MICROCHEM LABORATORY

STUDY REPORT

Study Title

Ability of Brock USA, LLC Test Substances to Resist Bacterial Growth

Test Method

ASTM International Method G22
Standard Practice for Determining Resistance of Plastics to Bacteria

Study Identification Number

NG9149

Study Sponsor

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Test Facility

Microchem Laboratory
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Testing Performed By: C. Craney

ASTM G22: General Information

ASTM International, formerly the American Society for Testing and Materials (ASTM), is an internationally recognized organization that develops and publishes product and testing standards. ASTM G22 is a qualitative test method designed to assess the ability of plastics to resist bacterial attack. The method is typically conducted over a 21 day period, where treated materials are placed on inoculated agar, incubated, then compared to untreated controls at intervals. The untreated controls serve as references for bacterial resistance. A diverse array of bacterial species are used in this method, so it is considered to be a good indicator of bacterial resistance in a variety of environments.

Study Timeline

20JUL2017 21JUL2017 18AUG2017 31AUG2017

Test Substance Information

The test substance was received on 31 MAY 2017.

Test Substances Received: PowerBase – molded EPP
ShockPad "SP Series" - EPP composite

Test Substances arrived in dimensions that were optimal for the conduct of the Study. Test substances were not cut down to ideal sizes for the Study.

Samples as received.

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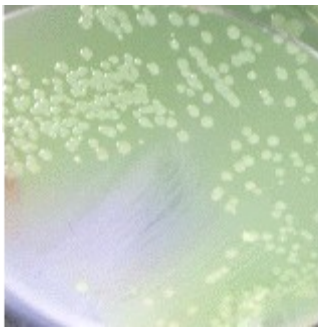
Test Microorganism Information

The test microorganism selected for this test:



Pseudomonas aeruginosa 15442

This bacteria is a Gram-negative, rod-shaped microorganism with a single flagellum. It grows optimally under aerobic conditions, however, it can use a host of electron receptors to respire anaerobically. *P. aeruginosa* can be found almost anywhere in nature and it is an opportunistic pathogen. Like many other bacterial-related diseases, the ability to form resilient biofilms within human tissues under anaerobic conditions is thought to be the primary cause for pathogenicity.

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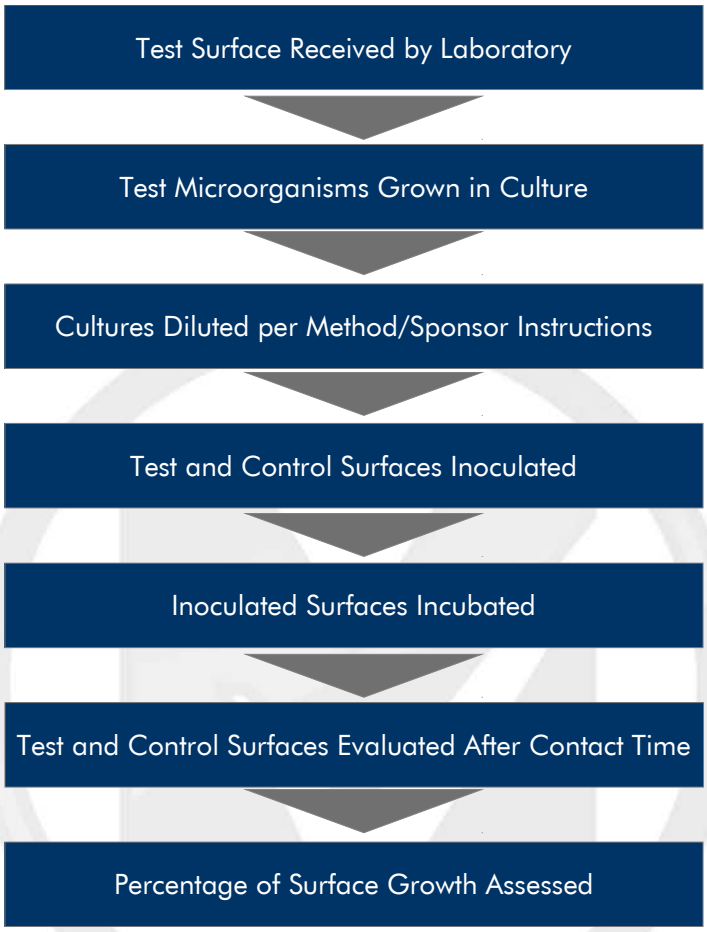
Diagram of the Procedure



Summary of the Procedure

- The test microorganism is prepared, usually by growth in a liquid culture medium.
- The suspensions of test microorganism is standardized by dilution in a buffered saline solution.
- Control and test surfaces are inoculated by placing them on agar which has been inoculated with the diluted test microorganism and allowed to solidify.
- Inoculated test and control substances are placed in a sealed, humid environment and incubated for the predetermined contact times.
- At the conclusion of each contact time, visual assessments of each sample are made, noting if bacterial growth is observed on the surfaces of both test and control substances.

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Criteria for Scientific Defensibility of an ASTM G22 Study

For Microchem Laboratory to consider an ASTM G22 study to be scientifically defensible, the following criteria must be met:

1. The average number of viable bacteria found in the inoculated agar shall be approximately 5×10^4.
2. Positive/Growth controls must demonstrate growth of the test microorganism.
3. Negative/Purity controls must demonstrate no growth of the test microorganism.

Passing Criteria

ASTM does not specify a performance criteria, therefore it may be established by the Study Sponsor.

Testing Parameters used in this Study

Test Substance:	See Page 3	Test Substance Size:	2 in x 2 in
Control Substance:	Viablilty Control	Control Substance Size:	N/A
Replicates:	Single		

Culture Growth Media:	Tryptic Soy Broth	Culture Growth Time:	18-24 Hours
Culture Suspension Media:	0.9% Saline Solution	Inoculum Application:	Inoculated Agar
Inoculum Concentration:	1.5×10^5 CFU/ml	Test Plating Media:	Mineral Salts Agar
Observation Times:	28 days	Contact Temp.:	36°C ± 1°C

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Study Modifications

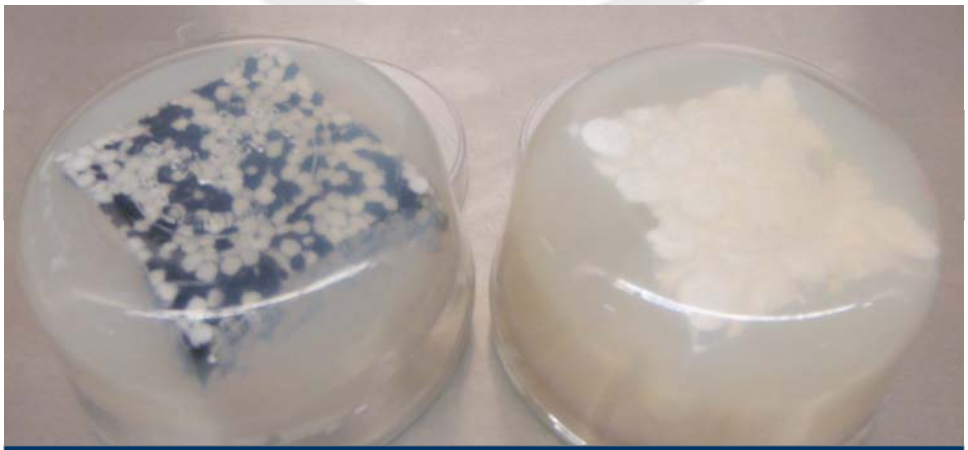
No further modifications were made to the method for this study.

Study Notes

Per the method, a positive control/viability control was performed on Tryptic Soy Agar at the time of initiation. After 48 hours of incubation, the inoculum was confirmed to be viable and pure.
The ASTM G22 Procedure B was used in this study. Procedure B provides total immersion of test substances in the *Pseudomonas aeruginosa* ATCC 15442 inoculated Mineral Salt Agar solution. This procedure provides intimate contact between the bacteria and the test substances on all sides.

Study Photographs-Day 28

Negative Control



Test Sample ShockPad "SP Series" on the left, PowerBase to the right. Both show No Growth on the underside.

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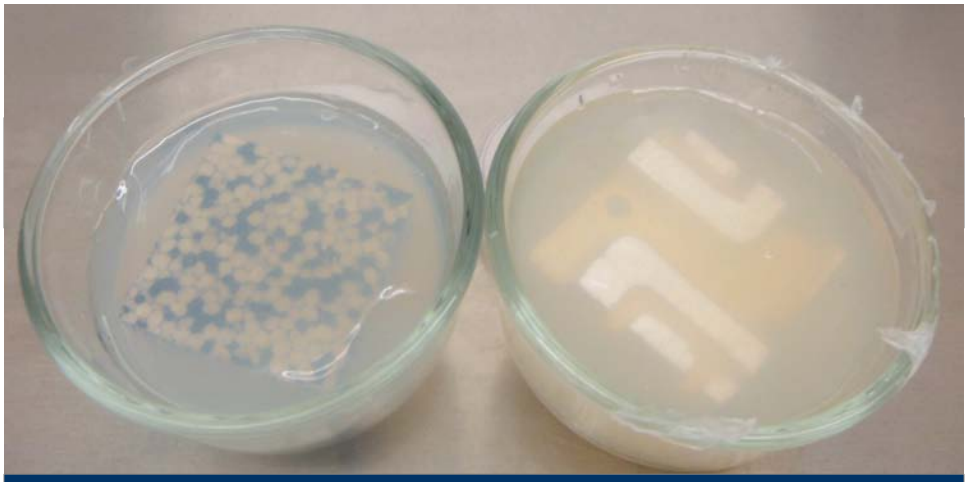
Study Photographs-Day 28



Test Sample ShockPad "SP Series" on the left, PowerBase to the right. Both show No Growth on the top side.

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Control Results

Neutralizer: N/A
Media Sterility: Sterile
Growth Confirmation: Viable & Pure

Calculations

No calculations are made for this study.

Observations of growth are classified as growth or no growth.

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Results of the Study

Sample	Incubation Time and Growth Score at Day 28
Microchem Negative Control	No Growth
PowerBase	No Growth
ShockPad	No Growth

The results of this study apply to the tested substances(s) only. Extrapolation of findings to related materials is the responsibility of the Sponsor.