Arthrex®

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To: All Distributors and Agents
Date: 9/7/05

RE: AR-6220 and AR-6421 Extension and Redeuce Tubing Performance (Contamination Control) Validation Test Results

Arthrex has contracted an independent testing facility (Infinity Laboratories, Inc.) to validate the contamination control of fluid when using our Extension and Redeuce Tubing Sets as intended. A copy of the test protocol and corresponding results are attached.

To summarize the results, the study concludes that after exchange of five Extension and Redeuce Tubing Sets with the same Main Pump Tubing Set, fluid contamination is controlled and maintained even in a retrograde fluid pressure environment. They also tested the same Extension and Redeuce Tubing Set five times with a new Main Pump Tubing Set and achieved the same results.

The test protocol called to exchange the Extension and Redeuce Tubing Set following aseptic techniques presently practiced in OR's. All tests were performed using the latest revision of Arthrex Extension and Redeuce Tubing.

Please call with questions or comments.

ATTACHMENTS

Performance (Contamination Control) Validation of Check Valve

Approved:

Infinity Laboratories Lawrence Eldon Date 7/5/05

Arthrex Manufacturing [signature] Date 7/5/05

Quality Assurance [signature] Date 7/5/05

DOCUMENT REVISION HISTORY

Revision Number	Description of Change	Date Effective
Initial Document		07-15-05

1 PURPOSE

The purpose of this document is to detail the procedures for validating the check valve on the pump tubing. The valve should block any reverse flow and prevent contamination of the tubing set. The challenge organism shall be inoculated and forced with pressure into the tubing set. This shall test the check valves ability to prevent contamination at the outflow end of the tubing set.

2 SCOPE

This protocol applies to the following Arthrex part numbers: AR-6410 (Arthroscopy Pump Tubing), AR-6220 (Arthroscopy Extension Tubing), AR-6411 (ReDeuce Tubing System, Pump), AR-6421 (ReDeuce Tubing System, Patient), and AR-6475 (Pump).

3 RESPONSIBILITY

3.1 Laboratory Technicians are responsible for following this procedure and notifying the Laboratory Director or Laboratory Manager of any deviation. Laboratory Technicians shall be responsible for reporting accurate results on all associated documents.

3.2 The Quality Manager is responsible for ensuring that Laboratory Technicians have been adequately trained prior to performing this procedure on test samples.

3.3 The Laboratory Manager is responsible for monitoring the technician's performance and ensuring these procedures are followed accurately.

3.4 The Laboratory Director or Laboratory Manager is responsible for approving the final results of this procedure and all associated procedures (i.e. Quality Control) prior to release of the Final Report to Arthrex.

4 REFERENCES

4.1 **United States Pharmacopeia 28**, NF 23, Official January 1, 2005

4.2 **AAMI TIR 12-1994** Designing, Testing, and Labeling Reusable Medical Devices for Reprocessing in Health Care Facilities: A Guide for Device Manufacturers

5 DEFINITIONS

5.1 **Culture Conditions:** Stated combination of conditions, including the growth medium with the period and temperature of incubation, used to promote germination, outgrowth, and /or multiplication of microorganisms.

5.2 **Sterile:** A state of being free from viable microorganisms.

5.3 **Aerobic Organism:** Microorganism that utilizes oxygen as the final electron acceptor during metabolism.

5.4 **Colony Forming Unit (CFUs):** A visible outgrowth of a population of organisms arising from a single or multiple cells.

5.5 **Growth Promotion:** Test performed to demonstrate that the prepared media will support

microbial growth.

5.6 **Test of sterility:** Test performed to establish the presence or absence of viable microorganisms on product units (or portions thereof) when subjected to defined culture conditions.

5.7 **Positive sterility test:** Sterility test samples which Form detectable microbial growth after incubation.

5.8 **Negative sterility test:** Sterility test samples which do not Form detectable microbial growth after incubation.

5.9 **Validation:** Documented procedure for obtaining, recording, and interpreting the results needed to show that a process will consistently yield a product complying with predetermined specifications.

6 ASSOCIATED DOCUMENTS

6.1 **LOP-121** Quality Management of Prepared Media

6.3 **LOP-120** Quantification of Microorganisms

7 FORMS

7.1 **F-143** Test Worksheet

7.2 **F-152** Quantification of Microorganism worksheet

8 EQUIPMENT

8.1 Instruments, (forceps, scissors, etc., as needed), sterile

8.2 Alcohol burner

8.3 Biohazard hood

8.4 Filter holders, sterile

8.5 Incubator (30-35°C, 28-32°C)

8.6 Vacuum pump with 1-L vacuum flask

8.7 Pressure vessel

9 SUPPLIES

9.1 Trypticase Soy Broth (TSB) prepared by Infinity Labs from dehydrated TSB (BBL or equivalent)

9.2 Alcohol, 95 percent reagent, sterile

9.3 Alcohol, 70 percent reagent, diluted, sterile

9.4 Antimicrobial soap (Fisher or equivalent)

9.5 Tubes or containers, glass, sterile

9.6 *Bacillus atrophaeus*

9.7 Filters, 0.45-µm w/grid, sterile (Gelman GN-6 or equivalent)

9.8 Trypticase Soy Agar (TSA) prepared by Infinity Labs from dehydrated TSA (BBL or equivalent)

10 PROCEDURE

10.1 Set-Up Procedure

10.1.1 Perform all steps of procedure in the biohazard hood.

10.1.1.1 Wipe down the work surface of the biohazard hood with 70 percent reagent alcohol or acceptable substitute, allowing the alcohol to moisten the surface before wiping dry.

10.1.1.2 Turn on the biohazard hood blower, the germicidal lamp, and the deionizing bar, where applicable. After waiting a minimum of five minutes, record that the magnehelic gauge reading is at the proper level. Turn off the germicidal lamp.

10.1.1.3 NEVER work in the hood with the vertical sliding viewing screen open more than 8 inches. THIS RESTRICTED OPENING IS ESSENTIAL FOR PROPER UNIT OPERATION.

10.1.1.4 Whenever bringing equipment or reagents into the hood, wait at least three minutes to purge airborne contamination from the work area.

10.1.1.5 Wash hands with antimicrobial soap before and after using hood. Wear long-sleeved lab coats and gloves to minimize shedding of skin flora into the work area and to protect the hands and arms from contamination.

10.1.1.6 After use, wipe down the hood with 70 percent reagent alcohol or acceptable substitute and turn on the germicidal lamp for a minimum of three minutes.

10.1.2 Media

10.1.2.1 Be certain all media are or have been quality control tested for sterility and growth promotion as specified in the document Quality Management of Prepared Media (LOP-121 or subsequent revision). Document the date of completion on the test worksheet.

10.1.2.3 All liquid media shall be clear and free of precipitation. Do not use cloudy media.

10.1.2.4 Media must be within current expiration date at the time of testing in

order to be acceptable for use.

10.1.2.5 Allow liquid media to come to room temperature before use.

10.1.3 Instruments and Supplies

10.1.3.1 Use sterile instruments at all times when handling test samples and controls.

10.1.3.2 Flame instruments with 95% sterile alcohol initially and between each sample handling.

10.2 Test Procedure

10.2.1 Record sample information (sample description, lot number,) on the test worksheet (F-143). Record any sample irregularity such as moistness, discoloration, or any potential source of contamination.

10.2.2 Suspension Preparation

10.2.2.1 Prepare a suspension of *Bacillus atropheaus* to a concentration of 10^6 per ml. Prepare 1L of the suspension.

10.2.2.2 Verify the number of viable spores in the suspension at time of preparation by inoculating each of two Trypticase Soy Agar (TSA) plates. Plate a dilution of the suspension, if necessary, to get an accurate count.

10.2.2.3 Invert the TSA plates and incubate at 30-35°C for at least 24 hours. Refer to LOP-120 (Quantification of Microorganisms) for additional instructions.

10.2.2.4 Count the number of CFUs on each plate and calculate population by multiplying the average of the two plates by the appropriate factor to determine the number of spores/0.1 ml (original suspension).

10.2.3 Challenge of the Check Valve on the Tubing Set

10.2.3.1 Place the pump and tubing into the biohazard hood. Assemble the pump and tubing as instructed by Arthrex.

10.2.3.2 Prime the Main Set of tubing with sterile WFI. Allow a small amount of water to flow into a collection vessel.

10.2.3.3 Aseptically attach the Extension Set to the Main Set. Turn the pump on to again allow water to flow through the Extension Set into the collection vessel. Take care to not contaminate the end of the Extension Set. Turn the pump off.

10.2.3.4 Once the tubing is primed, place the outflow end of the Extension Set into a sterile collection container. Allow approximately 100 mls of sterile water to flow through the connected tubing sets. Cap the container. Label the container "Negative Control".

10.2.3.5 Using a 22 gauge needle, puncture the Extension Set on the patient side of the check valve. This hole shall allow venting of the tubing when the *Bacillus* suspension is forced into the Extension Set. The Bacillus suspension is now in direct contact of the "back side" of the check valve.

10.2.3.6 Transfer the outflow end of the Extension Set to the pressure vessel containing the *Bacillus* suspension. Place the outflow end into the reservoir. Apply approximately 5-10 p.s.i. of pressure to the reservoir containing the challenge organism in order to fill the Extension Set to the venting hole.

10.2.3.7 Collect 0.1 mls of the *Bacillus* suspension from the venting hole into a sterile collection vessel. Cap the container. Label the container "Positive Control".

10.2.3.8 The punctured hole was then plugged with a pressure patch.

10.2.3.9 Again, apply approximately 5-10 p.s.i. of pressure to the reservoir containing the challenge organism for 10 minutes. Relieve the pressure. Disconnect the Extension Set from the Main Set. Place the outflow end of the Main Set into a sterile collection container. Turn the pump on. Allow approximately 100 mls of sterile water to pass through the main tubing set, collecting the extraction flush in a separate sterile receiving vessel. This vessel shall be capped and labeled Extension Set #1.

10.2.3.10 Turn the pump off. Connect a new Extension Set to the Main Set and repeat the above process until 5 different Extension Sets have been tested. The collected extraction flushes should be labeled Extension Set #2 through #5 appropriately.

10.2.3.11 After 5 Extension Sets have been tested, aseptically replace the Main Set with a new set. Follow the above procedure for a total of 5 flushes using the same Extension Set. Label these collection extraction flushes "Tubing Set #6A" through #6E.

10.2.4 Sterility Test of Extraction

Each extraction volume shall be filtered through a sterile 0.45 μm filter using a vacuum flask and pump. The filter shall be aseptically transferred to 100 mls of sterile TSB and incubated.

10.2.4.1 Assemble the filtering apparatus in the biohazard hood.

10.2.4.1.1 Attach the vacuum hose to the vacuum flask.

10.2.4.1.2 Turn on the vacuum pump.

10.2.4.1.3 Aseptically place a sterile filter holder onto the flask.

10.2.4.1.4 Using sterile forceps, place a 0.45μm filter, grid side up, on the center of the filter screen support. (A ridge around

the base will guide the filter into place). Place the reservoir on top. The reservoir and base will lock into place by magnetic force.

10.2.4.2 Aseptically add the extraction volume into the reservoir of the filter holder. Allow the liquid to pass through the filter.

10.2.4.3 Using sterile forceps, remove the filter and place it into an appropriately labeled container of TSB.

10.2.4.4 Remove the filter assembly from the vacuum flask and replace with a sterile filter assembly for the next sample.

10.3 Culture Conditions

10.3.1 Incubate the samples at 28-32°C for at least 14 days.

10.3.2 Caps on the containers shall be tight to prevent possible outside contamination.

10.4 Test Monitoring

10.4.1 Visually examine the test samples as needed during the incubation period for the presence of microbial growth. Record the results on the test worksheet, including the sample number of any positive sterility test.

10.4.1.1 The development of turbidity, precipitation, and/or surface growth are examples of microbial growth. The use of backlighting can aid in determining the presence of turbidity in the media.

10.4.1.2 Care shall be taken during examination so as to minimize the possibility of false positives. Do not jostle, shake, or open containers of tests in progress.

10.4.2 If growth is observed in any container (including negative controls), subculture and Gram stain the organism as described in procedure LOP-412 (Bacterial Gram Identification).

10.4.3 At Arthrex's request, the status of the sterility test, prior to completion, may be reported. A preliminary report may be communicated if requested.

10.5 Results

10.5.1 The Final Report shall include the results for the check valve performance validation. The number and Gram identification of positive sterility tests shall be reported where possible.

10.5.2 Abbreviations that may be used in the Final Report and on the test worksheet are:

- gpc = gram positive cocci
- gnc = gram negative cocci
- gnr = gram negative rods
- gpsr = gram positive sporogenous rod

- gpr = gram positive rod
- gpc-b = gram positive cocci-bacillus
- gnc-b = gram negative cocci-bacillus

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Attn: Doug Shufelt

Infinity LABORATORIES, INC.

Accession #: IT-29104
Protocol #: WI-1020-PUMP
P.O. # 10273

PERFORMANCE VALIDATION OF CHECK VALVE

SAMPLE DESCRIPTION: ReDeuce Tubing System, Pump (AR-6411) connected to ReDeuce Tubing System, Patient (AR-6421) with Check Valve

QUANTITY SUBMITTED FOR TESTING: 2 Main Sets (AR-6411) , 6 Extension Sets (AR-6421)

PROCEDURE:

A suspension of *Bacillus atropheaus* was prepared to a concentration of 3.5 x 10^6 per ml. The number of viable spores in the suspension was verified at time of preparation by inoculating each of two Trypticase Soy Agar (TSA) plates.

The pump and tubing sets was placed into the biohazard hood. The pump, Main Tubing Set, Extension Tubing Set and WFI reservoir were assembled as specified by Arthrex instructions. The Main Set and the Extension Set were primed with sterile WFI. Once the tubing was primed, the outflow end of the Extension Set was placed into a sterile collection container. Approximately 100 mls of sterile water was allowed to flow through the connected tubing sets as a Negative Control.

The Extension Set was punctured using a sterile 22g needle on the patient side of the check valve to allow venting of the tubing. The WFI water used for priming was allowed to drain out. The outflow end of the Extension Set was connected to a pressure vessel containing the *Bacillus* suspension. Using the pressure vessel, the suspension was forced into the Extension Set. When the *Bacillus* suspension had filled the Extension Set to the venting hole, 0.1 mls was collected as a positive control. The hole was then plugged with a pressure patch. This placed the *Bacillus* suspension in direct contact with the "back side" of the check valve.

Approximately 5-10 p.s.i. of pressure was applied to the vessel containing the challenge suspension for 10 minutes. This pressurized the challenge suspension against the check valve. After 10 minutes, the pressure was relieved from the tubing set, the Extension Set was disconnected from the Main Set, and the outflow end of the Main Set was placed into a sterile collection container. The pump was turned on to allow approximately 100 mls of sterile water to pass through the main tubing set, collecting the extraction flush in a separate sterile receiving vessel. A new Extension Set was connected to the Main Set and the process was repeated for a total of 5 extension sets.

After 5 Extension Sets were tested, the Main Set was aseptically replaced with a new set. The above procedure was repeated for a total of 5 flushes using the same Extension Set.

Each extraction flush (including controls) that was collected from the tubing sets was then filtered onto a sterile 0.45-µm filter. The filters were then immersed in a suitable sterile vessel containing 100 mls soybean casein digest broth (TSB). Media controls consisted of aliquot(s) of TSB. All products and controls were incubated at 28-32°C for at least 14 days. All products and controls were examined for microbial growth after at least 14 days of incubation. Any product demonstrating microbial growth was subcultured onto soybean casein digest agar (TSA) and gram stained.

STERILITY TEST RESULTS:

SAMPLE NUMBER	STERILITY RESULT
Negative Control	No Growth
Main Set #1 + Extension Set #1	No Growth
Main Set #1 + Extension Set #2	No Growth
Main Set #1 + Extension Set #3	No Growth
Main Set #1 + Extension Set #4	No Growth
Main Set #1 + Extension Set #5	No Growth
Main Set #2 + Extension Set #6A	No Growth
Main Set #2 + Extension Set #6B	No Growth
Main Set #2 + Extension Set #6C	No Growth
Main Set #2 + Extension Set #6D	No Growth
Main Set #2 + Extension Set #6E	No Growth
Positive Control	Growth

CONCLUSION:

When tested under the conditions described above, the check valve on the Extension Set (AR-6421) did not allow *Bacillus* to back flow into the Main Set (AR-6411).

Date Initiated: 08-03-05
Date Completed: 08-19-05
Technical Review JKH

Approved Lawrence Eilers
President

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Infinity
LABORATORIES, INC.

Accession #: IT-29103
Protocol #: WI-1020-PUMP
P.O. # 10273

PERFORMANCE VALIDATION OF CHECK VALVE

SAMPLE DESCRIPTION: Arthroscopy Pump Tubing (AR-6410) connected to Arthroscopy Extension Tubing (AR-6220) with Check Valve

QUANTITY SUBMITTED FOR TESTING: 2 Main Sets (AR-6410) , 6 Extension Sets (AR-6220)

PROCEDURE:

A suspension of *Bacillus atropheaus* was prepared to a concentration of 3.5×10^6 per ml. The number of viable spores in the suspension was verified at time of preparation by inoculating each of two Trypticase Soy Agar (TSA) plates.

The pump and tubing sets was placed into the biohazard hood. The pump, Main Tubing Set, Extension Tubing Set and WFI reservoir were assembled as specified by Arthrex instructions. The Main Set and the Extension Set were primed with sterile WFI. Once the tubing was primed, the outflow end of the Extension Set was placed into a sterile collection container. Approximately 100 mls of sterile water was allowed to flow through the connected tubing sets as a Negative Control.

The Extension Set was punctured using a sterile 22g needle on the patient side of the check valve to allow venting of the tubing. The WFI water used for priming was allowed to drain out. The outflow end of the Extension Set was connected to a pressure vessel containing the *Bacillus* suspension. Using the pressure vessel, the suspension was forced into the Extension Set. When the *Bacillus* suspension had filled the Extension Set to the venting hole, 0.1 mls was collected as a positive control. The hole was then plugged with a pressure patch. This placed the *Bacillus* suspension in direct contact with the "back side" of the check valve.

Approximately 5-10 p.s.i. of pressure was applied to the vessel containing the challenge suspension for 10 minutes. This pressurized the challenge suspension against the check valve. After 10 minutes, the pressure was relieved from the tubing set, the Extension Set was disconnected from the Main Set, and the outflow end of the Main Set was placed into a sterile collection container. The pump was turned on to allow approximately 100 mls of sterile water to pass through the main tubing set, collecting the extraction flush in a separate sterile receiving vessel. A new Extension Set was connected to the Main Set and the process was repeated for a total of 5 extension sets.

After 5 Extension Sets were tested, the Main Set was aseptically replaced with a new set. The above procedure was repeated for a total of 5 flushes using the same Extension Set.

Each extraction flush (including controls) that was collected from the tubing sets was then filtered onto a sterile 0.45-µm filter. The filters were then immersed in a suitable sterile vessel containing 100 mls soybean casein digest broth (TSB). Media controls consisted of aliquot(s) of TSB. All products and controls were incubated at 28-32°C for at least 14 days. All products and controls were examined for microbial growth after at least 14 days of incubation. Any product demonstrating microbial growth was subcultured onto soybean casein digest agar (TSA) and gram stained.

STERILITY TEST RESULTS:

SAMPLE NUMBER	STERILITY RESULT
Negative Control	No Growth
Main Set #1 + Extension Set #1	No Growth
Main Set #1 + Extension Set #2	No Growth
Main Set #1 + Extension Set #3	No Growth
Main Set #1 + Extension Set #4	No Growth
Main Set #1 + Extension Set #5	No Growth
Main Set #2 + Extension Set #6A	No Growth
Main Set #2 + Extension Set #6B	No Growth
Main Set #2 + Extension Set #6C	No Growth
Main Set #2 + Extension Set #6D	No Growth
Main Set #2 + Extension Set #6E	No Growth
Positive Control	Growth

CONCLUSION:

When tested under the conditions described above, the check valve on the Extension Set (AR-6220) did not allow *Bacillus* to back flow into the Main Set (AR-6410).

Date Initiated: 08-04-05
Date Completed: 08-19-05
Technical Review _____

Approved _____
President