

THE EFFECTS OF CERTAIN AGE FACTORS ON THE RESPONSE
OF THE GERMAN COCKROACH TO INSECTICIDES

by

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INTRODUCTION

The German cockroach, Blattella germanica (L.), is a common household pest over most of the United States. Efforts to control this noxious insect have been based upon the use of chlorinated hydrocarbon insecticides, especially chlordane, with excellent results. The discovery of a field strain of German cockroaches in Corpus Christi, Texas, that was resistant to chlordane (Heal et al., 1953) generated much interest in this species. Subsequently, chlordane resistance has become quite general, and extensive efforts have been made to find substitute insecticides for the control of this insect. The organic phosphate insecticides malathion and Diazinon are now used successfully in control work. However, a strain which is resistant to Diazinon has been discovered from Owensboro, Kentucky (Grayson, 1961) and forewarns of further difficulties.

In response to this continuing problem, studies have been conducted to evaluate many new insecticides which may be useful against resistant strains of the German cockroach (Grayson and Messersmith, 1959; Grayson and Perkins, 1960, 1961; Grayson and Townsend, 1962). From these and other studies, much has been learned about the response of this species to insecticides, and a great deal is now known about which insecticides are effective in its control. However,

one of the vexing problems in this work is the inconsistency of results from test to test (Cochran, 1962) which poses questions concerning the reliability of data. No satisfactory explanation for this situation exists, although a number of factors could be involved, such as sampling error, variation in the treatment procedure, age of test insects, age of the parents at the time of reproduction, etc. Critical studies of these factors have not been made for this species up to the present time.

The purpose of this study was to determine the effects of two age factors upon the response of adult German cockroaches to various insecticides. One of these factors was the length of time which the insect had been in the adult stage. Age groups of 1-3, 5-7, 10-12, 15-18, and 25-28 days of adult age were used for both sexes in testing this factor.

The other factor examined was the age of the parents at the time reproduction occurs. Precise control of this variable is difficult to achieve, but partial control of the age of the female parent can readily be obtained by taking advantage of the manner in which eggs are deposited by this species. In the German cockroach the females produce a series of egg cases each of which normally contains about 40 eggs (Girault, 1911). Each egg case is carried by the females until the embryos are nearly ready to hatch, which is 16-19 days at 30°C. according to Perkins and Grayson (1961). Thus, females of this species deposit eggs only periodically. By

separating progeny on the basis of the egg case number from which they were derived it was possible to examine the influence of parental age upon progeny response to insecticides.

REVIEW OF LITERATURE

Various workers have found that the age of the test insect at the time of testing is important. Simanton and Miller (1937) tested adult houseflies from one and one-half hours to ten days of age with pyrethrum sprays. It was found that, although very young flies were more easily paralyzed than were older flies, the young flies were less easily killed. Doner and Anderson (1947) suggested that, when using their micro method for biological testing of non-fumigant types of insecticidal residues on the housefly, test groups having an average age of no less than four nor more than six days be used. Individual flies in the test groups should not be less than three days or more than seven days of age at the time of testing. Cox (1944a) recommended that flies be three to seven days old when tested. Campbell and Sullivan (1938) used flies two to three days of age in their metal turntable method for comparative tests of liquid spray contact insecticides.

Pan and Yang (1962) reported that when adult *Drosophila* flies were exposed to dry films of endrin they were more resistant at one

day than at two to four days of age. Flies reared from another medium showed no difference in LD_{50} values in one-, two-, and three-day-old adults, but a decline in resistance was observed at four and five days of age.

Gough (1939), working on the resistance of the flour beetle Tribolium confusum Duv. to hydrogen cyanide fumigation, tested all stages of the insect. It was found that three- to four-day-old eggs were more resistant to HCN gas than were eggs from zero to one day in age. Two- to three-day-old pupae were more resistant than were zero- to one- or seven- to eight-day-old pupae. Young adults appeared to be slightly more resistant than were older adults.

Working with the plum curculio, Conotrachelus nenuphar (Hbst.), Smith and Fiori (1959) found no significant difference in the LD_{50} values for beetles 2, 26, and 95 days of age when treated with several different insecticides.

There are several conflicting references on the optimum age for testing German cockroaches with insecticides. Gould (1948) tested roaches of both sexes at two, five, and ten days after maturity and found that the older individuals were somewhat more resistant to insecticides. Hazard (1945a) tested both nymphs and adult males with pyrethrum. The nymphs showed an increase in resistance from 15-20 days up to 35-40 days. Adult males 1-3 days old were found to be more susceptible than roaches 7-9, 13-15, and 19-21 days of age. There were no significant differences between the last three age

groups. Tuma (1938) reported that, when tested with thiocyanacetic acid and pyrethrums, German cockroaches exhibited their greatest resistance at 17 weeks of age. A much higher mortality was exhibited when using roaches of mixed ages.

Several different ages of cockroaches have been recommended or used by research workers. McGovran and Fales (1942) isolated young nymph cultures from stock cultures twice a week. As soon as one-fourth or more of the nymphs had become adults, they were used for testing sprays. In all cases the adults were less than two months old. The National Association of Insecticide and Disinfectant Manufacturers recommends that males should have reached the adult stage at least three days prior to testing. Recently emerged adult males with undarkened cuticle should not be used (Anonymous, 1946). Butts and Davidson (1955) used adult, non-capsulated females one to two weeks old in their microinjection work. Campbell et al. (1941) used roaches from dated culture jars in which the adults were just beginning to appear. In their topical application work, Fisk and Isert (1953) used adult females approximately six weeks of age. Hazard (1945b) used adult males 7-14 days old in his work with liquid roach sprays. Cox (1944b) used adults two and one-half months old in testing baits.

Parental age has been found to play an important role in the life cycle of the offspring. O'Brian (1961) reared Drosophila melanogaster through nine successive generations, observing the effect of parental

age on the offspring from young, middle-aged, and old parents. He found that in offspring from middle-aged parents, the reproductive capacity was initially lower than that of the parental generation, whereas the reproductive capacity of the progeny from young and old parents was higher. Viable eggs were produced by the progeny of young parents for a longer period than they were by the parental generation. Progeny from middle-aged parents produced viable eggs for long periods initially, but this interval shortened in the last three generations studied. Viable egg production from the progeny of old parents was always of shorter duration than in the parental generation. It was also found that offspring from middle-aged and old parents had a shorter life span than that of the parental generation.

Saunders (1962) found that as the maternal generation of Nasonia vitripennis (Walker) (Hymenoptera, Pteromalidae) became senile, a greater proportion of the progeny entered diapause at the end of the fourth larval instar.

Working with the milkweed bug, Oncopeltus fasciatus (Dallas), Richards and Kolderie (1957) observed that the mother's age affected the weight, development rate, and hatchability of her eggs. Egg weight and production by young females were low, increasing until they reached a peak when the females were twenty days of age. Subsequently, both values decreased. The weights of terminal egg lots were found to be very low. The rate of development and

hatchability of the eggs roughly paralleled the weight changes.

Several workers have studied the effects of parental age on the life cycle of the mealworm Tenebrio molitor Linnaeus. Ludwig and Fiore (1960) studied beetles reared at 30°, 25°, and 20° C. They found that parental age had no effect on the duration of the egg stage, the weight of eggs, or the weight of newly hatched larvae. The percentage of eggs which hatched was found to decrease with an increase in parental age. The larvae from young parents grew at a slower rate than those from older parents. At 25° and 20° C. the larvae from young parents underwent more molts and required a longer time to complete development than those from the same parents after they were one month of age or older. In addition, an increase in parental age resulted in shortening of adult progeny life at these temperatures. Tracey (1958) also found that the progeny from young parents required more molts, had a longer larval stage, and a longer adult life than those from older parents. The pupal stage was not found to be effected by parental age. Ludwig (1956) observed that progeny from young parents had a longer larval stage than those from older parents.

METHODS AND MATERIALS

COCKROACH STRAINS

Three strains of the German cockroach were used in this study: a normal strain, an aldrin-resistant strain, and a DDT-resistant strain. The normal strain was one in which the breeding population had never been exposed to insecticides and was presumed to be susceptible to all insecticides. The DDT-resistant strain arose from placing roaches under selection pressure with DDT in the laboratory (Grayson, 1951; 1953). The aldrin-resistant strain was a strain which had become resistant to chlordane in the field (Heal et al., 1953; Grayson, 1954). It was then placed under selection pressure with chlordane followed by selection with aldrin in the laboratory. High levels of resistance were achieved in both resistant strains. Selection with the appropriate insecticide at periodic intervals has been employed to maintain high levels of resistance. However, the cultures used in these experiments had intentionally not been selected for approximately 18 months prior to testing, and the level of resistance in the DDT-resistant strain had decreased considerably.

REARING

Cockroaches for breeding purposes were obtained from cultures maintained in the laboratory. Breeders were kept in one-gallon, glass battery jars, with masonite panels being used to provide resting surfaces. Food, which consisted of dog food pellets, and water were provided. A thin layer of petroleum jelly about two inches wide around the inside of the jar at the top prevented the roaches from escaping. A layer of cheesecloth held over the top of the jar by a rubber band prevented stray roaches from contaminating the cultures.

Nymphs were removed from the breeder jars periodically and placed in battery jars. Instead of masonite panels, a V-shaped piece of screen wire was used as a resting surface. This permitted observation of the culture without disturbing it. Food and water were provided in the usual manner.

From these breeder jars the newly emerged adults were removed to battery jars set up in the same manner as for the nymphs. When egg cases were formed, the females were removed to another jar. After the egg cases hatched, the females were removed to await the formation of a second egg case, etc. In this manner it was possible to obtain cultures of nymphs from as many chronologically produced egg cases as needed. In the present studies only the first three were used.

When the nymphs from the various egg cases matured, the resulting adults were removed for testing. For the 15-18 day and 25-28 day old

groups the adults were removed every third day, while for the 1-3, 5-7, and 10-12 day old groups they were removed every other day. The adults were placed in one-quart, wide-mouth jars with a screen wire resting surface. No more than 100 roaches were placed in a jar. Water and food were provided.

The jars were labeled with the name of the strain of cockroach, the egg case number from which the roaches came, the age group in which they were to be tested, and the date they were to be tested. All roaches were kept in constant temperature cabinets at approximately 30° C. and 65% relative humidity.

TESTING

In this study three different insecticides were used; these were malathion, aldrin, and DDT. The normal strain of cockroaches was tested with all three insecticides, while the aldrin-resistant strain was tested with aldrin, and the DDT-resistant strain was tested with DDT.

The testing method of Clarke and Cochran (1959) was followed. Stock solutions of malathion and aldrin were made by dissolving a measured quantity of technical insecticide in acetone. The appropriate amount of this stock solution was then added to 500 ml. of distilled water to form a water suspension. When using aldrin, an

emulsifying agent, Emcol H-65C, was employed to form a better suspension. The DDT suspensions were made by adding a measured amount of 75% wettable powder to 500 ml. of distilled water. A Waring Blender was used to obtain good suspension of the DDT in water. After mixing, the insecticide suspensions were placed in a water bath at 30°C.

Roaches for testing were taken from the appropriate strain, age group, and egg case according to a pre-established schedule. The roaches to be tested were captured in small screen wire cages (three inches long and 3/4 inch in diameter) by means of an aspirating device. They were tested by immersing the cages in the insecticide suspension for 20 seconds, and then allowing them to stand at room temperature for one hour before removal to recovery jars. The recovery jars were held at 30°C. in a constant temperature cabinet until the mortality counts were made. Mortality counts were taken three days after testing with malathion, and six days after testing with aldrin and DDT.

STATISTICAL METHODS

A complete test on a given age group consisted of four to six concentrations of insecticide with at least three repetitions on different days. The results of these tests were compiled and the

percent mortality was plotted against concentration on logarithmic probit graph paper. A dosage-mortality line was then mathematically fitted to the points by the method of Bliss (1935). LC_{50} values, slopes of the lines, and other pertinent data were taken from these fitted regression lines.

The results were analyzed to determine whether either of the age factors was of importance in the response of the German cockroach to insecticides by transforming the individual mortality values to arcsine values which were arranged in two-way tables. An analysis of variance was then completed for each two-way table. In the cases where age or egg case number were found to be significant, the "New Multiple Range Test" of Duncan (1955) was run to determine which ages or egg case numbers were significantly different from others of similar nature.

RESULTS AND DISCUSSION

AGE OF THE TEST INSECT

When the normal strain was tested with malathion, the tests with males showed the 1-3 and 15-18 day age groups to be more susceptible than the other age groups. There was no significant difference between the 1-3 and the 15-18 day age groups, and none between the 5-7, 10-12, and 25-28 day age groups, but the former differed significantly from the latter. The females were found to be the least susceptible in the 1-3 and 25-28 day age groups (Table 1). This is contrary to what was reported by Gould (1948) who found that five- and ten-day old females were less susceptible to insecticides than were two-day old females. The slopes of the dosage-mortality lines for all age groups and egg case numbers were very similar (Figure 1; Table 2). LC_{50} values varied somewhat, but the highest value (.124 ml./liter) in the males was only twice the lowest value (.065 ml./liter). The LC_{50} values for the females followed the same general pattern.

The male normal strain roaches were found to be the least susceptible in the 1-3 day age group when tested with aldrin. There was no significance between the 5-7, 10-12, 15-18, and 25-28 day age groups (Table 1). This is also in disagreement with published

Table 1

Separation of Means by the New Multiple Range Test

Normal Strain Tested With Malathion

males	age group	10-12	25-28	5-7	1-3	15-18
	mean (coded)	<u>42.36</u>	<u>45.19</u>	<u>47.06</u>	<u>55.06</u>	<u>56.62</u>
females	age group	25-28	1-3	5-7	10-12	15-18
	mean	<u>37.65</u>	<u>38.16</u>	<u>45.27</u>	<u>46.29</u>	<u>49.36</u>

Normal Strain Tested With Aldrin

males	age group	1-3	5-7	15-18	25-28	10-12
	mean	<u>34.65</u>	<u>53.72</u>	<u>62.79</u>	<u>68.03</u>	<u>69.93</u>
females	age group	10-12	1-3	5-7	25-28	15-18
	mean	<u>36.44</u>	<u>36.52</u>	<u>45.99</u>	<u>54.24</u>	<u>60.43</u>

Normal Strain Tested With DDT

males	age group	15-18	10-12	5-7	1-3	25-28
	mean	<u>38.96</u>	<u>40.82</u>	<u>43.34</u>	<u>43.71</u>	<u>62.48</u>
females	age group	15-18	5-7	10-12	1-3	25-28
	mean	<u>30.25</u>	<u>33.22</u>	<u>33.30</u>	<u>35.41</u>	<u>51.98</u>

Aldrin-Resistant Strain Tested With Aldrin

males	age not significant					
females	age group	10-12	5-7	1-3	15-18	25-28
	mean	<u>28.26</u>	<u>28.63</u>	<u>35.74</u>	<u>36.31</u>	<u>47.66</u>

Any two means not underscored by the same line are significantly different. Any two means underscored by the same line are not significantly different.

FIGURE 1

The Response of Normal Strain German Cockroaches When Tested With
Malathion.

- A. First egg case progeny, 1-3 days of age
- B. First egg case progeny, 5-7 days of age
- C. First egg case progeny, 10-12 days of age
- D. First egg case progeny, 15-18 days of age
- E. First egg case progeny, 25-28 days of age
- F. Second egg case progeny, 10-12 days of age
- G. Second egg case progeny, 15-18 days of age
- H. Second egg case progeny, 25-28 days of age
- I. Third egg case progeny, 15-18 days of age
- J. Third egg case progeny, 25-28 days of age

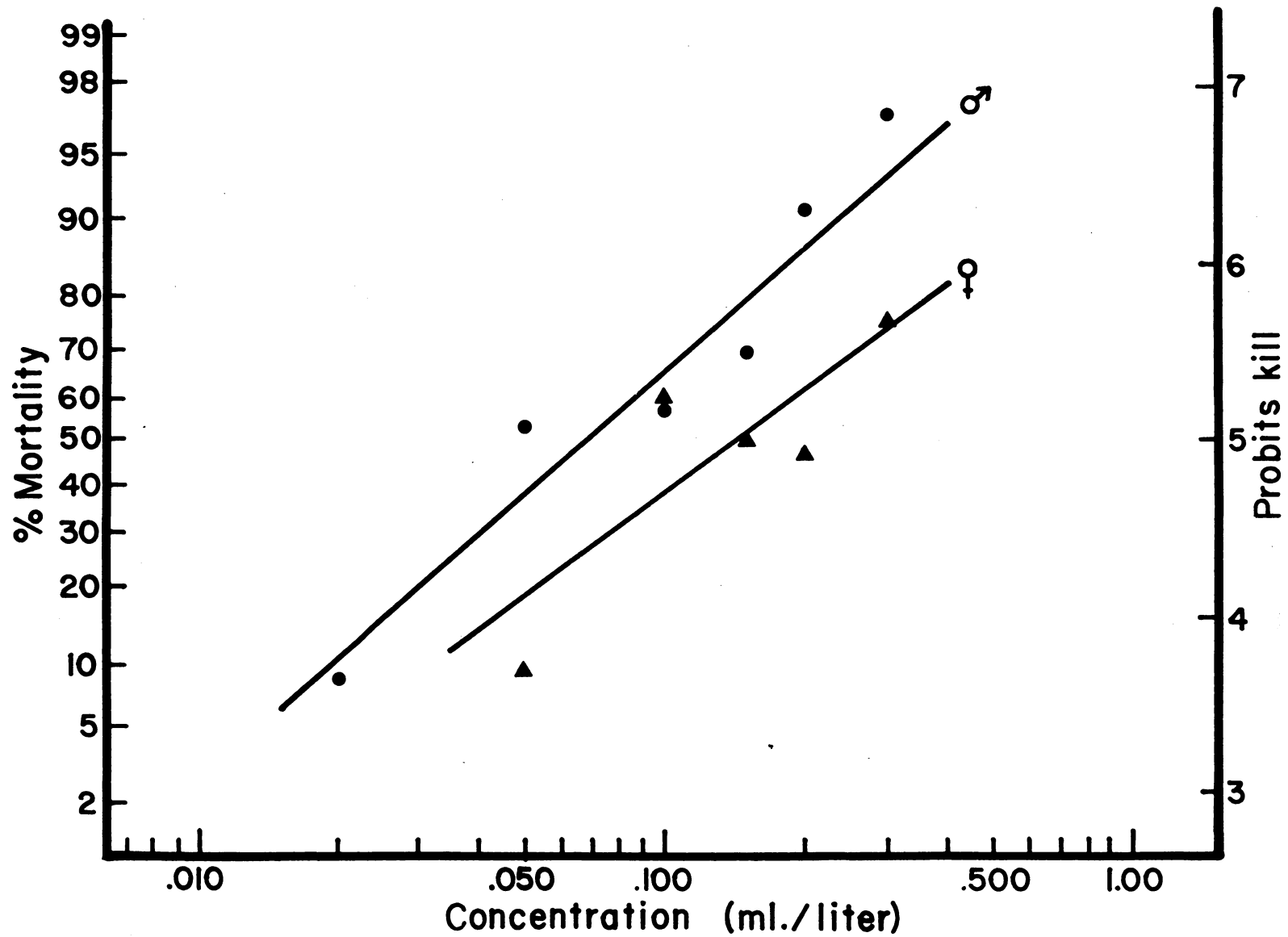


Figure 1 A

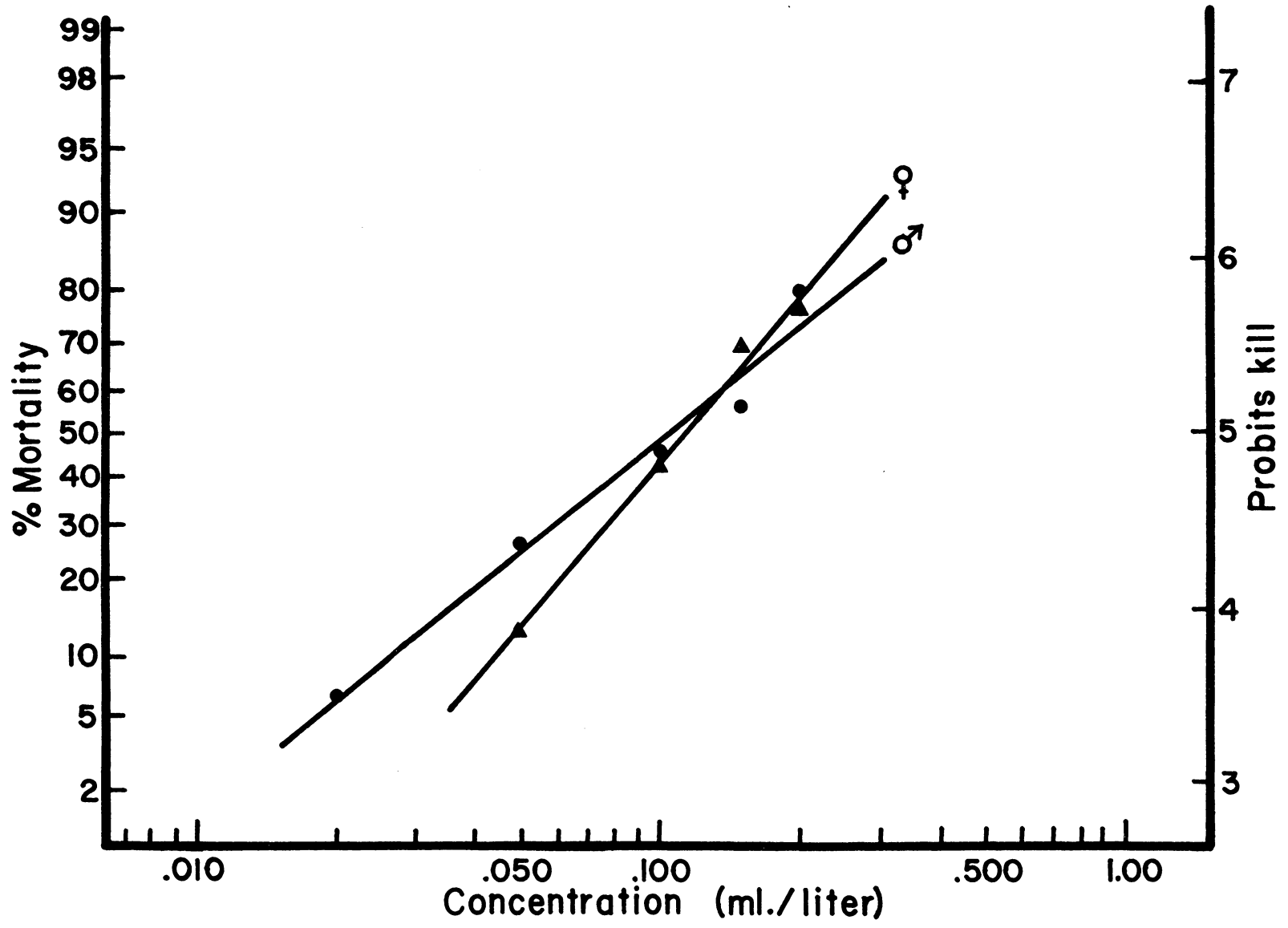


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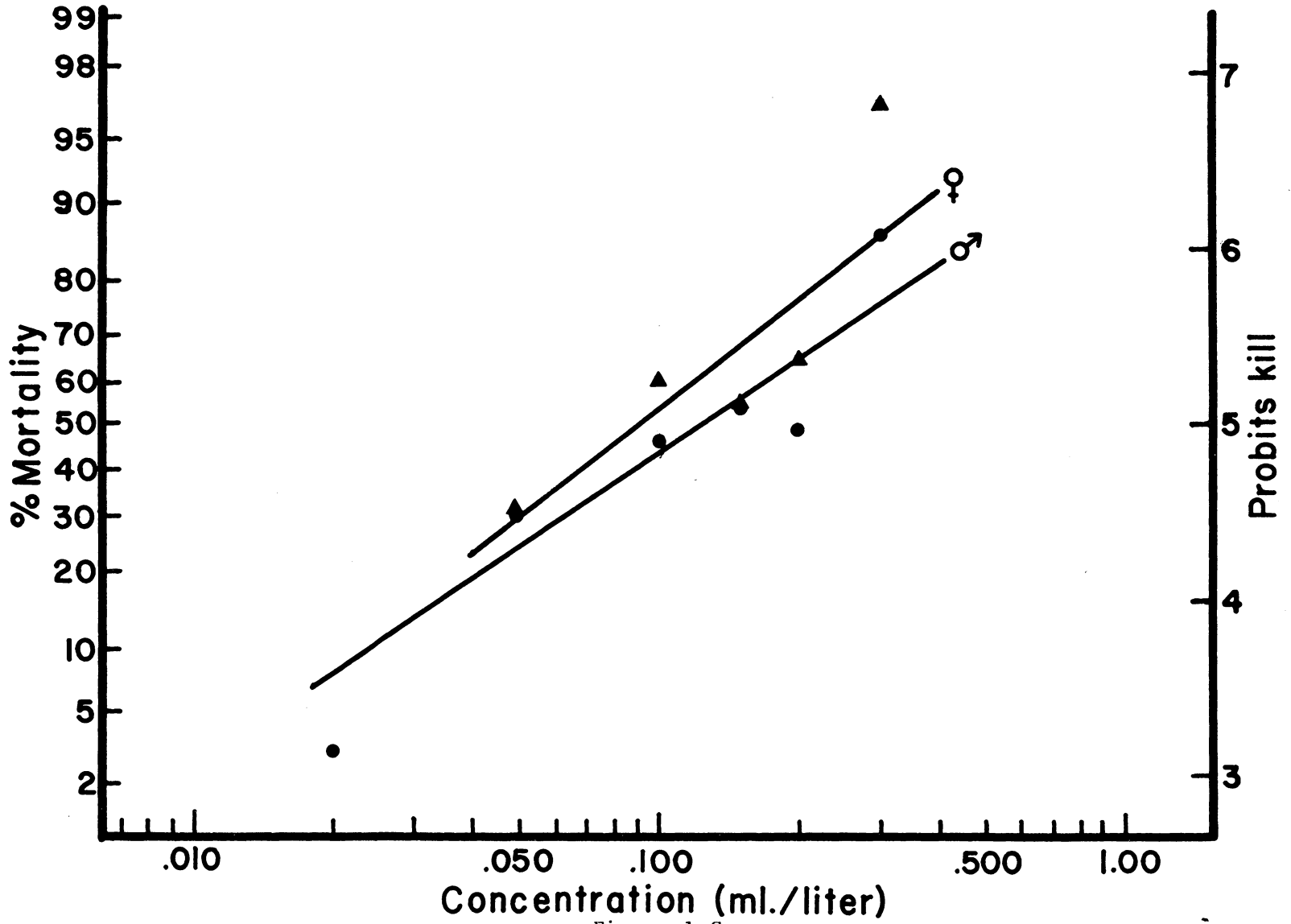


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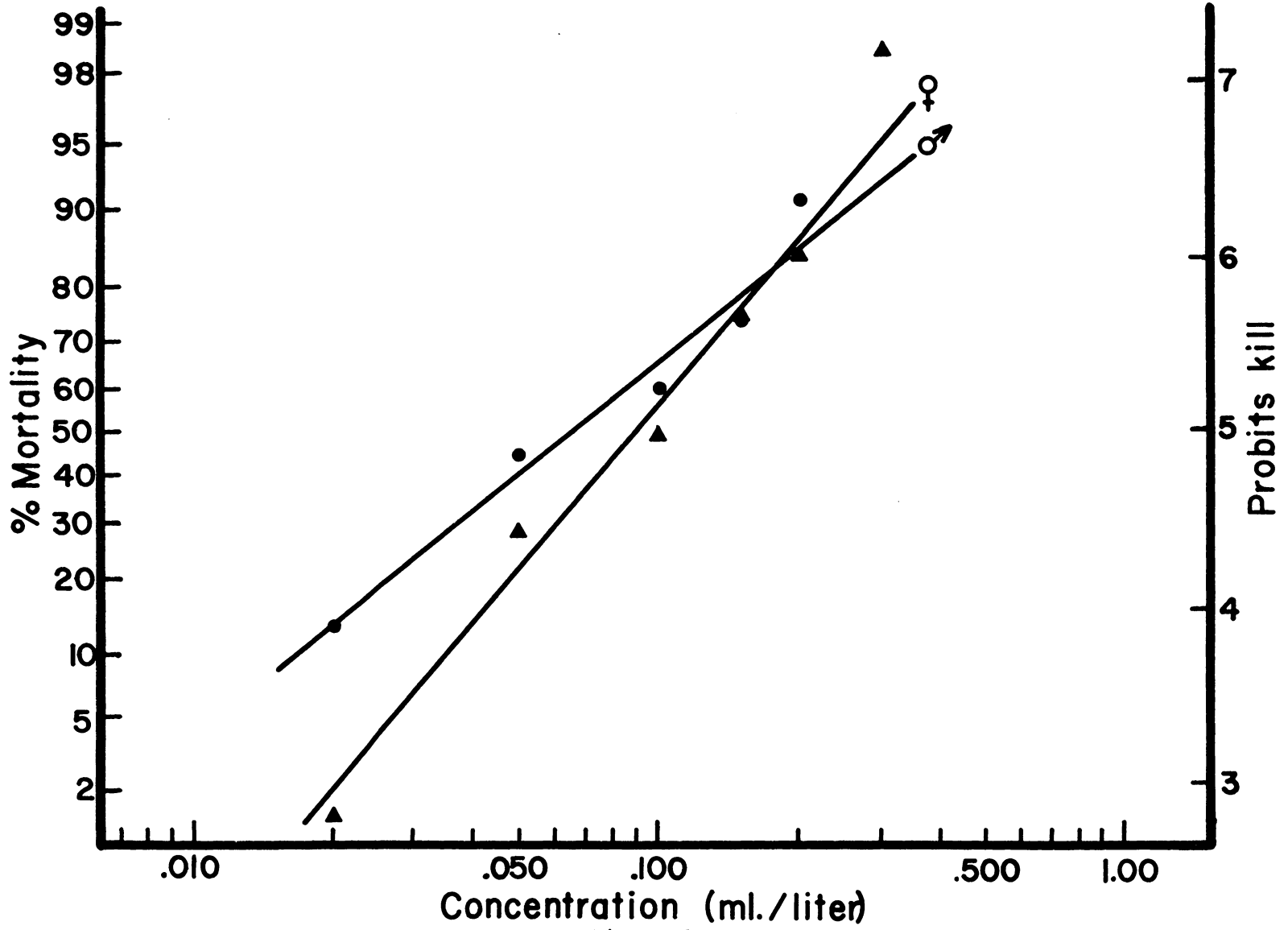


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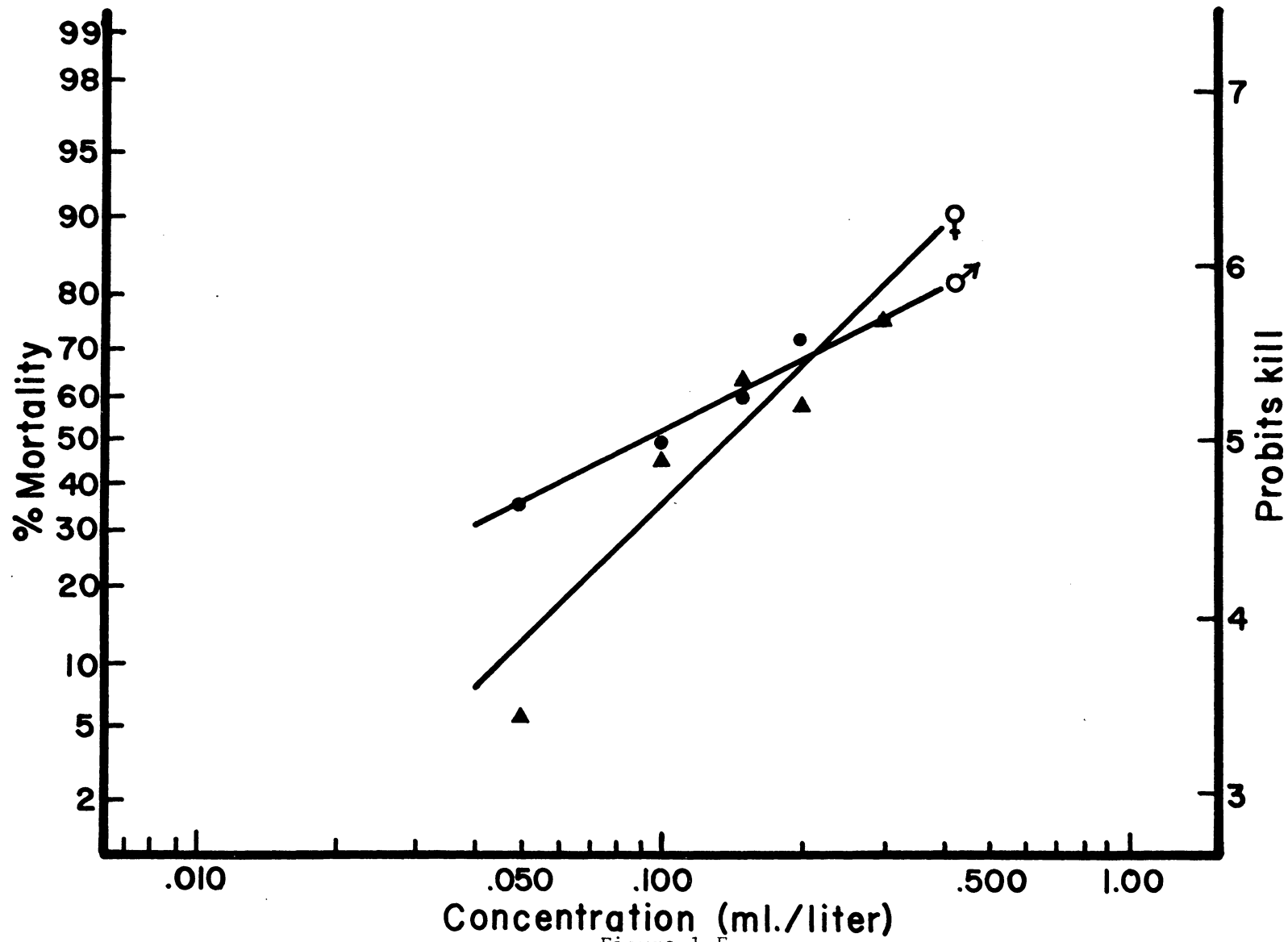


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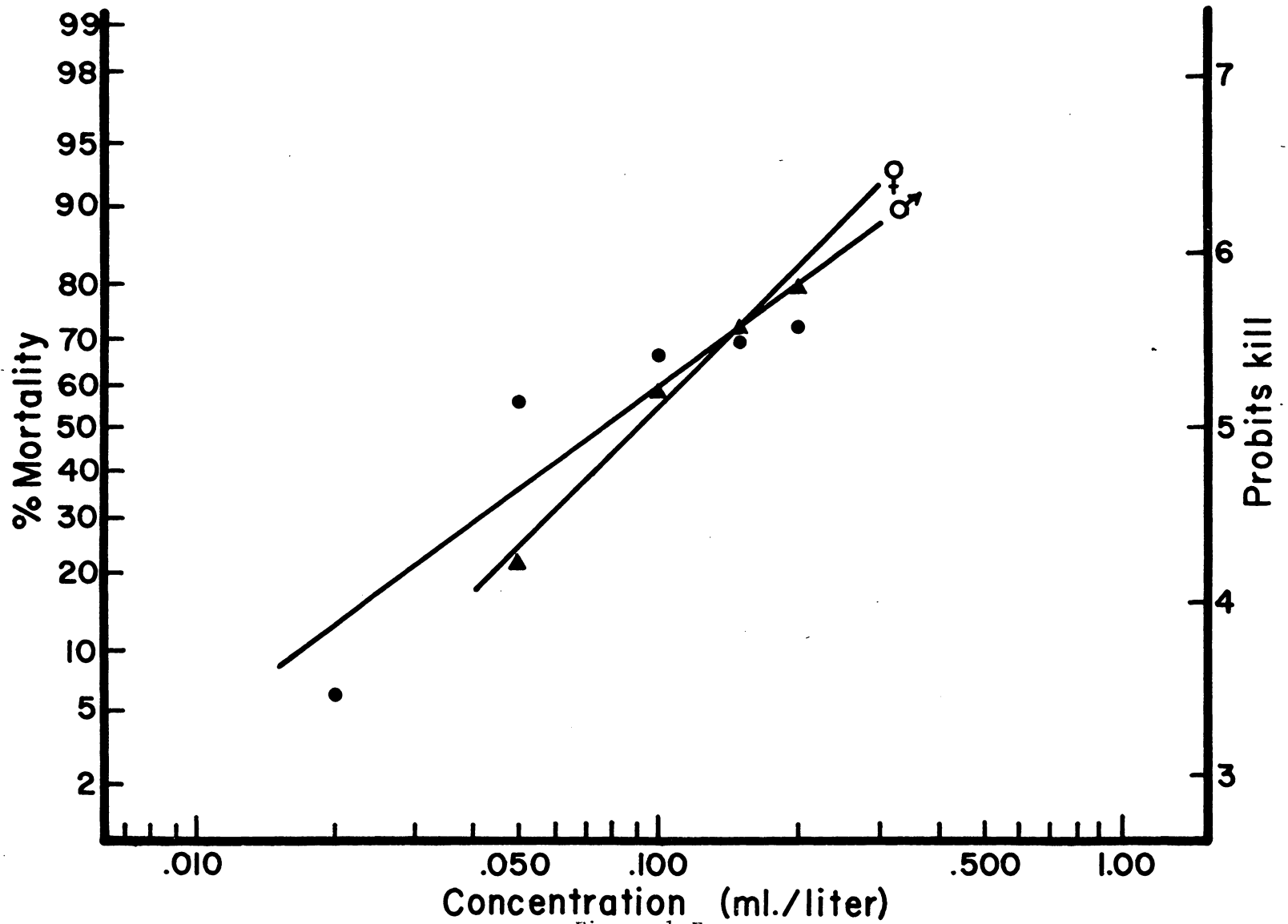


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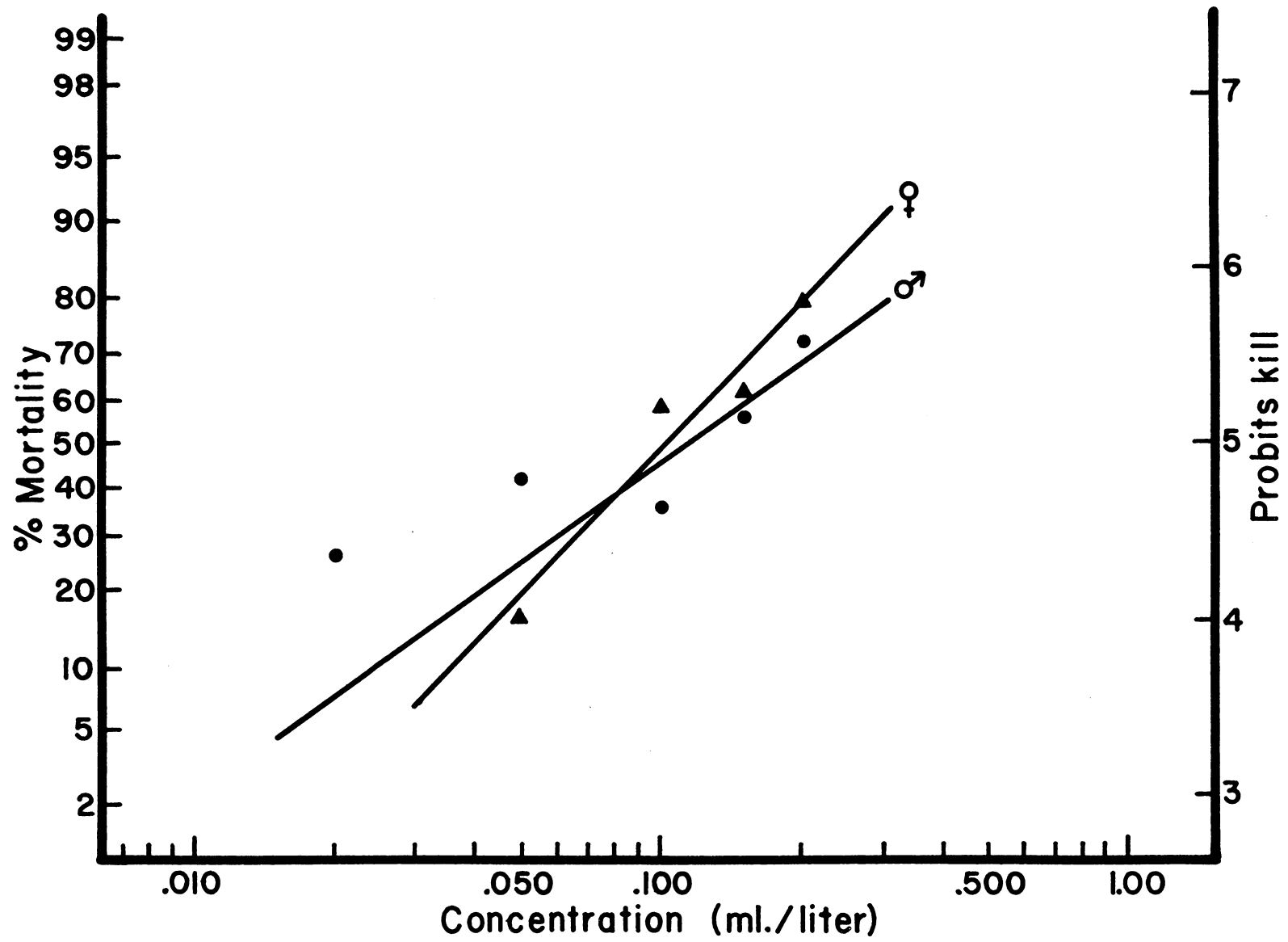


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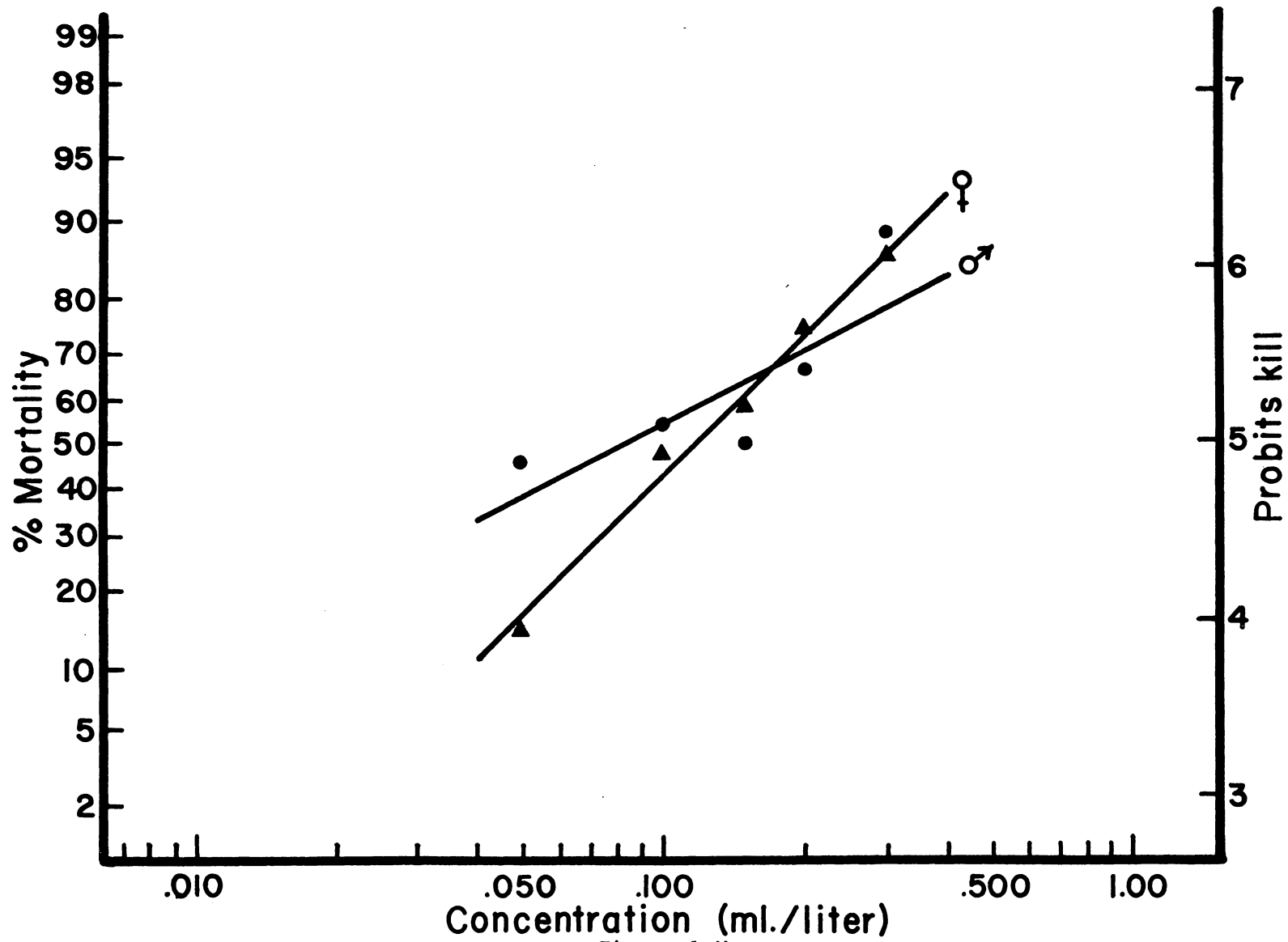


Figure 1 H

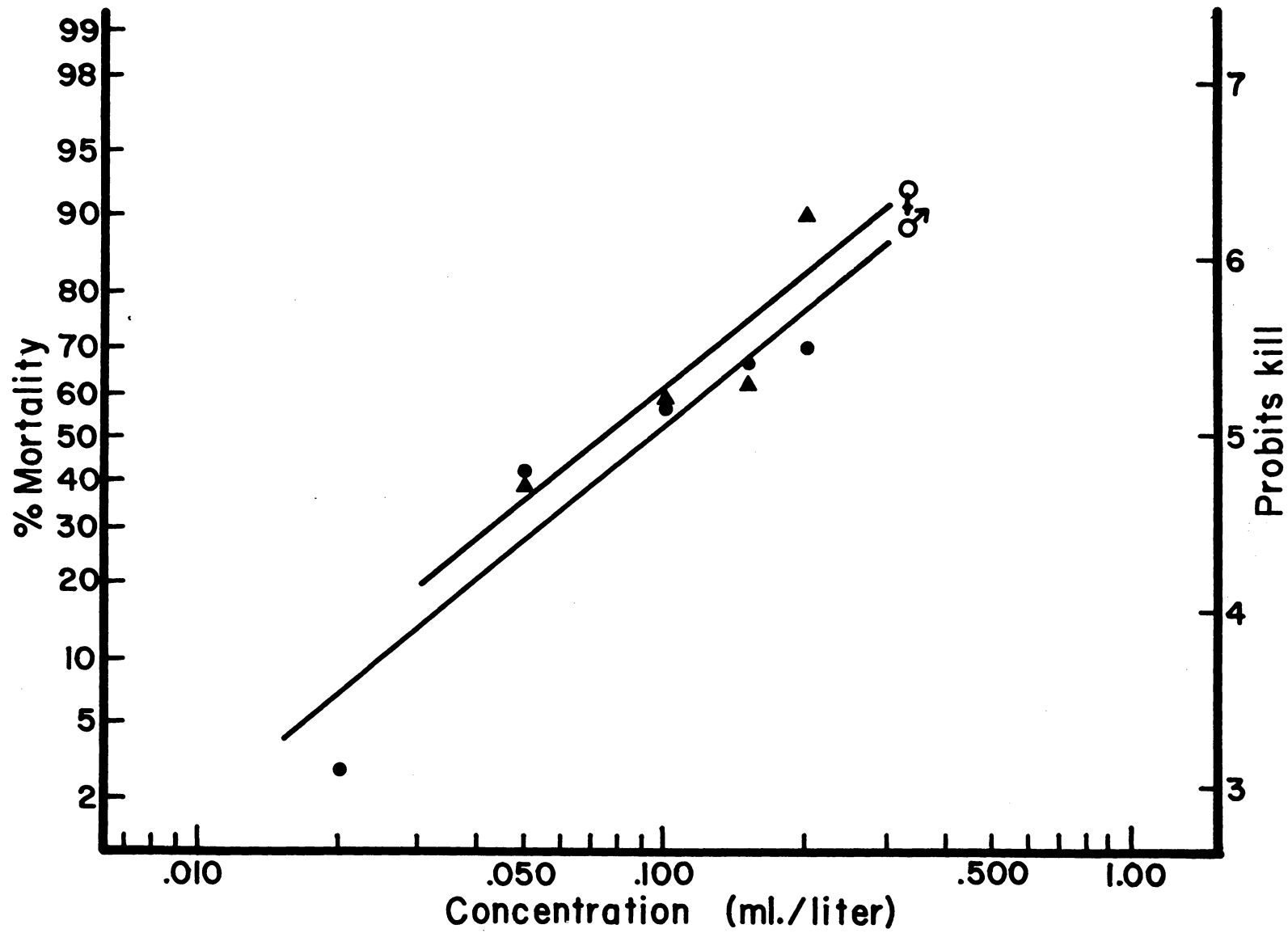


Figure 1 I

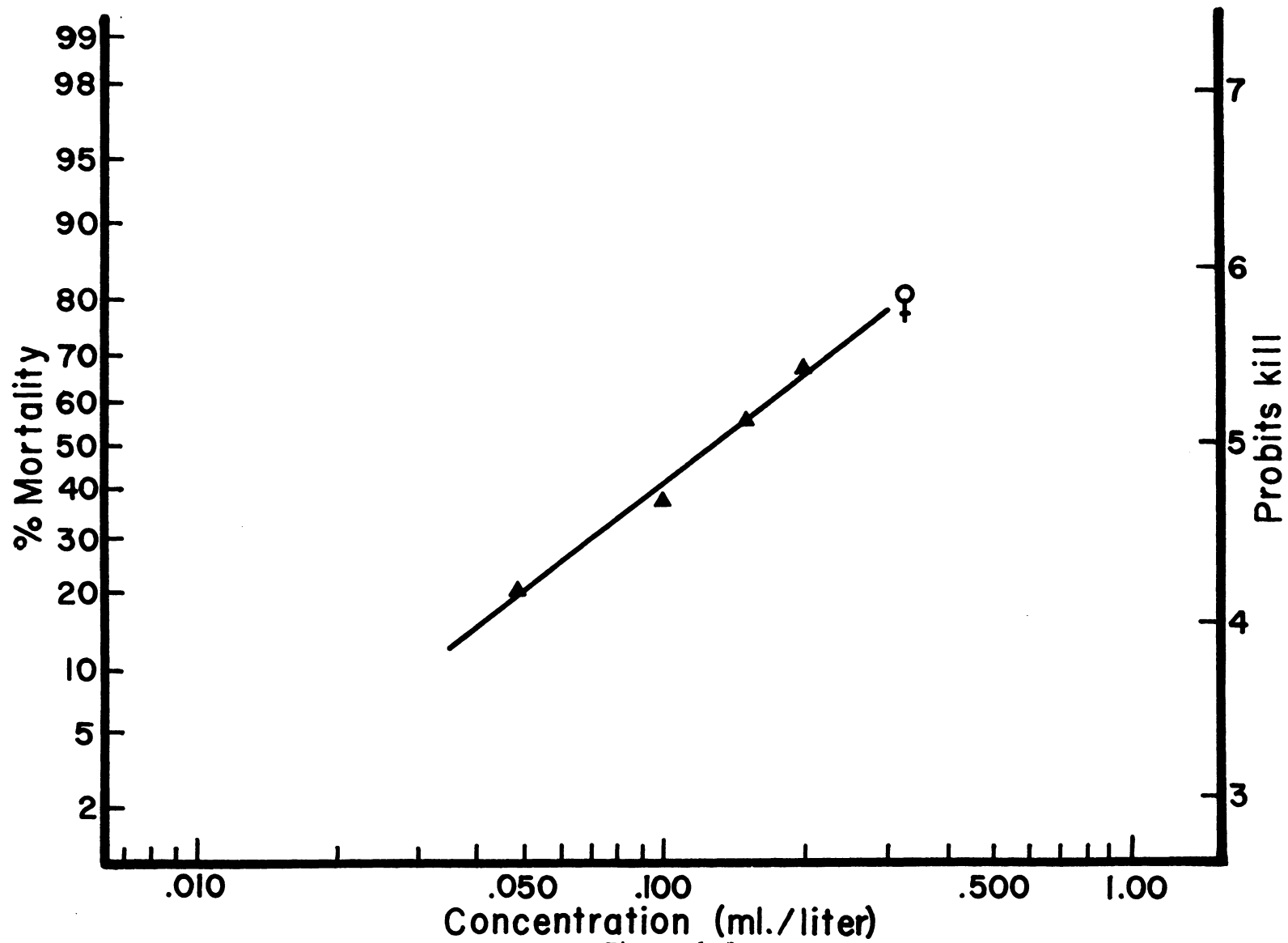


Figure 1 J

Table 2

The Response of Normal Strain German Cockroaches When Tested With Malathion

Egg Case Number	Sex		Age of Adults-Days				
			1-3	5-7	10-12	15-18	25-28
1	male	LC ₅₀ (ml./1.)	0.066	0.103	0.124	0.065	0.090
		Slope	2.30	2.16	1.79	2.13	1.37
	female	LC ₅₀	0.144	0.113	0.090	0.088	0.138
		Slope	1.96	3.18	2.05	3.18	2.64
2	male	LC ₅₀			0.074	0.110	0.080
		Slope			1.96	1.92	1.40
	female	LC ₅₀			0.089	0.100	0.116
		Slope			2.46	2.81	2.66
3	male	LC ₅₀				0.092	
		Slope				2.18	
	female	LC ₅₀				0.073	0.124
		Slope				2.15	2.11

results (Gould, 1948; Hazard, 1945a) in which the younger ages were found to be more susceptible. The three youngest age groups, 1-3, 5-7, and 10-12 days, were found to be the least susceptible when the females were tested with aldrin. There was no significant difference between these age groups, and none between the two older groups (15-18 and 25-28 days) (Table 1). The slopes of the dosage-mortality lines for both males and females varied within normal limits (Figure 2; Table 3). LC_{50} values varied considerably. There was a nine-fold difference between the lowest and the highest LC_{50} values in the males. This difference was about five-fold in the females. Thus it appears that as age increased, the LC_{50} values and the slopes of the dosage-mortality lines generally decreased. These data indicate that there was a significant age effect present; as the roaches grew older they gradually became more susceptible to aldrin.

Both the males and the females of the normal strain responded in the same manner when tested with DDT. There was no significant difference between the first four age groups (Table 1). The roaches in the oldest groups, 25-28 days, were found to be more susceptible to DDT than were the roaches in the younger age groups (Figure 3; Table 4). This is what would be expected, for as the roaches became older they began to die of natural causes. However, Table 4 shows that the 25-28 day age groups of the progeny of the second and third egg cases gave the same response as was found in the younger age groups. These data suggest that susceptibility to DDT in the

FIGURE 2

The Response of Normal Strain German Cockroaches When Tested With
Aldrin

- A. First egg case progeny, 1-3 days of age
- B. First egg case progeny, 5-7 days of age
- C. First egg case progeny, 10-12 days of age
- D. First egg case progeny, 15-18 days of age
- E. First egg case progeny, 25-28 days of age
- F. Second egg case progeny, 10-12 days of age
- G. Second egg case progeny, 15-18 days of age
- H. Second egg case progeny, 25-28 days of age
- I. Third egg case progeny, 25-28 days of age

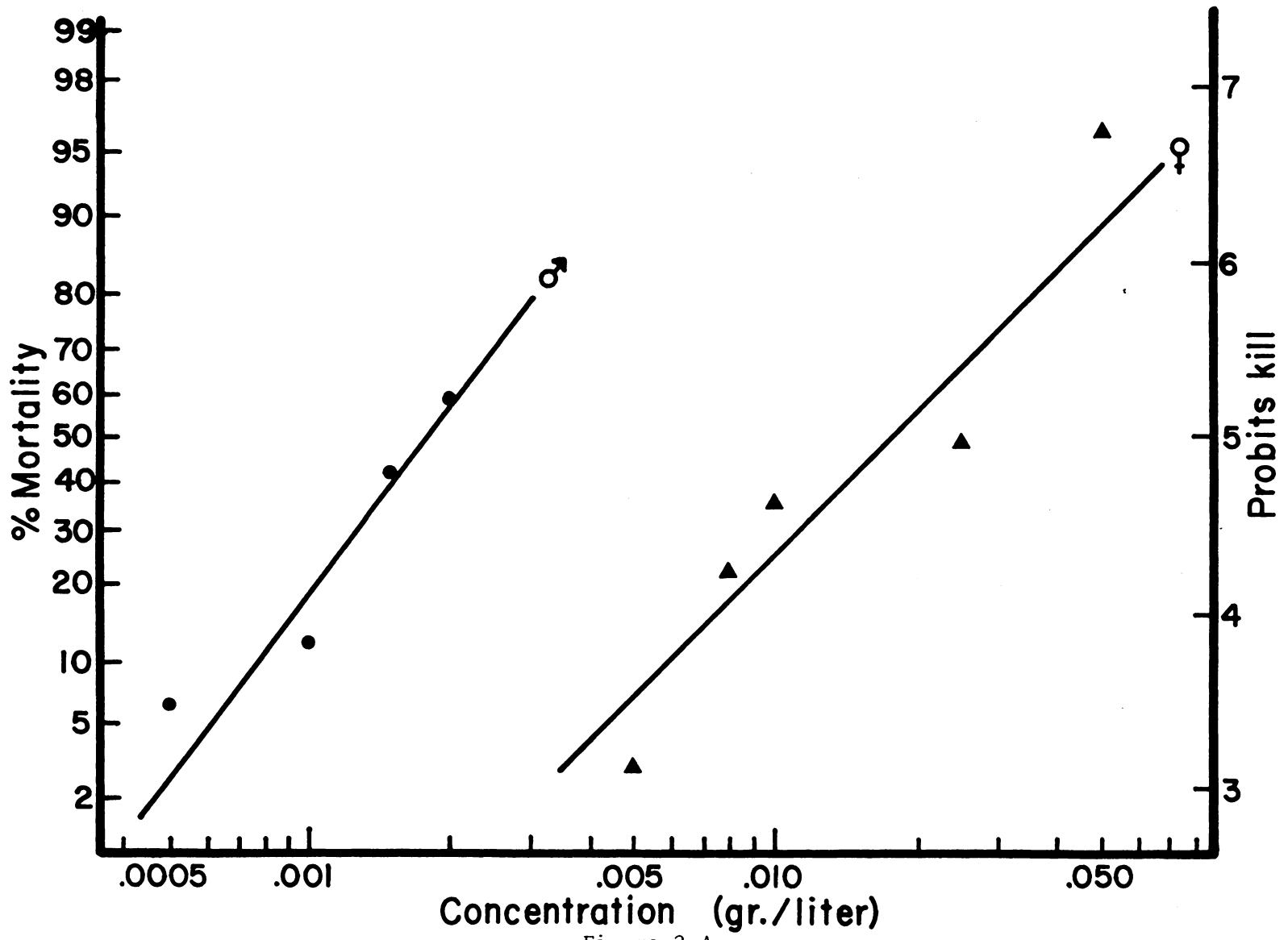


Figure 2 A

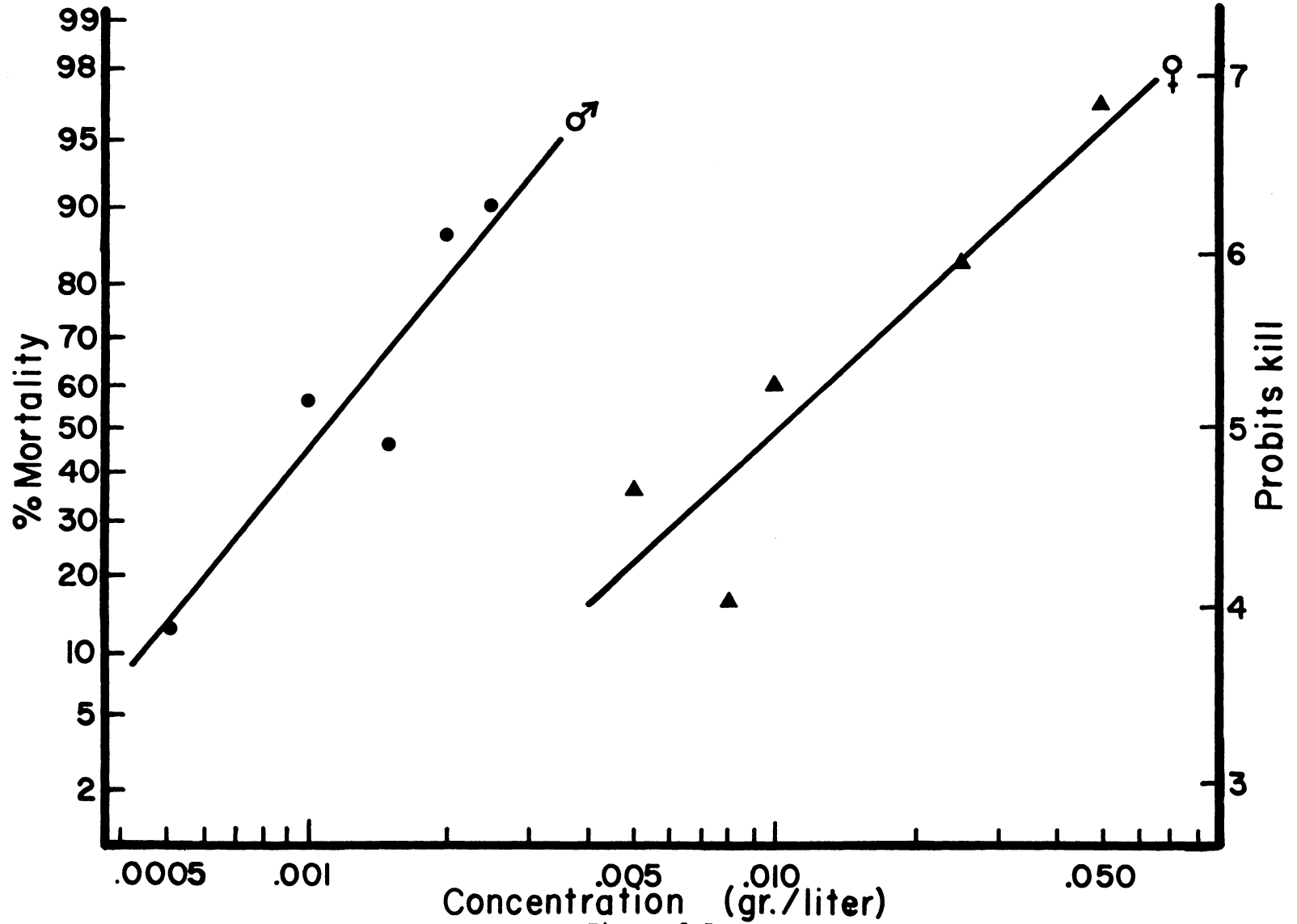


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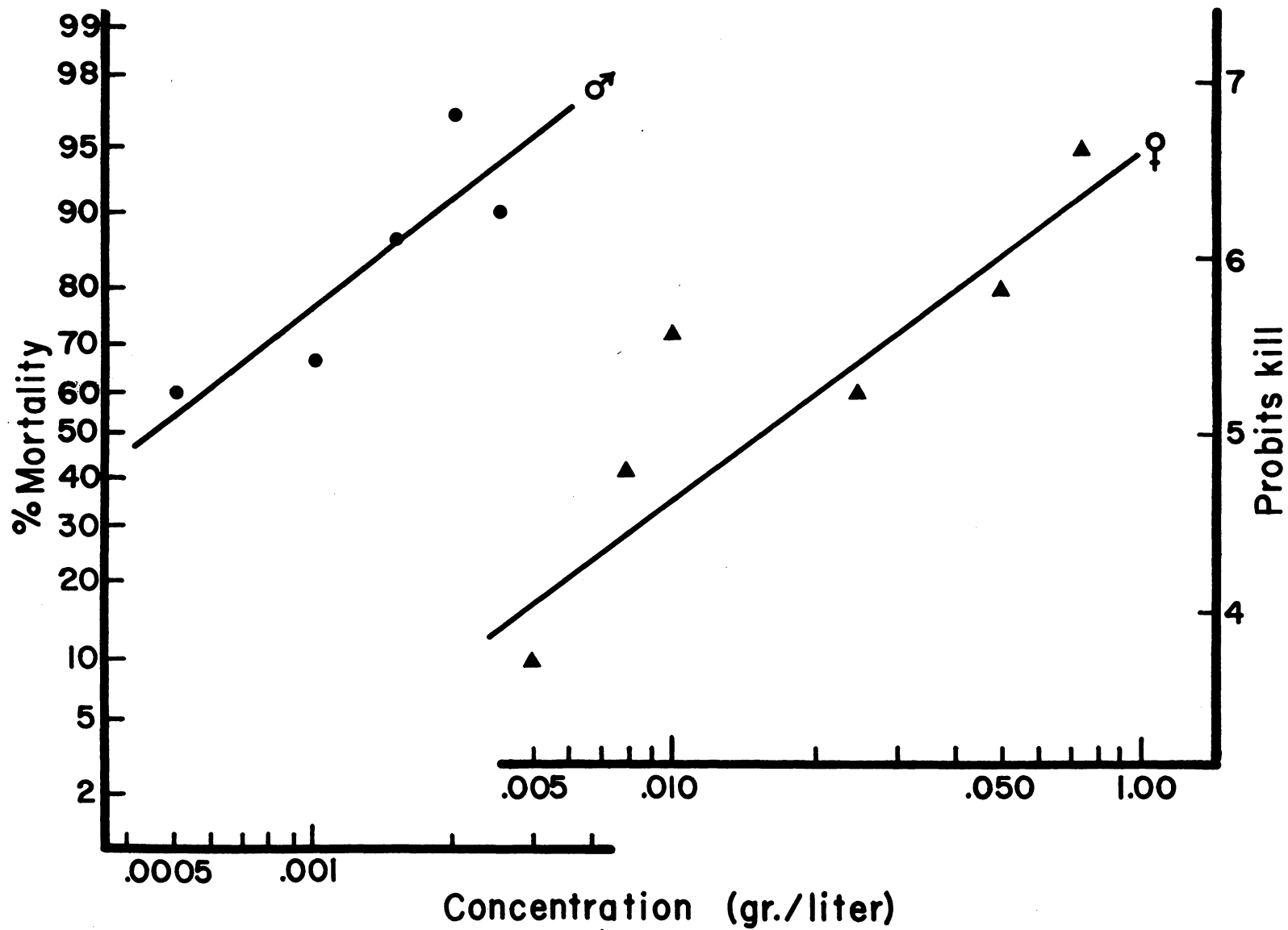


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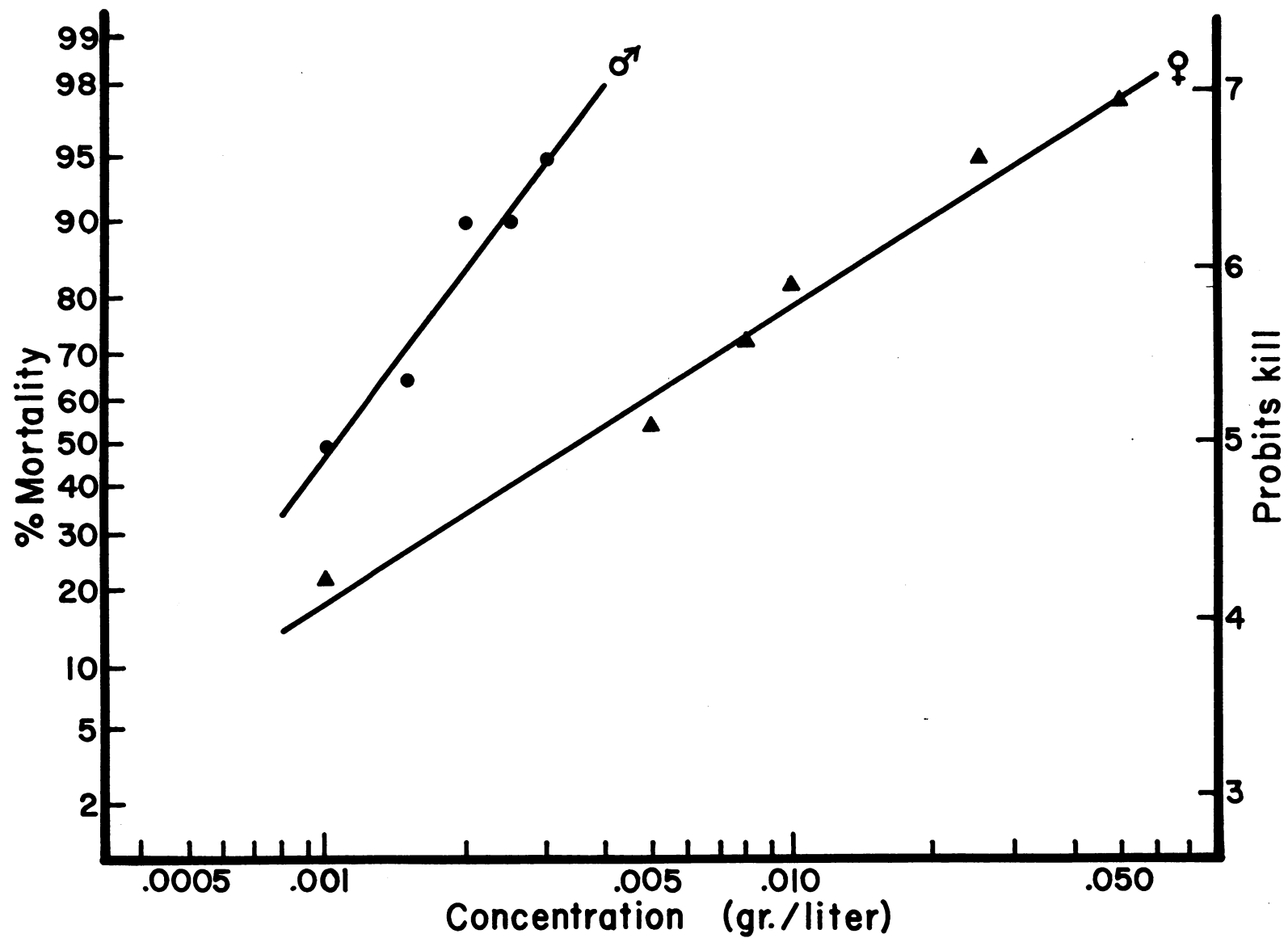


Figure 2 D

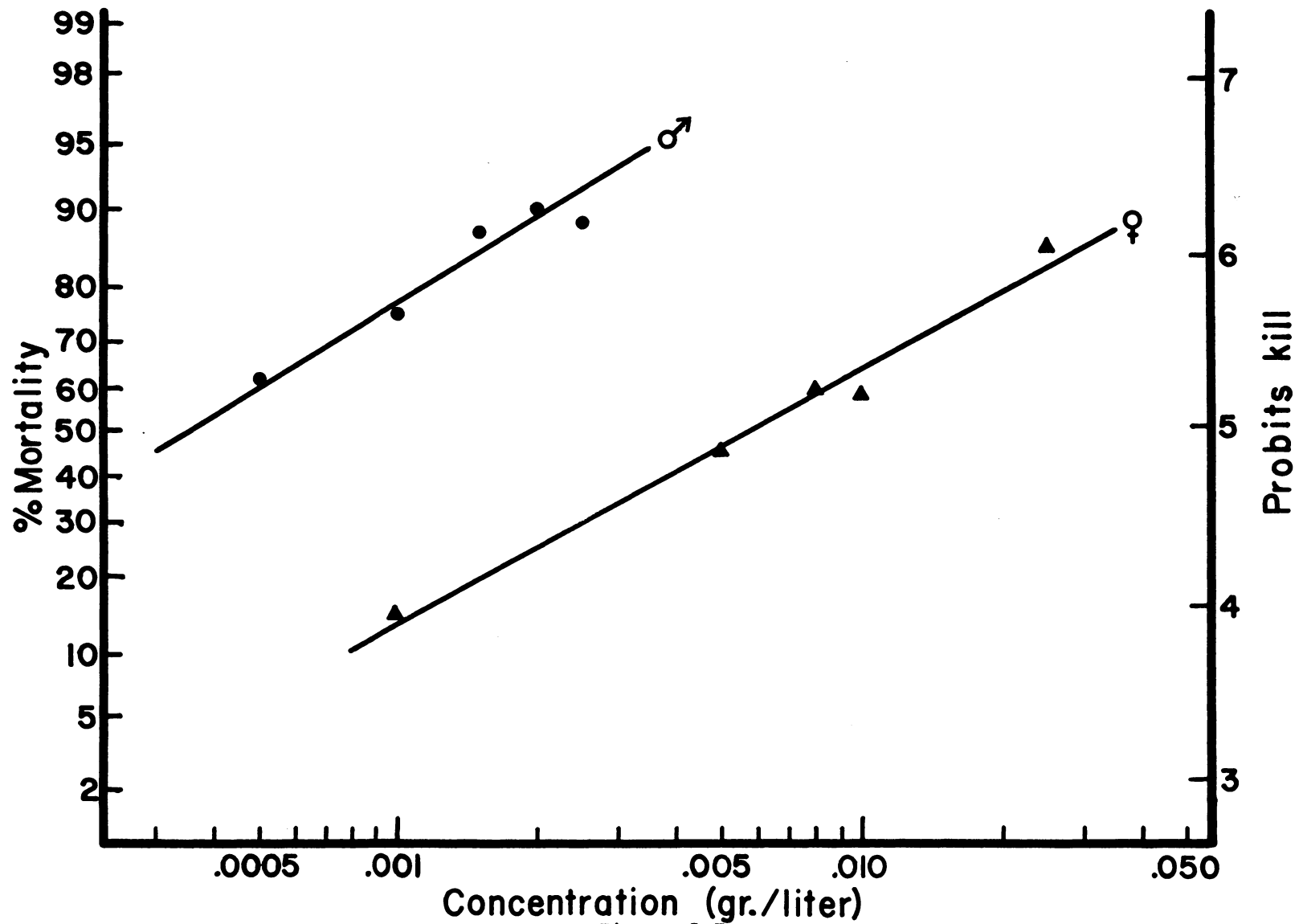


Figure 2 E

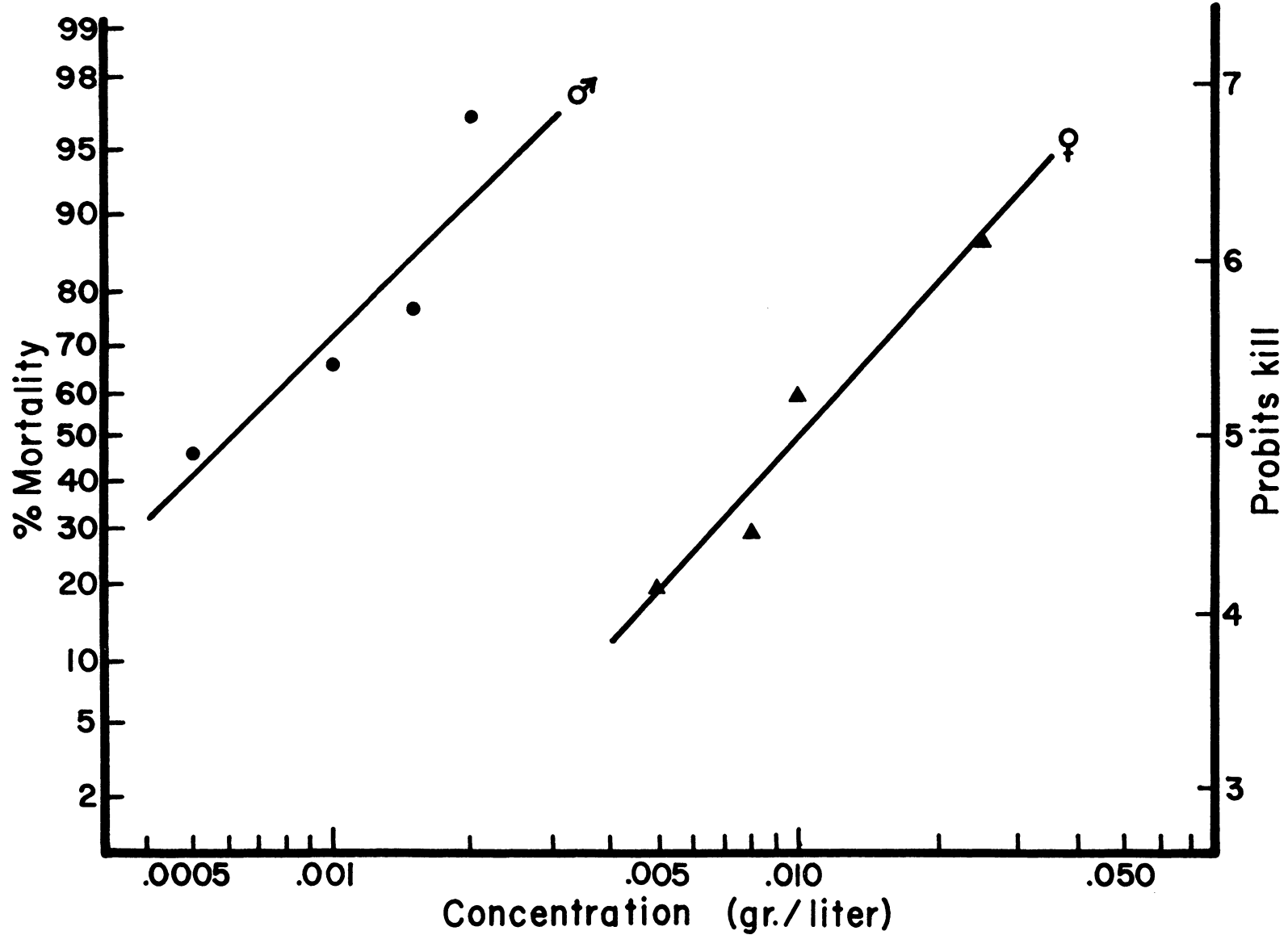


Figure 2 F

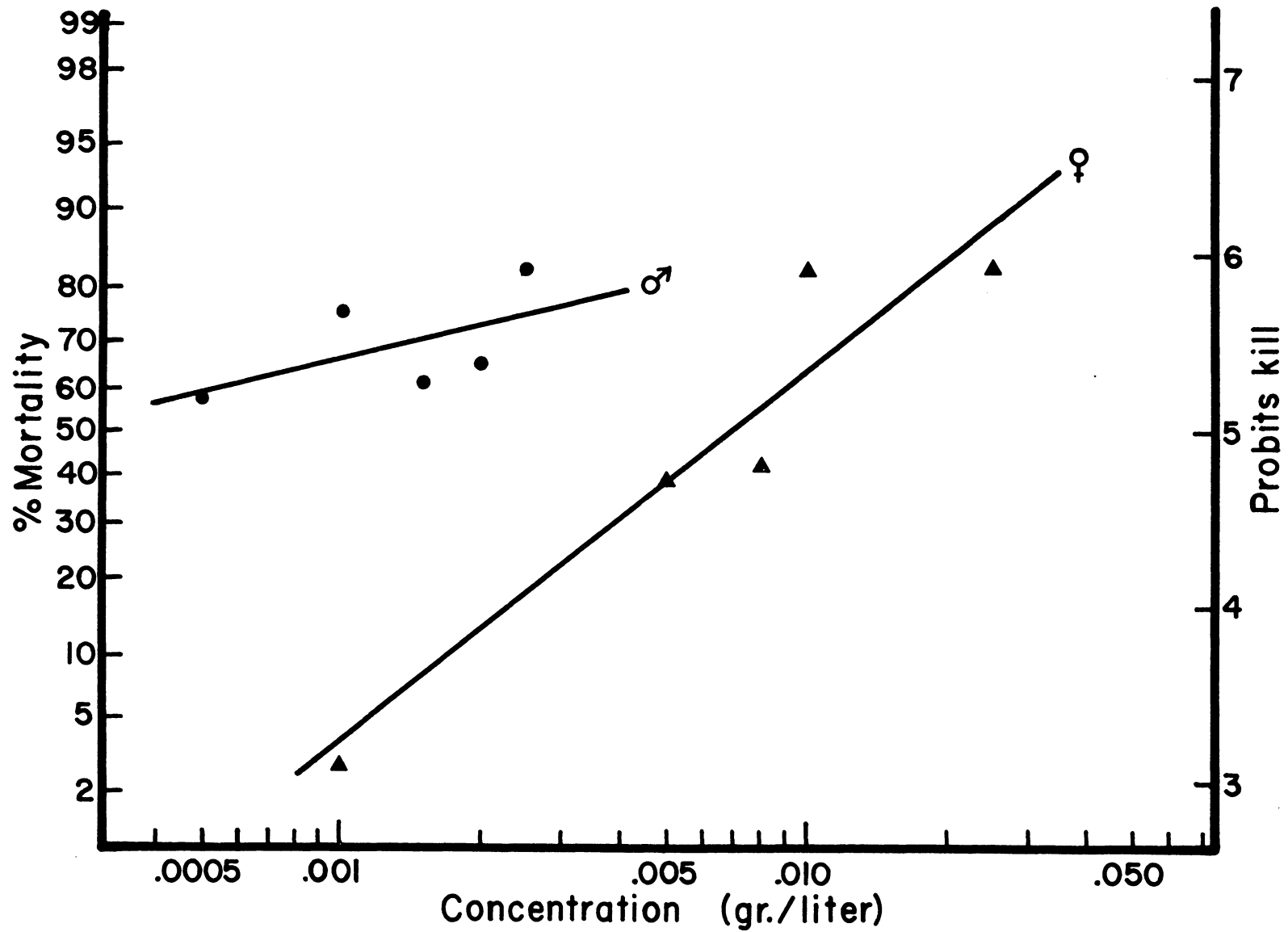


Figure 2 C

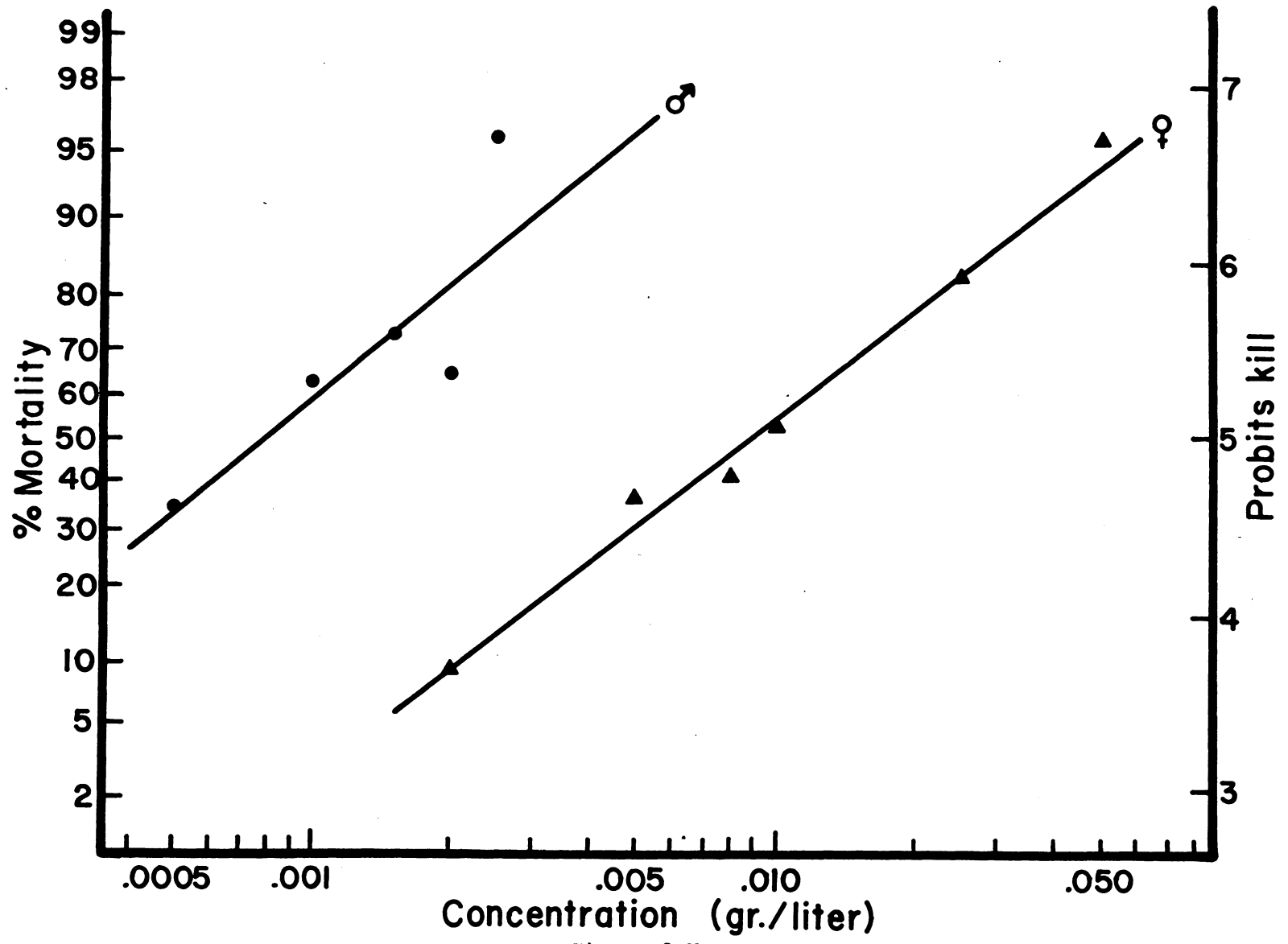


Figure 2 H

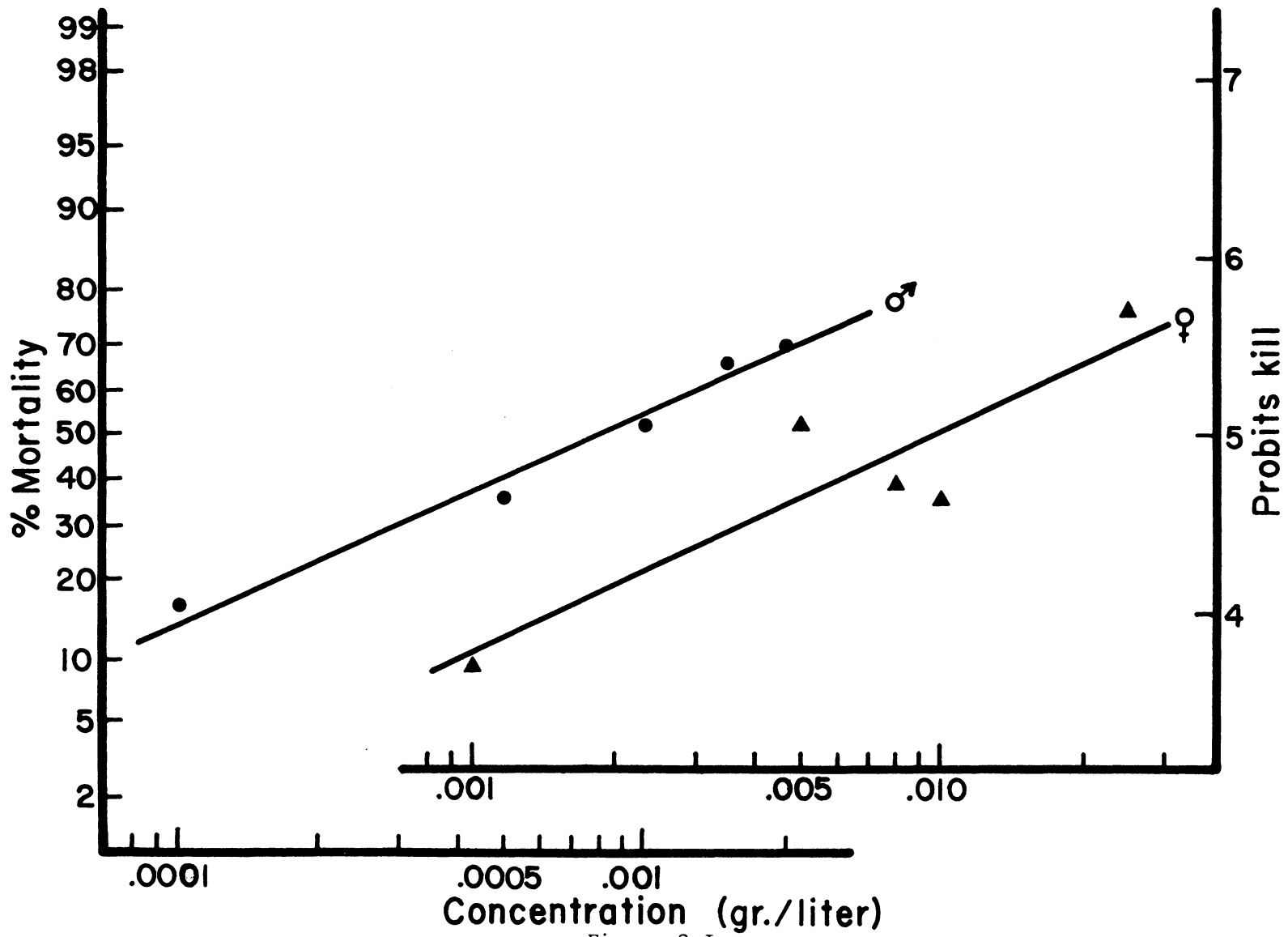


Figure 2 I

Table 3

The Response of Normal Strain German Cockroaches When Tested With Aldrin

Egg Case Number	Sex		Age of Adults-Days				
			1-3	5-7	10-12	15-18	25-28
1	male	LC ₅₀ (gr./l.)	0.0018	0.0011	0.0004	0.0010	0.0003
		Slope	3.47	3.21	2.04	3.59	1.44
	female	LC ₅₀	0.0175	0.0133	0.0155	0.0034	0.0056
		Slope	2.69	2.46	1.62	1.68	1.47
2	male	LC ₅₀			0.0006	0.0002	0.0008
		Slope			2.63	0.60	2.18
	female	LC ₅₀			0.0100	0.0068	0.0085
		Slope			2.93	2.12	2.03
3	male	LC ₅₀					0.0008
		Slope					1.19
	female	LC ₅₀					0.0092
		Slope					1.25

FIGURE 3

Response of Normal Strain German Cockroaches When Tested With
DDT

- A. First egg case progeny, 1-3 days of age
- B. First egg case progeny, 5-7 days of age
- C. First egg case progeny, 10-12 days of age
- D. First egg case progeny, 15-18 days of age
- E. First egg case progeny, 25-28 days of age
- F. Second egg case progeny, 10-12 days of age
- G. Second egg case progeny, 15-18 days of age
- H. Second egg case progeny, 25-28 days of age
- I. Third egg case progeny, 25-28 days of age

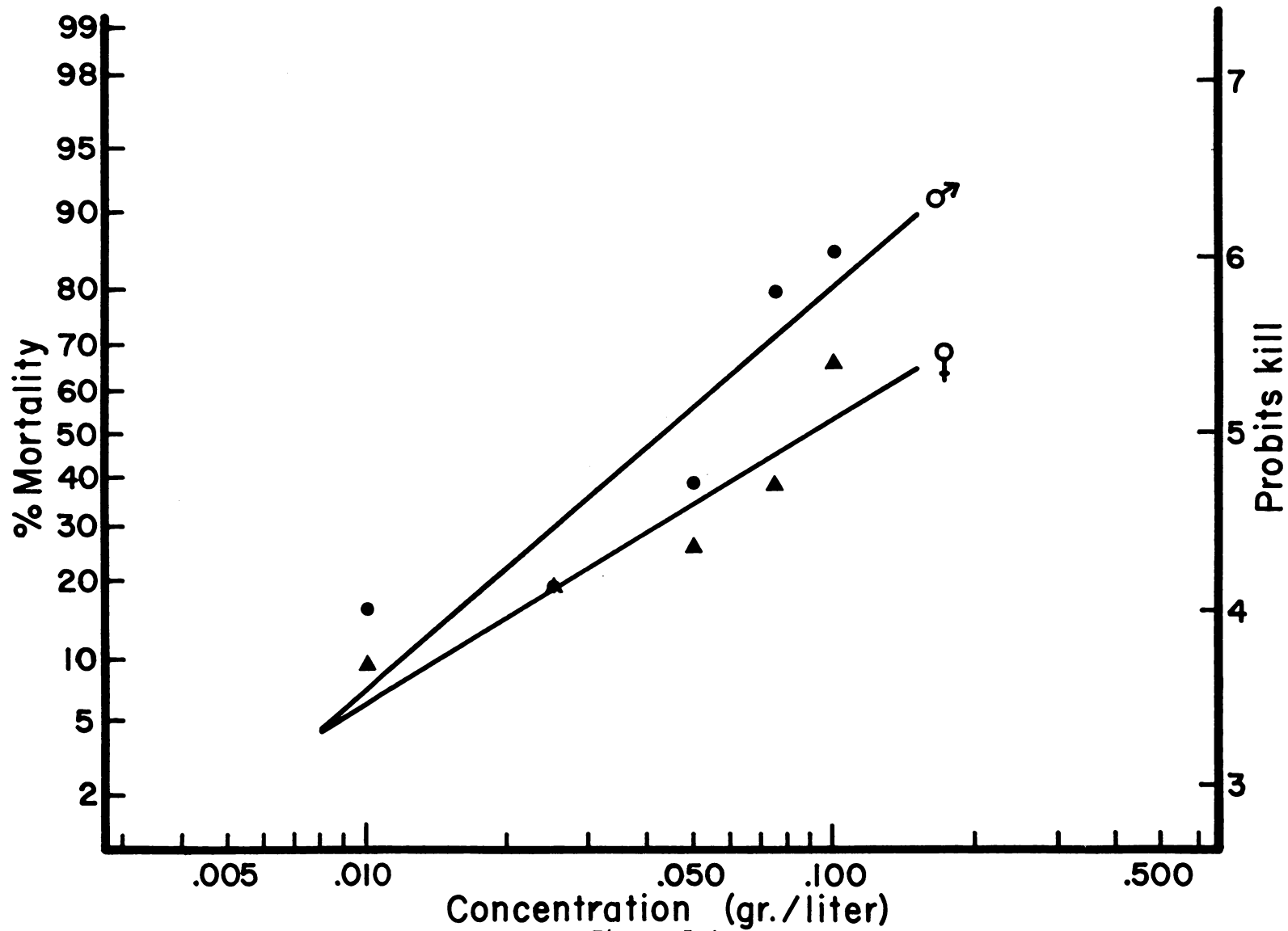


Figure 3 A

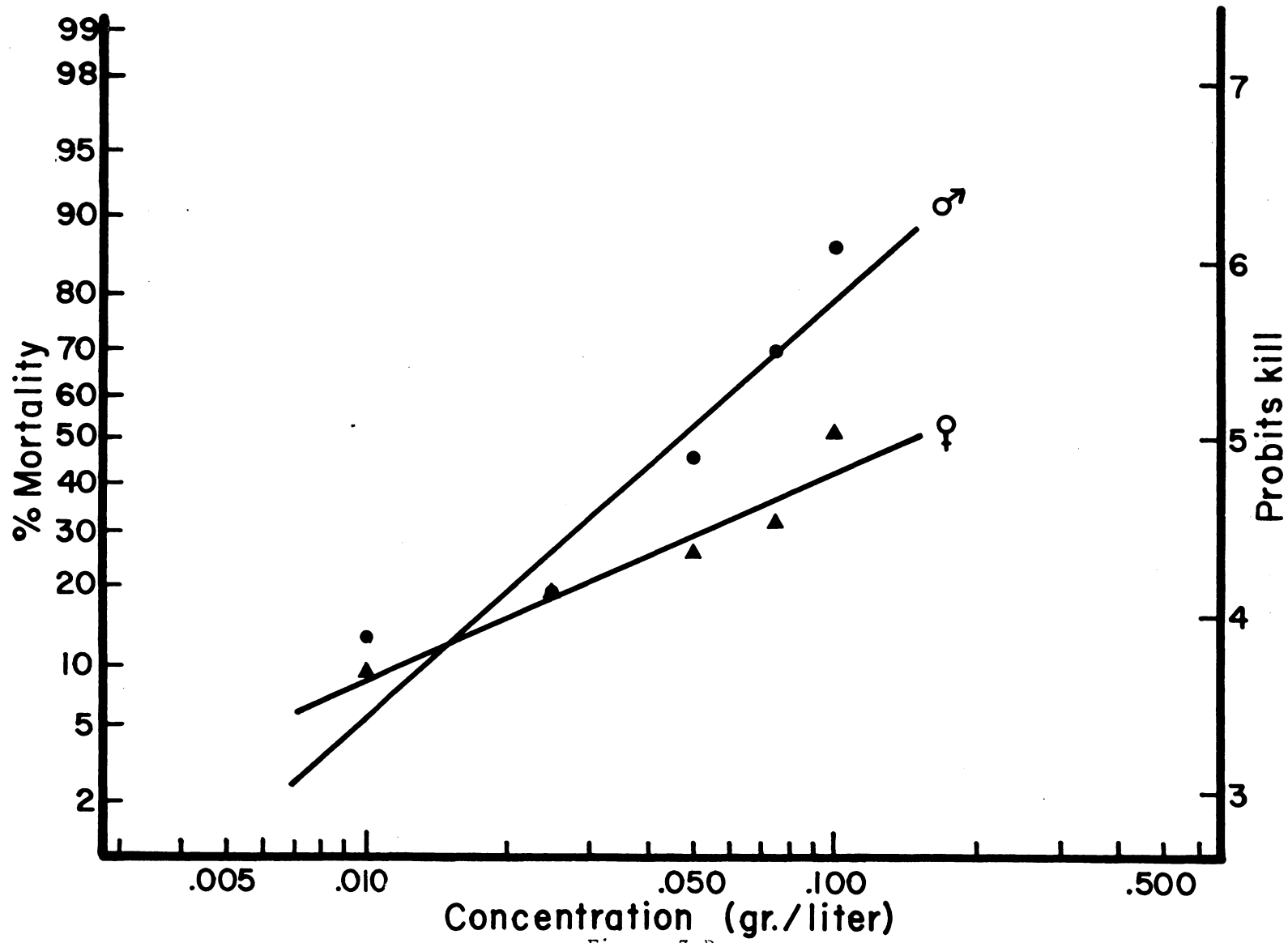


Figure 3 B

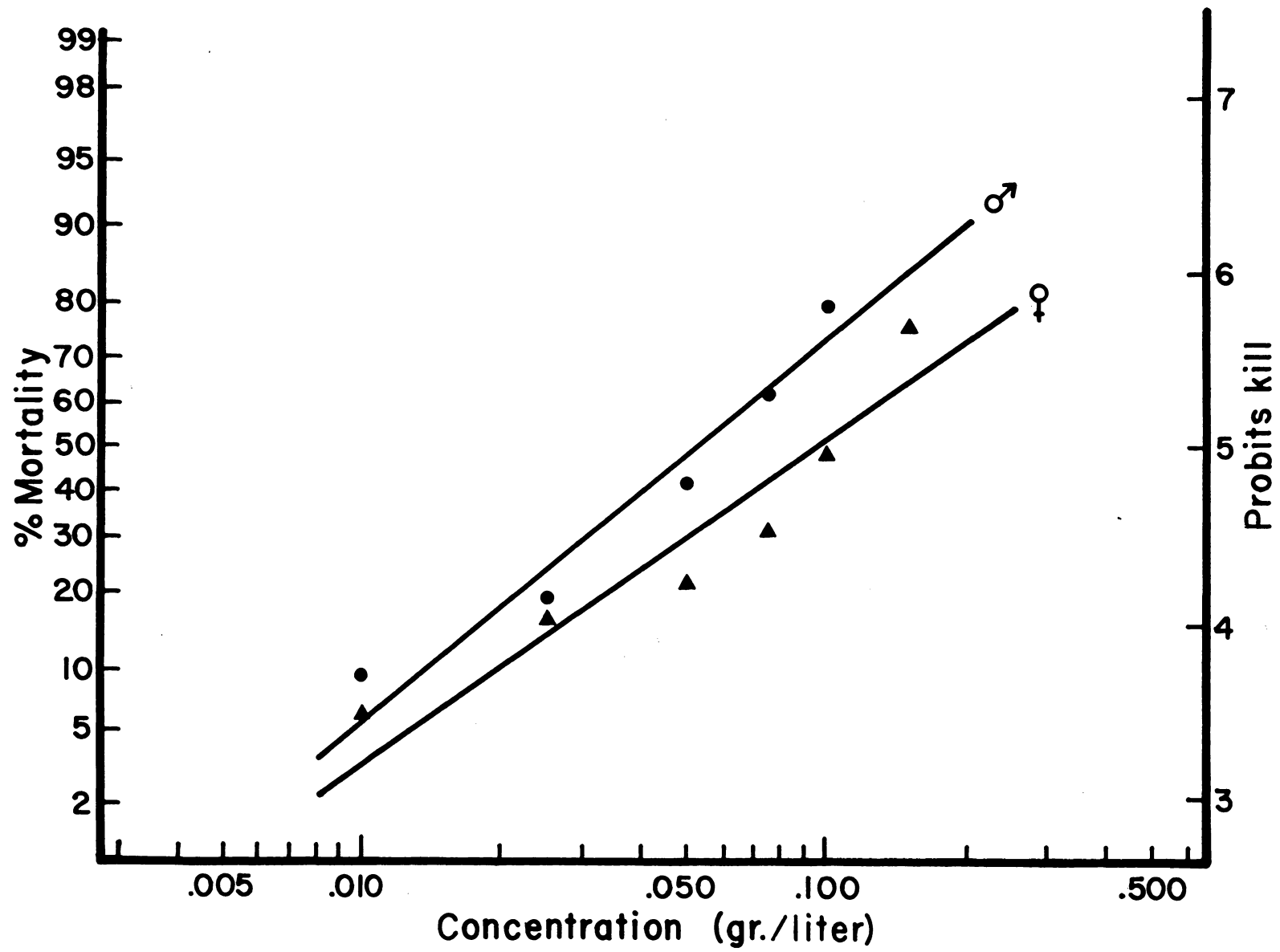


Figure 3 C

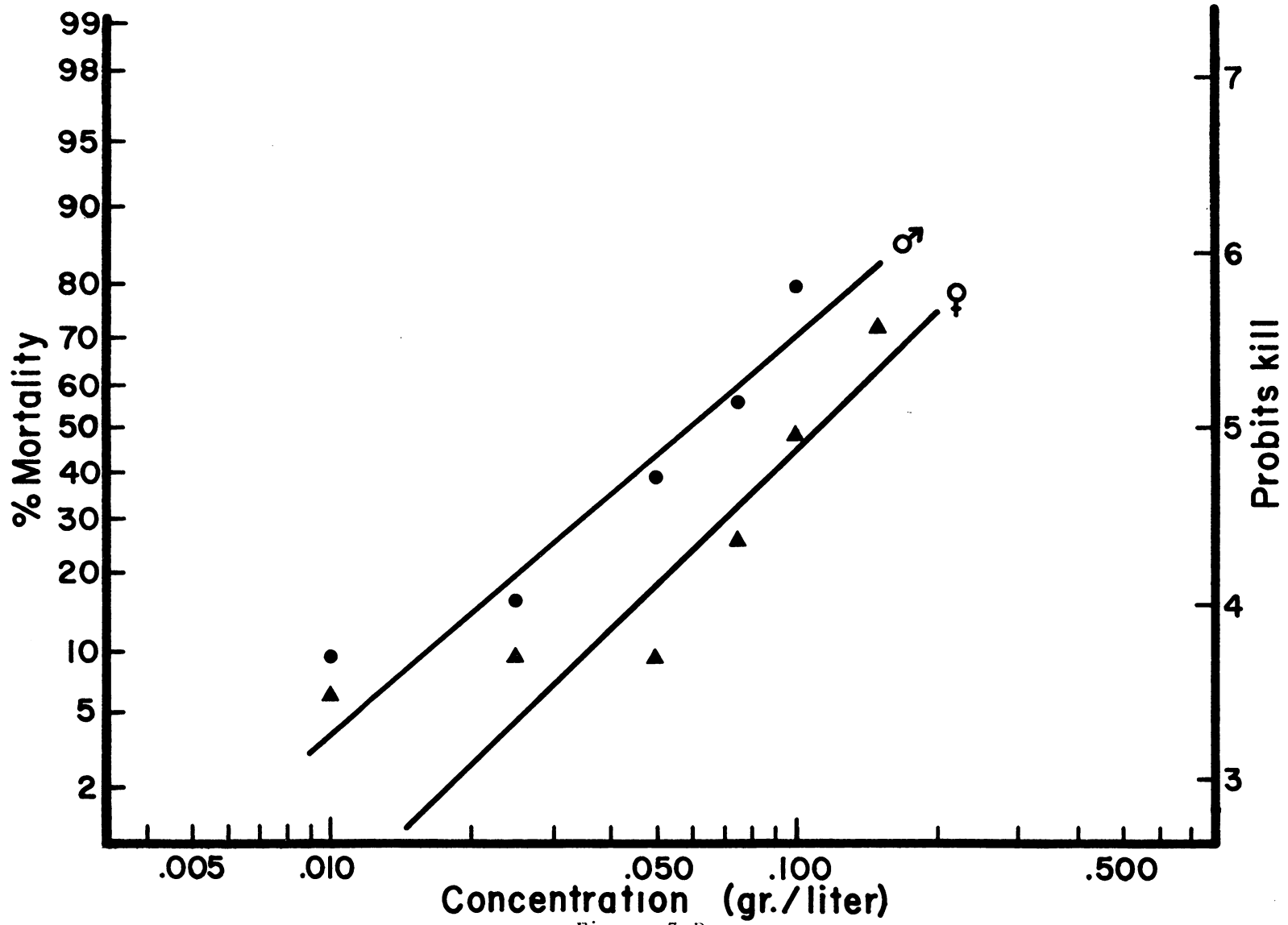


Figure 3 D

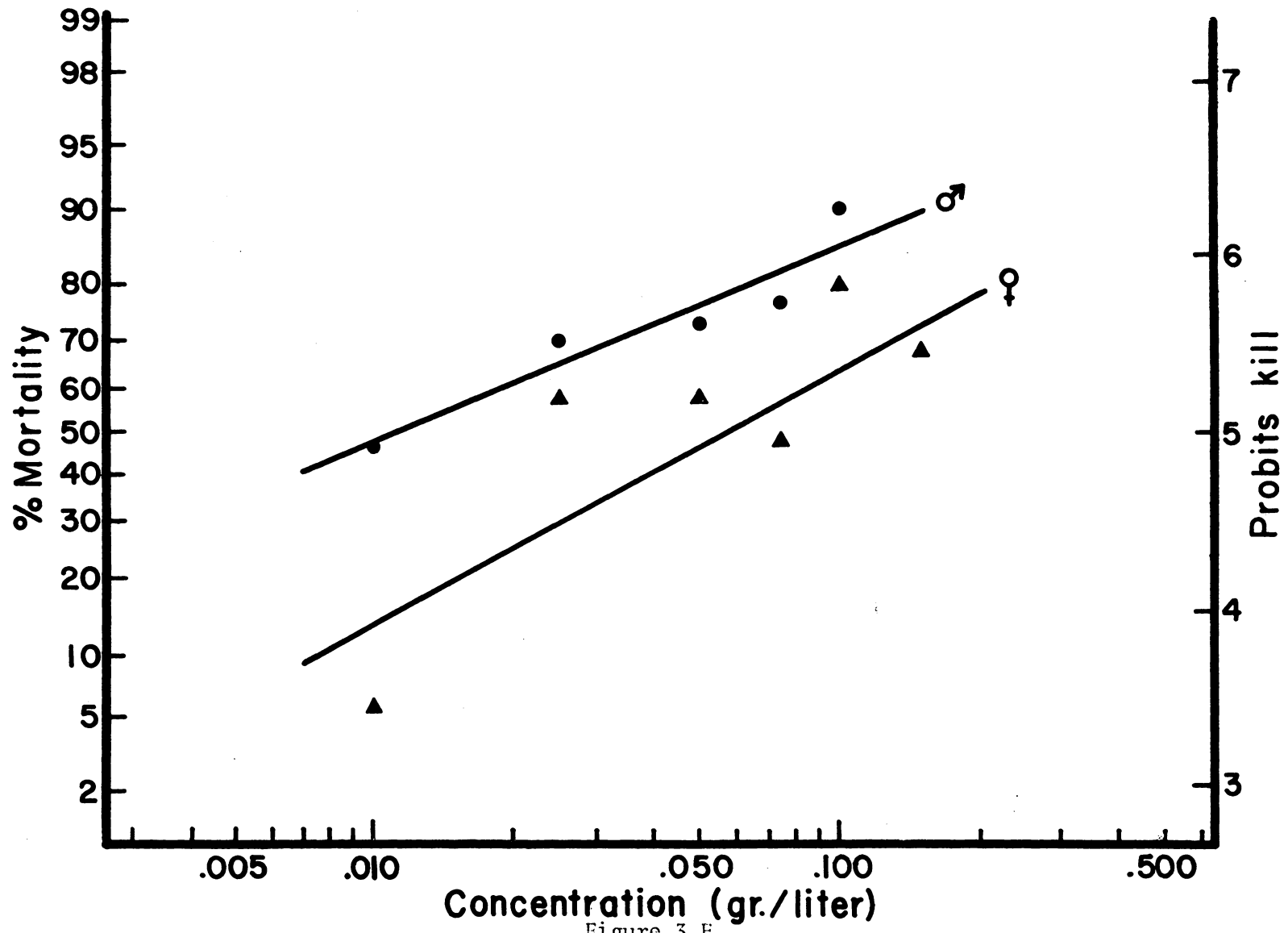


Figure 3 E

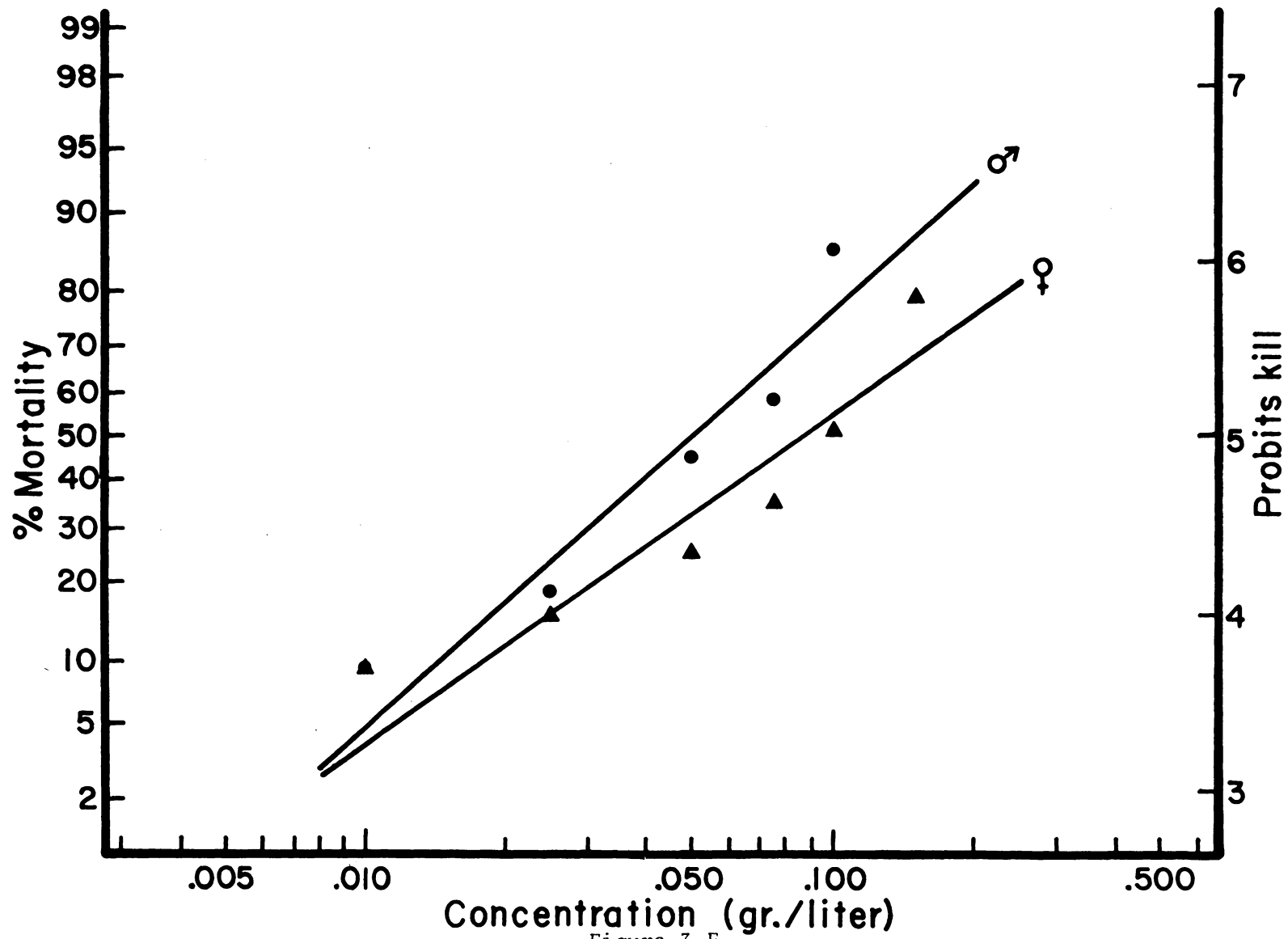


Figure 3 F

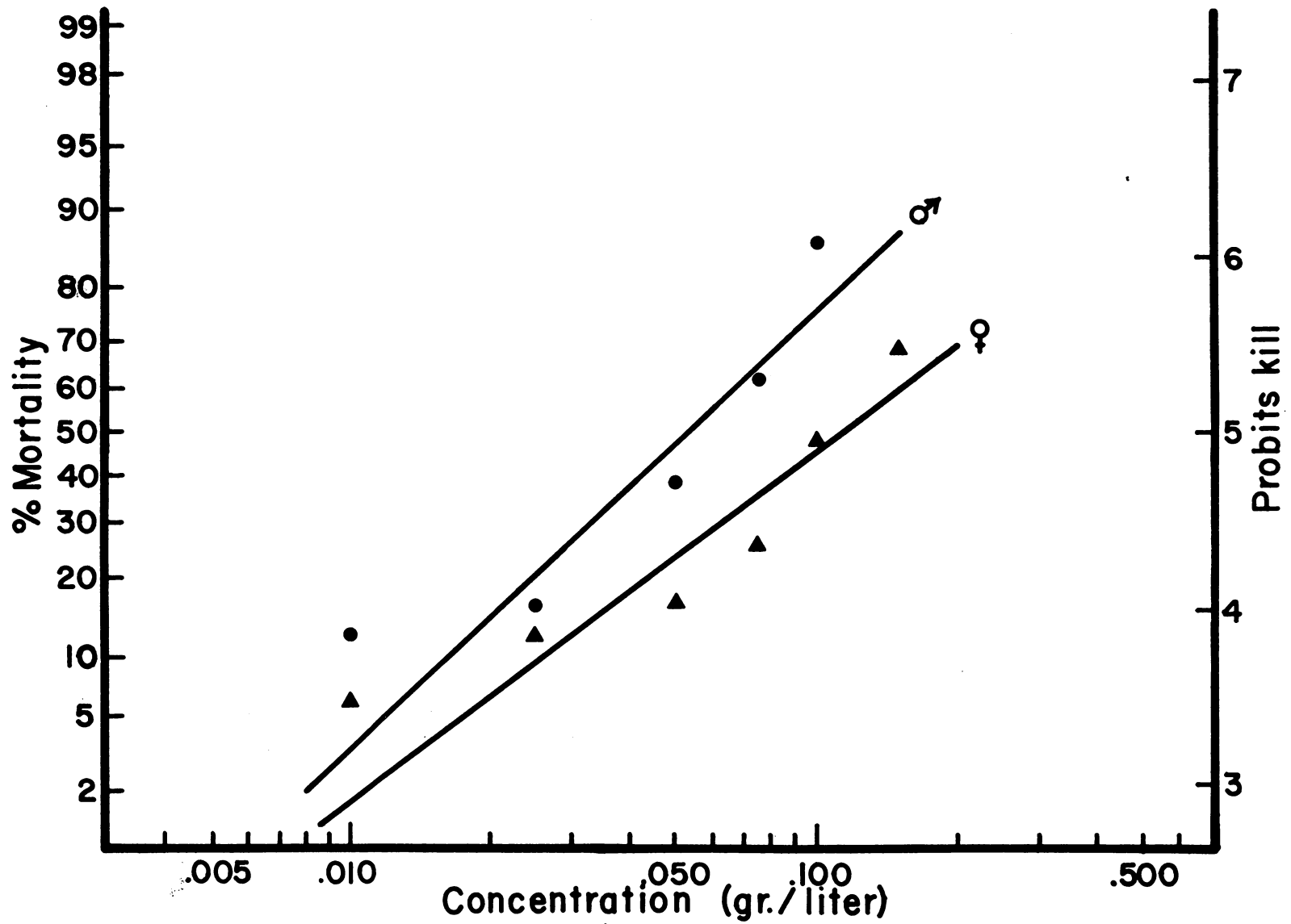


Figure 3 G

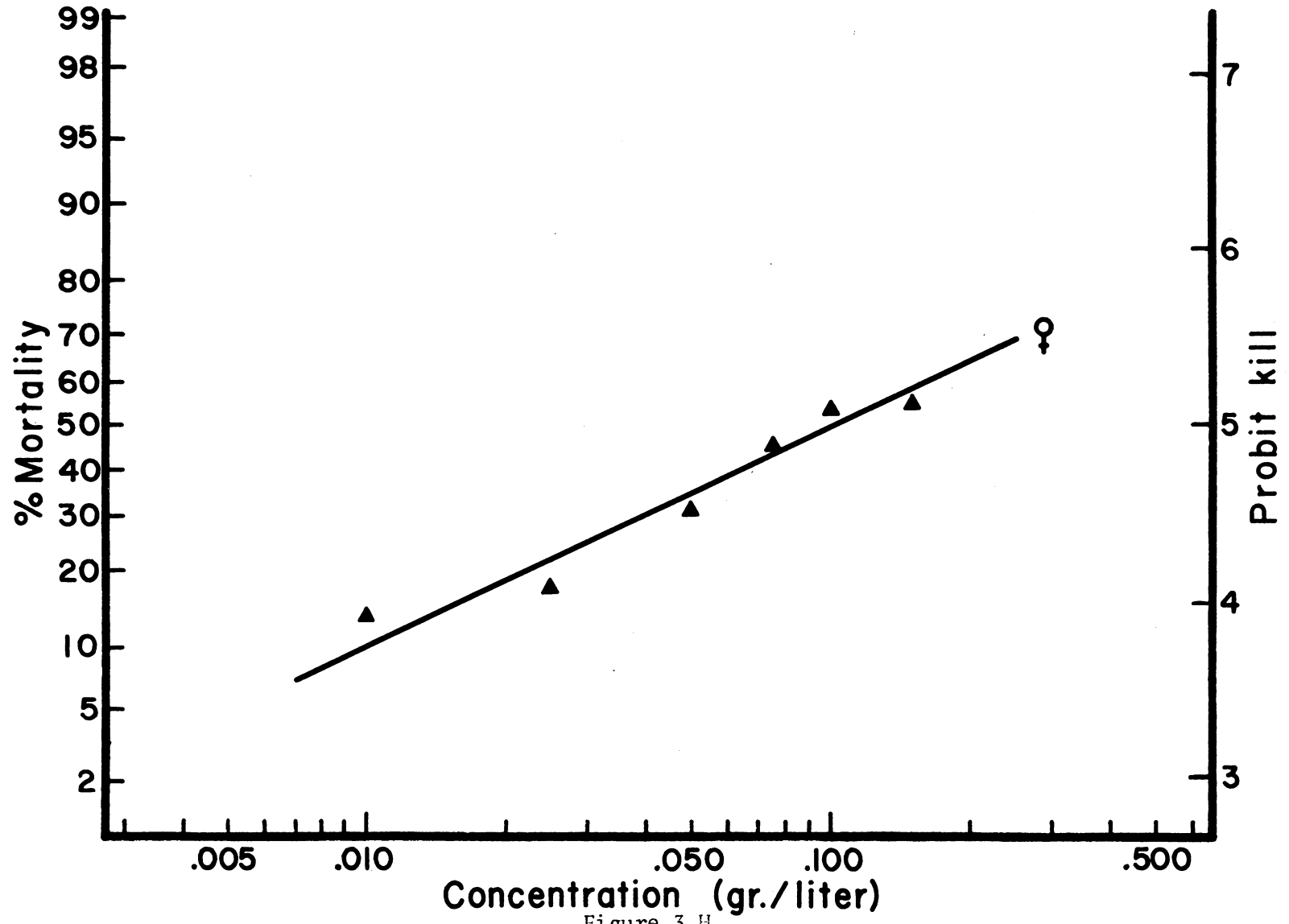


Figure 3 H

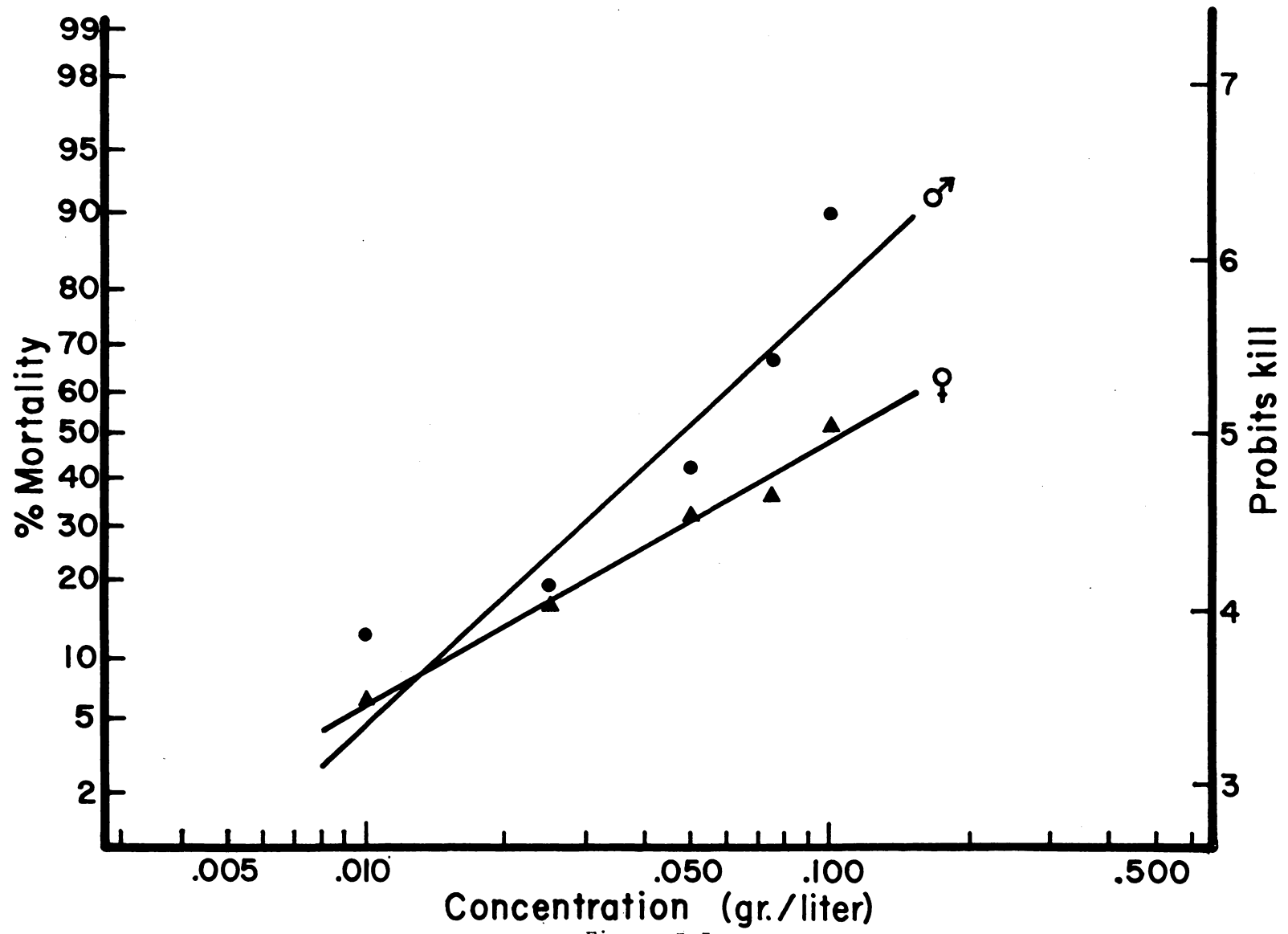


Figure 3 I

Table 4

The Response of Normal Strain German Cockroaches When Tested With DDT

Egg Case Number	Sex		Age of Adults-Days				
			1-3	5-7	10-12	15-18	25-28
1	male	LC ₅₀ (gr./l.)	0.042	0.046	0.050	0.057	0.011
		Slope	2.31	2.37	2.20	2.29	1.12
	female	LC ₅₀	0.086	0.140	0.090	0.110	0.054
		Slope	1.61	1.19	1.86	2.61	1.66
2	male	LC ₅₀			0.048	0.052	
		Slope			2.39	2.51	
	female	LC ₅₀			0.083	0.110	0.100
		Slope			1.87	1.99	1.23
3	male	LC ₅₀					0.047
		Slope					2.47
	female	LC ₅₀					0.100
		Slope					1.53

normal strain changes very little as age increases.

No significant age effect was found in males from the aldrin-resistant strain when they were tested with aldrin. However, there were three statistically significant age groupings when the females were tested (Table 1). The 5-7 and 10-12 day age groups were found to be the most resistant. The 1-3 and 15-18 day age groups were less resistant than the 5-7 and 10-12 day groups, but more resistant than the 25-28 day group. A study of Figure 4 and Table 5 indicates that there was not much difference in the slopes of the dosage-mortality lines except for those of the 10-12 day age group. These slopes were lower than the rest. LC_{50} values varied considerably, with a low of 3.9 gr./liter and a high of 35 gr./liter. The three highest LC_{50} values were all in the 10-12 day age group. Since these data were fairly consistent, this indicated that the aldrin resistant strain reached its peak of resistance at about 10-12 days of age and then decreased. The reason the males did not show this effect was probably due to the great variation within tests which resulted in a very large error term in the analysis of variance.

In tests on the DDT-resistant strain only the 15-18 and 25-28 day age groups were tested because of a lack of roaches of this strain. Neither of the age groups was found to differ significantly for either the males or the females (Figure 5).

It appears that age may influence the response of roaches to the insecticides tested to some extent. It is doubtful, however,

FIGURE 4

The Response of Aldrin-Resistant Strain German Cockroaches When Tested
With
Aldrin

- A. First egg case progeny, 1-3 days of age
- B. First egg case progeny, 5-7 days of age
- C. First egg case progeny, 10-12 days of age
- D. First egg case progeny, 15-18 days of age
- E. First egg case progeny, 25-28 days of age
- F. Second egg case progeny, 10-12 days of age
- G. Second egg case progeny, 15-18 days of age
- H. Second egg case progeny, 25-28 days of age
- I. Third egg case progeny, 10-12 days of age
- J. Third egg case progeny, 15-18 days of age
- K. Third egg case progeny, 25-28 days of age

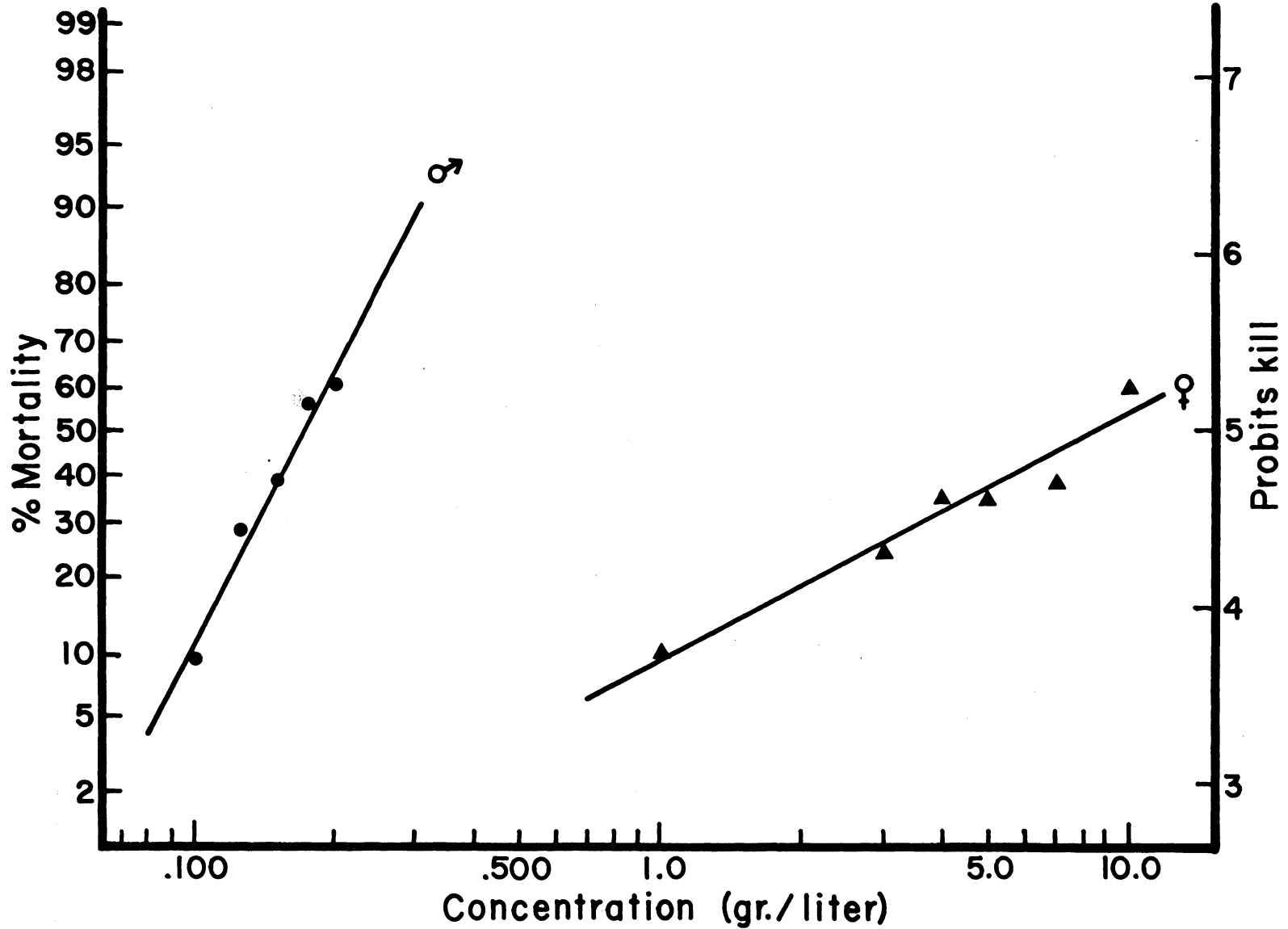


Figure 4 A

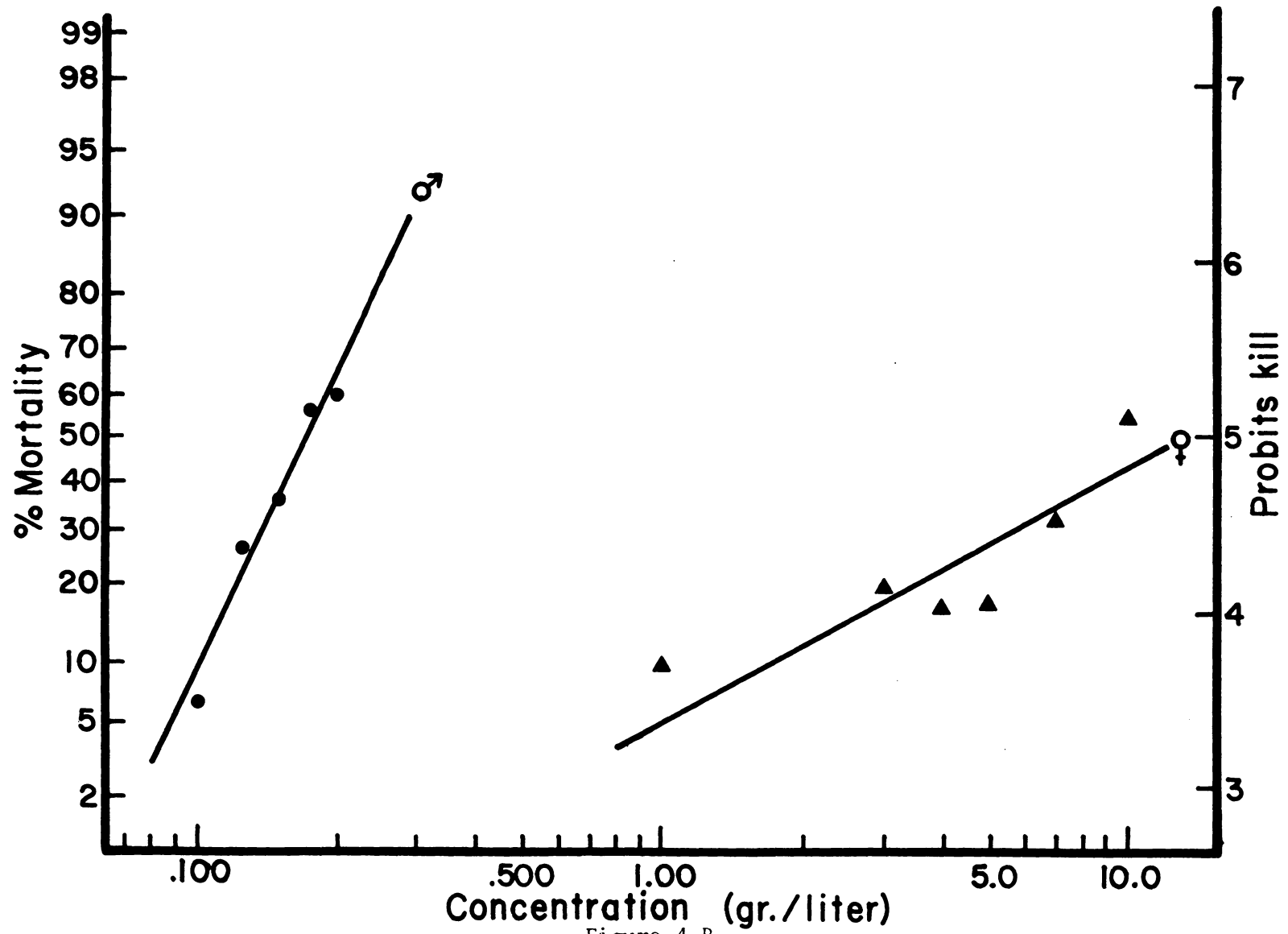


Figure 4 B

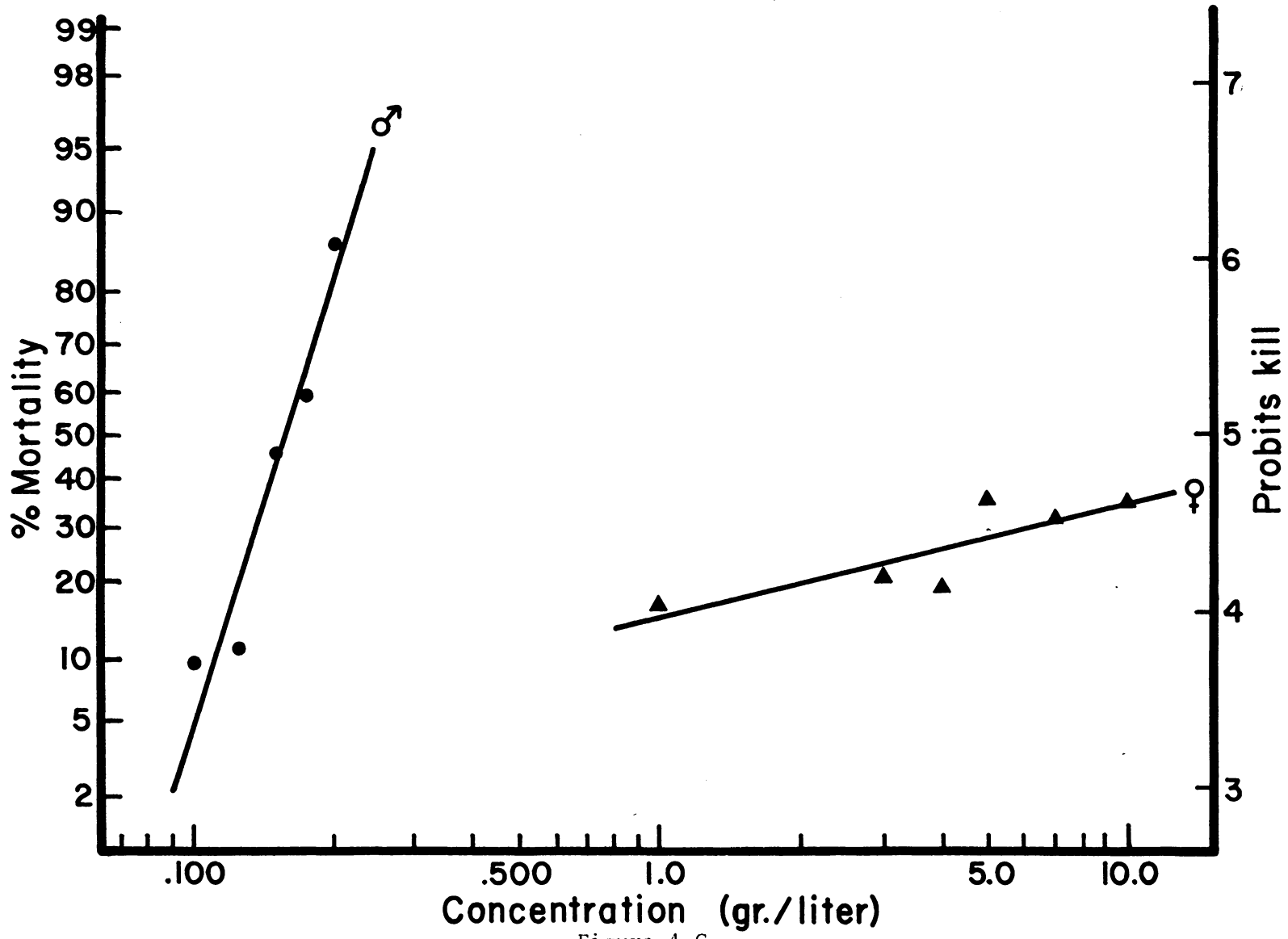


Figure 4 C

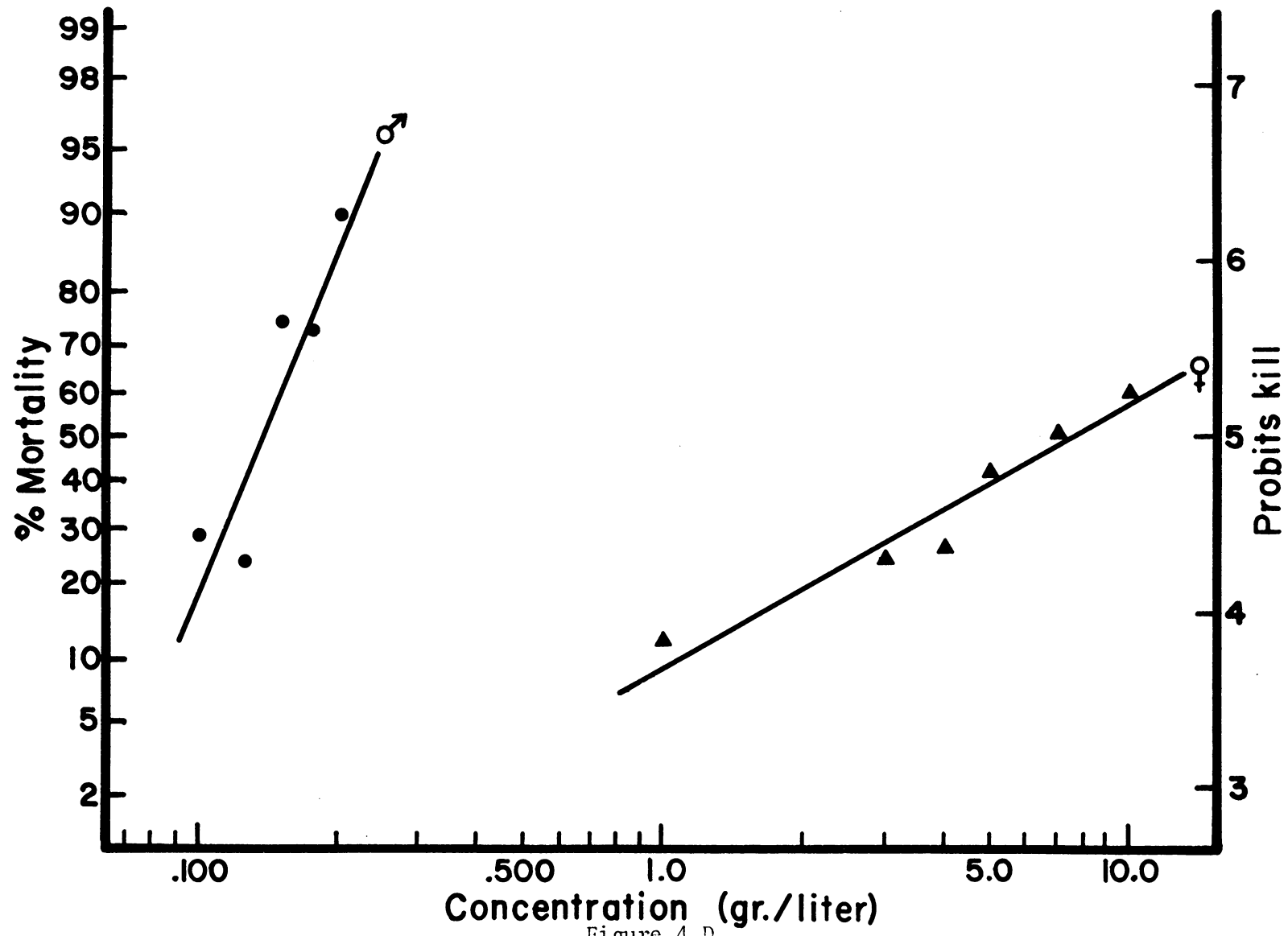


Figure 4 D

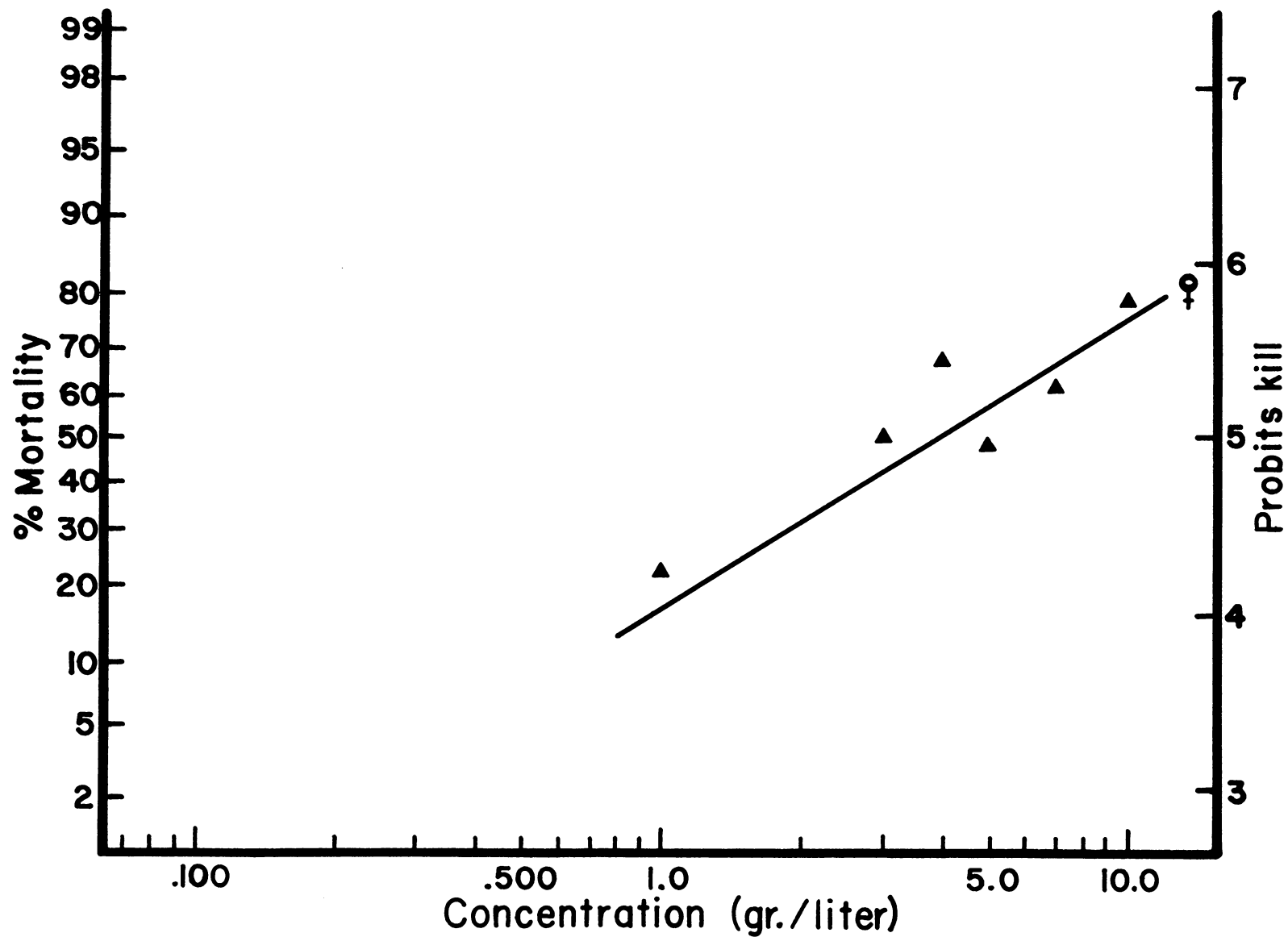


Figure 4 E

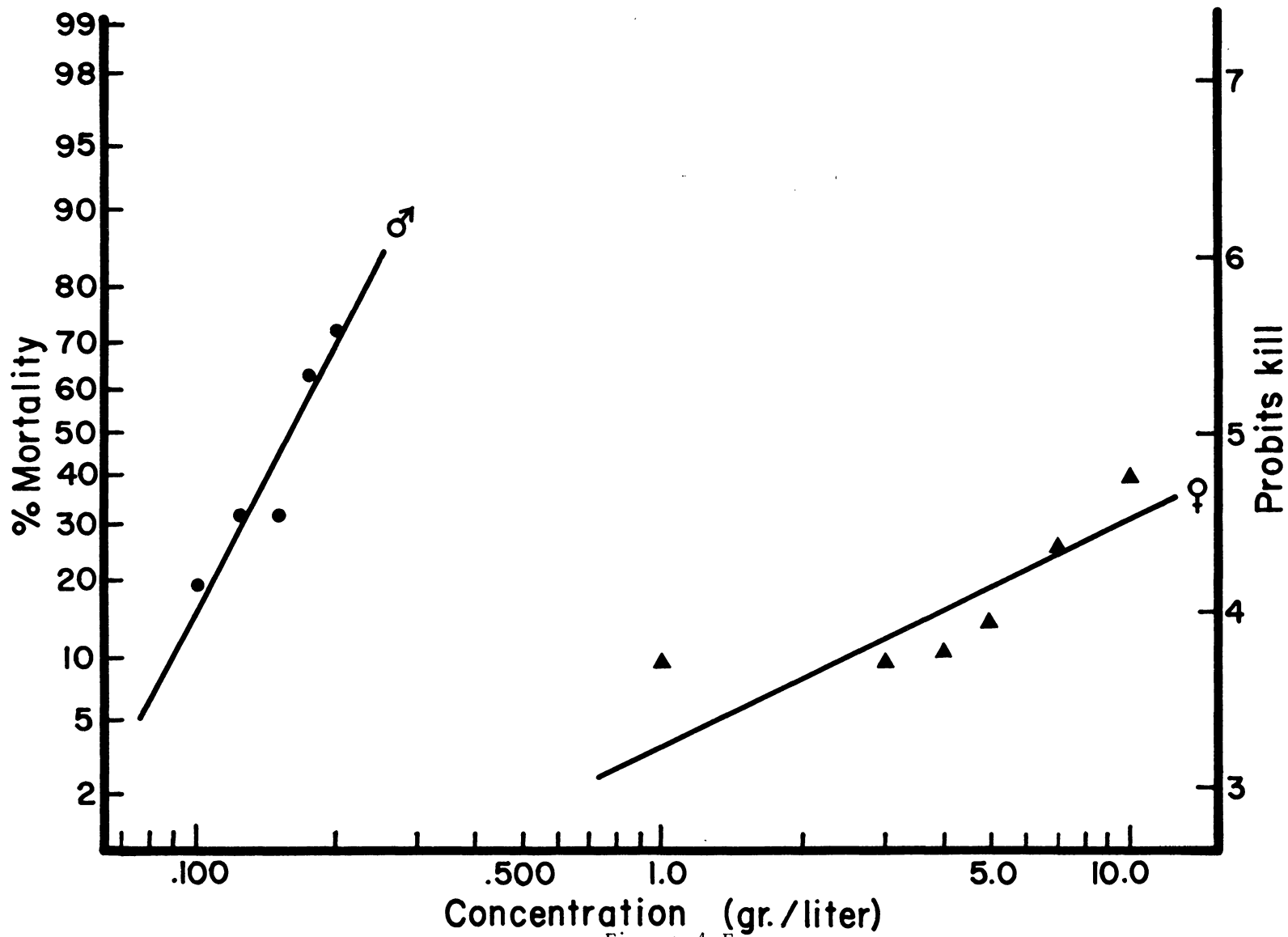


Figure 4 F

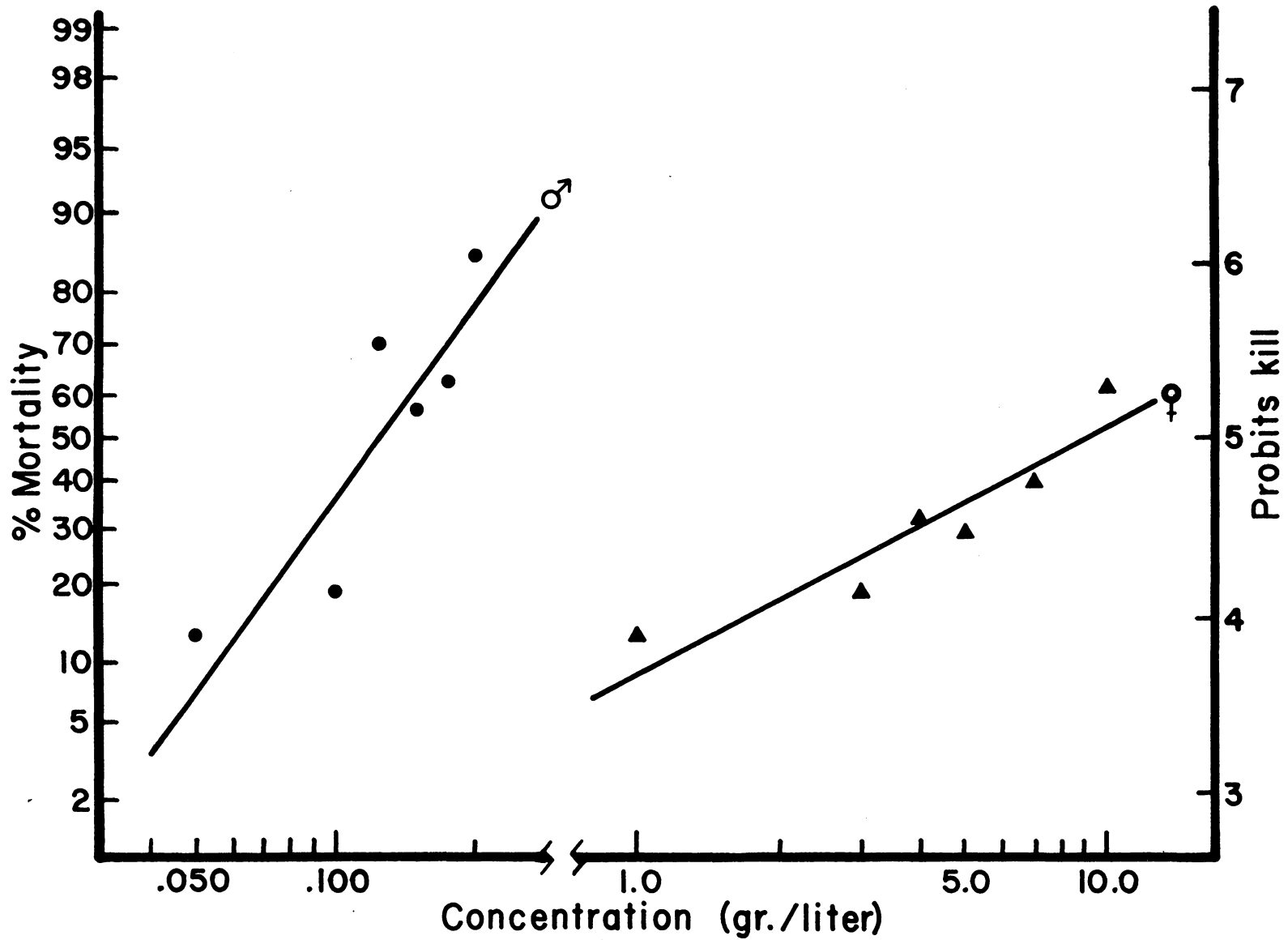


Figure 4 G

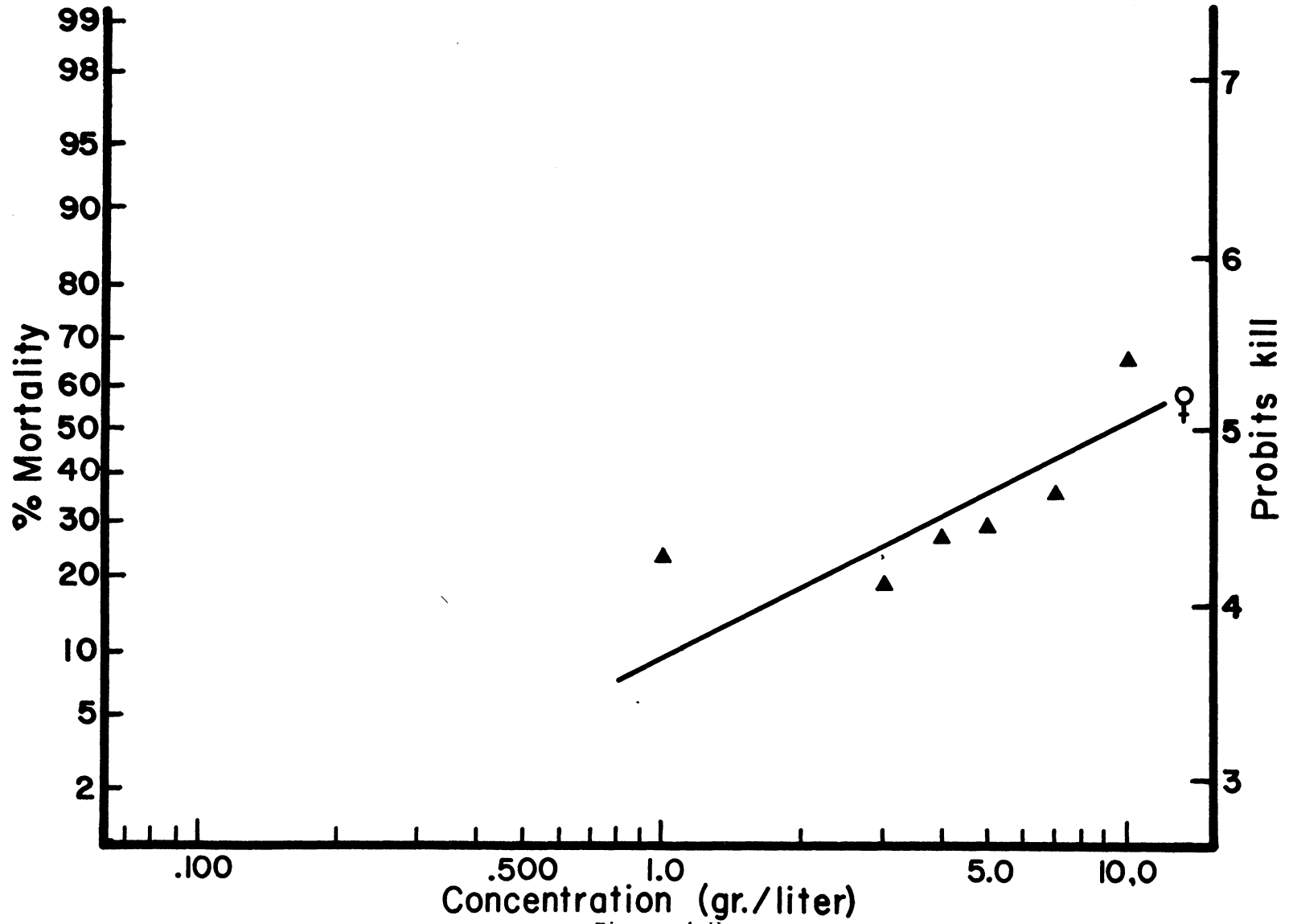


Figure 4 II

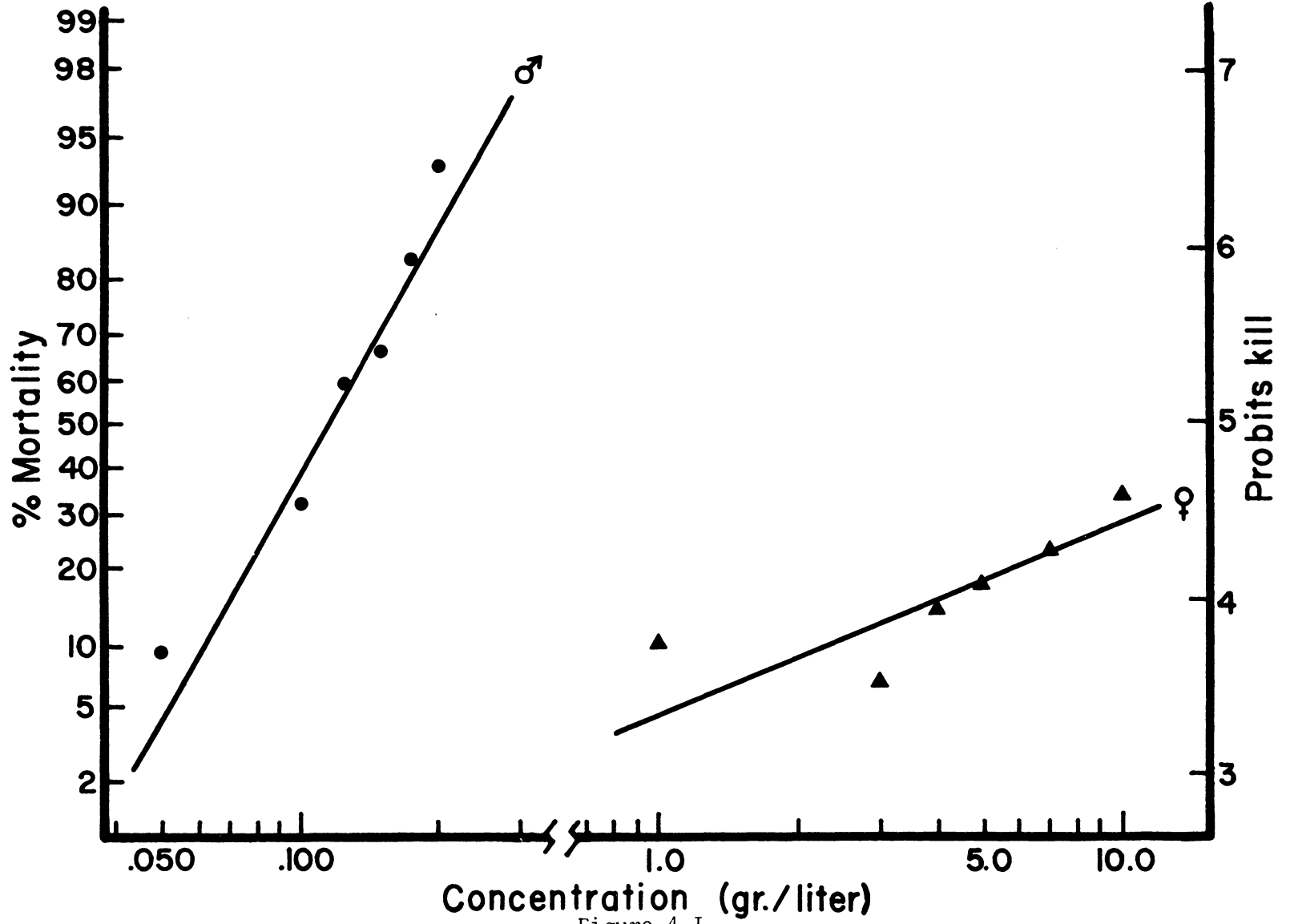


Figure 4 I

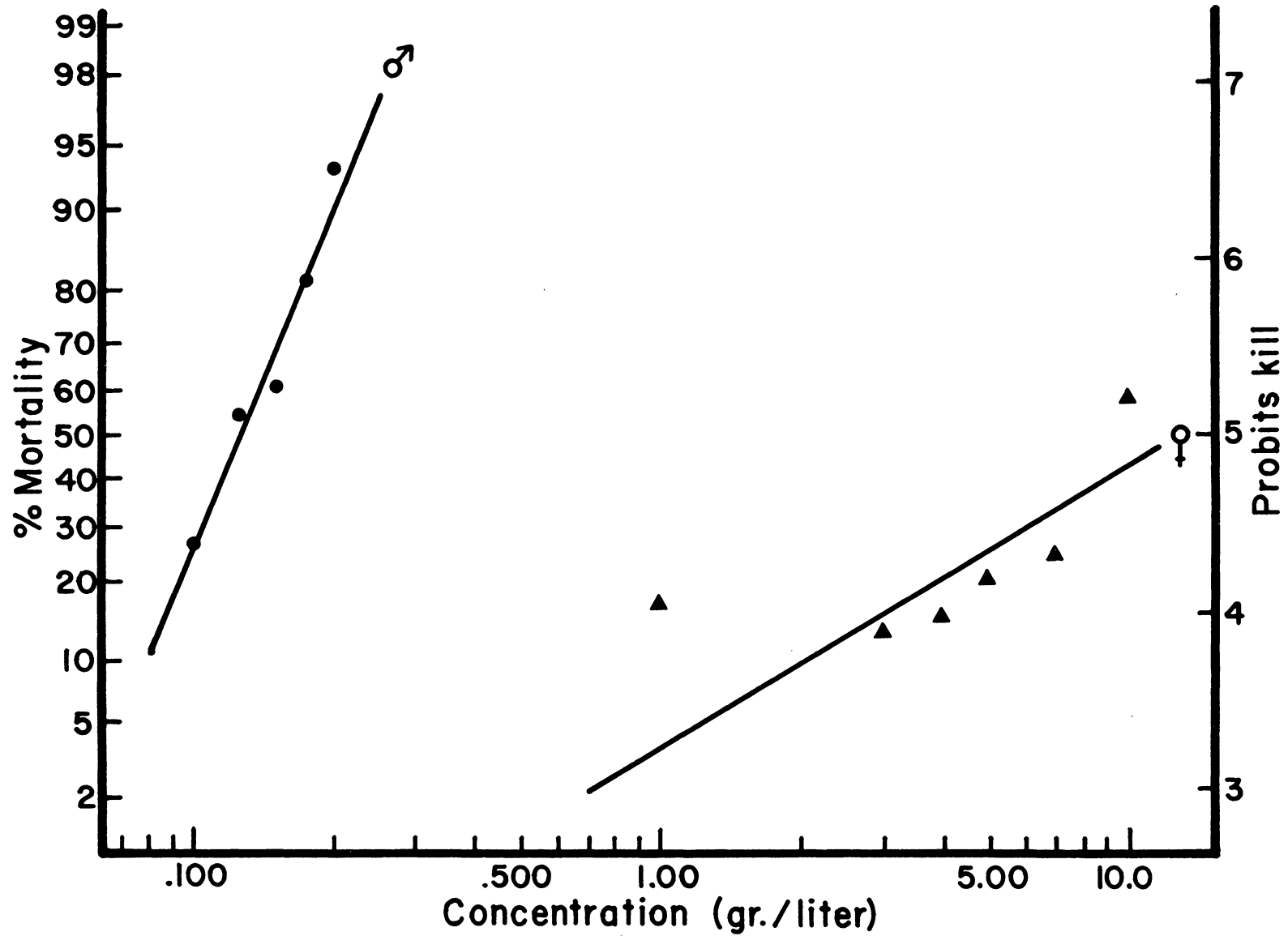


Figure 4 J

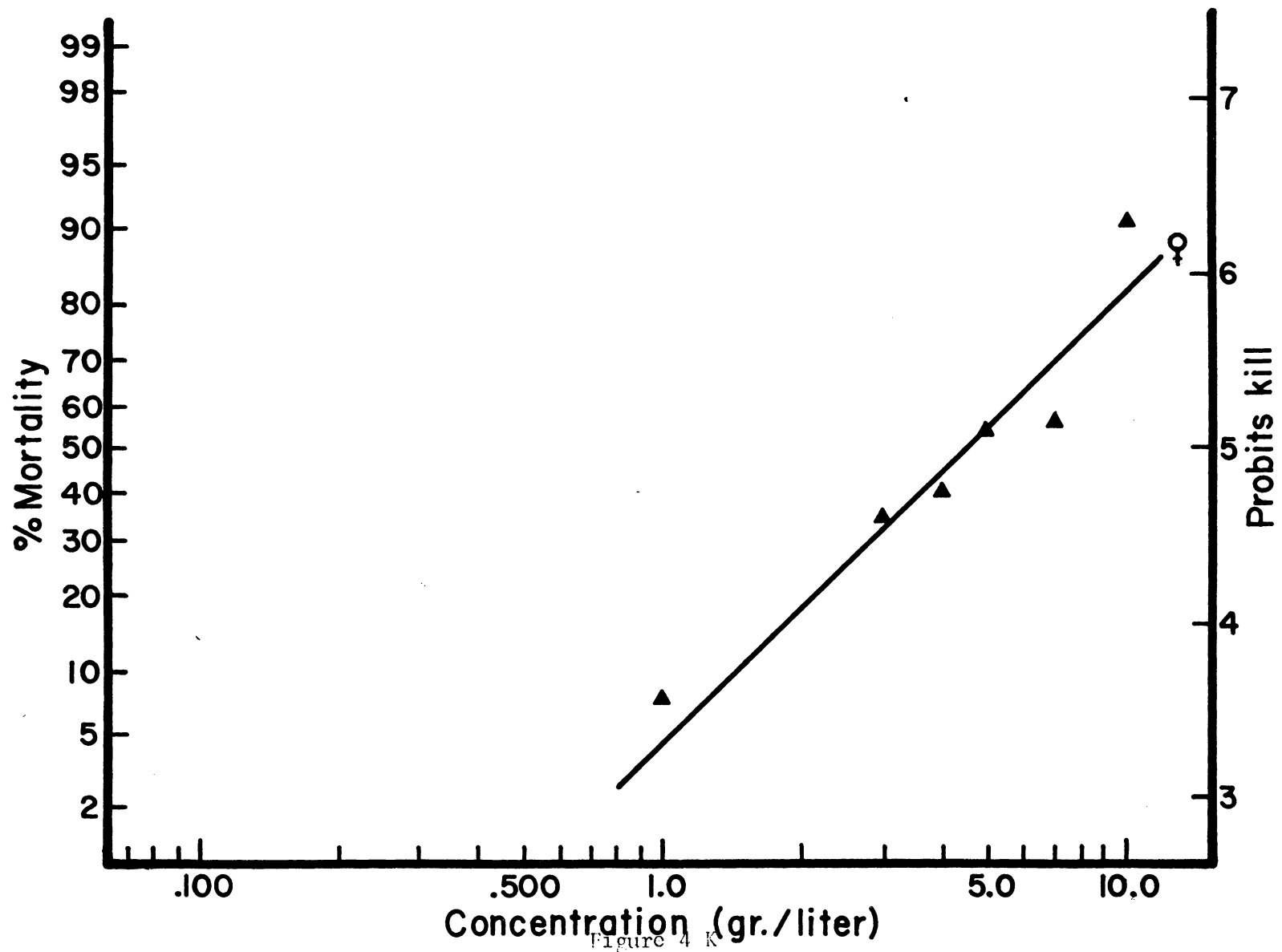


Figure 4 K

Table 5

The Response of Aldrin-Resistant Strain German Cockroaches When Tested With Aldrin

Egg Case Number	Sex		Age of Adults-Days				
			1-3	5-7	10-12	15-18	25-28
1	male	LC ₅₀ (gr./1.)	0.170	0.170	0.157	0.137	
		Slope	5.12	5.58	8.48	6.62	
	female	LC ₅₀	8.1	13.0	35.0	7.4	3.9
		Slope	1.43	1.44	0.66	1.54	1.35
2	male	LC ₅₀			0.157	0.122	
		Slope			4.93	3.64	
	female	LC ₅₀			23.0	8.6	9.0
		Slope			1.29	1.40	1.34
3	male	LC ₅₀			0.113	0.125	
		Slope			4.75	7.71	
	female	LC ₅₀			22.0	12.5	4.4
		Slope			1.09	1.59	2.60

FIGURE 5

The Response of DDT-Resistant Strain German Cockroaches When Tested
With
DDT

- A. First egg case progeny, 15-18 days of age
- B. First egg case progeny, 25-28 days of age
- C. Second egg case progeny, 15-18 days of age
- D. Second egg case progeny, 25-28 days of age
- E. Third egg case progeny, 15-18 days of age
- F. Third egg case progeny, 25-28 days of age

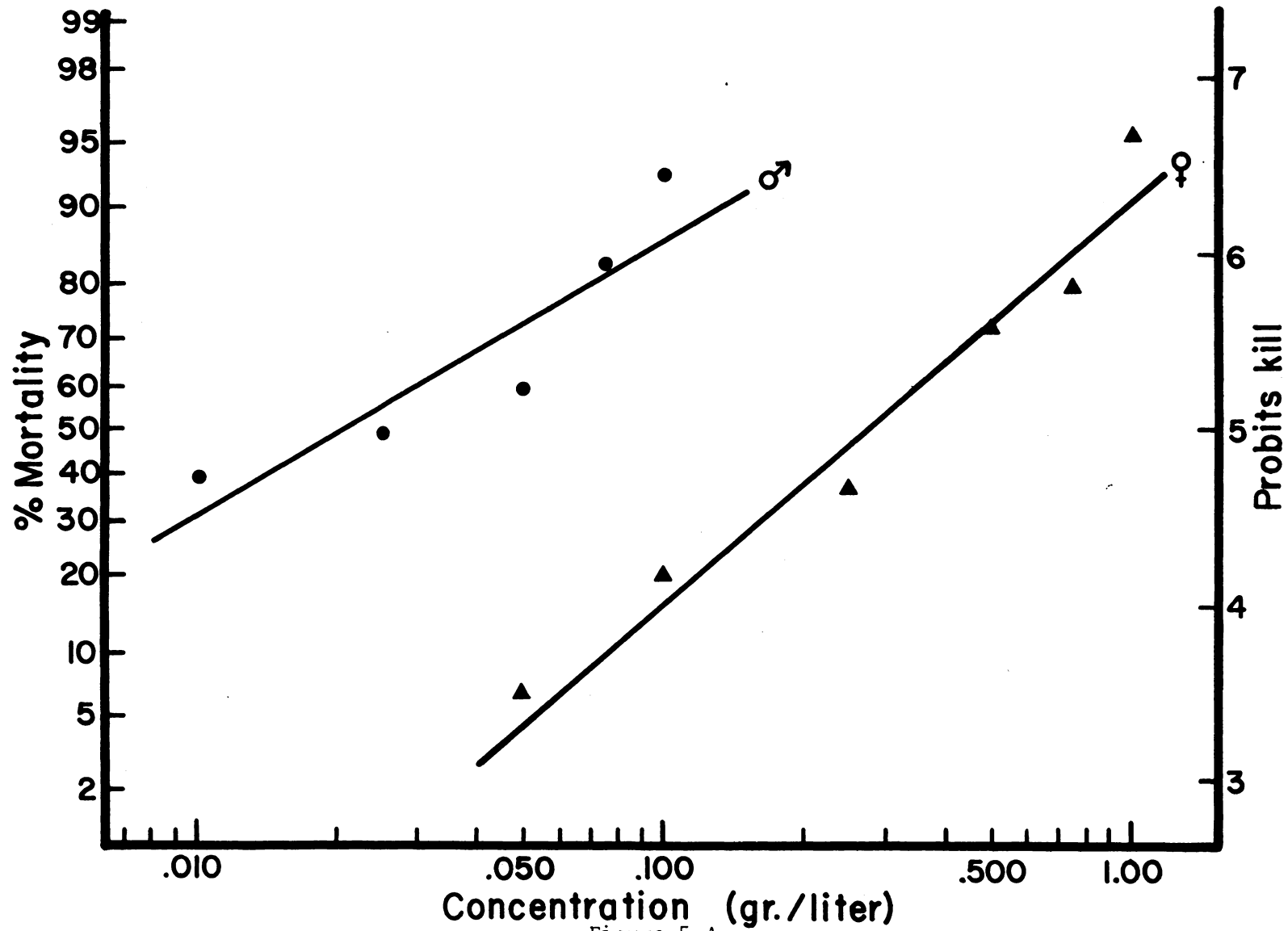


Figure 5 A

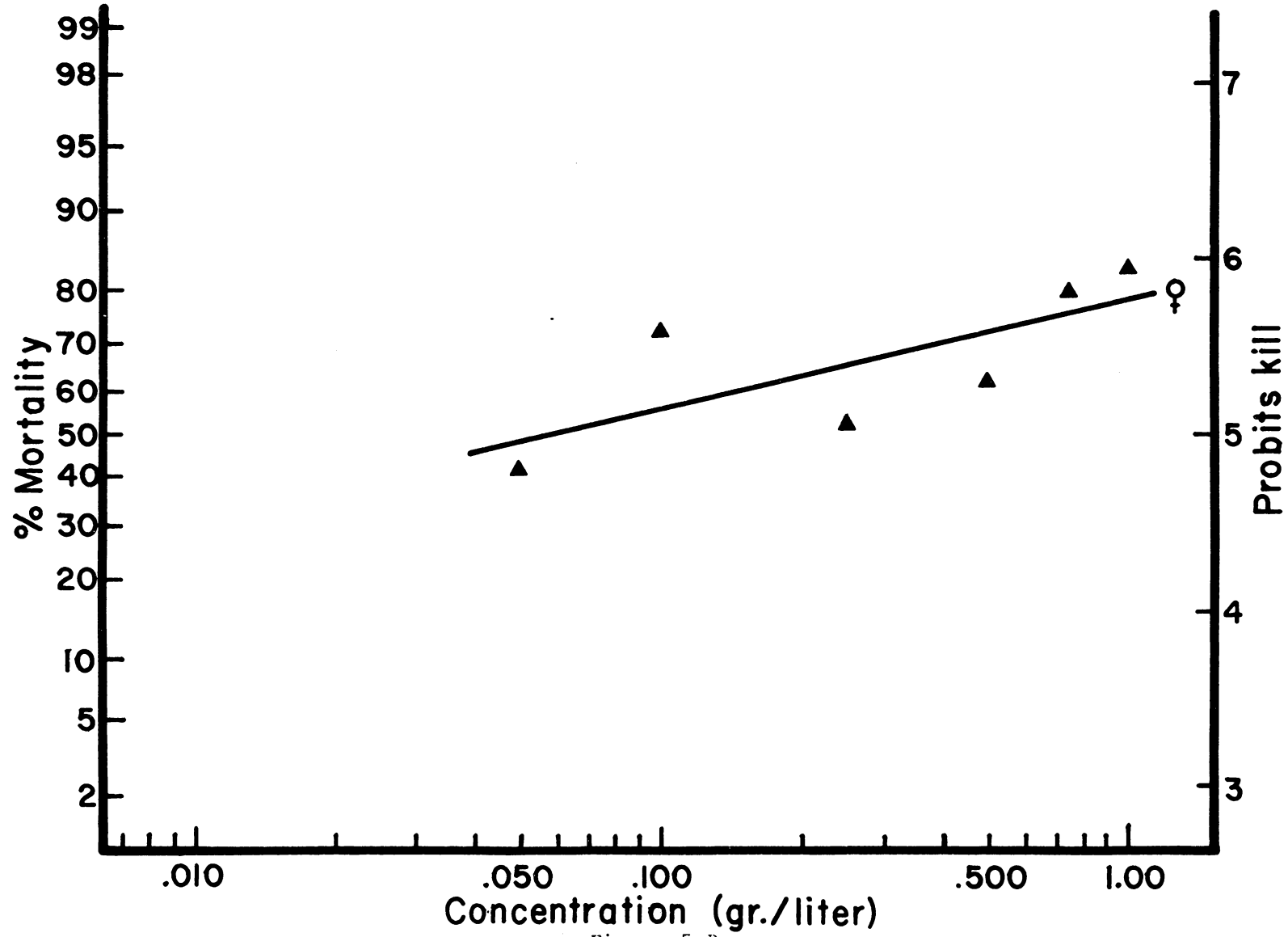


Figure 5 B

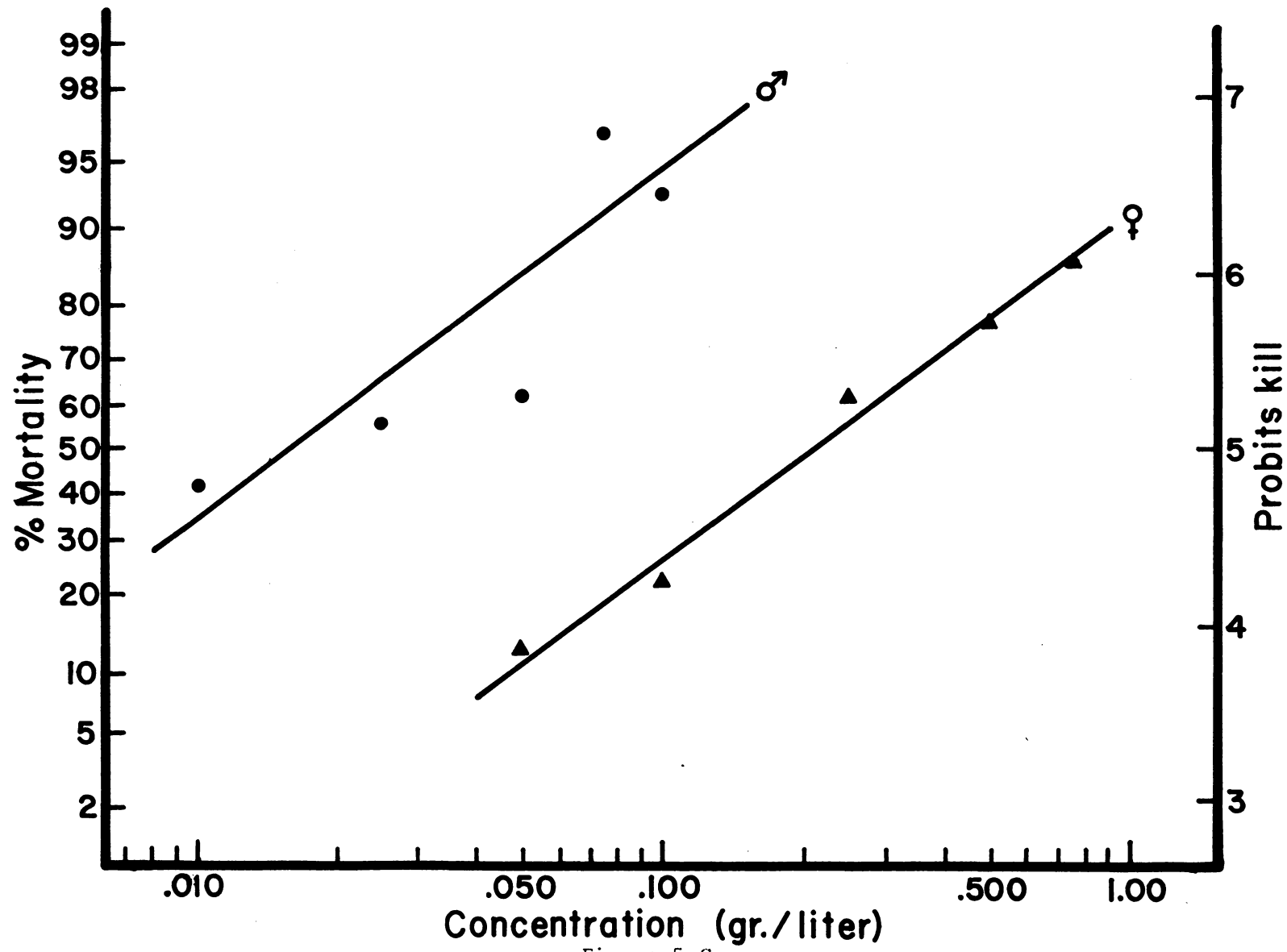


Figure 5 C

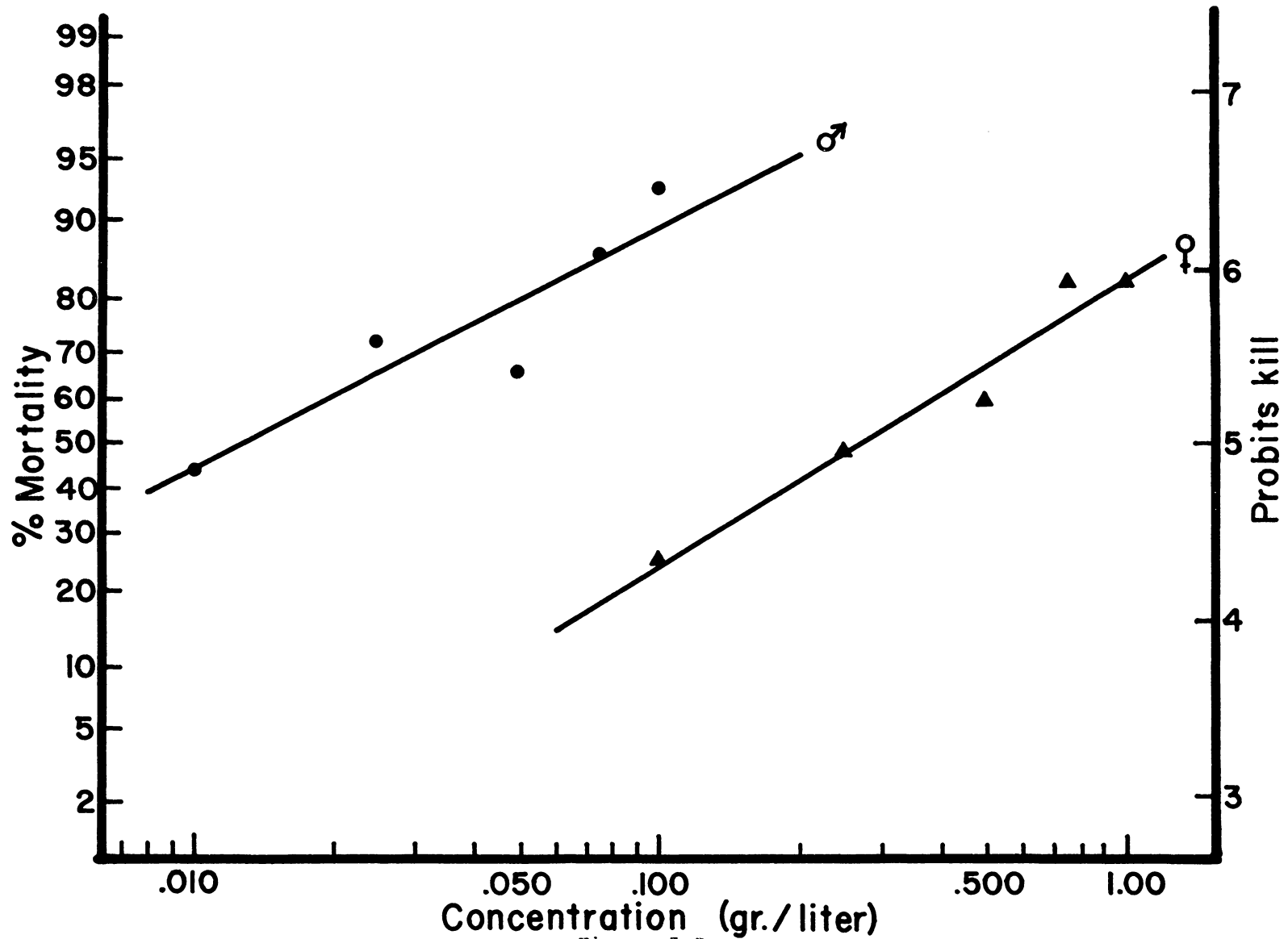


Figure 5 D

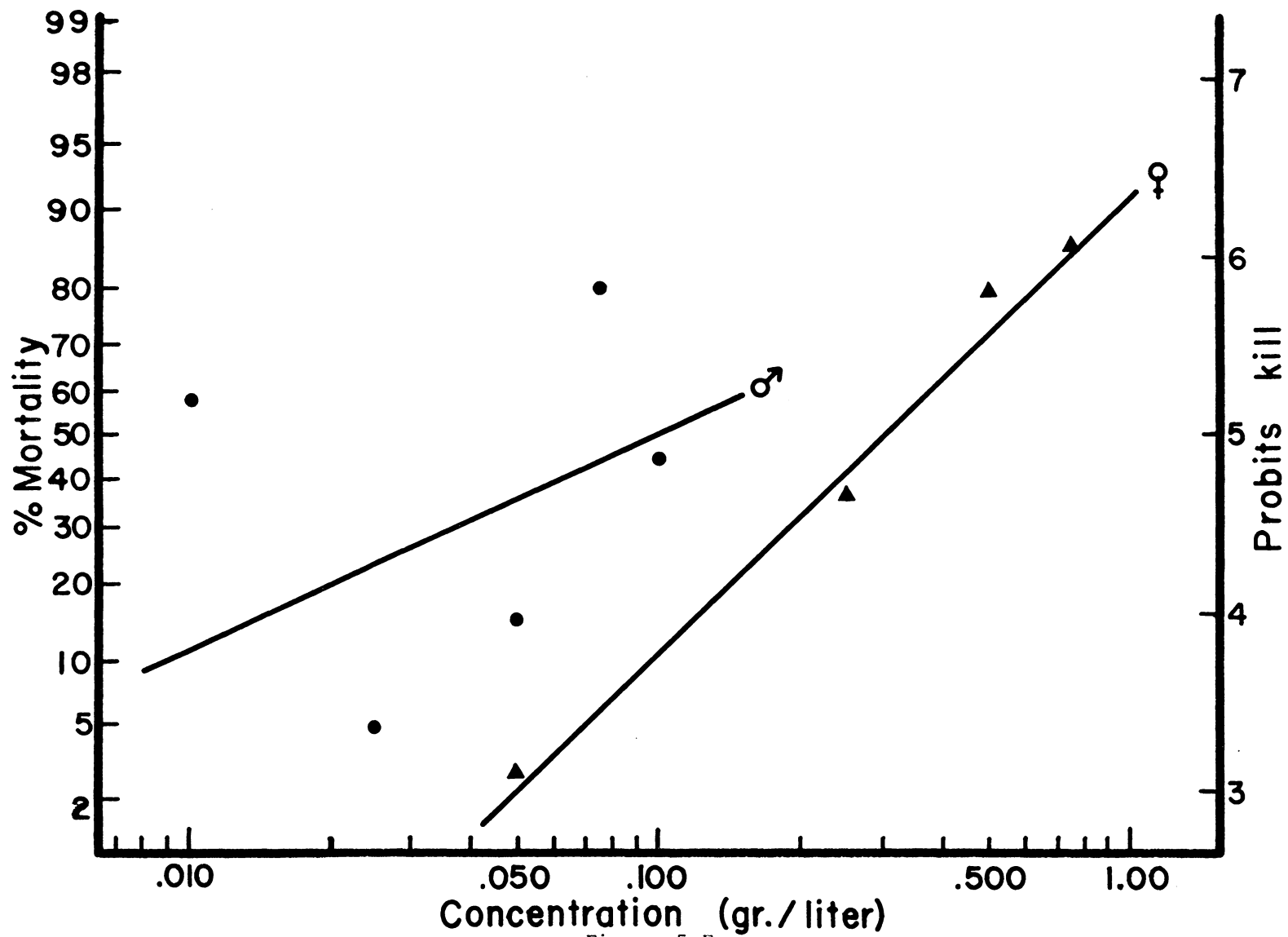


Figure 5 E

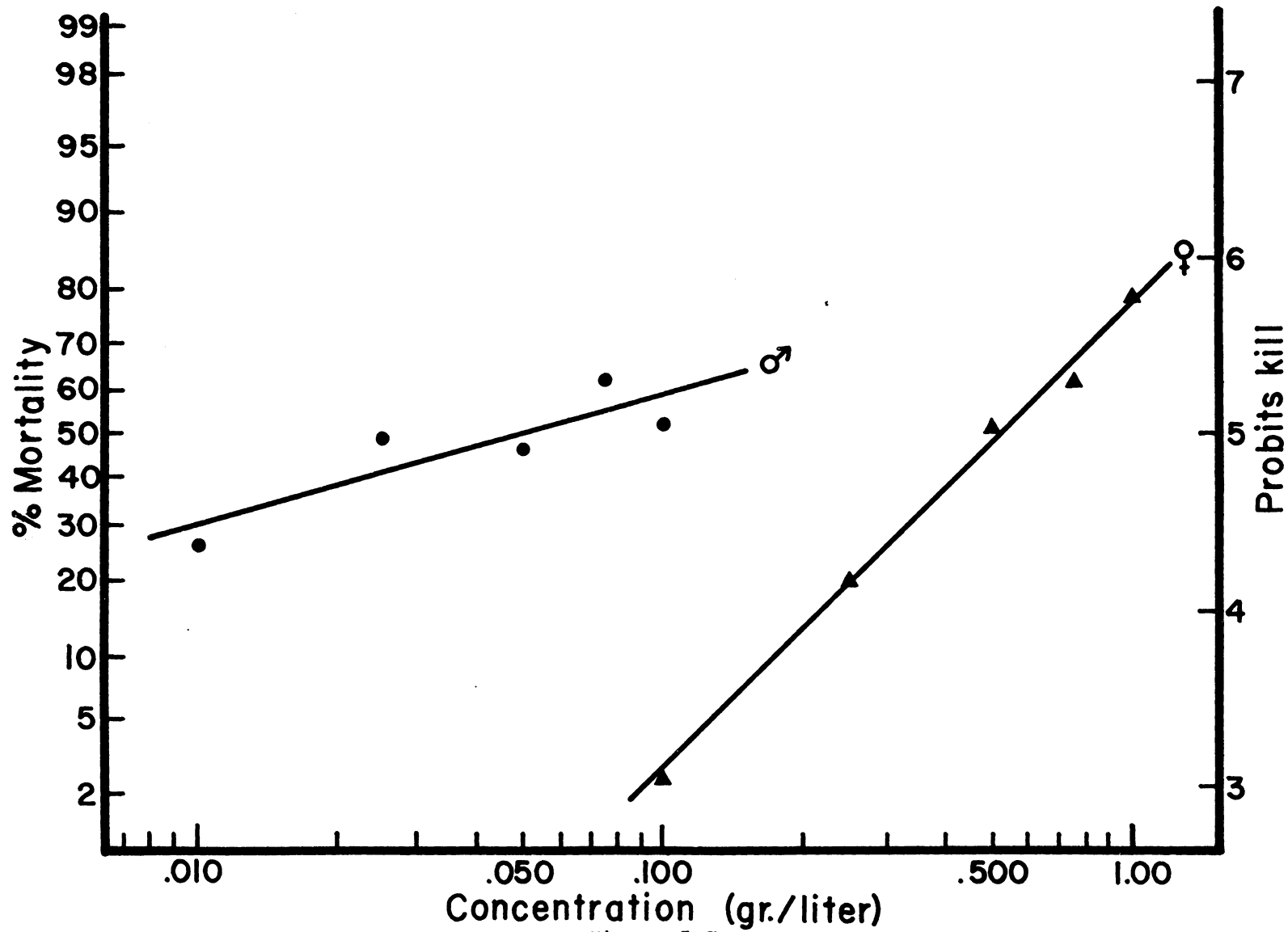


Figure 5 F

if all the instances where statistical significance was shown are of much biological significance because the magnitude of the changes were not often great. Unless the change in LC_{50} values is greater than two-fold and possibly more, the importance of such changes is doubtful (Hoskins and Gordon, 1956). In the malathion tests the differences were never more than two fold, thus it is probable that age had no effect in these tests. In the tests using aldrin on normal strain roaches, the LC_{50} values and the dosage-mortality line slopes gradually decreased as age increased. There was a nine-fold difference in the LC_{50} values in the males and a five-fold difference in the females. These large differences are probably significant, susceptibility having gradually increased as age increased. In the DDT tests on the normal strain the slopes and LC_{50} values remained about the same until the 25-28 day age group was reached. Since the differences were not very large and were not confirmed in the second and third egg cases it is doubtful if there was any significant difference indicated by the tests reported here. The differences found in the aldrin-resistant females were probably significant. Both the slopes and LC_{50} values exhibited greater than two-fold differences, especially in the 10-12 day age group. Thus resistance apparently reached a peak at about this age and then declined to or below the level of resistance at the youngest age group tested.

PARENTAL AGE

Statistically significant differences in the response of the progeny from the first three egg cases was found in about one-half of the tests. From the exposure of normal strain males to malathion, the progeny from the first egg case were found to be more susceptible than those from the second and third egg case. There were no differences due to egg case number in the females. The aldrin tests on normal strain roaches produced no difference due to egg case number in either sex. In the tests with DDT on normal roaches, egg case number was found to be significant in both sexes. When tested in the 25-28 day age group, the first egg case was more resistant than the third. When tested in the 15-18 day group, there were no differences in the progeny from the first and second egg cases. The tests with the females showed the progeny from the first egg case to be more susceptible than those from the second and third.

Significant differences due to egg case number were found in both sexes when the aldrin-resistant strain was tested with aldrin. In the males, the first and second egg case progeny were more resistant than the third. In the females, the second egg case progeny were more resistant than the first and third. The males of the DDT-resistant strain showed significance for egg case number, but the females did not. In the males, the third egg case progeny were more resistant than the first and second.

Although statistical significance at the 5% level was found for the parental age factor in about one-half of the tests, it was again probably of little biological significance. The lack of large variations in the LC_{50} values and the slopes of the dosage-mortality lines, coupled with the inconsistency of results between sexes and age groups, would seem to indicate a lack of biological importance for the parental age factor examined in these studies.

SUMMARY

The purpose of this study was to determine the effects of two age factors upon the response of adult German cockroaches from three different strains to various insecticides. The strains used were a normal strain, an aldrin-resistant strain, and a DDT-resistant strain. The first age factor studied was the age of the roach at the time of testing. Age groups of 1-3, 5-7, 10-12, 15-18, and 25-28 days of adult age were used for both sexes in testing this factor. The other age factor examined was the age of the female parent at the time reproduction occurred. Since the females of this species deposit eggs only periodically, progeny were separated on the basis of the egg case number from which they came. Only the progeny from the first three egg cases were examined.

Age was found to be a statistically significant factor when malathion, aldrin, and DDT were tested on both sexes of the normal strain. When aldrin was tested on the aldrin-resistant strain, significance was found only in the females. Only the 15-18 and 25-28 day age groups were tested in the DDT-resistant strain, and no difference was found between these ages. It is doubtful, however, if all the instances where statistical significance was shown are of much biological significance because the magnitude of the differences were not often great and the inconsistencies which were evident. Therefore, the differences found in the malathion tests on

the normal strain are probably of very little biological significance. On the other hand, the significance found in the aldrin tests on the normal strain is probably of biological importance. It appears that as the age of the roach increases, susceptibility to aldrin gradually increases. Because differences of fairly large magnitude were found in the tests with the aldrin-resistant strain, it is probable that these differences were of biological significance also. Resistance to aldrin in this strain appeared to increase from the 1-3 day age group until it reached a peak in the 10-12 day age group. Resistance then decreased to a level equal to or lower than that of the younger age groups.

Statistical significance was found for the parental age factor in about one-half of the tests. However, it was probably of little biological significance because the variations were of a small magnitude and inconsistencies between the males and females occurred often.

ACKNOWLEDGEMENTS

The author wishes to express his sincere appreciation to Dr. D.G. Cochran, his Major Professor, for encouragement, suggestions, and guidance during all phases of this project.

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LITERATURE CITED

- Anonymous. 1946. Testing of roach sprays. Soap and San. Chem. 22(7):145-7, 148E, 148G.
- Bliss, C. I. 1935. The calculation of the dosage-mortality curve. Ann. Appl. Biol. 22:135-67.
- Butts, W. L. and R. H. Davidson. 1955. The toxicity of five organic insecticides to resistant and non-resistant strains of Blattella germanica (L.). Jour. Econ. Ent. 48(5):572-4.
- Campbell, F. L. and W. N. Sullivan. 1938. Testing fly sprays: a metal turntable method for comparative tests of liquid spray contact insecticides. Soap. 14(6):119-25, 149.
- Campbell, F. L., C. S. Barnhart, and J. M. Hertzell. 1941. Tests on crawling insects. Part II. Soap and San. Chem. 17(8): 105, 107, 109, 111, 113, 115, 121.
- Clarke, T. H. and D. G. Cochran. 1959. Cross-resistance in insecticide-resistant strains of the German cockroach, Blattella germanica (L.). Bull. Wild Hlth Org. 20:823-33.
- Cochran, D. G. 1962. Personal communication.
- Cox, A. J. 1944a. Insecticide testing. Part I. Soap and San. Chem. 20(6):114-7, 149.
- Cox, A. J. 1944b. Insecticide testing. Part II. Soap and San. Chem. 20(7):123, 125, 129.

- Doner, M. H. and C. V. Anderson. 1947. Testing insecticidal residues. *Soap and San. Chem.* 23(10):124-5, 159.
- Duncan, D. B. 1955. Multiple range and multiple F tests. *Biometrics.* 11(1):1-42.
- Fisk, F. W. and J. A. Isert. 1953. Comparative toxicities of certain organic insecticides to resistant and non-resistant strains of the German cockroach, Blattella germanica (L.). *Jour. Econ. Ent.* 46(6):1059-62.
- Girault, A. A. 1911. Standards of the number of eggs laid by insects. Part IX. *Ent. News.* 22:14-5.
- Gough, H. C. 1939. Factors affecting the resistance of the flour beetle, Tribolium confusum Duv., to hydrogen cyanide. *Ann. Appl. Biol.* 26(3):533-71.
- Gould, G. E. 1948. The newer insecticides against roaches. *Soap and San. Chem.* 24(3):147, 149, 177, 179.
- Grayson, J. M. 1951. Response of the German cockroach to sublethal concentrations of DDT and benzene hexachloride. *Jour. Econ. Ent.* 44(3):315-7.
- Grayson, J. M. 1953. Effects on the German cockroach of 12 generations of selection for survival to treatments with DDT and benzene hexachloride. *Jour. Econ. Ent.* 46(1):124-7.
- Grayson, J. M. 1954. Differences between a resistant and a non-resistant strain of the German cockroach. *Jour. Econ. Ent.* 47(2):253-6.

- Grayson, J. M. 1961. Resistance to Diazinon in the German cockroach. Bull. Wld Hlth Org. 24:563-5.
- Grayson, J. M. and D. H. Messersmith. 1959. Latest on resistant roach control research at VPI. Pest Control. 27(2):26-7.
- Grayson, J. M. and B. D. Perkins. 1960. Results of 1959 cockroach control tests at VPI. Pest Control. 28(6):9, 11, 58.
- Grayson, J. M. and B. D. Perkins. 1961. Results of 1960 cockroach control tests at VPI. Pest Control. 29(1):30, 32, 34.
- Grayson, J. M. and H. G. Townsend. 1962. Results of recent cockroach control tests at VPI. Pest Control. 30:14, 16, 18.
- Hazard, F. O. 1945a. Liquid and powder methods for roach testing. Soap and San. Chem. 21(6):126-7.
- Hazard, F. O. 1945b. Modification of the liquid roach method. Soap and San. Chem. 21(12):159, 167.
- Heal, R. E., K. B. Nash, and M. Williams. 1953. An insecticide-resistant strain of the German cockroach from Corpus Christi, Texas. Jour. Econ. Ent. 46(2):385-6.
- Hoskins, W. M. and H. T. Gordon. 1956. Arthropod resistance to chemicals. Ann. Review Ent. 1:89-122.
- Ludwig, D. 1956. Effects of temperature and parental age on the life cycle of the mealworm, Tenebrio molitor Linnaeus (Coleoptera, Tenebrionidae). Ann. Ent. Soc. Amer. 49:12-5.

- Ludwig, D. and C. Fiore. 1960. Further studies on the relationship between parental age and the life cycle of the mealworm, Tenebrio molitor. Ann. Ent. Soc. Amer. 53:595-600.
- McGovran, E. R. and J. H. fales. 1942. Roach testing. Soap and San. Chem. 18(3):101, 103, 105, 107, 117.
- O'Brian, D. M. 1961. Effects of parental age on the life cycle of Drosophila melanogaster. Ann. Ent. Soc. Amer. 54:412-6.
- Pan, Y. S. and S. L. Yang. 1962. The relationship between the age of Drosophila adults and their susceptibility to endrin residues. Rept. Taiwan Sugar Expt. Stat. 27:85-90.
(In Chinese) (Original not seen. Abstract in Bio. Abs. 40(1):299).
- Perkins, B. D. Jr. and J. M. Grayson. 1961. Some biological comparisons of resistant and non-resistant strains of the German cockroach, Blattella germanica. Jour. Econ. Ent. 54(4):747-50.
- Richard, A. G. and M. Q. Kolderic. 1957. Variations in weight, development rate, and hatchability of Oncopeltus eggs as a function of the mother's age. Ent. News. 68:57-64.
- Saunders, D. S. 1962. The effect of the age of female Nasonia vitripennis (Walker) (Hymenoptera, Pteromalidae) upon the incidence of larval diapause. Jour. Insect Physiol. 8:309-18.

- Simanton, W. A. and A. C. Miller. 1937. Housefly age as a factor in susceptibility to pyrethrum sprays. *Jour. Econ. Ent.* 30(6):917-21.
- Smith, E. H. and B. J. Fiori. 1959. The use of the plum curculio in toxicological studies. *Jour. Econ. Ent.* 52(5):921-8.
- Tracy, K. M. 1958. Effects of parental age on the life cycle of the mealworm Tenebrio molitor Linnaeus. *Ann. Ent. Soc. Amer.* 51:429-32.
- Tuma, V. 1938. Roaches-a study of the relationship between the ages of cockroaches and their resistance to liquid insecticides. *Soap.* 14(6):109-111, 113, 115, 117, 151.

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THE EFFECTS OF CERTAIN AGE FACTORS ON THE RESPONSE
OF THE GERMAN COCKROACH TO INSECTICIDES

by

Howard Garfield Townsend, Jr.

ABSTRACT

Two age factors were studied: the age of the roach at the time of testing, and the age of the female parent at the time reproduction occurred. Age groups from 1-3 days to 25-28 days were used to test the first factor and the first three egg cases were used to test the latter.

Age was found to be a statistically significant factor when malathion, aldrin, and DDT were tested on both sexes of the normal strain. However, because of inconsistencies and a lack of differences of any great magnitude, it is believed that the differences in all but the aldrin tests were of no biological significance. In the aldrin tests it appeared that as the age of the roach increased, susceptibility gradually increased. The tests using aldrin on the Aldrin-resistant strain were significant for the females. Resistance in this strain appeared to increase from the 1-3 day age group until it reached a peak in the 10-12 day age group, then decreased to a level equal to or lower than that of the younger age groups. Statistical significance was found for the parental age factor in about one-half of the tests. However, it was of little biological significance because the variations were of a small magnitude and inconsistencies between the males and females occurred often.