Company Logo

Product Name API Manufacturing

Performance Qualification Protocol
for Agilent High Performance Liquid Chromatography System #10

Validation # PQ-LA-000-00

Performance Qualification Protocol

Prepared By:

_____ Originator

_____ Date

Approved By:

_____ Quality Unit

_____ Date

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1. Objective

1.1. Define the Performance Qualification requirements and acceptance criteria for the Agilent 1260 HPLC Chromatographic System #10.

1.2. To verify that the system operates in accordance with the intended use of the system.

2. References

2.1. IQ/OQ requirements should be successfully completed prior to PQ execution.

2.1.1. IQ to be completed by manufacturer. See attached Form PQ-02.

2.1.2. OQ to be completed by manufacturer. See attached Form PQ-02.

3. Description

The Agilent 1260 Chromatographic System #10 is used for analysis of spinosyns in in-process Fermentation samples.

The Agilent 1260 Chromatographic system #10 consists of the following:

- G1329B 1260 ALS Autosampler, used as the sample injector for the system.
- G1312B 1260 Binary Pump, used for eluent delivery to the system.
- G1314C 1260 VWD VL+ Detector is an optical measurement device.
- G1316A 1260 TCC Column Heater is used to control the column temperature.
- G1322A 1260 Degasser to remove gases from the mobile phase.

The Agilent 1260 Chromatographic System #10 is directly connected to a data server (serial number, ds000000), for transfer and interpretation of data by the Atlas Chromatography Data System.

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4. Responsibilities

4.1. The responsibilities of the Quality Unit include but are not limited to:

4.1.1. Creation of this protocol.

4.1.2. Assistance with the completion of the steps required to conduct this validation.

4.1.3. Providing personnel to complete the execution of the validation.

4.1.4. Reviewing and approving the protocol and any data generated during the execution of this report.

4.1.5. Generation of the final summary report for review and approval by those who originally approved the protocol or their designees

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5. Performance Qualification

Performance Qualification for the Agilent 1260 Chromatographic System #10 includes running a linearity study and percent recovery study in a known sample.

5.1. Verify that all procedures, logs and manuals are available, complete and correct.

5.2. Verify that there is no excessive baseline noise.

5.3. Verify that the system baseline is displaying between -5 and +5 mV.

5.4. Use procedure "Determination of Spinosyn Factors – Calibration and Atlas Setup" to check the system.

5.4.1. Verify that the resolution between the Factor A peak and the Factor D peak is greater than 3.0.

5.4.2. Verify that the peak tailing for Factor A and Factor D are both less than 1.8.

5.4.3. Verify that reproducibility of Factor and Factor D in the Standard has an RSD of less than 1.5%.

5.4.4. Verify that the correlation coefficient for the standard curve is equal to or greater than 0.9990.

5.4.5. Verify that the % Nominals for Factor A and Factor D are between 95 and 105%.

5.4.6. Verify that the assay of the Tox lot is equal to 88% ±1.5.

5.4.7. Verify that the assay of a previously ran fermentor sample is within 3% of the previous assay.

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6. Documentation

6.1. Documentation requirements for the PQ must be completed with all signatures in place.

6.2. Document that all acceptance criteria listed on the forms are met.

6.3. Any deviation or abnormality observed during the execution of this protocol must be investigated.

6.4. Any changes to the protocol, after it has been approved, will require a "Protocol Addendum" and require re-approval by representatives of the same departments who originally approved the protocol, or their designees.

6.5. The Quality Unit must file all required documentation.

6.6. Raw data will be reviewed by representatives of the same department who originally signed this protocol.

6.7. Upon completion of the protocol, a summary report is generated which will document the validation effort for this system. This includes the documentation gathered for the data and testing results of the PQ.

7. Validation Forms and Attachments

7.1. PQ-01: SOPs/Manuals

7.2. PQ-02: IQ/OQ Verification

7.3. PQ-03: Performance Qualification

Form Title: SOPs / MANUALS	FORM #: PQ-01
System/Equipment Name: Agilent 1260 System #10	Validation# PQ-LA-000-00
System/Equip. ID: Pump – G1312B, Ser. #DEAAE00000 Degasser – G1322A, Ser. #JP00000000 Autosampler – G1329B, Ser. #DEABE00000 Column heater – G1316A, Ser. #DEAAK00000 Detector – G1314C, Ser. #DEAAV00000	

SOP / Manual Title	Draft or Approved	Effective Date	Initial	Date
AGILENT 1260 OPERATIONS	COMPANY MANUAL	5/2009	LM	7/21/10
Determination of Spinosyn Factors – Calibration and Atlas Setup	MOC 162008020000	3/17/08		

Reviewed By: Lee Mecum **Date:** 7/28/2010

Form Title: IQ/OQ Verification	FORM #: PQ-02
System/Equipment Name: Agilent 1260 System #10	Validation# PQ-LA-000-00
System/Equip. ID: No. 7246 C - Room 3 Pump – G1312B, Ser. #DEAAE00117 Degasser – G1322A, Ser. #JP00000000 Autosampler – G1329B, Ser. #DEABE00000 Column heater – G1316A, Ser. #DEAAK00000 Detector – G1314C, Ser. #DEAAV00000	

Verification	Acceptance Criteria	Actual	Accept Y/N	Init	Date
Verify that the IQ has been completed for the system	Record IQ number and date completed.	IQ-LA-000-00	Y	JL	8/31/10
Verify that the OQ has been completed for the system	Record OQ number and date completed.	OQ-LA-000-00	Y	JL	8/31/10

Reviewed By: Joe Lowe Date: 8/28/10

Form Title: Performance Qualification	FORM #: PQ-03
System/Equipment Name: Agilent 1260 System #10	Validation# PQ-LA-00-00
System/Equip. ID: FERMENTOR #3 Pump – G1312B, Ser. #DEAAE0000 Degasser – G1322A, Ser. #JP0000000 Autosampler – G1329B, Ser. #DEABE00000 Column heater – G1316A, Ser. #DEAAK00000 Detector – G1314C, Ser. #DEAAV0000	

Verification	Acceptance Criteria	Actual	Accept Y/N	Init/Date
Verify that the system baseline is stable and free of excessive baseline noise.	Drift is <0.5 mV. No peaks >0.5 mV.	Drift = .045mV Peaks = 0.06 mV	Y	DB 10/9/10
Verify that the system baseline is displaying between -5 and +5 mV.	A value between -5 and +5 mV.	mV= 2 mV	Y	DB 10/8/10
Verify that the resolution between the Factor A peak and the Factor D peak is greater than 3.0	The resolution is greater than 3.0.	3.0	Y	DB 10/8/10
Verify that the peak tailing for Factor A and Factor D are both less than 1.8	Peak tailing is less than 1.8.	Fac A = 1.5 Fac D = 1.78	Y	DB 10/8/10
Verify that the system is capable of acceptable reproducibility.	Fac A and Fac D peaks have an RSD <1.5%.	Fac A RSD = 1.45 Fac D RSD = 1.1	Y	DB 10/8/10
Verify that the correlation coefficient for the standard curve is equal to or greater than 0.9990	The corr coef is equal to or greater than 0.9990	Corr coef = 0.99895	Y	DB 10/8/10
Verify that the % Nominals for the standards are acceptable.	All levels of Fac A and Fac D are 100% ±5.	Fac A = 99.0% Fac D = 98.0%	Y	DB 10/8/10
Verify that the assay of the Tox lot is equal to 88% ±1.5%	The assay result is 88% ±1.5%	86.5%	Y	DB 10/8/10
Verify that the assay of a previously run fermenter sample is within 3% of the previous assay.	The assay of the fermentor sample is within 3% of the previous assay.	Previous= 92.2% Results= 89.25% Difference= 2.952 3.0%	Y	DB 10/8/10

Identification of fermenter sample (Tank/Age): H - 162 hrs

Documentation: Atlas workbook name: Fermentor # 3

Reviewed By: Don Cox Date: 10/14/10

Form Title: Performance Qualification	FORM #: PQ-03
System/Equipment Name: Agilent 1260 System #10	Validation# PQ-LA-00-00
System/Equip. ID: FERMENTOR #3 Pump – G1312B, Ser. #DEAAE0000 Degasser – G1322A, Ser. #JP0000000 Autosampler – G1329B, Ser. #DEABE00000 Column heater – G1316A, Ser. #DEAAK00000 Detector – G1314C, Ser. #DEAAV0000	

Verification	Acceptance Criteria	Actual	Accept Y/N	Init/Date
Verify that the system baseline is stable and free of excessive baseline noise.	Drift is <0.5 mV. No peaks >0.5 mV.	Drift = 0.1mV Peaks = 0.55mV	—	JB 9/15/10
Verify that the system baseline is displaying between -5 and +5 mV.	A value between -5 and +5 mV.	mV= -5 & 5	Y	JB 9/15/10
Verify that the resolution between the Factor A peak and the Factor D peak is greater than 3.0	The resolution is greater than 3.0.	2.6	≃	JB 9/15/10
Verify that the peak tailing for Factor A and Factor D are both less than 1.8	Peak tailing is less than 1.8.	Fac A = 1.3 Fac D = 1.3	✓	JB 9/15/10
Verify that the system is capable of acceptable reproducibility.	Fac A and Fac D peaks have an RSD <1.5%.	Fac A RSD = <1.5% Fac D RSD = 1.3%	—	JB 9/15/10
Verify that the correlation coefficient for the standard curve is equal to or greater than 0.9990	The corr coef is equal to or greater than 0.9990	Corr coef = 0.9983	✓	JB 9/15/10
Verify that the % Nominals for the standards are acceptable.	All levels of Fac A and Fac D are 100% ±5.	Fac A = 99.4 Fac D = 99.3	✓	JB 9/15/10
Verify that the assay of the Tox lot is equal to 88% ±1.5%	The assay result is 88% ±1.5%	89.1%	—	JB 9/15/10
Verify that the assay of a previously run fermenter sample is within 3% of the previous assay.	The assay of the fermentor sample is within 3% of the previous assay.	Previous= — 1st Results= Run Difference=	✓	JB 9/15/10

Identification of fermenter sample (Tank/Age): H – 160hr

Documentation: Atlas workbook name: Ferm #3

Reviewed By: Don Cig Date: 10/5/10

Form Title: Performance Qualification	FORM #: PQ-03
System/Equipment Name: Agilent 1260 System #10	Validation# PQ-LA-00-00
System/Equip. ID: Form 3 Pump – G1312B, Ser. #DEAAE0000 Degasser – G1322A, Ser. #JP0000000 Autosampler – G1329B, Ser. #DEABE00000 Column heater – G1316A, Ser. #DEAAK00000 Detector – G1314C, Ser. #DEAAV0000	

Verification	Acceptance Criteria	Actual	Accept Y/N	Init/Date
Verify that the system baseline is stable and free of excessive baseline noise.	Drift is <0.5 mV. No peaks >0.5 mV.	Drift = <.5 mV Peaks = 70.5 mV	Y	JB 9/23/10
Verify that the system baseline is displaying between -5 and +5 mV.	A value between -5 and +5 mV.	mV= -5 AND +5 mV	Y	JB 9/23/10
Verify that the resolution between the Factor A peak and the Factor D peak is greater than 3.0	The resolution is greater than 3.0.	> 3.0	Y	JB 9/23/10
Verify that the peak tailing for Factor A and Factor D are both less than 1.8	Peak tailing is less than 1.8.	Fac A = <1.8 Fac D = <1.8	Y	JB 9/23/10
Verify that the system is capable of acceptable reproducibility.	Fac A and Fac D peaks have an RSD <1.5%.	Fac A RSD = <1.5% Fac D RSD = <1.5%	Y	JB 9/23/10
Verify that the correlation coefficient for the standard curve is equal to or greater than 0.9990	The corr coef is equal to or greater than 0.9990	Corr coef = 70.9990	Y	JB 9/23/10
Verify that the % Nominals for the standards are acceptable.	All levels of Fac A and Fac D are 100% ±5.	Fac A = 100% ±5 Fac D = 100% ±5	Y	JB 9/23/10
Verify that the assay of the Tox lot is equal to 88% ±1.5%	The assay result is 88% ±1.5%	89% ±1.5%	Y	JB 9/23/10
Verify that the assay of a previously run fermenter sample is within 3% of the previous assay.	The assay of the fermentor sample is within 3% of the previous assay.	Previous= 90.2% Results= 91.7% Difference= 1.5%	Y	JB 9/23/10

Identification of fermenter sample (Tank/Age): H – 164 hr

Documentation: Atlas workbook name: Form # 3

Reviewed By: Dan Cox Date: 10/5/10

Form Title: Performance Qualification	FORM #: PQ-03
System/Equipment Name: Agilent 1260 System #10	Validation# PQ-LA-00-00
System/Equip. ID: Fermentor #3 Pump – G1312B, Ser. #DEAAE0000 Degasser – G1322A, Ser. #JP0000000 Autosampler – G1329B, Ser. #DEABE00000 Column heater – G1316A, Ser. #DEAAK00000 Detector – G1314C, Ser. #DEAAV0000	

Verification	Acceptance Criteria	Actual	Accept Y/N	Init/Date
Verify that the system baseline is stable and free of excessive baseline noise.	Drift is <0.5 mV. No peaks >0.5 mV.	Drift = 0.4 mV Peaks = 0.55 mV	Y	9/30/10 RC
Verify that the system baseline is displaying between -5 and +5 mV.	A value between -5 and +5 mV.	mV= -4 – +3 mV	Y	9/30/10 RC
Verify that the resolution between the Factor A peak and the Factor D peak is greater than 3.0	The resolution is greater than 3.0.	3.6	Y	9/30/10 RC
Verify that the peak tailing for Factor A and Factor D are both less than 1.8	Peak tailing is less than 1.8.	Fac A = < 1.5 Fac D = < 1.5	Y	9/30/10 RC
Verify that the system is capable of acceptable reproducibility.	Fac A and Fac D peaks have an RSD <1.5%.	Fac A RSD = 1.3% Fac D RSD = 1.4%	Y	9/30/10 RC
Verify that the correlation coefficient for the standard curve is equal to or greater than 0.9990	The corr coef is equal to or greater than 0.9990	Corr coef = 0.7992	Y	9/30/10 RC
Verify that the % Nominals for the standards are acceptable.	All levels of Fac A and Fac D are 100% ±5.	Fac A = 102 % Fac D = 99.5%	Y	9/30/10 RC
Verify that the assay of the Tox lot is equal to 88% ±1.5%	The assay result is 88% ±1.5%	87.1%	Y	9/30/10 RC
Verify that the assay of a previously run fermenter sample is within 3% of the previous assay.	The assay of the fermentor sample is within 3% of the previous assay.	Previous= 91.7% Results= 89.9% Difference= 0.9%	Y	9/30/10 RC

Identification of fermenter sample (Tank/Age): H - 156 hrs

Documentation: Atlas workbook name: Ferm #3

Reviewed By: Dan Cox Date: 10/5/10

Form Title: Performance Qualification	FORM #: PQ-03
System/Equipment Name: Agilent 1260 System #10	Validation# PQ-LA-00-00
System/Equip. ID: Form 3 Pump – G1312B, Ser. #DEAAE0000 Degasser – G1322A, Ser. #JP0000000 Autosampler – G1329B, Ser. #DEABE00000 Column heater – G1316A, Ser. #DEAAK00000 Detector – G1314C, Ser. #DEAAV0000	

Verification	Acceptance Criteria	Actual	Accept Y/N	Init/Date
Verify that the system baseline is stable and free of excessive baseline noise.	Drift is <0.5 mV. No peaks >0.5 mV.	Drift = < 0.3 mV Peaks = > 0.5 mV	Y	DB 10/7/10
Verify that the system baseline is displaying between -5 and +5 mV.	A value between -5 and +5 mV.	mV= -5.4 mV	Y	DB 10/7/10
Verify that the resolution between the Factor A peak and the Factor D peak is greater than 3.0	The resolution is greater than 3.0.	3.2	Y	DB 10/7/10
Verify that the peak tailing for Factor A and Factor D are both less than 1.8	Peak tailing is less than 1.8.	Fac A = < 1.7 Fac D = < 1.82	Y	DB 10/7/10
Verify that the system is capable of acceptable reproducibility.	Fac A and Fac D peaks have an RSD <1.5%.	Fac A RSD = < 1.5% Fac D RSD = 1.4%	Y	DB 10/7/10
Verify that the correlation coefficient for the standard curve is equal to or greater than 0.9990	The corr coef is equal to or greater than 0.9990	Corr coef = 0.9993	Y	DB 10/7/10
Verify that the % Nominals for the standards are acceptable.	All levels of Fac A and Fac D are 100% ±5.	Fac A = 99.2% Fac D = 98.5%	Y	DB 10/7/10
Verify that the assay of the Tox lot is equal to 88% ±1.5%	The assay result is 88% ±1.5%	89.4%	Y	DB 10/7/10
Verify that the assay of a previously run fermenter sample is within 3% of the previous assay.	The assay of the fermentor sample is within 3% of the previous assay.	Previous= 91.7 Results= 89.9 Difference= 1.8%	Y	DB 10/7/10

Identification of fermenter sample (Tank/Age): H - 154 hrs

Documentation: Atlas workbook name: Form # 3

Reviewed By: Don Cox Date: 10/13/10