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## Antimicrobial Effectiveness of Isocide™ Powder Coating on Esco Biological Safety Cabinets

By: Dian Susanti, Fajar Mustika and Alexander Atmadi

### Abstract

Many undesired bioburden cause the contaminations on equipment and facilities used by lab professionals, whereas it is critical for them to keep everything clean and free from any health hazard. The bioburden commonly presence on many surfaces and it is hard to control their numbers effectively. Esco is presenting laboratory and cleanroom products equipped with organisms' growth control, by coating all of these products with Isocide™ powder coating.

Two types of testing were performed by adopting JIS and ASTM standards to evaluate antimicrobial effectiveness of Isocide™ powder coating. The tests showed that bacteria, yeast and mold, that came into contact with Isocide™ coated surfaces were inhibited or effectively eliminated.

### I. Introduction

For more than 30 years Esco has been manufacturing controlled environment, laboratory and cleanroom equipment solutions that include biological safety cabinets, laminar airflow and clean benches, animal handling workstations, laboratory fume hoods, ovens, incubators, PCR and IVF workstations. These products are widely used in many applications and facilities which are exposed to germs and microorganisms.

There is concern that contact with objects in the above facilities may result in illness aroused from microorganisms such as bacteria, viruses, fungi and higher undesired organisms. It is important to prevent biological contamination on the surface of lab equipment. Certain efforts have been undertaken to produce lab equipment with the ability to kill or inhibit the growth or reproduction of microorganisms which is termed "antimicrobial activity" herein.

There is no standard method published by either the EPA or BPD to determine the efficacy of antimicrobial paint. Many industry groups, such as ASTM, ISO, JIS, etc., publish their own standard methods that are primarily designed to determine the activity of antimicrobial agents in non-porous materials. Below, two testing procedures were developed based on basic principles of JIS and ASTM testing standards, to provide both qualitative and quantitative data relative to Isocide™ antimicrobial activity results.

## II. Materials and methods

**Preparation of microorganism.** Bacteria used for this experiment are *Bacillus subtilis var globigii*, *Serratia marcescens*, *Streptococcus epidermidis*, *Enterococcus faecalis*, *Escherichia coli*, and *Staphylococcus aureus*. While the yeast cultures used are *Saccharomyces cerevisiae*, *Candida albicans*, and *Rhodotularubra*. Bacteria and yeast cultures were prepared by passing 2 activation phases, and their vegetative cells were prepared in phosphate buffer solution at pH 7.3 with 24 hours of growth for bacteria, and 36 hours for yeast cells. *Bacillus subtilis var globigii* and *Aspergillus niger* spore suspension was also used in this experiment with each concentration  $1.5 \times 10^{10}$  spores/ml and  $5 \times 10^8$  spores/ml.

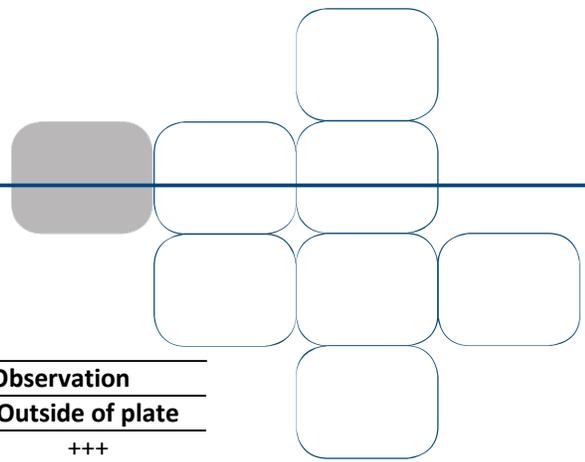
### Testing procedures

#### A. Qualitative Method: Isocide™ surface contact testing

1. Test plates were prepared from 50 x 50 mm electro galvanized steel plates, polished with zinc for rust protection, then sprayed with Isocide™ powder coating and baked in an oven over 180°C. These plates were swabbed with 70% Isopropyl alcohol to remove any visible dirt, left to dry and stored in the dark at 20°C prior to use.
2. All test microorganisms were pipetted with 0.1 ml into agar plates, and spread to the entire agar surface. Test plates were put in the center of inoculated agar. Bacteria cultures were spread into Trypticase soy agar, yeast culture inoculated to Sabouraud dextrose agar, and mold was spread to Potato dextrose agar surface.
3. Growth of test microorganisms was observed 24-48 hrs for bacteria, 48-72 hrs for yeast, while mold was observed until 1 week of incubation period. Any growth in surface contact between agar and test plate was documented.

#### B. Quantitative Method: Determination of Isocide™ log reduction of bioburden

1. Test plates were prepared from 20 x 50 mm electro galvanized steel plates, polished with zinc rust protector, and coated with Isocide™ powder coating and baked over 180°C. Those plates were swabbed with 70% Isopropyl alcohol to remove any visible dirt, left to dry and stored in the dark at 20°C.
2. A stainless pan was prepared to hold the test plates during contact time, and this pan was equipped with organic decontaminator sheet, to remove any contaminant at the back surface of test plates.
3. Test plates were arranged accordingly on top of the decontaminator sheet in the pan, and pipetted with 0.1 ml of test microorganisms, and then the pan was covered with plastic wrap to keep the plates safe from unnecessary materials and contaminants. These plates were put inside incubator without humidity control at 25°C.
4. Tested plates were sampled at 24 and 48 hrs of contact time, the remaining CFU of test microorganisms were quantified by serial dilution method, and log reduction was retrieved by comparing initial concentration with CFU remains in the test plates.



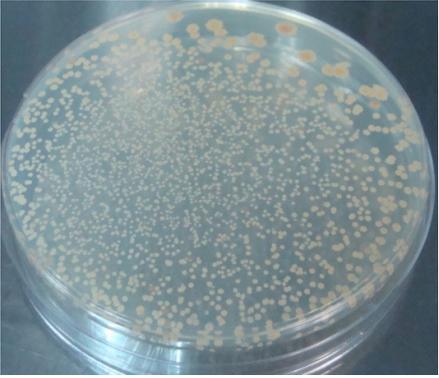
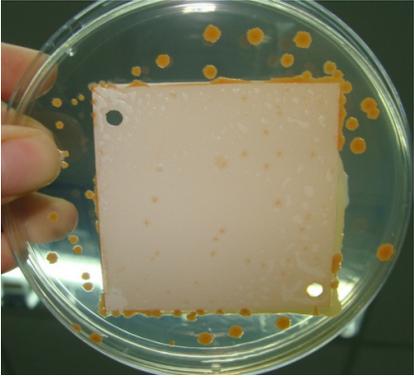
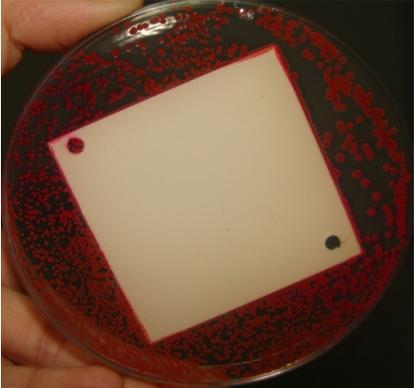
### III. Result

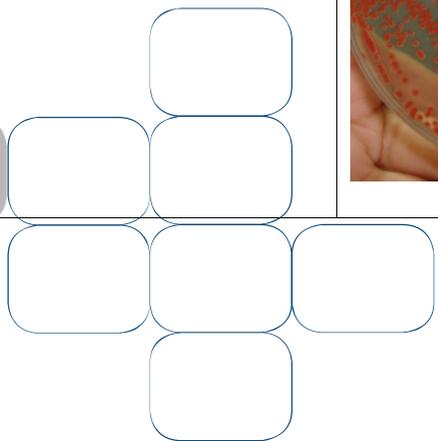
#### A. Qualitative Method:

Microorganisms	Growth Observation	
	Test plate	Outside of plate
<i>Bacillus subtilis</i> var <i>globigii</i>	+	+++
<i>Serratia marcescens</i>	-	+++
<i>Staphylococcus aureus</i>	-	+++
<i>Streptococcus epidermidis</i>	-	+++
<i>Enterococcus faecalis</i>	-	+++
<i>Escherichia coli</i>	-	+++
<i>Saccharomyces cerevisiae</i>	-	+++
<i>Candida albicans</i>	-	+++
<i>Rhodotularubra</i>	-	+++
<i>Aspergillus niger</i>	-	+++

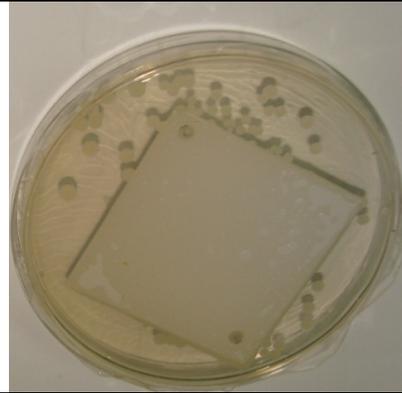
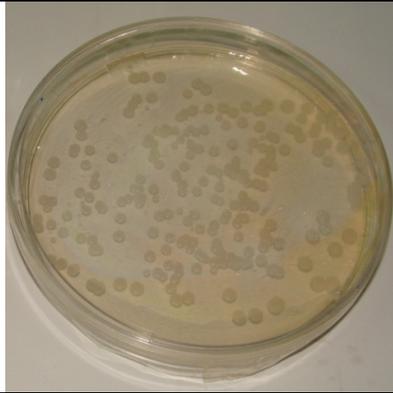
Exposed area : (-) No growth, (+) Some growth  
 Non-exposed / control area : (+++) Full growth

Detailed results are shown in figure below.

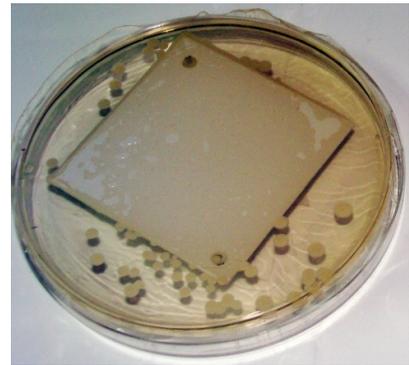
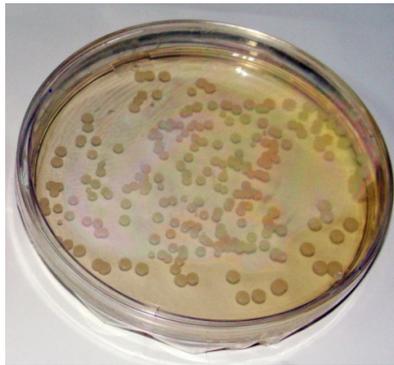
Microorganism	Agar Control	Agar with plate
<i>Bacillus subtilis</i> var <i>globigii</i>		
<i>Serratia marcescens</i>		



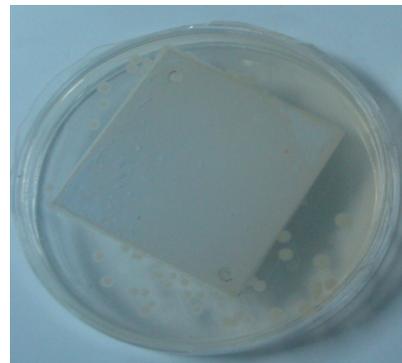
*Staphylococcus aureus*



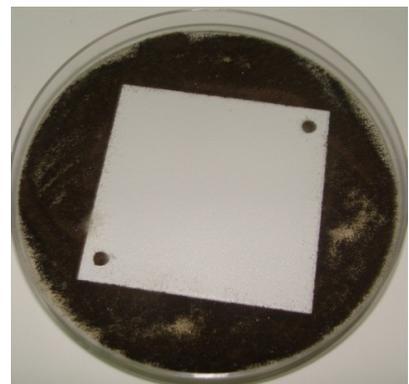
*Enterococcus faecalis*



*Escherichia coli*



*Aspergillus niger*



## B. Quantitative Method:

Table 2 Microbial log reduction after incubation with Isocide™ powder coating

Microorganisms	Initial concentration (CFU/ml)	Cells' logarithmic value	24 hr		48hr	
			CFU remain	Log reduction	CFU remain	Log reduction
<i>Bacillus subtilis var globigii</i>	$3.0 \times 10^5$	5.48	57	3.70	36	3.90
<i>Serratia marcescens</i>	$2.4 \times 10^5$	5.38	0	5.38	0	5.38
<i>Staphylococcus aureus</i>	$3.3 \times 10^5$	5.52	45	3.87	17	4.29
<i>Streptococcus epidermidis</i>	$1.2 \times 10^5$	5.08	40	3.48	15	3.90
<i>Enterococcus faecalis</i>	$1.6 \times 10^5$	5.20	0	5.20	0	5.20
<i>Escherichia coli</i>	$1.3 \times 10^5$	5.11	5	4.50	0	5.11
<i>Saccharomyces cerevisiae</i>	$9.0 \times 10^4$	4.95	10	3.95	0	4.95
<i>Candida albicans</i>	$1.0 \times 10^4$	4.00	50	2.30	12	2.90
<i>Rhodotularubra</i>	$4.0 \times 10^4$	4.60	90	2.65	30	3.12

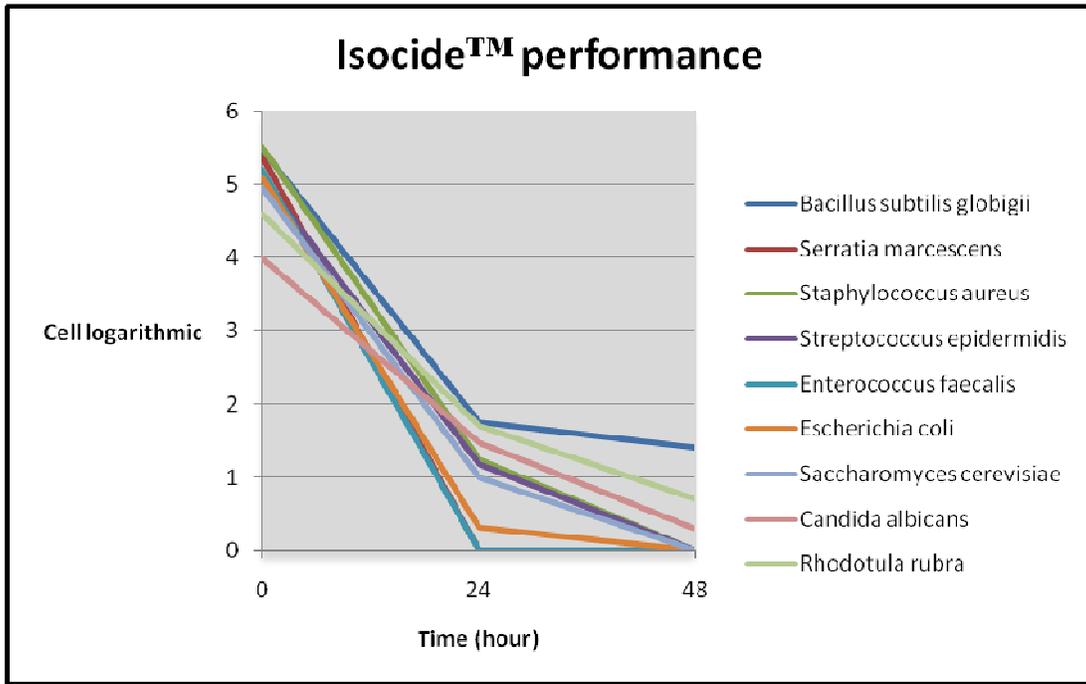


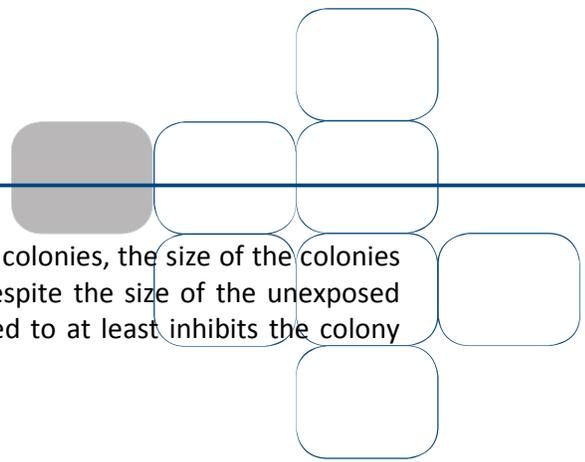
Figure 2 Isocide™ performance between different microorganisms

## IV. Observation

1. The qualitative and quantitative tests indicate that all challenge microorganisms experienced growth reduction or completely eliminated, after being exposed to Isocide™ powder coat for 24 – 48 hours.
2. Isocide™ performance varies, dependent upon type of microorganism. Best Isocide™ performance was shown by subduing *Serratia marcescens* colonies with 5.38 log reduction (no single colony growth) and *Enterococcus faecalis* with 5.20 log reduction after 24 hours incubation.



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3. Despite there is limited log reduction for *Bacillus subtilis* colonies, the size of the colonies that are exposed to Isocide™ experienced growth halt, despite the size of the unexposed colonies became bigger. This shows that Isocide™ managed to at least inhibits the colony growth.

## V. Conclusion

Esco Isocide™ powder coating was found to inhibit or even eliminate the microorganisms that it came in contact with, after 24 – 48 hours. The effectiveness of microbial elimination varies with the type of challenge microorganisms. Despite decontamination using formalin, chlorine dioxide, or hydrogen peroxide is still needed by the service personnel before accessing the contaminated areas, the Isocide™ powder coat reduces the bio burden, which translates to improved safety for the lab and service personnel, and better protection for products being worked inside Esco biosafety cabinets.

## VI. References

Barret, L., Emerentiana, S., Darrel, S. 2007. Antimicrobial Coating. <http://www.battelle.org/forecast/home.stm>.

Tesh, E.M. The Regulation of Antimicrobials in Paints and Surface Treatments. Paint and Coatings Industry 2004, 7

Sadasivan, L. 2007. Antimicrobial coating. <http://www.battelle.org/forecast/driver.stm>. Siva Microbiological Solutions LLC, Bristol, PA.

