal population of mitotically active, atypical cells amid abundant hemorrhage. Final pathology revealed a hematoma with acute, subacute, and chronic features with prominent extramedullary hematopoiesis (EMH) involving reactive brain parenchyma. Trilineage bone marrow components were identified: erythrocyte precursors, myeloid precursors, megakaryocytes, and macrophages were positive for CD71, MPO/CD117, CD61, and CD163, respectively.

Results: EMH is most often described in association with viral or congenital infections, neonatal anemias, hemolytic disease, neoplastic, autoimmune, and other hematologic disorders; however, its pathogenesis within the central nervous system remains poorly understood. In this case, the ICH was attributed clinically to neonatal alloimmune thrombocytopenia (NAIT), supported by parental platelet genotyping demonstrating HPA-1 incompatibility. While NAIT is rare overall, it frequently presents with ICH with the majority of cases occurring in utero, as in this case. Notably, cutaneous EMH has recently been reported in a neonate born to a mother with SARS-CoV-2 infection.

Conclusions: This case expands the spectrum of neonatal EMH by demonstrating a rare intracranial, mass-forming presentation within a chronic hematoma secondary to NAIT. Importantly, it emphasizes the consideration of EMH in differential diagnoses in the clinical context of ICH, critical to avoiding a misdiagnosis as malignancy during intraoperative consultations.

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Hemizygous ALG13 Variant in a Term Neonate with Grade IV Germinal Matrix Hemorrhage

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Background: Germinal matrix hemorrhage (GMH) remains a rare cause of intracerebral/intraventricular hemorrhage in full-term neonates. Although causes of GMH are debated, vascular fragility and hemodynamic lability are cited as primary contributors - conditions more frequently observed in preterm infants. Respiratory compromise, hypoxic insults, and coagulation abnormalities are commonly identified as etiologies in term infants. Understanding and addressing these causes is important since significant complications can arise, including posthemorrhagic hydrocephalus, epilepsy, and severe cognitive impairment.

Methods: We report a 39-week-old full-term male born via Cesarean section due to macrocephaly and failure to progress. Delivery was complicated by nuchal cord and meconium aspiration. Brain MRI/MRA/MRV imaging demonstrated grade IV GMH involving the left lateral ventricle body with ventricular system dilation. No vascular abnormalities or tumors/masses were identified. The hospital course was complicated by seizures and bronchopulmonary dysplasia. A ventriculoperitoneal shunt was placed after no improvement with a left frontal reservoir. He was discharged after 2 months, but expired at home one month later.

Results: The etiology of the GMH remained unclear. Infectious and hematologic workups were negative. Genetic testing yielded sickle cell trait and a hemizygous variant of uncertain significance in ALG13. Postmortem examination revealed significant ventriculomegaly in the brain with cortical thinning/compression. Microscopy showed remote periventricular and subarachnoid hemorrhages, and evidence of periventricular leukomalacia. There was loss of ependymal cells with proliferation of subependymal astrocytes and frequent ependymal rosettes/tubules.

Conclusions: Genetic risk factors for GMH have been explored involving inherited coagulation abnormalities, but ALG13 mutations have not been identified previously in these cases. Neurologic involvement predominates the phenotype of ALG13-associated disorders, with seizures being common. Cases of increased bleeding tendency have also been documented with ALG13 variants. The hematologic issues combined with the predominant neurologic effects of these ALG13 variants may increase risk for GMH, even in term infants, but this remains to be explored.

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Long-Term Survival in Holoprosencephaly: A Rare Case Extending Into Late Adolescence

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Background: Holoprosencephaly (HPE) is a rare congenital brain malformation characterized by incomplete medial cleavage of the prosencephalon into right and left cerebral hemispheres during early embryogenesis. It is etiologically heterogeneous, with an estimated prevalence of 1 in 15,000 –16,000 live births. HPE is associated with high postnatal mortality, with approximately one-third of affected neonates dying within the first 24 hours and an overall mortality of nearly 58% within the first month of life.

Methods: A 17-year-old female with a history of alobar holoprosencephaly and central diabetes insipidus presented after out-of-hospital cardiac arrest following a month-long productive cough, achieving return of spontaneous circulation after 26 minutes of CPR. Brain MRI revealed a large midline cerebrospinal fluid (CSF) filled mono-ventricle with absent falx cerebri and corpus callosum, fused thalami, and mass effect on the brainstem and cerebellum with posterior cerebellar atrophy. A right parietal ventriculoperitoneal shunt terminated within the mono-ventricle. Hyperintense FLAIR signal with restricted diffusion in the anterior cerebral cortex indicated acute infarction, without hemorrhage or mass.

Results: Autopsy of the brain (510 g) demonstrated alobar holoprosencephaly with a single holo-ventricle, absence of midline structures (falx cerebri, corpus callosum, septum pellucidum), absent olfactory nerves, abnormal cortical gyration, and fused basal ganglia and thalami. The brainstem and cerebellum were compressed but appear normally developed. Microscopic findings revealed global hypoxic-ischemic injury, diffuse astrogliosis, and scattered vascular mineralization perivascular lymphocytic aggregation. Bilateral cystic ovaries were also noted. Findings are consistent with severe congenital malformation due to alobar holoprosencephaly with superimposed hypoxic-ischemic injury contributing to the terminal event.

Conclusions: This case demonstrates rare survival into late childhood in alobar HPE and suggests that chronic brain changes, compounded by superimposed hypoxic-ischemic injury, contributed to the terminal event, underscoring the importance of long-term care and monitoring.

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Dystrophic calcification and cortical disorganization in epilepsy resections for Sturge-Weber syndrome: Genetic correlation

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Background: Sturge-Weber syndrome (SWS) is a congenital neurocutaneous disorder characterized by cortical disorganization of the brain, venous abnormalities of the leptomeninges, and dystrophic calcifications. Patients with SWS frequently develop epilepsy, which may necessitate surgical treatment. The underlying cause of SWS is a somatic activating mutation of the GPCR α subunit, GNAQ. Most SWS cases are caused by a c.548G->A (R183Q) mutation. However, mutations in GNA11 and GNB2 can also cause SWS. Given varying genetic causes of SWS, there is a need to investigate the relationship between genetic drivers, clinical outcomes and histopathological findings. Here, we report on a cohort of patients with SWS who were negative for the GNAQ R183Q mutation.

Methods: Six patients who underwent surgical resection of the neocortex were identified. Histopathological analysis was performed on all samples per routine H&E staining with immunohistochemistry for NeuN and GFAP performed on a subset of cases. Clinical data was obtained from the electronic medical record. Genetic testing for the GNAQ R183Q mutation was performed via Sanger DNA sequencing.

Results: Included patients tested negative for the R183Q mutation. The cohort was 66% male (4/6), with a median presenting age of 9 months (IQR 3 – 20 mo). Surgery was performed at a median age of 36 months (IQR 24 mo – 6 yr). All patients were seizure free following hemispherectomy, while two required ventriculoperitoneal shunting for postoperative hydrocephalus. All resection specimens demonstrated classic histopathologic findings of SWS including leptomeningeal angiomatosis, cortical calcospherites, parenchymal calcinosis, and neuronal disorganization. One case demonstrated abnormally few foci of parenchymal calcinosis.

Conclusions: In a cohort of patients with SWS who tested negative for the R183Q mutation, findings were largely in line with classic SWS. Hemispherectomy is an effective treatment strategy for seizure control in patients negative for the R183Q mutation. Genetic testing for GNB2 and GNA11 may provide additional value.

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Epilepsy-Associated Leptomeningeal Vascular Malformation Harboring an FGFR1 Mutation: A Case Report

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Background: Abnormalities of brain development represent a significant cause of drug-resistant epilepsy, particularly in the pediatric population. These anomalies may result from migrational and maturational defects of neurons. We describe an epileptogenic lesion characterized by leptomeningeal vascular anomaly, cortical calcification, and an FGFR1 mutation.

Methods: Evaluation included histologic examination, immunohistochemistry, and molecular testing by next-generation sequencing. Nucleic acids were extracted and quantified, followed by hybridization-based target enrichment, PCR amplification, and sequencing of exons and canonical splice sites only.

Results: The patient is a 12-year-old male with history of anxiety, learning disability, and longstanding, medically refractory epilepsy with increasing frequency over one year. An MRI showed right parieto-occipital cortical volume loss with presumed calcifications, overlying leptomeningeal enhancement, and a small non-enhancing T1-isodense nodule along the anterior third ventricle. Histology demonstrated superficial cortex with a band-like pattern of laminar calcifications extending into neuronal layers. The overlying leptomeninges showed numerous ectatic vessels with thick walls and internal elastic laminae. Neoplastic population was not identified by immunohistological stains for CD34, OLIG2, BRAF V600E, and GFAP. NeuN showed a reduced neuronal density, though cortical lamination could not be definitively assessed. Next-generation sequencing demonstrated an FGFR1 Asn577Lys*** mutation with a variant allele frequency of 7.5%.

Conclusions: Vascular anomalies are established causes of childhood epilepsy, including FCD IIIc adjacent to arteriovenous malformations (AVMs), and Sturge-Weber syndrome, which can produce vascular steal and band-like cortical calcifications. However, Sturge-Weber typically shows thin-walled leptomeningeal vessels and GNAQ mutations. These contrast the thick vessels and FGFR1 alteration seen in our case, which has not been described in a cerebral vascular malformation to our knowledge. FGFR1 mutations are reported in glial/glioneuronal tumors and encephalocutaneous lipomatosis, neither of which were present. AVMs commonly harbor KRAS mutations activating the MAPK pathway. While classical features such as tapering internal elastic lamina were absent, a similar mosaic mechanism remains possible.

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Classifying Previously Uncategorized Tumors: cases of MVNT and PLNTY diagnosed using the current WHO CNS tumor classification

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Background: The 5th edition of World Health Organization (WHO) classification of Central Nervous System (CNS) tumors introduced several new classes of gliomas and glioneuronal tumors based on newer molecular findings.

Methods: We present two exemplary cases of low-grade tumors - Pleomorphic Low-grade Neuroepithelial Tumor of the Young (PLNTY) and Multinodular Vacuolating Neuronal Tumor (MVNT). We describe our experience in incorporating the clinical presentation, imaging, and histology of these new diagnostic entities.

Results: These cases were previously difficult to classify and were either misclassified or “fitted” into other established classes. However, using the most recent CNS classification of tumors and molecular studies, we could accurately classify these tumors.

Conclusions: These cases underscore the value of the current WHO CNS tumor classification in providing diagnostic certainty and prognostic information to guide treatment in these previously difficult to classify tumors.

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A single center retrospective on the molecular stratification of meningiomas

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Background: Meningioma grading is critical and an increasingly complex component of neuropathology practice. The 2021 WHO and 2025 cIMPACT-NOW update 8 provide the most current guidelines for determining the prognostic significance of histologic and molecular findings. We examined meningiomas diagnosed at our institution to evaluate the utility of various histologic criteria against the final grades incorporating molecular criteria.

Methods: Meningioma cases from January 2023 to February 2026 were compiled. Cases sent for further molecular testing (chromosomal microarray, next-generation sequencing) were identified. Microscopic features were used to establish histologic grades. Cases were then assigned current molecular-based grades including stratification into histologic grade 1 (G1) with higher risk due to relevant chromosomal losses or histologic grade 2 (G2) with low molecular risk features due to the lack of higher grade alterations.

Results: 294 meningiomas were identified from which 80 cases (G1: 50; G2: 26; G3: 4) were sent for molecular characterization. Of the histologic G1 cases, 16 (32%) were upgraded to G2, 1 (2%) was stratified as higher risk G1, and 1 (2%) was upgraded to G3. Of the histologic G2 cases, 15 (57.7%) were stratified as lower risk G2 and 2 (7.7%) were upgraded to G3. All 4 histologic G3 cases had G3 alterations. Remaining cases had molecular alterations concordant with their respective histologic grades.

Conclusions: The most notable finding was that 34% of histologic G1 cases were found to have higher grade molecular features suggesting a sizable proportion of unsent cases may have poorer prognoses than expected, possibly warranting a lower threshold for molecular characterization. A high rate of histologic G2 cases lacking relevant molecular findings was also observed. The significance will become clearer as more prognostic data for cases stratified by cIMPACT-NOW guidelines are available. Communication with the clinical team regarding interpretation of such results will be increasingly necessary to guide appropriate management.

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Integrated meningioma classification outperforms clinical prognostication but loses power with time and is affected by Simpson grade.

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Background: Molecular testing has been proposed as superior to histological grade for assessing meningioma recurrence risk. However, clinical recurrence risk assessment usually incorporates variables other than histological grade alone. We compared histological grade, multidisciplinary (MDM) stratification (additionally incorporating tumour site and Simpson grade), cIMPACT-NOW 8 criteria, and integrated molecular-morphologic meningioma score (IMMS) in a continuous 3-year cohort of intracranial grade 1 and grade 2 meningiomas to determine which best predicted recurrence.

Methods: All intracranial meningiomas undergoing primary resection (1 January 2014 to 31 December 2016) with minimum 6-months radiological follow-up were identified. Relevant clinical data, including MDM risk stratification and recurrence, were recorded. All tumours were reclassified according to WHO CNS5 histological grade. Methylation profiling using the Illumina Infinium MethylationEPIC v2.0 Beadchip array and DKFZ classifier was performed. Methylation data and copy number plots were used to reclassify the meningiomas according to cIMPACT-NOW 8 and IMMS. The ability of the four methods to predict recurrence was compared.

Results: Included were 143 meningiomas (106 WHO grade 1; 37 WHO grade 2) with methylation profiling in 141. MDM assessment assigned 36 tumours as high-risk, 74 as low-risk and was unrecorded for the remainder. Stratification by IMMS gave a “low” risk result in 104 tumours, “intermediate” risk in 29 and “high” risk in 8. cIMPACT-NOW 8 criteria upgraded 18 histological grade 1 tumours to “grade 2”. IMMS (“low” versus “high” risk) significantly more accurately predicted recurrence than other methods up to 3 years post-surgery (accuracy at 3-years: IMMS 88%, MDM risk 68%, histological grade 73% (chi-square=14.9, p=0.0019)) but was not significant beyond this. Using IMMS, Simpson grade 1/2 resections had 7.48-fold higher odds of correct prediction than Simpson grade 3/4 at 3-year follow-up (p=0.0044)

Conclusions: IMMS is the optimal method of risk stratification for early meningioma recurrence but is most effective in Simpson grade 1/2 resections.

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Cellular crosstalk in the brain microenvironment promotes melanoma invasion and growth

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Background: Melanoma exhibits dynamic transcriptional adaptations to microenvironmental cues. Phenotypic heterogeneity impacts treatment response, particularly in melanoma brain metastasis (MBM), where therapeutic resistance is common.

Methods: To interrogate MBM cellular ecosystem, we profiled 114,455 single-cell transcriptomes from publicly available dataset. Melanoma cells were dichotomized by WNT5A expression levels to proliferative and invasive subtypes. Intercellular communication networks were inferred using CellChat, with top predicted interactions validated in vivo and in vitro.

Results: Invasive melanoma (WNT5A-high) showed strongest interactions with endothelial cells through laminin-integrin signaling. Proliferative melanoma (WNT5A-low) interacted most strongly with microglia, which induced a shift toward an invasive tumor state through MCAM activation. To assess melanoma phenotype—microenvironment interactions, control and WNT5A-overexpressing cells were injected intracranially into C57BL/6J mice. WNT5A tumors demonstrated significantly faster growth and shorter survival. Histologic examination revealed significantly greater proportion of invasive infiltrates in WNT5A tumors, frequently tracking along native vasculature. In vitro, WNT5A-overexpressing cells showed increased adhesion to endothelial cells and perivascular matrix, and ex vivo brain slice co-cultures demonstrated greater perivascular localization. Immunofluorescent imaging of microglia revealed three morphologies: homeostatic microglia in non-tumor regions, enlarged intratumoral microglia, and elongated microglia within invasive infiltrates aligned with MCAM-positive tumor tracks. While immunohistochemistry confirmed higher overall WNT5A expression in WNT5A-overexpressing tumors, melanoma cells within invasive infiltrates of control tumors exhibited comparable WNT5A expression, suggesting local microenvironmental induction of an invasive phenotype. Single-cell RNA sequencing of tumor-associated microglia identified three subclusters, including one with elevated WNT5A expression. Ongoing spatial transcriptomic analyses in human MBM aim to validate the spatial organization of melanoma phenotypes and their interactions with microglia and other brain cell types.

Conclusions: Integrated analyses show that MBM exhibits dynamic bidirectional signaling across tumor cells, endothelium, and microglia, collectively driving invasion and proliferation. Targeting these communication networks may offer a promising therapeutic strategy.

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Stratification by Ki-67 labeling index increases specificity of p16 expression as a surrogate marker for CDKN2A inactivation in meningioma

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Background: Meningiomas are the most common primary brain tumors. While most are benign, a subset behaves more aggressively with frequent recurrence. An important oncogenic driver of poor prognosis is CDKN2A homozygous deletion, which is an independent criterion for grade 3 meningioma in the current WHO classification. Next-generation sequencing (NGS) is the standard for detecting CDKN2A alterations, but cost and accessibility often limit routine use. While immunohistochemistry (IHC) assessment for p16, the protein product of CDKN2A, shows strong concordance between p16 loss and CDKN2A inactivation in high-grade meningiomas, its utility in low-grade meningiomas remains unclear. Therefore, we aimed to identify meningioma cases that may benefit from molecular testing, and hypothesized that Ki-67 proliferation index could serve as a stratification tool to identify meningiomas for which minimal expression of p16 indicates underlying CDKN2A inactivation.

Methods: We retrospectively analyzed 148 surgically resected meningiomas with paired NGS and available tissue for p16 and Ki-67 IHC. p16 expression was scored as low or high by three blinded pathologists. Ki-67 proliferation indices were digitally quantified using open-access platforms. Associations between p16 expression, Ki-67 index, histologic grade, and CDKN2A status were assessed, and optimal Ki-67 cutoffs were determined by sensitivity and specificity analyses.

Results: CDKN2A homozygous deletion was identified in 14 cases (9.5%). All CDKN2A-inactivated tumors showed p16-low expression, while all p16-high tumors retained CDKN2A. Tumors with CDKN2A-inactivation demonstrated significantly higher Ki-67 indices than wild-type tumors (20.5% vs 8.5%, $p < 0.0001$). Amongst p16-low meningiomas, a Ki-67 threshold of 8–10% optimized the balance of sensitivity and specificity for detecting CDKN2A inactivation. For broader screening purposes, a more stringent cutoff of 5% maximized sensitivity and captured all cases with CDKN2A inactivation.

Conclusions: An integrated IHC approach combining p16 and Ki-67 offers a practical and cost-effective screening tools to identify potentially aggressive meningiomas, and prioritize tumors to most benefit from NGS in resource-constrained settings.

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Concurrent somatic NF2 and SMARCB1 biallelic alterations in a series of higher grade meningiomas

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Background: Chromosomal and sequence alterations have gained significance in the grading of meningiomas in recent years. Somatic NF2 gene alterations are seen in ~50% of meningiomas, particularly those of the cerebral convexities and spine. Somatic SMARCB1 gene alterations are occasionally seen in meningiomas and have been described in tumors with an elevated Ki-67 index. NF2 and SMARCB1 alterations have been described in schwannomatosis-related tumors and in meningiomas without a clear association with a germline alteration.

Methods: Here we describe a series of seven cases (receiving copy number and tumor mutational profiling) of histologic grade 2 (n=6) and 3 meningiomas showing NF2 and SMARCB1 alterations alongside losses of chromosome 22.

Results: The patients were females with a median age of 60 years (38-79 years) and median tumor size of 3.6 cm. They were usually in the midline (n=3) or tentorial-based (n=4). Histologically, they were hypercellular transitional meningiomas with a mitotic count of 4-7 per 2 mm2 and a Ki-67 index of 5-9%, except for the grade 3 case (25 mitoses per 2 mm2 and 30% Ki-67 index). Other atypical histologic features were present variably. The grade 2 cases had loss of chromosome 22 with few or no other chromosomal alterations; the grade 3 case had a complex copy number profile. The NF2 gene alterations were variable in the tumors, but the SMARCB1 gene consistently showed a p.R377H point mutation.. Complete surgical resection resulted in disease-free follow-up (1-21 months). Cases (n=2) with incomplete surgeries or radiotherapy had a progression-free survival of 3.5-7 months.

Conclusions: This series of tumors with combined NF2 and SMARCB1 biallelic alterations were all grade 2 or 3 by histologic criteria, adding to the evidence base that these molecular findings may be driver events in a subset of higher grade meningiomas and could be considered as a part of future grading criteria.

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Multifocal Intraosseous Meningiomas with Sacrospinal and Calvarial Involvement. Case Report and Literature Review

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Background: Primary intraosseous meningiomas (PIM) represent only a small fraction of meningiomas, with multiplicity being exquisitely rare. The vast majority of PIMs arise within the calvarium.

Methods: Herein we present a unique case of histologically-proven multifocal intraosseous meningiomas involving the sacrum as well as vertebral bone.

Results: The patient is a 65-year-old woman who presented with 3-month history of coccydynia. Her symptoms progressed despite anti-inflammatories and physical therapy, prompting imaging studies which revealed a lobulated expansile lytic, mildly contrast-enhancing lesion occupying the majority of the sacrum. Tissue from biopsy and subsequent partial distal sacrectomy with intralesional resection represented a meningioma, WHO grade 2. Methylation profiling matched to meningioma, intermediate A/B, and NF2 p.S287fs mutation was detected (variant allele frequency 90.8%). Shortly after surgery, she experienced a ground level fall prompting additional imaging studies. Spinal magnetic resonance imaging (MRI) detected multiple enhancing mass lesions involving the spinous processes of T1 and T3 vertebrae, T3, T9, T11, and T12 vertebral bodies; no spinal dural lesions were detected. Brain MRI revealed a large extra-axial dural-based contrast-enhancing right parietal lesion with involvement of overlying bone, and separate contrast-enhancing lesion within the left parietal bone. Tissue from biopsy of T11 lesion represented a meningioma., WHO grade 1. Other notable incidental findings include a biopsy-proven cavernous hemangioma of the liver, and possible left hippocampal cavernoma detected on MRI brain. To date, she is pursuing radiation therapy for residual sacral tumor, and surgical resection of her right parietal lesion.

Conclusions: The multiplicity of lesions in this patient raises the question of whether these all may represent individual primary tumors versus metastases from one of the larger meningiomas. In either scenario, this unique case demonstrates the diagnostic and treatment difficulties that may be encountered with multifocal intraosseous meningioma.

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Atypical Meningioma with Granular Cell Features: A Case Report and Review of the Literature

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Background: Meningiomas are the most common primary central nervous system tumors and encompass a range of well-defined histopathologic subtypes. Rarely, meningiomas can demonstrate prominent granular cell change, characterized by abundant eosinophilic, PAS-positive granular cytoplasm. This pattern is not recognized as a distinct WHO subtype and has been described in a limited number of cases with no previously documented cases fulfilling criteria for an atypical meningioma. Granular cell change may mimic granular cell tumors causing diagnostic challenges. We report a rare case of an atypical meningioma with prominent granular cell features.

Methods: N/A

Results: A 72-year-old male, presenting with temporal headaches and a 4cm middle cranial fossa dural based enhancing lesion, underwent craniotomy and excision of tumor. Histology revealed a meningothelial neoplasm with transitional architecture, increased mitotic activity, necrosis, prominent nucleoli, focal brain invasion, and elevated Ki-67 proliferation. There were numerous clusters of cells with abundant eosinophilic granular cytoplasm, highlighted by PAS, CD68, SSTR2, and S-100 stains. Molecular testing revealed an NF2 mutation and chromosome 1p deletion. Ultrastructural analysis revealed increased mitochondria within the granular cells. Overall, the findings were consistent with atypical meningioma, CNS WHO grade 2, with granular cell features.

Conclusions: Granular cell change in meningiomas represents an uncommon histologic pattern rather than a distinct diagnostic subtype in the current WHO classification. The presence of abundant eosinophilic granular cytoplasm within tumor cells is thought to reflect metabolic or degenerative alterations of meningothelial cells. Awareness of this pattern is important to avoid misinterpretation as a primary CNS or metastatic granular cell tumor, oncocytic lesion, or histiocytic proliferation. According to a review of the literature, this is only the third report of a granular cell meningioma, and the first report of granular cell change in an atypical meningioma.

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Meningioma of the Jugular Foramen Presenting as a Neck Mass

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Background: Primary jugular foramen meningiomas are rare, originate from the arachnoid cap cells of the jugular bulb, and can spread into the posterior cranial fossa or the retropharyngeal space. Here we report a case of primary jugular foramen meningioma with extracranial extension, presenting as a neck mass.

Methods: Medical records, radiologic and pathologic findings were reviewed.

Results: A 42-year-old woman presented with dysphonia, dysphagia, and a 3.8 cm T2 hypointense, mildly enhancing, and partially calcified neck mass in the left jugular foramen extending intracranially and into the posterior left carotid space, encasing the internal carotid artery and occluding the internal jugular vein. The clinical and radiological differential diagnoses included glomus jugulare paraganglioma, schwannoma, and meningioma. Microscopic examination of the resection specimen showed a meningioma with meningothelial features. Mitotic rate was 0-1/1 mm2. No histological high-grade features were identified. Immunohistochemistry was positive for EMA, S100, and PR, with pertinent negatives including chromogranin A, GATA, SOX10, and synaptophysin. There was invasion of the jugular vein. Lymph nodes were unremarkable without involvement by meningioma. The diagnosis was meningioma, CNS WHO Grade 1. Molecular testing detected NF2 and TP53 mutations, but no other alterations. Follow-up scan showed stable residual 1.1 cm nodule adjacent to left cochlear aqueduct. She has hoarseness, left vocal cord paresis, and first-bite syndrome, but is otherwise well after initial resection alone.

Conclusions: The main considerations for neck masses include lesions of salivary, thyroid, and parathyroid glands, lymph nodes, paraganglioma, and peripheral nerve sheath tumors. Lesions arising in the jugular foramen are particularly uncommon, and are mostly paragangliomas, then schwannomas, and rarely meningiomas. Invasion into soft tissues, bone, or jugular vein alone is insufficient to establish high-grade behavior; however, careful consideration is required in planning surgical approach regarding patient quality of life with vascular and cranial nerve endangerment.

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B-lymphocytes Colliding with Meningiomas

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Background: A collision tumor between meningioma and CNS lymphoma is a rare occurrence with poorly understood pathological mechanisms presenting unique challenges for pre-operative assessment and post-operative management.

Methods: We describe an incidentally discovered right frontal convexity mass of a 75-year-old female.

Results: MR imaging revealed a 4.5 cm enhancing mass along the right frontal convexity, increased in size from 3.3 cm three years prior. She underwent resection and histology revealed spindled to epithelioid cells growing in intersecting fascicles and whorls with scattered psammoma bodies. Areas with extensive necrosis were also present and contained islands of enlarged cells with scant cytoplasm, apoptotic bodies, and mitotic figures, consistent with atypical lymphocytes. Meningioma cells were immunoreactive for SSTR2, EMA, and progesterone receptor, with BAP1, p16, and H3K27me3 intact and a low Ki-67 labeling index. The atypical lymphocyte population within the necrotic areas was immunoreactive for CD20, CD10, PAX5, and BCL6 with a Ki-67 labeling index of 50%. These cells lacked expression of cyclin D1, MUM1, c-MYC, SOX10, SSTR2, CAM5.2. EBV in-situ hybridization was negative. As a result of this DLBCL diagnosis, the patient received RCHOP chemotherapy and is doing well 1 year postoperatively.

Conclusions: We report a collision tumor containing components of DLBCL, germinal center subtype, and a meningioma, WHO grade 1. Collision tumors are exceedingly rare with proposed mechanisms involving changes in the tumor microenvironment facilitating or predisposing to the development of CNS lymphoma. This case highlights the need for additional study of the pathological mechanisms involved in the generation of these collision tumors.

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Metastatic Meningiomas

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Background: Herein we review extracranial metastatic meningiomas received at our institution over a 5-year period. Retrospective review of the Pathology Information Retrieval Optimizer (PIRO) with the key words “metastatic meningioma” identified three cases within the last 5 years: Two metastatic to the lung and one to the liver. Two of the cases metastasized from intracranial atypical meningiomas within 3 years of primary diagnosis. The third metastasized from a grade 1 meningioma 18 years after primary diagnosis.

Methods: N/A

Results: Clinical history, imaging, histomorphology, immunohistochemistry and molecular data were reviewed. Ki-67 index was above 20% when reported, rising up to 80% in one case with intracranial recurrence and metastasis to the liver. In all 3 metastases SSTR2A was positive; PR was positive in 2 of 2 cases; EMA was positive in 2 of 3. The reported molecular features of the primary meningiomas and metastases were maintained throughout the paired samples. Methylation profiling was available for 2 cases and matched to Meningioma, intermediate class, for both.

Conclusions: In a recent systemic review of the literature, 155 metastatic meningiomas are reported, with a prevalence of 0.18% of meningiomas having metastases [1,2]. The combination of the methylation profiling and molecular testing in our cases recapitulated similar findings. Particularly with our second case, the original diagnosis of grade 2 was based on 3 out of 5 high grade features, then confirmed on methylation with the loss of 1p and 12q, which was recently outlined in cIMPACT-NOW [3]. Further confirmation was shown with the metastatic lesion having additional whole arm losses of 6, 10, 14, and 18. With the rise of methylation profiling, analysis of the original and metastasis will be of interest in establishing higher risk features to guide surveillance.

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Malignant Meningioma of Paraspinal Soft Tissue

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Background: Meningioma is a common, typically benign, tumor that affects the dura mater of the brain and less commonly the spinal cord. Meningiomas rarely become malignant or metastasize. This report describes a case of malignant meningioma in the right erector spinae muscle. The patient was a 12-year-old male with a history of WHO grade 4 medulloblastoma, SHH-activated and TP53-wildtype diagnosed at age 3, with subsequent craniospinal radiation. During a routine follow-up visit, MRI demonstrated an enlarging right erector spinae intramuscular soft tissue lesion with possible peripheral enhancement concerning for metastatic medulloblastoma.

Methods: Surgical resection of the lesion was performed, and sections of the lesion were submitted for histological and immunohistochemical evaluation.

Results: Histological analysis showed a mitotically active, nodular arrangement of tumor cells with enlarged, atypical nuclei, speckled chromatin, and prominent nucleoli. Necrosis was identified. Immunohistochemistry showed P53 positivity in scattered tumor nuclei, Ki-67 positivity with a 10% increased proliferative index, SSTR2A positivity, EMA positivity, and PR positivity in scattered cells. Next generation sequencing (NGS) showed a heterozygous SMARCA4 pathogenic variant. Chromosomal microarray demonstrated loss of chromosomes 1p, 9p, 10q, 14q, and 22, and a gain of multiple regions of chromosome 1q.

Conclusions: Morphology, immunohistochemistry, NGS, and chromosomal microarray were consistent with malignant meningioma. Meningiomas outside of the dura mater are rare, especially those of the soft tissue. Meningiomas are a known sequela to craniospinal radiation in children with medulloblastoma; however, these are commonly low grade and of the central nervous system. This case is an aggressive, malignant meningioma of the paraspinal muscles, making it an anomaly in location and grade. Surgical resection of malignant meningioma significantly improves outcomes but is less likely to be definitively curative than benign types.

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Dural-based myxoid neoplasm harboring a rare SMARCA2::CREM Fusion

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Background: The 2021 WHO Classification of CNS Tumors introduced Intracranial Mesenchymal Tumors (IMT), FET::CREB fusion positive, as a provisional entity. Case studies have described IMTs with less common fusion partners that do not always correspond to the FET or CREB family, including rare cases with SMARCA2::CREM fusions. Herin we present a case of a dural-based myxoid neoplasm most consistent with IMT harboring a SMARCA2::CREM fusion.

Methods: We conducted histologic, immunohistochemical, and molecular analysis for a 51-year-old female with a history of arteriovenous malformation resection 9 years prior to her presentation with a new, left frontal dural-based tumor. Symptoms included new onset seizures, and imaging was suspicious for a left frontal meningioma.

Results: Microscopic examination revealed a dural-based neoplasm composed of spindled, epithelioid, and occasionally rhabdoid cells within a variably myxoid background. There were up to 7 mitoses per 10 high-power fields. The tumor was positive for desmin (patchy), SSTR2 (patchy), PR (focal), and showed areas of strong membranous CD99 staining. EMA was negative. Chromosomal microarray analysis showed chromosomal 3p deletion; however, no entity specific copy number changes were identified. DNA methylation profiling showed no match. To better classify this tumor, an RNA exome fusion panel was conducted and revealed a SMARCA2::CREM fusion.

Conclusions: Only rare IMTs harboring SMARCA2::CREM fusions have previously been described in the literature. While the patient described above has shown no evidence of recurrence for 7 months following a gross total resection, previously described cases harboring a SMARCA2 fusion partner had aggressive, early local recurrent following gross total resection. Thus, evaluation of prognostic outcomes of different fusion partners in IMTs warrants further investigation.

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A neuroepithelial tumor with PATZ:MN1 fusion a new tumor type and literature review

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Background: Neuroepithelial tumors (NET) represent a heterogeneous group CNS neoplasms originating from neuroepithelial cells. The recently described PATZ1::MN1 fusion–positive tumour has been discussed but not included in the 2021 WHO CNS classification owing to insufficient published evidence and clinicopathological heterogeneity. Although a few studies have been published, these tumours remain poorly defined and require further molecular and clinical characterization. We report here an additional case contributing to the expanding spectrum of PATZ1::MN1 fusion–positive neuroepithelial tumours.

Methods: We report the case of a 13-year-old patient who presented in 2020 with symptoms of intracranial hypertension. Brain MRI revealed a 7 cm solid-cystic mass in the left temporal lobe, showing contrast enhancement and focal lysis of the temporal bone.

Results: A subtotal resection (80%) was performed. Initial histopathological analysis revealed a round cell tumor expressing GFAP and S100, with heterogeneous expression of CD99.The diagnosis was challenging, based on morphology, a small round cell sarcoma was suspected; however, immunohistochemistry findings pointed toward a neuroepithelial origin. The patient was initially treated with radiotherapy after surgery. Upon recurrence in 2023, the tumor had extended to the petrous portion of the temporal bone, causing otorrhea, hearing loss, and facial paralysis. Histological examination revealed a spindle cell neoplasm exhibiting alternating cellular and fibrotic regions, with a Ki-67 proliferation index of 20%. Immunohistochemistry showed heterogeneous expression of SOX10 and negativity for S100 and other markers. GATA2 immunohistochemistry retrospectively performed on initial tumor was positive. RNA sequencing performed at the Institute of IRCAN , Nice, France identified a PATZ1::MN1 gene fusion.

Conclusions: This case highlights the diagnostic complexity of NET with PATZ1::MN1 fusion, spindle cell morphology may result in a sarcoma-like appearance, mimicking CNS sarcoma. Molecular profiling is crucial for accurate classification. These tumors share a methylation signature but diverge histologically and transcriptionally. Further studies are needed to clarify prognosis and guide treatment.

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GLI1 Fusion–Positive Mesenchymal Tumor Presenting as a Spinal Canal Mass: An Unusual CNS Manifestation of an Emerging Entity

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Background: GLI1-altered mesenchymal tumors represent an emerging group of neoplasms characterized by GLI1 fusions and activation of the Hedgehog signaling pathway. These tumors most commonly arise in the soft tissues of the trunk, extremities, and head and neck. Primary involvement of the central nervous system is exceedingly rare, with only isolated intracranial dural-based cases reported. We describe a diagnostically challenging tumor that to our knowledge is the first reported case arising in the spinal cord.

Methods: A 51-year-old male presented with back pain. MRI without contrast showed a spinal canal mass extending from L2 to S2. A CT-guided biopsy was performed. The diagnostic workup included NGS (UTSW 1500 gene DNA/RNA panel).

Results: Histology demonstrated a highly cellular neoplasm composed of small round to epithelioid cells with a high nucleus-to-cytoplasm ratio but no mitoses. A lobulated growth pattern was present focally. Immunohistochemistry showed diffuse positivity for S100, CD56, and vimentin. Despite the positivity for S100, SOX10 was negative. The MIB-1 index was 14%. Immunostains were negative for desmin, myogenin, MyoD1, GFAP, calponin, p63, ERG, CD31, CD34, chromogranin, INSM1, HMB45, and MelanA. NGS identified a PTCH::GLI1 fusion. The tumor was deemed unresectable; the patient received radiotherapy and shows no evidence of progression.

Conclusions: This case expands the anatomic spectrum of GLI1-altered mesenchymal tumors by documenting a spinal canal presentation. Malignant behavior including distant metastases has been described in a significant proportion of cases. Targeted therapy with the TKI inhibitor pazopanib has been proposed in the literature and is being considered for our patient. Molecular testing provides a definitive diagnosis, although the diagnosis can be suspected based on lobulated growth pattern and the unusual S100+/SOX10- immunohistochemical profile.

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A unique type of intracranial dural-based sarcomatoid neoplasm affecting pediatric patients harbors a distinct DNA methylation profile

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Background: Intracranial dural-based tumors are uncommon in the pediatric population and can be difficult to classify by current CNS WHO criteria.

Methods: We used unsupervised clustering of DNA methylation profiles from approximately 56,000 CNS samples to identify a group of 13 tumors from 10 patients that were epigenetically similar to each other but distinct from established tumor classes.

Results: Patient median age was 6 years (range 3-9 years) with an equal number of females and males. Tumor location was supratentorial and involved the dura in all seven patients with available records. Imaging characteristics were consistent with a subdural hematoma or fluid collection, and four of the patients had a history of prior trauma or surgical instrumentation to the affected area. DNA methylation-based classification (Bethesda v3.1) predicted the ‘mesenchymal atypical’ subtype of glioblastoma for all tumors. Tumors did not match a specific class on the sarcoma classifier (Heidelberg v12.6). Genome-wide aneuploidy was observed in all tumors. Recurrent copy number alterations included loss of chromosome (chr) 6 (7 of 10, with one additional tumor demonstrating loss of chr6q alone), gain of chr8 (7 of 10), and gain of a segment of chr11q harboring the CCND1 locus (8 of 10, 3 of which showed high-level amplifications). In tumors with sequencing data, tumor mutational burden was low. One case harbored an HRAS (G12S) mutation, another harbored a SMARCB1 mutation, and another showed high-level amplification of PDGFRA accompanied by multiple intergenic rearrangements suggestive of extra-chromosomal DNA. Seven tumors available for histologic review demonstrated variably epithelioid to spindled cells with eosinophilic cytoplasm and enlarged, pleomorphic nuclei embedded in an extensively myxoid stroma, often with abundant mitotic activity, and variable necrosis.

Conclusions: Herein, we identify a rare but epigenetically unique intracranial sarcomatoid neoplasm affecting pediatric patients. Additional clinical followup is needed to better assess tumor biology.

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Calcified Chondroid Mesenchymal Neoplasm/Chondroid Synoviocytic Neoplasm: An Emerging Entity With Predilection for Skull Base Involvement

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Background: Calcified Chondroid Mesenchymal Neoplasm (CCMN)/Chondroid synoviocytic neoplasm is a relatively recently described entity, which has a predilection for the temporomandibular joint (TMJ) region. The histologic features overlap with other chondroid neoplasms, making it relevant in the differential diagnosis of chondroid lesions that can involve the base of skull and intracranial space.

Methods: N/A

Results: A woman in her 60's presented to the emergency department with a 3-week history of headaches, nausea, and vomiting. Brain imaging showed an expansile, heterogeneous lesion within the temporal bone, with erosion through the bone adjacent to the temporal lobe. The lesion was subsequently resected, with surgical impression of a chondrosarcoma. Histologically, the tumor demonstrated variable morphologies, including areas of moderately cellular spindled cells arranged in storiform pattern, areas of more epithelioid-appearing cells with prominent multi-nucleated giant cells, transitioning into chondromyxoid stroma, and notable grungy calcifications. The nuclei were cytologically bland and mitotic activity was minimal. Differential diagnosis included primary bone and cartilaginous neoplasms such as tenosynovial giant cell tumor, chondrosarcoma, and chondroblastoma, as well as phosphaturic mesenchymal tumor due to the grungy calcifications. A panel of immunohistochemical stains showed a non-specific pattern of staining. Molecular testing identified a FN1::FGFR2 gene fusion, which led to the final diagnosis of FN1::FGFR2 rearranged CCMN/chondroid synoviocytic neoplasm.

Conclusions: Our case illustrates the typical histomorphologic and molecular findings of a relatively recently described entity, CCMN/chondroid synoviocytic neoplasm. This tumor type appears to be locally aggressive at most, with no metastatic disease reported to date. The predilection for TMJ/skull base location with potential intracranial involvement makes it an important entity for neuropathologists to recognize.

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HOXD12 activation status across brain tumor types reflects developmental origins

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Background: Reactivation of developmental transcriptional programs is a hallmark of cancer. HOX genes, master regulators of embryonic patterning, are typically silenced after development but are aberrantly expressed in multiple malignancies. We previously identified HOXD12 activation in a clinically aggressive subset of IDH-mutant, 1p/19q-codeleted oligodendrogliomas. However, the broader relevance of HOXD12 across primary and metastatic brain tumors has not been systematically examined.

Methods: We evaluated HOXD12 protein expression by immunohistochemistry in 162 primary CNS tumors and 84 brain metastases, with quantitative whole-slide digital image analysis (QuPath). To define subgroup-specific activation patterns, we interrogated whole-genome DNA methylation (n=1338) and bulk RNA-sequencing datasets (n=876) across medulloblastoma molecular subgroups.

Results: HOXD12 expression was significantly enriched in primary CNS tumors compared with brain metastases ($p < 0.0001$). Among primary tumors, medulloblastomas exhibited the highest HOXD12 levels, exceeding those observed in glioblastoma, astrocytoma, oligodendroglioma, ependymoma, and meningioma (one-way ANOVA). In contrast, no significant differences were observed among diffuse glioma subtypes or among metastases stratified by site of origin. Integrative epigenomic analysis demonstrated elevated HOXD12 gene body methylation in medulloblastoma, with peak levels in the WNT-activated subgroup ($p < 0.0001$). Concordantly, RNA-sequencing confirmed maximal HOXD12 transcript expression in WNT-activated tumors relative to other subgroups ($p < 0.0001$).

Conclusions: HOXD12 activation is preferentially associated with primary CNS tumors and is most pronounced in medulloblastoma, particularly the WNT-activated subgroup. These findings implicate HOXD12 as a lineage-linked developmental regulator in medulloblastoma and highlight subgroup-specific reactivation of embryonic patterning programs. Our results position HOXD12 as a candidate biomarker of developmental identity and a potential mechanistic contributor to medulloblastoma biology.

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Methylation profiling of biphasic classic medulloblastomas shows distribution across G3/G4 subgroups with enrichment for subgroups 1, 7, and 8

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Background: Histologically defined medulloblastoma includes desmoplastic/nodular, medulloblastoma with extensive nodularity, and large cell/anaplastic, though more nuanced patterns exist. “Biphasic classic” medulloblastoma is defined by pale nodules of neural differentiation without reticulin deposition, whereas triphasic medulloblastoma additionally contain compact hyperchromatic cell clusters with intense synaptophysin staining and reduced Ki-67 labeling. These rare histologic variants represent patterns of classic medulloblastoma in the non-WNT/non-SHH molecular group, for which correlation with granular methylation subgroups have not been established.

Methods: We searched St. Jude Children’s Research Hospital archives from 2007-2025, finding 59 biphasic medulloblastoma cases (classic = 58, anaplastic = 1) and 11 triphasic cases with sufficient material, performing DNA methylation profiling on each. DNA methylation group and subgroup were established using supervised and unsupervised methods. Copy number profiles were evaluated across the cohort using the conumee algorithm and correlated with methylation class.

Results: Tumors failed to classify into a specific methylation class. Biphasic and triphasic medulloblastomas were predominantly group 4 (n=44, 75% and n= 7, 64%), with fewer group 3 cases (n=15, 25% and n=4, 36%). Biphasic medulloblastomas demonstrated enrichment for subgroup 1 (n=13, 22%), subgroup 7 (n=13, 22%), and subgroup 8 (n=15, 25%), with the remaining falling into other subgroups (n=14, 24%) or receiving subthreshold classification scores (n=4, 7%). Only a single case classified into the high-risk subgroup 2 or 3. Triphasic medulloblastomas similarly showed enrichment for subgroup 7. Copy number analysis found recurrent chromosome 7 gain and chromosome 8 loss. Amplification events, including those in MYC or MYCN, were rare.

Conclusions: This study correlates biphasic and triphasic medulloblastoma patterns with methylation groups and subgroups. Our findings suggest neuronal differentiation is not associated with a specific methylation subgroup but is enriched in low-risk groups such as subgroups 7 and 8.

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CNS Embryonal Tumor Harboring a ZNF532::NUTM1 Fusion in the Cerebellum of a Young Adult

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Background: Embryonal tumors of the CNS are molecularly heterogeneous neoplasms that increasingly require integrated genomic characterization for accurate diagnosis. While medulloblastoma represents the most common embryonal tumor of the cerebellum, other morphologically similar entities may evade established molecular classification and are designated as not elsewhere classified (NEC). Neoplasms with NUTM1 rearrangements, most commonly arising in non-CNS midline structures, represent a clinically aggressive tumor type often with primitive morphologic features. We describe an exceptionally rare CNS embryonal tumor harboring a ZNF532::NUTM1 fusion in the right cerebellum of a 20-year-old male patient.

Methods: We reviewed clinical information, histomorphology, immunohistochemistry, methylation profiling (NIH/Bethesda classifier v2.0), and Tempus DNA/RNA sequencing data.

Results: Histomorphology demonstrated a poorly differentiated embryonal neoplasm composed of dense sheets of round cells with high N:C ratio. Brisk mitotic activity, apoptotic bodies, and necrosis were present. By immunohistochemistry, the tumor cells were positive for synaptophysin and focally positive for OLIG2; had retained expression of INI1/BAF47; showed a wildtype expression pattern of p53; and was negative for GAB1, YAP1, and β-catenin. The Ki-67 proliferation index was ~60%. The tumor matched to Family "Neuroblastic_embryonal_tumors" (0.955) but did not match to a Class by methylation. NGS demonstrated a pathogenic ZNF532::NUTM1 fusion.

Conclusions: CNS embryonal tumors with NUTM1 fusions have rarely occurred in children age ≤3 years or young adults, all located in the cerebral hemispheres. The ZNF532 fusion partner has been reported once in a 1.5-year-old male. To our knowledge, this represents the first case of a CNS embryonal tumor with a ZNF532::NUTM1 fusion and infratentorial location in a young adult patient. While extremely rare, molecular assays that detect NUTM1 rearrangements should be considered in CNS neoplasms with embryonal morphology, particularly in cases that would otherwise be designated as NEC. With additional cases, the clinicopathologic characteristics of this tumor type can be further elucidated to inform therapeutic management.

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Diagnostic Challenges in a Congenital Ependymoma with a Medulloblastoma Methylation Profile

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Background: Integrated histomolecular classification is the current standard for central nervous system tumor diagnosis. However, rare pediatric cases—particularly in the neonatal period—can exhibit striking discordance between histology, driver fusions, and DNA methylation, challenging established diagnostic frameworks.

Methods: The patient was included in the Children's Brain Tumor Network (CBTN) cohort. This case involved a 7-week-old female infant (37 weeks' gestation) presenting with a prenatally detected posterior fossa mass and obstructive hydrocephalus. Following a palliative biopsy and subsequent intraoperative arrest, tumor tissue was analyzed. Diagnostic workup included immunohistochemistry, RNA sequencing, and DNA methylation profiling utilizing both NIH and Heidelberg classifiers.

Results: Initial histopathologic evaluation demonstrated a primitive, highly cellular neoplasm; YAP1 and GAB1 immunopositivity favored a medulloblastoma (SHH subtype) diagnosis. However, RNA sequencing identified a YAP1–MAML2 fusion, a hallmark of supratentorial ependymoma. Retrospective histologic review identified focal perivascular arrangements and dot-like EMA expression consistent with ependymal origin. Conversely, DNA methylation profiling yielded a high-confidence match for medulloblastoma (SHH subtype).

Conclusions: This case identifies a rare diagnostic conflict in which a site-atypical, ependymoma-associated fusion occurs within a tumor harboring a medulloblastoma methylation signature. These findings highlight the limitations of current integrated diagnostic approaches in pediatric neuro-oncology and underscore the need for continued refinement of CNS tumor classification to account for such molecularly divergent phenotypes.

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YAP1::MAML2 Fused ETMR of the Posterior Fossa in Early Infancy: Case Report

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Background: Embryonal tumor with multilayered rosettes (ETMR) is a rare tumor of early infancy. We report the second infantile posterior fossa embryonal tumor matching to atypical ETMR by methylation profiling, harboring a YAP1::MAML2 fusion.

Methods: Clinical presentation and neuroimaging were integrated with histology, immunohistochemistry, fluorescence in situ hybridization (FISH), next generation sequencing (NGS), and methylation profiling to reach a diagnosis.

Results: A four month old male presented with feeding difficulty, impaired ocular movements, and somnolence. Brain magnetic resonance imaging showed a predominantly solid posterior fossa mass causing obstructive hydrocephalus. Resection revealed a primitive embryonal tumor composed of tightly packed round blue cells with nuclear molding, brisk mitotic activity, perivascular formations, necrosis, microcalcifications, and focal ganglion cell differentiation. Multilayered rosettes were not observed. By immunohistochemistry, the tumor expressed synaptophysin diffusely, membranous β catenin, retained nuclear INI1 and BRG1, and focal GFAP. GAB1 was positive and YAP1 showed nuclear and cytoplasmic positivity. Olig2 and NKX2.2 were negative. LIN28A showed weak nonspecific staining. The Ki-67 index exceeded fifty percent. DNA methylation profiling classified the tumor as subclass “ETMR, atypical” with high confidence. C19MC testing was negative by FISH. Whole exome NGS identified no pathogenic mutations, including in DICER1. However, solid tumor fusion analysis identified a YAP1::MAML2 gene fusion. Management to date includes subtotal resection with shunting, induction chemotherapy, and preparation for stem cell transplant. Interval MRI showed slightly decreased residual tumor and no evidence of spinal metastases. The patient was discharged in stable condition, with a resolved episode of febrile neutropenia and grade 2 hearing loss on audiometry as treatment complications.

Conclusions: This infant ETMR with YAP1::MAML2 fusion highlights integrating pathology modalities in classification and expands the observed molecular spectrum within ETMRs. Given a single case report of favorable prognosis relative to other ETMRs, additional data is needed to understand this molecular subtype of ETMR.

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Malignant Transformation of Recurrent Adamantinomatous Craniopharyngioma to Squamous Cell Carcinoma: Case Report

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Background: Malignant transformation of craniopharyngioma is a rare event which can occur in pediatric onset craniopharyngioma. We report a case in which histopathology and immunophenotype support squamous cell carcinoma arising from an adamantinomatous craniopharyngioma.

Methods: Clinical history from early childhood through adolescence, longitudinal neuroimaging, and prior and current surgical pathology were reviewed. Extensive sampling, immunohistochemistry, and comparison with earlier resections were used to assess growth pattern, cytologic atypia, proliferation, and squamous differentiation.

Results: The patient is a 16-year-old female with a history of adamantinomatous craniopharyngioma diagnosed at age 2. She initially underwent resection with early childhood proton radiation, subsequent placement of an intratumoral chemotherapy reservoir, and additional surgeries at ages 9, 12, and 15. She re presented at age 16 with escalating headaches, right sided proptosis, progressive visual decline, and multiple pituitary hormone deficiencies. Imaging showed a large solid and cystic skull base mass centered in the sellar/suprasellar region with cavernous sinus, orbital, and sinonasal extension. Debulking with orbital decompression was performed. Histology showed adamantinomatous craniopharyngioma with marked hypercellularity, prominent cytologic atypia, focal necrosis, bone and soft tissue invasion, and an elevated mitotic rate of up to 25 mitoses per 10 high power fields. Immunohistochemistry demonstrated strong nuclear and cytoplasmic β catenin expression, diffuse p40 and p63 positivity, p16 and p53 positivity in a subset of cells, and a Ki-67 proliferative index of 10-15%. The constellation of infiltrative growth, cytologic atypia, high proliferation, and diffuse squamous lineage markers support malignant transformation to squamous cell carcinoma from an adamantinomatous craniopharyngioma.

Conclusions: In this case, the synthesis of infiltrative growth, cytologic malignancy, and increased proliferation justify a malignant designation. Further outcome studies with histologic correlation are warranted for characterization of malignant transformation from craniopharyngioma.

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55-year-old female with a Multinodular and Vacuolating Neuronal Tumor (MVNT): A case report

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Background: Multinodular and vacuolating neuronal tumor (MVNT) is a CNS World Health Organization grade 1 neuronal neoplasm that typically involves the deep cortex, superficial white matter, and most often the temporal lobe. On MRI, there are typically clustered T2/FLAIR-hyperintense nodules with minimal mass effect or edema. The tumor is composed of nodules of monomorphic neuronal cells in a markedly vacuolated background. Tumor cells are often OLIG2-positive with weak synaptophysin and NeuN labeling, and CD34 can highlight ramified processes in adjacent cortex. MVNTs harbor activating alterations in the RAS/RAF/MAPK pathway, most commonly MAP2K1 indels/hotspot mutations, followed by non-V600E BRAF mutations and FGFR2 fusions.

Methods: A 55-year-old woman developed one year of right-sided numbness, headaches, episodic word-finding difficulty, and intermittent confusion. MRI revealed a T2/FLAIR-hyperintense lesion in the left anterior temporal white matter extending along the anteromedial temporal stem with multiple cystic foci and vasogenic edema. She underwent left temporal craniotomy; the collected tissue was submitted for histopathologic and immunohistochemical evaluation, next-generation sequencing, and DNA methylation profiling.

Results: Histology demonstrated a nodular proliferation of monomorphic neuronal cells within a prominently vacuolated background without mitotic activity, microvascular proliferation, or necrosis. Tumor cells were OLIG2-positive, weakly NeuN-positive, and GFAP-negative; CD34 highlighted ramified processes in adjacent cortex. Ki-67 proliferation index was < 1%. Next-generation sequencing identified an in-frame MAP2K1 deletion (p.Q56_V60del). DNA methylation profiling did not yield a classifier match but was indicative of a low-grade glioneuronal tumor. Overall findings were consistent with MVNT, CNS WHO grade 1.

Conclusions: The absence of a definitive methylation classifier match in this case underscores the opportunity for further epigenetic characterization of MVNT, as systematic DNA methylation profiling may enhance tumor classification and clarify the biological spectrum of this entity.

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Plot Twist: Intraparenchymal Schwannoma with CHD7::VGLL3 Fusion: A Case Report
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Background: Intraparenchymal schwannomas represent an exceedingly rare subset of CNS tumors, accounting for < 1% of intracranial schwannomas. Recently, VGLL-altered intraparenchymal CNS schwannomas have been identified as a distinct molecular entity characterized by specific gene fusions, most commonly CHD7::VGLL3.

Methods: N/A

Results: We report a 29-year-old man who initially presented to the ED on 6/27 with a seizure and underwent a nondiagnostic brain biopsy of a left parietal non-contrast-enhancing lesion. Repeat MRI on 8/12 revealed a solidly-enhancing mass within the left lingual gyrus with surrounding FLAIR hyperintensity, concerning for primary glial neoplasm. On 8/29 the patient underwent left frontal craniotomy and open biopsy. Histopathologically, the specimen was hypercellular with mild nuclear atypia and focal areas of cystic change. There were focal spindled cells resembling schwannoma but no Antoni A/B foci or Verocay bodies. Mitotic figures were rare. No endothelial proliferation or necrosis were seen. Immunohistochemical analysis revealed positivity for GFAP and negativity for mutated IDH-1 (R132H), BRAF (V600E), H3 (K27M), and SSTR-2. Staining for Ki-67 was positive in < 5% of tumor cells. The original favored diagnosis was a low-grade glioma based on its intraparenchymal location and GFAP-positivity. Molecular analysis by RNA next-generation sequencing identified a CHD7::VGLL3 gene fusion. A proprietary AI analysis indicated a 99% probability that the tumor was a schwannoma. VGLL fusions define a new class of intraparenchymal CNS schwannoma and can be GFAP-positive.

Conclusions: This case contributes to growing literature on VGLL-altered intraparenchymal CNS schwannomas, emphasizing the importance of molecular characterization in rare CNS tumors. CHD7::VGLL3 fusion in intraparenchymal schwannomas has diagnostic and prognostic implications, as these tumors demonstrate excellent outcomes following complete surgical resection with minimal risk of recurrence. Molecular testing should be considered for intraparenchymal tumors with schwannoma-like histology to identify this distinct entity.

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Evaluating MTAP as a Surrogate Marker for CDKN2A/B Deletion in MPNST and ANNUBP

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Background: Malignant peripheral nerve sheath tumors (MPNSTs) arise from schwann cells, often in the setting of neurofibromatosis type 1 (NF1), while atypical neurofibromatous neoplasms of uncertain biologic potential (ANNUBP) represent intermediate lesions that may progress to MPNST. Diagnosis remains challenging due to overlapping features with other sarcomas. Although markers such as S100, SOX10, and H3K27me3 assist in diagnosis, they lack sufficient specificity. S-methyl-5′-thioadenosine phosphorylase (MTAP), positioned adjacent to CDKN2A/B on chr9p21, has emerged as a surrogate marker for these deletions. This study compares MTAP and p16 IHC expression patterns, assessing their accuracy for detecting CDKN2A/B deletions compared with FISH analysis.

Methods: Cases diagnosed between 2014 and 2024 were stained for H3K27me3, MTAP, and p16. Complete loss of expression with positive internal control was considered a diffuse loss. MTAP and p16 expression in ≥20% of tumor cells was considered retained, ≤5% as loss, and 5–20% as equivocal. CDKN2A/B alterations were further evaluated with FISH.

Results: 36 cases were included (31 MPNST/5 ANNUBPs) with a mean age of 47.86 years. H3K27me3 expression was lost in 29/36(80.6%). MTAP expression was retained in 24 cases, lost in 10, and equivocal in 2. p16 expression was lost in 20/36(55.6%). FISH analysis was successful in 27 cases; 6(22.2%) demonstrated loss, while 21(77.8%) showed no alterations. 4/5 concordance between p16 and MTAP expression was seen in ANNUBP, while MPNSTs demonstrated concordance in 18/31(58.1%) cases. 12/14 discordant cases, exhibited complete loss of p16. FISH revealed, 8(57.1%) of discordant cases showed concordance with MTAP expression.

Conclusions: MTAP can serve as an alternative to p16 in detecting CDKN2A/B alterations. Although the overall concordance rate between MTAP and p16 expression was 61.6%, their combined assessment provides insight into deletion and aids in the decision of further testing.

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Disseminated and Metastatic Choroid Plexus Tumors: Patterns, Treatment, and Outcomes

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Background: Disseminated and metastatic choroid plexus tumors (CPTs) are rare, and published data on dissemination patterns, treatment, and outcomes remain limited.

Methods: We conducted a PRISMA 2020–guided systematic review, and included 47 full-text reports, yielding 51 pooled cases (37 adults, 14 pediatric). Dissemination was defined as ≥3 lesions at presentation or any number of lesions with drop metastasis. Patients were grouped as pediatric (< 18 years) or adult (≥18 years), and tumors were classified as choroid plexus papilloma (CPP), atypical choroid plexus papilloma (ACPP), or choroid plexus carcinoma (CPC). Dissemination pattern and clinical outcome (stable disease, progressed disease, expired) were recorded. Progression-free survival (PFS) was analyzed when available.

Results: Histology differed by age: adults predominantly had CPP (73.0%), whereas pediatric cases had more CPC (50.0% vs 10.8% in adults, p_adj=0.016). Adults more often presented with a single lesion (58.3%), while pediatric cases more frequently presented with disseminated disease (76.9%, p=0.05). Dissemination was mainly limited to the neuraxis (intracranial and/or spinal); extracranial metastasis was rare and occurred only in two CPC cases (abdominal shunt-related spread or detected at autopsy). Median PFS differed across histologic subtypes, with CPP longest and CPC shortest; this pattern was also seen on descriptive age stratification (adults: CPP 84.0 vs CPC 5.5 months; pediatrics: CPP 42.0 vs CPC 10.5 months). CPP had significantly longer PFS than CPC (p_adj = 0.0005), while CPP vs ACPP and ACPP vs CPC were not significant. PFS trended longer for single-lesion presentation (median 72 months, IQR 32.5–126) than for disseminated presentation (median 24 months, IQR 7.25–66), but was not significant (p=0.095).

Conclusions: Disseminated CPTs show age- and histology-associated differences in presentation and outcomes, with neuraxis-limited spread predominating and extracranial metastasis rare.

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Delineating the Expression of Lung Adenocarcinoma Markers in Choroid Plexus Tumors

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Background: Choroid plexus tumors are rare intraventricular papillary neoplasms that occur most commonly in younger children. While these tumors have a distinct morphology on microscopic examination, we present a sentinel case of an intraventricular mass with a papillary architecture lined with epithelial cells mimicking the morphology of choroid plexus tumors. This lesion displayed an immunohistochemistry (IHC) profile of a primary lung carcinoma (CK7-positive, CK20-negative, TTF1-postive and S100-negative) in an adult patient with smoking history but no dominant known primary lung mass. This initial profile was potentially compatible with the typical immunophenotypic profile of choroid plexus which is reported as positive for CK7, S100, Kir7.1, and transthyretin, and usually negative for CK20. Without in-house access to the Kir7.1 and transthyretin stain, a diagnostic dilemma was present as rare cases of solitary intraventricular lung adenocarcinoma metastasis have been reported, and an apparent lack of literature characterizing lung marker expression including TTF-1 and Napsin A was identified. While rare, a missed intraventricular lung adenocarcinoma metastasis could lead to missing the primary lung lesion and constitute radically different patient management strategies and outcome.

Methods: This study examined this gap by looking at expression of a standard battery of IHC markers for lung adenocarcinoma (CK7, CK20, TTF-1, and Napsin A) to stain previous cases of choroid plexus tumors from the UVMMC and NYU Langone Medical Center to further delineate the immunophenotype.

Results: Immunohistochemical examination of the evaluated cases revealed consistent patchy cytoplasmic expression of CK7 in a minority of cells, with rare cases displaying faint focal cytoplasmic expression of CK20. TTF1 and Napsin A were negative in all examined cases.

Conclusions: Overall these results affirm the previous characterization of CK7 and CK20 expression in choroid plexus tumors and support TTF1 and Napsin A as potential reliable markers in the differential between intraventricular lung adenocarcinoma metastasis and choroid plexus lesions.

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The rarest of the rare: Histologic and molecular features of pituicytomas with ependymal features

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Background: Pituicytomas are rare low-grade neoplasms composed of pituicytes of the posterior pituitary gland and infundibulum exhibiting various histologic types. While methylation-based classification studies on pituicytomas have shown close clustering of these tumors suggesting a single spectrum of the tumor types with a shared histogenesis, these studies have been restricted to the most common tumor types, excluding the rarest type with ependymal features. As such, little is known about the genetic and epigenetic profiles of pituicytomas with ependymal features.

Methods: Herein, we report two cases of pituicytoma with ependymal features with extensive workup including next-generation sequencing (NGS) and DNA methylation profiling (DNAMP).

Results: Patient 1 is a 59-year-old female who presented with severe headaches, nausea, vomiting, and hypotension with syncope. MR imaging showed a suprasellar mass. Patient 2 is an 80-year-old female with a history of breast cancer who was found to have a suprasellar mass on surveillance MR imaging concerning for metastatic disease. The pathology in both cases showed tumor cells with round to oval nuclei, perivascular pseudorosettes surrounding hyalinized blood vessels and inconspicuous mitotic figures. By immunohistochemistry, the tumor cells were positive for GFAP, partially positive for TTF1, and showed dot-like staining for EMA, D2-40, and CD99. In patient 1, NGS demonstrated a likely pathogenic missense mutation of NF2 accompanied by a frameshift mutation in EP300. An inactivating nonsense mutation in BRIP1 was identified in patient 2. DNAMP for patient 1 showed no match to a specific class, but was indicative of an ependymal-family neoplasm, while a suggestive score of pituicytoma, granular cell tumor of the sellar region, and spindle-cell oncocytoma was reported for patient 2.

Conclusions: Pituicytomas with ependymal features are underrepresented in methylation-based studies of pituicytomas. Further genetic and epigenetic studies are needed to better understand their biology.

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Germinoma Arising in Association With an Intracranial Epidermoid Cyst
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Background: Intracranial epidermoid cysts are benign congenital lesions derived from ectodermal inclusions and are typically characterized by slow growth and indolent clinical behavior. Malignant or neoplastic processes arising in association with epidermoid cysts are exceedingly rare. Recognition of such entities is critical, as failure to adequately sample suspicious areas may result in missed diagnoses with significant therapeutic implications. We report a rare case of germinoma arising in association with an epidermoid cyst in a 17-year-old male initially diagnosed with a third ventricular epidermoid cyst at age 15, who underwent endoscopic debulking. Two years later, follow-up MRI demonstrated interval enlargement of the residual cyst with progression of an eccentric enhancing component, prompting repeat endoscopic debulking and biopsy.

Methods: Case report

Results: Neuropathologic examination confirmed the persistent epidermoid cyst, while the enhancing component lacked squamous differentiation (p40 negative) and demonstrated tumor cells positive for OCT3/4 and CD117, with scattered β-hCG positivity, consistent with germinoma.

Conclusions: This case highlights the importance of close radiologic surveillance and targeted sampling of enhancing components in otherwise benign-appearing intracranial cystic lesions.

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Dural-Based Histiocytic Neoplasm with KRAS p.A146P Mutation: A Diagnostic Challenge
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Background: Histiocytic neoplasms represent a rare and heterogenous group of hematopoietic disorders, including Langerhans cell histiocytosis, Erdheim-Chester disease, Rosai-Dorfman disease, and histiocytic sarcoma. These neoplasms can be diagnostically complex, requiring histopathological and molecular profiling alongside systemic staging information for accurate diagnosis.

Methods: We present a case of a 62-year-old woman who presented for neurosurgical evaluation after experiencing worsening ataxia and anomia. Neuroimaging studies revealed a 5.0 x 3.6 x 3.4 cm enhancing extra-axial mass of the left tentorial leaflet with local parenchymal mass effect and vasogenic edema. Mild pituitary stalk thickening was also noted. A gross total neurosurgical resection was performed.

Results: Histopathological examination revealed sheets of atypical epithelioid cells with large, eccentrically located, rounded nuclei with moderately-sized nucleoli and abundant, amphophilic, bubbly to vacuolated cytoplasm. The atypical cells rested within a background of patchy fibrosis and extensive chronic inflammatory cell infiltrates. There were some foci consistent with emperipolesis. On immunohistochemistry, lesional cells strongly expressed CD68 and CD163, and showed abnormal reactivity for S100 protein. CD1a and ALK were negative. Cyclin D1 expression was variable. OCT2, a marker established for Rosai-Dorfman disease in extra-cranial cases, was negative. Next-generation DNA sequencing was performed, revealing a KRAS p.A146P mutation at variant allele frequency of 15%. This mutation has been reported in the setting of systemic Rosai-Dorfman disease and not yet reported in Erdheim-Chester disease.

Conclusions: This case describes a histiocytic neoplasm presenting as a dural-based mass, which was resected and evaluated by neuropathology prior to systemic staging, raising a diagnostic challenge. The histomorphologic appearance and molecular features favored Rosai-Dorfman Disease, while pituitary stalk involvement and negative OCT2 were consistent with Erdheim-Chester disease. This case highlights the diagnostic challenge posed by these tumors due to their phenotypic overlap, rarity, and requirement for integration of morphology, immunohistochemistry, molecular testing, and clinical-radiological correlation for accurate diagnoses.

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Erdheim-Chester Disease Presenting as a Chronic Subdural Hematoma

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Background: Erdheim-Chester disease (ECD) is a rare neoplastic histiocytosis that often involves multiple organs including the long bones of the extremities, kidneys, lungs, skin, and central nervous system. An initial presentation of neurological symptoms occurs in only around a fifth of ECD cases, while a minority of ECD cases demonstrate dural involvement, increasing the risk of atraumatic subdural hematomas (SDH). An initial presentation of ECD without bony involvement as an atraumatic SDH represents a significant diagnostic pitfall.

Methods: We report on a case of a 77-year-old woman who presented with headache, mild left-sided weakness, and issues with balance over the previous two weeks. She was found to have a large right hemispheric SDH by CT scan, for which she underwent evacuation and middle meningeal artery embolization. She was discharged and remained clinically and radiologically stable for several months, until her headaches worsened and the hematoma's size and mass effect increased. This necessitated re-evacuation and the specimen was submitted to neuropathology.

Results: The specimen consisted of a hematoma with both acute and organizing components, replete with prominent reactive fibroblasts, histiocytes, and xanthomatous cells. The apparent exuberant xanthomatous change and the presence of numerous Touton giant cells prompted further immunohistochemical investigations, revealing an abundance of cytoplasmic CD68, fascin, and factor 13A immunoexpression. S100 and CD1A were immunonegative throughout. Although initially called into question due to hemosiderin deposition, there was BRAFV600E immunopositivity throughout, leading to this mutation being confirmed by next-generation sequencing and to the diagnosis of ECD. The patient subsequently underwent an FDG PET/CT scan, revealing FDG-avid foci of long bone and possible perinephric involvement, a finding typical of this condition.

Conclusions: This case illustrates the importance of ancillary testing given sufficiently suspicious histological, immunohistochemical, and clinical findings in this rare initial presentation of a rare histiocytic neoplasm.

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Central Nervous System Histiocytosis: Diagnostic Challenges and the Importance of Accurate Classification

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Background: Histiocytic disorders in the central nervous system (CNS) are rare and include various entities with overlapping morphologic and immunophenotypic characteristics. Diagnosing histiocytic disorders is challenging as they may mimic reactive changes, like infectious/inflammatory conditions or the infiltrating growth pattern of some glial neoplasms. Accurate classification is essential for targeted therapies aimed at specific molecular alterations.

Methods: We present a case series demonstrating the variations in presentation and diagnostic features among histiocytic entities including Erdheim-Chester Disease (ECD), Juvenile Xanthogranuloma (JXG) and ALK-positive histiocytosis.

Results: With ages spanning 20s-40s and both sexes, represented manifestations included diplopia, dizziness, difficulty walking, and seizures. Locations varied from systemic disease to CNS localization. Initial clinical, radiologic, and pathologic differentials included brain tumors, metastatic disease, lymphoma, demyelinating disorders, inflammatory diseases, or infections. Morphology included oval to round or elongated/spindled nuclei with abundant, foamy, pale, eosinophilic cytoplasm. Admixed eosinophils and Touton-like giant cells were seen in some cases. Neoplastic histiocytes showed immunopositivity for CD163 and cyclin D1. Factor XIIIa, S100, and PU.1 were positive when performed. Molecular analysis of tissue showed alterations involving KRAS, TP53, CDK4, ALK, BRAF, ARID1A or NOTCH1. Histiocytoses require precise integration of morphologic, immunophenotypic, and molecular data for accurate classification. Finding KRAS, BRAF and ALK alterations qualified patients for targeted treatments. One patient experienced adverse effects with cobimetinib, melphalan, pembrolizumab, and radiation and succumbed to the disease after three months. A second patient passed away from the disease during the second line of treatment, chemotherapy after cobimetinib. Another patient showed partial response to radiation and alectinib but was lost to follow up. A patient has achieved partial response with cobimetinib, pexidartinib, cladribine, and cytarabine over the last two years, and is now enrolled in a trial.

Conclusions: This case series showcases the importance and complexities of CNS histiocytosis diagnoses, including potential pitfalls and critical diagnostic insights.

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Leptomeningeal Histiocytic Sarcoma: A case report

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Background: Leptomeningeal Histiocytic Sarcoma (HS) without brain involvement is a rare neoplasm with only 3 described cases (PMIDs 8888815/28548244/35062029). Important criteria to satisfy diagnosis of HS are expression of at least 2 histiocytic markers and exclusion of its mimics, requiring a panel of immunostains. Genomic profiling of HS has revealed 2 molecular subgroups based on presence or absence of NF1/PTP11 mutations; the wild type NF1/PTP11 group frequently shows activating mutations in RAS/MAPK pathway (PMID 31439678).

Methods: A 15-year-old boy presented with lower back pain for 4 months and intermittent headaches for a month. MRI showed an ill-defined, heterogeneously enhancing mass at T11 nerve root and diffuse involvement of leptomeninges of the brain. Cytopathologic examination of CSF revealed few large, atypical cells and flow cytometric examination was negative for hematolymphoid neoplasm. Biopsies of soft tissue adjacent to thoracic spinal nerve roots and of right cerebral temporal lobe were performed.

Results: The thoracic soft tissue biopsy showed marked crush artifact, but the temporal lobe biopsy showed a histiocytic tumor limited to the leptomeninges. The tumor cells were diffusely immunoreactive for CD4, PU.1, and p16; focally immunoreactive for Cyclin D1 and pS6RP; and weakly immunoreactive for CD68, CD163, and Lysozyme. An extensive panel of immunostains was performed to exclude primary brain neoplasm, myeloid sarcoma, lymphoma, dendritic cell tumors, germ cell tumors, and carcinoma. Targeted next generation sequencing showed IDH1R132C and SF3B1R625C mutations. Consistent with the IDH1R132C mutation, CSF D-2-hydroxyglutarate (D2HG) and D2HG:L2HG ratio were markedly elevated. Peripheral blood serum showed normal levels of D2HG but mildly elevated D2HG:L2HG ratio.

Conclusions: We present the 4th case of disseminated leptomeningeal HS and describe its immunophenotype. The presence of IDH1R132C and SF3B1R625C mutations suggest overlap with myeloid neoplasms. Importantly, in patients with IDH-mutated CNS tumors, the CSF D2HG and D2HG:L2HG ratio may serve as biomarkers for monitoring treatment response.

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Primary Diffuse Meningeal Melanomatosis with CNS Invasion and NRAS Q61L Mutation

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Background: Primary diffuse meningeal melanocytic neoplasms (PMMN) are rare tumors arising from leptomeningeal melanocytes. They range from melanocytosis (bland proliferation) to melanomatosis (atypical cells with CNS invasion) and are frequently driven by postzygotic NRAS somatic mutations, triggering constitutive activation of the MAPK and PI3K signaling pathways. Both carry poor prognosis once neurological symptoms develop.

Methods: A 60-odd-year-old male presented with a 3-month progressive history of postural and kinetic tremor, followed by dysgraphia and gait instability. Neurological examination showed left-sided dysmetria, bradykinesia, and hyperreflexia. Brain MRI demonstrated extensive diffuse leptomeningeal enhancement involving the cerebellopontine angles and internal auditory canals. Cerebrospinal fluid analysis revealed marked hyperproteinorrhachia (5 g/L) and atypical melanocytic cells. Meningeal and cortical biopsies were performed, and immunohistochemical and targeted next-generation sequencing (NGS) characterization followed.

Results: Histological examination showed a sheet-like melanocytic proliferation with moderate cytologic atypia and abundant melanin pigment diffusely expanding within the subarachnoid space, with focal invasion of brain tissue. Tumor cells were strongly positive for SOX10 and Melan-A, showed patchy HMB45 reactivity, and retained BAP1 expression. Conversely, epithelial, neuroendocrine, and glial markers were negative, with a 2% Ki-67 index. NGS identified a pathogenic NRAS p.Q61L mutation (VAF 33.3%), without BRAF, GNAQ, or GNA11 alterations. Skin examination, PET-CT, total-body CT, and endoscopy excluded a primary extracranial melanoma. Clinical course after biopsy was complicated by hydrocephalus and status epilepticus, requiring corticosteroids and antiepileptics. Despite palliative systemic anti–PD-1 immunotherapy, the patient deteriorated and died two months after diagnosis.

Conclusions: The diagnosis of primary diffuse leptomeningeal melanomatosis was based on diffuse meningeal involvement and CNS parenchymal invasion, which are hallmarks of malignancy regardless of mitotic activity in PMMN. This case supports NRAS codon 61 mutations as a characteristic molecular driver and underscores the importance of clinicopathological correlation in diffuse leptomeningeal lesions.

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Circumscribed Meningeal Melanocytic Neoplasms, WHO CNS5 Classification and Outcomes

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Background: Circumscribed meningeal melanocytic neoplasms (CMMN) represent a spectrum of disease including melanocytoma, intermediate-grade melanocytic tumor (IMT), and melanoma (WHO CNS5). Clinical behavior of these rare tumors and correlation with underlying molecular alterations remain limited.

Methods: In this single-institution analysis, slides and pathology reports from 31 patients (14 males, 17 females; median age 59) diagnosed with CMMN (2004-2025) were reviewed. NGS analysis (GNAQ/GNA11, BAP1, EIF1AX, SF3B1) was performed in 24 cases. Clinical histories and outcomes were reviewed.

Results: Tumors included 7 melanocytoma, 18 IMT, 5 melanoma, and 1 indeterminate (due to limited tissue). Mutations were detected in GNAQ/GNA11 (21 of 24), BAP1 (5 of 23: 2 IMT, 3 melanoma), EIF1AX (9 of 21: 1 melanocytoma, 8 IMT), and SF3B1 (3 of 23: 2 IMT, 1 melanoma). Primary sites were spinal (n = 21), posterior fossa (n = 7), or supratentorial (n = 3). Of 24 patients (6 melanocytoma, 13 IMT, 5 melanoma) with available follow-up: 5 melanocytoma recurred (12-51 months), 1 died (2-year PFS 83%, 5-year OS 80%); 9 IMT recurred (1-49 months), 8 died (2-year PFS 56%, 5-year OS 68.2%); 4 melanoma recurred (1-29.3 months), 3 died, 1 was alive with disease at 11 months and 1 had leptomeningeal dissemination, transitioned to hospice following biopsy and died 1 month later (2-year PFS 25%, 5-year OS 0%). BAP1 mutation correlated with worse OS (hazard ratio 9.67, p = 0.0043). Of 18 patients with documented recurrence, upfront treatment included resection, gross total in 7, subtotal in 8, biopsy-only in 3; adjuvant treatment included radiotherapy in 6, systemic therapy (temozolomide and/or immunotherapy) in 3, while 9 were observed only.

Conclusions: In this analysis, recurrence rate was high for all CMMN regardless of subtype and mutational profile. IMT and melanoma were associated with higher rates of mortality compared to melanocytoma. BAP1 mutation correlated with worse OS.

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A Rare Case of Malignant Metastatic Melanoma to the Central Nervous System with an IDH1 (R132C) mutation

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Background: Isocitrate dehydrogenase (IDH) mutations are rare in melanoma and occur predominantly in metastatic rather than primary tumors, with IDH1 R132C (arginine-to-cysteine substitution at codon 132) being the most frequently reported variant. Prior studies show that ~ 90% of IDH1-mutant melanomas harbor concurrent MAPK pathway alterations, supporting a cooperative role in melanoma pathogenesis. Additionally, BRAF mutant tumors with a concurrent TERT promoter mutation are associated with worse survival. To date, no published reports have specifically described IDH1 R132C–mutant melanoma brain metastases. Here, we report a case of an 85-year-old male with an enhancing, intra-axial left superior frontal lobe mass, found to have metastatic melanoma to the central nervous system (CNS) harboring an IDH1 R132C mutation.

Methods: A craniotomy with resection revealed a malignant neoplasm demonstrating sheets of pleomorphic, epithelioid to plasmacytoid cells with eccentric nuclei, prominent red nucleoli, and abundant eosinophilic cytoplasm. There were frequent mitoses with intratumoral pigment deposition and hemorrhage. Melanocytic markers were diffusely positive (Melan-A, SOX-10, HMB45). Next generation sequencing revealed alterations in BRAF V600K, TERT promoter, and IDH1 (R132C) genes.

Results: This case describes a rare finding of IDH1 R132C–mutant melanoma metastatic to the brain with concurrent BRAF V600K and TERT promoter mutations. This molecular profile supports prior evidence that IDH1 mutations in metastatic melanoma coexist with MAPK pathway activation and may contribute to highly aggressive tumor behavior when combined with TERT promoter alterations.

Conclusions: This report adds to the limited body of data on IDH1-mutant metastatic melanoma to the CNS. It may help inform future studies and potential clinical trials evaluating IDH-targeted therapeutic approaches in metastatic melanoma which is especially critical in BRAF V600K mutated tumors which reportedly shows lower response rates and shorter progression-free survival in response to BRAF and MEK inhibition compared to V600E mutants.

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A Classic Neural Crest Lineage Diagnostic Trap: Cellular Schwannoma Versus Metastatic Spindle Cell Melanoma: A Case Report

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Background: Neural crest–derived tumors present diagnostic challenges, as benign entities such as cellular schwannoma (CS) may mimic spindle cell melanoma (SCM) or low-grade malignant peripheral nerve sheath tumor (MPNST). Accurate distinction requires integration of histologic, immunophenotypic, and molecular findings. This case illustrates the diagnostic complexity and clinical implications of neural crest lineage overlap, particularly in the setting of molecular discordance.

Methods: A 65-year-old male presented with lower back pain and extremity weakness. An L4 spinal mass was resected and evaluated by light microscopy (LM) and immunohistochemistry (IHC). Three months later, two cutaneous lesions appeared and were diagnosed as SCM at an outside institution by LM, IHC, and molecular studies. A tertiary referral center re-evaluated all specimens, performing additional IHC and molecular studies on the spinal lesion.

Results: The spinal lesion initial diagnosis was CS, supported by spindle cell morphology with wavy nuclei, low mitotic activity, absence of necrosis, diffuse S100 and SOX10 positivity, and retained H3K27me3 expression. The cutaneous lesions were diagnosed as spindle cell melanoma, demonstrating SOX10, S100, and PRAME positivity, negative HMB45 and MART1 staining, and pathogenic BRAF, TERT, and CDKN2A mutations. Upon tertiary review, the spinal lesion showed PRAME positivity, negative HMB45 and MART1 staining, and was negative for BRAF V600E, RAS Q61R, and pan-melanoma markers, creating molecular discordance. Methylation profiling is pending to clarify lineage.

Conclusions: This case underscores the morphologic and immunophenotypic overlap between cellular schwannoma, low-grade MPNST, and metastatic spindle cell melanoma - a classic neural crest diagnostic trap. To minimize misclassification, we emphasize: (1) careful review of imaging; (2) assessment for encapsulation and growth pattern; (3) inclusion of PRAME when evaluating CS or MPNST; and (4) consideration of methylation profiling when morphology, IHC, and molecular testing are discordant. Metastatic SCM should remain in the differential diagnosis of any spinal extra-axial spindle cell neoplasms of neural crest origin.

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Corticotroph (Basophil) invasion of the posterior pituitary in the setting of pituitary adenoma (PitNET): A diagnostic pitfall

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Background: Pituitary adenoma is a clonal neoplastic proliferation of anterior pituitary cells with loss of normal pituitary architecture. Although pituitary tumors are histologically benign, they may exhibit aggressive behavior, including invasion into adjacent tissues, such as the sphenoid sinus, dura mater, posterior pituitary, and brain. However, the presence of corticotroph cells in the neurohypophysis should not be interpreted immediately as tumor invasion, as it may also represent physiologic migration from the adenohypophysis, a phenomenon known as corticotroph “basophil” invasion. Here, we report a middle-aged woman diagnosed with pituitary neuroendocrine tumor, with an incidental finding of basophil invasion of the neurohypophysis.

Methods: A 51-year-old female presented with 8 years of progressive enlargement of the hands and feet, headaches, voice changes, ring-size changes, and prominence of facial bones. IGF-1 is elevated (405 ng/mL), and the MRI demonstrated an asymmetric zone of diminished enhancement and heterogeneous signal abnormality within the right lateral pituitary gland consistent with a pituitary microadenoma (5-6 mm in size).

Results: Transsphenoidal hypophysectomy biopsy revealed monomorphic tumor cells arranged diffusely, with uniform nuclear morphology, inconspicuous nucleoli, and densely granulated cytoplasm. The tumor cells are immunoreactive for growth hormone, prolactin, and PIT1, consistent with the diagnosis of a mammo-somatotroph pituitary adenoma. There is also a normal neurohypophysis with sheets of monomorphic basophilic cells infiltrating the posterior pituitary parenchyma. These basophilic cells have a different immunoprofile with the pituitary adenoma and are positive on ACTH and TPIT, consistent with corticotroph invasion.

Conclusions: Corticotroph (basophil) invasion refers to the spread of normal ACTH-producing cells in the surrounding tissue, most commonly the posterior pituitary gland. It may be diagnostically challenging because these monomorphic adenohypophyseal cells are not native to the neurohypophysis, and their presence can mimic adenoma, necessitating careful histologic, immunophenotypic, and clinicoradiologic correlation.

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Skull base metastasis of adenocarcinoma arising from mediastinal teratoma with malignant transformation: A case report

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Background: Teratomas are tumors that contain at least two of the three germ cell layers: endoderm, mesoderm, and ectoderm. Pure mediastinal teratomas are classified as either mature or immature. Teratomas of either type may also occur as part of a mixed germ cell tumor. Mediastinal germ cell tumors with somatic-type solid malignancies most commonly arise from mature teratomas, with most of these malignancies being metastatic at the time of diagnosis. Common sites of metastasis include adjacent structures such as bone, lungs, and thoracic lymph nodes. Here, we report a case of a skull base metastasis from a gastrointestinal-type adenocarcinoma arising from a mature teratoma in a young male.

Methods: A 25-year-old male presented with a sudden two-week history of right eye blurriness and diplopia, and right facial droop. A prior resection of the right anterior mediastinal mass revealed adenocarcinoma arising in a mature cystic teratoma. Brain MRI revealed a solid enhancing mass within the right temporal petrous portion and right parietal and perieto-occipital calvarial masses with extracranial and intracranial extension, consistent with calvarial metastasis, without any evidence of parenchymal cerebral involvement. An MRI spine survey showed scattered osseous metastasis with pathologic compression fracture.

Results: The patient underwent right temporal craniotomy, and the biopsy of the right skull base showed metastatic adenocarcinoma identical to that present in the mediastinum.

Conclusions: To our knowledge, this appears to be the first reported case of a skull base metastasis of adenocarcinoma arising from teratoma with a somatic type solid malignancy of the mediastinum. This highlights a previously undocumented metastatic pattern expanding the known spectrum of metastatic behavior in mediastinal tumors and should raise clinical suspicion in prior teratomas presenting with new-onset focal neurological deficits or signs of increased intracranial pressure.

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Brain Metastases from Renal Cell Carcinoma: A Histologic and Molecular Analysis with Correlation to the Primary Tumor

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Background: Renal cell carcinoma (RCC) infrequently metastasizes to the brain. While the demographic and clinical characteristics of RCC brain metastases have been described, their detailed histologic and molecular features remain incompletely characterized.

Methods: We performed a retrospective review of resected RCC brain metastases at our institution from 2010 to 2026. Clinical data included patient demographics, disease course, and treatment. Pathologic variables included tumor grade, stage, histologic features, immunohistochemical (IHC) profile, and molecular findings. Molecular studies comprised DNA next-generation sequencing (NGS), RNA-based fusion assays, and chromosomal microarray analysis when available.

Results: Twenty-five patients underwent resection of RCC brain metastases over a 15-year period, including 24 clear cell RCCs and one NONO::TFE3-rearranged RCC. The median ISUP/WHO nucleolar grade of primary tumors was Grade 3 (n=13; 3 Grade 2, 7 Grade 3, 3 Grade 4). The median grade of the brain metastases was Grade 3 (n=15; 4 Grade 2, 9 Grade 3, 2 Grade 4). Among eight cases with both primary and metastatic specimens available for comparison, two demonstrated lower nucleolar grade in the metastatic lesion. Histologically, necrosis was identified in 9 of 15 cases (60%), and mitotic activity in 10 (67%), including atypical mitoses in 2 cases (13.3%). Nine patients underwent NGS, confirming eight clear cell RCCs and one NONO::TFE3-rearranged RCC. VHL mutations were most frequent (n=5), followed by PBRM1 mutations (n=2). Copy number variants involving FOXP1 and MITF were identified in two cases. Two patients with chromosomal microarray analysis demonstrated recurrent alterations, including loss of 8p and 14, segmental gains of Xq and 5q, and segmental losses of 3p and 6q.

Conclusions: RCC brain metastases most commonly retain clear cell morphology and intermediate to high nucleolar grade. In cases with non-classic histologic features, IHC is often sufficient for accurate classification; however, molecular profiling provides valuable adjunctive information and may be critical in diagnostically challenging cases.

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Chemoimmunotherapy-Associated Lineage Plasticity in NSCLC with RB1/TP53 Co-Alteration and Cerebellar Metastasis

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Background: Histologic transformation is a recognized mechanism of therapeutic resistance in non-small cell lung cancer (NSCLC). Co-alteration of RB1 and TP53 is characteristic of high-grade neuroendocrine carcinomas and may facilitate therapy-induced lineage plasticity under therapeutic pressure.

Methods: Clinical, radiologic, histopathologic, immunophenotypic, and comprehensive molecular profiling data were reviewed for a patient who developed a cerebellar metastasis following multimodal therapy for NSCLC.

Results: A 57-year-old man with a 40 pack-year smoking history was diagnosed with bulky right upper lobe NSCLC without mediastinal lymphadenopathy. Biopsy showed non-small cell carcinoma favoring squamous differentiation (p40+, p63+, TTF-1–, Napsin A–). He received neoadjuvant albumin-bound paclitaxel, cisplatin, and pembrolizumab followed by right upper lobectomy, lower lobe segmentectomy, and adjuvant radiotherapy. Nine months later, imaging revealed a 2.7 cm ring-enhancing left cerebellar mass with vasogenic edema. The first cerebellar resection demonstrated metastatic carcinoma composed of non-descript sheets of markedly pleomorphic cells with increased nuclear-to-cytoplasmic ratio, brisk mitotic activity, and extensive areas of necrosis. No overt glandular or squamous differentiation was identified. Ki-67 approached 100%. A second resection showed poorly differentiated carcinoma with extensive necrosis and persistent sheet-like growth, near-complete loss of squamous markers (p40 < 1%, CK5/6–, p63–), partial TTF-1 expression (40–50%), PD-L1 expression (10–20%), and negative synaptophysin. Molecular profiling revealed RB1 loss-of-function (p.E320fs9), TP53 mutation (p.Y163C), STK11 mutation (p.P281Rfs6), MYC amplification, ALK amplification, and FANCA mutation (p.R756H). The patient died six months after diagnosis of cerebellar metastasis.

Conclusions: This case illustrates therapy-induced clonal evolution and lineage plasticity in NSCLC, where RB1/TP53 co-alteration combined with STK11 mutation, MYC and ALK amplification, and FANCA mutation drove a highly proliferative cerebellar metastasis. Therapeutic pressure may select aggressive subclones, promoting adaptive resistance and treatment escape, highlighting the importance of integrated histopathologic and molecular evaluation to guide accurate diagnosis, prognostication, and therapeutic strategies in CNS metastases.

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Post-treatment Differentiation in Metastatic Sinonasal Small Cell Neuroendocrine Carcinoma with Central Nervous System (CNS) Involvement

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Background: We present a case of a 24-year-old man with a history of T3N0M0 sinonasal small cell neuroendocrine carcinoma (SNEC) who later developed leptomeningeal metastases with an unusual post-therapy histomorphology.

Methods: Not applicable.

Results: At the time of initial presentation, the patient had a 1-year history of worsening epistaxis, nasal congestion, and a sensation of facial pressure. Imaging studies showed an enhancing and expansile left nasal cavity mass. The patient was treated with endoscopic resection, gamma knife radiosurgery, and adjuvant chemoradiotherapy, which was completed 4 months later. Post-resection surveillance imaging showed the development of multiple left hemispheric dural-based masses, the largest of which measured 4.4 cm. These dural based masses were detected 11 months following the initial resection. Imaging findings at that time included mass effect on the left cerebral hemisphere with increased confluent T2 hyperintense signal in the left frontal lobe, increased effacement of the left lateral ventricle, and a mild increase in rightward midline shift measuring up to 7 mm. At the time the dural lesions were detected the patient was asymptomatic. Craniotomy for resection of the tumor was performed a few weeks following the completion of two cycles of chemoimmunotherapy. Pathology of the CNS resection consisted predominantly of hemorrhage and dura mater with dispersed small fragments of neoplasm. When compared to the histomorphology of the originally resected sinonasal carcinoma, the dural-based metastases were dissimilar. The treated metastasis demonstrated a morphology similar to differentiating neuroblasts within a background of neuritic processes.

Conclusions: This is a case of a man with sinonasal primary malignancy with a neuroepithelial/neuroendocrine component who later developed leptomeningeal metastases. A few prior case reports of sinonasal malignancies with primitive neuronal components demonstrating differentiation post-treatment have been published. We present this case as another rare example of such a phenomenon in a metastasis to the CNS.

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HER2-Positive Male Breast Cancer with Brain Metastasis: A Case Report
F Alkhutani, A Alkhotani; Umm Al-Qura University

Background: Male breast cancer is a rare entity, accounting for less than 1% of breast cancer vases. Due to its low incidence, evidence guiding management is largely extrapolated from studies conducted in female patients. Brain metastases from breast cancer are well recognized in advanced disease: however, centre nervous system involvement us male breast cancer remains exceedingly uncommon, with limited cases reported in the literature.

Methods: We report the case of a 49-year-old man with a history of left breast cancer who underwent modified radical mastectomy with en bloc axillary lymph node excision. Histopathology revealed invasive carcinoma of no special type (invasive ductal carcinoma, not otherwise specified), grade 2, which was estrogen receptor–positive (80%), progesterone receptor–positive (50%), and HER2-positive (immunohistochemistry 2+ with FISH positivity). The patient received adjuvant radiotherapy, trastuzumab, pertuzumab (17 cycles), and tamoxifen. He was subsequently lost to follow-up and re-presented three years later. Brain MRI demonstrated intra-axial lesions centered in the right temporoparietal region, with the largest measuring 2.3 × 2.2 × 2.5 cm. The lesions were predominantly cystic with peripheral enhancement and nodularity. The patient underwent right parieto-occipital craniotomy for tumour resection.

Results: Histological examination revealed metastatic carcinoma composed of malignant cells. Immunohistochemical analysis demonstrated CK7 positivity, estrogen receptor positivity (80%), progesterone receptor negativity (0%), and HER2 positivity (immunohistochemistry 2+). The diagnosis of metastatic carcinoma of breast origin was made. Unfortunately, the patient’s clinical condition deteriorated, and he passed away several months late.

Conclusions: This case highlights a rare occurrence of HER2-positive invasive carcinoma of the male breast with delayed brain metastasis. The findings underscore the aggressive nature of HER2 positive disease, the potential for receptor discordance in metastatic lesions, and the importance of comprehensive histologic and immunophenotypic evaluation in both primary and metastatic tumours. Reporting such cases contributes valuable insight into the clinicopathologic behavior of male breast cancer.

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Primary spinal cord adrenal cortical adenoma: A Case Report

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Background: Ectopic adrenal tissue, or adrenal rests, is a condition in which adrenal tissue is located outside its normal anatomic position. Ectopic adrenal tissue is not uncommon and is most often identified in the kidneys or gonads, in relatively close proximity to the native adrenal glands. Much literature attributes such anomalies to a defect in migration during embryogenesis. However, this mechanism does not adequately explain ectopic adrenal tissue in remote locations.

Methods: Immunohistochemistry, molecular testing, methylation profiling

Results: Here we report a case of a 71-year-old female, who while undergoing a workup for constipation and hematochezia, was noted to have an incidental spinal cord mass on abdominal CT. Subsequent MRI identified an intradural extramedullary enhancing nodule (11 mm) with a cystic component at level L1-L2. Based on the spinal MRI findings, the differential included a myxopapillary ependymoma, with an astrocytoma or schwannoma lower on the differential. Histologically, the tumor consisted of sheets of bland, oncocytic cells with abundant cytoplasm and round nuclei. After an extensive immunohistochemical workup, the lesional cells were diffusely positive for inhibin and negative for CD138, CD68, Pan-K, S100, SOX10, GFAP, OLIG2, EMA, TTF1, FOXJ1, and PAX8. MIB-1 was ~1%. Molecular testing with Oncopanel NGS sequencing of >400 cancer genes demonstrated no significant DNA variants, structural rearrangements, or copy number variants. Methylation analysis performed at the National Institutes of Health/National Cancer Institute showed a high confidence match to adrenal cortical adenoma and no similarity to common primary parenchymal brain tumors. Considering this new information, additional immunohistochemistry demonstrated that the lesion was positive for SF1 and synaptophysin and negative for Mart1/MelanA, chromogranin, and calretinin. Therefore, a diagnosis of primary spinal adrenal cortical adenoma was rendered.

Conclusions: Literature review shows this is one of a few cases reported. Prognosis is considered good, albeit followup and studies of cases in future will be needed.

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A Rare First Act: Brain Metastasis as Initial Clinical Presentation of Alveolar Soft-Part Sarcoma

P Loreto-Palacio, M Cohen, M Couce; University Hospitals Cleveland Medical Center/Case Western Reserve University

Background: Alveolar soft-part sarcoma (ASPS) is a rare mesenchymal neoplasm characterized by ASPSCR1::TFE3 gene fusion, typically arising in deep soft tissues of the extremities in young adults. Brain metastases are relatively common in advanced ASPS; however, initial presentation as an intracranial mass is exceptionally rare and presents unique diagnostic challenges.

Methods: Clinical, radiologic, and histopathologic evaluation of a patient with a solitary brain lesion, incorporating H&E staining, immunohistochemistry, and sarcoma fusion panel testing.

Results: A 35-year-old male presented with a two-week history of headaches. MRI revealed a right parietal mass (3.0 × 2.8 × 2.6 cm) with vasogenic edema and mass effect on the right lateral ventricle. Chest CT showed a heterogeneous left lower lobe mass (6.2 × 5.8 × 4.2 cm) extending to the major fissure and pleura, favoring metastatic disease. Brain histology demonstrated nests of polygonal cells with eosinophilic granular cytoplasm and vesicular nuclei with prominent nucleoli; PAS diastase-resistant staining was positive. Immunohistochemistry showed CD68/CD163 positivity and was negative for S100, synaptophysin, Melan-A, CAM5.2, GFAP, PAX8, OLIG-2, and INSM1. Molecular testing confirmed ASPSCR1::TFE3 fusion, diagnostic of ASPS. PET scan revealed uptake in the left lower leg; CT identified a heterogeneously enhancing soft-tissue mass in the proximal soleus (5.1 × 4.2 × 7.7 cm).

Conclusions: This case underscores the importance of integrating histopathology, immunophenotyping, and molecular diagnostics in evaluating intracranial masses without a known primary. While brain metastases occur in ASPS, initial presentation as a brain lesion is rare. Comprehensive workup, including PET, was essential to detect the primary tumor and guide management. Our findings add to limited literature on atypical ASPS presentations and reinforce the role of molecular profiling and whole-body imaging in sarcoma diagnosis.

Posters: Ophthalmic Pathology

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Sebaceous Carcinoma of the Eyelid Associated with Lynch Syndrome: Morphologic Features and Importance of MMR Testing

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Background: Sebaceous carcinoma (SC) is a rare malignant tumor arising from sebaceous glands, most commonly in the periocular region. SC has been associated with hereditary nonpolyposis colorectal carcinoma (HNPCC), also known as Lynch syndrome, an autosomal dominant disorder characterized by mismatch repair (MMR) deficiency and increased risk of visceral malignancies. We present two cases of periocular SC to highlight the well-differentiated morphology that may be associated with MMR-deficient syndromes such as Lynch and Muir-Torre.

Methods: Clinical and histopathologic evaluation of eyelid tumors, including immunohistochemistry for androgen receptor (AR), PRAME, epithelial markers, MMR proteins (MLH1, MSH2, MSH6, PMS2), and HPV in-situ hybridization.

Results: Both tumors demonstrated well-differentiated sebaceous carcinoma morphology and similar immunohistochemical profiles. In Case 1, the patient had a history of breast and colon carcinomas, while Case 2 involved a patient with history of sessile serrated adenoma and family history of colon cancer. Tumor cells in both cases were positive for Cam 5.2, EMA, AR. PRAME was positive in well-differentiated sebocytes. Immunohistochemistry for MMR proteins revealed combined loss of MSH2 and MSH6 in both tumors, confirming MMR deficiency. HPV in situ hybridization testing was also performed, and it was negative in both cases.

Conclusions: Sebaceous carcinoma may occur in association with MMR-deficient syndromes such as Lynch and Muir-Torre. These cases suggest that SC in this context can present with well-differentiated morphology, which could warrant consideration of MMR immunohistochemistry in sebaceous carcinomas, even when there is no known history of Lynch syndrome. This information is key to guide decisions regarding genetic counseling and testing in these patients.

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Xanthogranulomatous Epithelial Tumor (XGET) of the Orbit in a 4-Year-Old Boy
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Background: Xanthogranulomatous epithelial tumor (XGET) is a recently recognized entity characterized by giant cell morphology and HMGA2::NCOR2 gene fusion.

Methods: Herein, we report a case arising in the orbit of a child.

Results: A 4-year-old boy presented with proptosis. CT scan revealed a superior orbital mass with lytic bone erosion. Biopsy revealed a lesion rich in giant cells with fibrotic stroma. Immunohistochemistry showed positivity for CD68 in giant cells and negative staining for desmin, p63, and Langerhans cell histiocytosis markers. A diagnosis of “giant cell granuloma” was initially favored. The mass was resected five months after the initial biopsy. Morphology again showed a giant cell-rich lesion. Fibrohistiocytic and myofibroblastic features were more evident on the resection specimen than the biopsy, prompting further workup. The immunophenotype was not suggestive of solitary fibrous tumor (STAT6-negative), myofibroma (minimal SMA staining), or a dendritic cell proliferation (CD21-negative). An RNA fusion panel showed no rearrangements in STAT6 or USP6. Positive cytokeratin staining raised the possibility of XGET, but no HMGA2::NCOR2 rearrangement was identified on the fusion panel. A descriptive diagnosis of “fibrohistiocytic/myofibroblastic tumor with giant cells” was rendered. Fourteen months later, the reference laboratory indicated that an HMGA2::NCOR2 gene fusion was present, but had been missed due to technical problems. A final integrated diagnosis of XGET was rendered. Nineteen months post-resection, the patient was doing well with resolution of symptoms and no evidence of recurrence.

Conclusions: To our knowledge, this is only the second reported case of XGET involving the skull, and the first involving the orbit. It is also notable due to the young age of the patient. Although rare, XGET should be considered in the workup of giant cell-rich lesions. This case also highlights the morphologic overlap between XGET and other giant cell lesions, and the underappreciated value of epithelial markers in such tumors.

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Unexpected choroidal melanoma in evisceration for blind painful eye

G Yeaney, A Singh; Cleveland Clinic

Background: A 42-year-old female with history of retinopathy of prematurity and prior right eye evisceration presents with blind painful left eye for 8 months. Pain control was not possible after chlorazeapine injection, so evisceration was performed.

Methods: Left eye contents showed choroidal (uveal) melanoma, predominantly spindle B pattern, arising in the posterior choroid, measuring 2.7 mm diameter by 1.0 mm thick on slide (pTa1) in a background of disorganized retina with gliosis and calcification (phthisis bulbi). Immunohistochemical stains showed the following the tumor cells: double stain for MelanA and Ki67 index 5%; PRAME negative; SOX10 positive; BAP1 intact. Targeted Oncology Panel Next-Generation Sequencing revealed GNAQ p.Gln209Leu NM_002072.3:c.626A>T 7.6% VAF, Depth 997x, Ex5.

Results: Given the standard of enucleation for uveal melanoma and concern for potential tumor remaining in situ, enucleation-post-evisceration was performed to evaluate sclera and optic nerve. The entire sclera was submitted and showed macrophages, scarring, and granulation tissue, atrophic optic nerve, and no melanocytic proliferation. Discussion with ophthalmology and review of literature showed that standard of care in the case of blind painful eyes with prior uveal or conjunctival tumors is enucleation, and in those cases, evisceration is contraindicated. Imaging prior to surgery should be performed and may avoid unexpected outcomes in some cases. Some recommend that if suspicious ocular mass is found during evisceration surgery, the autologous sclera should not be reused, and it should be sent for histological analysis as well as the intraocular contents. However, this is not always realistic as surgeons may overlook small or non-pigmented ocular tumors. For this reason, unexpected melanoma in evisceration samples is rare but unavoidable.

Conclusions: We recommend entirely submitting evisceration tissue rather than taking representative sections. If tumor is found on evisceration and post-evisceration-enucleation is performed, submit entire sclera and remnants.

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Actinomyces-Associated Necrotizing Bilateral Corneal Infections in an Immunocompromised Patient

B Zeches, U Tran, P Paueksakon; Vanderbilt University Medical Center

Background: Actinomyces species are filamentous, gram-positive anaerobic bacteria that colonize the oral cavity and upper aerodigestive tract and can cause chronic, locally destructive infections characterized by sulfur granules. Ocular involvement is uncommon, and histologic features may overlap with organisms such as Nocardia. Accurate identification is important given distinct antimicrobial management strategies.

Methods: A 70-year-old man with chronic lymphocytic leukemia status post hematopoietic cell transplantation, diabetes mellitus, and sclerodermoid graft-versus-host disease with scleral lens use presented with bilateral necrotizing corneal infection. The patient had severe ocular surface disease and prior angiocatheter placement. Corneal tissue was submitted for histologic evaluation with special stains, followed by bacterial polymerase chain reaction (PCR) and 16S ribosomal RNA next-generation sequencing.

Results: Histologic sections demonstrated extensive tissue necrosis with abundant acute inflammation. Scattered sulfur granules were identified. Gram stain revealed numerous mixed bacterial organisms, composed of cocci, rods, and filamentous forms. Based on the grouped filamentous bacteria and sulfur granule-like material, Actinomyces was suspected, with Nocardia considered in the differential diagnosis. Molecular testing demonstrated a polymicrobial anaerobic infection, with Fusobacterium species related to Fusobacterium nucleatum in major abundance and trace levels of Actinomyces israelii and Eikenella species.

Conclusions: This case highlights the diagnostic challenges of necrotizing ocular infections in immunocompromised patients. The combination of graft-versus-host disease–related epithelial compromise, scleral lens use, and proximity to normal oral and sinonasal flora may predispose individuals to colonization by anaerobic organisms such as Actinomyces. Histologic suspicion of Actinomyces, particularly in the presence of sulfur granule-like structures, is critical to prompt appropriate molecular testing and to distinguish it from morphologic mimics such as Nocardia. Early recognition facilitates targeted antimicrobial therapy in severe corneal infections.

Posters: Infectious

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Withdrawn

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Molecular Detection of Naganishia globosa in a Destructive Pediatric Sellar Lesion: True Infection or Diagnostic Artifact?

R Joshi, B Lee, E Mohr, K Han; University of Wisconsin School of Medicine and Public Health

Background: Inflammatory sellar lesions frequently mimic neoplasms radiographically and clinically. The increasing use of molecular microbiology can be highly valuable, but it may also present diagnostic challenges when rare organisms are identified. Naganishia spp. are typically environmental yeasts that were formerly classified within the genus Cryptococcus but are now assigned to the family Filobasidiaceae. Reported human infections are rare and opportunistic, occurring mainly in immunocompromised hosts and involving pulmonary or cutaneous disease. An invasive central nervous system involvement is not well-documented.

Methods: A 13-year-old girl presented with panhypopituitarism and a 22-mm expansile intrasellar mass, favored to represent a pituitary macroadenoma or craniopharyngioma. The diagnostic workup included histologic examination along with special and immunohistochemical stains for infectious and neoplastic entities. Molecular microbiologic testing (e.g., broad-range PCR for bacterial, acid-fast bacilli, and fungal organisms) was also performed on paraffin-embedded tissue at a reference laboratory.

Results: Histologic sections revealed dense lymphoplasmacytic inflammation composed of plasma cells, lymphocytes, and histiocytes, with variable fibrosis. Scattered foci of necrosis were present, with adjacent obliteration of anterior pituitary architecture. Immunohistochemistry demonstrated polytypic plasma cells; IgG4 and IgG did not suggest IgG4-related disease. Special stains showed no diagnostic microorganisms, while evaluation for toxoplasma, syphilis, and viral etiologies was negative. No evidence of pituitary neuroendocrine tumor, craniopharyngioma, hematolymphoid neoplasm was observed. Broad-range fungal PCR with reflex sequencing identified Naganishia globosa with 100% sequence match. Although broad-range PCR can be prone to contamination, this organism is not considered a common laboratory contaminant. In the absence of an alternative etiology, CNS-dosed fluconazole therapy was initiated.

Conclusions: This case highlights a destructive lymphoplasmacytic hypophysitis with molecular identification of an exceedingly rare fungal species and underscores the interpretive challenges associated with molecular microbiologic assays in neuropathology. Careful integration of histopathologic, molecular, and clinical data is essential when rare organisms are detected without morphologic confirmation.

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A Rare and Deadly Pathogen: Diagnostic Challenge of Cladophialophora bantiana Brain Infection in an Immunocompetent Patient

A Pallante, S Mandavilli, P Patel; Hartford Hospital

Background: Cladophialophora bantiana is a rare neurotropic dematiaceous fungus associated with mortality rates up to 70%. It commonly presents as brain abscesses that radiographically mimic intracranial neoplasms, often leading to delayed diagnosis. We present a case of a 69-year-old immunocompetent male with progressive left-sided weakness found to have a right frontoparietal mass initially suspected to be malignant.

Methods: A 69-year-old male presenting with progressive left-sided weakness underwent neuroimaging, which revealed a right frontoparietal mass with extensive vasogenic edema and midline shift. Due to strong radiographic suspicion of a neoplastic process, the patient underwent surgical resection of the lesion. Histopathologic examination of the resected tissue was performed, including special stains for fungal elements. Additional brain biopsies were obtained and analyzed using fungal polymerase chain reaction (PCR) for definitive pathogen identification. Systemic imaging was conducted to evaluate for extracranial fungal involvement.

Results: Histopathologic analysis demonstrated abundant septate hyphae consistent with a dematiaceous fungal infection. Special stains and fungal PCR confirmed the presence of Cladophialophora bantiana. Despite surgical resection, the patient’s clinical condition deteriorated, with worsening cerebral edema necessitating repeat craniotomy for abscess evacuation and placement of an Ommaya reservoir. Systemic imaging did not reveal any extracranial fungal foci.

Conclusions: Cladophialophora bantiana infection can closely mimic intracranial neoplasms both clinically and radiographically, leading to delayed diagnosis and poor outcomes. This case shows the importance of maintaining a broad differential diagnosis when evaluating intracranial masses, even in the absence of systemic illness or extracranial findings. Early consideration of fungal pathogens may facilitate timely diagnosis and management in similar cases.

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Multiple Disseminated Opportunistic Infections as Initial HIV/AIDS Presentation in an Ecuadorian Immigrant

T Klug, U Alappan, J Magid-Bernstein, J Vinetz, A Huttner; Yale School of Medicine

Background: A 27-year-old man from Ecuador presented with vomiting, epigastric pain, fever and weight loss. He was found to have advanced AIDS with a CD4 T-cell count of 8 and HIV viral load of 504,000 IU/ml. Additionally, pulmonary histoplasmosis was diagnosed via culture of bronchoalveolar lavage fluid, and histopathologic analysis of transbronchial biopsy demonstrating small yeast forms with narrow-based budding staining strongly for GMS and weakly with PAS-fungus. CMV revealed a positive CMV PCR (2900 IU/mL), raising concern for CMV colitis with associated appendicitis. Twenty-one days into his hospitalization he became altered with confusion and mild expressive aphasia. CT and MRI showed rim-enhancing peripherally diffusion restricting supratentorial and infratentorial lesions with vasogenic edema and mass effect, leptomeningeal enhancement associated with several of the lesions, and stable midline shift and mass effect on the 4th ventricle.

Methods: Biopsy of a large right frontal brain lesion was performed.

Results: Histopathologic examination revealed cyst forms that stained positive with toxoplasma immunostaining and DNA PCR of the tissue was positive for toxoplasma. Few cells demonstrated positive staining for EBV alongside positive EBV PCR. Extensive inflammation was present with dense perivascular and intraparenchymal inflammatory infiltrate composed of a mixture of CD163 positive macrophages, CD3 positive T-lymphocytes, and CD20 positive B-lymphocytes. Scattered oligodendroglial cells with slightly enlarged, ‘glassy’ appearing nuclei with positive immunoreactivity for SV40 and p53 were present. GFAP demonstrated bizarre giant astrocytes. Alongside a JC virus index of 0.21, these findings support the diagnosis of progressive multifocal leukoencephalopathy (PML) due to reactivation of JC virus. Histoplasma was not identified in the brain parenchyma, as GMS staining was negative.

Conclusions: This case demonstrates the complexity of histopathologic diagnosis of opportunistic infections in the setting of advanced HIV/AIDS, as many opportunistic infections may coexist and variably contribute to patient presentation.

Posters: Tumors: Nonglial

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A Tale of Two Lesions: Synchronous Schwannoma and Glioneuronal Hamartoma

P Loreto-Palacio, M Cohen, M Couce; University Hospitals Cleveland Medical Center/Case Western Reserve University

Background: Glioneuronal hamartomas are rare lesions, whereas schwannomas are the most common tumors of the cerebellopontine angle (CPA) and internal auditory canal (IAC). Although glioneuronal hamartomas have been described, to our knowledge, this represents the first reported case of a synchronous schwannoma and glioneuronal hamartoma.

Methods: We present a case of synchronous schwannoma and glioneuronal hamartoma involving the right CPA and IAC. Histopathologic assessment was performed using hematoxylin and eosin (H&E) staining and immunohistochemistry.

Results: A 54-year-old female presented with decreased right-sided hearing and mild facial asymmetry. MRI revealed an enhancing 17 × 7 × 7 mm mass extending from the right IAC into the CPA; the contralateral CPA was unremarkable. The patient underwent a translabyrinthine craniotomy for tumor resection. Histology demonstrated multiple fragments of schwannoma and a single fragment composed of an admixture of peripheral nerve and disorganized non-neoplastic glial tissue.

Conclusions: This case represents, to our knowledge, the first report of a synchronous schwannoma and glioneuronal hamartoma in the CPA/IAC region, expanding the spectrum of lesions that may coexist in this location.

Posters: Peripheral Nerve/Muscle

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Severe Rhabdomyolysis in Inflammatory Myopathy with Abundant Macrophages
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Background: Inflammatory myopathy with abundant macrophages (IMAM) is a rare subtype of inflammatory myopathy characterized by diffuse macrophage infiltration within muscle and fascia, accompanied by fiber damage. The underlying pathophysiology remains poorly understood. We present a rare case of IMAM to contribute to the limited literature and highlight its clinical and pathological features.

Methods: Clinical, laboratory, and histopathological evaluation of a single patient presenting with acute systemic symptoms and severe rhabdomyolysis.

Results: A 24-year-old male with a history of congenital hydrocephalus and Rathke's cyst presented with nausea, abdominal cramping, and headache following a dental procedure without general anesthesia. Initial labs revealed severe metabolic acidosis (pH 7.28), elevated lactate (8.1 mmol/L), acute kidney injury (creatinine 3.64 mg/dL), leukocytosis (29.8 ×10^3/μL), and markedly elevated CK (>40,000 U/L). Muscle biopsy demonstrated prominent inflammation with interfascicular distribution involving endomysial, perimysial, and epimysial regions. Immunohistochemistry revealed numerous CD68+ macrophages, rare CD3+ T lymphocytes, and minimal CD20+ B cells. No granulomas, ragged red fibers, rimmed vacuoles, or amyloid deposits were identified. Fiber type distribution was preserved, and P62 staining highlighted inflammatory infiltrates without intrafiber aggregates.

Conclusions: IMAM represents a distinct inflammatory myopathy subtype with dermatomyositis-like clinical features, but unique histopathology dominated by macrophage infiltration and myonecrosis. Recognition of this entity is critical for accurate diagnosis and differentiation from dermatomyositis and other inflammatory myopathies. This case adds to the existing literature on IMAM, emphasizing its rarity and the need for further studies to better define its clinical spectrum and underlying mechanisms.

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Diagnostic Yield of Significant Neuromuscular Pathologies in Hospital Autopsy

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Background: Most hospital autopsies collect muscle tissue for histologic evaluation; however, the diagnostic yield may be perceived to be small given the rarity of fatal neuromuscular disorders. We examine the diagnostic yield of a prospective sample of autopsies at a large academic institution that is not enriched for neuromuscular disorders. We specifically endeavor to elucidate the most common entities found at autopsy in patients without known neuromuscular disease.

Methods: Adult autopsy cases were prospectively sent for neuromuscular consultation between September 2024 and December 2025. Results were recorded in the autopsy reports. All adult cases who consented to an unrestricted autopsy were included; fetal and pediatric cases, and cases that did not consent to abdomen or chest sampling, were not included.

Results: Seventy-five cases met inclusion criteria; there were 39 males and 36 females (male to female, 1.1), with an average age of 65 years. All patients were sampled at the psoas muscle. No patients had neuromuscular symptoms at the time of death. No specimens demonstrated changes grossly. On histology, 42 specimens (56.0%) had marked alterations in their skeletal muscle: 40 cases (53.0%) of type II fiber atrophy, 6 cases (8.0%) of neurogenic change, and 1 case (1.3%) of metastatic calcification. 6 cases (8.0%) had more than one pathology. Additional analysis on length of hospitalization and cause of death was also undertaken.

Conclusions: In this series, routine muscle specimens taken for hospital autopsies demonstrated frequent pathologic findings and suggest that key diagnoses, particularly type II fiber atrophy, are more common in this population than in targeted surgical biopsies.

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Genetic and Pathologic Diagnosis of Fatal Duchenne Muscular Dystrophy-Associated Dilated Cardiomyopathy at Autopsy

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Background: Duchenne muscular dystrophy (DMD) is a severe X-linked neuromuscular disorder characterized by progressive skeletal and cardiac muscle degeneration beginning in early childhood. It is caused by loss-of-function mutations in the DMD gene, resulting in absent or truncated dystrophin, a protein essential for maintaining sarcolemmal stability. Affected myofibers rupture easily; accumulating muscle injuries cause patients to develop debilitating systemic motor weakness. Progressive cardiomyopathy and respiratory muscle weakness are major contributors to morbidity and mortality.

Methods: N/A

Results: We present the postmortem diagnosis of DMD in a young child who suffered cardiopulmonary arrest while under general anesthesia for an elective procedure and could not be resuscitated. Ante-mortem history was limited to asthma and autism, with no known neuromuscular diagnosis. External examination demonstrated bilateral calf pseudohypertrophy. Internal examination revealed a globoid heart with ventricular dilation and diffuse fibrotic changes involving the left ventricle and interventricular septum. Histologic evaluation of myocardium demonstrated hypertrophic myocytes, interstitial and replacement-type fibrosis, granulation tissue, and myonecrosis. Examination of the diaphragm showed marked fiber size variability with hypertrophic and atrophic fibers and foci of myonecrosis, consistent with dystrophinopathy. Postmortem molecular testing identified a previously unreported nonsense mutation in the DMD gene, c.1738_1744del (p.Asp580*).

Conclusions: This case underscores the diagnostic value of systematic neuromuscular and cardiac evaluation at autopsy in sudden pediatric deaths. Approximately one-third of DMD cases are sporadic, and the disease progresses insidiously in early childhood. These factors contribute to the problem of diagnostic delay in DMD patients, with the mean age of clinical diagnosis approaching five years. Early detection through newborn screening grants DMD patients access to early and lifelong interventions to address the disease. The preventability of this death supports the inclusion of DMD on the U.S. Recommended Uniform Screening Panel and highlights the critical role of autopsy in recognizing dystrophinopathies postmortem.

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HIV-associated inflammatory myopathy with vacuolar change and diffuse nemaline pathology: a case report

M Sant [1], R Bell [2], O Lopes Abath Neto [1]; [1] University of Iowa, [2] Indiana University

Background: HIV infection is associated with a broad spectrum of skeletal muscle pathology, including inflammatory myopathy and sporadic late-onset nemaline myopathy (SLONM). These entities are usually described separately due to the uncommon overlap between inflammatory myositis and nemaline pathology. We report a case of HIV-associated myopathy demonstrating combined inflammatory, vacuolar, and diffuse nemaline features at the time of initial HIV diagnosis.

Methods: Clinical and laboratory data were reviewed, and a quadriceps muscle biopsy was evaluated using routine histology, histochemistry, immunofluorescence, and electron microscopy.

Results: A 40-year-old female presented with chronic neck weakness, progressive lower extremity weakness, facial droop, and weight loss. Brain imaging showed no acute intracranial pathology. MRI of the thighs demonstrated symmetric patchy STIR hyperintensity and enhancement consistent with myositis. Laboratory evaluation revealed a new diagnosis of HIV infection. Muscle biopsy showed marked myofiber size variability with hypertrophic and atrophic fibers, chronic perivascular perimysial inflammation, necrotic fibers invaded by macrophages, and numerous basophilic regenerating fibers. Scattered fibers exhibited vacuolar change, some rimmed on trichrome, with moth-eaten appearance on oxidative stains. Immunofluorescence revealed multifocal myofiber MHC class I upregulation, focal capillary C5b-9 deposition, and numerous embryonic myosin heavy chain-positive regenerating fibers. Electron microscopy demonstrated numerous nemaline bodies diffusely distributed within myofibers.

Conclusions: This case demonstrates an HIV-associated myopathy with overlapping features of inflammatory myositis, vacuolar change resembling inclusion body myositis, and diffuse nemaline pathology. HIV-associated SLONM is well-recognized but typically lacks significant inflammation. On the other hand, acute or subacute HIV myositis does not usually demonstrate prominent nemaline pathology. This combined pattern expands the morphologic spectrum of HIV-related muscle disease and highlights the diagnostic value of electron microscopy. Recognition of this overlap is important for disease classification and management, as HIV-associated myopathy may precede HIV diagnosis and mimic idiopathic inflammatory or degenerative myopathies.

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Gemcitabine-related acute ischemic myonecrosis: a case report

M Sant [1], R Bell [2], O Lopes Abath Neto [1]; [1] University of Iowa, [2] Indiana University

Background: Acute ischemic myonecrosis is an uncommon pattern of muscle injury. Chemotherapy-associated muscle injury is commonly attributed to direct myotoxicity or immune-mediated mechanisms. Gemcitabine is a nucleoside analog used in treatment of carcinomas and has recognized vascular toxicities, including thrombotic microangiopathy and hypercoagulability. We report a case of acute ischemic myonecrosis temporally associated with gemcitabine-based chemotherapy for pancreatic adenocarcinoma.

Methods: Clinical and laboratory data were reviewed, and a quadriceps muscle biopsy was evaluated using histology, histochemistry, and immunofluorescence.

Results: A 62-year-old female with metastatic pancreatic adenocarcinoma receiving a gemcitabine-based regimen developed bilateral lower extremity pain, swelling, and weakness within days of infusion. Symptoms improved, then recurred after subsequent infusion, accompanied by creatine kinase elevations of 600 to 1,195 U/L. Biopsy revealed large regional areas of necrosis involving entire fascicles with relative perifascicular preservation surrounding medium-sized vessels. Necrotic fibers were uniform in stage with rare regeneration. Macrophages were present in perimysial connective tissue without significant invasion of necrotic fibers. Oxidative and metabolic enzyme activities were lost in necrotic regions, whereas perifascicular fibers retained normal staining. MHC class I expression was confined to perimysium without myofiber upregulation. C5b-9 highlighted perifascicular capillaries and showed strong deposition in necrotic fibers.

Conclusions: Prior reports of gemcitabine-associated myopathy have variably emphasized inflammatory causes, whereas case reports with detailed histopathologic evaluation support a vascular mechanism, as demonstrated in this report. Recognition of this pattern is critical for neuropathologists, as gemcitabine-related vascular muscle injury may clinically mimic inflammatory myositis but requires different management.

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Diagnostic yield of electron microscopy in muscle biopsies evaluated for mitochondrial pathology

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Background: While electron microscopy (EM) is frequently employed in the evaluation of muscle biopsies for suspected mitochondrial disease, its diagnostic yield relative to histochemistry and its age-dependent contribution are not well established. In this project, we assessed the utility of muscle biopsy EM in a single-institution cohort.

Methods: Sequential muscle biopsies with ultrastructural evaluation at the University of Iowa from 2015–2024 were retrospectively reviewed. Cases were classified by clinical suspicion for mitochondrial disease and by the presence of mitochondrial abnormalities. Histochemical findings were ragged-red fibers or cytochrome c oxidase-negative fibers in excess of that expected for the age, and ultrastructural abnormalities included increased mitochondrial number or size, abnormal morphology of cristae, and presence of paracrystalline or other inclusions.

Results: A total of 425 muscle biopsies with EM were identified. Fifty-two cases were clinically suspicious for mitochondrial disease, of which 38/52 (73.1%) were confirmed by pathology in the biopsy. Among confirmed cases, 61.8% demonstrated exclusively ultrastructural mitochondrial pathology, indicating limited sensitivity of histochemical studies. Of confirmed cases in patients ≤18 years, 83.3% demonstrated EM-only findings, in contrast to 25% for patients >60 years. Diagnostic mitochondrial pathology was supported by ≥2 concurrent ultrastructural abnormalities in all EM-only confirmed cases, with enlarged mitochondria in 20/21 (95.2%) and abnormal cristae morphology in 19/21 (90.5%). Paracrystalline or other inclusions were identified in 7/38 (18.4%) of all confirmed cases, but only in 1/21 (4.8%) of EM-only confirmed cases. Among 373 cases without clinical suspicion for mitochondrial disease, diagnostic mitochondrial pathology was identified in 39 (11.5%), of which findings were EM-only in 27 (69.2%).

Conclusions: EM demonstrates a high diagnostic yield in muscle biopsies performed for suspected mitochondrial disease, particularly in younger patients in whom histochemistry is often nondiagnostic. Recognition of ultrastructural mitochondrial pathology is important for diagnosis, especially when supported by clinical correlation.

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MT-TK m.8363G>A Pathogenic Variant in an Adolescent Male Patient with Myoclonus Epilepsy with Ragged Red Fibers (MERRF)

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Background: Myoclonus epilepsy with ragged red fibers (MERRF) is a degenerative, multisystem, inherited mitochondrial disorder. The canonical features of MERRF include myoclonus, epilepsy, ataxia, and skeletal muscle biopsy demonstrating ragged red fibers; however, presentation is heterogeneous, and some patients may demonstrate only muscle weakness and/or exercise intolerance.

Methods: Clinical evaluation of a 15-year-old male by pediatric rheumatology revealed progressive proximal lower extremity fatigue and weakness, unintentional weight loss, and episodes of shortness of breath, tachycardia, and lightheadedness. His physical exam demonstrated limited range of motion, muscle atrophy, and concern for contractures. CK was mildly elevated (268 U/L). Magnetic resonance imaging was suspicious for myositis of the bilateral thighs. A biopsy of a left thigh muscle was obtained.

Results: Light microscopy showed wide variation in fiber size, with fiber diameters ranging from < 10 up to 100 microns. There were widely scattered necrotic and regenerating fibers. More than half of muscle fibers were cytochrome C oxidase (COX) negative. Scattered fibers had a ragged-red appearance. Ultrastructural analysis showed increased numbers of neutral lipid droplets and numerous mitochondria clustered between sarcomeres and in the subsarcolemmal space. Mitochondrial ultrastructural findings included abnormally large size, multilayered/concentric cristae, and paracrystalline inclusions.

Conclusions: COX-negative fibers, increased neutral lipid, and abnormal mitochondrial ultrastructure together suggest a primary mitochondrial myopathy, a complex class of disorders characterized by defects in oxidative phosphorylation, not all of which follow a maternal inheritance pattern. Genetic testing demonstrated a pathogenic variant in MT-TK (m.8363G>A), the mitochondrial DNA-encoded tRNA for lysine. The m.8363G>A MT-TK variant is a rare cause of MERRF, accounting for < 10% of cases. Both the noncanonical clinical presentation and pathogenic variant illustrate the clinical and genetic heterogeneity of MERRF and underscore the importance of ancillary testing – including molecular genetic testing and muscle biopsy with ultrastructural analysis – to arrive at a final diagnosis.

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MHC-II staining pattern in two cases of immune checkpoint inhibitor-associated myositis with classic histopathological findings.

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Background: Immune checkpoint inhibitors (ICI) have revolutionized cancer therapy with their ability to augment the immune response against tumor cells by blocking the inhibitory programmed cell death protein-1 (PD-1) or cytotoxic T lymphocyte-associated antigen 4 (CTLA-4) signaling pathways. However, they are also associated with a spectrum of immune-related adverse events, including neuromuscular complications such as myasthenia gravis (MG), ICI-associated myositis, and inflammatory neuropathy.

Methods: We studied two cases of ICI-associated myositis with extensive histopathologic workup, including ultrastructural evaluation.

Results: Patient 1 is a 75-year-old male with diabetes mellitus and metastatic urothelial carcinoma treated with transurethral resection and chemotherapy, enfortumab vedotin, and pembrolizumab, who presented with diplopia, ptosis, and shortness of breath concerning for pembrolizumab-associated MG, myocarditis, and myositis one month after treatment. Patient 2 is a 77-year-old male with a history of esophageal adenocarcinoma treated with robotic esophagectomy and nivolumab, who developed shortness of breath and generalized weakness 1 month after treatment. His pulmonary function testing suggested diaphragmatic weakness concerning for nivolumab-induced myopathy/MG versus paraneoplastic syndrome. In both cases, vastus lateralis biopsies showed the classic histopathologic features of ICI-associated myositis: multifocal chronic inflammatory infiltrates (predominantly macrophages and T cells, with occasional B cells) that were associated with clusters of degenerating/regenerating muscle fibers and sarcolemmal MHC-I and MHC-II upregulation. Interestingly, in patient 1, there were also frequent fibers with small sarcolemmal complement (C5b-9) deposits, raising the possibility that complement deposition in the NMJs is responsible for his MG-like symptoms. However, electron microscopy showed only a single NMJ with no ultrastructural abnormalities in patient 1; no NMJs were available for ultrastructural evaluation in patient 2.

Conclusions: With increasing usage of ICI, it is important to recognize the histopathologic features of ICI-associated myositis. Here, we demonstrate that MHC-II upregulation is a feature of this unique inflammatory myopathy.

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Muscle neurogenic features in idiopathic inflammatory myopathies

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Background: Idiopathic inflammatory myopathies (IIMs) with positive myositis-specific antibodies (MSA+) cause skeletal muscle damage and frequently involve extramuscular organs. 'Neuro-myositis' is rarely reported in MSA+ IIMs including antisynthetase syndrome (ASS) and dermatomyositis (DM), while neuropathy is commonly seen in patients with inclusion body myositis (IBM). Whether IIMs affect peripheral nerves is largely unknown. This study aims to investigate muscle neurogenic changes in MSA+ IIMs.

Methods: We examined muscle biopsies from patients diagnosed with IIMs according to the clinico-sero-morphological classification. Studied groups included: 18 ASS patients (aged 28-77 years, median: 55), 17 DM patients (aged 18-69, median: 42), and 10 patients with immune-mediated necrotizing myopathy (IMNM; aged 55-80, median: 71), compared to 11 IBM patients (aged 52-82, median: 68) and previously reported 58 patients with mitochondrial myopathy (MM; aged 3-78, median: 52 [PMID: 31100146]). Muscle histopathology was systematically assessed and correlated with clinical findings.

Results: Muscle fiber denervation atrophy (MFDA) was found in 100% of patients with ASS or IMNM, and in 88 % of DM patients; small fiber type grouping (FTG) was observed in 61%, 50%, and 24% of ASS, IMNM, and DM patients, respectively. These neurogenic features were at least partially age-related in patients older than 50 years, particularly IMNM patients, as they were similarly identified in IBM patients. Further analysis of 7 ASS and 9 DM patients younger than 50 years revealed statistically significant differences in MFDA between groups of ASS (7/7) and MM (13/27) patients; in FTG between groups of ASS (5/7) and MM (4/27) or DM (1/9) patients. ASS clinical manifestations included symptomatic neuropathy and electrodiagnostic findings of neurogenic changes in 5 patients including 3 younger patients.

Conclusions: Neurogenic features are common in IIMs, particularly ASS, and possibly share pathogenic mechanisms with some IIM extramuscular manifestations. Neuropathy and neurogenic changes may be a component of ASS.

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Neuromuscular Choristoma: A Rare Finding

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Background: Neuromuscular choristoma (NMC) is a benign, developmental peripheral nerve tumor comprised of a mixture of muscle fibers and peripheral nerves. NMC is commonly seen in pediatric patients and typically presents with neuropathic pain and sensory deficits.

Methods: We describe clinical, histologic, and molecular findings in a case of NMC.

Results: A 22-year-old woman presented with chronic right shoulder pain, right arm weakness, and numbness in both hands for 10 years. At age 8, she developed a right scapular abnormality, and imaging of the right shoulder identified possible plexiform neurofibromas. Genetic testing for neurofibromatosis (NF) was negative, but she continued to have a presumed diagnosis of NF. MRI in 2024 showed enlarged brachial plexus nerve roots at C6, C7, and C8 with no evidence of nerve sheath tumors. She underwent biopsy of the right C7 nerve root, which was noted to be approximately 3 times normal caliber. Histologic evaluation revealed a lesion comprised of skeletal muscle admixed with nerve fibers and cellular elements. Immunohistochemistry for neurofilament highlighted numerous nerve processes admixed with myocytes. The cellular elements were immunoreactive for S100 and SOX10. Beta-catenin showed nuclear labeling of rare cells within the admixed nerve tissue, and LEF1 highlighted rare nuclei, both suggestive of mutation. Plastic embedded sections with toluidine blue showed similar findings of myocytes intermixed with irregular bundles of angulated myelinated fibers. NGS sequencing detected a CTNNB1 p.Ser45Phe variant. The findings were consistent with a diagnosis of neuromuscular choristoma. A year after diagnosis, she continues to have right shoulder pain with no clinical evidence of transformation to desmoid fibromatosis. Whole exome sequencing did not reveal any pathogenic variants.

Conclusions: Neuromuscular choristoma (NMC) is a rare developmental peripheral nerve lesion with frequent mutations in CTNNB1. While considered benign, NMC has been associated with desmoid fibromatosis, highlighting the importance of diagnosis to guide clinical management.

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Targeting IL-17-Mediated Immune–Neural Crosstalk in Skin to Alleviate Diabetic Neuropathy

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Background: Diabetic neuropathy (DN) is a debilitating condition with no effective treatment due to the lack of actionable therapeutic targets. Our transcriptomics data revealed the proinflammatory cytokine interleukin-17 (IL-17) as the top altered signaling pathway in the skin of DN mice. This study aimed to elucidate the mechanisms by which IL-17 drives DN and determine the therapeutic efficacy of IL-17 blockades, which have been successfully used for treatment of psoriasis.

Methods: DN mouse models including high-fat diet (HFD) only and HFD followed by low-dose streptozotocin were assessed by behavioral assays, flow cytometry, and morphologic analyses. IL-17 expression was measured by qPCR and ELISA. scRNA-seq and spatial transcriptomics were used to assess the IL-17 receptor (IL-17R) expression in mice and humans, respectively.

Results: IL-17A and IL-17F exhibited species-specific upregulation in the glabrous skin of DN mice and human patients, respectively. IL-17 was mainly produced by skin γδ T cells, which were activated by dendritic cells recruited to injured skin nerves via damage-associated molecular pattern (DAMP) S100A8/A9. Treatment of DN mice with the S100A8/A9 inhibitor paquinimod reduced skin inflammation and reversed DN. IL-17R isoform expression in DRG neurons also differed by species: in mice, small-diameter peptidergic nociceptors (PEPs) primarily expressed IL-17RA, whereas in humans, multiple DRG neuronal subtypes predominantly expressed IL-17RC. The DRG neuronal IL-17R expression was upregulated in both DN mice and patients. Of note, knockout (KO) of PEP-specific Il17ra alleviated DN pain by reducing the excitability in small-diameter DRG neurons. Furthermore, DN phenotypes were successfully rescued by KO of Il17a or systemic IL-17A monoclonal antibody (mAb) blockade in mouse models.

Conclusions: Our data suggest that DN pain and pathology may result from IL-17-associated skin inflammation and the subsequent sensitization of DRG neurons. Our findings highlight the potential of repurposing the FDA-approved IL-17-targeting therapeutics as a novel treatment for DN.

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Granular structures in the human olfactory nerve across ages

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Background: Neuropathologic studies of human brain tissue are often limited by the availability of appropriate control specimens. Distinct granular structures were observed in the human olfactory nerve in the absence of corresponding pathologic changes in the cerebrum or cerebellum during routine autopsy, raising the question of whether they represent normal structures or postmortem changes.

Methods: Olfactory bulbs and tracts were obtained from 24 human autopsy cases ranging from 22 to 93 years of age, with postmortem interval (PMI) times between 7 and 81 hours.

Results: Small spherical granules were consistently observed in both olfactory bulbs and tracts on hematoxylin and eosin staining, with greater density in the tracts, particularly within loosely organized regions. The granules demonstrated weak blue staining with hematoxylin and eosin and Congo red, distinct blue staining with Luxol fast blue, and purple staining with periodic acid–Schiff. They were not localized within neuronal cell bodies or axons and appeared either singly or in clusters without membranous encapsulation. Quantitative analysis demonstrated smaller granule size, lower density, and reduced staining intensity with shorter PMI. High-magnification examination revealed that the granules were freely dispersed between cells and axons, indicating that these structures do not represent normal components of olfactory nerve anatomy. To assess the potential contribution of postmortem change, olfactory regions from C57BL mice of various ages (3 months, 5 months, 10 months, and 2 years) were examined at PMIs of 1 and 24 hours; no comparable granular structures were identified.

Conclusions: Although absent in murine tissue, species-specific differences in olfactory anatomy and protein-coding gene expression limit exclusion of intrinsic human specificity. These findings, observed across all cases, suggest that the granular structures may represent postmortem-associated changes and underscore the need for cautious interpretation of olfactory nerve findings in autopsy-based neuropathologic studies. Ongoing studies aim to further define their biochemical composition and mechanism of formation.

Posters: Neurodegenerative (FTLD/Lewy Body/Parkinson/Other)

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Neuropathology in a Diverse Cohort of Oldest Old: 2026 update on the LifeAfter90 Study

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Background: Examining the neuropathology of the oldest-old has advanced our understanding of multiple etiologies in very late life. Most studies have included exclusively White decedents; there is a dearth of knowledge on persons who come from heterogeneous demographic backgrounds. Our goal was to characterize neuropathology in a cohort of ethnically and racially diverse oldest-old decedents.

Methods: The LifeAfter90 study is an ongoing cohort study of Kaiser Permanente Northern California members of persons aged 90 and above with targeted recruitment of individuals across different demographics with no prior diagnosis of dementia in their medical record. Participants were evaluated approximately every six months during their life. Brain donation was available to all consenting participants. Neuropathology was assessed using National Alzheimer's Coordinating Center Neuropathology forms and NIA-AA guidelines for diagnoses.

Results: As of February 2026, 148 persons enrolled in autopsied had died and neuropathology forms were completed . The median age of death was 96 years (range 90-107) and 91 (61%) were women. Self-identified race and ethnicity was 24 (16%) Asian, 16 (11%) Black, 27 (18%) Latino, 10 (7%) Multiracial/Other, and 70 (47%) White. At the final clinical exam, 52 participants had dementia (35%), 34 had cognitive impairment but were not demented (23%), and 62 were cognitively normal (42%). Twenty-seven participants were not ADNC (18%) and High ADNC was found in 14 participants (9.5%). TDP-43 deposits were found in 41 participants (29%), with the highest frequencies among those having a final clinical diagnosis of dementia (41%), with 19% having cognitive impairment but were not demented, and 23% having normal cognition . Forty-seven participants had Lewy bodies (32%); five participants had hippocampal sclerosis (3.5%).

Conclusions: Many participants maintained normal cognition indicating neuropathologic burden may be present in the absence of overt cognitive impairment, especially in the oldest old.

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Differential Neuropathology in Female versus Male Brain Donors with Chronic Traumatic Brain Injury

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Background: Traumatic brain injury (TBI) is a growing public health concern and a risk factor for later development of neurodegenerative disease. How sex might influence the pattern of exposure, TBI-associated pathology, neuroinflammatory response and neurodegenerative disease is unknown; characterizing sex-specific exposures and neuropathologic outcomes may improve risk assessment, prevention strategies, and therapeutic approaches.

Methods: Brains were obtained from the Pacific Northwest Brain Donor Network and the Mount Sinai Brain bank. TBI history and exposure metrics were collected through structured family questionnaires and medical record review. Age-matched male (n=10 per group) and female (n=7 per group) cohorts, < 70 years of age, were categorized into control, mild or severe TBI groups. Mild TBI was defined as head injury with associated symptoms such as headaches, slower processing, and memory loss. Severe TBI was defined as the presence of a head injury with coma or skull fractures/contusions. All cases underwent detailed neuropathologic evaluation for TBI-associated pathology and neurodegenerative disease according to consensus criteria, including assessment for chronic traumatic encephalopathy (CTE).

Results: TBI exposures occurred through diverse mechanisms, including falls, motor vehicle collisions, assault and sports-related injuries. Repeated injury was more frequently documented in females across both mild and severe TBI groups compared to males, with intimate partner violence as the most common source. Neuropathologic assessment identified CTE and Lewy-body pathology exclusively in the male TBI cohorts. Interestingly, the female mild TBI group exhibited higher rates of hippocampal gliosis compared to the male mild TBI group, while contusions were more frequent in the severe male TBI group compared to severe female TBI group.

Conclusions: This cohort demonstrates sex-related differences in TBI exposure and neuropathological outcomes that need to be investigated further in larger cohorts, raising important questions for optimal prevention strategies and therapeutic approaches.

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Rodeo as a Risk Factor for Chronic Traumatic Encephalopathy, Alzheimer's Disease and Other Related Dementias.

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Background: Traumatic brain injuries (TBI) are associated with the pathogenesis of various neurodegenerative diseases, including Chronic Traumatic Encephalopathy (CTE). TBI exposure linked to CTE pathology is primarily repetitive head impacts (RHI) sustained in contact sports, including football, boxing, and ice hockey. Limited CTE research has been conducted on rodeo athletes, despite high rates of TBI, low rates of protective headgear usage, and exposure to significant impact forces.

Methods: Cases in the Biggs Institute Brain Bank were queried for participation in rodeo activities, identifying three cases. Two additional former contact sport athletes were included as a comparison group. Neurology and neuropsychology record data were extracted from Epic electronic health records. Postmortem evaluation was conducted using neurodegenerative disease standards, including immunohistochemistry against hyperphosphorylated tau (p-tau), amyloid-β, TDP-43, and α-synuclein pathologies.

Results: In this small rodeo case series, two had CTE pathognomonic lesions, while all three had other ADRD pathologies, including Alzheimer's Disease, Limbic-predominant Age-related TDP-43 Encephalopathy, and Lewy Body Disease. All cases exhibited profound cognitive decline with disease progression. Behavioral changes across cases included the development of major depressive disorder, increased agitation and aggression, and violent outbursts. One case is consistent with traumatic encephalopathy syndrome, with concurrent CTE neuropathology.

Conclusions: Findings from this case series demonstrate a range of neurodegenerative disease pathologies associated with antecedent TBI exposure in rodeo athletes, including CTE pathology. Given the high burden of RHI in this population, additional research is warranted to explore pathologic outcomes between rodeo events and other occupational exposome factors.

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CTE-Type Tau Filaments in LATE-NC

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Background: Neuropathologic co-morbidities of Alzheimer's Disease neuropathologic change (ADNC) are common and clinically significant; however, to date structural studies have been limited to "pure" ADNC. To determine the impact of co-morbidities on tau structure, we performed cryo-electron microscopy (cryo-EM) on four patients with the most common neuropathologic co-morbidity of ADNC: limbic-predominant age-related TDP-43 encephalopathy neuropathologic change (LATE-NC). LATE-NC is seen in ~50% of AD and is associated with more severe tau pathology and accelerated cognitive decline.

Methods: Our cohort included 3 cases of LATE+ AD (Cases 1-3; obtained from Michigan Brain Bank), and one case of LATE + Primary age-related tauopathy (PART; Case 4; obtained from University of Kentucky). Extended cortical sampling was performed for cases 1-3. Pathology burden was quantified with digital pathology. Cryo-EM was performed on sarkosyl-insoluble fractions from either amygdala (Cases 1-3) or hippocampus (Case 4); samples were applied to glow-discharged grids, then frozen into liquid ethane. Images were acquired on a Titan Krios G4i operated at 300 kV.

Results: In addition to the expected AD-fold tau filaments, all cases contained chronic traumatic encephalopathy (CTE) fold tau, which has never before been reported in ADNC. CTE-fold tau ranged from 10% to 95% of total tau species. Neuropathologic review showed one case with a pathognomonic CTE lesion, one with suggestive features, and two without neuropathologic features of CTE. The proportion of CTE-fold tau did not correlate with neuropathologic CTE, TDP-43 pathology, or astrocytic tau pathology. None of the individuals had a history of head trauma or contact sports participation.

Conclusions: Our findings suggest that CTE-fold tau is a conserved molecular feature of LATE-NC, seen both with and without neuropathologic CTE. This raises fundamental questions about the relationship between LATE-NC and CTE, the effect of TDP-43 on tau conformation, and the etiology of LATE-NC.

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Lesion-Associated Phosphorylated Tau in Traumatic Brain Injury and Repetitive Head Impacts: A Community Autopsy Cohort Study

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Background: Phosphorylated tau (ptau) characterizes multiple neurodegenerative and aging-related conditions. Based on incidentally reported p-tau near old traumatic lesions in cases screened for Chronic Traumatic Encephalopathy (CTE) in a community-based autopsy cohort, (PMID37846159, 27815395), we sought to determine the extent of this phenomenon in the Late Effects of Traumatic Brain Injury (LETBI) cohort.

Methods: Using a comprehensive neuropathologic workup, we examined ptau deposition (AT8) colocalized with 3 types of traumatic lesions, and associations with demographics, TBI patterns, neurodegenerative diagnoses (NDD), aging-related tau astrogliopathy (ARTAG), and cerebrovascular disease (CVD).

Results: Of 36 cases (ages 3rd-10th decade, median 7th; 26 male), 30 (83%) had gross and/or microscopic chronic traumatic lesions, 11 (37%) with lesion-associated ptau. This finding was more common for contusions (10 [33 %]), than torsional lesions (3 [10%]), and surgical sites (1 [3%]); 3 cases had ptau in more than one lesion type. Lesion-associated ptau was more pronounced in isolated TBI (7/14 [50%]), compared to mixed TBI/RHI (3/10 [30%]) and RHI-only (1/12 [8%]). Overall, 28 of 36 cases (78%) demonstrated ≥1 concurrent NDD (i.e., "polypathology"). Among lesional ptau cases, 10 of 11 (91%) exhibited polypathology; comorbid diagnoses included CVD (8/11 [73%], Alzheimer disease neuropathologic-change (ADNC) (4/11 [36%]), and chronic traumatic encephalopathy (CTE; 2/11 [18%]). Of the 19 without lesion-associated ptau, all had CVD, 8 with ADNC (42%), and 4 with CTE (21%). ARTAG was noted in 82% lesional ptau cases and 63% of cases without lesional ptau.

Conclusions: Lesion-associated ptau occurred in >1/3 of community brain trauma cases. Comorbidity with CTE was less common, whereas polypathology, ARTAG, and CVD were frequent, reinforcing a polypathologic landscape in post-traumatic neurodegeneration. Our systematic study highlights the previously unrecognized occurrence of frequent ptau colocalization with old trauma and suggests significance of direct mechanical injury in the etiology of pathologic ptau deposition.

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Bridging Central and Peripheral Pathology in Lewy Body Disease: Toward Personalized Biomarkers

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Background: Lewy body diseases (LBDs) are defined by neuronal accumulation of phosphorylated α-synuclein (p-syn), classically within the central nervous system (CNS). However, α-synuclein pathology is also well documented in peripheral nervous system (PNS) tissues, raising the possibility that peripheral sites may reflect CNS disease. As the CNS is largely inaccessible during life, peripheral tissues offer opportunities for biomarker development and therapeutic stratification. We hypothesize that CNS-derived molecular targets in LBD are recapitulated in the PNS in a tissue- and disease stage–dependent manner.

Methods: Twelve autopsy-confirmed LBD cases and four age-matched controls from the Johns Hopkins Autopsy Service were identified. CNS regions and multiple peripheral tissues—including skin, peripheral nerve, heart with epicardial nerves, gastrointestinal tract, and adrenal gland—will undergo multiplex immunohistochemistry for established and emerging LBD-associated targets: pS129 and pY39 α-synuclein, GPNMB, TSC2, tyrosine hydroxylase (TH), and axonal markers. Quantitative digital image analysis will assess regional burden, axonal involvement, and cell-type–specific expression patterns. Correlations with clinical parameters, including disease duration, will be explored.

Results: We anticipate measurable peripheral expression of canonical and emerging LBD biomarkers, with tissue-specific patterns of axonal involvement. We further expect that biomarker burden will correlate with disease duration and clinical phenotype, supporting biologically meaningful peripheral signatures of LBD.

Conclusions: Recent evidence demonstrating reliable detection of p-syn in skin biopsies underscores the translational potential of peripheral biomarkers in LBD. This study extends that paradigm by systematically evaluating both established and emerging CNS therapeutic targets in peripheral tissues. Defining peripheral molecular signatures of LBD may enable minimally invasive biomarker assays, facilitate patient stratification for targeted therapies, and advance personalized approaches to LBD care.

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Multisite Study: Predicting Lewy Body Disease Using Skin Biopsy alpha-Synuclein Seed Amplification Assays

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Background: Skin biopsies analyzed with α-synuclein seed amplification assays (SAA) might offer a simple way to determine the presence or absence of α-synuclein aggregates in patients with Lewy body disease (LBD). To date, though, the reported diagnostic accuracy of skin biopsies for PD, whether analyzed using immunohistochemistry (IHC) or SAA, has extensively varied between labs and methods, ranging from 24% to 98%.

Methods: We had blinded SAAs done in three independent laboratories to determine the accuracy of skin biopsy SAA in predicting the clinical diagnoses of Parkinson disease (PD; n = 23), PD with dementia (PDD; n =3) and dementia with Lewy bodies (DLB; n = 6). Subjects diagnosed with PD, PDD and DLB were analyzed together as Group 1, clinically unaffected subjects as Group 2 (n = 22) and those with risk factors for LBD as Group 3 (n = 51). Skin punch biopsies were taken from the posterior neck and four biopsies were analyzed in the 3 labs in 6 separate SAA's.

Results: Pairwise agreement between biopsies, labs and assays ranged from excellent (kappa 0.82) to moderate (kappa 0.40 - 0.68). Sensitivity across assays ranged between 50.0% and 65.6% while specificity ranged between 68.1% and 100%. Comparisons of Group 1 versus Group 2 produced the greatest specificities, between 77.3% and 100%, but with a maximum sensitivity of 61.3%. Exploratory analyses indicated that sensitivity for PDD and DLB was better (9/11 subjects positive in at least one assay; 81.8%), as was Group 3 sensitivity in subjects with both hyposmia and RBD (6/6). In 17 cases that subsequently came to autopsy, 93% of SAAs were positive in those at the neocortical LBD stage as compared to only 8% at any lower stage.

Conclusions: Skin biopsy α-synuclein SAA may be useful as a diagnostic and progression biomarker in Lewy body dementia clinical trials.

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Biofluid and Clinicopathological Correlates for NSD-ISS Validation in Lewy Body Disorders: Results from the AZSAND

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Background: Historically, PD and DLB were diagnosed only by clinical signs and symptoms. The introduction of CSF alpha synuclein seed amplification assays (SAA) now allows the detection of synuclein pathology in living individuals, even before symptom onset. The Neuronal Synuclein Disease Integrated Staging System (NSD ISS) was developed to incorporate this new capability and define Lewy body diseases based on both clinical and biological evidence, the latter with assessments of CSF SAA and striatal dopaminergic integrity. However, there is still an insufficient understanding of how the NSD ISS aligns with existing neuropathological staging systems for Lewy body disorders, and how it might be affected by the frequent coexistence of AD pathology.

Methods: We utilized longitudinal clinicopathological dataset and biofluid archive from the Arizona Study of Aging and Neurodegenerative Disorders (AZSAND) to compare NSD-ISS stage with internationally accepted clinicopathological PD, DLB, and AD consensus criteria, including those of the DLB Consortium and the NIA–Alzheimer's Association. Included were CSF and plasma samples from 130 cases clinicopathologically diagnosed with PD, 76 with DLB, 50 with Alzheimer's disease with Lewy bodies (ADLB), 69 with incidental Lewy body disease (ILBD) and 111 unaffected controls.

Results: The CSF SAA had high sensitivity for PD, DLB, ADLB and ILBD (100% for PD & DLB, 92% and 94% for ADLB and ILBD) but comparatively low specificity (69%), as 34 of 111 clinicopathological control subjects tested positive despite no Lewy pathology at autopsy. Plasma p tau 217 showed a modest correlation with neurofibrillary tangle burden ($r = 0.24$, $p < 0.001$).

Conclusions: Incorporating substantia nigra pigmented neuron loss score as a postmortem marker of dopaminergic dysfunction enabled classification of most cases within the NSD ISS. However, some early stage Lewy body cases did not align cleanly with NSD-ISS, indicating the need for refinement and multicenter validation, with postmortem confirmation to optimize accuracy.

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Microglial Morphology and Reactivity Across the Intersection of Lewy Body Dementia and Chronic Traumatic Encephalopathy

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Background: While investigating participants diagnosed with chronic traumatic encephalopathy (CTE) neuropathology from the Mayo Clinic Study of Aging (MCSA), over 53% of patients with CTE pathology showed comorbid Lewy body disease (LBD). Since activated microglia may play a role in mediating dementia and LBD, the goal of this study was to characterize differential microglial activation in the midbrain within a complex multiple-etiology CTE cohort.

Methods: 13 MCSA participants with focal or multifocal perivascular tau tangles at the sulcal depths were identified, and antemortem history of traumatic brain injury, repetitive head impact (history of contact sports or military service), infarct, and hemorrhage were documented. 12 of 13 participants had a clinical history of at least one injury/pathology, and all cases showed comorbid pathology: Alzheimer's disease neuropathologic change (53.85%; 7/13), FTLD (7.69%; 1/13), PART (7.69%; 1/13), and ARTAG (69.23%; 9/13). IF for panmicroglia (IBA1), dystrophic microglia (FTL), and amyloid-responsive microglia (CD163) was performed on midbrain sections from 11 cases with concomitant CTE and LBD. Controls were balanced for age and sex and level of LBD brainstem pathology.

Results: While there was no significant change in panmicroglial counts across LBD-alone vs LBD-CTE brains, dystrophic FTL-positive microglia trended higher in brains with no LBD but with CTE pathology compared to diffuse neocortical LBD with CTE pathology. Moreover, CTE cases without LBD pathology were enriched for dystrophic microglia (75.80%) versus cases with comorbid neuropathology (49.14%), underscoring the presence of CTE alone was associated with more enriched dystrophic microglia. Amyloid-responsive microglia (CD163-positive) were rare and found primarily in perivascular spaces.

Conclusions: Within the MCSA brain bank, CTE brains showed significant comorbid pathology and were enriched for LBD, including brainstem, limbic (transitional), and diffuse neocortical subtypes. Microglial morphologies showed various reactive states across the LBD spectrum in the presence of CTE pathology, while the total number of microglia remained relatively constant.

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Microglial sensome landscapes across tauopathies

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Background: Microglia detect and respond to chemotactic cues in neurodegenerative diseases. The microglial sensome, a defined set of genes, enables microglia to sense changes in brain parenchyma. In Alzheimer's disease (AD), "homeostatic" sensome genes are downregulated and "responsive" sensome genes are upregulated. However, it is unclear how the microglial sensome is altered in other tauopathies, including chronic traumatic encephalopathy (CTE), corticobasal degeneration (CBD), and progressive supranuclear palsy (PSP). We hypothesize that the microglial sensome protein landscape in CTE, CBD, and PSP individually differ from AD.

Methods: To characterize the microglial sensome, serial sections of formalin-fixed, paraffin-embedded brain tissue from frontal neocortex of decedents with pathologically confirmed AD (n= 13), CTE (n= 14), CBD (n= 9), and PSP (n= 11) were labeled with antibodies against sensome proteins, phosphorylated tau, and phosphorylated neurofilament. Labeled tissue was digitized and analyzed using Leica Aperio AT2 Slide Scanner and Imagescope software. Total percent positive staining was averaged for grey and white matter regions. Whole slide images were processed for cell segmentation via deep-learning U-net model and analyzed with Genentech's Spatial Quantitative Pathology Features (SpatialQPFs) R package.

Results: We characterized microglial landscapes using SpatialQPFs, highlighting sensome differences that may contribute to the disease microenvironment. Higher SMI-31 levels were observed in AD compared to CTE grey matter, and lower IBA1 and P2RY12 levels in CTE compared to PSP grey matter. Microglia with lower IBA1 levels were less likely to co-localize with tau pathology in AD grey matter, while higher IBA1 levels were more likely to co-localize with tau pathology in CBD white matter. Higher P2RY12 levels were more likely to co-localize with tau pathology in CBD grey matter.

Conclusions: These findings suggest neurodegenerative pathology associated with each disease may elicit distinct microglial adaptations contributing to divergent neurodegenerative pathways. Future work should explore sensome activity in selectively vulnerable regions of each disease.

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Clinicopathological correlates of FTLD-TDP type E: a deep phenotyping on an autopsy confirmed case

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Background: Nearly half of patients with frontotemporal lobar degeneration (FTLD) have underlying TDP-43 pathology (FTLD-TDP). Recently, FTLD-TDP pathological classification was updated to include a fifth subtype (TDP-43 type E) associated with motor neuron involvement and rapid clinical progression, with few cases reported in the literature.

Methods: We describe the clinical and neuropathological findings in a patient with FTLD-TDP-E. Clinical examination comprised electromyography, 18-fluorodeoxyglucose positron emission tomography (FDG-PET), laboratory workup, cerebrospinal fluid (CSF) Alzheimer's disease (AD) biomarker analysis, testing for C9orf72, and an FTLD-targeted gene panel. A neuropsychological evaluation and brain magnetic resonance imaging (MRI) had previously been conducted at another center.

Results: A 68-year-old woman presented with a 4-year history of prominent personality change consistent with the full behavioral variant frontotemporal dementia (bvFTD) syndrome. The informant also reported progressive motor impairment. Family history of neurodegenerative or psychiatric disorders was negative. At first visit, her Mini-Mental State Examination score was 16/30. Neuropsychological evaluation showed predominant executive dysfunction. Neurological examination was notable for vertical gaze palsy, orofacial apraxia, left-dominant akinetic-rigid syndrome, freezing of gait, postural tremor, hyperreflexia of upper limbs, and atrophy of intrinsic hand muscles. Electromyographic findings demonstrated lower motor neuron involvement. Brain MRI and FDG-PET showed bilateral frontotemporal atrophy and hypometabolism. Laboratory workup, CSF AD biomarkers, and genetic testing were unremarkable. Based on the comprehensive clinical assessment, the diagnosis was bvFTD with motor neuron disease (MND). She died at age 69 from sudden cardiac arrest. Neuropathology revealed severe FTLD with type E TDP-43 inclusions exhibiting predominant diffuse neuronal deposits, scarce compact inclusions, synaptic background, abundant oligodendroglial inclusions, and early upper and lower MND.

Conclusions: This case report adds to the characterization of clinicopathological correlates of FTLD-TDP-E. Our findings can help disentangle underlying pathological disease mechanisms.

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Novel GRN Mutation in a case of FTLD-TDP: TDP-43 Mass Spectrometry and GRN mRNA Analyses

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Background: DNA mutations occurring outside the open reading frame (ORF) of GRN have been found to result in decreased mRNA expression and protein production. We present a case of FTLD-TDP associated with a novel GRN mutation (NM_002087.4:c.-8+2T>A) located outside the ORF. Aims of this study are to determine whether this mutation affects GRN expression and to characterize the biochemical profile and post-translational modifications (PTMs) of TDP-43 in the frontal cortex.

Methods: Direct sequencing of the transcribed regions of GRN was carried out using DNA extracted from brain tissue postmortem. GRN expression was quantified by qRT–PCR in brain tissue obtained from three individuals: the proband (novel mutation carrier), a Q300X GRN mutation carrier, and a control. Sarkosyl-insoluble TDP-43 was extracted from the proband's frontal cortex. The biochemical profile of TDP-43 was assessed by Western blot (WB) using pS409/410 antibody. TDP-43 PTMs were identified by mass spectrometry (MS). Immuno electron microscopy of sarkosyl-insoluble fractions using TDP-43 pS409/410, pS305, and pS375 antibodies is in progress.

Results: GRN mRNA levels were reduced in both GRN mutation carriers as compared with the levels obtained in control. WB analysis showed that the TDP-43 biochemical profile observed in association with the novel mutation was consistent with that seen in other GRN mutation carriers. MS analysis of the sarkosyl-insoluble fraction identified phosphorylated, citrullinated, and ubiquitinated residues in TDP-43, including two phosphorylated sites not previously reported in humans.

Conclusions: The reduction in GRN mRNA levels was comparable in the two cases carrying GRN mutations; thus, additional studies may determine whether protein expression associated with the novel GRN mutation differs from the expression associated to other GRN mutations. Similarly, it will be of interest to determine whether the phosphorylation sites not previously reported represent a finding specific to the current case. Supported by PHS R01NS137469.

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Perry Syndrome in the Mayo Clinic Florida Brain Bank: A novel TDP-43 Proteinopathy

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Background: Perry syndrome (PS) is an atypical parkinsonism with depression, apathy, weight loss, and central hypoventilation caused by mutations in dynactin p150glued (DCTN1). The neuropathologic features include neuronal loss in the substantia nigra and basal ganglia and TDP-43 proteinopathy.

Methods: This report describes 13 cases of PS, obtained from USA (West Virginia, Louisiana, Minnesota) and from Canada, Japan, and Poland. The material for study included glass slides on all cases, paraffin blocks on 9 cases, fixed tissue on 7 cases, and frozen tissue on 7 cases. All cases were evaluated with routine histology (H&E) and with thioflavin-S fluorescent microscopy. Cases were also immunostained for tau, neurofilament, glial fibrillary acidic protein, IBA-1 and TDP-43. Select cases were submitted for electron microscopy, with immunoelectron microscopy for TDP-43.

Results: The most characteristic neuropathologic feature of PS is degeneration in the globus pallidus and substantia nigra (pallido-nigral degeneration). With TDP-43 immunohistochemistry are aggregates in neuronal cytoplasmic inclusions (NCIs), neuronal intranuclear inclusions, dystrophic neurites (DNs), as well as axonal spheroids, oligodendroglial cytoplasmic inclusions, and perivascular astrocytic inclusions (PVIs). The latter is a distinctive feature of PS. The pattern of TDP-43 pathology does not conform to any of the currently recognized subtypes of TDP-43 pathology seen in frontotemporal dementia and in the aging brain (e.g., aging-related TDP-43 encephalopathy (LATE)).

Conclusions: PS represents a TDP-43 proteinopathy with predominant glial TDP-43 pathology and a characteristic neuroanatomic vulnerability (globus pallidus, substantia nigra and variable subthalamic). The causative gene is p150glued (DCTN1), the largest subunit of the dynactin complex, encoded by the DCTN1 gene. It acts as a critical adapter that links the motor protein cytoplasmic dynein to its cargo and to microtubules, enabling essential cellular transport. It remains uncertain failure of cellular transport leads to the selective vulnerability of TDP-43 that is dissimilar to other TDP-43 proteinopathies.

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Facial-onset sensory and motor neuronopathy (FOSMN): a case report with postmortem findings including TDP-43 pathology

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Background: Facial onset sensory and motor neuronopathy (FOSMN) is a rare, slowly progressive neurodegenerative disorder characterized by early facial sensory deficits and rostro-caudal progression to include bulbar symptoms and sensory and motor neuron symptoms affecting the limbs. Autopsy reports (seven published to date) have identified degeneration of bulbar, spinal sensory and motor neurons, often with TDP-43 neuronal and oligodendroglial inclusions, supporting the inclusion of FOSMN under the umbrella of amyotrophic lateral sclerosis.

Methods: Electronic medical record review and autopsy evaluation of the brain, spinal cord, and associated nerve roots were undertaken. Microscopic examination included hematoxylin and eosin, luxol fast blue, and Bielschowsky stains, immunohistochemistry for TDP-43, and toluidine blue stains of epon-embedded nerves.

Results: A 62-year-old male presented with facial numbness that progressed craniocaudally to include bulbar symptoms and sensory and motor deficits affecting upper more than lower extremities. He died 13 years after initial presentation due to aspiration of a food bolus. Motor neuron loss was observed in the spinal cord (predominantly cervical) and hypoglossal nuclei. Corticospinal tracts were spared. Within the dorsal columns, the cuneate fasciculi showed greater axon loss than the gracile fasciculi. The single dorsal root ganglion (DRG) available for evaluation, from the lumbosacral region, showed mild ganglion cell loss manifested by nodules of Nageotte. A small subset of neurons within the DRG, lumbar ventral horns, and hypoglossal nuclei contained TDP-43 protein aggregates, while glial TDP-43 aggregates were identified in the spinal cord, medulla, and pons, mostly in association with motor neurons.

Conclusions: The findings of bulbar and spinal sensory and motor neuron degeneration with TDP-43 inclusions and corticospinal tract sparing were congruent with the patient’s clinical presentation and consistent with a diagnosis of FOSMN.

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Locus coeruleus TDP-43 pathology in a community-based cohort: clinical and pathological correlates

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Background: The locus coeruleus (LC) is the source of norepinephrinergic innervation in the human brain. Pathology in the LC has been associated with clinical conditions such as cognitive impairment and behavioral and psychiatric symptoms of dementia (BPSD). However, phosphorylated TDP-43 (pTDP-43) pathology in the LC has been understudied, particularly in the contexts of aging and limbic predominant age-related TDP-43 encephalopathy neuropathologic change (LATE-NC).

Methods: Here, a convenience sample (n=134) of autopsied participants from the University of Kentucky Alzheimer's Disease Research Center community-based cohort was analyzed for LC pTDP-43 and phosphorylated tau (pTau) pathologies.

Results: LC pTDP-43 pathology was found in 28/134 (20.9%) of brains and was generally sparse when present. LC pTDP-43 lesions typically appeared as round granular structures (RGs) that measured ~3-12 microns in diameter and the pTDP-43 immunoreactivity did not appear to colocalize with nucleated cells. LC pTDP-43 pathology was increased with advanced aging and was strongly, but imperfectly, correlated with LATE-NC staging. To our surprise, 6.5% of individuals without observed cortical pTDP-43 pathology (i.e. LATE-NC stage 0) had LC pTDP-43 immunoreactivity. Among those with LATE-NC stage 3, i.e. widespread cortical pTDP-43 pathology, 7/12 (58.3%) of cases had detectable LC pTDP-43 pathology. There was not a statistically significant correlation between LC pTDP-43 pathology and pTau pathologic severity in the LC or in cortical regions, among participants with either primary age related tauopathy (PART) or Alzheimer's disease neuropathologic change (ADNC). In terms of clinical-pathological correlations, LC pTDP-43 pathology was nominally associated with depressive symptoms but not with global cognition or with other BPSDs. Additionally, we found the first evidence of a positive association between LC pTau pathology and LATE-NC stages.

Conclusions: In summary, LC pTDP-43 pathology was seen in approximately 20% of brains in a community-based autopsy cohort and that pathology was associated with LATE-NC, with chronological aging, and with depressive symptoms.

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Analysis of tauopathies in the NACC Data Set indicates relatively high frequency of incident PSP pathology in older persons

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Background: The epidemiology of tauopathy subtypes remains incompletely understood. Autopsy cohorts are a primary data source because clinical features don't necessarily predict pathology. However, different autopsy cohorts deploy institution-specific study designs with distinct patterns of participant recruitment. Here we studied autopsy-confirmed tauopathies while also assessing the correlative influence of participant recruitment status across multiple research centers.

Methods: We focused on four different tauopathies -- Alzheimer's disease neuropathologic change, cortical basal degeneration (CBD), Pick's disease, and progressive supranuclear palsy (PSP) -- and analyzed the NACC Neuropathology Data Set with aggregated data from NIH/NIA-sponsored Alzheimer's Disease Research Centers (ADRCs). We compared neuropathologic and clinical results of autopsied participants stratifying on their cognitive status at the time of recruitment.

Results: A total of 6994 individuals were included in the analyses, sourced from 37 different ADRCs (median number of included cases per ADRC=156.5), and participants were followed longitudinally for 4.1 yearly visits on average before autopsy. Among participants who were recruited already with dementia (n=4309) or mild cognitive impairment (MCI; n=1084), 38.8% had Braak NFT Stage VI, 2.9% CBD, 2.6% Pick's disease, and 4.6% PSP. By contrast, among those recruited while cognitively normal (n=1452), 7.6% had Braak NFT stage VI, 0.7% CBD, and 0.1% Pick's disease, whereas, remarkably, 3.3% had PSP pathology. In comparing between incident (recruited normal) and prevalent (recruited impaired) autopsy-confirmed PSP, those with incident PSP died older (89.6 years old, on average) than those recruited with dementia (76.2 years) or MCI (77.9 years). Furthermore, those with incident PSP pathology were less likely than prevalent PSP to manifest stereotypical PSP clinical features, indicating that the older age-at-onset phenotype can be clinically conflated with other dementias.

Conclusions: In conclusion, an analysis of the NACC Neuropathology Data Set indicated that incident PSP pathology is a relatively common FTLD-Tau subtype, often differing clinically from "classic" PSP among older persons.

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Patterns and prevalence of TDP-43 Copathology in Progressive Supranuclear Palsy Reveal Regional Vulnerability and Pathologic Heterogeneity

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Background: Progressive supranuclear palsy (PSP) is a primary 4-repeat tauopathy characterized by brainstem-predominant neurodegeneration with characteristic tufted astrocytic pathology. Although tau pathology defines PSP, emerging evidence suggests that mixed proteinopathies may be more common than previously recognized. Prior studies have identified TDP-43 pathology in a subset of PSP cases, often in association with advanced age or Alzheimer-type copathology; however, reported frequencies vary and systematic evaluation of its regional distribution remains limited. We systematically evaluated TDP-43 pathology in a cohort of neuropathologically confirmed PSP cases to determine its frequency and regional distribution.

Methods: Postmortem brain tissue from the University of Washington Brain Repository and Integrated Neuroscience (BRaIN) laboratory, included 32 neuropathologically confirmed PSP cases. Phosphorylated TDP-43 immunohistochemistry was performed on sections from the middle frontal gyrus, amygdala, hippocampus/entorhinal cortex, medulla, and spinal cord. Cases were evaluated for neuronal cytoplasmic inclusions, dystrophic neurites, intranuclear inclusions, and regional distribution of pathology. Alzheimer-type pathology (Braak stage and CERAD score) and hippocampal sclerosis were recorded.

Results: TDP-43 pathology was identified in 16/32 cases (50%). Deposition demonstrated a consistent limbic predominance, with amygdala involvement in nearly all positive cases and progressive extension to the hippocampus and neocortex in a subset of cases. Pure neocortical-predominant TDP-43 was not observed. TDP-43–positive cases tended to be older but were not uniformly associated with advanced Alzheimer-type pathology, and hippocampal sclerosis was uncommon. Notably, additional TDP-43 pathology was identified in the medulla, frequently involving the inferior olivary nucleus, and in the spinal cord in select cases.

Conclusions: TDP-43 copathology is common in PSP in this systematically screened cohort and exceeds previously reported frequencies. The predominantly limbic distribution, with additional involvement of brainstem and spinal cord regions, suggests a broader regional distribution of TDP-43 deposition in PSP than previously appreciated. These findings expand the neuropathologic spectrum of PSP and support biologic heterogeneity within this primary tauopathy.

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A Quantitative Pilot Study: Traumatic Brain Injury as a modifier of Tau Burden in Confirmed cases of Progressive Supranuclear Palsy

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Background: Progressive supranuclear palsy (PSP) is a neurodegenerative disorder characterized by primary 4-repeat tau aggregation. Traumatic brain injury (TBI) is believed to be a key risk factor for subsequent neurodegenerative diseases and can lead to accumulation of phosphorylation tau within the brain. However, its role as a modifier of PSP tau pathology remains unclear. This study aims to quantitatively determine whether PSP patients with a history of TBI demonstrate altered regional cortical tau burden compared with PSP patients without TBI.

Methods: Post-mortem brain tissue was obtained from the University of Washington Brain Repository and included two PSP cases with TBI and two matched PSP cases without TBI, all with low concomitant Alzheimer's pathology (Braak < III). Formalin-fixed, paraffin-embedded sections from the middle frontal gyrus, anterior temporal cortex, and orbitofrontal cortex were stained for phosphorylated tau (AT8). Whole-slide images were acquired on the Leica Aperio platform, and tau burden in gray matter was quantified using the HALO image analysis program (% AT8-positive area).

Results: Preliminary analyses suggest higher overall AT8 tau burden in PSP cases with prior TBI compared to without TBI, with a trend in significance for TBI effect with a two-way ANOVA repeated measures analysis. This was largely driven by higher levels of tau specifically in the middle frontal gyrus.

Conclusions: Our pilot analysis suggests that prior history of TBI in those with PSP is associated with higher tau burden in the frontal cortex. More cases will be needed to confirm these findings and assess possible correlations with clinical symptomatology.

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Distinct Regional Tau Distribution of Pallidopontonigral Degeneration Compared to Progressive Supranuclear Palsy

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Background: Pallidopontonigral degeneration (PPND) is a rare hereditary 4-repeat tauopathy caused by the MAPT p.N279K mutation. While PPND and progressive supranuclear palsy (PSP) share 4-repeat tau pathology, a comprehensive comparison of regional tau distribution has not been performed.

Methods: We compared regional tau pathology of 17 patients with PPND (9 females; age at onset 44.8±5.7 years, age at death 51.5±5.6 years) with early-onset PSP (EPSP, n=18; 8 females; age at onset 51.7±3.8 years, age at death 59.2±4.8 years) and typical-onset PSP (TPSP, n=19; 8 females; age at onset 69.2±3.8 years, age at death 75.2±3.9 years). Tau burden was assessed using semi-quantitative scoring (0-3) across 18 brain regions for four pathological features: neuronal tau, coiled bodies, astrocytic tau, and threads. To objectively evaluate whether these regional differences could distinguish PPND from PSP, unsupervised hierarchical clustering and principal component analyses were performed.

Results: For neuronal tau, coiled bodies, and threads, PPND showed significantly higher severity in cortical regions and striatum, with temporal cortex demonstrating the most prominent differences (all $p < 0.0001$). Brainstem and cerebellar regions showed less severe pathology in PPND. Astrocytic tau pathology showed a distinct pattern: the temporal cortex showed higher severity in PPND, while deep gray matter and brainstem regions demonstrated significantly lower burden compared to PSP groups. Unsupervised hierarchical clustering successfully separated 94% (16/17) of patients with PPND from PSP groups. Principal component analysis confirmed this clear separation.

Conclusions: PPND showed cortical predominance with relative brainstem sparing, contrasting with PSP's brainstem-predominant tau pathology. This study demonstrates that PPND and PSP represent distinct disease entities with different regional tau distribution patterns despite both being 4-repeat tauopathies.

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Profiling of tau neuropathology in human 4R-tauopathies: progressive supranuclear palsy (PSP) and corticobasal degeneration (CBD)

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Background: Progressive supranuclear palsy (PSP) and corticobasal degeneration (CBD) are neurodegenerative diseases characterized by the accumulation of hyperphosphorylated-tau within neurons, astrocytes, and oligodendrocytes. The complex mechanisms occurring within each cell type and the effects exerted onto neighboring cells and the cellular network are largely unknown. Using spatial proteogenomic technologies, simultaneous quantitative analyses of protein and RNA expression can be performed to better understand the molecular mechanisms underpinning the symptomatology seen within 4R-tauopathies, such as PSP and CBD.

Methods: We performed high-plex multi-omics using NanoString's GeoMx Digital Spatial Profiler (DSP) assessing the expression of 1,200 proteins, and CosMx Spatial Molecular Imager (SMI) to assess RNA and protein expression in neurons and glia with and without p-tau aggregates from the middle frontal gyrus, hippocampus, cerebellum, pons, and medulla of PSP, CBD, and control cases (n=11).

Results: Preliminary findings reveal several proteins that are expressed in all cell types that are positive for p-tau accumulation, as well as several distinct proteins in the different inclusion types (tufted astrocytes, coiled bodies, astrocytic plaques and neurofibrillary tangles). For example, HSP60 is significantly upregulated in glia (coiled bodies, tufted astrocytes, and astrocytic plaques), carbonic anhydrase 2 is elevated specifically in coiled bodies, GLUD1/2 is especially high in astrocytic inclusions, whereas TREM2 and PINK1 are elevated in neurofibrillary tangles.

Conclusions: Thus far, proteins involved in proteostasis, inflammatory response, and oxidative stress display differential expression in the cell types. Studies are ongoing with the goal of identifying differential RNA and protein expression in the affected regions as compared to unaffected, as well as p-tau positive cell types as compared to negative. The mechanistic and eventual therapeutic potential of the findings are significant, as identifying disease-specific molecular signatures could enable the development of biomarkers for accurate ante-mortem diagnosis, as well as novel therapeutic targets for disease-modifying treatment.

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Neuronal cell cycle re-entry in a multiple system atrophy seeding mouse model

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Background: Evidence shows that neurons in Alzheimer disease aberrantly re-enter the cell cycle, expressing markers such as cyclins, CDKs, and Ki-67—features rarely observed in healthy brains. Neuronal cell cycle reactivation (CCR) has been sporadically reported across multiple neurodegenerative diseases, yet the disease specificity and mechanistic relevance remain poorly understood. We leverage a multiple system atrophy (MSA)-seeding mouse model, which recapitulates key neurological and motor symptoms and exhibits robust neuronal alpha-Synuclein (αSyn) aggregation in brainstem, to investigate the extent and regional specificity of neuronal CCR in the context of synucleinopathies.

Methods: Aggregates of αSyn derived from brain homogenates or CSF of patients with MSA neuropathologic diagnosis, after cycles of seed amplification, were inoculated into the dorsal striatum of a transgenic mouse model overexpressing human αSyn with the A53T mutation (line M83, hemizygous). Animals (n=7) were sacrificed after the development of severe symptoms. Control animals (n=3) were the same transgenic mice receiving PBS inoculation. The left brain was formalin fixed and paraffin embedded for histological analysis. Ki67 immunohistochemical stain was used as a cell cycle marker. Whole-brain images were obtained, and positive cells in the brainstem were quantified using QuPath digital pathology software.

Results: MSA-seeded mice demonstrated abundant neuronal αSyn aggregates in brainstem regions, whereas none of the PBS-inoculated controls revealed αSyn pathology. MSA-seeded mice had, on average, a higher Ki-67 positivity in brainstem than controls (mean (SE), 5.9 (0.2) vs. 1.6 (0.8) cells per mm2, P-value= 0.01). Most Ki67-positive cells exhibited morphological features consistent with neurons, both in MSA-seeded mice and controls.

Conclusions: Elevated Ki67 positivity in the brainstem of MSA-seeded mice with abundant αSyn pathology suggested a potential link between degenerating neurons and aberrant cell cycle entry. Our findings may be important to understand the broader biological significance of the neuronal CCR in neurodegenerative disease.

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Clinical and neuropathologic phenotypes associated to a novel GRN mutation

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Background: Primary progressive aphasia (PPA) is defined by adult onset and progressive impairment of language and absence of other consequential behavior or cognitive deficits for approximately the first two years. We present a case with onset of progressive aphasia, forgetfulness, and a tremor of upper extremities that poses a question for a nosologic classification.

Methods: Neurohistology and immunohistochemistry were used. Antibodies to TDP-43, tau, β-amyloid, α-synuclein, and GFAP were used. Direct sequencing of the transcribed regions of the GRN gene was performed with genomic DNA extracted from the brain tissue from the patient.

Results: A 67-year-old man experienced forgetfulness and word finding difficulty. A neurological exam revealed cognitive decline, language impairment, and tremor of the upper extremities. An MRI revealed asymmetric cerebral atrophy, left greater than right. Subsequent neurological exams revealed a progression in the language impairment which led to the diagnosis of semantic PPA. He died at age 78. Neuropathology revealed severe atrophy of the frontal and temporal lobes with “knife-blade” atrophy in the left frontal and temporal lobes. Severe neuronal and white matter loss with gliosis was most severe in language areas. Atrophy of the head of the caudate nucleus was also extremely severe. Frontotemporal lobar degeneration with Type A + B TDP-43 proteinopathy was characterized by the presence of intracytoplasmic and intranuclear neuronal inclusions. This was most severe in frontal and temporal lobes, caudate nucleus, putamen, amygdala, and hippocampus. Direct sequencing analysis identified a novel base substitution 3’ to non-coding exon (NM_002087.4:c.-8+2T>A).

Conclusions: Previous studies showed that PPA subtypes associated with GRN mutations present considerable heterogeneity. We conclude that the current case presented with a progressive aphasia and not primary progressive aphasia. The severe atrophy observed in language areas and caudate nucleus suggests that vulnerability of these areas may be related to the GRN mutation and unknown pathogenetic mechanisms. PHS R01NS137469.

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Neuropathologic Review of Spinocerebellar Ataxia Type 7 in a Five Generation Family

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Background: Spinocerebellar ataxia type 7 (SCA7) is an autosomal dominant neurodegenerative disorder characterized by progressive cerebellar ataxia and macular degeneration leading to vision loss, along with dysarthria, dysphagia, pyramidal signs, and ophthalmoplegia. We describe clinical and neuropathologic findings from a five-generation family with genetically confirmed SCA7, emphasizing correlations between clinical manifestations and disease pathology.

Methods: Eleven affected individuals from generations III–V had medical records available for review.

Results: Neurologic abnormalities included gait and limb ataxia, dysarthria, dysphagia, hyperreflexia, spasticity, ankle clonus, and up-going toes. Ocular findings comprised decreased visual acuity, visual field deficits, impaired color perception, abnormal extraocular movements, the Flynn phenomenon, and macular pigmentary mottling. One patient underwent complete autopsy. Gross examination of the central nervous system revealed spinal cord atrophy and marked atrophy of the cerebellum, pons, and medulla. Microscopically, affected regions showed neuronal loss, fibrillary astrogliosis, and polyglutamine-immunoreactive neuronal nuclei with or without intranuclear inclusions. The spinal cord demonstrated extensive atrophy with preserved posterior columns and loss of myelinated fibers in the ventral and dorsal spinocerebellar and spino-olivary tracts, most pronounced in cervical and upper thoracic segments. Cerebellar sections revealed severe dentate nucleus and Purkinje cell loss accompanied by Bergmann gliosis. Polyglutamine-positive intranuclear inclusions were widespread, most prominent in the dentate and brainstem nuclei.

Conclusions: Neuropathologic features in this case aligned with previously published SCA7 autopsy studies. This series contributes additional longitudinal clinical and visual data and expands the limited literature providing detailed neuropathologic characterization of SCA7.

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Neuronal intranuclear inclusion disease with motor-predominant symptoms in a middle-aged woman

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Background: Neuronal intranuclear inclusion disease (NIID) is a rare adult-predominant neurodegenerative disease characterized by a GGC trinucleotide repeat expansion in the 5' untranslated region of the NOTCH2NLC and clinically characterized by cognitive, autonomic, and motor manifestations. Glial and neuronal intranuclear eosinophilic inclusions predominating in the basal ganglia and the cerebral cortices are the hallmark of the disease, although the PNS, viscera and skin may be also involved. We present the case of an adult woman with slowly progressive upper motor neuron and bulbar symptoms, clinically diagnosed as primary lateral sclerosis and complicated by repeated aspiration pneumonia which resulted in the patient's demise.

Methods: A brain-only autopsy was performed with a postmortem interval of 19 hours. A midline section through the corpus callosum was performed and the right hemibrain was stored for research purposes. The left hemibrain and 4 levels of spinal cord were fixed in 10% neutral buffered formalin for 10 days. A neurodegenerative-oriented sampling protocol was performed and H&E-, luxol fast blue-stained slides, and immunostains for p62, ubiquitin, p-Tau, p-TDP43, FUS and a-Beta were performed for assessment.

Results: The brain's fresh weight was 1,090 grams and gross examination was unremarkable. Microscopic assessment showed glial and neuronal cells harboring intranuclear eosinophilic inclusions highlighted with p62 and ubiquitin immunostains. These were present throughout the CNS including gray and white matter regions of the brainstem, limbic, and cortical regions. However, a higher frequency was identified in the amygdala, subcortical white matter, and inferior olivary nuclei. No other inclusions were present. Remarkably, upper motor neuron loss was minimal but prominent microglial activation in subcortical white matter and bilateral lateral column degeneration throughout the spinal cord was present.

Conclusions: While genetic testing is pending, the clinical-pathological features are most consistent with NIID. The aim of this presentation is to bring awareness of this uncommon neurodegenerative condition.

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Cryo-EM Studies of PrP Amyloid: Comparison of Filaments' structures from Cases with the Q217R and the F198S PRNP Mutations

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Background: Prion protein (PrP) aggregation is central to the pathogenesis of the dominantly inherited PrP amyloidoses. The clinical and neuropathologic phenotypes of these neurodegenerative diseases vary for the anatomical distribution of PrP aggregates and for the presence or absence of tau neurofibrillary pathology. The coexistence of PrP aggregates with tau neurofibrillary tangles has been reported in a group of dominantly inherited PrP amyloidoses. This group includes Gerstmann-Sträussler-Scheinker disease associated with the F198S and Q217R PRNP mutations. The structure of PrP filaments from the amyloid of F198S carriers has been previously studied by cryogenic electron microscopy (cryo-EM) and the aim of the current study is to compare it with structure of PrP filaments extracted from two symptomatic Q217R carriers.

Methods: Neuropathologic studies of seven symptomatic Q217R carriers from a large pedigree were carried out using thioflavin S to reveal the presence of amyloid plaques and immunohistochemistry to determine the amyloid's nature. Antibodies to PrP including PrP 23-40, 3F4, and PrP 220-231 were used. PrP filaments were extracted from the cerebellum of two family members and were studied by cryo-EM.

Results: Numerous PrP amyloid plaques were present in most areas of gray matter but the most severe involvement was seen throughout the cerebellar cortex. Neurofibrillary tau pathology was severe throughout the cerebral cortex and cortical nuclei. Cryo-EM analysis of amyloid extracted from the cerebellar cortex of the two patients revealed that PrP filaments are composed of dimeric left-handed protofilaments with their protomers sharing a common spiral fold. Current studies are in progress to determine the amino acid composition of the protomers and the composition of each protomer's β-strands.

Conclusions: The data obtained by cryo-EM provide new insights into the structure of the Q217R PrP amyloid and reveal a strong similarity between PrP filaments associated with the Q217R and those associated with the F198S mutation. PHS R01AG071177

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Single-Channel Dual Labeling of GS and GFAP Improves Astrocyte Detection in Spectrally Limited Multiplex Assays

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Background: Defining cell-type localization of neuropathological drivers is essential for understanding disease mechanisms. Determining how a pathological molecule (e.g., a misfolded protein) distributes across brain cells—and how this distribution shifts across biological contexts—clarifies disease processes. Multiplex immunofluorescence (IF) enables spatial localization of molecular targets in archival tissue; however, antibody compatibility and limited spectral bandwidth restrict panels to ~3–4 markers before overlap becomes prohibitive. Broader cell-typing strategies streamline translational research because standard IHC markers are less resource-intensive than multimodal subclassification approaches. Such insight guides mechanistic studies and development of clinically relevant genetic models (e.g., cell-type–specific inducible drivers). No single immunomarker ubiquitously labels mature astrocytes. Glial Fibrillary Acidic Protein (GFAP) is a canonical marker of activated astrocytes but captures only a subset. Glutamine Synthetase (GS), a metabolic astrocyte marker, exhibits partially overlapping and regionally distinct expression. To improve astrocytic coverage under limited spectral conditions, we performed dual immunofluorescent staining with digital pathology–based quantification to evaluate complementarity between GFAP and GS and feasibility of combining them on a single spectral channel.

Methods: Formalin-fixed paraffin-embedded cortical sections were dual stained for GFAP and GS on separate channels, and overlap was quantified. S-Split Far-red (650 nm; OPAL 690 + Alexa 647) and S-Split Orange (545 nm; OPAL 570 + Alexa 546) configurations were compared to optimize same-channel labeling. Cyclic IF was integrated to generate a 7-plex, 6-color panel with GFAP and GS combined on one channel.

Results: Among GS-positive cells, 65% co-expressed GFAP (37% weak, 17% moderate, 11% strong). Among GFAP-positive cells, 73% co-expressed GS (58% weak, 11% moderate, 4% strong). Regions with lower GS immunoreactivity demonstrated higher GFAP immunoreactivity, supporting spatial complementarity.

Conclusions: Dual labeling of GS and GFAP enhances astrocyte detection without increasing spectral burden and provides a practical strategy for maximizing astrocytic coverage in multiplex IF assays.

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Whole Slide Vector Visualization: A Visualization Platform for Interpreting and Utilizing AI Models in Histopathologic Image Analysis

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Background: Foundation models are becoming increasingly popular for analyzing whole slide images (WSI), but beyond their use for classification tasks the potential utility of extracted image features (embeddings) remain poorly understood. In this work, we developed a novel visualization platform that relates embeddings back to input image tiles, enhancing Artificial Intelligence (AI) explainability by allowing users to explore the nature of the foundation model outputs.

Methods: A highly-performant database and server-side computational platform were developed in parallel with a web-based front end, facilitating WSI preprocessing, feature extraction using foundation models, storage of embeddings, and integrated visualization tools. Histopathologic image feature extraction models were evaluated for their usefulness in characterizing histopathologic features from whole slide images from generic (tile-level) and specific (tau-positive) regions of interest. We compared state-of-the-art pathology AI models including Conch, Uni, Uni2, ProvGigapath, Virchow, and Virchow2, as well as an unreleased transformer-based image feature extractor focused on neurofibrillary tangle pathology developed by our lab.

Results: We used our computational platform to compare feature extraction model performance on WSIs from our Alzheimer's Disease Research Center. Model outputs were visualized by dimensionality reduction and attention-based heatmaps on associated image regions. Our visualization tool enabled intuitive visual exploration of embeddings that varied across different brain regions highlighting their utility in describing tissues, but revealed that image types not represented in the model training data lead to extracted features that are difficult to interpret.

Conclusions: Visualizing AI-extracted image features with an interface that associates model outputs with their associated image regions enhances AI model explainability, helps identify comparative strengths and weaknesses of different image feature extractors, and demonstrates potential usefulness in workflow automation and hypothesis exploration.

Posters: Tumors: Glial

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From Atypical Ganglioglioma to ZFTA Fusion-Positive Ependymoma, How a Tumor Eventually Histologically and Molecularly Declared Itself

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Background: This is a male born in 1980 who in 2009 underwent right parietal resection of brain tumor at outside hospital that was later sent to academic center for consultation and felt to be most consistent with atypical ganglioglioma, unusual as that diagnosis was. The report is available for review but slides are not. He did not complete adjuvant therapy and was lost to follow up but in 2021 presented with seizures., worsened vasogenic edema and recurrence of the tumor. He underwent resection of the tumor and many atypical ganglion-like cells (some binucleate) and glial cells with increased mitotic activity were seen. Ki-67 was focally over 30% positive nuclei. Classic ependymal histology was minimal at best. Per report, the histology was similar to the 2009 resection. NGS sequencing was not performed but methylation profiling at the NCI was performed and in the 2021 classifiers was suggestive of ZFTA-fused ependymoma but did not match to high confidence level. This time, he underwent radiation therapy but no chemotherapy. Some radiographic evidence of recurrence was present in 2023 but by 2025 it had recurred again and was resected again in June 2025. This time, it had some of the same glial and ganglion cell features, but had grade 3 ependymal features. Both NGS and methylation profiling were performed in 2025 and this time it matched with high confidence to supratentorial ependymoma, ZFTA fusion-positive and the NGS was also positive for ZFTA-RELA fusion gene. It recurred again in Jan 2026, was resected, and this time the grade 3 ependymal features dominated the histology. Further molecular workup was not performed.

Methods: histology, methylation profiling, NGS

Results: see background

Conclusions: through time, the tumor histologically evolved into a pattern consistent with the molecular phenotype. See background for details.