STUDY SUMMARY

Microbial Ingress Study for ChemoLock® Devices

Report of a study commissioned by ICU Medical Inc. and conducted by AAIPharma Services Corp.

PURPOSE

The purpose of this study was to evaluate microbial ingress on ChemoLock devices following multiple activations over a seven-day period.

METHODS

A protocol was developed and executed by AAIPharma Services using four bacterial strains:

- Staphylococcus aureus ATCC #6538
- Staphylococcus epidermidis ATCC #12228
- Klebsiella pneumoniae ATCC #4352
- Pseudomonas aeruginosa ATCC #9027

Microbial Recovery Studies

- The device was inoculated in duplicate with an appropriate volume of inoculum to yield 1 - 5 x 10^3 colony forming units (CFU).
- The device was allowed to dry for 1 minute.
- After drying, the device was placed in 10 mL of sterile 0.9% saline and vortexed.
- The positive control was prepared by inoculating a 10 mL volume of 0.9% saline with the same inoculum used to inoculate the device.
- 1 mL aliquots were plated to Trypticase Soy Agar (TSA) and incubated at 30-35°C for 1-2 days.
- Acceptance Criteria: Percent recovery should be greater than 70%.

Microbial Ingress Procedure

- Six devices for each organism were inoculated with an appropriate volume of inoculum to yield 1 - 5 x 10^3 colony forming units (CFU) prior to each activation.
- The devices were allowed to dry for 1 minute.
- The septa was wiped vigorously in a circular motion for not less than 3 seconds with a 70% IPA prep pad and allowed to dry.
- The device was activated using a syringe with 10 cc saline. The fluid was collected in a filter funnel and filtered through a 0.45 micron cellulose nitrate filter. The filter was rinsed with 100 mL of 0.9% saline.
- The filter was transferred to a solidified plate of TSA and incubated at 30-35°C for 2-3 days.
- The process was repeated for a total of seventy times (ten activation per day) per device per organism for seven days.
- Two devices were prepared for the positive control by performing the activation and testing procedure without the 70% IPA disinfection step.
- Negative controls were performed.
- Acceptance Criteria: Report results.

RESULTS

The results for the microbial recovery studies are shown in Table 1.

Table 1. Microbial Recovery Studies Results

Test Organism	Recovery (CFU/Device)	Positive Control Recovery Average (CFU)	Percent Recovery	Meets Criteria
Staphylococcus aureus	1.9 x 10^3	2.0 x 10^3	95%	Yes
Staphylococcus epidermidis	2.0 x 10^3	2.0 x 10^3	100%	Yes
Pseudomonas aeruginosa	1.4 x 10^3	1.3 x 10^3	108%	Yes
Klebsiella pneumoniae	1.9 x 10^3	2.4 x 10^3	79%	Yes

The results for the Microbial Ingress are shown in Tables 2 - 5.

There was no microbial recovery on the negative controls. Standard inoculum counts were performed by inoculating 10 mL saline with the same amount of organism used for testing, and 1 mL was plated using the pour plate method.

Table 2. Staphylococcus Aureus Microbial Ingress Results

Activation		Replicate (CFU/filter) 1	2	3	4	5	6	Inoculum Verification (CFU/Device)
DAY 1	1-10	0	0	0	0	0	0	1.1×10^3
DAY 2	11-20	0	0	0	0	0	0	1.7×10^3
DAY 3	21-30	0	0	0	0	0	0	1.4×10^3
DAY 4	31-40	0	0	0	0	0	0	1.2×10^3
DAY 5	41-50	0	0	0	0	0	0	1.2×10^3
DAY 6	51-60	0	0	0	0	0	0	1.2×10^3
DAY 7	61-70	0	0	0	0	0	0	1.1×10^3
Positive Control Replicate 1 (CFU/filter)								39
Positive Control Replicate 2 (CFU/filter)								3

Table 3. Staphylococcus Epidermidis Microbial Ingress Results

Activation		Replicate (CFU/filter) 1	2	3	4	5	6	Inoculum Verification (CFU/Device)
DAY 1	1-10	0	0	0	0	0	0	1.2×10^3
DAY 2	11-20	0	0	0	0	0	0	1.7×10^3
DAY 3	21-30	0	0	0	0	0	0	1.4×10^3
DAY 4	31-40	0	0	0	0	0	0	2.4×10^3
DAY 5	41-50	0	0	0	0	0	0	1.4×10^3
DAY 6	51-60	0	0	0	0	0	0	1.1×10^3
DAY 7	61-70	0	0	0	0	0	0	2.6×10^3
Positive Control Replicate 1 (CFU/filter)								0
Positive Control Replicate 2 (CFU/filter)								3

Table 4. Pseudomonas Aeruginosa Microbial Ingress Results

Activation		Replicate (CFU/filter) 1	2	3	4	5	6	Inoculum Verification (CFU/Device)
DAY 1	1-10	0	0	0	0	0	0	1.9×10^3
DAY 2	11-20	0	0	0	0	0	0	1.5×10^3
DAY 3	21-30	0	0	0	0	0	0	1.9×10^3
DAY 4	31-40	0	0	0	0	0	0	1.7×10^3
DAY 5	41-50	0	0	0	0	0	0	1.7×10^3
DAY 6	51-60	0	0	0	0	0	0	1.2×10^3
DAY 7	61-70	0	0	0	0	0	0	1.1×10^3
Positive Control Replicate 1 (CFU/filter)								6
Positive Control Replicate 2 (CFU/filter)								6

Table 5. Klebsiella pneumoniae Microbial Ingress Results

Activation		Replicate (CFU/filter) 1	2	3	4	5	6	Inoculum Verification (CFU/Device)
DAY 1	1-10	0	0	0	0	0	0	1.3×10^3
DAY 2	11-20	0	0	0	0	0	0	1.1×10^3
DAY 3	21-30	0	0	0	0	0	0	1.1×10^3
DAY 4	31-40	0	0	0	0	0	0	1.5×10^3
DAY 5	41-50	0	0	0	0	0	0	1.3×10^3
DAY 6	51-60	0	0	0	0	0	0	1.1×10^3
DAY 7	61-70	0	0	0	0	0	0	4.2×10^3
Positive Control Replicate 1 (CFU/filter)								8
Positive Control Replicate 2 (CFU/filter)								24

CONCLUSION

Based upon the results of the microbial ingress testing, as reported above, it is determined that the ChemoLock system meets established criteria for microbial recovery studies. Per the device's FDA 510(k) clearance, the ChemoLock system prevents the transfer of environmental contaminants, including bacterial and airborne contaminants into the system.