<u>APHELINUS ASYCHIS</u> (WALKER), THE IMPORTED PARASITE AND EFFECT OF ITS PARASITISM ON THE FECUNDITY OF THE SORGHUM GREENBUG, <u>SCHIZAPHIS</u> <u>GRAMINUM</u> (RONDANI)

By

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PREFACE

Sorghum has become one of the most important economic crops of the Great Plains during the past few years. All stages of growth of sorghum are infested and damaged by several kinds of insects. The greenbug, <u>Schizaphis graminum</u> (Rondani) is considered to be one of the most injurious insects attacking sorghums in the Great Plains. Chemicals have been the main means of control; however, the recent banning of many insecticides has brought attention to the use of other types of control. Several states (i.e., California, Indiana, etc.) have had success in biological control attempts by more effectively utilizing native and introduced parasites and predators. Thus, research along this lines have been initiated to investigate the possibilities of effectively utilizing parasites and predators in controlling the greenbug. In the present research on greenbug and the effectiveness of parasite action will be studied.

I wish to thank the Entomology Department for the facilities and assistance provided for this study.

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CHAPTER I

INTRODUCTION

Sorghum was seriously damaged by the greenbug, <u>Schizaphis</u> <u>graminum</u> (Rondani), over extensive areas of the United States for the first time during 1968. Outbreaks were reported in Oklahoma, Texas, Arizona, Kansas, Colorado, Nebraska and South Dakota (USDA 1968). The Cooperative Economic Insect Report, (USDA 1969) reported that over 7¹/₁ million acres of sorghum were infested by greenbugs and the estimated loss to production was up to 68 million dollars. All stages of growth were infested, but the most serious damage resulted from infestation on small plants. Populations as high as 40,000 greenbugs per plant were estimated in these areas during 1968. Outbreaks were also reported from these states in 1969, but the infestation and damage were not as severe as they were in 1968. The above facts suggest that greenbugs could be a major pest to sorghum and could reduce incomes and profits to farmers in the future.

Due to the severe outbreak of greenbugs on sorghum in 1968, Harvey and Hackerott (1969b) reported that in Kansas, an estimated 1,056,515 acres of sorghum were infested at damaging levels, 227,313 acres of sorghum were destroyed, and 595,555 acres of sorghum were treated with insecticides for greenbug control. These treatments by using some kinds of insecticide could contaminate the environment in infested areas. This problem has caused many entomologists to seek other means

of improving control methods, especially the use of parasites and predators as a means of biological control. Therefore, several native and imported parasites have been studied to evaluate the use of biological control as an agent in controlling this important pest. Raney et. al. (1971) and Langston (1970) have investigated laboratory studies on the fecundity, longevity, developmental period, sex ratio, and host preference of an imported parasite <u>Aphelinus asychis</u> (Walker) and a native parasite <u>Aphelinus nigritus</u> (Howard). Hight et. al. (1972) conducted laboratory studies on the effect of parasitism by a native parasite, <u>Lysiphlebus testaceipes</u> (Cresson), on the fecundity of the greenbug. The objective of this study was to determine the developmental period and sex ratio of <u>A. asychis</u> while using the biotype C greenbug as a host, and to determine the fecundity and reproductive life of the parasitized sorghum greenbug.

CHAPTER II

LITERATURE REVIEW

The greenbug, <u>Schizaphis graminum</u> (Rondani) is one of the most destructive pests of small grains in the central and southwestern states of the United States (Hight et. al. 1972). The first record of its appearance is dated 1847 and Rondani described it in Italy in 1852. The greenbug was first noted in the United States in Virginia in 1882 (Webster and Phillips 1912).

Sorghum was first recorded as a greenbug host by Passerini in Italy in 1863 (Hunter 1909, Harvey and Hackerott 1969b). Although the greenbug was not considered to be an important pest of sorghum prior to 1968, Hays (1922) reported the greenbug damaged sorghum in 1916.

Oklahoma recorded its first outbreak on small grains in 1901. Damaging infestations have occurred in 1907, 1916, 1939 and 1942 when more than 61 million bushels of grain were lost (Daniels et. al. 1956). Daniel and Jackson (1968) reported that greenbugs were found on sorghum in Texas in 1967.

Severe greenbug outbreaks occurred for the first time on sorghums in Oklahoma in the summer of 1968 and throughout a major portion of the sorghum producing region of the United States (Wood et. al. 1969, Harvey and Hackerott 1969a). The greenbugs attacking sorghum were considered to be a different biotype from those collected on wheat and were designated as biotype C (Harvey and Hackerott 1969a,b). The

biotype C greenbug is taxonomically similar to the biotype A and B greenbugs which are collected from wheat. However, several differences are listed: the biotype C greenbugs are yellowish green; they have little or no black on cornicles; tips are not expanded, and wrinkles are present throughout their entire length. The biotype A and B greenbugs are dark green; they have distinctly black on the distal one-third of the cornicles, tips are expanded and wrinkles are present on basal portions only (Daniels and Jackson 1968, Harvey and Hackerott 1969a, b, and Wood and Chada 1969). There are also ecological differences. The greenbugs on sorghum develop and reproduce under conditions in which the temperature is as high as 110° F in the field. Greenbugs on small grains develop and reproduce at temperatures ranging from 68° F to 75° F, but usually leave the host when temperatures reach 85° F to 90° F (Wood and Chada 1969, Harvey and Hackerott 1969b).

The parasite <u>Aphelinus asychis</u> (Walker) is classified in the subfamily Aphelininae, family Eulophidae. Members of this subfamily are parasites of aphids, scales and whiteflies. Apparently all species of the genus <u>Aphelinus</u> are internal parasites of aphids (Schlinger and Hall 1959). This parasite was introduced from Iran into the United States by the Insect Identification and Parasite Introduction Research Branch, USDA (Raney et. al. 1971). <u>Aphelinus asychis</u> is similar to <u>A</u>. <u>semiflavus</u> (Howard) which Hartley (1922) described and reported on the biology of this species.

Recently, Ferriere (1965) synonymized <u>A. asychis with <u>A. semiflavus</u> and Force and Messenger (1968) concurred. <u>A. asychis</u> and <u>A. semiflavus</u> are very similar in morphology and in color and may well be conspecific. However, they seem to differ in their host ranges. Mackauer and</u>

Finlayson (1967) left <u>A</u>. <u>asychis</u> as a separate species because of possible differing geographic races or strains and also stated that <u>A. asychis</u> preferred hosts of the families Callaphididae and Aphididae but did not attack any Phylloxeridae or Pemphigidae.

Jackson and Eikenbary (1971) found that A. asychis shows a definite preference for the biotype C greenbug over the corn leaf aphid, Rhopalosiphum maidis (Fitch); oat-bird cherry aphid R. padi (L); and yellow sugar cane aphid, Sipha flava (Forbes). This strain attacks both apterous and alate greenbugs. van den Bosch et. al. (1957, 1959) reported that A. semiflavus seemed to show varying climatic adaptation and was encountered in fair abundance in Southern California. This species was successfully mass produced in the insectaries at Albany and Riverside, California and released in large numbers at many localities throughout the state. Manglitz and Schalk (1970), reported A. semiflavus was the most abundant species of parasites reared from both Therisaphis richni and T. maculata (Buckton) in collections made in Eastern Nebraska (Lancaster, Saunders, and Burt counties) in the fall of 1967. Force and Messenger (1968) evaluated the field potential of A. asychis by the use of laboratory studies. A rather complete biology of A. semiflavus has been presented by Hartley (1922), who used Myzus persicae (Sulz.) as its host. Schlinger and Hall (1959), added some information on the form of A. semiflavus that was introduced from the Middle East (mainly Israel). On the basis of the biological evidence, there is reasonable doubt whether this Old-World Strain is conspecific with the American form. A. asychis may have more than 20 generations a year, and does not enter diapause either in summer or winter; it usually manages to remain reproductively active in early and late

winter; it has the special ability to discriminate between parasitized and unparasitized hosts and will rarely deposit eggs in hosts that are already parasitized either by its own species or other species (Force and Messenger 1964, 1965). However, further studies on <u>A</u>. <u>asychis</u> and its parasitization of the greenbug are still required.

CHAPTER III

MATERIALS AND METHODS

In determining the fecundity of the parasitized greenbug, two constant temperatures (21.1°C. and 26.7°C) and one constant alternating temperature (21.1-26.7⁰C.) were used in this study. After Raney et. al. (1971) found that at the 35°C. constant and 26.7-37.8°C. alternating temperatures yielded a few mummies (parasitized aphids), but no adults emerged. The constant alternating temperature was maintained under 12 hour low temperature and 12 hour high. All temperatures were maintained in controlled environmental chambers with a 12 hour photoperiod yielding approximately 1110 ft. candles per chamber. To provide illumination, each chamber contained four 25 watt incandescent lamps together with Sylvania's lifeline FR40CW - 235 florescent lamps. The greenbugs used in this study are of the 'C' biotype and obtained from the laboratory culture. These aphids were maintained and reared on RS-610 sorghum plants growing in 4-inch pots in the laboratory. The test sorghum plants were covered with ventillated cellulose nitrate cages (Raney et. al. 1971). The soil surface under each plant was covered with fine white sand in order to facilitate recovery of fallen mummies and to check on the small parasites. The imported parasite, Aphelinus asychis, was used as the test parasite in the study. This parasite was maintained and reared on greenbug colonies in the laboratory.

To obtain greenbugs at the desired age, twenty adult greenbugs were

placed on sorghum plants in each pot for 12 hours. Figure 2 shows a population of greenbugs 12 hours after the adults were placed on the plants. The progeny from these adult greenbugs were then sized and all but 10 aphids were removed per pot in order to get uniformity.

At 26.7°C. and 21.1-26.7°C., five age groups of aphids were used as follows: (1) one day old (24-30 hours); (2) two days old (48-54 hours); (3) three days old (72-78 hours); (4) four days (96-102 hours); and (5) five days old (120-126 hours). At 21.1° C., greenbugs required seven days to reach the adult stage, therefore, seven age groups of aphids were used. The ages of aphids used in the 21.1° C. test were the same as for the 26.7°C. and 21.1-26.7°C. tests, except the periods of (6) six days old (144-150 hours), and (7) seven days old (168-174 hours) were added to obtain adult aphids. This method of obtaining aphids of a known age was similar to that used by Fox et. al. (1967) and Hight et. al. (1972).

Three pairs of 1- to 3-day old parasites, <u>A</u>. <u>asychis</u>, were introduced into the cage just prior to or at the beginning of 12 hours of light. Then the parasites were removed after 12 hours of parasitization. A mouth aspirator, as described by Childs (1932) and Raney et. al. (1971), was used to transfer the parasites into and out of the cage through a small hole in the side of the cage. The aphids in each pot were observed daily until they developed into mummies (parasitized greenbugs). The greenbug was considered to be a mummy when the exoskeleton had changed into black or bluish black color.

The fecundity of parasitized greenbugs was measured by the daily number of progeny produced by each of the parasitized greenbugs. Progeny were counted and removed daily until parturition ceased. All mummies

were left on the plants to develop to adult parasites. The emerging parasites were collected, sexed and recorded. Figures 3 and 4 show the profile of male and female parasites while Figures 5 and 6 show the female in the act of parasitization. Data concerning fecundity of parasitized aphids, developmental period, and sex ratio of parasites were collected and recorded at the same time. Five replications consisting of 50 greenbug nymphs or adults of the desired age were placed on caged RS-610 sorghum plants growing in 4-inch pots in the controlled environmental chamber for each experiment. Figure 1 shows test pots in the controlled environmental chamber.

CHAPTER IV

RESULTS AND DISCUSSION

Oviposition by <u>Aphelinus</u> spp. can occur during the night or day. Typically, the female, after sensing the presence of the host, turns quickly, folding the apices of her wings anteriorly, and with the ovipositor extended inserts it either into the venter, side, or dorsum of the aphid by lunging directly backward. The ovipositor may remain inserted for as long as 15 minutes (Hartley 1922, Hagen and van den Bosch 1968). Hartley (1922) stated that, <u>Aphelinus semiflavus</u> (Howard) showed a distinct preference for lst- and 2nd-instar aphid nymphs. Adult aphids were rarely attacked. If a second instar numph was attacked, the aphid seldom matured and did not produce young, while aphids parasitized in the third instar may mature and produce several young before dying, and if later stages were parasitized, many young aphids are produced.

Table I shows the effect of age and temperature on the reproductive life of the parasitized sorghum greenbug. The reproductive life of parasitized greenbugs is indicated when the first aphid at a certain age began reproducing progeny. Aphids parasitized at age 4-to-5 days at 21.1°C. and at age 3-to-5 days at 26.7°C. and 21.1-26.7°C. produced a near normal number of offspring (4 to 5 aphids/day) until the 2nd to 3rd day at 21.1°C. and the 4th to 5th day at 26.7°C. and 21.1-26.7°C. after parasitization, then the reproduction declined to zero; while

aphids parasitized at age 6-to-7 days at 21.1° C. produced many more offsprings than normal. The minimum and maximum reproductive life of parasitized greenbugs were 3 days and 5 days, respectively at 21.1° C. and 26.7° C. Aphids attacked at age 1-to-3 days at 21.1° C. and at age 1-to-2 days at 26.7° C. and $21.1-26.7^{\circ}$ C. did not live to produce offspring. The average number of progeny produced by parasitized aphids at each age of the aphid and each temperature is shown in Figure 7. Hight (1971) found that, the average progeny of parasitized aphids increased with increasing host age for aphids parasitized that were 4 days or older at tested temperatures. This agrees with number of progeny produced by parasitized aphids over all temperatures in this study.

The parasitization decreases with increasing aphid age at all temperatures (Table I). The basis for low parasitization of older aphids might be due to several factors. The older greenbugs are larger and may have defense mechanisms to protect themselves from parasitization. Wilbert (1964) describes three types of defense mechanisms exhibited by various aphids against <u>A</u>. <u>asychis</u>: (a) mechanical, by moving legs and antennae before parasite attacks; (b) intensified movement of the legs, antennae, or whole body at the time of parasite ovipositor insertion; and (c) physiological reaction where eggs or larvae are destroyed by encapsulation.

Table II gives a theoretical model of a greenbug population originating from one greenbug under laboratory conditions.

Figure 9 shows a theoretical average reduction of greenbug population due to parasitization by <u>A</u>. <u>asychis</u>. If one adult greenbug reproduces an average of 5 offspring per day for 25 days and the reduction resulting from parasitism has similar figures, then the average percent

reduction may be calculated. Population reduction would be 100% for all parasitized greenbugs from the 1- and 2-day old aphid numphs while a reduction of 86% of better would be possible for all others except those parasitized at the age of 5 days old at 21.1°C., where a 56% reduction was obtained.

Table III contains compilation of data gathered from the study of development of A. asychis at different host ages and various temperatures. The egg-mummy category indicates the average period from parasitization to appearance of the mummy. The mummy-adult column indicates the average period for pupal development, and the eggadult category indicates the average period from oviposition to emergence of the adult parasite. Variation of egg-adult period depend upon host age and temperature. The laboratory studies have shown that the development time for A. asychis was decreased with higher temperatures. The parasite took about 16 days to develop at 21.1°C. and 13 days at 26.7°C. in all ages of greenbug hosts. At the alternating temperature, 21.1-26.7°C., the parasite required about 14 days to develop from egg to adult. Raney et. al. (1971) reported that the duration of development time for A. asychis was about 16 days at 23.9° C. and 13 days at 26.7°C. Hagen and van den Bosch (1968) stated that, the developmental time for this species was 11.3 days at 21.1°C., and 10.4 days at 29.4°C. The respective time revealed by the both studies are much less than that was resulted in this study. However, Force and Messenger (1964) indicated the developmental periods of A. semiflavus and A. asychis are quite similar. It took about 18 days for the first one to develop at 21.1°C. and 11 days at 26.7°C.

The age of the aphid when parasitized did not have any effect on

the emergence of the parasite. The percentage of emerging parasites was high at all ages of aphids and temperatures. The maximum and minimum percentages were 100% and 86%, respectively. The size of parasites from all ages of aphid host was almost the same except those emerging from 1-day old aphids were slightly smaller than others.

Figure 9 illustrates the number of males and females emerging from each age of aphid and temperature. The temperatures have great influence on male and female production of <u>A</u>. <u>asychis</u>. Male production, from this study, was increased with higher temperatures. On the contrary, more females were produced at lower temperatures. For example, the sex ratio was approximately 1:2 when parasitized aphids were reared at 21.1° C., but it was changed to 1.5:2 at 26.7° C. This meant that <u>A</u>. <u>asychis</u> produced over 50% females at all conditions tested for all ages of aphid hosts and temperatures. Schlinger and Hall (1959) found that temperature influenced the sex ratio of this species. Hartley (1922), with a similar result, reported that <u>A</u>. <u>semiflavus</u> reared on <u>Myzus</u> <u>persicae</u> produced a few males.

Raney et. al. (1971) stated that the ratio of males to females was 1:2.4 at 23.9° C. and 1:1 at 32.2° C. But Force and Messenger (1964) reported that most of emerging <u>A</u>. <u>semiflavus</u> at 32.2° C. were males.

Clausen (1939) stated that one of the important factors influencing sex ratio is the size of the host in which the parasite develops. Large hosts favor female parasite development. As supported by the Clausen's work, the parasites emerging from older ages of aphid hosts were mainly females in this study.

A. asychis reduced the number of greenbugs occurring on the sorghum

plant in two ways. The 1- and 2-day old parasitized greenbugs died and produced no offspring at all temperatures tested, while 4-day old and older parasitized greenbugs at 21.1°C produced an average of 1.8, 2.6, 8.8 and 11.5 offsprings per greenbug, respectively.

In this study <u>A</u>. <u>asychis</u> appears to be a good parasite of greenbug in that: (1) it attacks all ages, (2) reduces the fecundity of the greenbug, (3) adult emergence is good, and (4) adult developing from all ages are healthy. This parasite may be one of the important factors in controlling of the greenbug in the future.

CHAPTER V

SUMMARY

Laboratory studies of <u>A</u>. <u>asychis</u> were conducted on the effect of parasitism on fecundity and reproductive life of sorghum greenbug, parasite developmental period, and sex ratio at 3 controlled temperatures. Greenbugs parasitized at the age of 1 day old and 2 days old died and produced no offspring. Greenbugs parasitized at the age of 3 days old, 4 days old, and 5 days old at 21.1-26.7°C. reproduced an average of 1.7, 3.8 and 5.9 offsprings per greenbug, respectively. The reproductive life of parasitized greenbug varied from 0 to 5 days versus about 25 to 30 days in nonparasitized greenbug.

The parasite's developmental period was approximately 16 days at 21.1° C. and 13 days at 26.7°C. The greatest amount of female production was obtained at the lower temperature tested. On the contrary, more males were produced at the higher temperature tested. The ratio of males to females was 1:2 at 21.1° C. and 1.5:2 at 26.7° C.

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APPENDIX

TABLES AND FIGURES

TABLE I

19 13 - 1917 - 2017 - 2017 - 2017 - 2017 - 2017		CTIVE LIFE O SITIZED SORG GREENBUG			
Temperature	Age of	No. of	Progenyof	Reproduct (in d	
	parasitized aphids (days) ^{<u>a</u>/}	parasitized aphids	parasitized aphids	Start	End
	1	32	0	-	-
	2 3	33	0	-	-
0		38	0	-	
21.1°C	4	23	41	8	10
	5	19	50	8	10
	6	13	115	7	10
	7	14	161	7	10
	1	40	0	-	-
	2	38	0	-	-
26.7 ⁰ C	3	34	44	6	9
	4	28)	89	6	10
	- 5	25	105	5	10
	1	32	0	-	-
	2	36	0	-	-
21.1-26.7°C	3	30	51	6	9
	4	21	79	6	10
	5	19	112	6	10

THE EFFECT OF AGE AND TEMPERATURE ON THE REPRODUCTIVE LIFE OF THE

<u>a</u>/ Five replicates, 10 aphids/replicate per age class

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TABLE II

A THEORETICAL MODEL OF AN APHID POPULATION ORIGINATING FROM ONE GREENBUG UNDER THREE CONTROLLED TEMPERATURESa/ IN THE LABORATORY

					Tatastu	Izeu by	A. asycuis			
Days after 1st nymph	Non- parasitized	<u></u>	3rd instar	•		4th insta	r		5th insta	
born	21.1-26.7°	21.10	26.7 ⁰ 2	21.1-26.7°	21.1°	26.7°	21.1-26.7 ⁰	21.1 ⁰	26.7 ⁰	21.1-26.7 ⁰
• 5	26	2.	6 1.3	1.7	8.	8 3.3	2 3.8	.11.	5 4.2	2 5.9
10	426	19	9	12	64	23	28	103	30	43
15	5,826	112	56	73	379	138	164	740	181	254
20	74,351	654	327	428	2,214	805	956	5,214	1,056	1,484
25	931,001	3,841	1,921	2,511	13,002	4,278	5,614	36,809	6,206	8,717

Parasitized by A. asychis

a/ Temperature expressed in Centigrade.

	TAB	LE	Τ	Ι	Ι
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	AND SEX	RATIO	OF APHE	LINUS ASYCHIS			
Temperature	Age of parasitized aphid(days) <u>a</u> /	mummy		Egg- adult (days) en	% norgence	% ma le	% female
	1	8.5	7.1	15.6 <u>+</u> 0.80	100	37.5	62.5
	2	8.7	7.5	16.2 <u>+</u> 1.25	94	39.4	54.6
	3	9.1	7.2	16.3 <u>+</u> 0.61	92	36.8	55.2
21.1 ⁰ C	4	8.6	7.3	15.9 <u>+</u> 0.83	91	39.0	52.0
	5	9.1	7.4	16.5 <u>+</u> 0.84	100	42.1	57.9
	6	8.9	7.6	16.5 <u>+</u> 0.52	100	30.8	69.2
	7	9.2	7.5	16.7 <u>+</u> 1.03	100	21.4	78.6
	Average	8.9	7.4	16.3 <u>+</u> 0.84	97	35.3	61.7
	1	5.9	6.6	12.5 ± 0.81	93	35.2	57.8
	2	5.6	6.8	12.4 ± 0.77	95	39.6	55.4
26.7 ⁰ C	.3	6.1	6.7	12.8 ± 1.20	91	35.2	55.8
	.4	5.8	6.8	12.6 <u>+</u> 1.14	89	39.2	49.8
	5	6.0	6.9	12.9 <u>+</u> 0.82	96	44.0	52.0
	Average	5.8	6.8	12.6 <u>+</u> 0.95	93	38.6	54.4
	1	6.7	6.9	13.6 <u>+</u> 1.10	91	37.7	53.3
	2	7.0	7.1	14.1 <u>+</u> 1.05	97	30.5	66.5
21.1-26.7 ⁰ C	.3	7.3	6.8	14.1 <u>+</u> 0.77	93	33.2	59.8
	4	7.4	6.6	14.0 <u>+</u> 1.01	86	23.9	62.1
	5	7.2	7.0	14.2 <u>+</u> 0.72	90	26.5	63.5
	Average	71	6.9	14.0 <u>+</u> 0.93	91	30.3	60.7

THE EFFECT OF TEMPERATURE AND AGE OF THE PARASITIZED SORGHUM GREENBUG ON THE DEVELOPMENT, EMERGENCE, AND SEX RATIO OF APHELINUS ASYCHIS

<u>a</u>/ Five replicates, 10 aphids/replicate per age class.

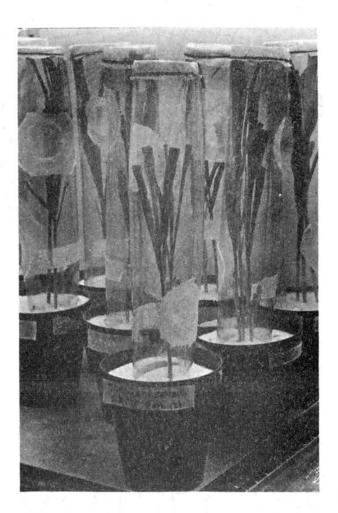


Figure 1. Caged Sorghum Plants Growing in 4-Inch Pots in the Controlled Environmental Chamber

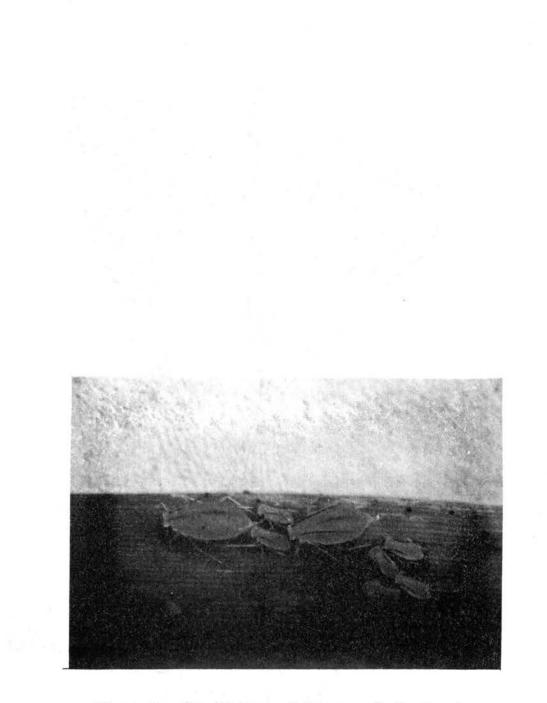


Figure 2. The Adults and Progeny of the Sorghum Greenbug

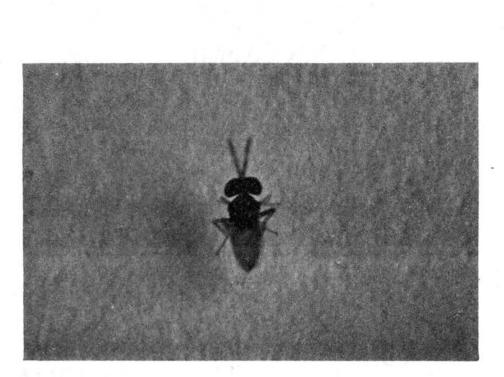


Figure 3. The Profile of a Male Aphelinus asychis



Figure 4. The Profile of a Female Aphelinus asychis

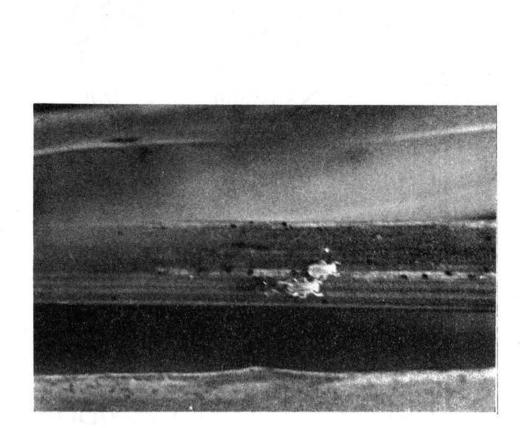
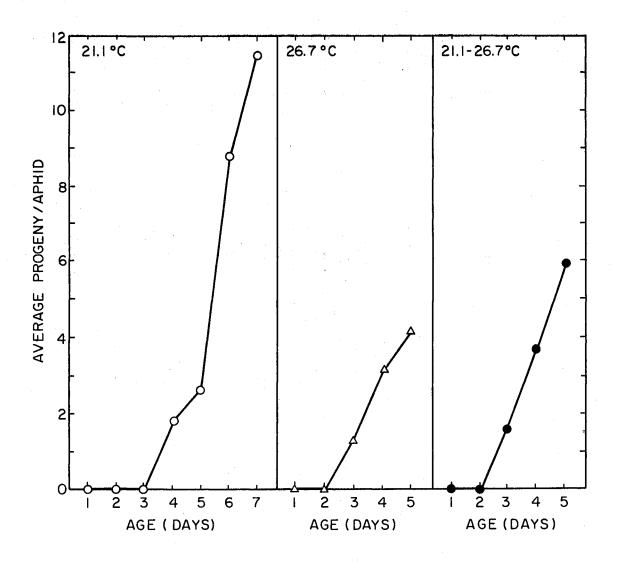
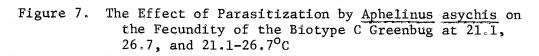


Figure 5. <u>Aphelinus asychis</u> Attacking a Two-Day Old Greenbug



Figure 6. <u>Aphelinus asychis Attacking</u> an Adult Greenbug





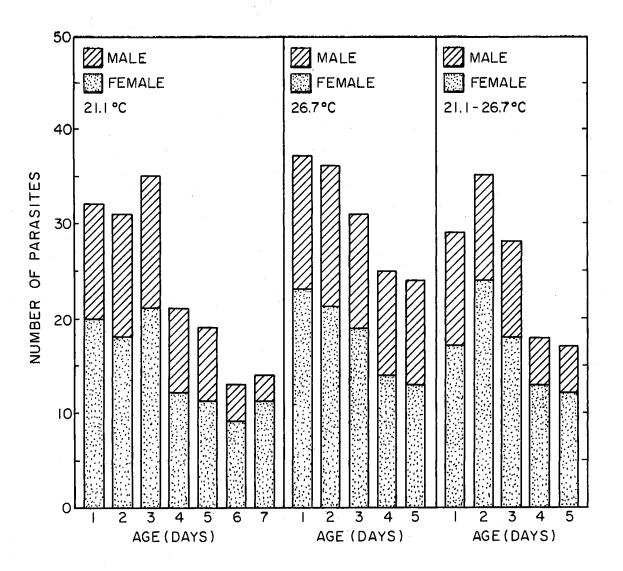
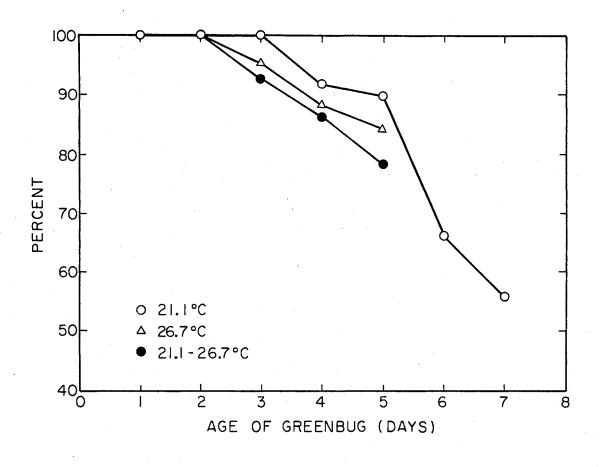
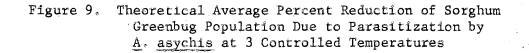


Figure 8. The Number of Male and Female <u>A</u>. <u>asychis</u> Emerging From Parasitized Biotype C Greenbugs at 3 Controlled Temperatures





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Master of Science

Thesis: <u>APHELINUS ASYCHIS</u> (WALKER), THE IMPORTED PARASITE AND EFFECT OF ITS PARASITISM ON THE FECUNDITY OF THE SORGHUM GREENBUG, SCHIZAPHIS GRAMINUM (RONDANI)

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