



FINAL REPORT

Determining the Activity of Incorporated Antimicrobial Agents

PROTOCOL
Modified ASTM E2180

ORDER Number
371111495

PREPARED FOR:

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Certificate of Analysis

Client: Gamet S.A.

Contact: Michal Rakowski

Project: ASTM E2180 Bacterial Resistance

Product : Door coatings

EMSL NO: 371111495

Sample received: 9/7/2011

Start date: 9/14/2011

Report date: 9/29/2011

Challenge Bacteria: Gram Positive – *Enterococcus faecalis* ATCC 29212

Experimental Summary: The testing procedure was designed after discussions between EMSL Analytical, the testing company, and the client, Gamet, S.A. The testing procedure is based on ASTM E2180, with the testing conducted on door handle coatings submitted by Gamet for their ability to reduce bacteria. The testing was conducted in our Cinnaminson, NJ Microbiology Laboratory.

Procedure:

E2180:

Culture preparation: *Enterococcus faecalis* (*E. faecalis*) was observed for the determination of antimicrobial efficacy of the different door coatings; brushed, satin and gloss. Cells were first plated onto tryptic soy agar (TSA) and incubated at 35°C for 24 h. A well isolated colony was then taken and placed into 10 mL of tryptic soy broth (TSB) and incubated as stated above. After 24 h incubation 1 mL of the bacterial solution was placed into a separate bottle of 99 mL Agar slurry (0.3% agar and 0.85% NaCl), resulting in a concentration of $\sim 1.0 \times 10^7$ cfu/mL.

Innoculation of Test Material: Individual test material was placed in standard 100 x 15mm Petri dishes and inoculated with 1 mL of the bacterial solution, prepared as stated above. Simultaneously, six control non-coated metal coupons were similarly prepared and inoculated. Three of the control coupons were immediately recovered and plated out as stated below to determine the starting population. Each Petri dish from the test material and control material was then wrapped with plastic wrap, to avoid evaporation (dehydration), and incubated at 35°C for 24 h.



Recovery of Test Organism: Following incubation the entire inoculated test material was removed with pre-sterilized forceps and washed with 10 mL of phosphate buffer in the same Petri dish. The wash was then removed with a pipette and placed into a sterile 15mL centrifuge tube. The wash solution was then vortexed for 30 seconds to mix all bacterial cells into solution, and then serial diluted. Respective dilutions were plated onto Tryptic Soy Agar + 5% Sheep Blood (TSAB) and incubated at 35°C for 24 h before colonies were counted. All tests were completed in triplicate.

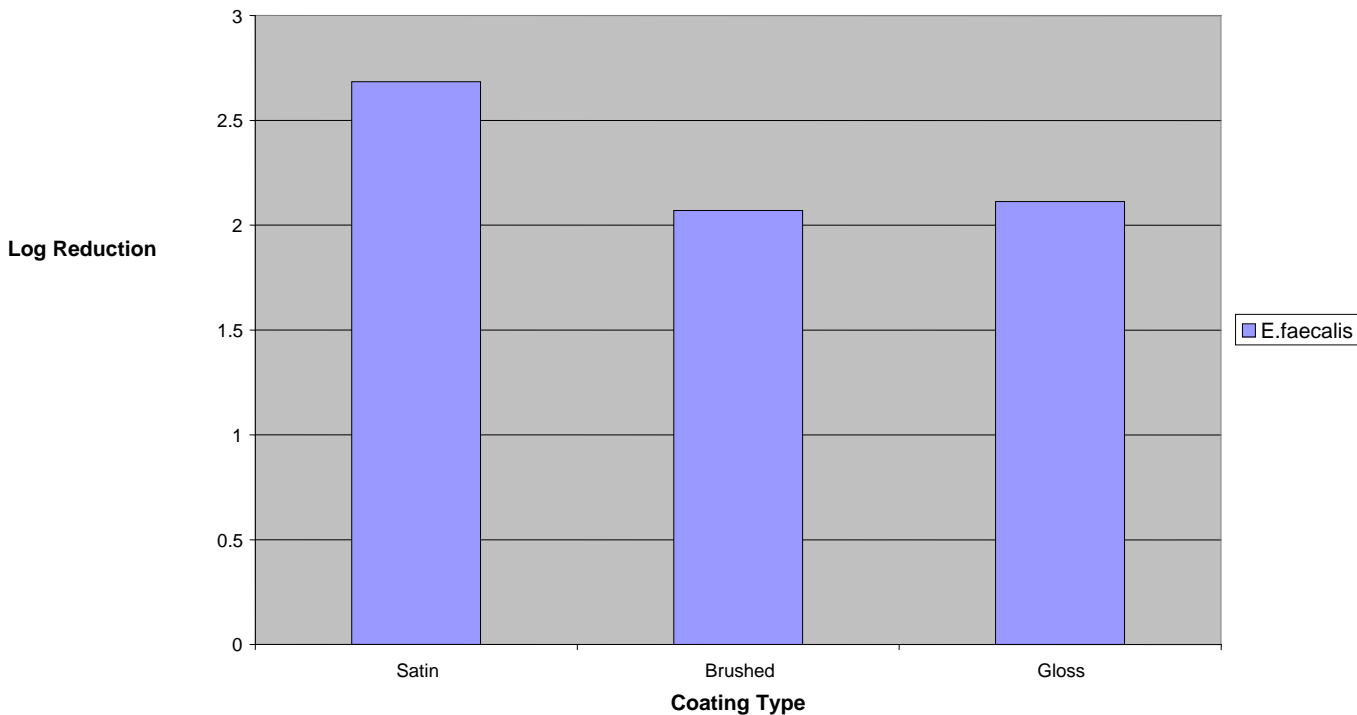
Experimental Results:

Table 1.1

<i>E. faecalis</i>	Time (hours)	Average CFU/mL	Log10/CFU	Log Reduction	% Reduction
Control	0	3.3×10^7	7.53	1.31	95.15%
	24	1.63×10^6	6.21		
Satin Coating	24	6.97×10^4	4.84	2.68	99.79%
	24	2.87×10^5	5.46	2.07	99.15%
Gloss Coating	24	2.60×10^5	5.41	2.11	99.23%

% Reduction = difference between Log10/CFU after 24 h – Log10/CFU after 0 h

Fig. 1.1 Comparison of Log Reduction for Each Type of Coating

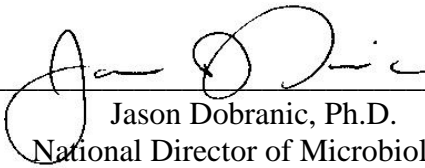




Conclusions/Observations:

The three different door knob coatings sent in by Gamet, S.A. were tested under ASTM E2180 protocol to determine the activity of incorporated antimicrobials against *E. faecalis*. After 24 h contact time it was observed that the satin coating produced a log reduction of 2.68, while the brushed coating demonstrated a log reduction of 2.07, and the gloss coating demonstrated a log reduction of 2.11 (Table 1.1 and Fig. 1.1).

In conclusion, the satin coating demonstrated the greatest ability to disinfect (kill) *E. faecalis* with a percent reduction of 99.79%.



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