INHIBITION OF GROWTH OF A MOLD QUANTITATED TO DEMONSTRATE THE EFFECT IN INSECT SPECIMEN BOXES¹

By CDR John D. DeCoursey, MSC, USN² and CDR A. P. Webster, MSC, USN³

NAVAL MEDICAL FIELD RESEARCH LABORATORY CAMP LEJEUNE, NORTH CAROLINA

INTRODUCTION

One way to protect insect collections contained in specimen chests from molding is to incorporate a substance within the chest, the vapors of which are fungicidal or fungistatic. Further, the substance should have the property of low volatility so that prolonged action is obtained.

Two such substances, having the required physical properties, which have been used for years in insect collections to protect them from museum pests, and which are in common use by every housewife to protect clothes, are naphthalene and paradichlorobenzene.

Bolcato (1) found that naphthalene vapors inhibited sporification of aspergilli; and Bishopp (2) says that both naphthalene and paradichlorobenzene are mold inhibitors, but that naphthalene is considered preferable for insect collections because of its lower volatility. The following study was undertaken in order to evaluate the quantitative effect of naphthalene vapor as a fungistatic agent against a single mold—Penicillium.

EXPERIMENTAL METHOD

Four Petri dishes were prepared with Sabouraud's dextrose media (Difco dehydrated media) by seeding with a piece of mycelial mat transferred from a contaminant (*Penicillium*) on a Sabouraud's agar plate. Two of the dishes were sealed with scotch tape and placed aside as controls. The other two were treated with naphthalene by suspending 0.2 gram of naphthalene

¹ This work is not to be construed as necessarily reflecting the views of the Navy Department.

² Head, Department of Entomology.

³ Research Director.

crystals from the inner surface of the Petri dish cover by means of a small square of bobbinet held in place by adhesive tape. These dishes were also sealed with scotch tape and placed beside the controls at room temperature (approximately 78° F.). The diameter of the mold, during its growth, was measured in the

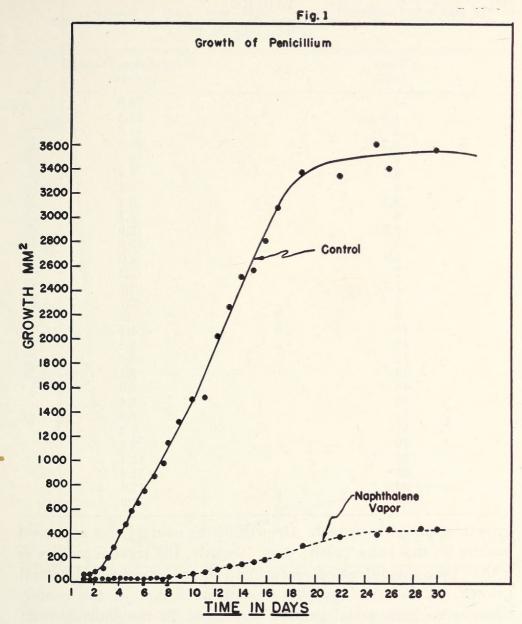
TABLE 1
GROWTH OF Penicillium

Time	Growth	
	Naphthalene Treated	Control
	mm ²	mm ²
0		
1	21	26
1.5	22	47
2	23	74
2.5	25	96
3	23	194
3.5	24	269
4	24	417
4.5	$\frac{24}{24}$	464
5	$\frac{\overline{24}}{24}$	583
5.5	$\frac{1}{26}$	645
6	28	747
6.5	29	740
7	29	876
7.5	27	979
8	31	1158
9	42	1314
10	62	1514
11	88	1518
12	102	2023
13	139	2261
14	150	2498
15	165	2561
16	192	2804
17	222	3068
19	296	3370
22	373	3349
$\frac{22}{25}$	412	3605
26	421	3406
30	423	3563

four dishes by taking multiple readings, always at the same points, with a millimeter rule. Two readings a day were taken for the first seven days when the mold growth was most rapid and occasional readings thereafter. Subsequently the mean diameters of the treated and untreated molds were obtained and the surface area of the mold calculated.

RESULTS AND DISCUSSION

Table 1 shows the mean growth of *Penicillium* treated with naphthalene vapor in a closed Petri dish, compared with the untreated control. Figure 1 is a plot of the data from Table 1. Table 2 shows a comparison by day of growth of the naphthalene



treated in per cent of the control. Figure 2 is a plot of the growth of the naphthalene treated mold in percent of the control growth. The area of the control mold increased rapidly. The amount

of growth was approximately 200 mm² per day. The growth of the naphthalene vapor treated mold was almost completely inhibited until the eighth day when a slight increase was noted. After the eighth day the naphthalene treated mold slowly increased in surface area for about 16 more days at which time

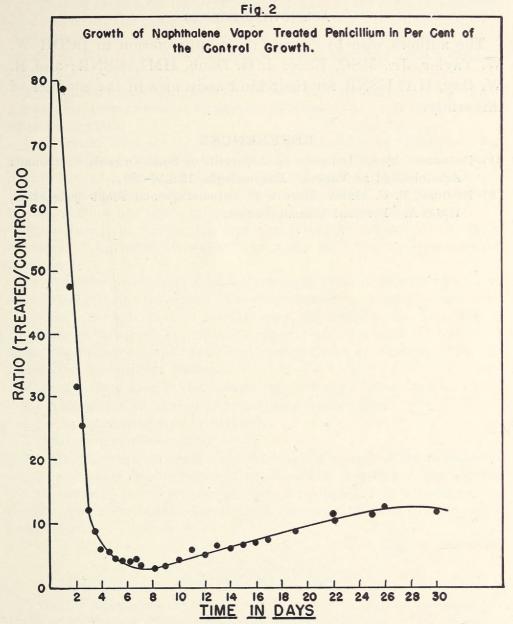
TABLE 2
COMPARISON OF NAPHTHALENE TREATED AND CONTROL GROWTH
OF Penicillium

Time Days	Naphthalene/Control per cent	
1	78.8	
1.5	47.4	
2	31.6	
2.5	25.7	
3	12.1	
3.5	8.8	
4	5.8	
4.5	5.1	
5	4.1	
5.5	4.0	
6	3.8	
6.5	4.0	
7	3.3	
7.5	3.3 2.7	
8	2.7	
8	3.2	
10	4.1	
11	4.1 5.8	
12	5.0	
13	6.1	
14	6.0	
15	6.4	
16	6.9	
17	7.2	
19	8.8	
22	11.1	
25	11.4	
26	12.4	
30	11.9	

growth apparently ceased. Growth of the control also appeared mature on this same (24th) day. Grossly, the treated mold was whiter than the untreated control and showed considerable aerial growth. This aerial growth was about 5 mm in the center, whereas no such aerial growth was noted in the controls. At no time during its course of growth after the third day was the naphthalene treated mold over approximately 12 percent of the surface area of the control.

SUMMARY

The vapor of naphthalene crystals has a marked fungistatic action on *Penicillium* as shown by a comparison study of the growth of the mold on Sabouraud's media in a Petri dish. The



growth of the mold under the influence of naphthalene vapor was completely inhibited up to the eighth day when slow growth took place to an apparent mature value of about 420 square millimeters on the 24th day, as compared with rapid growth of

the control to an apparent mature value of about 3560 square millimeters on the 24th day. At no time during its course of growth after the third day was the naphthalene treated mold over approximately 12 percent of the surface area of the control.

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