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1.0 Purpose

The HIAC Royco 9703 Particle Counter uses light obscuration sensors to determine the cumulative particulate count in parenteral pharmaceuticals and utilizes PharmSpec software to provide an interface for determining compliance with the USP General chapter <788> (USP 38 or the current edition of USP) requirements for the following tests.

- Environmental (Glassware) Test
- Volume Accuracy Test
- Counting Accuracy Test
- Sensor Resolution Test
- Moving Window Test
- Small Volume Injections (SVI) Less than 100 mL
- Small Volume Injections (SVI) Less than 25 mL
- Large Volume Injections (LVI) Test

NOTE: The Volume Accuracy, Counting Accuracy, Sensor Resolution and Moving Window Tests are a part of the bi-annual calibration procedures and are not involved in routine functions. Accessibility to these functions is restricted to the manufacturer's Calibration Technician and the System Administrator.

2.0 Scope

This equipment is to be used by Process Analytics/Quality Control (PA/QC) personnel to quantitate particulate matter in aqueous solutions and parenteral pharmaceuticals as per USP General chapter <788> (USP 38 or the current edition of USP).

3.0 Authority and Responsibility

3.1 The Director, Process Analytical/Quality Control (PA/QC) has the authority to define this procedure.

3.2 PA/QC is responsible for training laboratory personnel and documenting the training to Biopharmaceutical Quality Assurance (BQA).

3.3 PA/QC personnel are responsible for implementation and performing this procedure.

3.4 Biopharmaceutical Quality Assurance (BQA) is responsible for auditing records pertaining to this procedure.

3.5 The System Administrator has the authority to assign operator access.

4.0 Materials and Equipment

4.1 HIAC Royco 9703 Liquid Particle Counter (9064 Counter, HRLD-400 Sensor, 3000A Sampler), Pacific Scientific Co. (NCI C122562, BDP 65390).

4.2 Purifier Vertical Clean Bench (Labconco NCI S058211, BDP 80350).

4.3 Fisher Scientific FS60 Ultrasonicator.

4.4 Distilled, Deionized Water (Sartorious Arium Pro, or Water for Injection (WFI).

4.5 Clean and sterile glassware with a minimum volume of 50 mL.

4.6 Dell Inspiron 3500 Computer (NCI Equipment ID S048081) or equivalent.

4.7 PharmSpec [software Version 2.0 (or current version of the software.)]

4.8 Powder-free gloves.

5.0 HIAC Royco 9703 Liquid Particle Counter

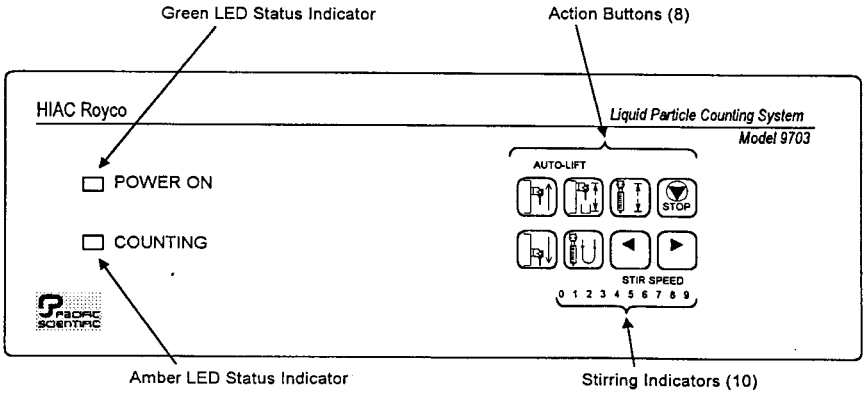
The 9703 Liquid Particle Counting System, which is placed in a vertical flow clean bench in order to eliminate the introduction of foreign particulates, operates as both “particle counter” and “sampler” in one self-contained unit.

The “particle counter” controls many functions of the 9703, as well as the count **START** and count **STOP** instructions to the sampler section and sensor. Two status indicators are provided for the Operator on the front panel. The green (**POWER ON**) LED status indicator shows that the system is “on” and the amber (**COUNTING**) LED status indicator shows that the system is performing “counting” operations. The “particle counter” also processes the analog signal from the sensor so that each particle is individually counted and sized as it passes through the sensing zone within the sensor. The count and size data from the 9703 is transmitted to a computer where the PharmSpec™ software provides the Operator with the measured particle size distribution.

The “sampler” has a sensor arm that holds the sensor and sample probe. A lift mechanism controls the height of the sensor arm. By raising and lowering the sensor and probe, the operator can change sample bottles easily without disturbing the sample handling system. In addition, the sampler section has a magnetic stir motor that allows the operator to use nine different stir speeds or turn the stirrer motor off.

5.1 Front Panel Familiarization

The front panel of the 9703 contains two status indicators, one set of controls for the stirring apparatus and a set of eight action buttons.



5.1.1 Status Indicators

5.1.1.1 The green (POWER ON) status indicator denotes application of power to the 9703.

5.1.1.2 The amber (COUNTING) status indicator denotes that the counter section of the 9703 is performing particle counting operations.

5.1.2 Stirring Indicators

The stirring indicators are a set of numbers from 0 to 9. The number 9 represents maximum speed (about 555 rpm), the number 1 represents minimum speed (about 170 rpm), and the number 0 indicates that the stir bar motor is off.

5.1.3 Action Buttons

The PharmSpec Software does NOT need to be initiated for the “Action Buttons” to be operational.

5.1.3.1 [STOP] Button

Pressing the [**STOP**] button causes any sample run to abort and the sensor lift operations to terminate. The stir bar motor will continue to operate.

5.1.3.2 [SET] and [LOAD] Buttons

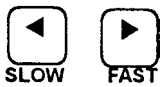
The Operator uses the [**SET**] and [**LOAD**] buttons to manipulate the sensor lift mechanism.

5.1.3.2.1 The [**SET**] button establishes the bottom position of the sensor lift mechanism. The operator will want to change this position depending on the type of bottle or beaker used for sampling. Two set speeds are available:

- Press and hold the [**SET**] button for rapid motion.
- Press and release the [**SET**] button for precise placement. The bottom position determined with the [**SET**] button is saved until a new bottom position is established.

5.1.3.2.2 The [**LOAD**] button causes the sensor lift mechanism to travel from its bottom set point to home position (the highest point in its range of motion) or vice versa.

5.1.3.3 [**SLOW**] and [**FAST**] Buttons



5.1.3.3.1 The [**SLOW**] button decreases the stir bar motors by ~48 rpm for each button pressed.

5.1.3.3.2 The [**FAST**] button increases the stir bar motors by ~48 rpm for each button pressed.

5.1.3.4 Syringe LOAD Button

When the operator presses the syringe **LOAD** button, the sampler syringe drive will send the syringe plunger to either its bottom or top most position, thus loading or unloading the syringe.

5.1.3.5 Syringe CLEAN Button

When the operator presses the syringe **CLEAN** button, the sampler will draw a full syringe of fluid into the sampler through the sensor and push this fluid out through the drain tubing.

NOTE: The operator should use the "syringe clean" function to flush the storage solution (70% Isopropyl alcohol) from the sampler with clean, particle-free water prior to each analytical use or to fill the system with a storage solution after usage. All procedures performed on the system (preventative maintenance, calibration, cleaning, sample analysis, storage) must be recorded in the equipment logbook.

6.0 PharmSpec™ (System Setup)

The PharmSpec™ software has been installed onto the system-dedicated computer. The HIAC Royco 9703 Particle Counting System is configured and calibrated by a manufacturer's field representative to provide information concerning the hardware, channel settings, sensor calibration, and run parameters. These settings may change with each bi-annual calibration by the manufacturer's field representative and will overwrite the pre-existing settings. The current settings may be found in the most recent Calibration Report, located in the Master Equipment File (MEF), and may NOT be changed by any operator.

6.1 System Initiation

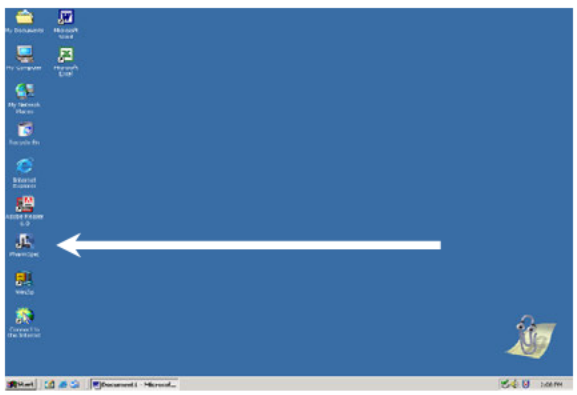
6.1.1 Turn on the HIAC Royco 9703 Liquid Particle Counter by pressing the toggle switch located on the right-rear corner of the counter.

6.1.2 Turn on the computer that controls the instrument and software.

6.1.3 Log into the network using your BDP screen name and password.

6.2 Operator Log-In

6.2.1 Double click on the PharmSpec™ (desktop icon to initiate the software package.)



6.2.2 Upon software initiation, an “Operator ID” Screen will prompt for an Operator ID and a password. The System Administrator prior to any attempt to operate the system must assign a new user (Select “Set Up” on the menu bar.) Click on “Administration.” Click on “Add User.”

6.2.3 Enter your operator name and password in the appropriate fields and click on OK.

6.2.4 New users operating the system for the first time will be prompted to change their password the first time they activate the software

6.2.5 Type and “Verify New Password” in the appropriate fields.

6.2.6 Click on OK.

NOTE: The system is calibrated bi-annually by a manufacturer’s service technician. Assure that the equipment is still within calibration (The next calibration due date is recorded on the calibration sticker affixed on the front of the equipment). If the expiration date has passed and the equipment has not been calibrated since, DO NOT USE THE EQUIPMENT until the service technician has returned to re-calibrate the system.

6.3 Hardware Setup

6.3.1 The manufacturer’s Calibration Technician sets the Hardware Setup during the bi-annual calibration and the operators may NOT change the majority of these settings. The ONLY settings that may be changed by the operator are those listed under “Operating Parameters” on the Hardware Setting screen. They are listed in the following sections 6.3.1.1 through 6.3.1.5.

6.3.1.1 **Sample Volume (mL)**

The “Sample Volume” for routine analyses is set at 5.0 mL per injection. This value may be changed to accommodate the volume of sample submitted for testing.

6.3.1.2 **Number of Runs**

The “Number of Runs” for routine analyses is set at 3 injections per run. This value may be changed to accommodate the volume of sample submitted for testing; however, it should be noted that the first injection of each run is considered as a “system purge” to remove any residual sample from the previous run and the data is discarded and not used in any subsequent calculations.

6.3.1.3 **Dilution Factor**

The “Dilution Factor” for routine analyses is set at 1.0 mL; however, this value may be changed accordingly when the volume of sample submitted for testing is too small and requires diluting to obtain the minimum volume required for testing.

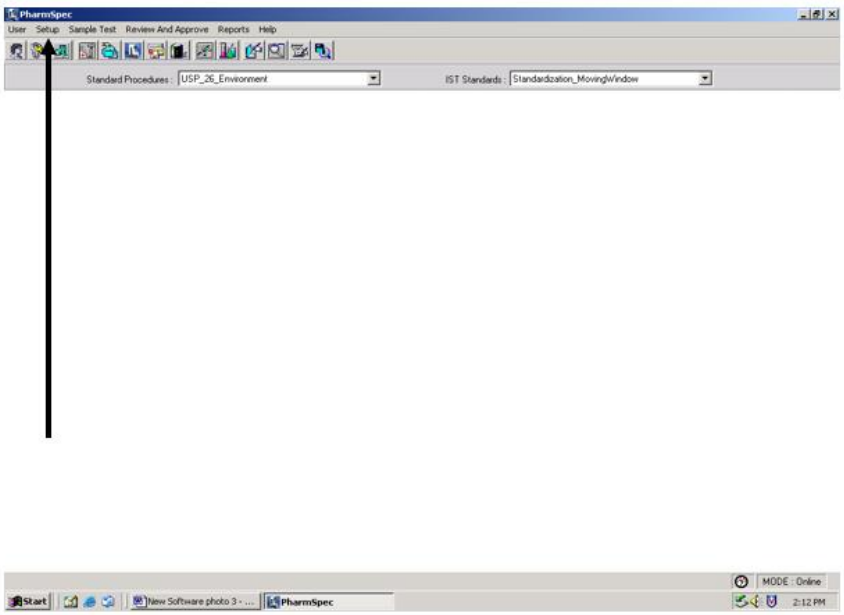
6.3.1.4 **Tare Volume (mL)**

A “Tare Volume” is a volume of fluid that is sampled before the counter begins counting, which ensures that the sample fluid is moving at the desired flow rate and that no liquid meniscus passes through the sensor during counting operations. The “Tare Volume” for routine analyses is set at 1.0 mL per injection; however, a smaller tare volume may be used when the sample volume is extremely small and/or expensive.

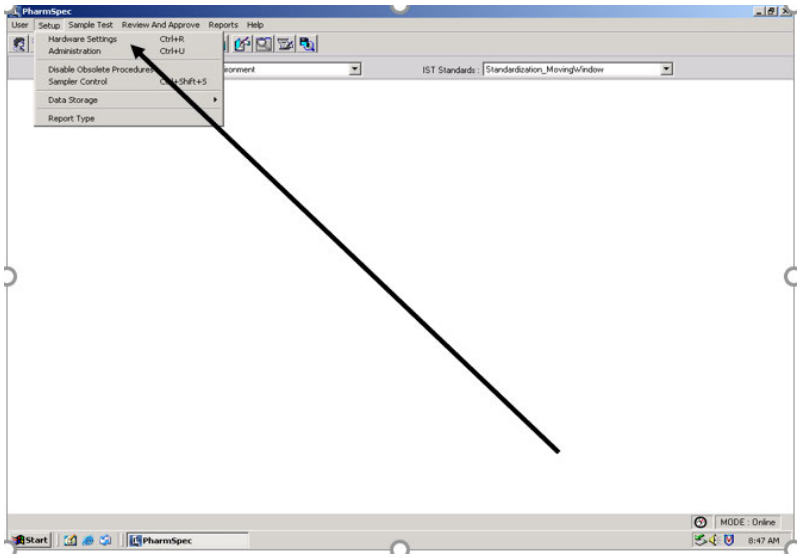
6.3.1.5 **Multi-stroke Tare (mL)**

The “multi stroke tare” is used when sampling volumes larger than the syringe size, which requires multiple strokes of the syringe drive. The “Multi-stroke Tare” for routine analyses is set at 0.10 mL per injection.

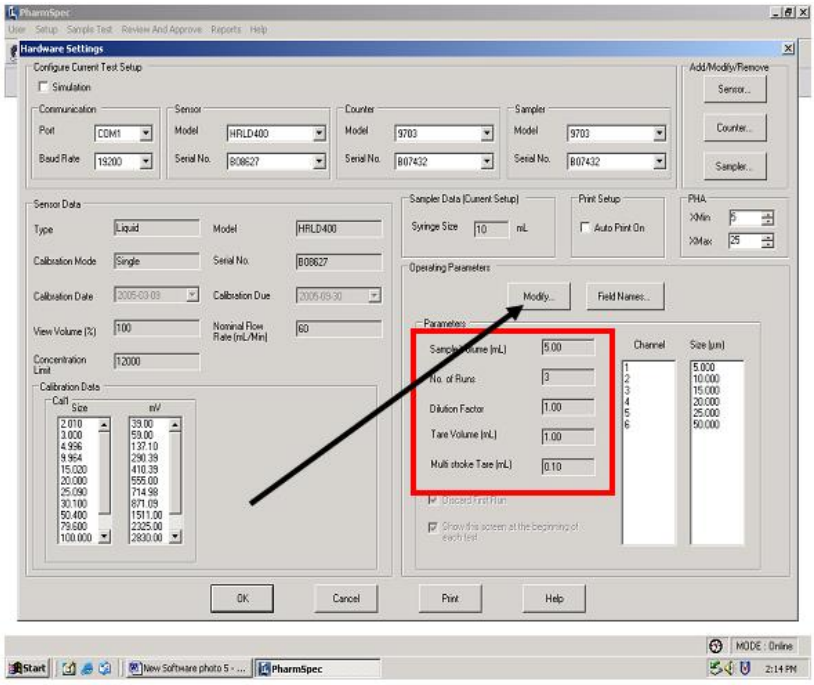
6.3.2 Click on “Setup” on the main menu bar.



6.3.3 Click on the “Hardware Settings.”



6.3.4 To make any necessary adjustments to the “Operating Parameters” (see Section 6-.3.1), click on the “Modify” button.



6.3.5 Enter the corrected/adjusted values into the appropriate fields (highlighted in above diagram).

6.3.6 Click on “OK” to save the changes.

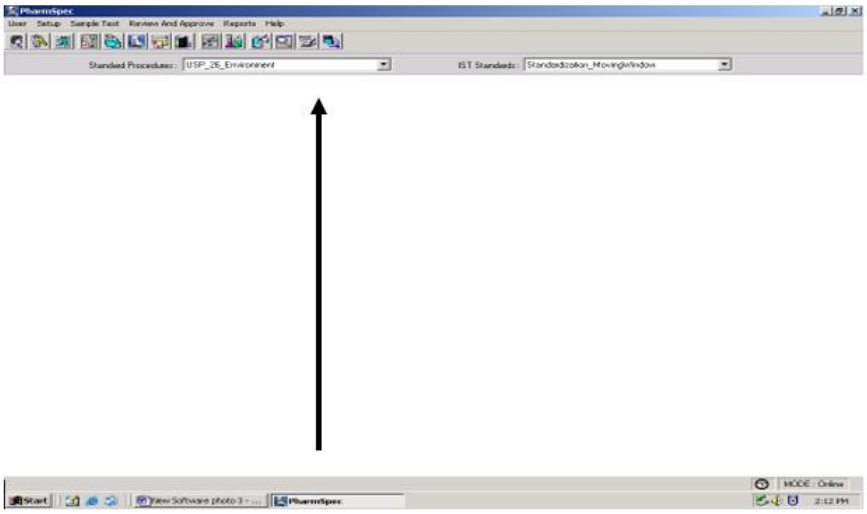
7.0 Running the PharmSpec™ Program

NOTE: All operations under sections 7.1, 7.2, and 7.3 are conducted inside the vertical flow clean bench. The blower for the clean bench must be turned on approximately one hour before initiating the assay and must remain on during the performance of the assay.

7.1 Environmental (Glassware) Test

This test is performed to assure that the glassware used to contain the "test article" is clean and free of possible contaminating particulate matter. The glassware is rinsed thoroughly with Direct-Q water and then inverted to dry before the test is initiated.

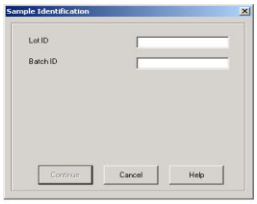
7.1.1 Click on the "Standard Procedure" drop-down menu button located on the PharmSpec™ Home Page.



7.1.2 Select "USP38Environment" from the list of options.

Select "**Run Procedural Test"** from the main menu bar.

7.1.3 A "Sample Identification Screen" will be presented.

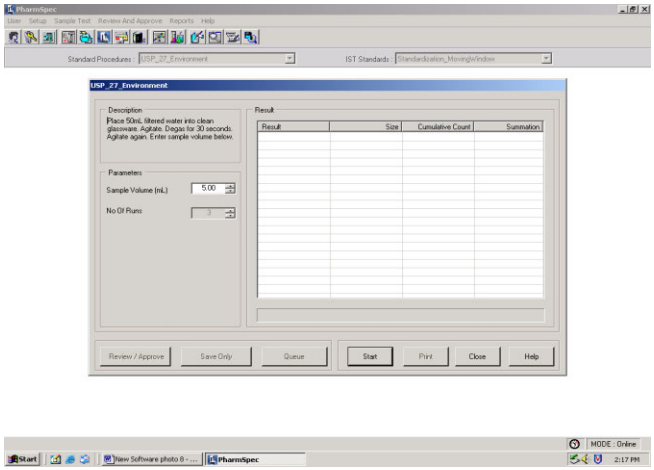


7.1.4 Insert the sample Information:

The default sample information headings are “Lot Number” and “Batch Number.” They cannot be changed or altered by the user. To accommodate for these, enter the “**QC Requisition Number**” in the space designated for “Lot Number” and enter the “**Sample Lot Number**” for which this piece of glassware is to be used in the space designated for “Batch Number”. The user will note that the “Continue” button is initially grayed out and inactive but when the sample information is entered, the “Continue” button will become active and darken in color.

7.1.5 Click “Continue.”

7.1.6 The following screen will be presented.



NOTE: Make sure that the blower for the vertical flow clean bench is on before initiating the test.

7.1.7 Follow the instructions that appear in the upper left corner of the screen:

7.1.7.1 Place a minimum of 50 mL of distilled water in a clean glass beaker that has been thoroughly rinsed with distilled water.

7.1.7.2 Slightly swirl the water and degas for 30 seconds by placing in the beaker in a sonicating waterbath OR allowing the beaker to sit, undisturbed for a minimum of 3 minutes. For maximum effect, do both procedures.

7.1.7.3 Slightly swirl the solution again and place the beaker on the sample platform of the particle counter.

7.1.7.4 If a “Sample Volume” that differs from the default value (5.0 mL) is required, enter that volume in the appropriate space. Note that the number of runs is set at a default value of 3, but if this value needs to be modified, see “Hardware Settings,” section 4.3.

7.1.8 Click on the ”Start” button. The sampler arm will lower and three preset volumes (tare volume + sample volume) will be drawn into the sample syringe, one at a time, then immediately passed through the sensor where particles at 10 micron (µm)m and 25 micron (µm) diameter are counted after the pre-set tare volume has passed through the sensor.

7.1.9 The results will be displayed on the monitor and indicate whether or not the glassware met USP specifications. The first run is discarded as it is used to purge the system. The remaining two runs are averaged and used to determine if the test has met the USP specifications. If the result of the glassware test does not meet USP specifications, then it has to be repeated after additional cleaning of the glassware or selecting a different container.

7.2 Test for Small Volume Injections (SVI)

NOTE: The "SVI" is the option to use for testing "In-process" and "Final" samples of materials being produced by BDP for compliance with USP Standards. In-process "FIO" sample data, with no reference to USP compliance, can be obtained simply by pressing the "Run Counter…. [F9]" button after the appropriate "Sample Information" has been entered.

The SVI is performed for the purpose of counting particulate matter in injectable solutions. The results of the test determine whether or not a solution conforms to the requirements set for that solution. The SVI test is for injections of 100 mL or less that are used over a prolonged period of time as well as drugs that are used continuously or repeatedly (e.g., potassium chloride or other electrolytes used as additives to IV's). The SVI test allows the user to select the number of runs, but requires at least three runs with the first run being discarded and averages the remaining runs. The USP standard specifies the number of containers to be tested.

- If the volume in the container is less than 25 mL, test a solution pool of 10 or more units. This is to ensure that the number of test specimens adequately provides a statistically sound assessment of whether a batch or other large group of units represented by the test specimens meets or exceeds the limits.
- Single small-volume injection units may be tested if the individual unit volume is 25 mL or more.
- In either case, the **total volume tested must be at least 25 mL**.

7.2.1 Go to the USP 38 <788> menu and click on SVI Test.

7.2.2 Press the [F8] key or click on the "Sample ID" button and enter the necessary information in the appropriate fields. Click on OK.

7.2.3 Enter the required information into the appropriate fields on the subsequent screen:

- Number of Runs.
- Number of Containers.
- Total Volume.
- Sample Volume.

7.2.4 Wearing powder-free gloves, place each unit inside the clean bench. Mix each unit by inverting 25 times within 10 seconds. Remove the crimped cap and the label (if necessary) from the sample container. Clean the outside of the sample container with Sartorious Arium Pro. This is to remove any residue from the container, which may introduce particles into the vertical flow clean bench. Leave the sample container in the clean bench and proceed with sample preparation.

7.2.5 Combine the units (if more than one vial is submitted for testing) into a clean beaker or flask that has passed the USP Glassware Test.

7.2.6 Place the beaker or flask onto the sample platform and click on the “Run Counter” or press the [F9] button at the bottom of the screen.

7.2.7 The sampler will draw the designated number of consecutive portions of designated volume of the suspension, discard the data from the first sample, and average the counts of the remaining samples. The PharmSpec™ (program will determine whether the test was compliant with USP 38<788> and print the results accordingly (PASS or FAIL).

7.2.8 Record the results in the system logbook as well as the appropriate QC Test Request.

7.3 Test for Large Volume Injections (LVI)

The LVI test also focuses on counting particulate matter within injectable solutions; however, this test is for solutions of more than 100 mL designated for single-dose infusion. The LVI test allows the user selectable number or runs, but requires at least three runs with the first run being discarded and averages the remaining runs.

7.3.1 Go to the USP 38<788> menu and click on LVI Test.

7.3.2 Press the [F8] key or click on the “Sample ID” button and enter the necessary information in the appropriate fields. Click on OK.

7.3.3 Enter the required information into the appropriate fields on the subsequent screen:

- Number of Runs.
- Sample Volume.

7.3.4 Place the test solution inside the clean bench. Mix the test solution by inverting 25 times within 10 seconds.

7.3.5 Aliquot a sufficient volume of the solution into a clean beaker or flask that has passed the USP Glassware Test.

7.3.6 Place the beaker or flask onto the sample platform and click on the “Run Counter…[F9]” button at the bottom of the screen.

7.3.7 The sampler will draw the designated number of consecutive portions of designated volume of the suspension, discard the data from the first sample, and average the counts of the remaining samples. The PharmSpec (program will determine whether the test was compliant with USP 38 <788> and print the results accordingly (PASS or FAIL).

7.3.8 Record the results in the system logbook as well as the appropriate QC Test Request.

8.0 Data Reporting, Analysis and Review

8.1 Data Recording

8.1.1 The data recorded by the sensor and PharmSpec software is based on the preset Channel Settings and is reported for ”individual” and “averaged” runs as follows.

- Cumulative Counts – the total number of particles counted from any particular channel size to the maximum channel size.
- Differential Counts – the total number of particles counted from any particular channel size to the next larger channel size.
- Cumulative Counts/mL Counts – the total number of particles counted from any particular channel size to the maximum channel size, as above, divided by the sample volume.
- Differential Counts/mL per Channel Counts – the total number of particles counted from any particular channel size to the next larger channel size, as above, divided by the sample volume.

8.1.2 The values for the Differential Counts at the 10 micron (μm) and 25 micron (μm) settings are used to determine the number of particles per container and evaluate compliance with USP guidelines for the Small Volume Injection (SVI) and Large Volume Injection (LVI) Tests as follows.

8.1.2.1 Differential counts at a specific channel setting (10 micron (μm), 25 micron (μm)) divided by the Sample Size (5.00 mL) gives the Differential Counts/mL for that channel setting.

8.1.2.2 The Differential Counts/mL for a specific channel setting multiplied by the Total Volume of pooled-sample tested gives the Total Differential Counts/mL for that channel setting.

8.1.2.3 The Total Differential Counts/mL for a specific channel setting divided by the number of containers used to produce the Total Volume tested gives the Final Counts per Container.

$$\frac{(\text{Cumulative Counts} / \text{Sample Volume})(\text{Total Volume})}{(\text{Number of Containers})(\text{Dilution Factor})}$$

8.2 Data Analysis

8.2.1 The criteria for passing the SVI test are:

- Less than 6000 particles per container of 10 μm size.
- Less than 600 particles per container of 25 μm size.

8.3 The criteria for passing the LVI test are:

- Less than 25 particles per milliliter of 10 μm size.
- Less than 3 particles per milliliter of 25 μm size.

8.4 Data Review

8.4.1 The hard copy printout of the report including the assay result, sample name, technician's name, and test date will be forwarded to a PA/QC Supervisor for review and signature. Attachment 1 is an example of a liquid particulate assay report. An electronic copy of the report is stored on QCPublic/Lab433QC/Particles. An example of file name for a liquid particulate assay report is as follows: QCPublic/Lab433QC/Particles/QCXXXXXX.

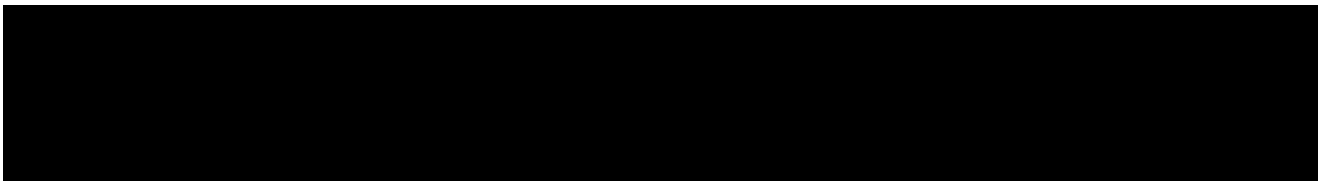
8.4.2 The hard copy printout of the assay results, including the sample name, technician's name, test date, and PA/QC Review will be forwarded to BQA for review and signature.

9.0 Attachment

An Example of Liquid Particle Assay Report

NOTE: The following report format is included as an example. Any modifications deemed necessary should be incorporated in the report for an actual assay. Also, any necessary raw data must accompany the report. The report summary is generated only for lot release or stability studies. No summary report is generated for FIO or in-process samples.

10.0 Change Summary



Attachment 1

Process Analytics Report Summary

QC Test Request Number: TBD

Test Article Name / Lot#: Sample 1, Lot# TBD,
Concentration: N/A, Volume: TBD

Study Title: Operation of the HIAC/Royco Liquid Particle Counter / USP 38 <788> Small Volume for Injection (SVI)

Study Protocol Number(s): SOP Number 22719

Date of Analysis: TBD

Test Result:
Sample 1) TBD particles at 10 μm diameter and TBD particles at 25 μm diameter per container. Sample meets/does not meet the specification for the USP 38 <788> SV1 Test.

Calibration Due Date for HIAC/Royco Liquid Particle Counter: Month / Year

I. **Assay Description**

The HIAC Royco 9703 Particle Counter uses light obscuration sensors to determine the cumulative particulate count in parenteral pharmaceuticals and utilizes PharmSpec™ software to determining compliance with the USP 38<788> Small Volume for Injection (SVI) specifications: ≤ 6000/container at 10 um, ≤ 600/container at 25 um.

II. Procedure

All glassware used in the study was thoroughly rinsed with Sartorios Arium Proand subjected to the USP 38<788> Environmental Test to document that particles were not being introduced by the glassware itself. The samples were pooled with a minimum of 20 mL required for all testing procedures. Three injections of 6 mL (5 mL Sample Volume plus 1 mL tare volume) were passed through the Particle Counter and the number of particles counted. The first injection of each run is treated as a system purge to clear the lines of any previous sample and the data not included in any calculations. The average of the remaining runs is used to calculate the number of particles at 10 and 25 microns in size per container as follows:

Attachment 1 (Continued)

((Cumulative Counts/Sample Volume)(Total Volume))
(Number of Containers)(Dilution Factor)

III. Controls

- **Positive Control(s):** TBD

  Test Result: TBD

- **Negative Control(s):** TBD

  Test Result: TBD

IV. Conclusion / Discussion

Sample 1, Lot# TBD, Concentration: N/A, Volume: TBD
Passed/did not pass the USP 38<788> SVI test.

V. Quality Statement and Approvals

This study has been performed under current Good Manufacturing Practice (cGMP) regulations, as per 21 CFR Part 211, and in accordance with the standard operating procedure(s) listed above. The results obtained have been reviewed for accuracy, completeness, and compliance with established standards. Biopharmaceutical Quality Assurance (BQA) will maintain all original report summaries and associated data for each study performed. Raw data, figures, tables, etc. have been attached to this summary, and meet the requirements for good documentation practices.

Approvals:

_____
Report Prepared By (PA/QC Personnel) Date

_____
PA/QC Supervisor/Director Date

_____
BQA Representative Date

ATTACHMENT 1 (CONTINUED)

USP 38 <788> Environment Test

Comment 1	[redacted]	Sensor Model	: HRLD400
Comment 2	[redacted]	Sensor Serial Number	: B08627
		Operator Name	: palab
		Sample Date	: 9/1/2015 11:20:52 AM
		View Volume (%)	: 100.00
Sample Volume (mL)	: 5	No Of Runs	: 5
Dilution Factor	: 1		
Run1	: Included		

Run No.	Particle Size(µm)	Cumulative Count	Summation
Run 1	10.000	5.00	5.00
Run 2	10.000	4.00	9.00
Run 3	10.000	5.00	14.00
Run 4	10.000	3.00	17.00
Run 5	10.000	2.00	19.00

Pass Criteria

10.000 µm Counts <= 25 per 25 mL

USP 38 <788> Environment Test PASSED

QC Test Request # [redacted]
Page 4 of 5
Initial / Date FA 9.1.15

ATTACHMENT 1 (CONTINUED)

USP 38 <788> Test 1.B

Lot ID	[redacted]	Sensor Model	: HRLD400
Batch ID	[redacted]	Sensor Serial Number	: B08627
		Operator Name	: palab
		Sample Date	: 9/1/2015 11:24:30 AM
		View Volume (%)	: 100.00
Sample Volume (mL)	: 5	No Of Runs	: 4
No. of Containers	: 1	Total Pooled Volume (mL)	: 50
Container Volume (mL)	: 50	Dilution Factor	: 1
Run1	: Discarded		

Run No.	Particle Size(µm)	Cumulative Count	Particles Per Container
Run 1	10.000	10.00	100.00
	25.000	2.00	20.00
Run 2	10.000	8.00	80.00
	25.000	0.00	0.00
Run 3	10.000	15.00	150.00
	25.000	0.00	0.00
Run 4	10.000	3.00	30.00
	25.000	0.00	0.00
Average	10.000	8.67	86.67
	25.000	0.00	0.00

Pass Criteria

10.000 µm Counts <= 6000 per 1 Container

25.000 µm Counts <= 600 per 1 Container

USP 38 <788> Test 1.B PASSED

Number of Particles per container: $\frac{(\text{cumulative count} / \text{sample volume})(\text{Total Volume})}{(\text{Number of containers})(\text{dilution Factor})}$

10µm: $\frac{(8.67/5)(50)}{(1)(1)} = 86.67$ Particles

25µm: $\frac{(0.00/5)(50)}{(1)(1)} = 0.00$ Particle

Passes USP 38 <788> Test 1.B Specifications.

QC Test Request # [redacted]
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