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Superparasitism by *Opius oophilus,*
A Parasite of the Oriental Fruit Fly[1]

HARRY K. KAYA and TOSHIYUKI NISHIDA

INTRODUCTION

The phenomenon of superparasitism is often encountered by investigators in biological control. However, the meaning of superparasitism appears to differ among various investigators. According to Fiske (1910), who first used the term, it meant the simultaneous attack of a host either by two or more species of primary parasites or by one species more than once. Subsequently, this term was given a more restricted meaning than the one used by Fiske. According to Smith (1916), superparasitism meant the superabundance of individuals of a single parasite species attacking a single host so that there are more parasite larvae than can reach maturity. The phenomenon in which there was a simultaneous attack by two or more species of primary parasites in a single individual host was referred to as multiple parasitism. Thus, the original term of Fiske was split into superparasitism in a restricted sense and multiple parasitism. Other terms used synonymously with superparasitism are: "accidental secondary parasitism," "cannibal superparasitism," "mixed superparasitism" (Pierce, 1910), "epiparasitism," "hyperparasitism" (Salt, 1934), and "multiple parasitism" (Sweetman, 1958). In the present study the term superparasitism is used in accordance with the definition by Smith (1916).

Superparasitism has been discussed in the literature by various investigators, but the true nature of this phenomenon is not clearly understood. Fiske (1910) stated that superparasitism occurs because the females, being incapable of discerning between parasitized and unparasitized hosts, oviposit at random in parasitized and unparasitized hosts. Therefore, the incidence of superparasitism was dependent upon the laws of probability. Although this viewpoint was accepted by Thompson (Salt, 1934), other workers such as Salt (1934), Ullyett (1936), and Lloyd (1938) working with *Trichogramma evanescens* Westwood, *Microplectron fuscipennis* Zett., and *Ooencyrtus kuvanae* Howard, respectively, found that these insects do not distribute their eggs at random. They concluded that superparasitism occurred when unparasitized hosts were not available to the parasites. In 1932, Salt pub-

lished data on the natural superparasitism by *Collyria calcitrator* Graven-horst, an ichneumonid parasite of *Cephus pygmaeus* L., the wheat stem saw-fly. It was shown mathematically that there was a certain degree of dis-crimination between parasitized and unparasitized hosts by the ovipositing parasite. In addition to discrimination between parasitized and unparasitized hosts, there are other factors that influence the extent of superparasitism. Varley (1941) stated that superparasitism can occur when: (1) the eggs are not laid singly, (2) the hosts are not equally available, (3) the search for hosts is not evenly distributed in space, or (4) the parasite has little or no tendency to avoid superparasitism. DeBach and Smith (1947) stated that superparasitism occurs when there is a high ratio of parasites to hosts.

The present study is concerned with some aspects of superparasitism in the eggs of the Oriental fruit fly, *Dacus dorsalis* Hendel, by the braconid parasite, *Opius oophilus* Fullaway. Besides *D. dorsalis*, it is known that *O. oophilus* will oviposit in the eggs of other fruit flies, *Dacus cucurbitae* Coq. and *Ceratitis capitata* Wied. In addition to *D. dorsalis*, a limited amount of investigation was carried out on superparasitism by *O. oophilus* in the eggs of these fruit flies.

METHOD OF STUDY

The major portion of this study was carried out on the island of Oahu. However, a limited number of host egg samples were obtained from the is-lands of Maui and Molokai. All the climatological data mentioned in this study were obtained from Annual Summary, Climatological Data, U. S. Weather Bureau, 1953–1955 and 1960–1964; Taliaferro, 1959; and unpub-lished data, Waimanalo Experimental Farm, Hawaii Agricultural Experi-ment Station, University of Hawaii. The general methods of study are given in this section. Details of others are given under the appropriate sections.

The Sampling Areas

The sampling of host eggs in guava fruits (*Psidium guajava* L.) was carried out on Oahu in areas where guava shrubs were found in abundance. The guava areas are found in vegetation zones C and D, ranging from sea level to approximately 5,000 feet elevation (Ripperton and Hosaka, 1942). In these areas the guava was observed to fruit throughout the year. However, as will be mentioned later, there were seasonal fluctuations in the amount of fruits produced. Four guava fruit sampling sites were established within these zones which are best suited for guavas in order that fruits in sufficient quantity would be available all year round. The sampling sites were located in Helemano, Kailua-Kaneohe, Waimanalo, and Tantalus (Figure 1). The initial sampling was started in January, 1965, and was continued at monthly intervals throughout the following 12-month period.

Helemano is located at an elevation of about 960 feet on the plateau between the Koolau and Waianae Ranges on Oahu. Here dense groves of
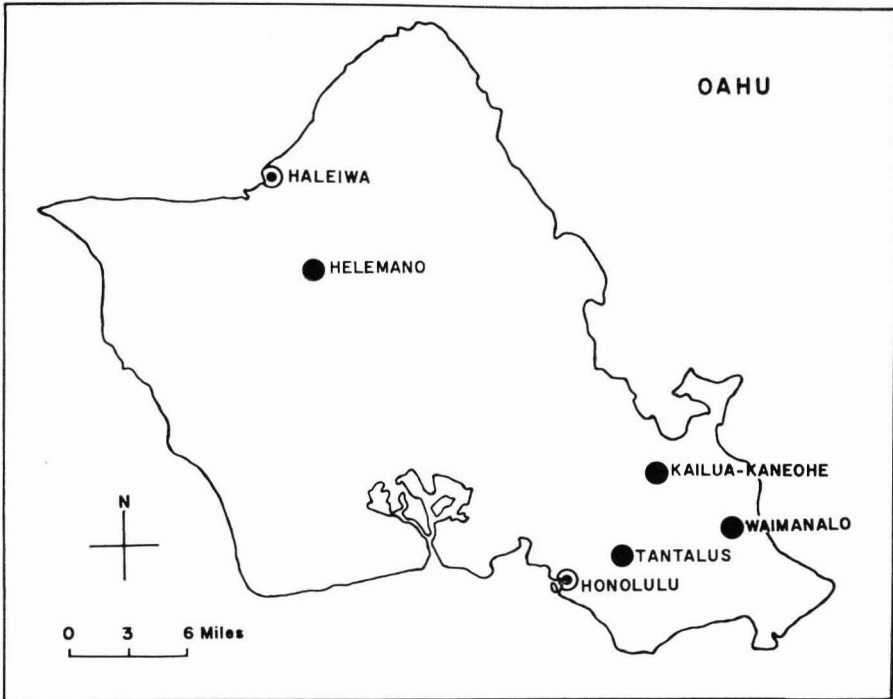
FIGURE 1. Island of Oahu showing the four sampling locations.

guava trees are found in gulches which are surrounded by pineapple fields. The annual mean temperature of this area is 70.9°F, and the annual mean rainfall is 53.5 inches.

Kailua-Kaneohe is located on the windward side of Oahu at an elevation of about 280 feet. Here dense guava groves are found on pasture land. The annual mean temperature is 72.0°F, and the annual mean rainfall is 51.3 inches.

Waimanalo is located on the windward side of the island at the foot of the Koolau Range. The guava samples were taken from the Waimanalo Experimental Farm, Hawaii Agricultural Experiment Station, University of Hawaii, at 60 feet elevation. The fruits were collected from the cultivated variety of guavas which were being grown for seedling selections. The annual mean temperature is 75.3°F, and the annual mean rainfall is 45.1 inches.

Tantalus is located on the leeward side of the Koolau Range near Honolulu. The sampling area is about 1,000 feet elevation, where the guava trees grow wild on the hillside. The annual mean temperature is 67.0°F, and the annual mean rainfall is 96.1 inches.

Collection of Host Fruits

Since guava fruits are generally available throughout the year, most of the studies were carried out using this host fruit. Mature green to firm ripe fruits, usually 40, were collected from each sampling area. The fruit samples were taken to the laboratory and the required number of host egg samples was taken from each of these lots of fruits.

Examination of Host Eggs

The extent of parasitism and superparasitism was determined by dissecting the fruit fly eggs in the laboratory under a binocular microscope. The fruit fly eggs were removed from the host fruits and placed in Ringer's solution in a Petri dish. From this group of eggs a sample of 25 was then taken at random and examined. The number of parasite eggs and larvae in each host egg was recorded. In sampling the host eggs, only those which had a glistening, normal appearance were considered. The dull-looking eggs or those covered with fungal growth were not considered in the sampling. To obtain the percentage of superparasitism, the number of host eggs superparasitized was divided by the total number parasitized in the sample and multiplied by 100.

Guava Fruit Abundance

At each sampling, an index of fruit abundance was taken to obtain data on the relative changes in abundance of fruits. Since ripe fruits were more readily visible from a distance than green ones, the index used was based only on ripe fruits.

The method of obtaining this index consisted of standing on a high vantage point and scanning an area approximately one-tenth of an acre. From this point the number of ripe yellow fruits was rapidly counted. The index was based on an arbitrary scale from 0–5; 0, no fruits; 1, 1–20 fruits; 2, 21–50 fruits; 3, 51–100 fruits; 4, 101–200 fruits; and 5, 201 or more fruits.

BIOLOGY OF HOST AND PARASITE

The biologies of *D. dorsalis* and *O. oophilus* were studied by Haramoto (1953) and Bess and Haramoto (1961). A brief review will be given here stressing those aspects pertinent to the understanding of superparasitism.

The fruit fly punctures the epidermis and lays eggs in the rind or mesocarp of the host fruits (Figure 2*B*). The ovipositional puncture in the guava fruit usually has a small aperture in the epidermis, but in the mesocarp it is a pouchlike cavity. The number of eggs per cavity is highly variable, ranging from 5 to 25 per cavity. The fly has the habit of laying eggs either in cavities made by herself or in those made by other flies, and since the eggs may be laid by different females at different times, it is possible to find newly laid eggs, empty egg chorions, and dead eggs covered with fungi in the same cavity.
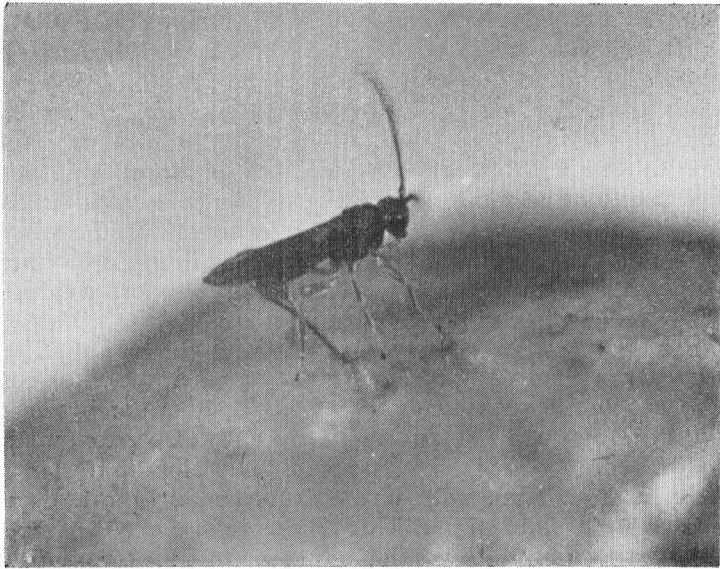
A

B

FIGURE 2. *A, O. oophilus* ovipositing in *D. dorsalis* eggs in guava fruit; *B*, cross section of guava fruit showing an oviposition puncture containing eggs of *D. dorsalis*.

The egg of *D. dorsalis* hatches in about 30 to 40 hours after deposition. The newly hatched larva then bores deeply into the fruit. When fully developed the mature larva leaves the fruit and pupates in the soil. After 10 to 14 days, the adult fly emerges, and egg laying begins about 10 days later depending on diet.

According to Bess and Haramoto (1961), the degree of fruit maturity has a marked effect on egg laying in various fruits by *D. dorsalis*. In passion fruits, *Passiflora edulis* f. *flavicarpa* Degener, eggs are laid in the soft young fruits, but in others such as guava, the fly oviposits in green to fully ripe fruits. However, the mature green fruits appear to be more conducive to egg laying than either the green or ripe fruits.

O. oophilus is an egg-larval parasite. It lays eggs in the eggs of *D. dorsalis* after inserting the ovipositor into the fruit cavity where the host eggs are present. The egg of the parasite usually hatches after the host egg has hatched. Out of a total of 1,438 parasites, 1,199, or 83.4 percent, were eggs, and 239, or 16.6 percent, were larvae. The parasite develops in the larval and pupal stages of its host. The adult parasite then emerges 10 to 12 days after the puparium is formed. In the field the adult parasite may be frequently seen either walking on or inserting the ovipositor in fruit fly punctures in guava and other fruits (Figure 2*A*).

In Hawaii this parasite is known to oviposit in the eggs of three economically important fruit flies: *D. dorsalis, D. cucurbitae,* and *C. capitata* (Haramoto, 1953). Although egg deposition occurs in these three species, this parasite develops in *D. dorsalis* and *C. capitata,* but not in *D. cucurbitae* (Nishida and Haramoto, 1953; Haramoto, 1953).

Being a solitary parasite, only a single adult *O. oophilus* emerges from the host puparium. According to van den Bosch and Haramoto (1953), superparasitism frequently occurs, but the supernumerary individuals are eliminated soon after the first parasite larva hatches. It was stated that death of the supernumerary individuals was caused by some physiological reactions.

There are a number of parasites of *D. dorsalis* in Hawaii and other countries (Clausen *et al.*, 1955). In Hawaii the species commonly associated with *D. dorsalis* are *Opius oophilus, Opius vandenboschi* Fullaway, and *Opius longicaudatis* (Ashmead) (Bess and Haramoto, 1961). Since *O. oophilus* is the only known egg-larval parasite, it is unlikely that the activities of these other parasites could have influenced the superparasitism data obtained in this study.

EGG DISTRIBUTION AND ITS EFFECT ON PARASITISM

The incidence of superparasitism is closely associated with the manner in which the female parasite distributes eggs among its hosts. Egg distribution may be random or nonrandom. Random distribution implies that the female did not discriminate between parasitized and unparasitized hosts and that whether or not a particular host received parasite eggs depended on the

laws of probability. On the other hand, nonrandom distribution implies that the parasite exercised at least a certain amount of discrimination and avoided ovipositing in already parasitized individuals. The egg distribution will then be different from that which would occur when eggs are distributed purely by chance. From these considerations it becomes evident that the incidence of superparasitism would be dependent upon the ability of the parasite to distinguish between parasitized and unparasitized hosts.

The manner in which a female parasite distributes progeny among host individuals has been a subject of controversy. This subject is of fundamental importance because mathematical models describing host-parasite relationships take into consideration the manner in which eggs are distributed. Salt (1934) pointed out that ". . . superparasitism has been observed and defined and, on the assumption that parasite progeny (*sic*) are distributed at random, treated mathematically." He stated further that parasitism has been assumed to be governed by chance but has not been experimentally examined.

The field data obtained in this study were examined to determine whether *O. oophilus* distributed its progeny at random or whether it had the ability to distinguish between parasitized and unparasitized hosts. The method of analysis used was the one developed by Stoy and used by Salt (1932).

It was shown by Stoy that if a parasite distributes progeny at random, one at a time, the number of hosts that will contain a given number of eggs is described by the following relationship:

$$Z = N \, C_p^x \left(\frac{1}{N}\right)^p \left(1 - \frac{1}{N}\right)^{x-p}$$

where N is the number of hosts; x, the number of parasite eggs distributed; Z, the number of hosts containing p eggs; and where C_p^x represents the number of ways p eggs can be distributed by x number of parasite eggs. The equation for the possible number of combinations is:

$$C_p^x = \frac{x \, (x-1) \, (x-1) \ldots (x-p+1)}{p \, (p-1) \, (p-2) \ldots 1}$$

Stoy (Salt, 1932) pointed out that when x and N are large, Z can be determined by fitting the data to the Poisson series. The above-mentioned formula and that of the Poisson series (Snedecor, 1962) were used in the analysis of the data obtained in this study.

To determine the extent of superparasitism by *O. oophilus* in the field, the data obtained during 1965 from dissecting 1,192 eggs of *D. dorsalis* were used. Of these, 941, or 78.9 percent, were parasitized by *O. oophilus.* Of the 941 parasitized eggs, 607 were infested with one, 213 with two, 85 with three, 28 with four, 5 with five, 1 with six, and 1 with seven individuals. There were 1,438 individuals of *O. oophilus* distributed among 1,192 hosts, and out of the 941 parasitized eggs, 333, or 35.4 percent, were superparasitized. These data indicate that a little more than one-third of all parasitized *D. dorsalis* eggs in the field were superparasitized.

In order to test the hypothesis of random or nonrandom distribution of eggs by *O. oophilus,* the observed and expected frequencies of hosts containing various numbers of parasites were compared. These frequencies, presented in Figure 3, show that in the observed distribution the frequencies of zeros, twos, and threes were too low and the frequency of the ones was too high. The frequencies of fours, fives, sixes, and sevens approximated closely the calculated values. A Chi-square test showed that the deviation be-
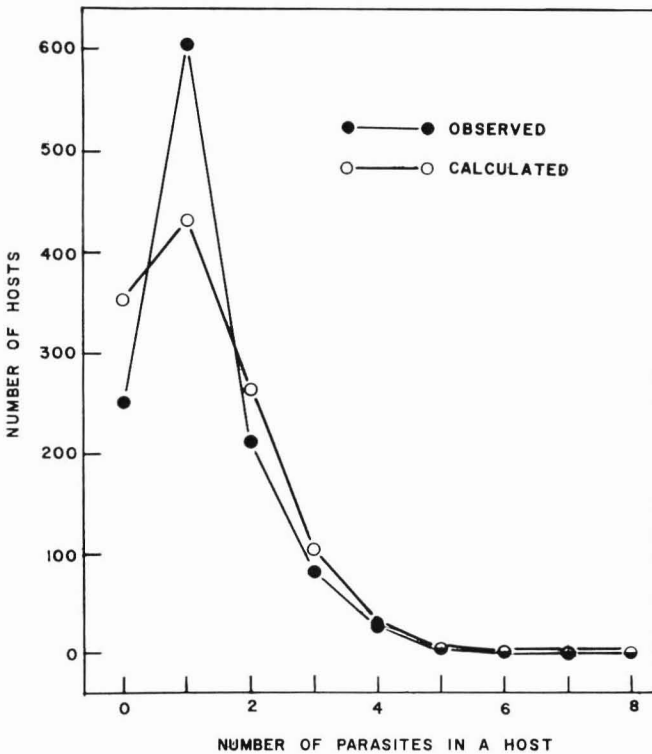
FIGURE 3. The observed and expected frequencies of hosts containing various numbers of parasites. Chi-square was 116.96, significant at P < 0.01.
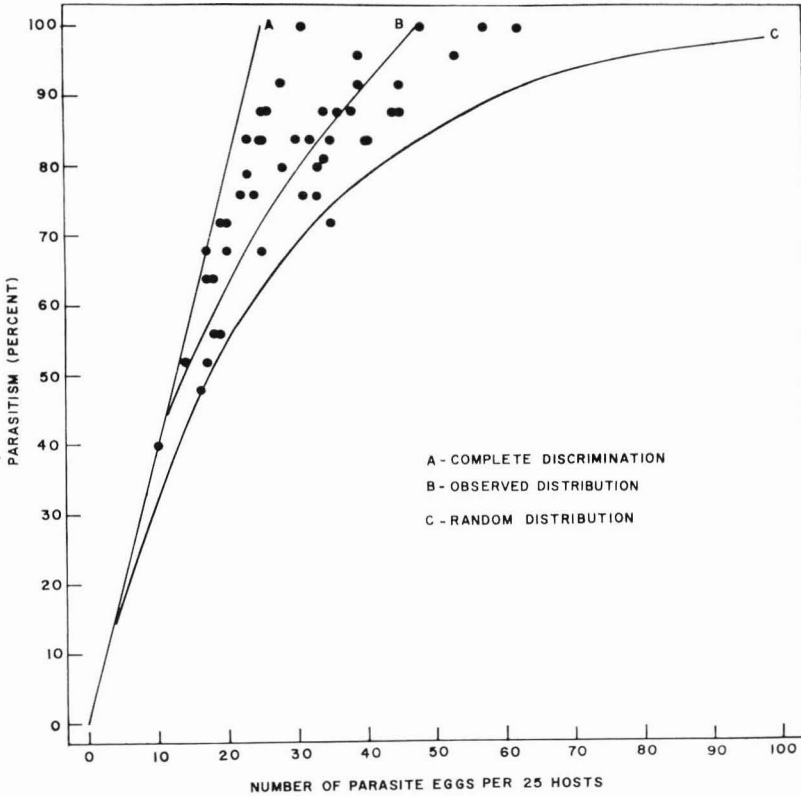
FIGURE 4. Relationship between the observed and hypothetical egg deposition and the percent parasitism by *O. oophilus*. *A*, The hypothetical distribution with complete discrimination between parasitized and unparasitized hosts; *B*, the observed distribution; and *C*, the hypothetical random distribution.

tween the observed and calculated distributions was highly significant (Chi-square equaled 116.96; P<0.01). These data suggest that *O. oophilus* did not distribute eggs at random. The data obtained in this study were also fitted to the Poisson series and the same conclusion was reached. An identical conclusion was made by Salt (1932, 1934) and Walker (1938) in the case of *Collyria calcitrator* Grav., *Ibalia leucospoides* Hoch., *Limnerium validum* (Cres.), and *Dacnusa areolaris* Nees.

Using the field data obtained in this study, the relationship between egg distribution and extent of parasitism was examined. Figure 4 shows the relationship between egg distribution and percent parasitism when the eggs were distributed in accordance with (1) complete discrimination, *A*; (2) partial discrimination, *B*; and (3) random distribution, *C*. Line *A* was obtained from the formula:

$$y = bx$$

where y equals the number of parasitized hosts; x, the number of parasite eggs per 25 hosts; and b, the slope of the line. Line B, "eyeball fitted," represents the observed egg distribution by $O.$ $oophilus$. Line C was obtained from the formula originally devised by Thompson and later modified by Stoy (Salt, 1932):

$$y = Ne^{-\frac{x}{N}}$$

where y equals the number of parasitized hosts; N, the number of hosts; x, the number of parasite eggs distributed; and e, the Naperian logarithmic base.

The data shown in Figure 4 show the relationship between the number of parasite eggs laid and the percent parasitism. The number of eggs laid by $O.$ $oophilus$ increased markedly as the percentage of parasitized hosts increased. The number of eggs, however, was less than that distributed at random and greater than that distributed with perfect discrimination. This situation exists at parasitization levels exceeding 45 percent because the lines, A, B, and C, tend to converge at low levels of parasitism.

FACTORS INFLUENCING SUPERPARASITISM

Seasonal and Locality Variations

An investigation of the seasonal changes in superparasitism of $D.$ $dorsalis$ eggs by $O.$ $oophilus$ was conducted by taking monthly guava samples from four localities for a 12-month period. Guava fruits were taken at random from four localities on Oahu: Helemano, Kailua-Kaneohe, Waimanalo, and Tantalus (Figure 1).

The data, presented in Figure 5, show that the highest and lowest incidence of superparasitism at the four localities did not occur during the same period. The incidence of superparasitism was low at Helemano during March and April; at Kailua-Kaneohe, in June; at Waimanalo, in July; and at Tantalus, in September. The highest incidence of superparasitism occurred at Tantalus in June; Kailua-Kaneohe, in October; and Helemano and Waimanalo, in November.

Although seasonal differences in superparasitism within each locality were noted, there appeared to be no differences in the mean superparasitism among the four localities. For the 12-month period, the mean percentages of superparasitism were Helemano, 38.7 ± 6.4; Kailua-Kaneohe, 35.2 ± 5.8; Waimanalo, 34.8 ± 6.6; and Tantalus, 24.6 ± 5.0. From these data it appears that although there were seasonal fluctuations, the overall population of $O.$ $oophilus$ at the four localities during the year was the same.
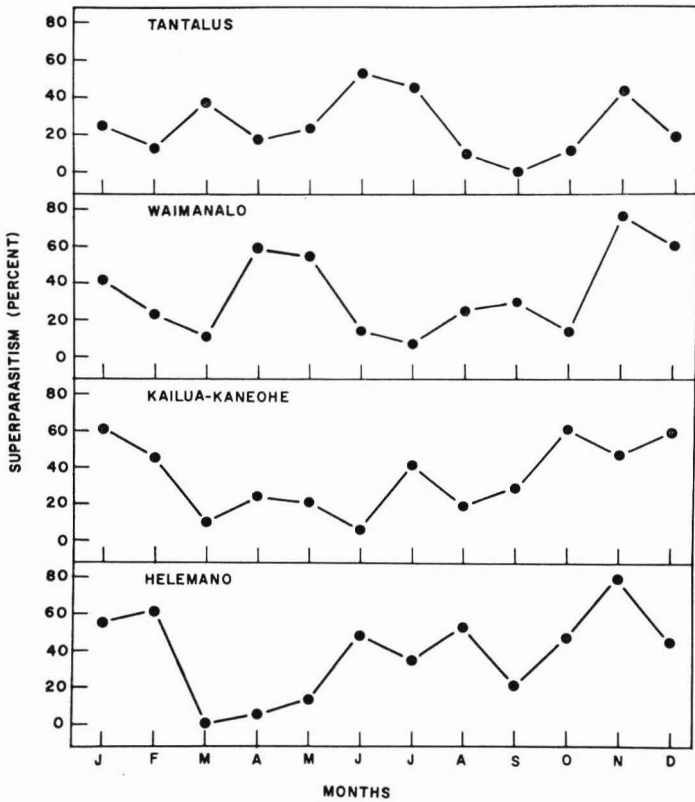
FIGURE 5. Seasonal fluctuations in superparasitism by *O. oophilus* at four localities on Oahu from January to December, 1965.

Effect of Elevation

The incidence of superparasitism in *D. dorsalis* eggs by *O. oophilus* at various elevations was studied by sampling eggs in guava fruits collected at various elevations. Egg samples were obtained from Manoa, at 280 feet; Tantalus 1, at 920 feet; Tantalus 2, at 1,000 feet; Tantalus 3, at 1,200 feet; Tantalus 4, at 1,400 feet; and Tantalus 5, at 1,600 feet. Additional data were obtained from egg samples collected at 2,000 feet elevation at Makawao, Maui. Data were also obtained from egg samples from wild peaches collected at Kula, Maui, at 3,000 to 3,600 feet elevations. The fruit samples from which egg samples were obtained were collected during September to December, 1965. A total of 1,819 host eggs was dissected.

The data obtained, depicted in Figure 6, indicate no apparent relationship between these two variables up to an elevation of about 2,000 feet. At elevations above 2,000 feet, there was a decline in the percentage of super-
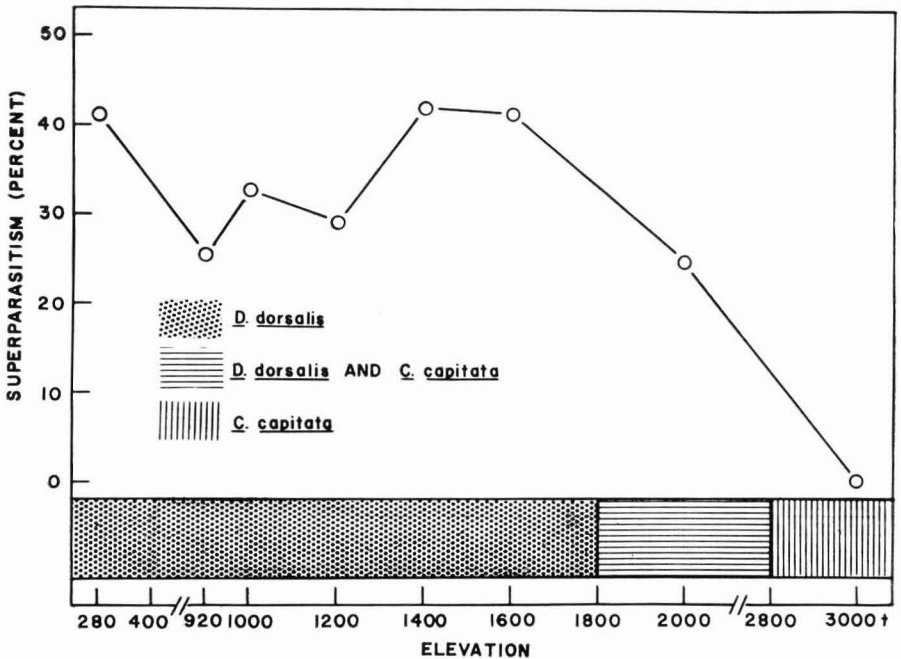
FIGURE 6. Relationship between superparasitism and elevation. The number of host eggs dissected at various elevations ranged from 70 to 318.

parasitism. It should be mentioned that at elevations higher than 2,000 feet, *C. capitata* eggs rather than *D. dorsalis* eggs were being sampled because it has been shown by Bess (1953) that at high elevations the latter species become scarce. In this study, it was noted that parasitism by *O. oophilus* was low above 2,000 feet elevation. Because the species of fruit flies changed with elevation, it is difficult to state whether elevation *per se* was the cause of the low incidence of superparasitism at high elevations. However, it was noted that under laboratory conditions, *O. oophilus* superparasitized eggs of *C. capitata* to the same extent as those of *D. dorsalis*. It appears that the low incidence of superparasitism in *C. capitata* at high elevations was due to adverse environmental conditions which affected the ovipositional behavior of *O. oophilus*.

Effect of Fruit Abundance

This study was conducted in a cultivated guava grove on the Waimanalo Experimental Farm, Hawaii Agricultural Experiment Station, University of Hawaii, Waimanalo. In this particular grove, there were trees with many fruits as well as trees with few fruits. The intensity of superparasitism between trees with many ripe fruits was compared with trees with few ripe fruits.

Trees with less than 14 fruits were considered "low fruit abundance" while trees with more than 30 mature fruits were considered "high fruit abundance." All the fruits from the low abundance trees were picked, and an equal number of fruits from the high abundance trees were picked at random. A total of 180 *D. dorsalis* eggs was sampled from fruits from the low abundance trees and 222 eggs from the high abundance trees.

The results of this study showed that superparasitism was 40.7 percent on trees with high fruit abundance and 54.8 percent on trees with low fruit abundance. Statistical analysis showed that these differences in superparasitism were not significant. In other words fruit abundance *per se* had no effect on superparasitism.

The effect of fruit abundance on superparasitism was also investigated by comparing data on the extent of superparasitism and fruit abundance. The data on superparasitism and seasonal fluctuations in guava fruit abundance, shown in Figure 7, indicate that there was no clear-cut relationship between superparasitism and fruit density. Superparasitism appeared to fluctuate independent of fruit density. It was noted that, in general, super-
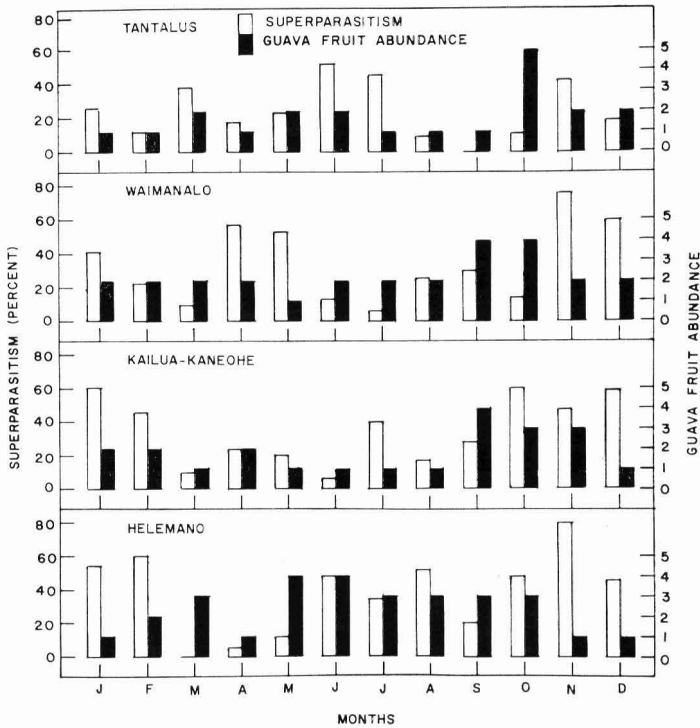

FIGURE 7. Seasonal fluctuations in superparasitism by *O. oophilus* and guava fruit abundance from January to December, 1965.

parasitism was higher during the winter months than during the summer months, but fruit density did not seem to affect the efficiency of the parasite. These conclusions are in agreement with those reached by Bess, Haramoto, and Hinckley (1963), who stated that fruit density had no effect on the efficiency of *O. oophilus*.

Effect of Fruit Maturity

According to Haramoto (1953) the ovipositional activity of *O. oophilus* is not influenced by the degree of ripeness of guava fruits. He found that the adult parasite is influenced by the ovipositional activity of the fruit fly adults. He presented field data that showed proportionately more *O. oophilus* were present on mature green guava fruits than on either semiripe or ripe guava fruits. Since no data were presented on the actual egg-laying activity, it is not known whether there were any differences in parasitism and super-parasitism in fruits of different degrees of ripeness. The purpose of this study was to determine whether there was any difference in superparasitism by *O. oophilus* in *D. dorsalis* eggs in guava fruits in various stages of ripeness.

To determine the effect of fruit ripeness on superparasitism, guava fruits in various stages of ripeness were sampled on Oahu at Poamoho (Poamoho Experimental Farm, Hawaii Agricultural Experiment Station, University of Hawaii), Waimanalo (Waimanalo Experimental Farm, Hawaii Agricultural Experiment Station, University of Hawaii), and Manoa. The guava fruits were arbitrarily segregated on the basis of color into three categories, green, semiripe, and ripe. Green fruits were those that were mature and pale green; semiripe fruits, those that were yellow-green; and ripe fruits, those that were yellow with no shades of green. The guava fruit samples were taken at various times during June to September. Usually, about 30 fruits were picked for each category of ripeness. A total of 2,421 host eggs was sampled from all the fruit categories.

TABLE 1. The effect of guava fruit maturity on superparasitism

SITE	MATURITY	TOTAL DISSECTED	TOTAL PARASITIZED	TOTAL SUPERPARASITIZED	PERCENTAGE SUPERPARASITIZED
Poamoho					
	Green	317	206	91	44.2
	Semiripe	325	246	111	45.1
	Ripe	167	111	42	37.8
Waimanalo					
	Green	325	226	62	27.4
	Semiripe	325	231	52	22.5
	Ripe	165	100	31	31.0
Manoa					
	Green	325	192	29	15.1
	Semiripe	316	195	17	8.7
	Ripe	156	94	21	22.3

The data obtained, shown in Table 1, indicate that there were no significant differences in superparasitism among the host eggs from green, semiripe, and ripe fruits. This indicates that the intensity of attack of host eggs in fruits in various stages of maturity is the same. This finding is of interest for it shows that the mere presence of more adults on a fruit is no indication that ovipositional activity will be greater in that fruit.

The data presented also indicate variations in the intensity of superparasitism in different areas. At Poamoho, superparasitism was 44, 45, and 38 percent; at Waimanalo, superparasitism was 27, 22, and 31 percent; and at Manoa, superparasitism was 15, 9, and 22 percent for the green, semiripe, and ripe fruits, respectively. The reason for the differences between areas is probably due to different parasite–host ratios.

Effect of Species of Host Fruits

The effect of host fruits on superparasitism was studied by collecting various species of infested fruits from as many areas as possible. Since the fruiting seasons varied, it was not possible to collect all samples at the same time. The host egg samples were taken from the respective fruits, and the extent of superparasitism was determined.

The data obtained from 19 species of host fruits collected on Oahu, Maui, and Molokai are given in Table 2. The fruits in which the highest superparasitism occurred were acerola, *Malpighia punicifolia* L.; rose apple, *Eugenia jambos* L.; bananas, *Musa* spp.; Terminalia, *Terminalia melanocarpa* F. Muell.; Surinam cherry, *Eugenia uniflora* L.; coffee, *Coffea arabica* L.; passion fruit, *Passiflora edulis* f. *flavicarpa* Degener (vines on fruit trees); mock orange, *Murraya exotica* L.; fig, *Ficus sycomorus* L.; and false kamani, *Terminalia catappa* L. Low superparasitism was encountered in ball kamani, *Calophyllum inophyllum* L.; mango, *Mangifera indica* L.; mountain apple, *Eugenia malaccensis* L.; natal plum, *Carissa grandiflora* A. DC.; strawberry guava, *Psidium cattleianum* Sabine; and papaya, *Carica papaya* L.

Superparasitism was not encountered from host eggs sampled from balsam apple, *Momordica balsamina* L.; loquat, *Eriobotrya japonica* Lindl.; passion fruit, *Passiflora edulis* f. *flavicarpa* (vines on trellises) ; or peach, *Prunus persica* (L.) Batsch. The absence of superparasitism and parasitism in balsam apple is probably because this fruit is a host of *D. cucurbitae* and not a host of *D. dorsalis*. It is known, however, that in the laboratory *O. oophilus* oviposits in eggs of *D. cucurbitae* (Nishida and Haramoto, 1953). The failure of *O. oophilus* to oviposit in *D. cucurbitae* eggs in the field is not known. In peaches and loquats, superparasitism was not encountered probably because they were obtained from Kula, Maui, at elevations of about 3,000 feet. It has been shown in a previous section that, in general, superparasitism was low at high elevations.

TABLE 2. Superparasitism by *O. oophilus* in some tephritid fruit fly eggs oviposited in various fruits. Fruit collections were made at various times during 1965. All localities are on Oahu except those otherwise indicated. (*Continued*)

COMMON NAME	SCIENTIFIC NAME	LOCALITY	NO. DISSECTED	NO. PARA-SITIZED	NO. SUPER-PARASITIZED	PERCENT SUPER-PARASITIZED
Acerola	*Malpighia punicifolia* L.	Waimanalo	25	18	3	16.7
		Waimanalo	23	2	0	0.0
		Waimanalo	12	8	5	62.5
		Waimanalo	18	13	6	46.2
		Waimanalo	23	16	7	43.8
Bananas	*Musa* spp.	Manoa	25	23	13	56.5
		Waimanalo	25	17	3	17.6
		Waimanalo	25	19	0	0.0
		Waimanalo	25	20	3	15.0
Balsam Apple[1]	*Momordica balsamina* L.	Honokowai, Maui	13	0	0	0.0
		Kunia	16	0	0	0.0
		Kunia	25	0	0	0.0
		Waimanalo	7	0	0	0.0
		Waimanalo	6	0	0	0.0
		Waimanalo	25	0	0	0.0
		Moku, Molokai	17	0	0	0.0
Coffee	*Coffea arabica* L.	Waimanalo	12	4	0	0.0
		Waimanalo	16	3	3	100.0
		Waimanalo	15	4	0	0.0
		Waimanalo	25	4	0	0.0
Fig	*Ficus sycomorus* L.	Kalihi	25	5	1	20.0
		Kalihi	7	0	0	0.0
		Waimanalo	25	10	2	20.0
		Waimanalo	25	15	2	13.3
Kamani, Ball	*Calophyllum inophyllum* L.	Manoa	25	2	0	0.0
		Kaneohe	19	18	1	5.6

(*Continued*)

[1] *D. cucurbitae* eggs.

TABLE 2. Superparasitism by *O. oophilus* in some tephritid fruit fly eggs oviposited in various fruits. Fruit collections were made at various times during 1965. All localities are on Oahu except those otherwise indicated. *(Continued)*

COMMON NAME	SCIENTIFIC NAME	LOCALITY	NO. DISSECTED	NO. PARA-SITIZED	NO. SUPER-PARASITIZED	PERCENT SUPER-PARASITIZED
Kamani, False	*Terminalia catappa* L.	Manoa	25	18	2	11.1
		Kuliouou	25	15	3	20.0
		Waimanalo	15	0	0	0.0
Loquat[2]	*Eriobotrya japonica* Lindl.	Kula, Maui	25	7	0	0.0
Mango	*Mangifera indica* L.	Honolulu	25	5	1	20.0
		Manoa	25	13	2	15.4
		Manoa	25	18	0	0.0
		Makiki	25	13	1	7.7
Mock Orange	*Murraya exotica* L.	Manoa	25	4	1	25.0
		Waimanalo	12	0	0	0.0
		Manoa	14	4	0	0.0
		Manoa	21	3	1	33.3
Mountain Apple	*Eugenia malaccensis* L.	Manoa	25	15	0	0.0
		Manoa	25	14	1	7.1
Natal Plum	*Carissa grandiflora* A. DC.	Tantalus	24	14	0	0.0
		Tantalus	25	11	0	0.0
		Tantalus	25	16	0	0.0
		Tantalus	25	2	0	0.0
		Tantalus	25	6	1	16.7
Papaya	*Carica papaya* L.	Waimanalo	25	16	4	25.0
		Waimanalo	25	9	0	0.0
		Waimanalo	25	15	0	0.0
		Waimanalo	25	9	0	0.0
		Waimanalo	25	18	0	0.0

(Continued)

[2]Most likely *C. capitata* eggs.

TABLE 2. Superparasitism by *O. oophilus* in some tephritid fruit fly eggs oviposited in various fruits. Fruit collections were made at various times during 1965. All localities are on Oahu except those otherwise indicated. *(Continued)*

COMMON NAME	SCIENTIFIC NAME	LOCALITY	NO. DISSECTED	NO. PARA-SITIZED	NO. SUPER-PARASITIZED	PERCENT SUPER-PARASITIZED
Passion Fruit[3]	*Passiflora edulis* f. *flavicarpa* Degener	Waimanalo	25	19	6	31.6
		Waimanalo	25	12	0	0.0
		Manoa	25	10	3	30.0
		Waimanalo	25	16	2	12.5
Passion Fruit[4]	*Passiflora edulis* f. *flavicarpa* Degener	Waimanalo	25	2	0	0.0
		Waimanalo	25	0	0	0.0
Peach[5]	*Prunus persica* (L.) Batsch.	Kula, Maui	285	17	0	0.0
Rose Apple	*Eugenia jambos* L.	Hana, Maui	25	10	2	20.0
		Kunia	25	16	2	12.5
		Kunia	25	23	12	52.2
		Kunia	25	9	1	11.1
Strawberry Guava	*Psidium cattleianum* Sabine	Waimanalo	16	0	0	0.0
		Waimanalo	25	7	0	0.0
		Manoa	25	12	1	8.3
		Waimanalo	25	2	0	0.0
		Makiki	25	3	0	0.0
Surinam Cherry	*Eugenia uniflora* L.	Manoa	25	0	0	0.0
		Manoa	25	9	2	22.2
Terminalia	*Terminalia melanocarpa* F. Muell.	Honolulu	25	2	1	50.0
		Manoa	25	6	0	0.0
		Manoa	25	17	5	29.4

[3]Vines on coffee and guava trees.
[4]Vines on trellises.
[5]Most likely *C. capitata* eggs.

The variation in superparasitism in passion fruit appears to differ with the situation in which this plant grows. Both parasitism and superparasitism were moderately high when passion fruit was growing in association with other fruit trees such as guava and coffee. However, when it was growing on trellises as a monocultural crop, parasitism was very low and super-parasitism was not observed. This observation shows that parasitism and superparasitism by *O. oophilus* can be influenced by other plants. It seems that the presence of other trees can make the habitat more attractive to *O. oophilus*.

Effect of Number of Host Eggs

Mature green guava fruits were collected from the field from various areas to determine the effect of the number of *D. dorsalis* eggs per oviposi-tional puncture on superparasitism by *O. oophilus*. Each puncture was examined under a binocular microscope and all the host eggs were removed. The eggs were then counted and dissected, and the number of *O. oophilus* eggs in each *D. dorsalis* egg was recorded.
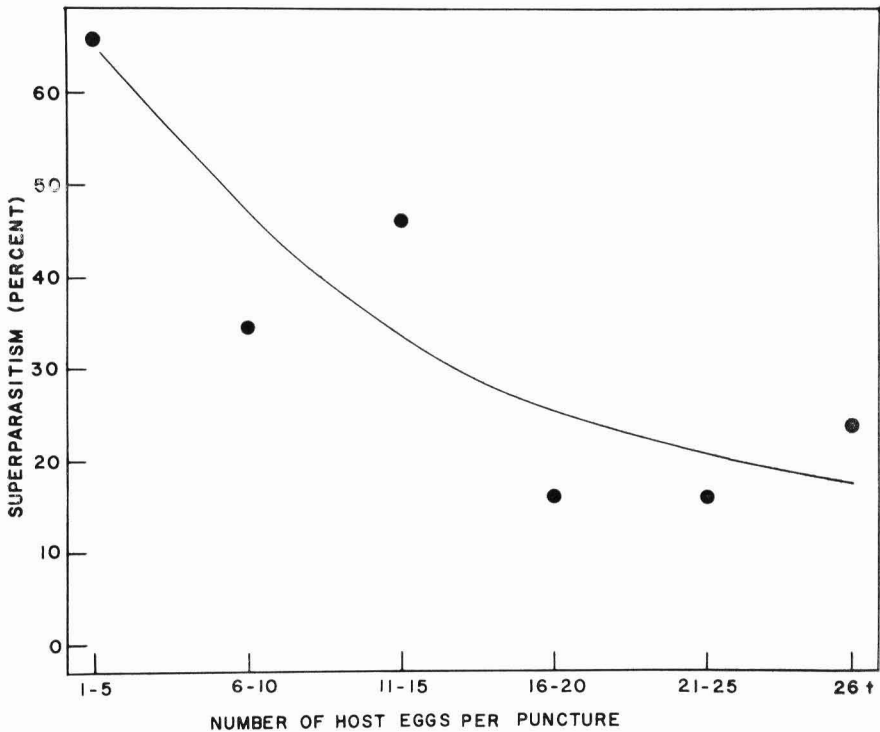

FIGURE 8. Relationship between superparasitism and host egg density.

The relationship between egg density and intensity of superparasitism, shown in Figure 8, indicates a decrease in superparasitism with an increase in host egg density. However, this relationship appears to be nonlinear. Superparasitism was 67.8 percent, 35.2 percent, 47.4 percent, 16.7 percent, 16.3 percent, and 24.6 percent in ovipositional punctures containing 1–5, 6–10, 11–15, 16–20, 21–25, and 26+ host eggs, respectively.

To determine whether host egg abundance at various elevations had an effect on superparasitism, guava fruit samples were taken from September to December, 1965. Data on *D. dorsalis* egg abundance and incidence of superparasitism were taken from localities situated at various elevations.

The data, presented in Figure 9, show changes in superparasitism and host egg density at various elevations. At 280 feet elevation, superparasitism was 41.3 percent at an egg density of 3.4 per fruit. At 920 feet, superparasitism was 25.9 percent at an egg density of 2.9 per fruit. At elevations between 920 and 1,600 feet, there was a tendency of superparasitism to increase with a decrease in host egg density from 2.9 to 0.5 per fruit.

The reason for the occurrence of a high percentage of superparasitism at 280 feet is not known. It may be that Manoa Valley from which the sample was taken was exceptionally favorable for *O. oophilus* ecologically, and for this reason there was a high rate of superparasitism in spite of the high host egg density. Since this study involved elevation and host egg density, it is not possible to distinguish the effects of elevation and host egg density.
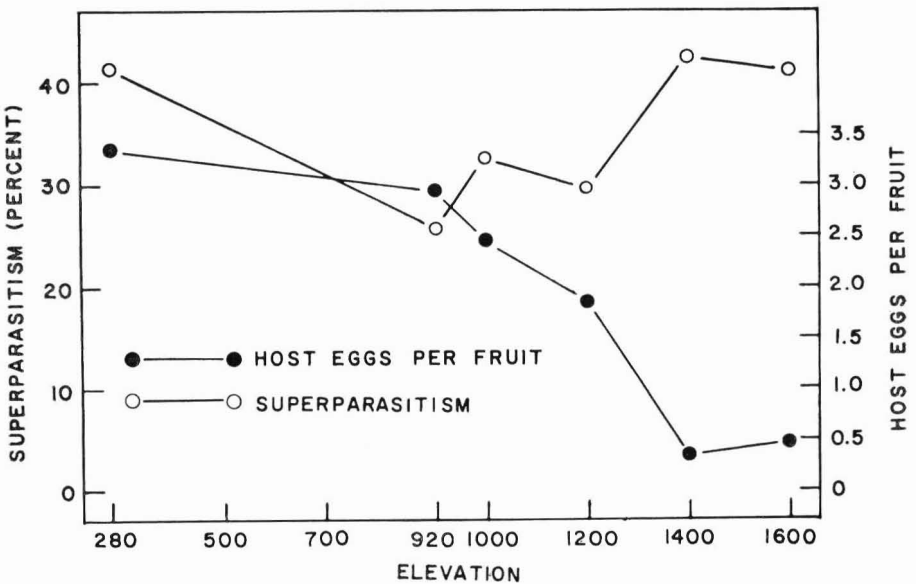

FIGURE 9. Relationship between host egg abundance and superparasitism at various elevations.

Relationship to Total Parasitism

In a previous section the relationship between the number of eggs laid and the percentage of parasitism was discussed. It was found that the number of eggs laid increased as the percentage of parasitism increased. The present discussion is a closely related aspect of the egg-laying habit of *O. oophilus.*

The incidence of superparasitism is closely related to the intensity of parasitism in the case of parasites that distribute eggs at random and in the case of those that possess an imperfect sense of discrimination between parasitized and unparasitized hosts. The relationship between superparasitism and parasitism of *O. oophilus,* a parasite which has an imperfect sense of discrimination, is shown in Figure 10. Although there were considerable variations, it is evident that the percentage of superparasitism increased as the percentage of parasitism increased. At 50 percent parasitism the average rate of superparasitism was about 10 percent, but as the percentage of parasitism increased, the incidence of superparasitism increased. At 100 percent parasitism the average rate of superparasitism was about 52 percent.
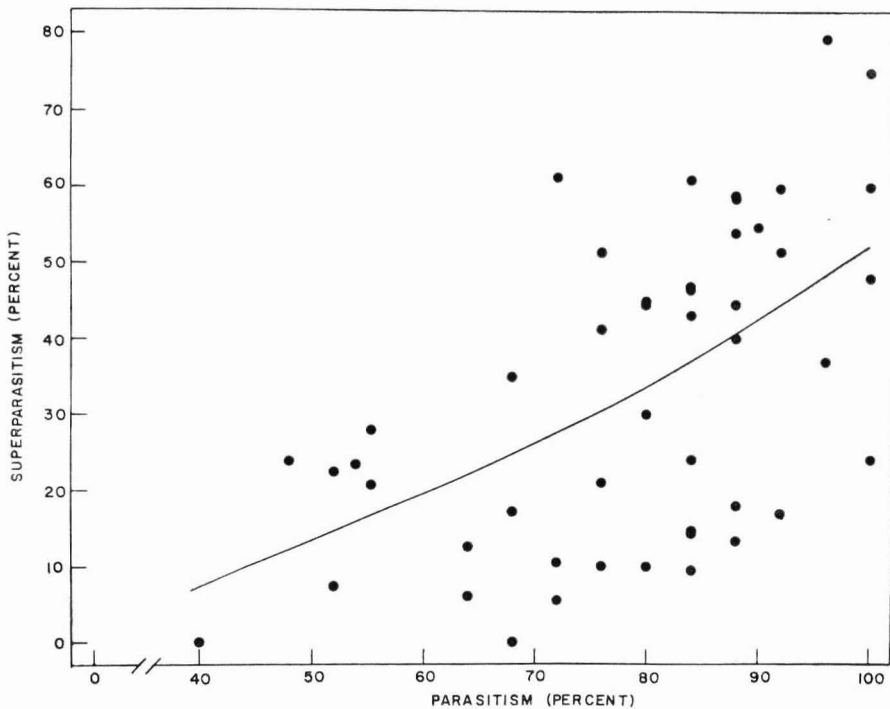

FIGURE 10. Relationship between superparasitism and parasitism of host eggs by *O. oophilus.* Data based on samples collected during 1965.

DISCUSSION

In the present study, the effect of the age of the eggs on superparasitism is not known because in the field samples the exact time of egg deposition could not be determined. Since the host eggs take about 30 to 40 hours to hatch, they could have been a few seconds to almost 2 days old. However, the nonviable eggs would have been exposed to the parasites longer than 2 days. If the parasite did not discriminate between viable and nonviable eggs, the "tail-end" of the frequency distribution would be prolonged because egg deposition by the parasite would have continued for 2 days or more.

The arrangement of the host eggs in the puncture probably affected the distribution of the eggs by *O. oophilus*. The host insect, *D. dorsalis*, lays the eggs into the mesocarp or rind of the fruits in a pouchlike cavity (Figure 2*B*). It was noted that some of the eggs were at the bottom while others were near the top of the cavity. The eggs at the bottom of the cavity obviously would not be exposed to parasitization to the same extent as those located near the aperture of the cavity.

It was found that superparasitism was affected to a greater extent by host egg density than by guava fruit density. The rate of superparasitism fluctuated at about the same level during the periods of fruit scarcity and fruit abundance. It was also found that there was no significant difference in the rate of superparasitism between trees with low fruit abundance and those with high fruit abundance. Although fruit abundance did not seem to affect superparasitism, the data obtained indicated that with an increase in egg density, the rate of superparasitism declined. These observations suggest that *O. oophilus* tends to remain in areas where host eggs are present rather than in areas where host fruits are abundant. These findings are in line with those obtained by Bess, Haramoto, and Hinckley (1963) and by Bess and Haramoto (1961).

This study showed no clear relationship between superparasitism and elevation. The incidence of superparasitism from sea level to 2,000 feet was about the same. However, at elevations above 2,000 feet, the rate of superparasitism declined. Although *C. capitata* is the predominant species at higher elevations (Bess, 1953), the rate of superparasitism cannot be attributed entirely to this factor for it is known that *O. oophilus* super-parasitizes both *C. capitata* and *D. dorsalis* eggs. It appears that the environmental conditions and possibly host fruits rather than the species of host eggs were unfavorable to the activity of *O. oophilus* at the higher elevations.

Although superparasitism of *D. dorsalis* eggs by *O. oophilus* was common under field conditions, the distribution of progeny by the parasite was not at random. When the expected random and the observed distributions were compared, there were highly significant differences indicating that *O. oophilus* discriminated to a certain extent between parasitized and un-parasitized individuals. As shown in Figure 4, the observed distribution fell between the random distribution and the complete discrimination lines.

Furthermore, since approximately 35 percent superparasitism did occur, this ability to discriminate must be considered an imperfect one. A similar conclusion was reached by Salt (1932) in his work with *Collyria*. In his studies with *Trichogramma*, Salt (1936) concluded that although discrimination was perfect, the power of restraint was imperfect. Whether or not *O. oophilus* is imperfect in regard to discrimination or restraint is not known.

The actual mode of discrimination between parasitized and unparasitized hosts by *O. oophilus* is not known. Salt (1937) found that *Trichogramma* discriminated between parasitized and unparasitized host eggs by two faculties, odor and sensory receptors. The odor left by a female parasite upon examining the host was perceived by another female probably by means of the antennae. This second female avoided the same host because of the presence of the odor. If the odor was removed, the second parasite attacked the host as readily as an uncontaminated one. At this point the other faculty came into play. If the parasite inserted the ovipositor into the host already parasitized, it withdrew the ovipositor without laying any eggs into that host. Therefore, a parasite may utilize both the antennae and the ovipositor to discriminate between parasitized and unparasitized hosts. The situation with *O. oophilus* is slightly different from that of *Trichogramma*. Because the host eggs are located in the rind of the fruits, the parasite cannot come in direct contact with the host except by means of the ovipositor. The discrimination between parasitized and unparasitized hosts, therefore, is most likely accomplished by sensory receptors located on the ovipositor. Probably the sensory hairs on the tip of the ovipositor, mentioned by Haramoto (1953), may be involved in discrimination. The possibility of sensory receptors on the ovipositor in discrimination was also mentioned by Salt (1937) in reference to *Collyria* and *Ibalia*. In the case of *O. oophilus* there is still a possibility that a contaminant left by the female parasite at the opening of the ovipositional puncture might prevent superparasitism by other females. However, whether or not such a contaminant is involved is not known.

The effects of superparasitism on the biology of parasites have been discussed by various investigators (Fiske, 1910; Salt, 1937; Chacko, 1964). It was pointed out that when superparasitism occurs, the average size, vigor, longevity, and fecundity of the parasites are reduced. In addition, since the males are intrinsically superior, there is an increasing proportion of males emerging. Being a solitary parasite, *O. oophilus* may lay more than one egg per host egg; however, only one individual survives and emerges as an adult. The exact manner in which supernumerary individuals are destroyed is not known. It is possible that some form of selection occurs within a host egg containing several individuals with the emergence of an intrinsically superior individual. It could be speculated that superparasitism by a parasite like *O. oophilus* is selecting individuals that are intrinsically superior.

The extent of mortality on *D. dorsalis* caused by *O. oophilus* has not

been determined. According to Salt (1936), the egg parasite *Trichogramma* caused a certain amount of mortality of the host when superparasitism occurred. Since in this study superparasitism was concerned with host eggs, it was not possible to obtain data on whether or not there was a higher mortality among host eggs that contained a high number of parasite eggs than those containing a few. It is possible that host eggs containing many parasite eggs might succumb merely by the physical presence of many foreign bodies because they could adversely affect the embryonic develop-ment. Furthermore, the frequent insertion of the ovipositor increases the probability of rupturing vital organs. There is also the possibility that the frequent puncturing of the chorion might increase the entry of micro-organisms.

Parasites may be grouped into three categories on the basis of the mode of egg distribution among the hosts. These three categories are distributed with (1) complete discrimination, (2) partial discrimination, and (3) no discrimination. Parasites in the first category usually possess a perfect sense of discrimination between parasitized and unparasitized hosts and will usually avoid attacking an already parasitized host; those in the second category have an imperfect sense of discrimination and may or may not attack an already parasitized host; and those in the third category do not possess any sense of discrimination between parasitized and unparasitized hosts and will distribute eggs at random. These categories are applicable only at higher levels of parasitization. As indicated by Figure 4, *O. oophilus* belongs in the second category.

This classification of parasites raises the question as to the efficiency of parasites as a biological control agent. Theoretically, since parasites in category 1 distribute eggs very efficiently without "wastage," it would seem that they would be the most effective parasites from the biological control standpoint. However, there seem to be no actual cases when this is true. Actually, those parasites in categories 2 and 3 seem to be the most effective parasites. For example, *O. oophilus* which is in the second category is the most effective parasite of *D. dorsalis* known in spite of the "wastage of eggs." At the present time it is very difficult to draw conclusions on the effectiveness of parasites in the various categories because of the lack of information on the mode of egg distribution of many effective and ineffective parasites. It should also be mentioned that aside from the mode of egg distribution there are other attributes that determine the effectiveness of parasites as biological control agents.

Superparasitism in one sense might be considered as an index of the suitability of the habitat to a parasite. Since by definition superparasitism means a superabundance of reproductive units within a host, it is reasonable to expect a high degree of superparasitism in areas suitable to a particular species of parasite. The variations in the incidence of superparasitism in different fruits which occurred in this study might be looked upon as an

indication of the variations in the suitability of the habitat where the fruits were collected. Furthermore, the present study has shown that host eggs in passion fruits growing on other fruit trees had a higher incidence of super-parasitism than host eggs in passion fruits growing as a monocultural crop on trellises.

SUMMARY

This was a 12-month study on some aspects of superparasitism by *Opius oophilus*, an egg-larval parasite of the tephritid fruit fly, *Dacus dorsalis*. The major portion of the data was obtained from the guava (*Psidium guajava*) areas of Oahu.

The data on egg distribution indicated that *O. oophilus* distributes its eggs in a nonrandom manner. It appears that this parasite is capable of exercising a certain amount of discrimination between parasitized and un-parasitized host eggs. It was found that as the number of eggs deposited by the parasite increased, the percentage of parasitism increased, but not linearly. Since *O. oophilus* discriminates between parasitized and un-parasitized eggs imperfectly, the number of parasite eggs laid was less than that which would occur when the parasite distributes eggs at random. For the same reason the number of parasite eggs laid was higher than that which would occur when the parasite distributes eggs with perfect discrimination between parasitized and unparasitized eggs.

The various factors that influence superparasitism were studied. Studies carried out on Oahu indicated no significant differences in the percentage of superparasitism among the four localities. However, there were seasonal differences. Superparasitism was higher during the winter months than the summer months. Elevation had no effect on superparasitism up to 2,000 feet. Above this elevation the incidence of superparasitism decreased, possibly due to changes in the species of host fruit and climatic factors. Fruit abundance did not affect the rate of superparasitism. Although the egg-lay-ing activity of *D. dorsalis* was greater on trees with abundant fruits, the intensity of superparasitism between high abundance trees and low abun-dance trees was the same. The maturity of guava fruits also had no effect on superparasitism. There were no differences in the intensity of superparasitism among the green, semiripe, and ripe fruits. Superparasitism was found to vary considerably with the species of host fruits in which *D. dorsalis* eggs were laid. The highest superparasitism was found in acerola, *Malpighia punicifolia,* and the lowest, in passion fruit, *Passiflora edulis* f. *flavicarpa,* which was growing on trellises. It was found that there was a decrease in superparasitism with an increase in the host egg density. However, this rela-tionship was nonlinear. In general, there was an increase in percent parasitism with increased superparasitism.

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