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Microbial Control

Laboratory Report

**The Resistance of RENOLIT AG Semi-Rigid PVC to  
Microorganisms**

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Confidentiality Agreement:

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## **Introduction**

RENOLIT AG manufactures semi-rigid PVC films for decoration. Three films were submitted containing various dosage levels of VINYZENE™ IT 4010 DIDP (DCOIT<sup>1</sup> active ingredient). Bacterial resistance (*Staphylococcus aureus*), pink stain testing, and mixed fungal testing were performed on the three submitted samples.

## **Summary**

The bacterial resistance testing showed that only Sample 1 had no zone of inhibition and had growth on the contact area. The pink stain test showed that only Sample 1 had no zone of inhibition and moderate staining. Sample 1 supported fungal growth in mixed fungal testing. Samples 2 and 3 resisted growth and staining throughout bacterial resistance, pink stain, and mixed fungal testing. It is recommended that RENOLIT AG semi-rigid PVC film for decoration is treated with 1.2% (1200 ppm) VINYZENE IT 4010 DIDP.

## **Experimental Methods**

### **Bacterial Resistance (qualitative)**

#### **Kirby-Bauer Method**

The samples were placed on nutrient agar inoculated with:

*Staphylococcus aureus*      ATCC 6538

After 24 hours of incubation at 37°C, antibacterial activity was evaluated by measuring (in mm) the size of a clear zone of no growth (Zone of Inhibition) around each sample, and visually determining growth in the contact area.

Bacterial growth is rated by the following scale:

#### **No Growth Contact Area (NGCA)**

This is a designation frequently used in bacterial tests. Bacterial organisms are difficult to determine on the sample itself, so the area immediately under the sample is examined for growth. This is usually a passing designation and indicates that there were no bacterial colonies found under the sample.

#### **Growth Contact Area (GCA)**

This indicates failure of the sample since colonies of bacteria are detected immediately under the sample in contact with the same.

## **ASTM E-1428-91**

Standard Test Method for Evaluating the Performance of Antimicrobials in or on Polymeric Solids Against Staining by *Streptovorticillium reticulum* (A Pink Stain Organism).

The samples were placed on nutrient agar inoculated with the pink staining organism, *Stv. reticulum* ATCC 25607. After 14 days of incubation at 28°C, antimicrobial activity was evaluated by visually rating the degree of stain.

Surface stain is rated by the following scale.

<u>ASTM Rating</u>		
No Stain	(NS)	0
Trace of Stain	(TS)	1
Light Stain	(LS)	2
Moderate Stain	(MS)	3
Heavy Stain	(HS)	4

## **MIXED FUNGAL TEST**

### **Method for Determining Resistance of Polymeric Materials to Fungi**

The samples are placed on non-nutrient agar and inoculated with an aqueous mixed fungal spore suspension of:

<i>Aspergillus niger</i>	ATCC 9642
<i>Aureobasidium pullulans</i>	ATCC 9348
<i>Cladosporium cladosporioides</i>	ATCC 16022

After 28 days incubation at 28°C, antifungal activity is evaluated by visually rating the degree of fungal growth.

Surface fungal growth is rated by the following scale:

No Growth	(NG)
Traces of Growth (less than 10% coverage)	(TG)
Light Growth (10 to 30% coverage)	(LG)
Moderate Growth (30 to 60% coverage)	(MG)
Heavy Growth (60% to complete coverage)	(HG)

## Results

The results of bacterial resistance, pink stain, and mixed fungal testing on the semi-rigid PVC samples are below.

### RENOLIT AG Semi-Rigid PVC Film Testing Results

SAMPLE	ZONE OF INHIBITION (mm) / GROWTH OR STAIN		MIXED FUNGAL GROWTH
	<i>Staphylococcus aureus</i>	PINK STAIN TEST	
Sample 1	0 / GCA	0 / MS	HG
Sample 2	1 / NGCA	0.25 / NS	NG
Sample 3	3 / NGCA	0.5 / NS	NG

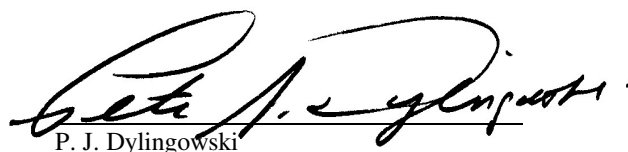
The bacterial resistance testing showed that only Sample 1 had no zone of inhibition and had growth on the contact area. The pink stain test showed that only Sample 1 had no zone of inhibition and moderate staining. Sample 1 supported fungal growth in mixed fungal testing. Samples 2 and 3 resisted growth and staining throughout bacterial resistance, pink stain, and mixed fungal testing.

## **Conclusions/Recommendations**

Samples 2 and 3 resisted growth throughout testing. It is recommended that RENOLIT AG semi-rigid PVC film for decoration is treated with 1.2% (1200 ppm) VINYZENE IT 4010 DIDP.



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Microbiologist



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DCOIT<sup>1</sup> - 4,5-dichloro-2-(n-octyl)-4-isothiazolin-3-one

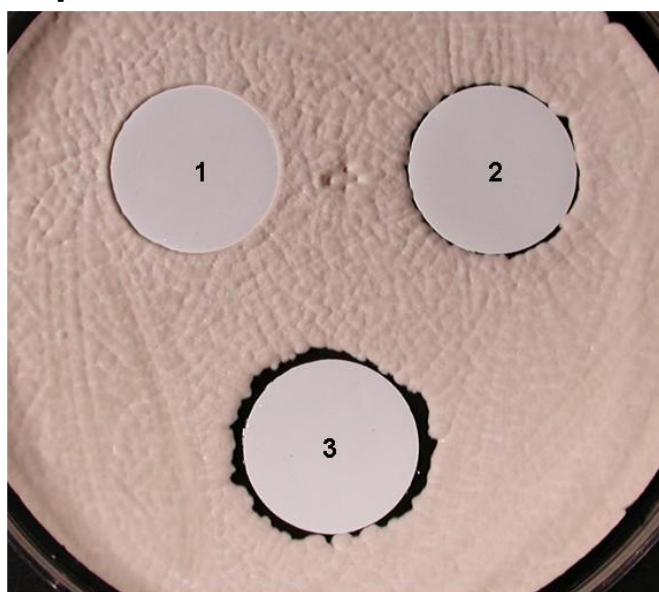
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TEST NO. 09-068  
BACTERIAL RESISTANCE  
*Staphylococcus aureus*



TEST NO. 09-068  
PINK STAIN TEST  
*Streptoverticillium reticulum*



# TEST NO. 09-068 FUNGAL TEST MIXED FUNGAL TESTING

