Assessment of the Efficacy of ENV Services Equipment Bio-Decontamination Service (EBDS) with the BIOQUELL Clarus "S" Vapor Phase Hydrogen Peroxide System to Decontaminate Allentown Caging Equipment BCU Animal Rack.

Abstract: Gaseous disinfection is widely used in microbiology laboratories and vivairiums for two main reasons. First, it is used to ensure the inactivation of potentially harmful bacteria and viruses on surfaces in order to prevent the exposure of laboratory and maintenance staff to infectious agents. The second reason is to ensure a clean working environment to reduce the chances of contamination of the work area with background agents.

In this study Vapor Phase Hydrogen Peroxide (HPV) has been studied for efficacy as a gaseous disinfectant against Geobacillus Stearothermophilus spores on metal carrier strips, with a loading of 10^6 spores per carrier.

Introduction: ENV Services Inc's., EBDS system has been developed to use the BIOQUELL Clarus "S" suite in order to decontaminate biological safety cabinet, incubators, and various laboratory equipment using vapor phase hydrogen peroxide. In this study the system was tested to show its efficacy at decontaminating a BCU Animal Rack with mouse cages using 255ml H_2O_2 fill volume. The test cycle was conducted using Log 6 Geobacillus Stearothermophilus biological indicators to prove the efficacy of the process cycle.

Materials and Methods: The following Allentown Caging Equipment BCU Rack was used:

Allentown Caging Model BCU711598EZ S/N BC-0014-05-00

Biological Indicators (BI's): Apex Laboratories, Log 6 Geobacillus Stearothermophilus (ATCC#12980 Lot# H0017 Expiration 31-Aug-2007).

Trypitcase Soy Broth (TSB) Becton Dickenson 15ml vials, (B21823X, Lot# 6255873, Exp. 3/08/08)

The Clarus "S" suite (ENV Kit #15) was used to generate the HPV and was operated to the following parameters for the test. Plastic sheeting (4 mil) and tape were used to tent the rack for decontamination.

Test A: 255ml of H_2O_2 30% w/w Lot # CG60712B 1307 was vaporized in 140 minutes at 2.59 mls/min with an inclusive dwell time of 40 minutes. During this test the rack was kept at negative pressure (from between –2Pa to –10Pa) by extracting air through the end of the rack tent to a H_2O_2 catalytic converter that is part of the Clarus "S" suite. At the end of the 40-minute dwell time the rack was aerated over night to reduce the H_2O_2 levels below 2ppm and the samples were collected for incubation. If H_2O_2 levels are not below 2ppm after 60 minutes continue to aerate until levels are acceptable.

All work was performed in a laboratory at Allentown Caging Equipment Co. Inc., in Allentown, New Jersey. Incubation of BI's was performed at ENV Services Inc., in Hatfield, Pennsylvania.

One biological indicator (BI) was used for each of the twenty (20) positions within the rack as indicated by photo #1. After the test run the BI's were removed using aseptic technique and place in vials containing 15ml of Trypticase Soy Broth (TSB).

Photo #1: Showing placement of BI's in each corner and middle cages of rack. Same BI placement on both sides of rack. (A1 to G7) & (H1 to N7).

Photo #2: Showing placement of BI's in each exhaust manifold (EX-1 thru EX-7).

The TSB vials were incubated for seven days at 55-60 degrees Celsius and regularly examined for growth. One unexposed BI was used as a positive control in each test run. Refer to tables 1.1 for the BI incubation results.

Table 1.1 BI Incubation Results from Test A: Run 1 of 1

BI Position	Day1 Reading	Day 2 Reading	Day 6 Reading	Day 7 Reading
A1	No Growth	No Growth	No Growth	No Growth
A7	No Growth	No Growth	No Growth	No Growth
D4	No Growth	No Growth	No Growth	No Growth
G1	No Growth	No Growth	No Growth	No Growth
G7	No Growth	No Growth	No Growth	No Growth
H1	No Growth	No Growth	No Growth	No Growth
H7	No Growth	No Growth	No Growth	No Growth
K4	No Growth	No Growth	No Growth	No Growth
N1	No Growth	No Growth	No Growth	No Growth
N7	No Growth	No Growth	No Growth	No Growth
EX-1	No Growth	No Growth	No Growth	No Growth
EX-2	No Growth	No Growth	No Growth	No Growth
EX-3	No Growth	No Growth	No Growth	No Growth
EX-4	No Growth	No Growth	No Growth	No Growth
EX-5	No Growth	No Growth	No Growth	No Growth
EX-6	No Growth	No Growth	No Growth	No Growth
EX-7	No Growth	No Growth	No Growth	No Growth
EXH-V	No Growth	No Growth	No Growth	No Growth
EXH-CL	No Growth	No Growth	No Growth	No Growth
Supply-V	No Growth	No Growth	No Growth	No Growth
Positive Control	Pos Growth	Pos Growth	Pos Growth	Pos Growth

Start date of Incubation: 05-Apr-2007

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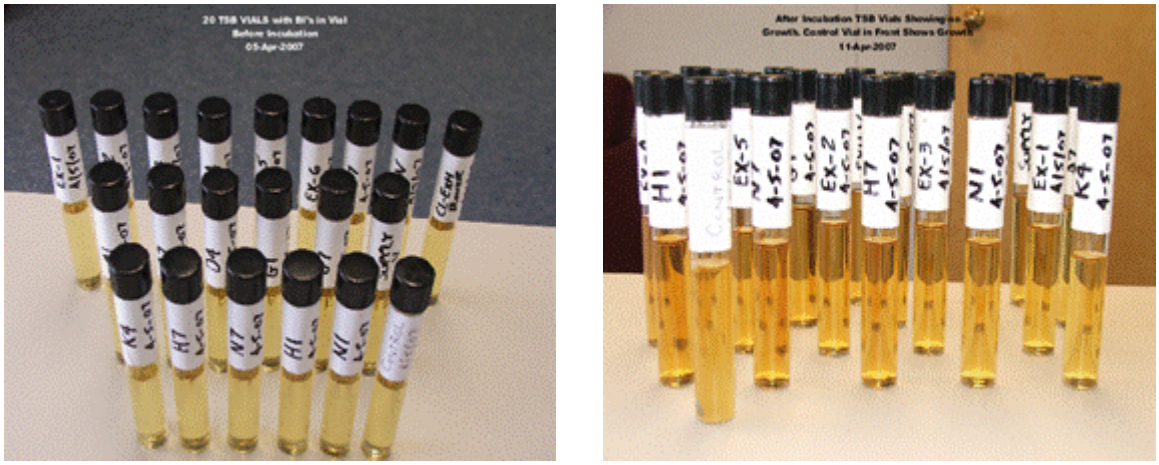

Photo #3: (Lt. 20 TSB vials before incubation Rt. 20 TSB vials after incubation control vial showing growth)

Photo #4: Showing TSB vial on left with no signs of growth. TSB control vial on right showing growth.

Passing Criteria: Biological Indicators (BI's) that have been place within the rack are incubated for seven (7) days. The results of the BI incubation after two (2) and seven (7) days must show no signs of biological growth in all indicators placed within the rack system.

Cycle Optimization: Cycle optimization was not considered for this test run. Cycle run optimization could be achieved with further testing to possibly reduce both the H_2O_2 volume used and the overall cycle run time.

Conclusion: Hydrogen peroxide vapor (H_2O_2) is an excellent choice for microbial bio-decontamination of animal racks and enclosures. Filter media, size and rack design, all have effects on the efficacy results of the decontamination process. The effectiveness of the decontamination by (H_2O_2) can be evaluated easily through the use of biological indicators placed at key locations within the rack. As indicated by the results of this study efficacy of the vapor phase hydrogen peroxide process was successfully achieved. The results equate to a complete bio-decontamination of all surface areas both inside and out of the BCU rack system.