

# Antimicrobial Test Laboratories

## Fast, Reliable Antimicrobial Efficacy Testing

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Study ID: NG1380

Client: Microgen Inc.

Protocol Reference: External

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### **FINAL RESEARCH STUDY REPORT – NON-GLP**

#### Study Title

Determination of the effect of D-125 (US REG. 61178-1) on peanut allergen using a peanut allergen specific Enzyme-Linked ImmunoSorbent Assay (ELISA).

#### Product Identity

Microgen D-125 Disinfectant

#### Author

Jason Williams, B.S.  
Study Director, Antimicrobial Test Laboratories

#### Study Completion Date

03/09/2009

#### Testing Facility

Antimicrobial Test Laboratories  
3000 Joe DiMaggio Blvd., Suite #32  
Round Rock, Texas 78665

#### ELISA Kit Manufacturer

Neogen Corporation  
ALERT® Peanut Allergen Screening Test<sup>1</sup>  
620 Leshar Place  
Lansing, MI 48912

#### Antimicrobial Test Laboratories Study ID#

NG1380

#### Conclusion in Brief

Peanut allergen levels, as determined by the Neogen ALERT® Peanut Allergen Screening Test, were reduced approximately ten-fold (~90% reduction) after a ten minute contact with D-125. Reductions were calculated relative to a water control that received the same concentration and volume of peanut allergen for the same period of contact. The reduction in peanut allergen (~90%) observed in this preliminary study is of approximately the same magnitude as reported for common leading “anti-allergen” spray products.<sup>2</sup>

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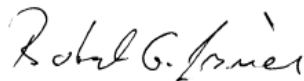
Study ID: NG1380

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### STUDY PERSONNEL

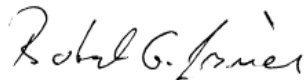
SUBMITTER: Microgen Inc.



Robert Prince  
President

DATE: 03/09/2009

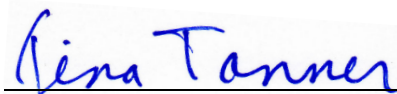
SPONSOR: Microgen Inc.



Robert Prince  
President

DATE: 03/09/2009

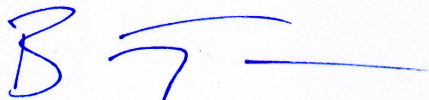
STUDY ADMINISTRATIVE MANAGER:



Gina Tanner  
Business Manager

DATE: 03/09/2009

STUDY DIRECTOR:



Benjamin D. Tanner, Ph.D.  
Scientific Director

DATE: 03/09/2009

ASSISTING LABORATORY PERSONNEL: S. Nalley, J. Williams.

# Antimicrobial Test Laboratories

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### STATEMENT OF NO DATA CONFIDENTIALITY CLAIMS

No claim of confidentiality is made for any information contained in this study.

Company: Microgen Inc.

Agent: Robert Prince

Title: President

Date: 03/09/2009

Signature: 

# Antimicrobial Test Laboratories

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### **LABORATORY QUALITY ASSURANCE STATEMENT**

This study was performed in accordance with Antimicrobial Test Laboratories Standard Operating Procedure (SOP) #050, Methods and Procedures for Laboratory Quality Control.

Antimicrobial Test Laboratories SOP 050 specifies the following:

- Laboratory equipment and devices are verified to function properly and calibrated internally or externally as appropriate to ensure experimental quality.
- Each experiment is evaluated relative to rigorous in-process experimental controls
  - o Media sterility controls (negative controls)
  - o Vehicle (carrier) sterility controls
  - o Media growth controls (positive controls)
  - o Neutralization of active ingredient controls
  - o Verification of positive cultures as target organism
- Review by Antimicrobial Test Laboratories' Scientific Director, Benjamin Tanner, Ph.D. for scientific clarity, accuracy, and completeness.

*This study is exempt from 40 CFR Part 160. Per sponsor communication, data not intended to support a United States antimicrobial pesticide registration.*

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### **FINAL RESEARCH STUDY REPORT – NON-GLP**

#### Study Title

Determination of the effect of D-125 (US REG. 61178-1) on peanut allergen using a qualitative peanut allergen specific Enzyme-Linked ImmunoSorbent Assay (ELISA).

#### Study Number

NG1380

#### Study Sponsor

Bob Prince  
Microgen  
33 Clinton Road, Suite 102  
Clinton Square Executive Center  
West Caldwell, NJ 07006 USA

#### Test Facility

Antimicrobial Test Laboratories  
3000 Joe DiMaggio Blvd., Suite 32  
Round Rock, TX 78665 USA

#### Test Manufacturer

ALERT® Peanut Allergen Screening Test  
Neogen Corporation  
620 Leshar Place  
Lansing, MI 48912

#### Study Objective

To investigate whether or not D-125 causes a reduction in detectable levels of peanut allergen after a contact time of 10 minutes.

#### Technical Note

Approximations of the percent allergen reduction were estimated from a qualitative allergen test kit, calibrated with an allergen dilution series. Due to the nature of the test kit, it was not necessary to control for denaturation of kit components by the test substance. Therefore, a separate study was conducted to evaluate the impact of D-125 alone on denaturation of kit components. That study showed that the D-125 had no negative effect on the kit itself.

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### Key to Abbreviations Used in the Report

ELISA = Enzyme-Linked ImmunoSorbent Assay

ml = milliliter

N/A = Not Applicable

10X = Ten times more concentrated than the working stock

# Antimicrobial Test Laboratories

## Fast, Reliable Antimicrobial Efficacy Testing

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Study ID: NG1380

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### Summary of the Test Method

Based on: External Protocol Provided by the Manufacturer of the ALERT® Peanut Allergen Screening Test, Neogen Corporation Product #8431

*Note: All reagents used in this assay were prepared according to the manufacturer's protocol unless otherwise noted.*

#### Summary of the Test Procedure:

- Peanut allergen was extracted from unsalted roasted peanuts according to the manufacturer's protocol, and diluted 1:1000 in the provided buffer solution to a concentration of ~10X that of the manufacturer's positive control (empirically determined).
- 0.090ml of test product was added to two 1.5 ml tubes (one for the addition of peanut allergen "test" and one for the addition of buffer solution only "control")
- 0.010ml of a 1:1000 dilution of peanut allergen extract was added to the "test" tube for a contact time of 10 minutes at ambient temperature (23°C +/- 1°C), followed by the addition of 0.010ml of the same dilution to 0.090ml water for use as a positive control.
- The entire 0.100 ml reaction volume was added to the appropriate well after 10 minutes had elapsed, followed by the addition of 0.1ml of the manufacturer provided positive control sample containing 200ng/ml (5 ppm) to use as an interpretational reference and internal positive control.
- The manufacturer's protocol was followed and the results recorded for interpretation following the addition of the final reaction stop solution.
- A blue color indicates the sample tests positive for peanut allergen. If the sample well contains more blue color than the control well, the sample contains more peanut allergen. If the sample well contains less blue color, or more red color, the sample well contains less peanut allergen.
- Success criteria – determined by study sponsor.
- A kit interference control was run separately, wherein the antibody portion of the test kit was exposed to D-125, then rinsed, then exposed to peanut allergen and compared to a normal positive control.

# Antimicrobial Test Laboratories

## Fast, Reliable Antimicrobial Efficacy Testing

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### Test Information

#### **Client Information**

Company Name: Microgen Inc  
Sponsor: Robert Prince  
Sponsor's Phone: (973) 575-9025  
Sponsor's Email: robert@microgeninc.com

#### **General Test Information**

Test Performed: Qualitative Enzyme-Linked ImmunoSorbent Assay  
ATL Study ID: NG1380  
SOP Basis: External Protocol  
Performed By: J. Williams  
Date Initiated: 03/09/2009  
Date Completed: 03/09/2009

#### **Sample**

Test Substance: Microgen D-125 Disinfectant ((US REG. 61178-1)  
Device ID: N/A  
Date Received: 04/25/2008 & 02/27/2009

#### **Test Parameters**

Microorganisms: N/A  
Subculture Number: N/A  
Growth Medium: N/A  
Dilution Tested: 1:1000 of peanut allergen extract  
Contact Time: 10 minutes  
Organic Soil Load: N/A  
Neutralizer(s): N/A  
Exposure Temperature: 22 - 24C  
Type of Carrier: N/A  
Carrier Dry Time: N/A  
Incubation Temperature: N/A  
Incubation Time: N/A

#### **Controls**

Manufacturer's positive control: Passed  
Extract positive control: Passed  
Test Sample Negative Controls: Passed

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### Notes Recorded By Study Director

- The empirically determined 1:1,000 dilution of the peanut allergen extract was determined to be 10X concentrated based on the result of a 1:10,000 dilution of the extract giving similar colorimetric results as that of the manufacturer provided positive control.
- 0.1ml of the manufacturer provided positive control (200ng/ml) was used instead of 3 drops (as recommended by the manufacturer) to yield a known concentration of 20ng total peanut allergen added to the manufacturer provided positive control well.
- It was empirically determined and estimated that each test sample received ~200ng of peanut protein (allergen).

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### Colorimetric Results

Photograph 1. ELISA results post-study on 03/09/2009.



**1.**

**2.**

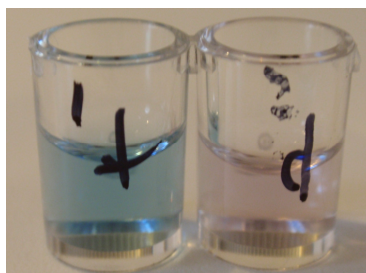
**3.**

**4.**

#### Photograph 1 legend

1. Manufacturer provided positive control well (20ng detected)
2. Peanut allergen extract positive control well (~200ng detected)
3. D-125 "test" well
4. D-125 negative "control" well

Photograph 2. Manufacturer provided positive control dilution results taken on 3/06/2009



**1.**

**2.**

#### Photograph 2 legend

1. Manufacturer provided positive control well (20ng detected, 5 ppm)
2. Manufacturer provided positive control diluted 1:10 (<5 ppm detected)

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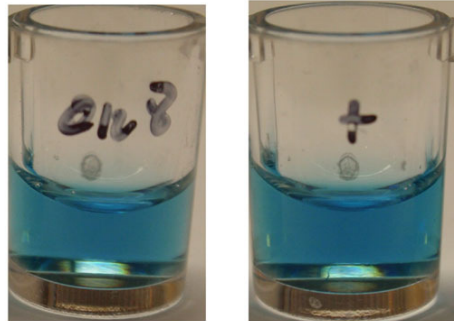
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Photographs of the study, continued:

Photograph 3. Test kit interference control results.



**1.**      **2.**

Photograph 3 legend

1. D-125 exposed and rinsed, then allergen-exposed positive control well.
2. Normal positive control well.

# Antimicrobial Test Laboratories

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### **PROTOCOL CHANGES, DEVIATIONS, AND AMENDMENTS**

#### **Changes**

None Recorded for This Study

#### **Deviations**

0.1 ml of the manufacturer provided positive control was used instead of 3 drops to ensure the addition of 20ng/reaction.

#### **Amendments**

None Recorded for This Study

# Antimicrobial Test Laboratories

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### STUDY ACCEPTANCE CRITERIA

The following controls must be normal in order for the study to be deemed acceptable by the Scientific Director:

1. Manufacturer provided positive control
2. Test sample negative controls
3. Peanut extract positive control
4. Test kit interference control

### Study Control Results

5. Manufacturer provided positive control - **Passed**
6. Test sample negative controls - **Passed**
7. Peanut extract positive control – **Passed**
8. Test kit interference control – **Passed**

## **CALCULATIONS AND STATISTICAL ANALYSIS**

No calculations or statistical analyses were used in this study

# Antimicrobial Test Laboratories

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### **STUDY RECORD AND SPECIMEN RETENTION**

#### **Study Record Retention**

This study report and corresponding data sheets will be held in the archives of Antimicrobial Test Laboratories at the following address for at least 2 years after the date of this report:

Antimicrobial Test Laboratories  
3000 Joe DiMaggio Blvd, Suite #32  
Round Rock, Texas, 78665 USA

#### **Specimen Retention**

The test specimen will be held for 30 days after study completion at the following facility:

Antimicrobial Test Laboratories  
3000 Joe DiMaggio Blvd, Suite #32  
Round Rock, Texas, 78665 USA

# Antimicrobial Test Laboratories

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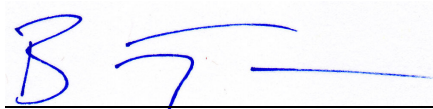
Protocol Reference: External

### STUDY CONCLUSION

Peanut allergen levels, as determined by the Neogen ALERT® Peanut Allergen Screening Test, were reduced approximately ten-fold (~90% reduction) after a ten minute contact with D-125. Reductions were calculated relative to a water control that received the same concentration and volume of peanut allergen for the same period of contact and a separate, laboratory-made calibration curve. The reduction in peanut allergen (~90%) observed in this preliminary study is of approximately the same magnitude as reported for common leading "anti-allergen" spray products.<sup>4</sup>

### Report Submitted By

Scientific Director:



Benjamin D. Tanner, Ph.D.

Study Completion Date:

03/09/2009

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### REFERENCES

1. Reference: Neogen Corporation. "ALERT® Peanut Allergen Screening Test" Product #8431. A-PeaAll-0808
2. Reference: Clorox™ Anti-allergen spray product information website.  
([http://www.clorox.com/products/overview.php?prod\\_id=aaafs](http://www.clorox.com/products/overview.php?prod_id=aaafs)) Downloaded 3/17/09.