EEE BRANCH REVIEW

DATE: IN _____ OUT _____ IN _____ OUT _____ IN 7/3/78 OUT 8/11/78

FISH & WILDLIFE ENVIRONMENTAL CHEMISTRY EFFICACY

FILE OR REG. NO. 34292-1

PETITION OR EXP. PERMIT NO. _____

DATE DIV. RECEIVED 5/11/77 and 3/2/78

DATE OF SUBMISSION 5/5/77

DATE SUBMISSION ACCEPTED _____

TYPE PRODUCT(S): I, (D), (H), (F) N, R, S Industrial Antimicrobial

PRODUCT MGR. NO. 31 (Tavano)

PRODUCT NAME(S) Dow Corning 5700 Antimicrobial Agent

COMPANY NAME Dow Corning Corporation

SUBMISSION PURPOSE Amendment with data (use on carpets)

CHEMICAL & FORMULATION Concentrate for manufacturing use only

Active Ingredient: 3-(Trimethoxysilyl)-propyldimethyloctadecyl ammonium chloride.....42%

DATA ACCESSION NO(S). 232333, 232334, 232335

200.0 Introduction

200.1 Use(s)

The product is registered as a bacteriostat, algistat, and fungistat for manufacturing use as a preservative for unfinished textile fibers, fabrics, and threads.

Claims have also been accepted for its use in a finished article, socks, to prevent deterioration and discoloration caused by fungi and to inhibit odor causing bacteria.

The purpose of the current submission is extend the claims to include finished carpeting, as follows:

"Carpeting.....Treated with Dow Corning 5700 Antimicrobial Agent (1) for lasting freshness and to prevent deterioration and discoloration caused by fungi and bacteria. (2) To inhibit the growth of bacteria and mildew to prolong the life of the carpet. (3) To provide a durable, non-leachable antimicrobial treatment. (4) To provide hygienic freshness. (5) To provide a treatment that lasts the lifetime of the carpet and is not destroyed by repeated cleaning or shampooing. (6) To inhibit the growth of odor causing bacteria and mildew."

200.2 Background Information

It should be noted that this review addresses only the specific data which are the subject of this proposed amendment. No attempt has been made in this review to re-evaluate previously submitted data or previously accepted claims or other proposed amendments for added uses which are also under consideration for this product.

201.0 Data Summary

201.1.1 Brief Description of Test

(A) "Antibacterial activity of throwrugs treated with Dow Corning 5700 Antimicrobial Agent." Reference E-2069-5. Dated 5-5-78 (No Accession Number).

(B) Various tables of summary data headed "Surface Activity: Bacteriostatic Activitiy, Wear Durability Test (Cont'd), Fungistatic Activity", pp. 15 a, 15b, 15c, and 18a. Data appear extracted from a larger report. Undated and unattributed (Accession No. 232335).

(C) "Antibacterial Finishes on Fabrics, Evaluation of ", CTM-0829, dated 11-17-75, pp 1-4. Test protocol (Accession No. 232334).

(D) "Antibacterial Finishes on Fabrics, Evaluation of", AATCC Test Method 100-1974, pp. 272-273. Test protocol (Accession No. 232333).

201.1.2 Data Summaries

(A) "Antibacterial activity of throw-rugs treated with Dow Corning 5700 Antimicrobial Agent".

Purpose

To determine the antimicrobial activity of throw-rugs treated with Dow Corning 5700 antimicrobial agent before and after home laundering.

Test Materials

Polyester throw-rugs and nylon throw-rugs were obtained from Burlington Industries for use in this study. The throw rugs were treated with Dow Corning 5700 antimicrobial agent 0.3% wt./wt. pickup of active ingredient.

Laundering Method

The treated throw-rugs were laundered five cycles and ten cycles according to the Home Laundering section of AATCC Test Mehtod 124-1975 using Tide detergent as directed.

Microbiological Methods

The laundered and unlaundered throw-rugs treated with Dow Corning 5700 antimicorobial agent and

untreated control throw-rugs were tested according to modified AATCC Test Method 100-1974 (CTM-0829) against Klebsiella pneumoniae (ATCC-4352).

Results

The bacterial counts from treated and untreated throw-rugs obtained by CTM-0829 are presented in Table I attached.

Summary

Procedural detail was lacking with respect to the following:

(1) The method of treatment of the rug samples with the product was not described.

(2) A detailed description of the test rug samples was not provided.

(3) The number of samples inoculated and the volume of inoculum per sample were not indicated. Raw data was not included.

(4) The effectiveness of Letheen broth as a neutralizer for the active ingredient was not documented.

(5) A copy of the laundering method (AATCC 124-1975) was not included.

Assuming that the above mentioned details could be provided, the test results indicate that reduction of numbers of Klebsiella pneumoniae was observed when liquid inocula were placed on treated (0.3%a.i.) carpet samples for 4 hours at 37 C and kept under wet conditions, i.e., in a screw-capped bottle with the cap screwed on tightly to prevent evaporation. Under these conditions, count reductions of 99.3-99.4% on nylon rugs and 97.8-99.3% on polyester rugs were obtained after up to 10 launderings compared to initial ("0" contact time) counts. Untreated, unlaundered control rugs supported survival of the inoculum

for the 4 hour period under the test conditions, which were relatively favorable for survival and/or multiplication of the test organism.

The test did not address the proposed claims for the product, such as prevention of deterioration, discoloration, or odor; the causative agent(s) of such problems; the conditions likely to be encountered in actual use; and testing for extended periods of time to support the intended use life of the treated carpet.

(B) Summary data headed "Surface Activity: Bacteriostatic Activity, Wear Durability Test (cont'd.), Fungistatic Activity."

Procedure

Reference to the CTM-0829 protocol. No other procedural details were provided for these studies.

Results

See attached tables designated 15a, 15b, and 15c. Table 18a concerns fungistatic activity and, therefore, is not included here.

Summary

Procedural details and raw data are lacking for these studies.

Assuming that such information can be provided, the results can be tentatively summarized, as follows:

Carpeting treated with the product (0.5-0.75% a.i.) exhibited bacteriostatic activity against artificial inocula of both Staphylococcus aureus and K. pneumoniae, presumably under the same conditions and limitations described for the the study summarized in 201.1.2 (A) above; such activity appeared to be retained after unspecified cleaning procedures and/or exposure to "traffic" (Tables 15a and 15c).

Studies concerning the levels of unidentified "indigenous microflora" on untreated and treated (0.5% a.i.) carpeting, initially, and after exposure to "traffic," suggest that treated carpet yielded lower "microflora" counts. However, the significance of these results was obscured by the stipulation that baseline counts were adjusted to zero (Table 15b). The data relative to the effect of unspecified cleaning of untreated and treated carpet on "indigenous microflora" suggest that differences between treated and control carpets before and after cleaning may be questionable or marginal (Table 15b). The question of what constitutes "indigenous microflora" and the significance of these organisms, if any, in terms of discoloration, deterioration, odor production, or any other problem under actual use conditions was not addressed.

No further evaluation can be attempted in the absence of detailed protocols and raw data for the carpet studies.

(C) "Antibacterial Finishes on Fabrics, Evaluation of," CTM-0829.

This document consists of a protocol which is a modification of AATCC Test Method 100-1974 for laboratory bacteriological evaluation of antibacterial finishes on fabrics (copy attached).

(D) "Antibacterial Finishes on Fabrics, Evaluation of," AATCC Test Method 100-1974.

This document is the basic laboratory method referenced in 201.1.2 (C) above (copy attached.)

202.0 Recommendations

202.1 Efficacy Supported by the Data Submitted

If adequate procedural details, as well as raw data, for the submitted studies are provided, the data would be adequate only to support intrinsic value of the product as a bacteriostat for manufacturing use in the impregnation of carpet material.

202.2 Efficacy not Supported by the Data Submitted

The submitted data do not support effectiveness of the product in prevention of deterioration, discoloration, or odor production by bacteria in finished carpet during use. Until such efficacy is substantiated, the duration of effectiveness of the treatment for the intended purpose(s) cannot be determined.

Effectiveness of the product in providing vague benefits such as "lasting freshness" and "hygienic freshness" to carpets during use do not readily land themselves to verification by scientific methods. Such claims should be clarified or deleted.

202.3 Additional Data Required to Support Efficacy

(A) The product must be tested for effectiveness in/on the treated carpet after application as indicated in the directions for use. The testing must simulate actual in-use conditions. There is no standardized testing protocol for determination of residual bacteriostatic activity of carpets for the proposed claims. Therefore any proposed testing protocol(s) should be submitted for review and comment prior to the initiation of the tests. The protocol(s) should include the following basic elements:

(1) The bacterial problem associated with untreated carpets must be described and documented, i.e. odor production,

deterioration, discoloration, or whatever the problem may be. The test bacteria selected for study must be those which have been isolated or otherwise documented to be the cause of the problem(s) in carpets.

(2) A controlled in-use, or simulated-use study must be conducted for the specified period of time that residual effectiveness is claimed in labeling. Bacteria of the type identified in (1) above must be employed as test microorganisms. The test inoculum must consist of a mixture of the test microorganisms plus representative carpet soil. The inoculum level must be sufficiently high to simulate the more extreme conditions of contamination encountered in actual use, as well as to provide adequate numbers for quantitative comparison. The test inoculum must be applied to both treated and untreated carpet samples of representative types as specified in labeling. The treated and untreated carpet samples must be recontaminated with the test inoculum at multiple intervals each day for the duration of the test. The study must incorporate quantitative recovery techniques (elute surviving microorganisms into appropriate liquid media containing appropriate neutralizer) and quantitative enumeration procedures (plate counts on appropriate agar containing the same neutralizer). Rodac plate determinations are not acceptable. At least 3 different manufacturing lots (one of which should be at least 60 days old) of product must be tested with each type of carpet and test microorganism. Parallel controls are required. Periodic cleaning of the carpet should be addressed in the protocol(s). The relative humidity, temperature, and all other of the test procedures employed

in the study must be provided along with the results. The untreated inoculated controls must show bacterial growth, survival, or metabolism and must demonstrate the bacterial problem (odor, deterioration, discoloration, etc.) associated with carpets. It must be shown that treated carpet inhibits bacterial growth and/or metabolism, or reduces the number of survivors, and prevents the problem associated with carpets. If the control carpets fail to support bacterial growth, survival, or metabolism, or if no problem can be demonstrated, then no effect of the bacteriostatic treatment can be proven.

(3) If the intended use for product is on indoor carpeting of the types associated with residential living quarters or lobbies, hallways, and offices of commercial or institutional buildings, such carpet would be considered as likely to remain dry between treatments. In this case, dry test inocula must be applied to dry treated and untreated carpet samples in the study described in (2) above. No moisture must be added to the system during the test period.

(4) If the recommended areas of use for the product are specifically restricted in labeling to indoor-outdoor types of carpeting associated with patios, swimming pools, shower and locker rooms, etc., which is likely to become and remain wet for considerable periods, wet test inocula may be employed and moisture may be included in the test described in (2) above.

(B) The recommended area(s) of use for the carpet, such as hospitals, medical-type institutions, motels, office buildings, homes or whatever the intended market for the product may be, must be specified in the labeling.

(C) Subsequent to the development of appropriate and adequate efficacy data, label revisions may be necessary to accurately reflect the effectiveness demonstrated.

Dennis G. Guse 8/11/78

Dennis G. Guse
Technical Support Section (Efficacy)
Disinfectants Branch, RD

concur D.J. Jenkins 8/11/78

TABLE I

Antibacterial Activity of DOW CORNING® 5700 Antimicrobial Agent
Treated Polyester and Nylon Throw-Rugs

Samples	No. of Laundering Cycles	Number of Bacteria Remaining at: 0 Time	4 Hours	% Reduction
Nylon Throw-Rugs:				
Control	0	196,000	625,000	--
Treated	0	196,000	1,050	99.4
Treated	5	178,500	1,100	99.4
Treated	10	186,000	1,350	99.3
Polyester Throw Rugs:				
Control	0	189,000	106,000	—
Treated	0	207,500	1,250	99.3
Treated	5	184,500	4,300	97.8
Treated	10	184,000	1,400	99.3

SURFACE ACTIVITY

BACTERIOSTATIC ACTIVITY

1. Staphylococcus aureus ATCC 6538 (a) Nylon Carpeting CTM-0829

Sample	Treatment	Bact. Recovery 0 Time	Bact. Recovery 6 Hours	% Reduction
Nylon Carpeting	None	203,000	200,000	-
Nylon Carpeting	0.75% Q9-5700	203,000	3,400	98.3
Nylon Carpeting	0.50% Q9-5700	203,000	600	99.7

5. Klebsiella pneumoniae ATCC 4352 (a) Nylon Carpeting CTM-0829

Sample	Treatment	Bact. Recovery 0 Time	Bact. Recovery 6 Hours	% Reduction
Nylon Carpeting	None	139,000	140,000	-
Nylon Carpeting	0.75% Q9-5700	139,000	600	99.6
Nylon Carpeting	0.50% Q9-5700	139,000	850	99.4

WEAR DURABILITY TEST (Continued)

A STUDY OF THE INDIGENOUS MICROFLORA ON SAMPLES OF TREATED AND UNTREATED CARPETING WITH BASELINE ADJUSTED TO ZERO

Treatment 0.5% Dow Corning® Q9-5700 Antimicrobial Agent

Treatment	Number Of Traffics*	Cleaned (+ or -)	No. Of Microorganisms Per Sample	% Reduction (Treated vs Untreated)
None	0	-	0	
Q9-5700	0	-	0	
None	10,000	-	18,000	
Q9-5700	10,000	-	2,500	86.1
None	20,000	-	22,000	
Q9-5700	20,000	-	1,000	95.5

*Actual count of people measured by electronic calculator.
*Ac

EFFECT OF CLEANING ON THE INDIGENOUS MICROFLORA OF TREATED AND UNTREATED CARPETING

Treatment 0.5% Dow Corning® Q9-5700 Antimicrobial Agent

Treatment	Number Of Traffics*	Cleaned (+ or -)	Number Of Microorganisms Per Sample
None	10,000	-	30,000
Q9-5700	10,000	-	14,500
None	10,000	+	6,100
Q9-5700	10,000	+	4,650

*Actual count of people measured by electronic calculator.

WEAR DURABILITY TEST (Continued)

ANTIMICROBIAL ACTIVITY OF
CARPETING TREATED WITH 0.5% DOW CORNING® Q9-5700 ANTIMICROBIAL AGENT

Treatment	Number Of Traffics*	Cleaned (+ or -)	No. Of Bacteria[1] Per Sample Remaining After Incubation[2]	% Reduction (Treated vs Untreated)
None	0	+	280,000	0
Q9-5700	0	+	1,900	99.3
None	10,000	+	1,050,000	0
Q9-5700	10,000	+	545,000	48.1
None	20,000	+	1,800,000	0
Q9-5700	20,000	+	545,000	69.7
None	30,000	+	2,800,000	0
Q9-5700	30,000	+	660,000	76.4

*Actual count of people measured by electronic calculator.

[1]Samples (four sq. in.) were inoculated with Klebsiella pneumoniae ATCC 4352. The initial inoculum for all samples was approximately 260,000 bacteria.

[2]The exposure or incubation period for all samples was six hours.

TRI QUATS PC 107401

8/11/78 REG # 34292-1

Page _____ is not included in this copy.

Pages 15 through 18 are not included in this copy.

The material not included contains the following type of information:

_____ Identity of product inert ingredients.

_____ Identity of product impurities.

_____ Description of the product manufacturing process.

_____ Description of quality control procedures.

_____ Identity of the source of product ingredients.

_____ Sales or other commercial/financial information.

_____ A draft product label.

_____ The product confidential statement of formula.

_____ Information about a pending registration action.

X____ FIFRA registration data.

_____ The document is a duplicate of page(s) _____.

_____ The document is not responsive to the request.

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.

Antibacterial Finishes on Fabrics, Evaluation of

Developed in 1961 by AATCC Committee RA31; revised 1965; reaffirmed 1970, 1974; editorially revised 1959, 1971, 1974. (Same as ANSI L14.144-1965/R1971)

Foreword

The problem of evaluation of antibacterial finishes on fabrics involves the degree of antibacterial activity intended in the use of such fabrics. If only bacteriostatic activity (inhibition of multiplication) is intended, a qualitative procedure which clearly demonstrates antibacterial activity as contrasted with lack of such activity by an untreated specimen may be acceptable. Several qualitative procedures must be available, due to the diversity of chemicals used as antibacterial agents.

However, if bactericidal activity is intended or implied, quantitative evaluation is necessary. Quantitative evaluation also provides a clearer picture for possible uses of such treated fabrics. Through interlaboratory tests a quantitative procedure, which stimulates in-use conditions as much as possible, was developed. It is an outgrowth of the Majors test for bacteriostatic activity.

Committee RA31 has concluded that it would be realistic to recommend acceptance of a two-part procedure as follows:

a) qualitative or presumptive tests, and

b) quantitative or reference test.

Of the four qualitative methods listed below, the Committee has carried out interlaboratory tests on three of them (see 11.1, 11.2, and 11.3), and one of the Committee members was a coworker in the development of the fourth (see 11.4). The AATCC has approved the Agar Plate Method (AATCC Method 90) (see 11.1), which is dependent on the production of a clear zone of inhibition around the treated fabric. Another procedure ("streak method") depends on inhibition along the line of inoculation (see 11.2). A third method involves changes in pH, when the inoculum is suspended in a highly buffered medium (see 11.3). Still another method depends on growth from an inoculated swatch embedded in agar (see 11.4). A brief summary of these procedures is given in Appendix A. Since all of these methods are qualitative, they cannot accurately measure the amount of antibacterial activity.

1. Purpose and Scope

1.1 This method recognizes four qualitative procedures for the demonstration of bacteriostatic activity. They are used to select fabrics which show promise of antibacterial activity. This method provides a quantitative procedure for the evaluation of the degree of activity.

2. Limitations

2.1 Both the qualitative and quantitative tests should be carried out by persons with training and experience in the use of bacteriological techniques.

3. Principle

3.1 Swatches of test and control fabrics are tested qualitatively for antibacterial activity by any of the four procedures described in Appendix A. Those showing activity are evaluated quantitatively. Test and control swatches are inoculated with the test organisms. After incubation, the bacteria are eluted from the swatches by shaking in known amounts of liquid. The number of bacteria present in this liquid is determined, and the percentage reduction by the treated fabric is calculated.

4. Safety Precautions

4.1 Both *Staphylococcus aureus* and *Klebsiella pneumoniae* are bacteia capable of infecting man and producing disease. Therefore, every necessary and reasonable precaution must be taken to eliminate this risk to the laboratory personnel and to personnel in the associated environment.

5. Test Organisms

5.1 Test bacteria: (a) *Staphylococcus aureus* strain 209, American Type Culture Collection No. 6538; (b) *Klebsiella pneumoniae*, American Type Culture Collection No. 4352.

6. Culture Medium

6.1 AATCC Broth (see 11.5).

Peptone (Bacto-peptone (see 11.6) or Thiotone (see 11.7))	10 g
Beef Extract (see 11.8)	5 g
Sodium Chloride	5 g
Distilled Water	1000 ml

6.2 Heat to dissolve ingredients. Adjust to pH 6.8 with NaOH.

6.3 Dispense in 10-ml amounts in 125 mm x 17 mm tubes and sterilize at 103 kPa (15 psi) for 20 min.

7. Maintenance of Culture of Test Organisms

7.1 Using a 4 mm loop, transfer the test culture daily in AATCC broth for not more than two to three weeks. At the end of this time, make fresh transplants from the stock culture. Incubate cultures at 37C.

7.2 Maintain stock cultures on AATCC agar slants, which are the same composition as the AATCC Broth, plus 1.5% agar. Store the stock culture at 5C and transfer once a month (see 11.9).

8. Qualitative Procedure (Screening or Presumptive Test)

8.1 For detection of bacteriostatic activity, apply one of the four qualitative test methods summarized in Appendix A to the test fabric and to a control fabric, in accordance with the recognized procedure for the method, using the organisms referred to above. For demonstration of bactericidal activity, proceed to the quantitative test described below.

9. Quantitative Procedure (Reference or Confirmatory Test)

9.1 Size of inoculum per sample. Apply 1 ml of an appropriate dilution of a 24-hour broth culture of the test organism so that recovery from (1) untreated control fabric swatches or (2) treated test fabric swatches at "0" contact time (plated as soon as possible after inoculation) will show counts of 10-20 x 10^4 organisms. The dilution of test organism should be made in AATCC broth.

9.2 Size and Shape of Treated Swatches. Cut circular swatches, 4.8 cm (1 7/8 in.) in diameter, from the test fabric (preferably with a steel die). Stack the swatches in a .237 l (8 oz) wide-mouth glass jar with screw cap. The number of swatches to be used is dependent on the fiber type and fabric construction. Use that amount of fabric which will absorb the 1 ml of inoculum, and leave no free liquid in the jar. For example, 4 swatches of cotton print cloth will absorb 1 ml.

the number of swatches used per jar should be reported.

9.3 Controls. Inoculated swatches of the same fiber type and fabric construction as test sample but containing no antibacterial finish; uninoculated test fabric (negative control).

9.4 Sterilization of samples. The method to be used depends on the type of fiber and finish. Cotton, acetate and many man-made fibers can be sterilized in the autoclave. Wool can be sterilized by ethylene oxide or by intermittent (fractional) sterilization in flowing steam. The latter is also least damaging to certain finishes.

9.5 Inoculation of fabrics. Using a 1-ml pipette, pad the inoculum carefully on to the fabric to *insure even distribution*. Screw the jar tops on tightly to prevent evaporation.

9.6 Sampling at "0" contact time.

9.6.1 As soon as possible after inoculation, add 100 ml of neutralizer solution to jars containing the inoculated untreated control swatches and to jars containing the inoculated treated test swatches. Add 100 ml of neutralizer solution to jars containing the uninoculated treated test swatches.

9.6.2 Shake the jars vigorously just for one minute. Make serial dilutions and plate (in duplicate) in tryptone glucose extract agar. Dilutions of 10^0, 10^1, 10^2 are usually suitable.

9.6.3 The neutralizer solution should include ingredients to neutralize the specific antibacterial fabric treatment and to take care of any pH requirements of the fabrics (from finishes, antibacterial agents, etc.). The neutralizer solution employed should be reported.

9.7 Incubation over contact periods. Incubate additional jars containing inoculated untreated control swatches and jars containing inoculated treated test swatches at 37C for 18-24 hours. Similar jars may be incubated over other periods (e.g., 1 or 6 hours) to provide information about the bactericidal activity of the treatment over such periods.

9.8 Sampling of inoculated and incubated swatches. After incubation, add 100 ml of neutralizer solution to jars containing untreated control swatches and to jars containing treated test swatches. Shake the jars vigorously for one minute. Make serial dilutions and plate (in duplicate) in tryptone glucose extract agar. Dilutions of 10^0, 10^1, 10^2, are usually suitable for treated test fabrics. Several different dilutions may be required for untreated control fabrics depending upon the incubation period.

9.9 Incubate all plates for 48 hours at 37C.

10. Quantitative Evaluation

10.1 Report bacterial counts as the number of bacteria per fabric sample, (swatches in jar) not as the number of bacteria per ml of neutralizer solution. Report "0" counts at 10^0 dilution as "less than 100."

10.2 Calculate percent reduction of bacteria by the fabric treatment as follows:

$$\frac{B \text{ or } C \text{ or } \frac{B+C}{2} - A}{B \text{ or } C \text{ or } \frac{B+C}{2}} \times 100$$

A = the number of bacteria recovered from the inoculated treated test fabric swatches in the jar incubated over the desired contact period.
B = the number of bacteria recovered from the inoculated treated test fabric swatches in the jar immediately after inoculation (at "0" contact time).
C = the number of bacteria recovered from the inoculated untreated control fabric swatches in the jar immediately after inoculation (at "0" contact time). If "B" and "C" are not similar, the larger number should be used. If "B" and "C" are not significantly different, B + C/2 should be used.

10.3 For a valid test there should be: (1) "0" colonies recovered from the uninoculated treated test fabric swatches and (2) A significant increase in the numbers of bacteria recovered from the inoculated untreated control fabric swatches incubated for 24 hours over the numbers of bacteria recovered from the inoculated untreated fabric swatches at "0" contact time (immediately after inoculation).

10.4 Report percent reduction of bacteria by the fabric treatment against each test organism.

11. References

11.1 AATCC Test Method 90 "Antibacterial Activity of Fabrics, Detection of: Agar Plate Method."

11.2 Engel, W., "Self-Sterilizing Surfaces," Witherby, London, 8 pp. (1952).

11.3 Majors, Paul, "Evaluation of the Effectiveness of Antibacterial Finishes for Cloth," *Am. Dyestuff Reptr. 48:* 91-3 (1959).

11.4 Quinn, Herbert, "A Method for the Determination of the Antimicrobial Properties of Fabrics." *Applied Microbiology 10:* 75-78 (1962).

11.5 Dehydrated AATCC Broth and Agar may be obtained from: Baltimore Biological Laboratories, 2201 Aisquith St., Baltimore, Maryland, and Difco Laboratories, 920 Henry St., Detroit 1, Michigan.

11.6 Bacto-Peptone may be obtained from: Difco Laboratories (address above).

11.7 Thiotone may be obtained from: Baltimore Biological Laboratories (address above).

11.8 Beef Extract may be obtained from: Baltimore Biological Laboratories (address above); Difco Laboratories (address above); or Oxo (USA) Ltd., 1330 Beacon Street, Brookline, Massachusetts.

11.9 Consistent and accurate testing require maintenance of a pure, uncontaminated, nonmutant test culture. Avoid contamination by use of good sterile technique in plating and transferring. Avoid mutation by strict adherence to monthly stock transfers. Check culture purity by making streak plates periodically and observing for a single species-characteristic type of colonies.

Appendix A

Summaries of Qualitative Test Procedures.

A1. AATCC Agar Plate Method. This is a qualitative test to demonstrate bacteriostatic activity against *Staphylococcus aureus* and *Klebsiella pneumoniae*. A clear zone of no growth is demonstrable around treated fabric when a specimen of such fabric is placed on the surface of agar seeded with the test organism, and incubated at 37C for 24 hours. An untreated control fabric must be included in in the test.

A2. Streak Test. This is a modification of the Agar Plate Method. Agar Plates are streaked with the test organism, and treated and untreated swatches are placed on the surface of the plate at right angles to the line of streak, clear areas of no growth adjacent to the fabric are demonstrable.

A3. Majors Test. This is a semiquantitative test procedure for evaluation of bacteriostatic activity. The amount of growth of the test organism is a highly buffered medium, held in the interstices of the fabric is estimated by titration of the amount of acid or alkali produced from the medium substrate (glucose or urea) by the test organism.

A4. Quinn Test. This method is suitable for bacteriostatic evaluation, and is an attempt to relate a test procedure to in-use conditions. Small treated and untreated swatches are inoculated with the test organism and then dried under conditions of known relative humidity. They are then placed on sterile agar plates, covered with a thin layer of agar, and incubated. After incubation; the numbers of colonies on treated and untreated swatches are counted under the low power lens of a microscope.

(20)