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Send Correspondence to:

Melody A. Keena

Department of Entomology

University of California

Davis, CA 95616

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Cyhexatin and Propargite Resistance in Populations of Spider Mites (Acari: Tetranychidae) from California Almonds

MELODY A. KEENA and JEFFREY GRANETT

Department of Entomology,

University of California,

Davis, California 95616

ABSTRACT Cyhexatin (Plictran^{R} 600 Flowable) or propargite (Omite^{R} 30 Wettable Powder) resistances or both were detected using a residual-bioassay procedure in spider mite (Tetranychus spp.) colonies collected from California almonds. Cyhexatin-susceptible T. urticae and T. pacificus responded with statistically identical log concentration/probit mortality lines. Statistically significant cyhexatin resistance was only detected in T. pacificus collected. Propargite-susceptible T. urticae were 8-fold more susceptible to propargite than the most susceptible T. pacificus colony collected. Propargite-resistant individuals of both T. urticae and T. pacificus were found. Colonies collected in the field were heterogeneous in their response to cyhexatin and propargite and contained varying frequencies of susceptible types. These results have implications for control of spider mites with these acaricides under commercial conditions in California almonds.

Keywords: Spider mites, Cyhexatin, Propargite, Resistance, Almonds, Tetranychidae

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THREE SPECIES of spider mites--Tetranychus urticae Koch, T. pacificus McGregor, and T. turkestani Ugarov and Nikolski--are annual pests of almonds. In California, cyhexatin (Plictran^{R}) and propargite (Omite^{R}) are commonly used selective acaricides currently registered for control of spider mites infesting California almonds.

Cyhexatin has been used world-wide since the early 1970's. A 2-fold cyhexatin resistance was first reported in T. cinnabarinus Boisduval from greenhouse roses in Israel in 1973 (Mansour & Plaut 1979). Since 1973 higher levels (7.8 to 107.8-fold) of cyhexatin resistance have been observed in T. urticae from apple and pear orchards in Australia (Edge & James 1982), pears in Oregon (Croft et al. 1984, Hoyt et al. 1985), and strawberries in California (Miller et al. 1985). Cyhexatin has been registered for use on California almonds since 1973. Resistance problems with control of spider mites by this acaricide in California almonds have not been reported until recently.

Propargite has been used since the late 1960's. A colony of T. cinnabarinus from greenhouse roses in Israel was found to be moderately resistant to propargite (ca. 5-fold) in 1973 (Mansour & Plaut 1979).

Tetranychus urticae collected from blackcurrants in New Zealand showed similar levels of resistance in 1982 (Chapman & Penman 1984). Propargite has been registered for use on California almonds since 1970. In 1983, spider mites collected from almonds exhibited propargite resistance in a laboratory bioassay (Keena and Granett 1985).

In this paper, we describe intraspecific and interspecific differences in the toxicity of cyhexatin and propargite of T. urticae and T. pacificus populations collected from California almond orchards. Implications of these results for control of spider mites with these acaricides under commercial conditions in California almonds are discussed.

Materials and Methods

In April and August of 1985, spider mites were collected from 20 almond orchards located throughout the almond growing region of California. The same orchards were sampled on both occasions. Seven single species colonies were chosen to represent the extreme and intermediate susceptibilities found in populations from the field. Colonies were chosen for each species/acaricide set based on previous work (Keena &

Granett 1985). The following are the nearest city and county for each of the colonies: T. urticae were collected from Livingston, Merced County (A), Clovis, Fresno County (B), and Pixley, Tulare County (C); T. pacificus were collected from Clovis, Fresno County (D), Arvin, Kern County (E), Wasco, Kern County (F), and McFarland, Kern County (G). Colonies B and C were collected in August; the other colonies were collected in April. Colonies B, C, E, and F were used as representatives for only one acaricide. Therefore, data for these colonies tested with the other acaricide are not presented here.

Spider mites were sampled and colonies established using procedures described by Keena & Granett (1985). The spider mites collected from each site were visually separated by species and initially reared in isolation using the floating-island method (Dennehy & Granett 1982). Once spider mite populations had reached maximum capacity on the islands, the colonies were transferred into styrofoam box enclosures.

Each styrofoam box (3-cm-thick, 40 cm by 33.5 cm by 26.5 cm high on the inside) had a hole cut in the center of each side (15 cm by 30 cm) and each end (15 cm by 21 cm) for ventilation. The holes were covered

with fine mesh polyester cloth (32 threads per cm). The lid consisted of a wooden frame (46 cm x 55 cm) with polyester cloth stretched over it. Three circular, clear acetate (0.5 mm thick, 12 cm in diameter) windows were sealed into the cloth of the lid to provide an observation window and illumination. Anhydrous lanolin was applied between the rim of the box and the lid to create a mite-proof seal.

The spider mite colonies were reared on 5- to 15-day-old 'Acala SJ-2' cotton seedlings in the box enclosures. Colonies were kept in a greenhouse at 29 $\pm$ 5 C, 40 $\pm$ 10% RH and constant light. Colonies were transferred from the box enclosures into larger cages measuring 60 cm per side (Dennehy & Granett 1984).

Concentration-mortality lines were estimated using the residual cell method described by Keena & Granett (1985). Cell bioassays using 20-30 adult female spider mites were replicated six times for each concentration. Water controls were included with each bioassay replication.

The concentrations of cyhexatin (Plictran 600 Flowable, Dow Chemical USA, Midland, Mich.) mixed in distilled water were 1.78, 3.16, 5.62, 10.0, 17.8,

31.6, 56.2, 100, 178, 316, and 562 ppm AI. The concentrations of propargite (Omite-30 Wettable powder, Uniroyal Chemical, Fresno, Calif.) mixed in distilled water were 10, 17.8, 31.6, 56.2, 100, 178, 316, 562, 1,000, 1,780, 3,162, 5,620, and 10,000 ppm AI.

Log concentration/probit mortality (lc/pm) lines were estimated by the POLO (Russell et al. 1977) probit program for both cyhexatin and propargite. The chi-squared (X^2) goodness of fit test for all but three of the lc/pm lines thus estimated resulted in large X^2 values with significance of < 0.005 indicating a poor fit of the data to the linear regression model. Three of the lc/pm lines gave a better fit of the data colonies D with cyhexatin (0.01) and propargite (0.1), and G with porpargite (0.025) with significances as indicated. Because of the poor fit of the data to a line, a plot of the means, standard deviations, and 95% confidence intervals for each concentration and colony were determined. First, the results were corrected for control mortality using Abbott's formula (Abbott 1925). The mortality values were then transformed to probits. For the purposes of averages of probit data, mortalities of 98% and above were considered probit

7.05 and mortalities of 2% and less were considered probit 2.95.

Results and Discussion

Cyhexatin. Tetranychus urticae colony A (Table 1) responded with the most cyhexatin susceptibility and had the steepest lc/pm line slope of all T. urticae colonies we collected from the field in 1985. No significant cyhexatin-resistance was detected in T. urticae were found in any of the 20 orchards sampled during the 1985 season. For this purpose resistance was defined as a significant shift in the lc/pm line as compared to the lc/pm line of the most susceptible colony collected. These results indicate that naturally occurring resistant individuals were too rare for our sampling and bioassay procedures to detect in significant numbers.

The most cyhexatin-susceptible T. pacificus colony was colony D (Table 1 and Fig.1). The lc/pm lines for susceptible T. urticae (colony A) and T. pacificus (colony D) were statistically the same based on the results of the likelihood ratio test of equality (Russell et al. 1977), indicating equal responses for the two species.

Colony F (Table 1 and Fig. 1), a T. pacificus colony, responded with intermediate cyhexatin-resistance; colony G was the most cyhexatin-resistant T. pacificus colony collected (greatest shift in lc/pm line). The LC_{50} of colony F was only 3-fold greater than that of colony D while the LC_{50} of colony G was 5-fold greater. The difference between colonies D and F remained the same at the LC_{90} level, while the difference between colony D and colony G increased to 12-fold. However, at the LC_{10} level the 95% confidence intervals for colonies D, F, and G overlap as shown in Table 1 and Fig. 1, indicating that susceptible individuals were present in each of these populations. The frequency of the most resistant individuals in colony G (defined here as survivors of 178 ppm) was less than 4%, as indicated by the plateau on the lc/pm line at 96% mortality. The fact that the LC_{10s} remained similar while the LC_{50s} and LC_{90s} increase from colony D to F to G, in addition to the discontinuity of the lc/pm lines for the resistant colonies indicate that colonies F and G were not genetically homogeneous with respect to cyhexatin susceptibility (Hoskins & Craig 1962). Therefore, linear probit-regression analysis is inappropriate for the data from colonies F and G (Neter

& Wasserman 1974). The results for cyhexatin suggest that T. urticae is still controllable while, cyhexatin-resistance in T. pacificus may reduce the efficacy of this acaricide in some orchards.

Propargite. The most propargite-susceptible T. urticae individuals were collected from site A (Table 2 and Fig. 2). The most susceptible T. pacificus colony collected (D, Table 2 and Fig. 3) had an LC_{50} 8-fold greater than that for the propargite-susceptible T. urticae colony (A). This indicates that T. pacificus may have a natural tolerance to propargite, thus requiring higher treatment rates to control susceptible T. pacificus than T. urticae.

Slopes of the propargite lc/pm lines indicated that the populations were relatively homogeneous with regard to susceptibility. The X^2 value for colony A, however, was significant at < 0.005 indicating deviation from the linear probit-regression model.

Colony B (Table 2 and Fig. 2) responded with intermediate propargite-susceptibility; colony C exhibited the highest levels of propargite-resistance of the T. urticae populations sampled in 1985. At the LC_{50} level, colony A was 3-fold more susceptible than

colony B and 42-fold more susceptible than colony C. At the LC_{90} level, the difference between colonies A and B increased to 7-fold and the difference between colonies A and C increased to 785-fold using the extrapolated LC_{90} values for colony C (which was beyond the range of testable concentrations). However, the 95% confidence intervals for the LC_{10s} of colonies A, B, and C overlapped (Table 2 and Fig.2), indicating the presence of susceptible individuals in both colonies B and C. This demonstrates that colonies B and C were heterogeneous with regard to propargite susceptibility.

Colony E, a T. pacificus colony, responded with intermediate propargite-susceptibility (Table 2 and Fig. 3); colony G was the most propargite-resistant T. pacificus collected in 1985. Although the propargite LC_{50} for colony E was similar to that of colony D (only 1.5-fold difference), colony E was considered of intermediate resistance because the line reached a plateau at ca. 92% mortality, and had a lower slope resulting in similar LC_{10s} but a 3-fold difference in LC_{90s}. Between colonies D and G there was a 14-fold difference in LC_{50s} and using an extrapolated LC_{90} value for colony g there was a 165-fold difference in LC_{90s}. At the LC_{10} level, however, colonies D and G

responded with similar levels of susceptibility, indicating that colony G was a highly heterogeneous population with regard to propargite susceptibility.

Another indication that colonies E and G were heterogeneous is the fact that when we treated a similar populations with a high concentration of propargite, the susceptible individuals were eliminated, and the resulting slope was steeper (Keena & Granett 1985). Between colony D and the selected T. pacificus colony tested on cotton (Keena & Granett 1985) there is a 29-fold difference in LC_{10s}, a 44-fold difference in LC_{50s}, and a 67-fold difference in LC_{90s}.

Relevance of Findings to Efficacy in the Field. Extrapolations from laboratory detected resistance to the field cannot be made without field trials. Field trials will determine whether a specific frequency and intensity of resistance estimated by laboratory bioassays will result in a loss of chemical efficacy in the field. Independent of such trials, however, we can discuss certain factors that pertain to control. First, because of interspecific differences in susceptibility, the relative frequencies of each species present in a particular orchard are important.

Our data show that the most susceptible T. pacificus we collected are more tolerant of propargite than T. urticae and that populations of T. pacificus have developed cyhexatin resistance while T. urticae have not. Thus T. pacificus is likely to be more difficult to control with either chemical.

Second, colonies from the field were heterogeneous with regard to either intensities or frequencies of propargite and cyhexatin resistance. Based on the definition of resistance (a shift to higher concentrations in the lc/pm line), various intensity levels of cyhexatin and propargite resistance are present in the orchards. Differences in intensity are indicated by shifts in any portion of the lc/pm line. Differences in the frequencies of individuals responding with a given intensity of resistance are indicated by shifts in the height of the plateau preceding the steeply rising linear portions of the lc/pm response line. In general, when populations of spider mites are heterogeneous with respect to resistance the acaricides will remain effective at controlling a portion of the population. Control failure will only occur when the frequency of individuals able to survive the field application rate

exceeds the tolerance threshold for the spider mites on the crop of interest.

Finally, susceptible individuals were present in all colonies. The presence of susceptible individuals in the same population with resistant individuals may allow the two types to intermate and reduce the frequency of resistant individuals in the population. However, treatment with the acaricide, for which resistance is present, selectively removes susceptible individuals from the population and maintains the resistant individuals at relatively high frequencies. By temporarily discontinuing the use of the acaricide, for which resistance is present and causing loss of efficacy of the acaricide, susceptible individuals will not be selectively removed from the population. Subsequent intermating with resistant and susceptible individuals may reduce the frequency of resistance in the population to a controllable level.

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Table 1. Log concentration/probit mortality regressions of T. urticae and T. pacificus Colonies collected from California almonds exposed to residues of cyhexatin

Species	Colony	n	Slope (± SE)	LC_{10} (ppm AI) (95% CL)	LC_{50} (ppm AI) (95% CL)	LC_{90} (ppm AI) (95% CL)
T. urticae	A	1,250	3.10 (0.28)	2.02 (0.94-3.00)	5.23 (3.78-6.38)	13.54 (11.18-18.39)
T. pacificus	B	788	3.20 (0.28)	2.20 (1.34-3.01)	5.53 (4.34-6.61)	13.90 (11.72-17.40)
T. pacificus	F	958	3.15 (0.22)	5.86 (2.80-8.62)	14.97 (10.80-18.87)	38.21 (29.56-58.14)
T. pacificus	G	1,813	1.56 (0.09)	3.94 (1.97-6.38)	26.14 (19.17-33.54)	173.37 (130.76-251.37)

Table 2. Log concentration/probit mortality regressions of T. urticae and T. pacificus colonies collected from California almonds exposed to residues of propargite

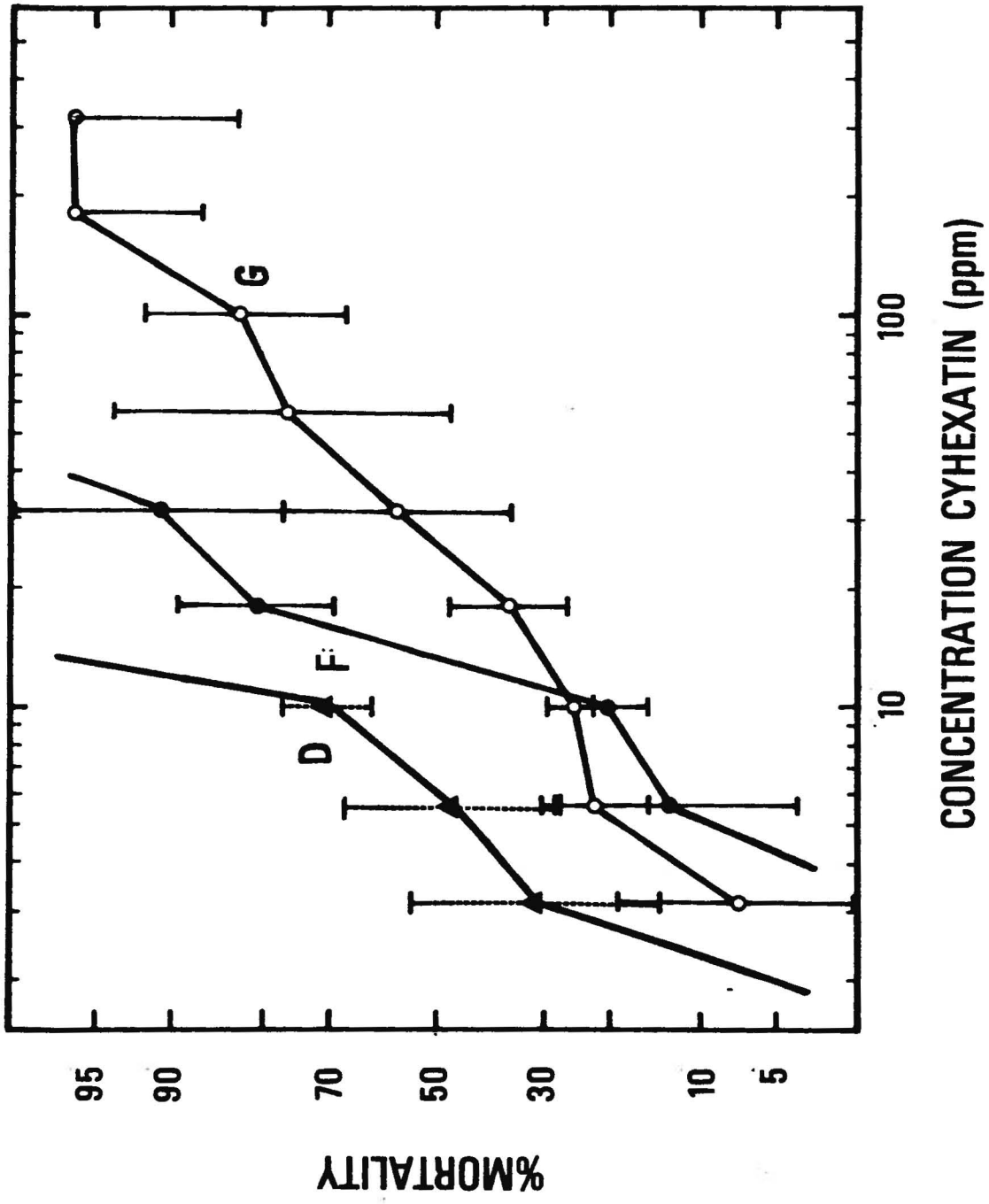
Species	Colony	n	Slope (± SE)	LC_{10} (ppm AI) (95% CL)	LC_{50} (ppm AI) (95% CL)	LC_{90} (ppm AI) (95% CL)
T. urticae	A	1,213	2.63 (0.17)	10.69 (6.77-14.47)	31.72 (26.22-37.19)	97.08 (79.42-128.58)
T. urticae	B	2,103	1.50 (0.11)	13.67 (6.07-23.61)	98.33 (68.91-128.56)	707.11 (538.67-1,016.60)
T. urticae	C	2,914	0.74 (0.06)	24.83 (6.00-63.34)	1,351.33 (840.12-1984.39)	>10,000.00 ---
T. pacificus	D	1,141	2.18 (0.15)	65.71 (45.51-86.58)	255.50 (217.42-293.73)	1,017.16 (855.21-1,265.76)
T. pacificus	E	1,676	1.34 (0.07)	41.85 (24.72-62.80)	379.83 (299.58-468.07)	3,447.69 (2,640.10-4,797.07)
T. pacificus	G	1,384	0.77 (0.08)	75.98 (24.56-159.73)	3,569.57 (2,393.65-5,553.34)	>10,000.00 ---

Figure Captions

/Fig. 1. Susceptibility of T. pacificus colonies D, F, and G to cyhexatin, with means and 95% confidence intervals.

/Fig. 2. Susceptibility of T. urticae colonies A, B, and C to propargite, with means and 95% confidence intervals.

/Fig. 3. Susceptibility of T. pacificus colonies D, E, and G to propargite, with means and 95% confidence intervals.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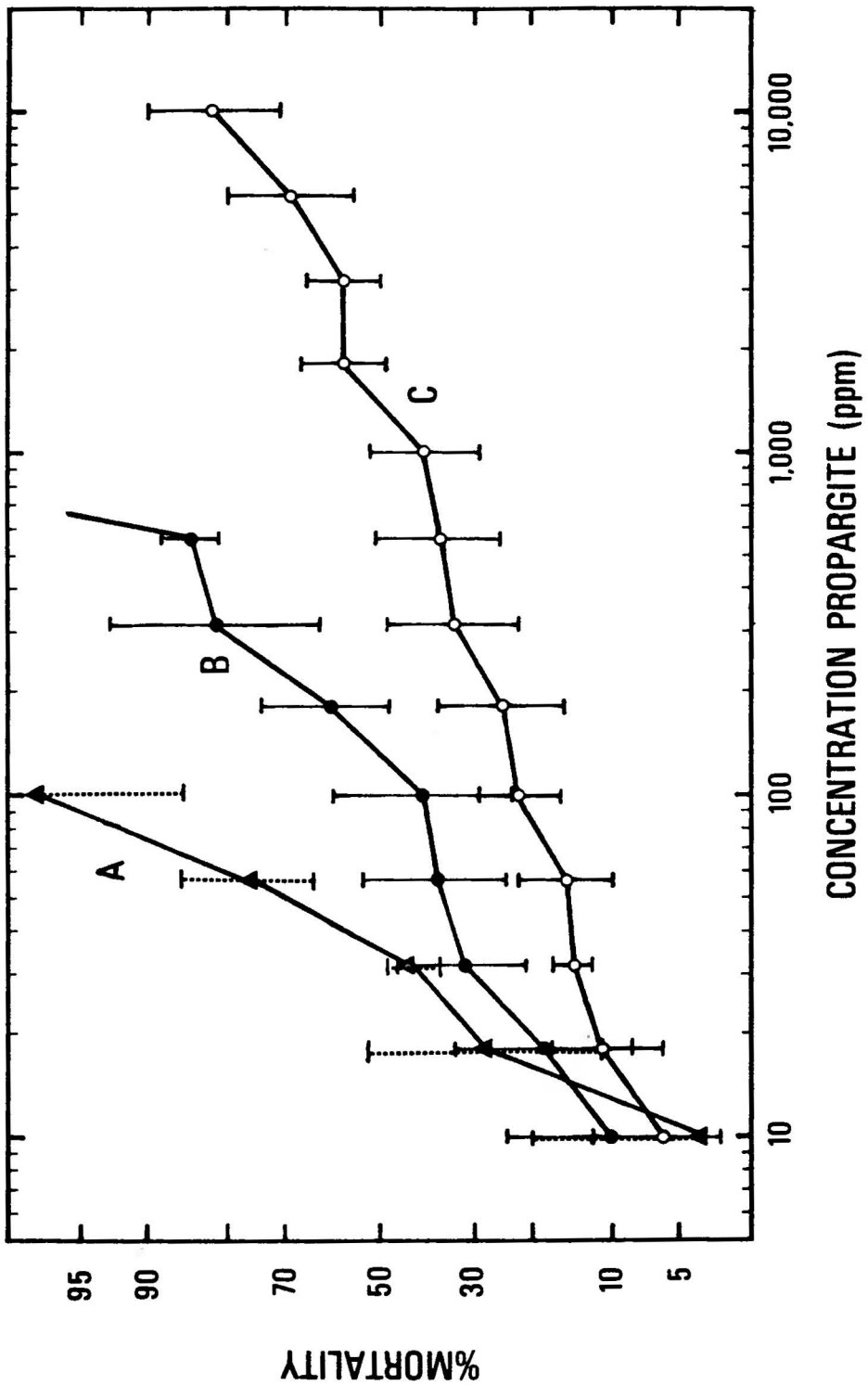
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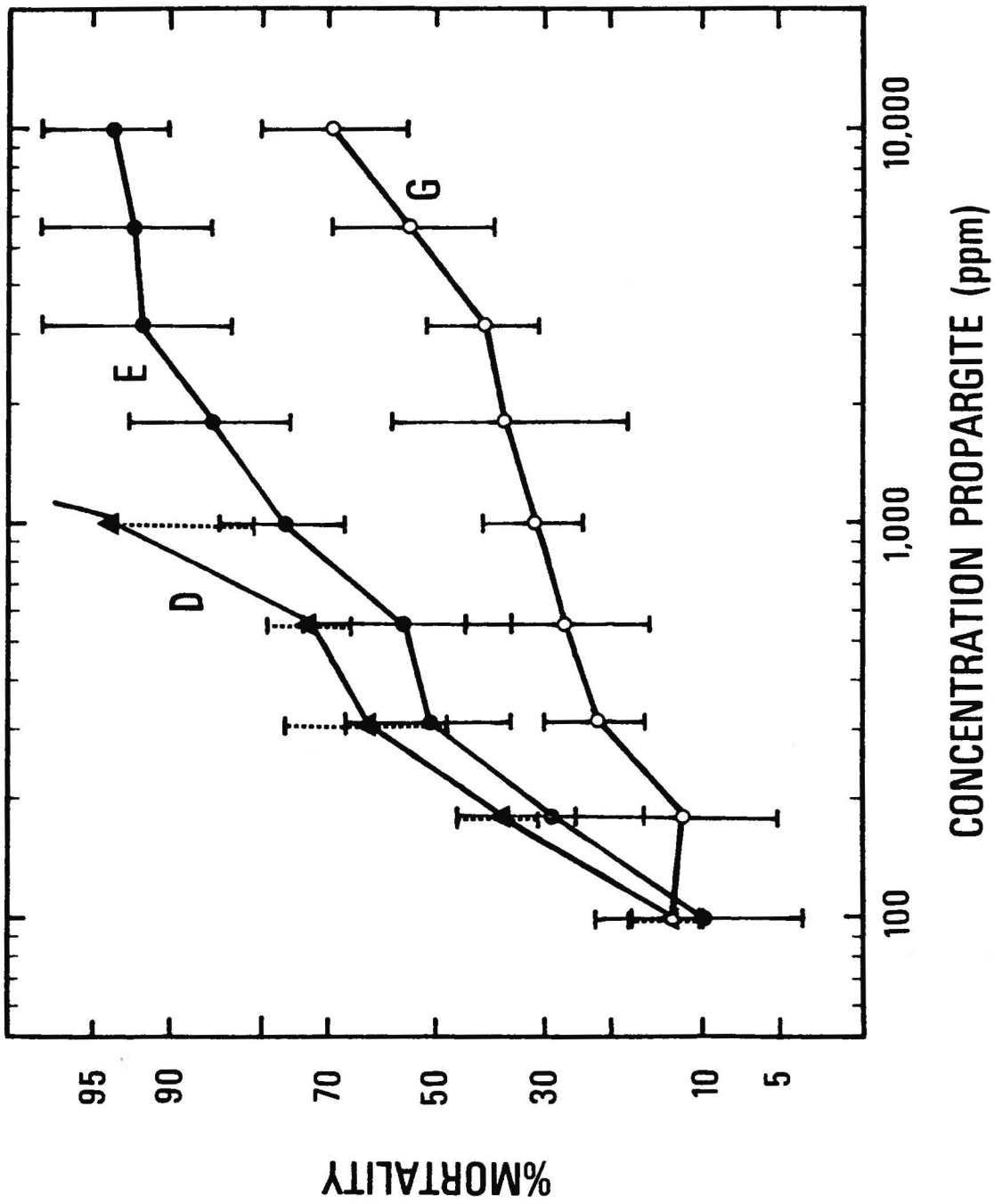
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