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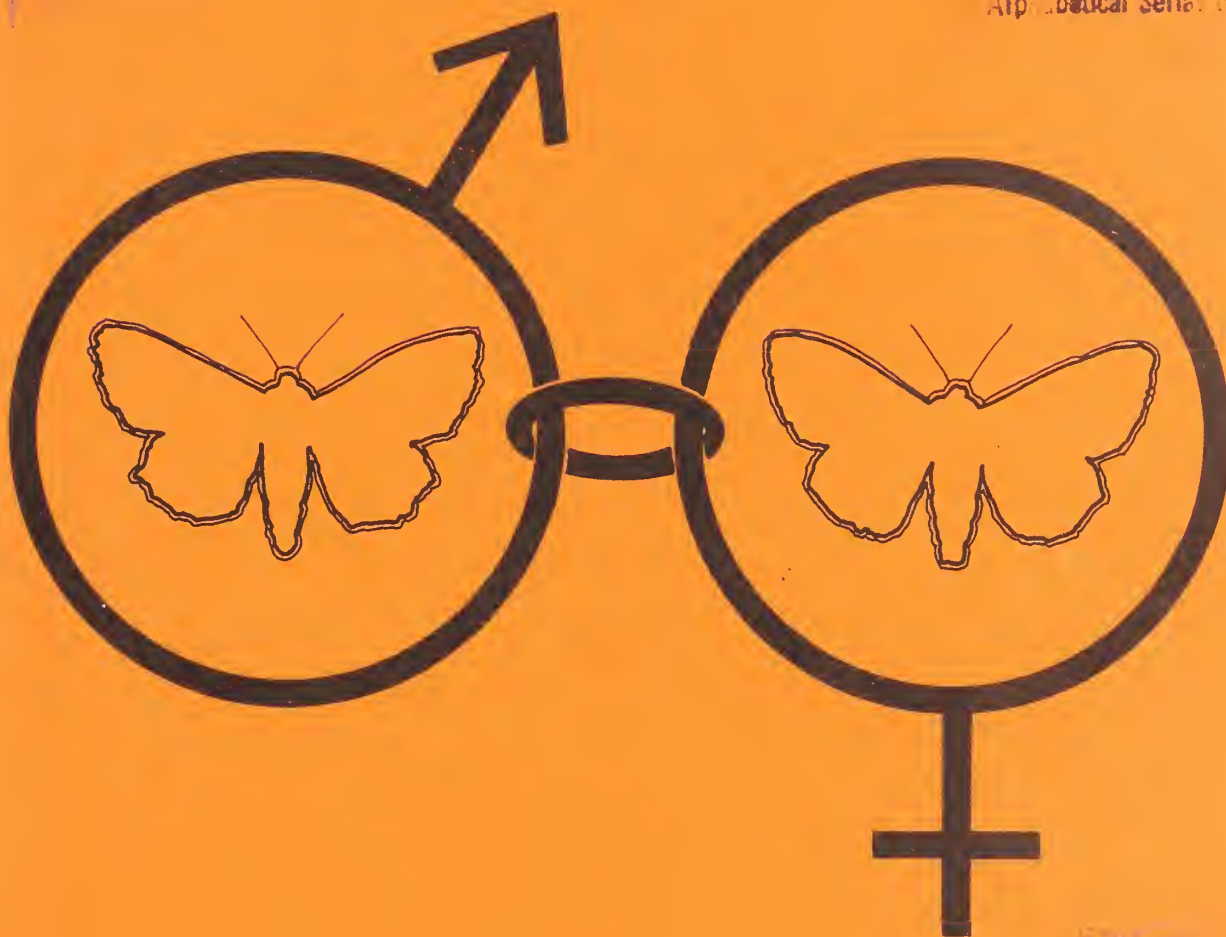
Gainesville, Florida

FIRST SEMI-ANNUAL REPORT - 1975

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FIRST SEMI-ANNUAL REPORT

1975

Insect Attractants, Behavior, and Basic Biology

Research Laboratory

USDA-ARS, Southern Region, Florida-Antilles Area

P.O. Box 14565

Gainesville, Florida 32604

In Cooperation with the Departments of

Entomology and Nematology

and

Agricultural Engineering

University of Florida

This progress report includes tentative results of research not sufficiently complete to justify general release. Such findings when adequately confirmed, will be released through established channels. Therefore, this report is not intended for publication and should not be referred to in literature citations. Individual reports herein may refer by code numbers to previous reports. Copies of these previous reports may be requested.



Growth and Development

- Effect of juvenile hormone and its mimics on mitochondrial metabolism in the Indian meal moth, Plodia interpunctella Part I: Mechanisms of action. D. L. Silhacek and K. Kohl 1
- Effect of juvenile hormone and its mimics on mitochondrial metabolism in the Indian meal moth, Plodia interpunctella Part II: Metabolic effects. D. L. Silhacek and D. Firstenberg 2
- Juvenile hormone binding in the subcellular components of the insect. S. M. Ferkovich and P. R. Putter 3
- Effects of juvenile hormone on the metamorphosis of malathion-resistant Indian meal moths. D. L. Silhacek, J. Zettler and H. Oberlander 4
- Interaction of juvenile hormone with proteins in tissue culture. H. Oberlander and S. M. Ferkovich 5
- Hormonal control of chitin synthesis in vitro. H. Oberlander and C. E. Leach 6
- Dissociation and reaggregation of fat body cells during metamorphosis. H. Oberlander and C. E. Leach 7
- Control of egg maturation in Dermestid beetles. K. Vick, J. Coffelt and D. D. Tobin 8
- Reproductive biology of the cigarette beetle, Lasioderma serricornis (Fabr.). J. A. Coffelt and W. T. McClellan 9
- Bacteremia as a deterrent to laboratory rearing of Biosteres (Opisus) longicaudatus: Diagnosis, control and evaluation of control measures. P. D. Greany, G. Allen, J. C. Webb, J. L. Sharp, and D. L. Chambers 10
- Histological, bacteriological, and physiological studies on the invasion of host tissues by bacterial pathogens of the Caribbean fruit fly. G. E. Allen, P. D. Greany, T. C. Carlisle, and M. Chance 11



Introduction of <u>Opius oophilus</u> into Florida for control of the Caribbean fruit fly. P. D. Greany, R. M. Baranowski, D. L. Chambers, T. Wong, and W. J. Schroeder	12
Effect of temperature and moisture concentration of the pupation medium on the successful development of <u>Biosteres (Opius) longicaudatus</u> and <u>Anastrepha suspensa</u> . T. R. Ashley, P. D. Greany, and D. L. Chambers	13
Host selection studies on <u>Biosteres (Opius) longicaudatus</u> in relation to attack of the Caribbean fruit fly. P. O. Lawrence and P. D. Greany	14
Emigration: The major factor in regulation of growing <u>Tribolium castaneum</u> populations. David W. Hagstrum, Edward E. Gilbert, and Burrell J. Smittle	15
CO ₂ output during maturation of colonized cabbage looper larvae. H. C. Leppla and W. K. Turner	16
<u>Behavior: Chemical Cues</u>	
Micro techniques and analytical methods. R. R. Heath, J. H. Tumlinson, and R. E. Doolittle	17
Sex pheromone studies of the navel orangeworm. J. A. Coffelt, K. W. Vick and L. L. Sower	18
Borer pheromones (family Sesiidae). J. H. Tumlinson, R. E. Doolittle, J. W. Devore and A. T. Proveaux	19
A potent sex attractant for the carpenterworm moth, <u>Prionoxystus robiniae</u> . R. E. Doolittle, J. D. Solomon, W. L. Roelofs, M. Beroza, M. McKnight, and A. Tagestad	20
Isolation and identification of the sex pheromone of the white peach tree scale. R. R. Heath, J. R. McLaughlin, J. H. Tumlinson, T. R. Ashley, and D. L. Chambers	21
Exploitation of the sex pheromone behavior of Sesiidae: Disruption of mating communication. J. R. McLaughlin, R. E. Doolittle, E. R. Mitchell, R. Gentry, J. H. Tumlinson and C. R. Yonce	22



	<u>Report Number</u>
Control of stored-product insects with sex pheromones. L. L. Sower, K. W. Vick, J. A. Coffelt and P. Whitmer	23
Resistance to atmospheric permeation as a means for mating control of Lepidoptera. J. P. McLaughlin and L. L. Sower	24
Use of sex pheromones for behavioral control of loopers and related noctuid species. J. P. McLaughlin, E. R. Mitchell and D. L. Chambers	25
Disruption of mating in <u>Heliothis zea</u> and <u>H. virescens</u> . E. R. Mitchell, A. H. Baumhover, and M. Jacobson	26
Field evaluation of sex pheromones of the fall armyworm, beet armyworm and southern armyworm. E. R. Mitchell and R. E. Doolittle	27
Seasonal abundance of lesser peachtree borer and peach-tree borers in north central Florida. J. L. Sharp, J. P. McLaughlin and J. H. Tumlinson	28
Sticky traps baited with <u>E,Z</u> or <u>Z,Z</u> isomers of 3-13-octadecadien-1-ol acetate catch several different sesiid moths in north central Florida. J. L. Sharp and J. P. McLaughlin	29
Air puff sensor for insect electrophysiological studies. E. W. Hamilton	30
Attracting fall armyworms with controlled release plastic strips containing <u>Z</u> -9-DDA. F. C. Tingle and E. R. Mitchell	31
Laboratory-reared fall armyworm females vs. wild females as bait in sticky traps. F. C. Tingle and E. R. Mitchell	32
Waserlike frequency from an ethyl alcohol-acetone mixture. P. S. Callahan	33

Behavior: Physical Cues

Response of fall armyworm moths to 337 μ m laser radiation. W. K. Turner, P. S. Callahan and F. L. Lee	34
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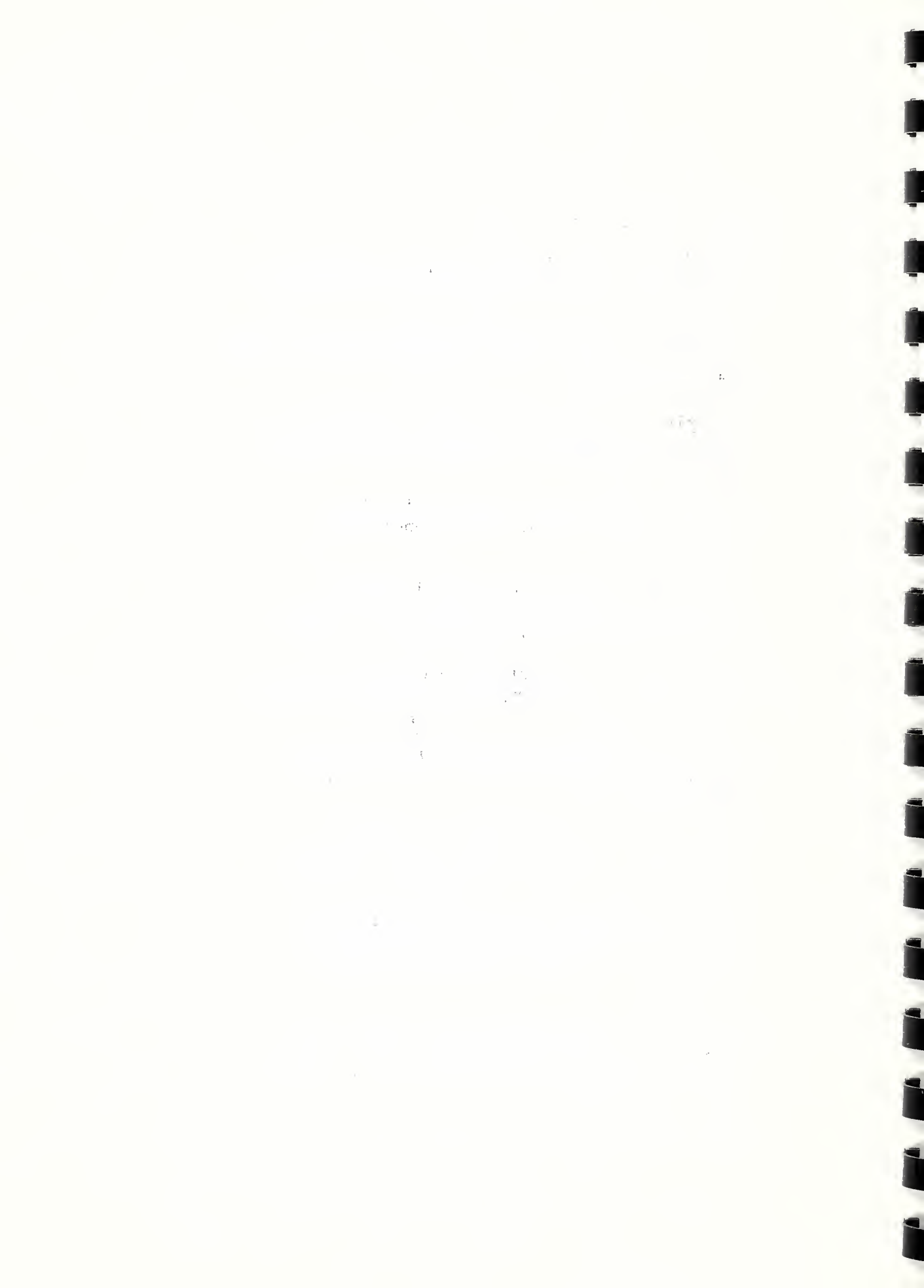


	<u>Report Number</u>
Sound production by <u>Conotrachelus nenuphar</u> : Associated behavior. C. O. Calkins, J. C. Webb, H. R. Agee and T. C. Carlisle	35
Sound production by <u>Conotrachelus nenuphar</u> : II. Analysis and identification. J. C. Webb, C. O. Calkins and T. C. Carlisle	36
Analysis of screwworm fly flight sounds. J. C. Webb, J. L. Goodenough and J. C. Penner	37
Sounds and electrical charges produced by electric- grid and black light traps that repel and/or attract insects. J. M. Stanley, H. R. Agee and J. C. Webb	38
Attraction of fall and beet armyworm to electro- magnetic energy. J. M. Stanley, E. R. Mitchell and H. R. Agee	39
Sensory physiology of the compound eye of <u>Biosteres (Opus) longicaudatus</u> and <u>O. oophilus</u> . H. R. Agee, P. D. Greany, M. L. Parks and D. L. Chambers	40
Field responses of the Caribbean fruit fly and it primary parasites to colored surfaces. P. D. Greany, H. R. Agee, D. L. Chambers and A. K. Burditt	41
Use of the electroretinogram to measure the quality of fruit fly vision. H. R. Agee and M. L. Park	42
Morphology of the antenna of <u>Trichoplusia ni</u> . M. S. Mayer and T. C. Carlisle	43
Histology of the antenna of <u>Trichoplusia ni</u> . T. C. Carlisle and M. S. Mayer	44
<u>Behavior: Mechanisms of Perception</u>	
Correlations of the EAG of <u>Trichoplusia ni</u> to enzymic pheromone hydrolysis. M. S. Mayer	45



Behavior: Organismal Dynamics

Comparative life histories of "wild type" and "colonized" Caribbean fruit flies. N. C. Leppla, M. D. Huettel, D. L. Chambers, and W. K. Turner	46
Comparison of "wild type" and colonized populations of cabbage loopers reared in field cages. R. H. Guy and N. C. Leppla	47
Comparison of the daily activity of wild and laboratory cabbage looper moths. W. K. Turner and N. C. Leppla	48
Circadian rhythms of locomotion in adult Caribbean fruit flies. N. C. Leppla and W. K. Turner	49
Comparison of almond moth activity under continuous weak light and continuous darkness. W. K. Turner and D. W. Hagstrum	50
Flight ability of <u>Anastrepha suspensa</u> in the laboratory. J. L. Sharp, J. C. Webb, H. P. Agee and B. J. Smittle	51
Flight metabolism of the Caribbean fruit fly, <u>Anastrepha suspensa</u> (Loew). K. W. Vick and J. L. Sharp	52
Activity of stable fly adults at reduced temperatures. W. K. Turner and B. J. Smittle	53
Effect of pre-release acclimation on response of male and female <u>Pseudoplusia includens</u> to BL-traps baited with (Z)-7-dodecen-1-ol acetate. J. L. Sharp, E. R. Mitchell and J. James	54
Optimization of parasitoid-host densities for <u>Biosteres (Opus) longicaudatus</u> and its host <u>Anastrepha suspensa</u> . T. R. Ashley, P. D. Greany and D. L. Chambers	55
Simple devices for transferring moths. W. K. Turner	56



Population Dynamics

Hibernation of the plum curculio, <u>Conotrachelus</u> <u>nenuphar</u> (Herbst). C. O. Calkins and W. McCoy	57
Reciprocal negative interactions in competitive populations of cabbage and soybean loopers. H. C. Leppla, W. K. Turner, E. P. Mitchell, W. J. Pons and F. L. Lee	58
Trapping equipment for the almond moth, <u>Cadra cautella</u> : Evaluation of trap design. J. M. Starley and D. W. Hagstrum	59
Solid state photoswitch and photoswitch timer for insect survey units. F. W. Hamilton	60
Collection of beet armyworm and southern armyworm larvae from wild hosts. F. C. Tingle, E. R. Mitchell and T. R. Ashley	61
Inheritance of alleles at four allozyme loci in the plum curculio. W. D. Huettel and C. O. Calkins	62
Sperm precedence in the plum curculio, <u>Conotrachelus nenuphar</u> (Herbst). W. Huettel, C. Calkins and A. Hill	63
Effect of killing agents on capture of <u>Heliothis</u> <u>virescens</u> , <u>H. zea</u> , and <u>Spodoptera exigua</u> in pheromone traps. F. C. Tingle and E. R. Mitchell	64
Staff List	App. 1
Manuscript Status Report	App. 2
Publications List	App. 3



Effect of Juvenile Hormone and Its Mimics on Mitochondrial
Metabolism in the Indian Meal Moth,
Plodia interpunctella
Part I: Mechanisms of Action

D. L. Silkacek and K. Kohl

(Continuation of report 91 74(7-12))

Objective: In studies on the effects of hormones on intermediary metabolism of Indian meal moths, we found that, in vitro, juvenile hormone (JH) inhibits some mitochondrial oxidations while stimulating others. These results suggested that the JH effects on mitochondrial metabolism might provide valid criteria for evaluating potentially active JH-mimics. The purpose of the present experiments is to determine the biochemical mechanisms of the JH actions.

Methods: Mitochondria were isolated from Indian meal moth larvae by differential centrifugation. Mitochondrial oxidative activities with succinate and pyruvate-malate as substrates were determined with a vibrating platinum electrode. Initial experiments were conducted with mitochondria isolated from larvae of different known ages. Subsequent experiments were conducted with mitochondria isolated from newly molted last-instar larvae.

Results: Studies on the JH stimulation of succinate oxidation in isolated mitochondria were continued. We have established that juvenile hormone alters mitochondrial membrane permeability. This alteration, which can be monitored spectrophotometrically, depends upon magnesium ions and pH, but does not require an energy source.

Plans: Studies will continue on determining what molecular structural characteristics are needed for affecting mitochondrial metabolism and elucidating the mechanism of JH-stimulated succinate metabolism. A manuscript reporting these results is near completion.



Effect of Juvenile Hormone and its Mimics on Mitochondrial
Metabolism in the Indian Meal Moth, *Plodia interpunctella*
Part II: Metabolic Effects

D. L. Silhacek and D. Firstenberg

(Continuation of report 02 74(7-12))

Objective: In studies on the effects of hormones on intermediary metabolism of Indian meal moths, we found that, in vitro, juvenile hormone (JH) inhibits some mitochondrial oxidations while stimulating others. These results suggested that the JH effects on mitochondrial metabolism could have profound effects on the overall metabolism of the insect. The purpose of the present experiments is to investigate possible effects of JH on the metabolism of the insect.

Methods: Mitochondria were isolated from Indian meal moth larvae by differential centrifugation. Mitochondrial oxidative activities with succinate and pyruvate-malate as substrates were determined with a vibrating platinum electrode. Cytochrome content was measured spectrophotometrically. Experiments were conducted with mitochondria isolated from larvae of different known ages.

Results: Portions of this work were written up and successfully submitted as a Ph.D. thesis by D. Firstenberg. A manuscript for publication has been prepared. No new areas were investigated during this reporting period.

Plans: Studies on the hormonal mechanisms governing cytochrome synthesis will be continued.



Juvenile Hormone Binding in Subcellular
Components of the Insect

S. M. Ferkovich and D. P. Putter

(Continuation of report 03 74(7-12))

Objectives: To establish the site of juvenile hormone (JH) action at the cellular and subcellular levels and isolate and identify receptor molecules responsible for binding the hormone in the Indian meal moth, Plodia interpunctella.

Methods: Haemolymph was collected from 12-day-old larvae (12 mg) and stored at -20°C until an adequate quantity was obtained for preparative isolation of the JH-carrier protein. JH degradative enzymes were separated from the carrier protein by gel permeation chromatography. Preparative ultracentrifugation was used to separate homogenates of epidermis into subcellular fractions. Binding of radiolabeled JH to the enzymes, carrier protein and epidermal fractions was measured by liquid scintillation counting. Metabolites of the hormone were detected by thin layer chromatography. Degradation of DNA, RNA and proteins in the homogenates was accomplished with enzymes obtained commercially.

Results: We previously reported on the influence of a JH-carrier protein on JH binding in epidermal target tissue 74(7-12). Subsequent experiments have been concerned with the effect of varying levels of carrier protein and JH on binding of the hormone in subcellular fractions obtained by differential centrifugation. When the concentration of JH was varied from 9.4×10^{-10} to 2×10^{-9} M (733 μg protein 12×10^{-20} JH), the following order of specific activity (dpm/ μg protein) in the fractions resulted: $600 \times \text{g} > 105,000 \times \text{g} > 20,000 \times \text{g}$ pellet. These results suggested that the binding affinity of the JH-carrier-protein-complex was highest in the $600 \times \text{g}$ fraction that primarily contains nuclei. In all three fractions, the carrier protein provided 95%+ protection against degradative enzymes. In other experiments, we determined the effects of three enzymes on JH binding in subcellular fractions of epidermal tissue. Pretreatment with RNase, DNase and trypsin induced a combined increase of 55.9, 45.8 and 41%, respectively in ^3H -JH labeling in fractions F_1 through F_4 obtained from the epidermal homogenates. This increase in JH-binding was accompanied by a 40, 37 and 453% increase in protein conc. as detected by the Lowry method in the RNase, DNase, and trypsin treated homogenates, respectively. These findings are not in agreement with those published by another laboratory and require additional study.

Plans: Components in the fractions of epidermal tissue that bind the JH-carrier protein complex will be biochemically characterized. The cytosol fraction ($105000 \times \text{g}$ supernatant) will be examined for the presence of receptors for JH. In addition, further studies on the effects of RNase, DNase and proteases on JH binding will be conducted.



Effects of Juvenile Hormone on the Metamorphosis
of Malathion-Resistant Indian Meal Moths

D. L. Silhacek, J. Zettler^{1/} and H. Oberlander

(Continuation of report 95 74(7-12))

Objective: To determine whether malathion-resistant strains of the Indian meal moth, Plodia interpunctella (Wübner) exhibit cross-resistance to juvenile hormone.

Methods: Malathion-resistant Indian meal moths were reared on juvenile hormone-treated diet. The percentage of insects reaching maturity was compared with the effect of the hormone on nonresistant insects.

Results: We concluded from this study that resistance to malathion does not confer resistance to juvenile hormone in Indian meal moths. However, our data did indicate the possibility that resistance might develop with sufficient selective pressure. A manuscript describing this work has been submitted for publication.

Plans: No additional work in this area is planned.

1/ Research Entomologist, Stored-Products Insects Research and Development Laboratory, Savannah, GA.



Interaction of Juvenile Hormone With Proteins In Tissue Culture

H. Oberlander and S. M. Ferhovich

Objectives: To determine (1) if fat body tissue is the source of the JH-carrier protein in the haemolymph, and (2) if components of tissue culture media influence the availability of JH to cultured tissue.

Methods: Fat body was taken from fifth-instar larvae. The tissue was cultured in plastic petri dishes in Grace's medium. The medium was modified with the addition of various proteins. Binding of the JH to proteins synthesized by the fat body was measured by gel permeation chromatography and liquid scintillation counting.

Results: When fat body from 15-18 mg larvae was incubated for 3 weeks in Grace's medium plus fetal calf serum and albumins, a ^3H -JH binding protein was detected with an elution volume similar to that for the JH-carrier protein found in the haemolymph. However, only 15% of the JH associated with the presumed carrier protein was unmetabolized. In contrast 95% of the JH associated with the carrier protein found in the haemolymph was unmetabolized JH. Elimination of the fetal calf serum and albumins in the tissue culture medium and shortening the time of incubation with fat body from 3 weeks to 10 and 5 days did not change the quantity of carrier-protein released into the medium. Also, fat body from younger larvae (10 mg) produced the same level of JH-binding protein as did tissue from 15-18 mg larvae. In addition, incubation of beta-ecdysone with fat body for 10 days did not enhance production of the protein.

Finally, we observed that ^3H -JH bound to albumins and other constituents in Grace's tissue culture medium. This phenomenon likely influences the availability of the hormone to the tissue being cultured.

Plans: To determine if the fat body is indeed synthesizing the carrier protein, binding of ^3H -JH to ^3H -leucine incorporated proteins produced by the fat body will be examined. Binding of ^3H -JH to various other tissue culture proteins will be studied.



Hormonal Control of Chitin Synthesis In vitro

H. Oberlander and G. E. Leach

(Continuation of report 07 74(7-12))

Objective: To determine the mode of action of ecdysone and juvenile hormone on the initiation and inhibition of metamorphosis. Ecdysone stimulates and juvenile hormone inhibits cuticle desposition in imagonal disks in vitro. We are focusing on the action of these hormones on the biosynthesis of cuticle.

Methods: Wing disks of the Indian meal moth, Plodia interpunctella (Hübner), are cultured in vitro in a modified Grace's medium. We examined the effects of inhibitors of RNA, DNA and protein synthesis on cuticle formation in hormone-treated tissue.

Results: Actinomycin D (0.1 µg/ml) suppressed more than 95% of the incorporation of tritiated uridine into RNA from cultured wing disks. Treatment with actinomycin D prevented cuticle formation by disks incubated with beta-ecdysone. We found that RNA synthesis during the ecdysone-dependent period was necessary for subsequent cuticle deposition. Cycloheximide (110 µg/ml) inhibited incorporation of tritiated leucine into protein from the cultured wing disks. Inhibition of protein synthesis during the ecdysone-dependent period prevented cuticle deposition. This effect of limited exposure to cycloheximide was reversed by subsequent incubation of the disks with additional beta-ecdysone. Inhibition of DNA synthesis had no effect on ecdysone-induced cuticle deposition.

Plans: The effects of beta-ecdysone and juvenile hormone on protein constituents of cuticle will be investigated.



Dissociation and Reaggregation of
Fat Body Cells During Metamorphosis

H. Oberlander and C. E. Leach

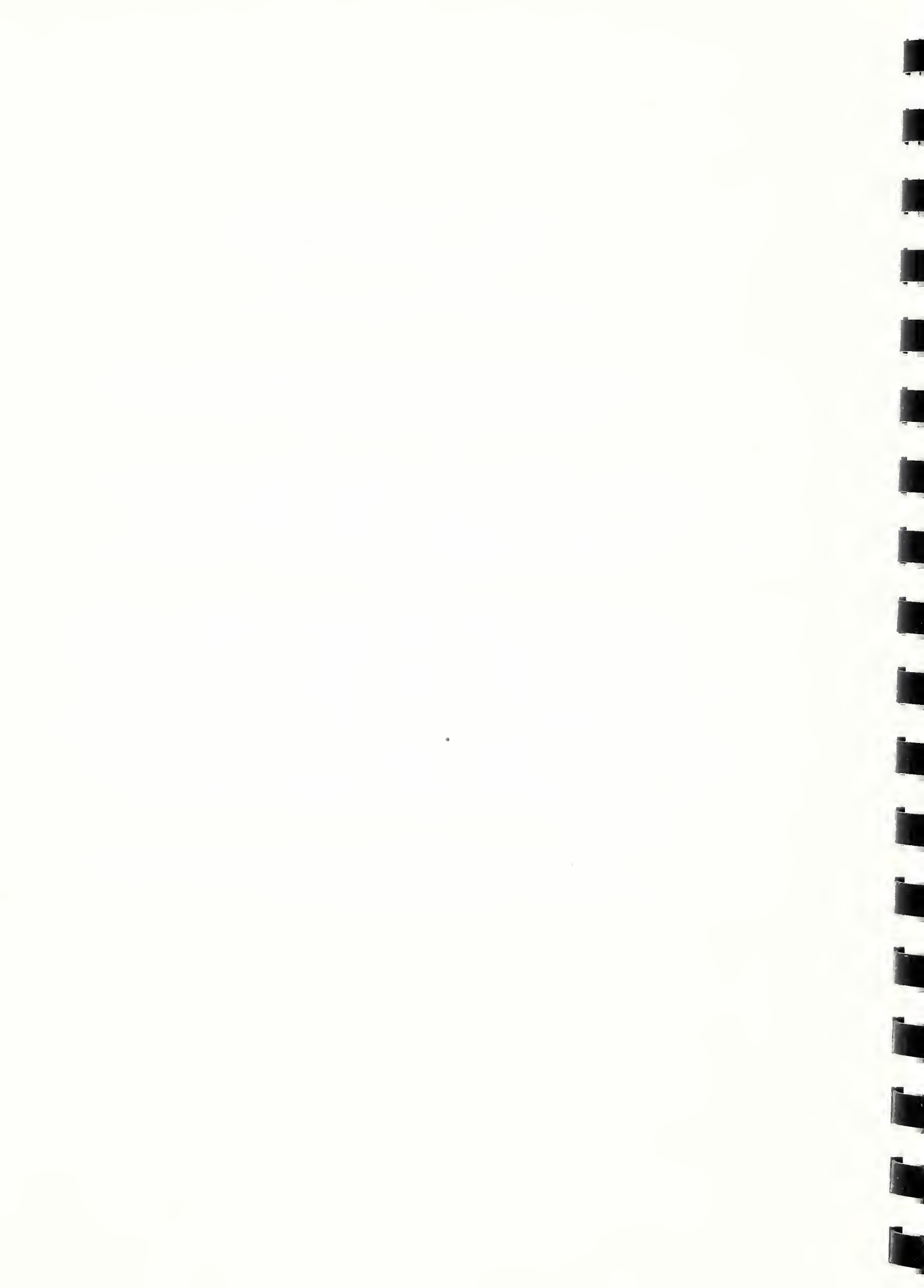
(Continuation of report 9 74(7-12))

Objective: To determine the mode of action of ecdysone and juvenile hormone on dissociation and reaggregation of fat body cells during metamorphosis.

Methods: Fat body tissue from larvae of the Indian meal moth, *Plodia interpunctella* (Hübner), was cultured in vitro in a modified Grace's medium. The degree of dissociation or reaggregation was monitored by microscopic examination. The degradation of juvenile hormone by the fat body was examined with thin layer chromatography and scintillation counting techniques.

Results: Beta-ecdysone at concentrations of 0.005 µg/ml to 5.0 µg/ml caused dissociation of cultured larval fat body. A minimum incubation period of 15 hours with hormone was required to elicit dissociation, but a 48 hour treatment was most effective. The fat body metabolizes 50% of the tritiated juvenile hormone in culture within 4 to 16 hours and greater than 75% of the hormone by 24 hours. However, when the juvenile hormone was incubated with haemolymph carrier protein only 15% of the hormone was degraded at 24 hours. This was reflected in our observations that it took one-quarter as much juvenile hormone to prevent ecdysone-induced dissociation when carrier protein was present compared to treatment with juvenile hormone alone.

Plans: The interactions between juvenile hormone and beta-ecdysone on the dissociation and reconstruction of the fat body will be examined.



Control of Egg Maturation in Dermestid Beetles

K. Vick, J. Coffelt and D. D. Tobin

Objective: To elucidate the hormonal control of egg maturation in black carpet beetles including the role of the male in stimulating egg production.

Methods: The presence of food, topical treatment with juvenile hormone (JH) and irradiation of the males (100 Kr of gamma radiation) to which females mated, were studied for their effect on egg production.

Results: The presence of food had no effect on the number of eggs produced by adult females. Topical application of JH caused a more rapid production of eggs than did mating. Females treated with 2 doses of JH at an interval of 72 hr between treatments laid more eggs than did those females that had received only one treatment or that had mated. Irradiation of male insects had no effect on their ability to induce egg production in females with which they had mated. However, females that were mated to irradiated males remated more frequently than those mated to normal males.

Plans: Research on the various components of vitellogenesis will continue.



Reproductive Biology of the Cigarette
Beetle, Lasioderma serricorne (Fabr.)

J. A. Coffelt and W. T. McClellan

(Continuation of report 19 74(7-12))

Objectives: These studies are part of a continuing series of investigations of the reproductive biology of the cigarette beetle and have as current specific objectives: 1) demonstration between remating propensity and pheromone content, and (2) elucidation of the relationship between density and mating frequency.

Methods: Relative quantities of pheromone in mated and unmated females were established by bioassay of extracts prepared from each group. Additional replications of previously described experiments designed to reveal the relationship between density and mating frequency were made.

Results: The bioassay data showed no clear distinction between the pheromone content of virgin and mated beetles. Within treatment, variation was greater than that between groups and was considerably higher than expected. Final replicates of mating frequency vs. density were supportive of previously reported results, i.e., the insects are capable of mating at extremely low densities.

Plans: Attempts will be made to disrupt mating communication by using sex pheromone and several density time exposures in closed areas of ca 1.0 m³.



Bacteremia as a Deterrent to Laboratory Rearing of
Biosteres (Opus) longicaudatus: Diagnosis, Control,
and Evaluation of Control Measures

P. D. Greany, G. Allen^{1/}, J. C. Webb, J. L. Sharp, and
D. L. Chambers

(Continuation of report 11 74(7-12))

Objectives: To discover means of reducing bacterially-related mortality of Caribbean fruit fly puparia parasitized by B. longicaudatus, and to evaluate the relative merits of alternate approaches to control.

Methods: In addition to studies reported earlier, tests were conducted on the influence of methenamine mandelate (M41) at 250, 500, and 1000 PPM in the larval rearing medium upon signaling sound produced by caribfly males. A test was also conducted on the influence upon longevity of incorporating M41 in the honey and water fed to B. longicaudatus adults.

Results: M41 was found to produce aberrations in the signaling sounds produced by caribfly males. Changes were noted in the harmonic content as well as in the fundamental frequency (Fig. 1), and the two phases (arbitrarily labeled x and y) observed when the controls' sounds became progressively less distinct at the higher doses. Significant changes in harmonic content are evident in Fig. 2, a frequency signature plot of representative sound pulses from each type of fly. Also, the y portion of the pulse diminishes considerably (noted by disappearance of the peak under the arrow).

The behavioral significance of these changes is not yet known, but the changes are very similar to those observed in sounds produced by flies which received sterilizing doses of radiation prior to emergence. In contrast, feeding M41 to adult parasites appears to be beneficial, at least in terms of increasing longevity. Significantly more parasites survived to 14 days of age when treated with M41 (Fig. 3). However, it is not known what effects upon parasite fecundity might occur as a result of treatment. Thus, considerable caution should be employed prior to using an antibiotic for disease control due to the potential for harmful side effects which may not be obvious. In the present study, we found that merely eliminating overcrowding and preventing the ambient temperature from exceeding 26°C sufficed to control excess mortality.

Plans: These studies are complete and a manuscript is in preparation. Histopathological and physiological studies have been initiated to better understand the relationship of stress to pathogenicity.

^{1/} Department of Entomology, University of Florida, Gainesville, FL.



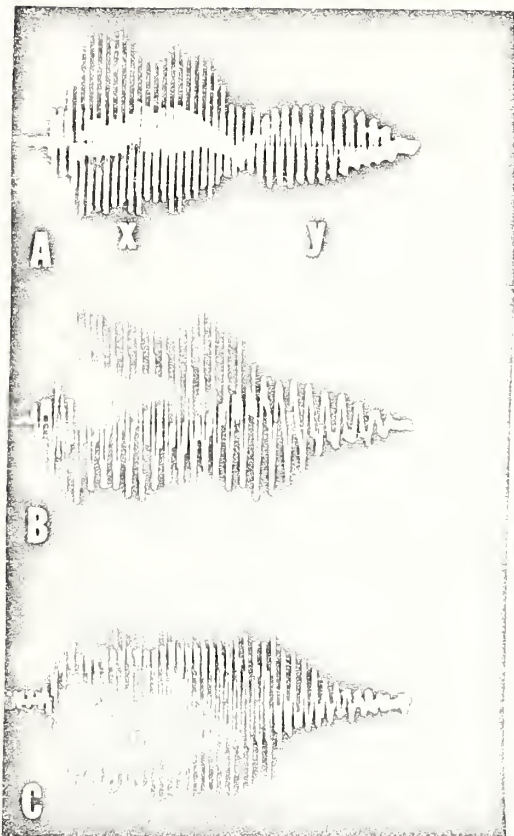


Fig. 1. Effect of methenamine mandelate (MM) upon male-produced signaling sound in the caribfly. A: untreated controls; B: 500 PPM; C: 1000 PPM.

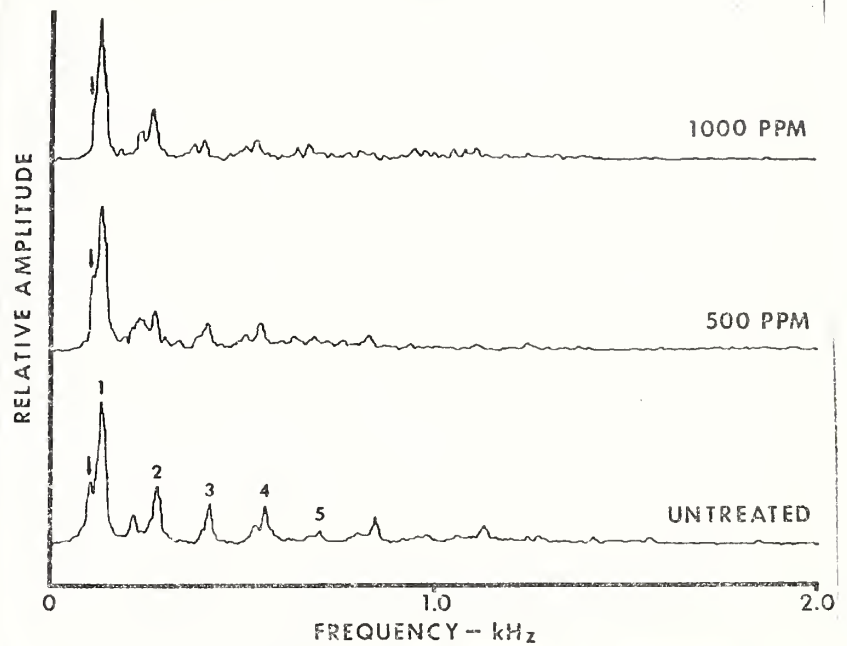
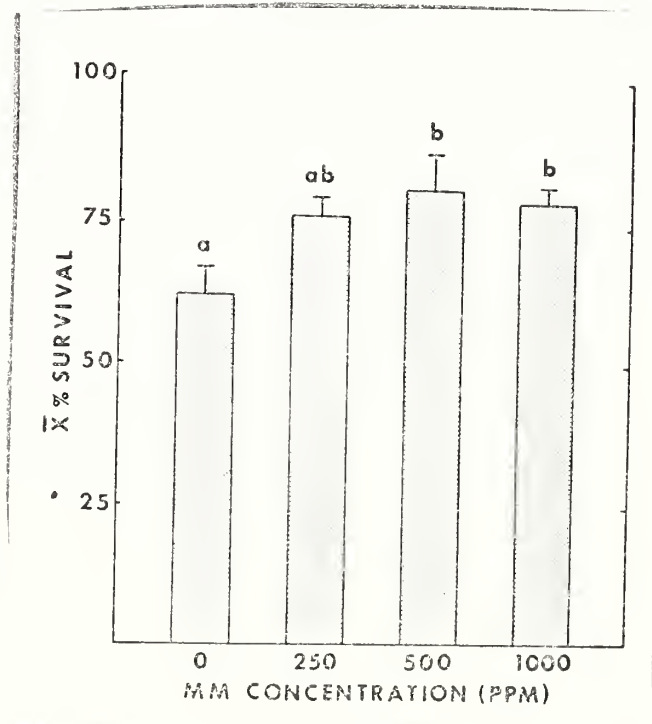


Fig. 2. Frequency signature analysis of the signaling sound in relation to MM concentration.

Fig. 3. Effect of incorporating MM in adult parasite honey and water upon survival of populations of *B. longicaudatus* to 14 days.





Histological, Bacteriological, and Physiological Studies
on the Invasion of Host Tissues by Bacterial Pathogens of
the Caribbean Fruit Fly

G. E. Allen^{1/}, P. D. Greany, T. C. Carlisle, and M. Chance^{1/}

Objectives: To define the anatomical site(s) within caribfly larvae and puparia at which bacterial invasion occurs, and to relate this phenomenon to the physiology of the host under stress.

Methods: Conventional histological procedures are being employed using paraffin sectioning techniques to conduct microscopic examination of stressed and non-stressed mature caribfly larvae and puparia. Samples of the bacteria present in specific regions of the gut are being acquired through dissection using sterile technique, and the pH of the gut in each region is being determined. Stress will be imposed by use of high rearing temperatures (32°C) and by parasitization by Piosteres longicaudatus, a larval parasite of the caribfly.

Results: Histological results have been good for the larval and nearly fully-formed puparial stages, but difficulty has been encountered in working with newly-formed and intermediate puparia. Consequently, efforts will be focused initially upon nearly mature larvae. It has been found that caribfly larvae are able to neutralize their diet (formulated at a pH of 4.5), so that the pH of the midgut is approximately 7.0. As the species of bacteria isolated from diseased hosts (compare earlier reports by Greany et al.) are destroyed by strongly acidic conditions, it is noteworthy that caribfly larvae inadvertently create hospitable conditions for potential pathogens.

Plans: Considerable additional effort is anticipated in order to more fully describe the pathology of this system. Innovations will be required to facilitate histological studies of newly-formed puparia, the stage most heavily afflicted by pathogens upon application of stress.

^{1/} Professor and Laboratory Technician, respectively, Department of Entomology, University of Florida, Gainesville, FL.



Introduction of Opius oophilus into Florida for
Control of the Caribbean Fruit Fly

P. D. Greany, R. H. Baranowski^{1/}, D. L. Chambers,
T. Wong^{2/}, and W. J. Schroeder^{3/}

(Continuation of report 12 74(7-12))

Objective: To establish Opius oophilus in Florida for control of the Caribbean fruit fly.

Methods: Field-collected parasites are being imported from Hawaii at ca. 1500 females and 500 males per month and are being released directly into the field at Homestead at sites with high fly populations. In addition, laboratory studies on rearing are being conducted with the intent of acquiring a strain of O. oophilus adapted to the caribfly and to provide additional parasites for release to supplement those imported from Hawaii.

Results: As reported earlier, O. oophilus females held in the laboratory responded best to caribfly eggs in guavas, as compared to eggs in other types of fruit. Efforts to devise an artificial host exposure technique have not yet been successful. Field results, however, have been encouraging in that small numbers of O. oophilus progeny have been recovered from samples of fruit collected in release areas, indicating that released parasites find the new conditions hospitable.

Plans: Releases will be terminated at the conclusion of the major guava season this fall. Fruit samples will be taken on a regular basis over the next several months to determine whether establishment has occurred.

1/ Department of Entomology, University of Florida, Homestead, FL.

2/ USDA Hawaiian Fruit Flies Investigations Laboratory, Honolulu, HI in cooperation with the Department of Entomology, University of Hawaii.

3/ USDA Horticulture Research Laboratory, Orlando, FL, formerly of the Hawaiian Fruit Flies Investigations Lab.



Effect of Temperature and Moisture Concentration of the
Pupation Medium on the Successful Development of Biosteres
(Opius) longicaudatus and Anastrepha suspensa

T. R. Ashley, P. D. Greany, and D. L. Chambers

(Continuation of report 13 74(7-12))

Objectives: To optimize the conditions during the pupation period of Anastrepha suspensa so as to achieve maximum survival and quality in the adult parasitoid, B. longicaudatus, and also, to ascertain if the exposure of A. suspensa larvae to the adult parasitoids stresses these larvae so that there is a difference in the required environmental conditions between these larvae and those not exposed to the parasitoids.

Methods: After exposure to the parasitoids, larvae were permitted to continue feeding until they migrated from the diet. At this stage they were placed into containers with vermiculite having moisture concentrations of 25, 50, and 75%. These containers were kept at temperatures ranging from 22-32°C (in 2°C increments) and at 60% ambient RH. Daily emergence and sex of the flies and parasitoids were recorded. After emergence ceased each puparium from which nothing had eclosed was opened and its contents recorded.

Results: The following general conclusions can be made about both flies and parasitoids: (1) the upper thermal limit for development is near 32°C; (2) maximum survival occurred between 24 and 26°C with 50 or 75% water concentration in the pupation medium; (3) the amount of moisture in the pupation medium did not affect the duration of the developmental period; (4) each 2°C increase in temperature from 22 - 28°C resulted in a significant reduction in the length of the developmental period; (5) no significant differences were found in the length of the developmental period between 28 and 30°C; (6) at 22°C the 25% moisture concentration caused significant mortality. Decreasing the temperature caused a significant increase in the length of the emergence period of the parasitoids but not of the flies. The moisture concentration of the pupation medium was more important than temperature in causing the parasitoid to diapause with up to 30% of puparia without emergence holes containing diapausing parasitoids at 25% moisture concentration and 26°C. No diapause was noted in A. suspensa. Exposing A. suspensa larvae to the adult parasitoids resulted in a reduction in total emergence compared with larvae not exposed to the parasitoids. Maximum survival occurred at the same combinations of temperature and moisture concentration for larvae exposed and not exposed to the parasitoids.

Plans: Further studies dealing with the relationship between diapause and temperature and moisture concentration are contemplated. A manuscript is being prepared.



Host Selection Studies on Biosteres (Opius) longicaudatus
in Relation to Attack of the Caribbean Fruit Fly

P. O. Lawrence^{1/} and P. D. Greany

(Continuation of report 15 73(7-12))

Objectives: To determine whether B. longicaudatus females are able to discriminate between parasitized and non-parasitized hosts and to define the maximal daily egg-laying capability of the parasite females.

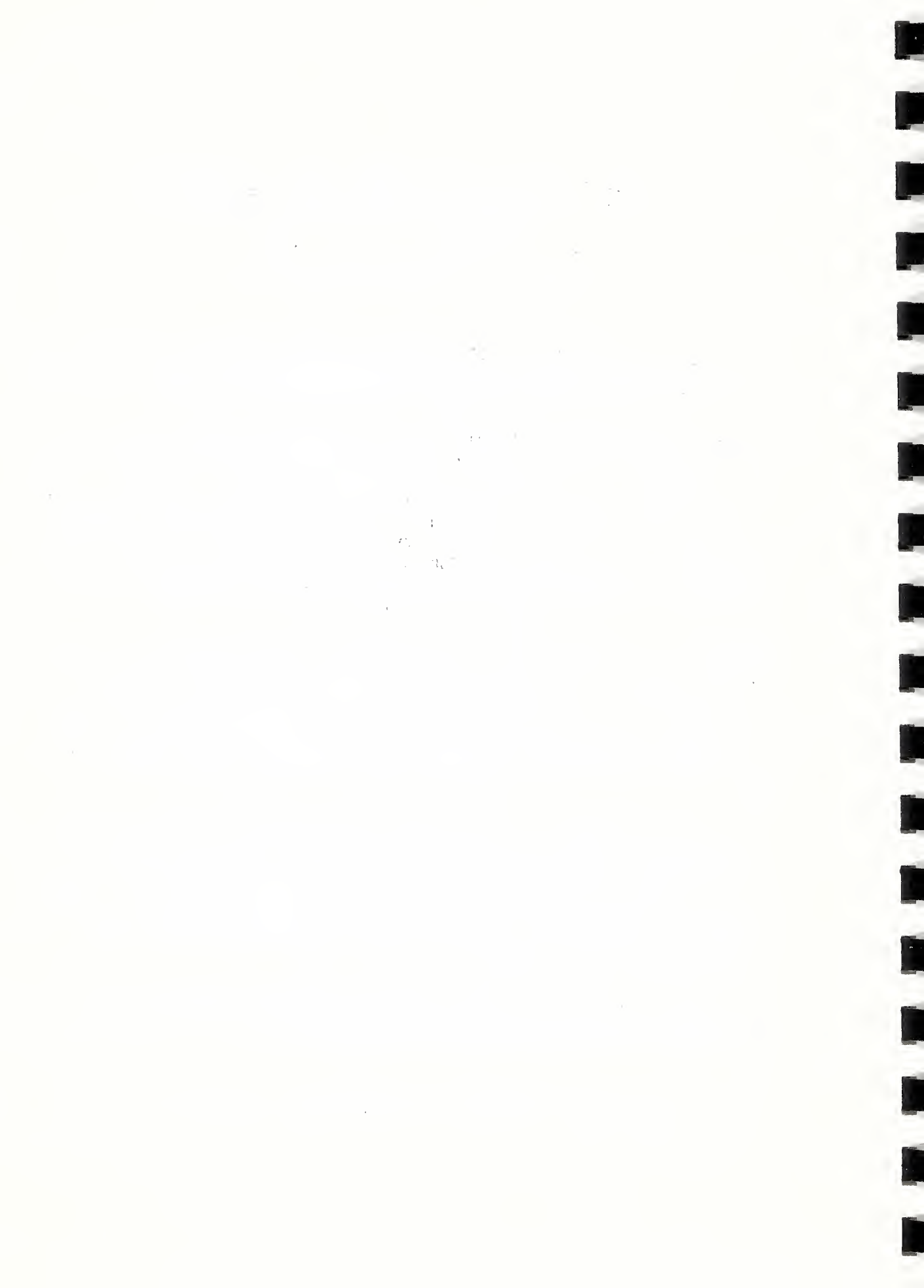
Methods: Discrimination studies were conducted by exposing 150, 350, or 700 late 2nd-to-early-3rd (final) instar caribfly larvae in diet for 24 hrs to 24 pairs of 5-day-old B. longicaudatus adults. This stage of the host was found earlier to be optimal for parasitization. After exposure, the larvae were allowed to develop for 1 day to allow parasite eggs to enlarge, and then samples of the larvae were preserved in 70% ETOH for dissection. The observed distribution of parasite eggs was compared with a theoretical random (Poisson) distribution using a chi-square analysis. Each density level was replicated 3 times.

Results: Host discrimination was evidenced in each replicate when 700 and 350 larvae were exposed to the parasites, with more larvae receiving only one egg than would be expected if the parasites had oviposited randomly. However, discrimination broke down in all replicates at the lowest host density level. Apparently, B. longicaudatus females have the sensory capability to distinguish the condition of the host, but under conditions of host deprivation accept even already parasitized hosts.

Dissections revealed that the egg-laying potential of B. longicaudatus is considerably greater than had been indicated in earlier studies. At the highest host density, an average of ca. 30 eggs were laid per female per day, whereas in effective parasitization studies only about 5 adult progeny were produced per female per day, even when hosts were exposed ad libitum, indicating a high degree of unsuitability of even the optimal stage of the caribfly.

Plans: Studies on the post-emergence egg production capability of B. longicaudatus females are planned, as well as studies on the physiological bases for and manifestations of unsuitability.

^{1/} Graduate student, Department of Entomology, University of Florida, Gainesville, FL.



Emigration: The Major Factor in Regulation of Growing
Tribolium castaneum Populations

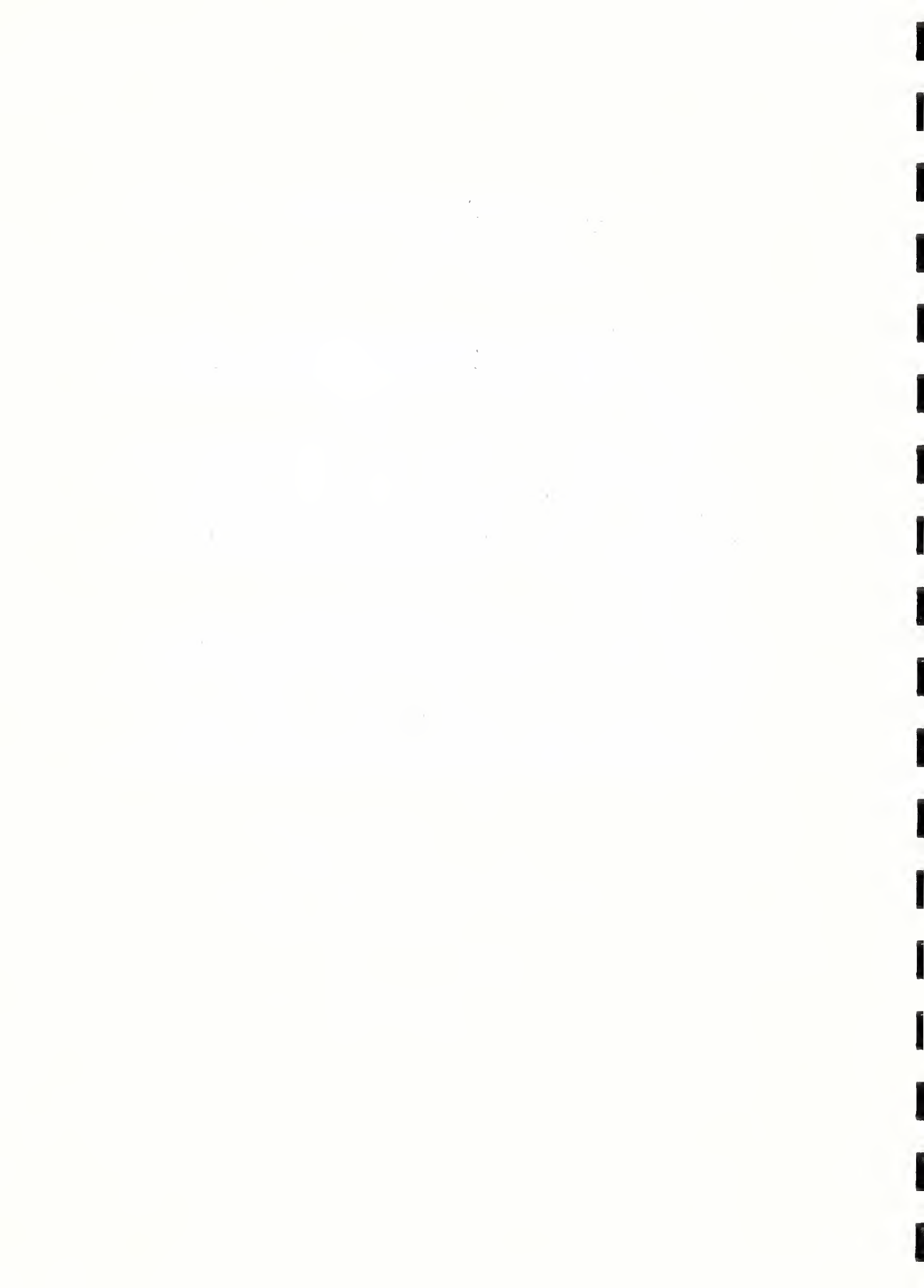
David W. Hagstrum, Edward E. Gilbert, and Burrell J. Smittle

Objective: To evaluate the functional relationship between emigration rate and age structure in a growing population with particular emphasis upon possible mechanisms for numerical regulation. This would provide insight into the potential for contamination when a single female is introduced into a warehouse.

Methods: One-day-old pupae were placed in woven stranded plastic bags that contained 40 gm of flour. The number of pupae per bag ranged from 50 to 600 in multiples of 50. Adult emigration was recorded daily. Similar studies were made using one-day-old pupae and three, five- or seven-day-old pupae in the same bag. Mutants with black adult body color were used so the 2 age groups could be distinguished.

Results: The number of adults emigrating during the 1st 6 days was negligible. Between the 7th and 12th days the number of emigrants averaged approximately 20% of the resident population. The presence of older adults did not result in earlier emigration.

Plans: Using an autoradiographic technique, we are currently comparing the tunneling activity of adults of different ages and plan to study the effects on tunneling of contacts between adults.



CO₂ Output During Maturation of Colonized
Cabbage Looper Larvae

N. C. Leppla and W. K. Turner

Objective: To relate CO₂ production to the fundamental behavioral events that occur during growth and development of cabbage looper, Trichoplusia ni (Hübner), larvae subjected to an established rearing protocol.

Methods: By using a CO₂ gas analyzer, the CO₂ output was monitored continuously from populations of 75 cabbage looper larvae maintained on artificial diet. The environment established in standard 16-oz rearing containers was 27±1° C and 70±5% RH with a 12-hr photophase (cool white lamps). The study included all stages from eggs through mature moths (2 replicates).

Results: During maturation, CO₂ output occurred in 6 characteristic phases including hatch and establishment (I, days 1-5), growth (II, days 6-9), prepupation (III, days 10-13), pupation (IV, days 14-18), emergence (V, days 19-21), and adult (VI, days 22-29+) (Table 1). Phase I involved a progressive daily increase in CO₂ production. Output was also continuous during phase II but the increase was 4-18 times greater each day. Phase III included a 12 to 5 unit/day decline, IV was stable, V indicated intermittent and circadian CO₂ discharge, and during phase VI the moths were photoperiodically entrained.

Plans: Mean growth rates and yields of each developmental stage will be measured and related to patterns of CO₂ production. CO₂ output will also be correlated with locomotor activity and other larval behavior patterns.



Table 1. Characterization of CO₂ output from a population of 75 cabbage looper larvae that developed on artificial diet (50 units = maximum CO₂ output).

Type	Duration (days)	Mean	Range	Maximum	Max 24-hr difference
Hatch-Establishment	5	3.33	2-6	6	1
Growth	4	28.75	12-49	49	13
Prepupation	4	23.75	14-37	37	12
Pupation	5	12.00	12-12	12	1
Emergence	3	29.33	25-36	36	11
Adult	7+	25.00	25-25	25	0



Micro Techniques and Analytical Methods

P. R. Heath, J. H. Tumlinson, and P. E. Doolittle

(Continuation of report 17 74(7-12))

Objective: To develop analytical techniques for structural elucidation of microgram quantities of pheromones and other natural products and for analyzing and purifying naturally derived and synthetic compounds, particularly isomeric mixtures.

Methods: During the past 6 months primary emphasis has been on high pressure liquid chromatography (HPLC). Slurry packing techniques for high resolution HPLC columns have been developed and are still being studied for further improvements.

Results: HPLC columns, 25 cm long by 0.25 in. o.d., have been packed with 5 μ m silica (Partisil 5). 10,000 theoretical plates per column, based on benzene & naphthalene peaks, have been obtained.

Plans: Further improvements in column packing techniques will be developed to give more theoretical plates per column.



Sex Pheromone Studies of the
Navel Orangeworm

J. A. Coffelt, K. W. Vick and L. L. Sower

(Continuation of report 18 74(7-12))

Objectives: To isolate and identify the female sex pheromone of the navel orangeworm, and to develop and understand the physiological, behavioral, and environmental factors that may influence male response to, or female production of the pheromone.

Methods: Pheromone was collected from individual females according to previously described methods. The pheromone was purified by means of liquid and gas chromatography. Fluctuation in female pheromone titer was determined by gas chromatographic analysis of the pheromone obtained from females at different times throughout a 24-hr period.

Results: Mass UV spectra of the pheromone were obtained. Significant quantities of pure pheromone were obtained. Female pheromone content varied considerably during a 24-hr period. Maximum pheromone quantities were recovered from females during that portion of the scotophase during which females were receptive to mating. GLC analyses of field-collected females (3 reps of 10-15 females each) indicate that wild females contain considerably more pheromone than do those reared in the laboratory.

Plans: Continue to collect pheromone for additional chemical testing and complete laboratory biology studies.



Borer Pheromones (Family Sesiidae)

J. H. Tumlinson, R. E. Doolittle, J. M. DeVore
and A. T. Proveaux

(Expansion and continuation of report 19 74(7-12))

Objective: As part of a program to develop an integrated pest management system for peach insects, the sex pheromones of the lesser peachtree borer and peachtree borer were isolated, identified, and synthesized and will be incorporated into survey and control programs for these insects. Additionally, these pheromones and isomeric compounds will be tested for survey and control of other sesiid species.

Methods: All of the isomers of 3,13-octadecadien-1-ol acetate and all of the isomeric alcohols have been synthesized and purified (>99%) and sent to the entomologists listed in Table 1 for field testing with the various Sesiidae species this summer. Liquid chromatography with AgNO₃-silica was used to purify the acetates and the alcohols were prepared by saponification of the respective acetates.

Results: Preliminary reports have been received from the field. The (E,Z) acetate is trapping large numbers of the lesser peachtree borer and small numbers of other species. The (Z,Z) isomer seems to be attractive to the greatest number of species including the peachtree borer and the western peachtree borer. It is being used in a large survey program for the western peachtree borer in California. The alcohols seem to be implicated as pheromone components in some species. Disruption of both the lesser peachtree borer and the peachtree borer with the (Z,Z) acetate has been successful in small orchards in Byron, Ga.

Plans: Several intermediates that were part of the original synthesis will be purified and tested for potential application as disruptants for the lesser peachtree borer and the peachtree borer.



Table 1. Borer pheromone cooperating scientists.

Mr. R. E. Dolphin Fruit Insect Research Invest. P.O. Box 944 Vincennes, Indiana 27591	Dr. E. R. Mitchell Insect Attractants and Basic Biol. Research Laboratory P.O. Box 14565 Gainesville, Florida 32604
Dr. T. D. Eichlin Laboratory Services/Entomology Division of Plant Industry California Dept. of Food and Agric. 1229 H. Street Sacramento, California 95814	Dr. H. P. Moffitt Insect of Fruits, Vegetables, & Field Crops Yakima Agric. Res. Laboratory Yakima, Washington 98902
Mr. G. R. Gentry S. E. Fruit and Tree Nut Research Laboratory P.O. Box 87 Byron, Georgia 31008	Dr. D. G. Nielsen Entomology Department Agric. Res. & Development Center Wooster, Ohio 44691
Dr. R. L. Holloway Dept. of Entomol. and Zool. Clemson University Clemson, South Carolina 29631	Dr. J. L. Sharp Insect Attractants and Basic Biol. Research Laboratory P.O. Box 14565 Gainesville, Florida 32604
Dr. P. L. Horsburgh College of Agriculture and Life Sci. Va. Polytechnic Inst. & State Univ. Dept. of Entomology Shenandoah Valley Res. Stn. Steeles Tavern, Virginia 24476	Dr. Yoshio Tamaki Division of Entomology National Inst. of Agric. Science Mishigahara, Kita-Ku Tokyo, Japan
Dr. M. G. Karandinos Department of Entomology University of Wisconsin 1630 Linden Drive Madison, Wisconsin 53706	Dr. James Tette N.Y.S. Apple Pest Management Project N.Y.S. Agric. Exp. Stn. Department of Entomology Geneva, New York 14456
Dr. J. R. McLaughlin Insect Attractants and Basic Biol. Research Laboratory P.O. Box 14565 Gainesville, Florida 32604	Mr. C. E. Yonce S.E. Fruit and Tree Nut Res. Lab. P.O. Box 87 Byron, Georgia 31008



A Potent Sex Attractant for the Carpenterworm Moth
Prionoxystus robiniae

R. E. Doolittle, J. D. Solomon^{1/}, W. L. Poelofs^{2/}, M. Beroza^{3/},
 W. McKnight^{4/} and A. Tagestad^{5/}

(Continuation of report 20 74(7-12))

Objectives: Isolate, identify, and synthesize the female produced sex pheromone of the carpenterworm moth and evaluate its usefulness as a survey and/or control tool for the insect.

Methods: For isolation, identification, and bioassay methods, see (23 73(1-6)). Separation (purification) of the (Z,E) and (E,E) isomers to greater than 98% purity of 3,5-tetradecadien-1-ol acetate was accomplished by a combination of spinning band distillation and high pressure liquid chromatography.

Results: As described in the plans section of report 20 74(7-12), several grams of synthetic attractant was prepared and purified by spinning band distillation. This material was distributed to our cooperators for the 1975 field tests; however, no formulation was carried out. In addition, some assistance was given to the supplier who prepared a quantity of the attractant under a contract with our cooperators. This involved the analysis of the results of several trial preparations. The commercially prepared sample has been delivered thus relieving us of any further responsibility for synthesizing additional attractant.

Plans: The last remaining obligation we have is to fractionate the commercially prepared sample by spinning band distillation, analyze the fractions and distribute them to our cooperators. We will continue to render any consultative help we can to enable our cooperators to conduct their tests.

- 1/ Forest Service Insect Research Laboratory, P.O. Box 227, Stoneville, MS.
- 2/ New York Agricultural Experiment Station, Geneva, N.Y.
- 3/ Organic Chemical Synthesis Laboratory, Agricultural Environmental Disease Research, Forest Service, Washington, D.C.
- 4/ Division of Forest Insect Research, 1621 North Kent, Arlington, VA.
- 5/ Shelterbelt Laboratory, Bottineau, N.D.



Isolation and Identification of the Sex Pheromone
of the White Peach Tree Scale

R. R. Heath, J. R. McLaughlin, J. H. Turlinson, T. R. Ashley,
and D. L. Chambers

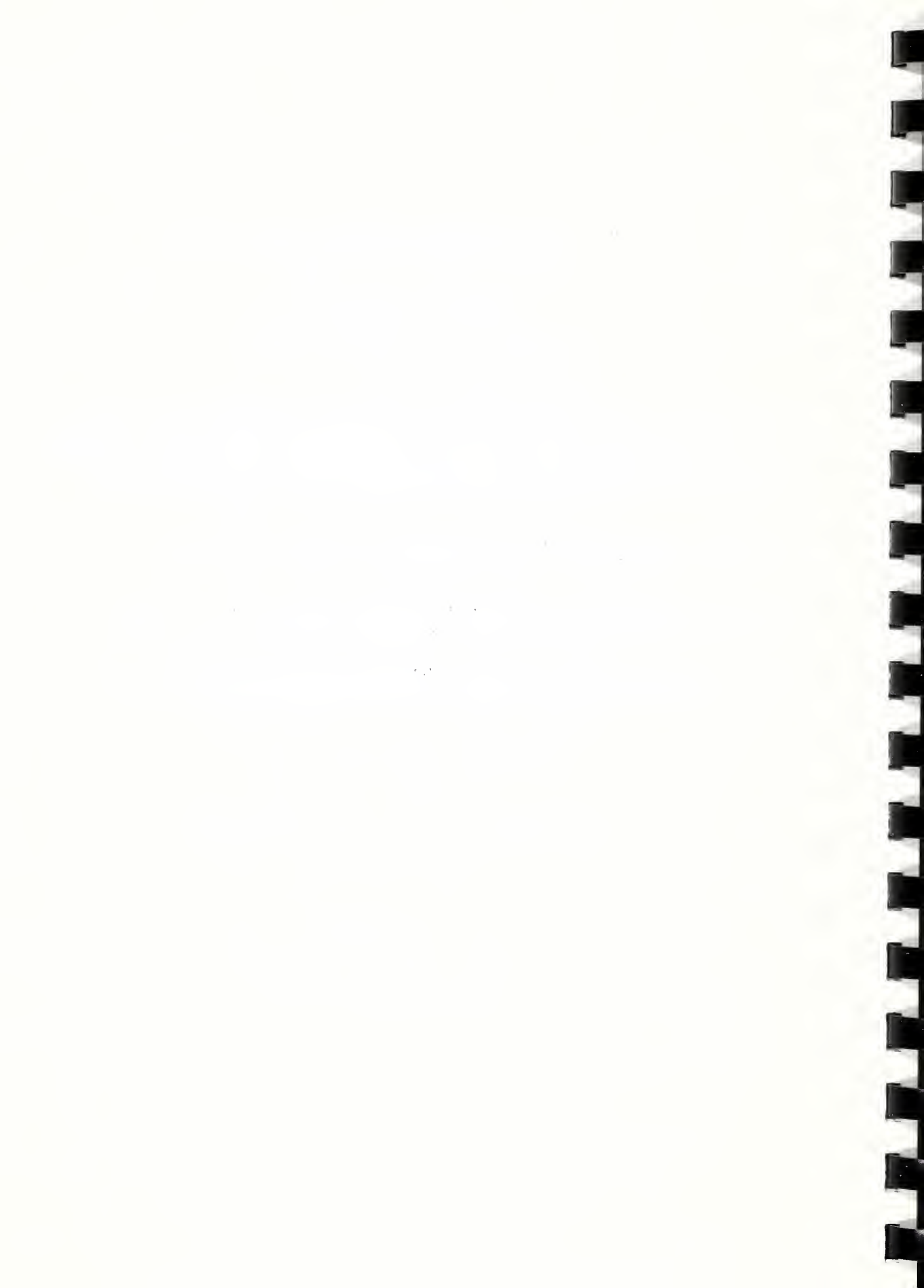
(Continuation of report 22 74(7-12))

Objectives: As part of a program to develop an integrated pest management system for peach insects, the sex pheromone produced by the female white peach scale, Pseudaulacaspis pentagona, will be isolated, identified, synthesized, field tested, and incorporated into survey and control programs.

Methods: Isolation procedures previously described were used to obtain additional quantities of the pheromone.

Results: Material is still being collected from females. Rearing procedures have been improved to provide more females.

Plans: The pheromone will be analyzed using microspectroscopic techniques when sufficient material has been collected and purified.



Exploitation of the Sex Pheromone Behavior
of Sesiidae: Disruption of Mating Communication

J. P. McLaughlin, R. E. Doolittle, E. P. Mitchell, R. Gentry^{1/}
J. H. Tumlinson and C. R. Yonce^{1/}

(Continuation of report 24 74(7-12))

Objective: To develop techniques for disrupting mating communication in the lesser peachtree borer and peachtree borer.

Methods: The lesser peachtree borer pheromone (E,Z)-3,13-octadecadien-1-ol acetate, or peachtree borer pheromone (Z,Z)-3,13-ODDA have been dispensed from laminated plastic (Hercon[®]) strips and from packets of capillary tubes (Conrel[®]). These dispensing methods have been compared for longevity of effect as trap baits and have been utilized to quantify mating disruption experiments using the atmospheric permeation technique.

Direct observation of baited Pherocon[®] 1C traps was used to determine their efficiency in trapping lesser borers.

Results: Nearly 100% reduction in trap captures of male LPTB can be achieved by surrounding a pheromone-baited trap with dispensers releasing either the (Z,Z) or (E,Z) isomer at the rate of 5 ng/min/m². The disruptive effect is markedly reduced with a 10-fold reduction in the pheromone concentration below this level.

Both the Conrel and Hercon dispensers are very effective for baiting traps. They appear to have equal longevity. We have achieved good catches of male LPTB with Pherocon 1C traps baited with dispensers releasing at 100 ng/min.

Observation of the Pherocon trap baited with 100 ng/min revealed that only about 50% of entries by male LPTB into the trap resulted in capture. Total males captured were only about 25% of those observed flying in the vicinity of the traps; however, we cannot correct these figures for males that may have returned on several occasions. Nonetheless, the trap is not highly efficient. The pheromone bait should be placed as near the sticky surface as possible and the trap should be primed with a dark object resembling a LPTB.

Plans: Practical field testing of atmospheric permeation will continue at Byron. Gainesville personnel will continue to develop methodology for permeation and trapping approaches.

^{1/} Research Entomologists, Southeastern Fruit and Tree Nut Research Laboratory, Byron, Georgia.



Control of Stored-Product Insects
with Sex Pheromones

L. L. Sower, K. W. Vick, J. A. Coffelt, and P. Whitmer

(Continuation of report 25 74(7-12))

Objectives: To make sound recommendations as to whether or not certain lepidopterous pests of stored products can be directly controlled by introducing synthetic sex pheromone into their environment. The immediate objectives are to determine the effects of pheromone concentrations and population densities on the mating frequencies and behavior of Indian meal moths, and Angoumois grain moths.

Methods: Indian meal moths were introduced into 90 m³ enclosed environment holding 180 lbs of unshelled peanuts. Several population densities of Indian meal moths were released into these rooms in the presence and absence of synthetic pheromone. The population growth trends of these insects were then monitored for 1-3 generations.

Results: Parent populations of 500 Indian meal moths released 100 at a time over a 6 week period, ultimately produced 3,680 first generation adults in the presence of synthetic pheromone and 7,390 progeny in the absence of pheromone. Similarly, 50 moths produced 540 and 2,320 adult progeny, respectively in the presence and absence of pheromone. In addition (incomplete) tests populations of 20 adults have thus far produced 48 and 298 adult progeny, respectively in the presence and absence of pheromone. Mating frequency vs. population density data were consistent with previous results. Populations of almond moths accidentally present in some of the tests behaved similarly to the Indian meal moths. The two species appeared to have no effect on one another except their mutual contribution to food shortages where extremely high larval densities occurred. Both the almond and Indian meal moth females were successful in mating with their own males in the presence of large numbers of both species where no synthetic pheromone was present.

Plans: Research in simulated warehouses will continue with the three lepidopterans (almond moth, Indian meal moth and Angoumois grain moth), and two coleopterans (Trogoderma inclusum and cigarette beetle) to determine ways to best use sex pheromones and mating inhibition to control stored-product insects.



Resistance to Atmospheric Permeation as a
Means for Mating Control of Lepidoptera

J. R. McLaughlin and L. L. Sower

(Continuation of report 26 74(7-12))

Objective: To determine the potential selective influence of a pheromone-permeated atmosphere on the premating behavior of a moth.

Methods: We have begun a series of mating selection experiments using a strain of the almond moth, Cadra cautella, newly colonized from the field vs. an established laboratory colony of Cadra.

As in our previous tests with Plodia interpunctella, adults from each generation are mated in standardized cages. In each generation some of the lines must mate in a sex pheromone-permeated atmosphere while others are allowed to mate with no interference. Periodic tests of the mating rate of these lines at various pair densities and pheromone concentrations are made.

Other behavioral characteristics of the wild vs. lab strains of Cadra are also being observed.

Results: Through the 7th generation we have observed no characteristics indicating development of resistance to the pheromone treatments. Certain interesting differences of growth and behavior between the wild and lab strain do occur.

Plans: We will pursue the objective as stated. Selection will be carried 3 to 5 more generations.



Use of Sex Pheromones for Behavioral Control
of Loopers and Related Noctuid Species

J. P. McLaughlin, E. P. Mitchell and D. L. Chambers

(Continuation of report 28 74(7-12))

Objectives: This study explores methods for controlling population levels by manipulating the sex pheromone communication systems of the cabbage looper, soybean looper, and several related species. The technique whereby the sex pheromone is continually evaporated into the air over a test area at a concentration above the male behavioral threshold (environmental permeation) is being examined. To test this technique adequately against wide-ranging species such as the cabbage and soybean loopers, large land areas must be treated. We are developing a system for broadcasting the pheromone over such areas.

Methods: Tests of microencapsulated (Z)-7-dodecen-1-ol acetate formulations were made during the report period. The materials were applied to cabbage foliage with a Hudson (prototype model) ULV backpack sprayer. Ten NCP formulations were tested. These included several polymeric wall materials, two solvent systems, two levels of post-treatment, and an antioxidant. They were applied at 3.2 to 3.6 g actual pheromone per 0.1 ha. Five Pennwalt formulations with varying wall porosity and containing dilute or neat pheromone were applied at rates from 1.1 to 3.0 g actual per 0.1 ha. All formulations contained a thickener to suspend the capsules, and a latex sticker to increase their adherence to the foliage. Can traps baited with (Z)-7-dodecen-1-ol acetate were placed at the center of each plot and in nearby untreated areas.

Results: All treatments reduced the capture of male cabbage loopers relative to the controls for the 24 days of the test. The following statements regarding NCR formulations are based on results of the previous as well as the present report. No other wall material developed by NCR was superior to that originally used with the Gypsy moth pheromone. The 2.2% pheromone in xylene solvent system gave results much superior to a xylene-trichloroethylene system. No clearly separable effect of a high vs. a low level of post treatment (polymer coating) of the capsules was observed. Formulations containing UOP 688 antiozonant performed slightly better than those without the antiozonant. The most consistent results for the tests in both soybean and cabbage were obtained with a high post-treatment capsule containing antiozonant.

The Pennwalt material has been tested only once on cabbage. Two highly effective formulations were found. Both utilized a high (relative to others tested) porosity capsule. In one sample the pheromone was encapsulated at a 2% solution in xylene and sprayed at 1.6 g actual pheromone per 0.1 ha while the capsules in the other sample contained neat pheromone and were applied at 3.0 g actual pheromone per 0.1 ha.

Plans: Continued testing of microcapsular formulations will be conducted. Tests of Hercon[®] and Conrel[®] dispensing systems are also planned.



Disruption of Mating in Heliothis zea and H. virescens

E. P. Mitchell, A. H. Baumhover^{1/}, and H. Jacobson^{2/}

(Continuation of report 29 74(7-12))

Objective: To evaluate the effects of Z-9-tetradecen-1-ol formate (Z-9-TDF), Z-9-tetradecenal (Z-9-TDAL), and Z-11-hexadecenal (Z-11-HDAL) on pheromone communication and mating in H. zea and H. virescens.

Methods: A treated plot and a control were set up ca. 60 m apart, perpendicular to the prevailing wind. Chemicals tested were evaporated into the air from dispensers (25 or 36) arrayed at intervals of ca. 3 m in either a 5 X 5 or 6 X 6 checkerboard pattern. The chemicals were evaporated from either Hercon® plastic strips (Z-9-TDAL, 1 in²) or polyethylene vials (Z-9-TDF, 25 mg ea) held ca. 1 m above the soil surface on wooden stakes. Each type of dispenser released the test chemical at ca. 300 ng/min. The treatment and control plots were rotated daily. The disruptive effect of the compounds was assessed by determining whether laboratory-reared females confined on cotton plants located near the center of the plots would mate with wild males. The plants were encircled with an aluminum ring (1.1 m diam., 15.2 cm high). The inner surface of the ring was dusted with talc to discourage moths from climbing up the band and escaping. The wings of the females (10-15/plot) were clipped, and then they were released onto the plants just before sundown. (In a separate trapping experiment, we found that females with clipped wings were as attractive to wild males as normal females). The females were collected the following morning and dissected to determine if they had mated. Because several of the females either escaped or were eaten by predators, the % reductions in matings necessarily were based upon the numbers of females actually recaptured.

Results: Z-9-tetradecen-1-ol formate was highly effective in disrupting mating in H. zea (Table 1). Disruption of mating in H. virescens also was achieved although the mean % disruption was less than that recorded for H. zea. A component of H. virescens' sex pheromone, Z-9-TDAL, also was very effective in disrupting mating in this species. The effect of this chemical on pheromone communication by H. zea is under investigation. The other component of the H. virescens' pheromone, Z-11-HDAL, currently is being evaluated as a mating disruptant for both H. virescens and H. zea.

Results shown in Table 2 confirm our previous assertion that Z-9-TDF is indeed a mating disruptant for H. zea and H. virescens. The reductions in moth captures reported here were very similar to those previously reported (2nd Semi-Annual Report 1974). Moreover, the reductions in matings obtained in these experiments correlate closely with the reduced trap captures.

Plans: The effects of the subject chemicals on the mating behavior of H. zea and H. virescens will continue. Various microencapsulated formulations of Z-9-TDF also will be evaluated.



Table 1. Disruption of mating in *Heliothis zea* and *H. virescens* in field plots treated with Z-9-tetradecen-1-ol formate or Z-9-tetradecenal.

Test no. and chemical	Replicates†	Total no. females			Reduction in mating (%)‡
		Released	Recaptured	Mated	
<u>H. zea</u>					
Control	14	184	166	68	
<u>Z-9-TDF</u>	14	184	147	2	96.7*
<u>H. virescens</u>					
Test 1					
Control	12	144	122	78	
<u>Z-9-TDF</u>	12	144	108	13	81.2*
Test 2					
Control	11	134	104	50	
<u>Z-9-TEAL</u>	11	134	101	2	95.9*

* Number of insects/replicate: H. zea, 12-15; H. virescens, 10-14.

† Percentage reduction in mating = $\frac{\% \text{ ♀ mated controls} - \% \text{ ♀ mated treatment}}{\% \text{ ♀ mated controls}} \times 100$.

‡ Mating significantly reduced by treatment ($P = 0.05$, Student's t-test).



Table 2. Efficacy of Z-9-tetradecen-1-ol formate in disrupting pheromonal communication (relative to controls) in Heliothis zea and H. virescens based on captures of ♂ in ♀-baited traps and mating by ♀ (wings clipped) released on cotton plants.*

Species	Reduction in trap captures (%)	Reduction in mating (%)
<u>H. zea</u>	88.6 (10)†	96.7 (14)
<u>H. virescens</u>	86.6 (16)	81.2 (12)

* The chemical was evaporated into the air of each plot from 36 dispensers arrayed at intervals of 3 m in a 6 × 6 checkerboard pattern.

† Replicates shown in ().



Field Evaluation of Sex Pheromones of the Fall Armyworm,
Beet Armyworm, and Southern Armyworm

E. R. Mitchell and R. E. Doolittle

Objective: (Z,E)-9,11-tetradecen-1-ol acetate is a sex pheromone of some Spodoptera sp. in Asia and Africa, but it has not been reported as a pheromone for species of Spodoptera in North America. The objective of this study was to determine the relative attractancy of this compound to wild males of the FAW, BAW, and SAW.

Methods: Samples of (Z,E)-9,11-TDDA were obtained from M. Jacobson^{1/} and B. Nesbitt.^{2/} Pherocon[®] 1C sticky traps were baited with plastic vials containing either 1 or 25 mg of the chemical (3 traps/treatment) and positioned in an area at Hastings, Fla., known to have high populations of the FAW and BAW.

Results: (Z,E)-9,11-TDDA did not attract moths of any species in these trials.

Plans: The isolation and identification of the natural pheromones for these insects will be undertaken.

^{1/} Agricultural Environmental Quality Institute, Beltsville, Maryland.

^{2/} Tropical Products Institute, London, England.



Seasonal Abundance of Lesser Peachtree Borer and
Peachtree Borers in North Central Florida

J. L. Sharp, J. R. McLaughlin, and J. H. Tumlinson

Objective: To determine the seasonal abundance of lesser peachtree borer and peachtree borer males in north central Florida.

Methods: Pherocon® sticky traps baited with E,Z or Z,Z isomers of 3-13-octadecadien-1-ol acetate were positioned at several locations in woods and near or in peach trees in orchards in Hawthorne, Gainesville, Lowell, Belleview, and Pedro, Florida, beginning April 1975. Initially, each trap was baited with 200 µg of either pheromone isomer in planchets in the field. Trap catches were counted biweekly.

Results: Populations of lesser peachtree borers fluctuate weekly and have been 15-20 x greater in numbers than peachtree borers as of July 1975. Catches of LPTB usually were low ($\bar{x} = 3/\text{trap}$) in wooded areas next to peach orchards but higher ($\bar{x} = 20/\text{trap}$) in woods isolated from orchards. Catches were the greatest in peach orchards.

Plans: This study will continue.



Sticky Traps Baited With E,Z or Z,Z Isomers of 3-13-Octadecadien-1-ol Acetate Catch Several Different Sesiid Moths in North Central Florida

J. L. Sharp and John R. McLaughlin

Objective: To determine which sesiid moth species other than Synanthedon pictipes or Sanninoidea exitiosa are captured in sticky traps baited with E,Z or Z,Z isomers of 3-13-octadecadien-1-ol acetate (ODDA).

Methods: Pherocon[®] sticky traps baited with E,Z and Z,Z isomers of ODDA were positioned at several locations in woods and near or in peach trees in orchards in 5 different areas in north central Florida beginning April 1975. Initially, each trap was baited with 200 µg of either pheromone isomer in planchets. Bi-monthly, 50 µg of either isomer were added to planchets in the field. Trap catches were counted bi-weekly. Clearwing moth identification was done by Dr. T. D. Eichlin, Division of Plant Industry, Sacramento, California.

Results: As of Aug. 1, 1975, 9 different clearwing moths have been captured in sticky traps baited with the E,Z isomer: Parathrene dolli, P. simulans, P. asilipennis, Synanthedon arkansasensis, S. pictipes, S. alleri, S. sapygaeformis, Sannia uroceriformis and Vitacea scepsiformis. Seven different clearwing moths have been captured in sticky traps baited with the Z,Z isomer: P. simulans, Sanninoidea exitiosa, Carmenta texana, S. sapygaeformis, S. geliformis, Podosesia syringae, and V. scepsiformis.

Plans: This study will continue.



Air Puff Sensor for Insect Electrophysiological Studies

E. W. Hamilton

The air puff sensor unit described is sensitive to very small air currents. Although the unit can be used for determinations of air movement it was designed for use in electrophysiological measurements as a triggering and stimulus onset-monitoring device.

Description: The RCA[®] CA3018 transistor array is the heart of the sensor system (Fig. 1). Two Fenwal[®] No. GB31P2 1-Kohm glass probe thermistors are used, thermistor A for automatic ambient temperature correction and thermistor B as the air current sensor. V_{out} , applied to an oscilloscope or a microammeter, is used to indicate the passage of an air current. The circuit is designed so that the thermistors are slightly heated. Small air currents then cause a cooling of the thermistor and a resultant change in thermistor resistance. Thermistor A must be enclosed in a matrix such as beeswax or cement that is responsive to ambient temperatures but not to air currents.

Use: The system, when used in olfactory electrophysiological studies (Fig. 2), will detect the onset of air movement created when air is forced across an odorant source. The thermistor sensor is very small so it can be placed near the insect antenna. Thus the electrophysiological response can be monitored from the precise moment of stimulation, and the latency of the neural response can be more precisely determined. The unit can be either battery operated or AC powered with a well-filtered DC power supply.

Plans: A manuscript has been prepared and is now submitted to the journal for publication.



Figure 1. Air current sensor.

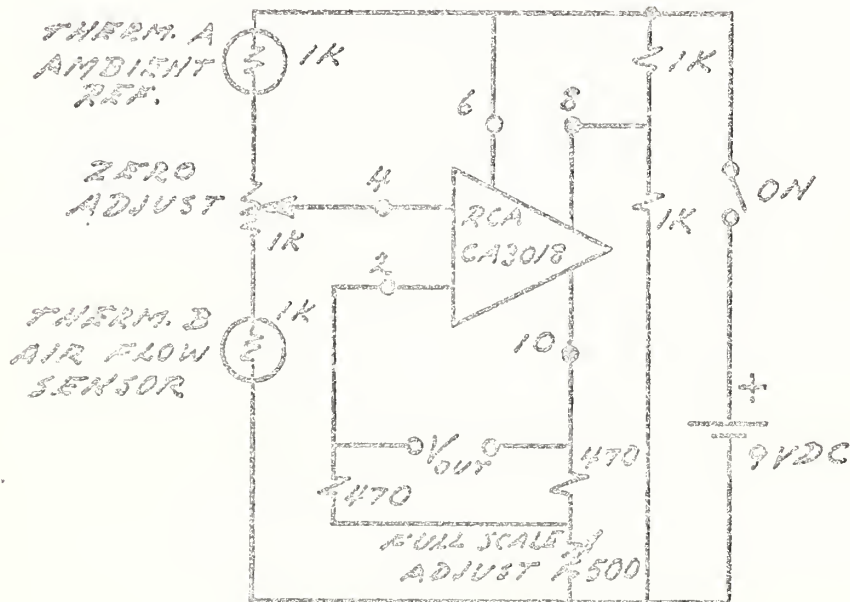
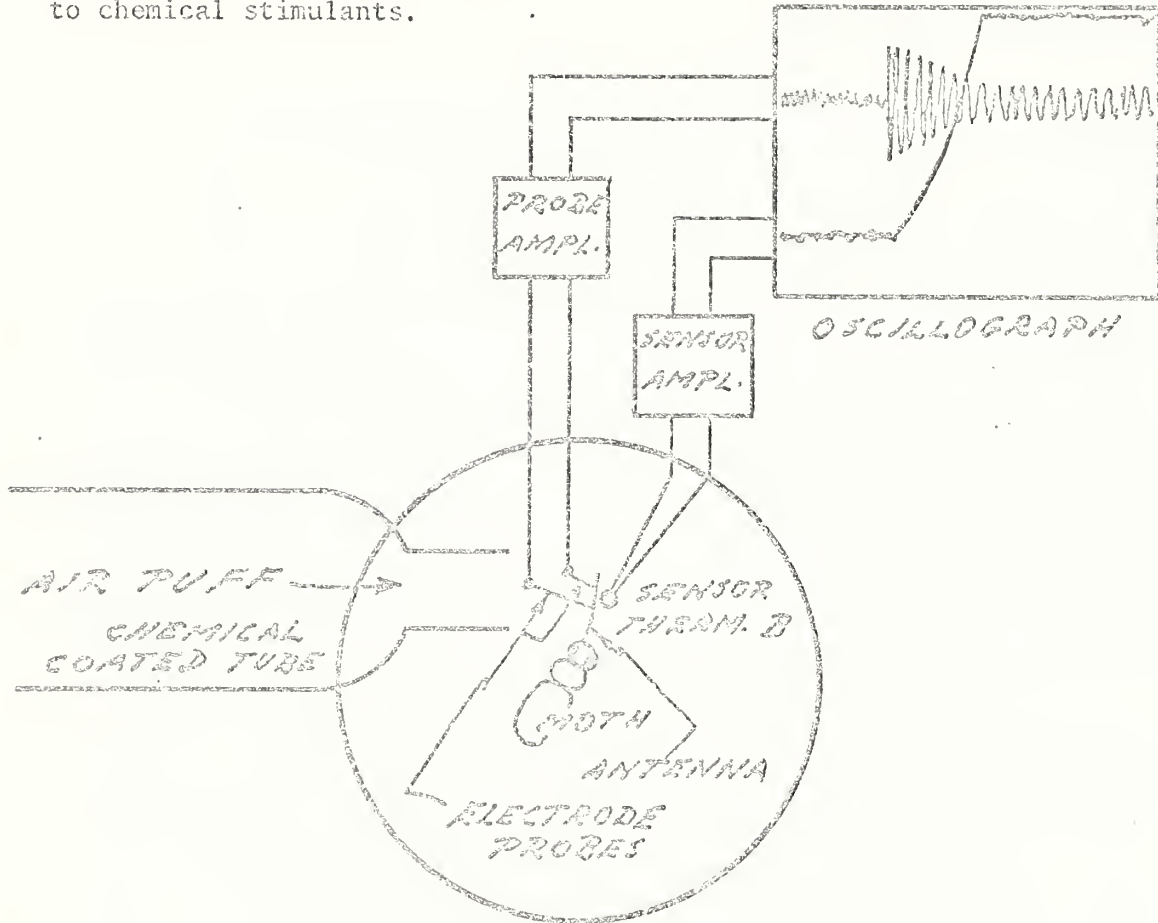




Figure 2. Use of air puff sensor to determine response of moth antenna to chemical stimulants.





Attracting Fall Armyworms with Controlled Release
Plastic Strips Containing Z-9-DDA

F. C. Tingle and E. R. Mitchell

Objectives: To evaluate Herculite strips containing Z-9-DDA for longevity as compared to plastic vials containing the pheromone for attracting the fall armyworm, Spodoptera frugiperda.

Methods: Plastic strips containing 24 or 25 mg/in.² and 122 or 127 mg/in.² of Z-9-DDA were sized into 1 x 2, 1.0, 0.5, or 0.25 in.² strips and used as bait in Pherocon[®] 1C sticky traps. These strips were compared to plastic vials containing 25 mg Z-9-DDA. Baited traps were placed near the edges of corn fields for capture of fall armyworm moths. Strips and vials were then aged outside until they were tested 4 and 9 weeks later when they were compared with fresh baits. Tests were conducted at Hastings, Florida, except for the 9-week test which was made in Gainesville.

Results: Although both strips and vials containing Z-9-DDA remained effective at 4 weeks, both were ineffective after 9 weeks (Table 1). Much variation in catch occurred during the 9-week test, especially between replications of the new vials.

Plans: Strips and vials containing Z-9-DDA will be tested at weekly intervals between 4 and 9 weeks to determine exactly when each bait loses its effectiveness. Also, vials with an increased amount of pheromone will be evaluated, as well as the addition of an anti-oxidant to the pheromone.

Plastic strips aged outside, as well as strips aged at a constant temperature of ca. 80°F, will be examined chemically at 2-week intervals to determine when they may be expected to lose their attractiveness.



Table 1. Mean % capture of male fall armyworms on sticky traps baited with Herculite strips containing Z-9-DDA*

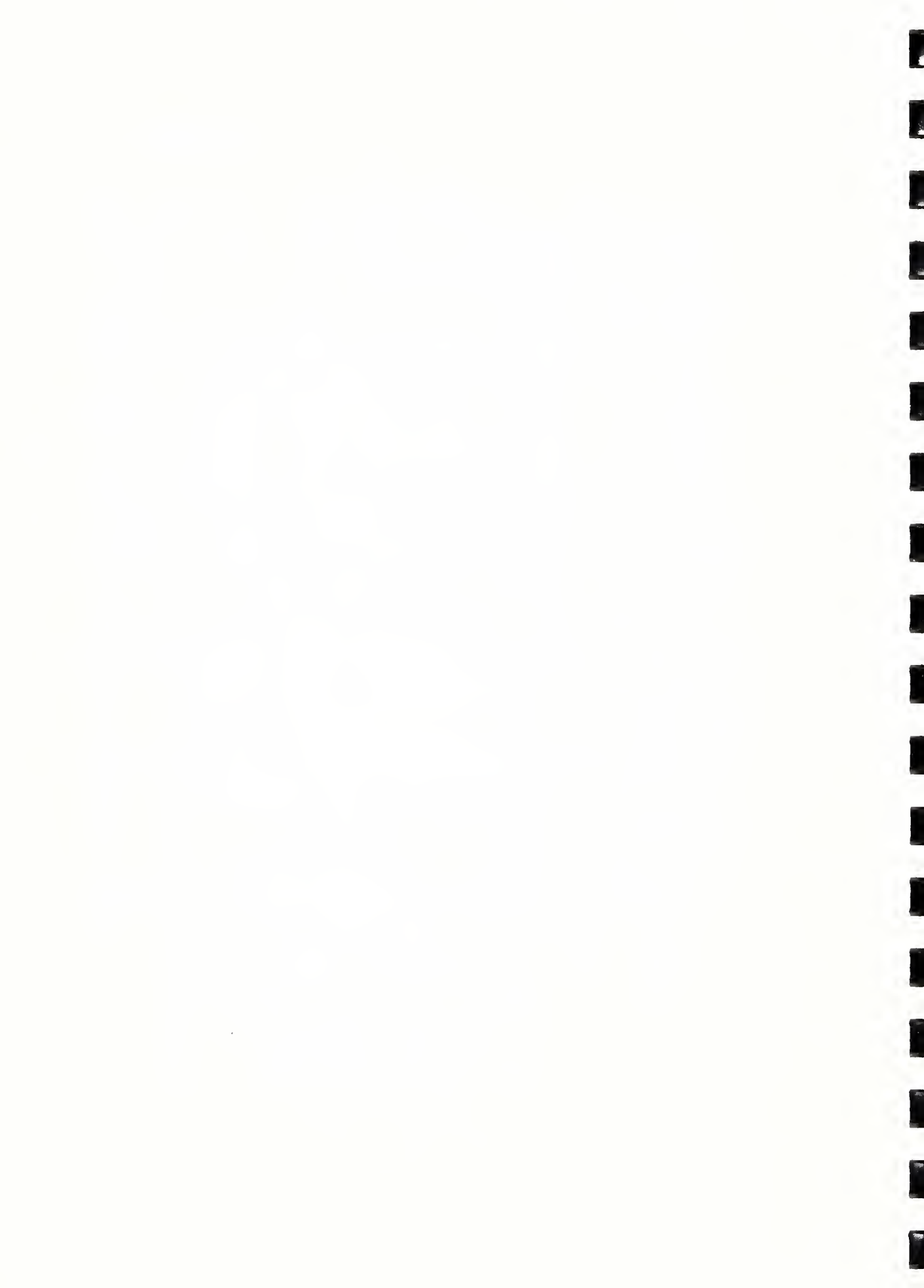
Week	24 mg/in. ² strip size (in. ²)		122 mg/in. ² strip size (in. ²)		Vial with 25 mg Z-9-DDA		
	Aged	New**	Aged	New†	Aged	New	
0 (9 reps)	18.3 c	11.3 b	6.4 a	20.5 c†	15 bc	10.8 ab	17.2 c
4. (9 reps)	15.9 a	15.3 a	17.3 a	19.4 a	15.8 a	16.4 a	16.4 a
9 (6 reps)	0.5 a	40.1 b	3.8 a	29.7 b	1.3 a	24.6 b	24.6 b

* Means in the same row followed by the same letter are homogeneous. (P=0.05, Duncan's multiple range test).

** 25 mg/in.²

† 127 mg/in.²

‡ In a subsequent test, 1 x 2 in. strips attracted almost identical numbers of moths as 1 x 1 in. strips.



Laboratory-reared Fall Armyworm Females vs.
Wild Females as Bait in Sticky Traps

F. C. Tingle and E. R. Mitchell

Purpose: To determine if our laboratory-reared fall armyworm, Spodoptera frugiperda, females are as effective as wild females when used as bait in pheromone sticky traps; also to determine if females with wings clipped to prevent flying attract equal numbers of males.

Methods: Large fall armyworm larvae (4th and 5th instar) collected from corn plants at Hastings, Florida, were held on artificial diet until adults emerged. Female moths (2-4 day old) were then used as bait in Pherocon® 1C sticky traps, as were females of the same age from the laboratory culture (3/trap). Each of the two treatments was replicated 6 times. This test was conducted June 9-10 at Hastings.

The test in which females with wings clipped were compared to normal females as baits in sticky traps was conducted July 21-22 at Gainesville. Three laboratory-reared females (2-4 day old) were caged in each trap; each treatment was replicated 8 times.

Results: Fall armyworm females from the laboratory culture and those collected from corn were equally as attractive as the lab females, capturing 52.7% of the total catch.

Females with wings clipped and normal females attracted equal numbers of males (50.6% and 49.4%, respectively).

Plans: Laboratory-reared insects used in field studies will be checked from time to time to determine their performance as compared to wild insects. This information will make data obtained in other tests more meaningful.



Maserlike Frequency From an
Ethyl Alcohol-Acetone Mixture

P. S. Callahan

Objective: To determine if maserlike frequencies are emitted by combinations of chemicals other than known insect pheromones and host plant attractants.

Method: A 95.5% pure solution of acetone was rubbed on a No. 3 steel insect pin. The acetone-primed insect pin was attached to a vibrating relay (modulator) driven by an electronic oscillator. The vibrator (modulator) frequency was set at 55 cps to the peak antenna vibration frequency of the cabbage looper. The pin was positioned in the source infrared beam of a Fourier Transform Spectrophotometer and vibrated in the beam. The vapors from (USP) absolute (pure) ethyl alcohol combusted in an alcohol lamp were directed across the pin in the source chamber and the infrared spectrum scanned at high resolution (1 cm^{-1}) across the region 700 cm^{-1} to 300 cm^{-1} .

Results: Extremely narrow band high intensity maserlike radiation was detected at 525 cm^{-1} ($19.05 \mu\text{m}$). The maserlike emission occurred where expected and exhibited characteristic side bands and slight rotation on either side of the strong emission frequency (Figure 1). "Close" side bands are evident at 545 cm^{-1} and 505 cm^{-1} , 20 cm^{-1} on either side of the main emission line and "far" side bands at 700 cm^{-1} and 350 cm^{-1} , 175 cm^{-1} on either side of the main emission line. Such side bands are characteristic of lased molecules and are categorical proof that maserlike frequencies emit from thin layer molecular mixtures modulated on a "sensilla like" surface.

Plans: Further work is in progress to test other chemicals that the cabbage looper responds to and which may emit far infrared maserlike emissions.



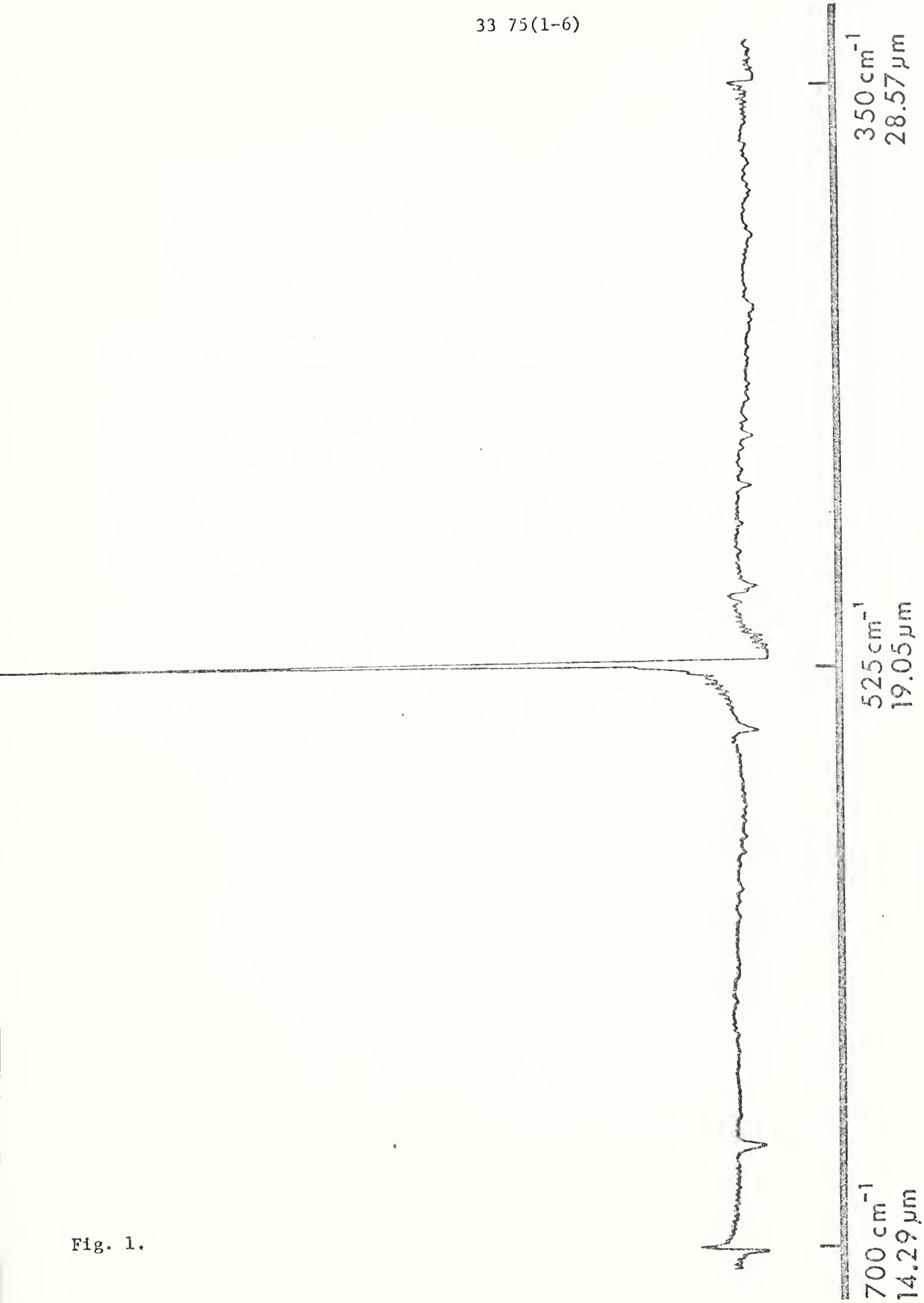


Fig. 1.



Response of Fall Armyworm Moths to 337 μm Laser Radiation

W. K. Turner, P. S. Callahan, and F. L. Lee

(Continuation of report 32 74(7-12))

Objective: To determine if fall armyworm, Spodoptera frugiperda, moths sense and respond to far infrared (IR) laser radiation with a wavelength of 337 μm .

Methods: Groups of 10 σ and 10 ρ laboratory reared fall armyworm moths were placed in a 10 cm by 1.2 m plexiglass tube and allowed several hours for adapting to the test environment. During the nocturnal phase the moths were irradiated with 337 μm radiation from a hydrogen cyanide laser by directing the laser beam along the longitudinal axis of the tube. Insect distribution was recorded before introducing the radiation and after 5, 15, 30, 45, and 60 minutes of irradiation. The test was replicated 4 times.

Results: The 337 μm laser radiation did not cause a bias in the distribution of insects toward either end of the test chamber. Visual observations did not reveal any apparent influence by the radiation.

Plans: A manuscript on the far IR work with cabbage looper, corn earworm, and fall armyworm moths will soon be submitted for review.



Sound Production by Conotrachelus nenuphar:
Associated Behavior

C. O. Calkins, J. C. Webb, H. R. Agee, and T. C. Carlysle

(Continuation of report 37 74(1-6))

Objective: To determine the behavior of plum curculios, particularly mating behavior, as it is associated with sound production.

Methods: Detailed observations were made of adult behavior in an anechoic chamber with the aid of microphones, speakers, and recorders.

Results: Additional sounds and behavior associated with the plum curculio were observed. Under certain conditions, individuals of either sex will attempt to mate with a member of its own sex. This elicits definite behavioral and sound patterns which appear to repel the aggressor. This behavior is not used when a beetle is being mounted by an individual of the opposite sex. Another sound is produced by the female as she partially mounts a male or crawls over his back. She then positions herself before the male and elevates her abdomen slightly and is receptive to mating.

Plans: Observations and sound recordings will continue to be made until all aspects of the phenomena are understood and described.



Sound Production by Conotrachelus nenuphar:
II. Analysis and Identification

J. C. Webb, C. O. Calkins, and T. C. Carlisle

(Continuation of report 37 74(1-6))

Objectives: To analyze and identify the sounds produced by adult plum curculios, and to establish frequency signatures for each sound.

Methods: An appropriate number of adult male and female beetles were placed in containers so as to elicit a given behavioral response. The sounds produced from each behavioral response were recorded on magnetic tape. The sounds will be analyzed and frequency signatures will be developed for each.

Results: The sounds of 4 behavioral responses were identified and recorded. These sounds were identified as stress, defensive, premating, and aggression. Other sounds have been observed but positive identification has not been made.

Plans: Observations and sound recordings will continue to be made of the identified sounds and attempts will be made to identify all observed sounds.

1950

Analysis of Screwworm Fly Flight Sounds

J. C. Webb, J. L. Goodenough^{1/} and J. C. Benner

Objective: Analyze and develop frequency signatures of flight sound of screwworm flies reared on different diets. The data obtained will be used in quality control programs.

Methods: Flies were reared on three diets and sound recordings were made from three age groups of those reared on each diet at the Screwworm Research Laboratory, Mission, Texas. Data tapes were shipped to this laboratory to be analyzed for frequency and amplitude content. The frequency signatures will be compared to determine if differences exist between treatments.

Results: The analyses of these tapes has begun and should be completed by September 1, 1975.

Plans: Compare the acoustical properties of the flight sounds of the screwworm flies and determine if a parameter exists that can be used to measure changes in physical conditions of the flies.

^{1/} Screwworm Research Laboratory, USDA, ARS, Mission, TX.



Sounds and Electrical Charges Produced by Electric Grid and
Black Light Traps that Repel and/or Attract Insects

J. M. Stanley, H. R. Agee, and J. C. Webb

(Continuation of report 38 73(7-12))

Objectives: To determine which stimuli (sounds, electromagnetic, and electrostatic) produced by electric grid and black light traps affect the behavior of insects. Determine the trapping units most attractive and least repellent to better survey and control insects.

Methods: Acoustic sense cell response of corn earworm, Heliothis zea, cabbage looper, Trichoplusia ni, fall armyworm, Spodoptera frugiperda, and beet armyworm, Spodoptera exigua, will be determined by presenting sound stimuli produced by various trap units to determine the maximum distance that the acoustic receptors can detect the sound of arcing and non-arcing grid traps and black light traps. The sounds will be recorded and analyzed for frequency, pulse duration, pulse rate, and sound level. The active components of the sounds will be determined physiologically and bioassayed. Similar physiological and bioassay methods will be used to evaluate the effect of electromagnetic and electrostatic charges on corn earworms, cabbage loopers, fall armyworms, and beet armyworms.

Results: Recordings have been made of the arcing caused by corn earworm, cabbage looper, fall armyworm, and beet armyworm moths across the 1/2 in. electrode spacing of grids. This has also been done for mechanically produced areas across known gaps on the grids. These data were taken when supplying power to the grid from each of three transformers having 4, 10, and 30 ma. secondary current ratings, respectively.

Plans: Data will be analyzed and prepared insect specimens will be electrophysiologically treated to determine their response.



Attraction of Fall and Beet Armyworm to Electromagnetic Energy

J. M. Stanley, E. R. Mitchell, and H. B. Agee

(Continuation of report 43 74(1-6))

Objective: To determine the relative attraction of fall armyworm, Spodoptera frugiperda (J. E. Smith), and beet armyworm, S. exigua (Hübner) males to the radiation from lamps that have elicited a significant electro-physiological response from these moths in laboratory tests.

Methods: Survey light traps equipped with black light (BL), blue, strontium blue, and green 15-watt fluorescent lamps were operated in field locations near Hastings, Florida, where large fall and beet armyworm populations existed. Traps were installed and rotated according to an approved statistical design.

Results: The superiority of the attractiveness of both species to the BL lamp was very obvious from the tabulation of data. Statistical analyses showed that the BL lamp was significantly better than the other three lamps for attracting both species. No difference was shown in the attractiveness of the blue, strontium blue, or green lamps to either species. The data on the beet armyworm confirmed the results obtained during the previous season.

Plans: From this work the BL lamp will be the lamp of choice of the four for use on light traps. Further study is desirable on correlation of electro-physiological data and field trapping results toward understanding insect attraction to electromagnetic energy.



Sensory Physiology of the Compound Eye
of Biosteres (Opus) longicaudatus and
O. oophilus

Herndon R. Agee, P. D. Greany, M. L. Park, and
D. L. Chambers

Objective: To determine the spectral sensitivity of the compound eyes of the hymenopterous fruit fly parasites, Biosteres longicaudatus and O. oophilus.

Methods: Electrophysiological techniques and instrumentation described in earlier reports were used to determine the spectral sensitivity of the compound eyes of these two species to monochromatic light thru the wavelength range 350 nm to 650 nm. The B. longicaudatus were reared on a laboratory colony of Caribbean fruit fly larvae and from field infested fruit. The parasites were tested for visual sensitivity when 1, 4, and 10 days old. The O. oophilus were collected as adults in Hawaii and were estimated to be 5-10 days old when tested. Each test group consisted of ten specimens (5 male, 5 female).

Results: Both species were most sensitive to 480 to 530 nm wavelengths, the blue-green region of the spectrum, and displayed a peak sensitivity at 500-510 nm. The four-day-old B. longicaudatus were slightly more sensitive over the entire spectrum than the 1-day-old insects, while the 10-day-old insects were less sensitive than the 1- or 4-day olds.

Plans: Color preferences of field populations of B. longicaudatus will be established using sticky panels.



Field Responses of the Caribbean Fruit Fly
and its Primary Parasites to Colored Surfaces

P. D. Greany, H. P. Agee, D. L. Chambers, and
A. K. Burditt^{1/}

Objective: To establish in the field the color preferences of the Caribbean fruit fly and of Biosteres (Obius) longicaudatus and Parachasma cereum, its primary parasites in south Florida.

Methods: Eight of 15 x 20 cm colored sticky panels have been suspended in known host trees in the Miami-Fomestead area. Each array includes one panel of each of the following colors: Gloss Black, Architectural White, Regal Blue, Green Thumb (dark green), Sharp Green (light green), Lacquer Lemon Yellow, Hot Orange, and Banner Red (Pittsburgh Paint Co., Pittsburgh, Pa.). Reflectance spectra were determined from 350 to 700 nm for each color using uncoated panels and panels coated with Tack Trap[®] (Animal Repellents Co., Griffin, Ga.). Position effects and color continuity effects were avoided by randomization. Traps are serviced weekly.

Results: Too few trials have been completed to allow comparisons at this point, but sufficient flies and parasites have been caught to establish that caribflies and the parasites can be attracted to and captured upon these panels. Electoretinogram studies conducted in this laboratory (compare related report by Agee et al.) indicated peak responsiveness by the eyes of the flies and of P. longicaudatus to light at about 510-530 nm. The Green Thumb panels showed a peak reflectance in the 510-530 nm region.

Plans: To continue these studies throughout the next several months or until a clear preference is established.

^{1/} Subtropical Horticulture Research Unit, USDA, ARS, Miami, Florida



Use of the Electroretinogram to Measure the
Quality of Fruit Fly Vision

Herndon L. Agee and Mary L. Park

Objective: To develop an efficient method to measure a quality parameter of an insect that could be used in rearing programs to compare the reared insects to the field counterpart.

Methods: The electroretinogram was selected to measure the visual sensitivity of Caribbean fruit flies reared at 3 laboratories on 5 artificial diets. The ERGs of these flies were compared to those of flies reared from field infested-fruit.

Results: Wide variations were noted in the visual sensitivity between flies reared on artificial diets and those reared on fruit, the latter were at least 10 times more sensitive than certain artificial diet-reared groups. As ERGs can be obtained 3-4 days before the Caribbean fruit fly is sexually mature, the use of the visually "substandard" fly in behavioral studies or field releases could be avoided.

Plans: This work has stimulated further studies on the effects of nutrition and handling procedures on the vision and behavior of the Caribbean fruit fly in the laboratory and under field conditions.



Morphology of the Antenna of Trichoplusia ni

H. S. Mayer and T. C. Carlisle

Objective: To study the morphology of the antenna of male and female T. ni in detail.

Methods: The methodology will be varied and most techniques associated with light and electron microscopes will be used. For the beginning work reported here, the antennae were cleared in KOH and mounted between two microscope cover slips. The various sensilla were then counted on every fifth segment with a transmission light microscope.

Results: To date the Type I and Type III sensilla trichodea have been counted on the male antenna (terminology that of Jefferson, R. N., et al., Ann. Entomol. Soc. Am. 63: 1227 (1970)). The distribution and total numbers of Type I and III sensilla trichodea obtained by polynomial regression analysis reveal that each male antenna has a complement of 4265 Type I and 1225 Type III sensilla trichodea.

Plans: This work will continue, primarily toward measuring the antenna and sensilla as well as to characterize them morphologically.



Histology of the Antenna of
Trichoplusia ni

T. C. Carlisle and M. S. Mayer

Objective: To study the innervation pathways of the antennae sensilla of male and female T. ni.

Methods: Thin sections in the range of 0.5 to 1.0 μm will be made from epox embedded antenna using a sorval ultra-microtome. Sections will be stained using a technique developed by Carlisle and Roppel. Information gathered from this technique will lead to an in-depth study using transmission electron microscopy.

Plans: This work will continue until nerve pathways can be traced from the sensilla on the antenna to the brain.



Correlations of the EAG of Trichoplusia ni to
Enzymic Pheromone Hydrolysis

M. S. Mayer

(Extension of report 39 74(7-12))

Objectives: To determine if the EAG decay rate following stimulation by various isomers and analogs of the pheromone can be correlated with the rate of in vivo hydrolysis.

Methods: EAGs were obtained in the usual way using Ag-AgCl glass capillary electrodes. The EAGs were recorded on an X-Y recorder and the analog waveforms converted to digital at 0.1-sec intervals. The decay rates of the EAGs were then analyzed by ordinary digital techniques and regressions were fitted by regression analysis. The theoretical basis of this work is found in: Kaissling, K.-E., (1969) In: Olfaction and Taste III, pp 52-70, Rockefeller Univ. Press, New York.

Results: The decay times of EAGs following stimulation with 10 replications of 6 concentrations of pheromone were obtained. The decay rate was exponential with an average T 1/2 of 0.57 sec. The decay times of EAGs following stimulation with 3 replications of 2 concentrations of (Z)-5-dodecen-1-ol acetate resulted in an average T 1/2 of 0.29 sec. These decay times resulted in theoretical enzyme turnover numbers of 75 for the pheromone and 54 for the isomer. The ratio of these two turnover nos. is $54/75 = 0.72$. This ratio corresponds very closely to the ratio of in vivo enzyme hydrolysis of these 2 compounds which was 0.6.

Thus, with only 2 chemicals yet compared the decay rate of the EAG seems to be correlated with the rate of in vivo enzyme hydrolysis on the antenna (Mayer, M.S., *Experientia* 31:452 (1975)). This lends credence to the hypothesis that enzymic hydrolysis of pheromone is at least a part of the transducing process.

Plans: To continue the work with 5 more isomers and analogs of the pheromone to obtain further correlations with in vivo enzyme hydrolysis.



Comparative Life Histories of "Wild Type" and "Colonized"
Caribbean Fruit Flies

N. C. Leppla, M. D. Huettel, D. L. Chambers, and W. K. Turner

(Continuation of report 56 74(1-6), 43 & 54 74(7-12))

Objective: To compare the development of each stage of the life cycle of a wild type population of *Anastrepha suspensa* (Loew) that was adapting to colonization with an established laboratory strain.

Methods: The inbred reference laboratory colony was compared with the newly colonized wild strain by determining the relative mating frequency, fecundity, egg viability, duration of the larval and pupal stages, pupal weights, and yields of pupae and adults (by sex) for both populations during each generation. Also, as an overall index of adult vigor, the amount of expired CO₂ was measured from equivalent populations of 50 pairs.

Results: By the F₅ generation, wild type flies were essentially the same size as the colonized insects, but they consistently oviposited nearly 40% fewer eggs and required 6-7 extra days to complete the generation. However, mean mating frequency, egg viability, pupal yield and weight, and adult emergence were statistically equivalent (Table 1). Average values recorded for these parameters were $0.425 \pm 0.06\%$, $55.61 \pm 23.53\%$, $10.25 \pm 7.50\%$, 0.0127 ± 0.0017 mg/pupa and $94.17 \pm 2.95\%$, respectively. CO₂ production was monitored simultaneously from wild and laboratory flies of the F₂, F₃, and F₅ generations during days 12 or 13 postemergence. During the photophase (active phase), ca. 20% more CO₂ was evolved by the wild flies than by the laboratory strain (Fig. 1).

Patterns of CO₂ output were generally similar; however, the total quantity varied between generations of both populations. Even though statistical data varied greatly, increased adaptation to the laboratory environment was indicated for each successive generation of wild type flies. Trends in several parameters suggested genetic selection that favored flies with the capability of accepting confinement, artificial provisions, and other "unnatural" sensory input. Only those insects matured, mated, and survived to produce their eggs and accept the artificial oviposition substrate.

Plans: The comparative portion of this study has been completed and a manuscript is in preparation.



Table 1. Comparison of the life histories of wild type and colonized strains of Anastrepha suspensa, maintained in the laboratory.

Generation	Mean Percentage Difference in Parameters ^{a/}				
	Eggs Deposited	Eggs Hatched	Pupal Yields	Pupal Weights	Generation Time
P ₁	>50	<10	14.63-	2.94-	--
F ₁	>50	<10	<20	8.64-	15.79+
F ₂	58.38-	<10	<20	<10	15.79+
F ₃	68.52-	0.44+	1.73+	5.25-	15.79+
F ₄	39.66-	8.00-	<20	1.76+	18.42+
F ₅	37.22-	9.33-	13.42-	0.04+	15.79+

^{a/} + or - indicates wild relative to lab, entries without signs are estimates.



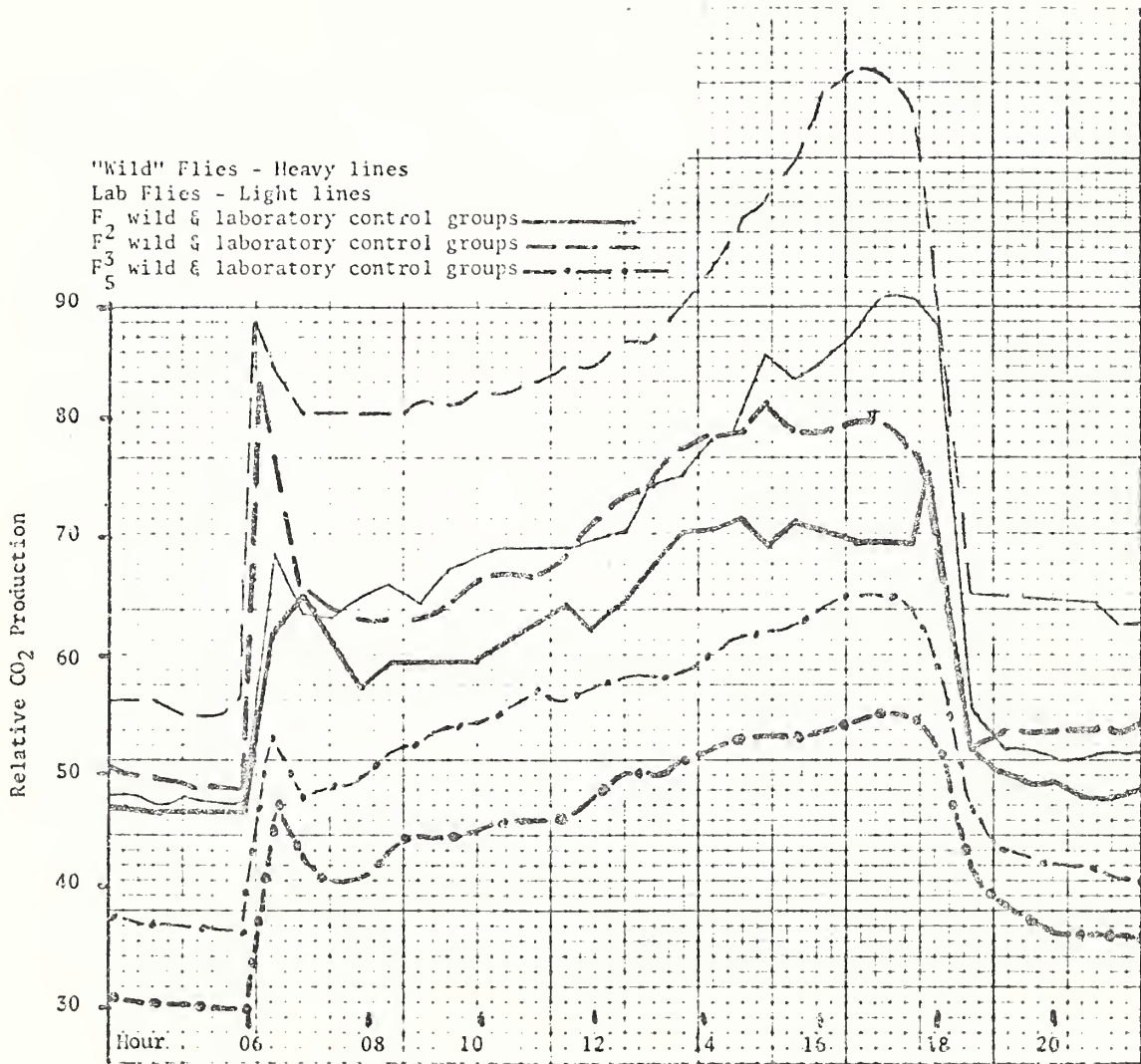


Fig. 1 Relative CO₂ production by groups of 100, 12 or 13 day-old "wild" and laboratory Caribbean fruit flies.



Comparison of "Wild Type" and Colonized
Populations of Cabbage Loopers Reared in Field Cages

R. H. Guy and N. C. Leppla

Objective: To compare the life histories of field-collected and laboratory-reared cabbage loopers, Trichoplusia ni (Hübner), reared for successive generations on collard plants in 12 x 6 x 5 ft field cages.

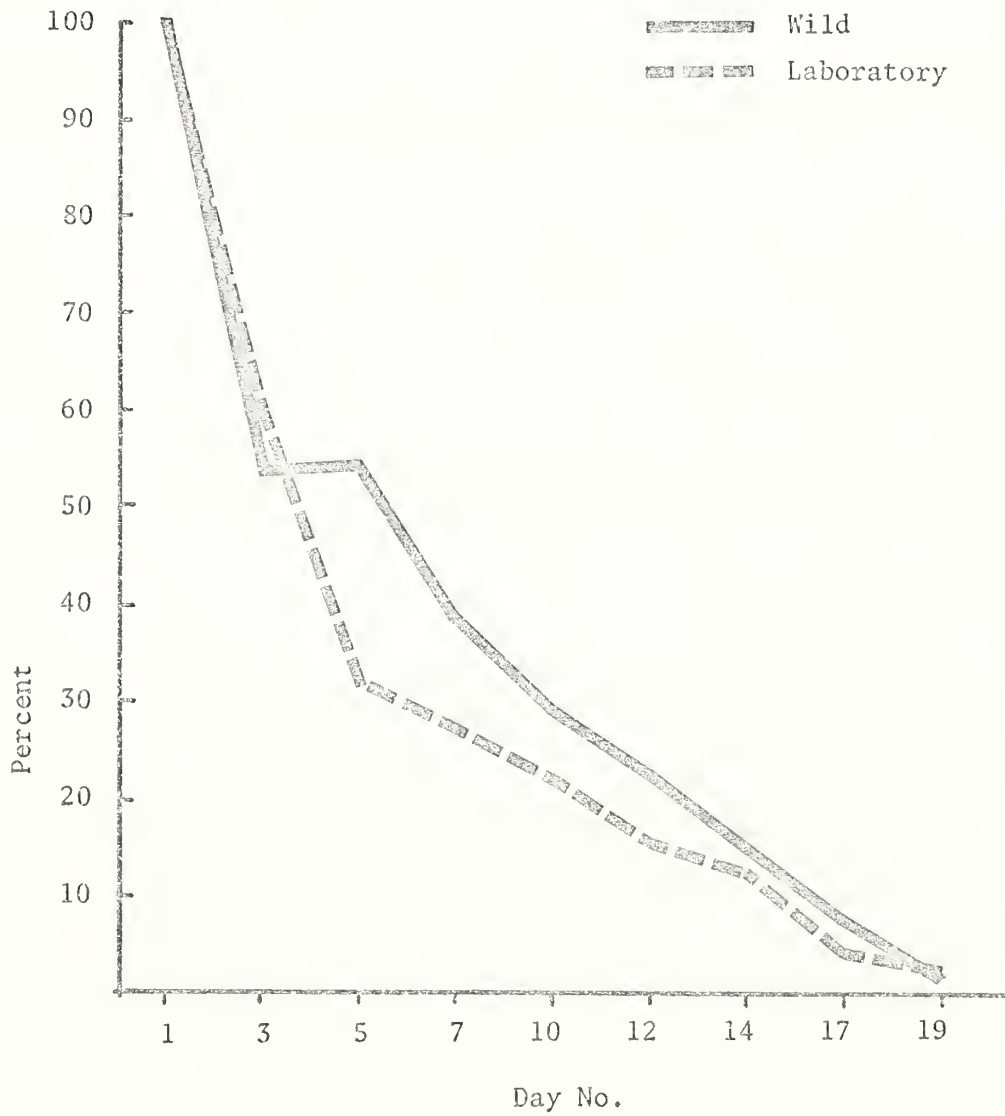
Methods: A colony of 10 wild-type moths was established by collecting and holding ca. 200 larvae encountered on cabbage and rutabaga crops in Hastings, Florida. Adults were caged in the laboratory on young collards, and 2 plants supporting ca. 200 eggs each were used to infest individual field cages. A canopy was provided by surrounding these 2 plants with 12 additional collards. The number of eggs deposited and their distribution on the plants, percentages of eggs that hatched, larval and pupal locations and survival (sex ratio and fecundity by subsample), and the mating frequency and fecundity of adults were recorded at appropriate intervals. Ambient temperature and relative humidity was also monitored in the cages. Identical procedures were repeated simultaneously for the laboratory colony, except the 400 eggs were applied to paper toweling and attached to the leaves.

Results: A complete life cycle (egg to egg) was obtained with a wild strain of Trichoplusia ni reared in the field. The percentage of larval survival for the "wild" and "lab" strains, respectively, was 29% and 23.5% after 10 days and 2.4% and 2.6% after 19 days (Fig. 1). Eighteen adults emerged from the wild strain and produced 231 viable eggs. The 13 adults that emerged from the laboratory strain produced no eggs.

Plans: This study will be continued as cool weather arrives and "wild type" cabbage loopers begin to emerge.



Figure 1. - Percentage of survival of wild and laboratory cabbage looper larvae reared in the field.





Comparison of the Daily Activity of Wild and
Laboratory Cabbage Looper Moths

W. K. Turner and N. C. Leppla

Objectives: To compare the circadian rhythms of activity of adult cabbage loopers taken from the field or one generation removed from the field (F_1) with a laboratory strain.

Methods: Cabbage looper moths collected from a walk-in light trap were compared with a laboratory strain by simultaneously monitoring the CO_2 production from each group. Five wild moths of each sex were compared with equal numbers of laboratory moths over a 3-day period, beginning with 2-day-old laboratory moths. Also, F_1 generation adults from pupae collected from rutabaga in the field and reared on live collard plants in the laboratory were compared with the laboratory strain by the same method. Four ♂ moths were used in each group. Other comparisons of activity were made using a vibration-sensitive actograph system with groups of 5 ♂ and 5 ♀ moths, the wild moths being from pupae collected from field cabbage.

Results: Activity patterns were similar for the wild and laboratory strains, but the wild moths produced ca. 30% more CO_2 . The moths taken directly from the field were more active during the diurnal phase than the laboratory moths. The actograph data gave similar results, and further showed that the differences in activities were related to frequency of flights.

Plans: More tests will be conducted as collection of field loopers permits.



Circadian Rhythms of Locomotion in Adult
Caribbean Fruit Flies

N. C. Leppla and W. K. Turner

Objective: To describe the temporal patterns and relative intensities of locomotion in aging populations of Anastrepha suspensa (Loew).

Methods: Pupae and flies were held in provisioned cages maintained at $26 \pm 1^\circ\text{C}$ and $80 \pm 10\%$ RH with a 14-hr photophase (310-750 nm, 17-29 ft-c); 1-2-day-old adults were transferred to monitored cages isolated in the same environment. Circadian rhythms of locomotion (impacts against the cages) were recorded from populations of 40 males, 40 females, or 20 pairs per cage by using a vibration-sensitive actograph system.

Results: Caribbean fruit flies are restrictively diurnal and exhibit 3 typical phases of development during their ca. 4-wk existence. Phase I, maturation of the integument and associated skeletal muscles requires 5-6 days. Flies ambulate and feed, but fly only intermittently. Peak flight, mating, feeding, and oviposition activities occur throughout the next 11-12 periods of phase II (days 7-18). The next 10+ periods of phase III (days 19-23) include increased mortality and a gradual decrease in the expenditure of energy. Isolated males or females and interacting pairs all produce similar patterns of locomotion.

Plans: This study has been concluded and a manuscript is in preparation.



Comparison of Almond Moth Activity Under
Continuous Weak Light and Continuous Darkness

W. K. Turner and D. W. Hagstrum

Objective: To compare the activity of almond moths, Cadra cautella (Walker), under continuous weak light and continuous darkness.

Methods: Groups of 5 ♂ and 5 ♀ laboratory-reared almond moths were maintained under either continuous light supplied by a Sears Cool-White[®] fluorescent lamp and neutrally filtered to ca. .015 fc or continuous darkness. Tests lasted for 72 hr and were monitored by recording CO₂ produced by the test insects. Data from the second 24 hr period of the tests were plotted and compared. Other similar tests were conducted with light levels of ca. .0001 fc and with weak UV radiation.

Results: Activity of almond moths was inhibited by continuous darkness as compared with continuous weak light. Under weak light, an activity rhythm occurred with the peak activity being 2-3 times the approximate constant level occurring under continuous darkness. Activity rhythms also occurred under the weaker white and weak UV radiation.

Plans: A study has been initiated to compare activities of almond moths under several nocturnal levels of visible and UV radiation.



Flight Ability of Anastrepha suspensa in the Laboratory

J. L. Sharp, J. C. Webb, H. R. Agee, and B. J. Smittle

Objective: To study the flight ability of Anastrepha suspensa in the laboratory.

Methods: Adult A. suspensa that had been provided with food and water were flown on the flight mill system for 3 hr at 25-26°C, 60-65% RH, and ca. 4300 lux of light intensity. Wild stock from rose apple were tested when they were 14 days old; treated adults (irradiated with 10 kr as pupae 2 days before adult eclosion) were flown when they were 1, 5, or 10 days old; laboratory colonized flies were flown when they were 1, 3, 5, 7, 9, 11, 13, or 15 days old. Tests with wild flies have been repeated 2 times; those tests with lab flies have been repeated 6 times. Flies were not allowed to feed or drink during tests and were discarded after tests.

Results: Figure 1 illustrates distances flown by laboratory-colonized flies. Table 1 presents results for other flight categories. Females fly better ($P < 0.01$) than males when both sexes are 3-4, 12-14, or 14-16 days old. Wild flies performed better than 14-day-old lab flies. Irradiation improved the distance flown by females 1 day old and irradiated females flew further than irradiated males when both were 10 days old.

Plans: Tests will be conducted at different RHs and temperatures. Previous reports numbered 44 74(7-12), 45 74(7-12), and 47 74(7-12) will be incorporated, and a manuscript will be prepared.



Table 1. Flight ability of Anastrepha suspensa males and females flown at different ages.

Age (day)	Flight categories ^{a/}	
	% Flying time	Overall velocity (m/min)
1	29-32	29-31
3	45-66	37-40
5	51-57	42-40
7	55-61	42-43
9	71-62	42-41
11	87-87	43-42
13	40-64	41-46
15	65-98	41-45

a/ ♂♂ - ♀♀



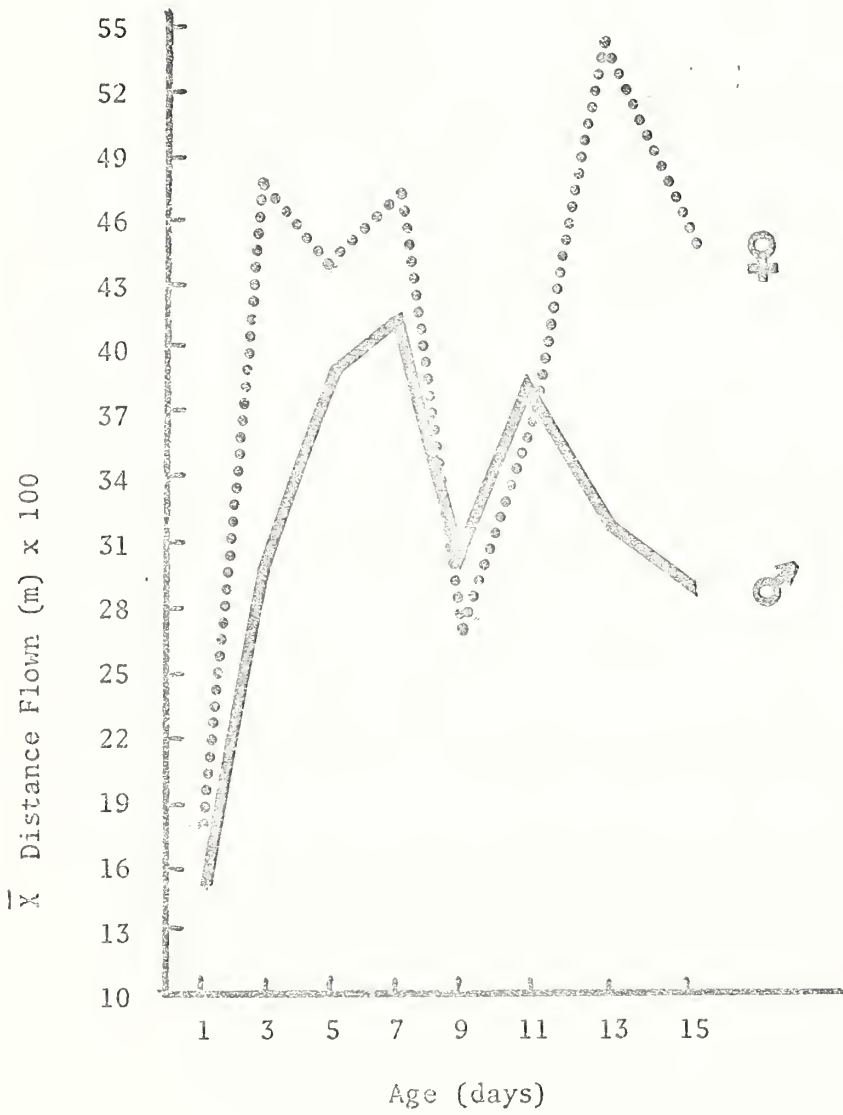


Fig. 1. Distances flown (m) by *A. suspensa* males and females flown at different ages.



Flight Metabolism of the Caribbean Fruit Fly
Anastrepha suspensa (Loew)

K. W. Vick and J. L. Sharp

Objective: To determine the kinds and amounts of energy reserves used by the Caribbean fruit fly during flight and the effect of gamma radiation and age on flight metabolism.

Methods: The amounts of sugar, lipid, and glycogen utilized during flight by irradiated (10 kr 2 days before adult eclosion) sterilized male flies allowed to fly for 3 hr were determined by chemical analysis. Also the effect of age on the consumption of energy reserves during flight by male and female flies was determined. Flight data including distance flown and average velocity were collected from each fly during the above experiments for correlation with the chemical data.

Results: Gamma radiation had little or no effect on energy reserves present at adult emergence on utilization of these reserves during flight. Energy reserve utilization followed the pattern seen in other flies and in mosquitoes with energy for flight being obtained from sugars and glycogen but not lipids. Data are still being collected to determine the effect of insect age on energy reserves and utilization.

Plans: The experiment on the effect of age on energy reserves will be finished. The data obtained will be analyzed to determine correlations between energy reserves and flight.



Activity of Stable Fly Adults at Reduced Temperatures

W. K. Turner and B. J. Smittle

Objective: To determine how low the temperature must be to prevent physical activity of stable fly, *Stomoxys calcitrans* (Linnaeus), adults. This objective is related to problems of handling and shipping of the flies.

Methods: Groups of 25 ♀ and 25 ♂ laboratory-reared stable flies were held under either constant or slowly decreasing temperatures for periods of time exceeding 24 hr. A 12-hr photophase was used. RH varied linearly with temperature, being ca. 35% at 22°C and 50% at 5°C. Activity was monitored by recording CO₂ produced by the flies and also with a vibration-sensitive actograph.

Results: CO₂ production indicated ca. a 100-fold decrease in metabolic rate and physical activity at 5°C compared with 22°C with the decrease approximately linear with temperature. Actograph data generally agreed with the CO₂ data. Even though metabolism was greatly reduced at 5°C, some physical activity occurred during the initial hours of each light phase. After 4 days at 5°C, the flies responded to an increase in temperature to 22°C with an activity level approaching what was considered normal.

Plans: More study is needed to determine the effects of extended cooling on the subsequent performance of the flies. However, none is planned.



Effect of Pre-Release Acclimation on Response of
Male and Female Pseudoplusia includens to BL-Traps Baited
with (Z)-7-dodecen-1-ol Acetate

J. L. Sharp, E. R. Mitchell, and J. James

Objective: To determine the effect of pre-release acclimation on response of male and female Pseudoplusia includens to BL-traps baited with (Z)-7-dodecen-1-ol acetate.

Methods: Four BL-traps baited with 0.05 ml of (Z)-7-dodecen-1-ol acetate were positioned in a square shape, each trap 200 ft from the center of the figure on the release site. Three groups of pupae (475 ♂ + 400 ♀/group) were marked with different colors of Day Glo[®] fluorescent powders. Two groups each were placed in 2 separate holding or release cages (sexes separated by a partition), provided with 10% sucrose, and allowed to emerge in the laboratory for 3 days at conditions of 13:11 L:D photoperiod, 60-65% RH, and ca. 23°C. The other group of pupae were held outside in a cage with 10% sucrose, and adults emerged there. Twenty-four hours before release time, one cage of males and females was transferred from the laboratory to outside and placed next to the cage. Then 2 hr before release time, the third cage of adults was removed from the laboratory conditions and set outside next to the 2 other cages. All 3 cages were protected from ant or bird predation. About 1 hr after sunset on the third night after initial emergence, the 3 cages were transferred to the release site where the moths were allowed to disperse from the cages. The next day, remaining pupae and moths were counted and from 69 to 85% of the moths dispersed. Capture records were taken for 5 consecutive days. Catch was examined under UV lights.

Results: The results from 1 replicate are presented in Table 1.

Plans: Four more replications will be carried out.



Table 1. Recapture of soybean looper moths (marked with fluorescent powders) acclimated outside or held in the laboratory.

Treatment	Days after release		
	% of marked adults recaptured ^{a/}		
	1	2	3
Emerged in lab and acclimated 2 hours	0.3-10.7	0.3-0.5	0-0
Emerged in lab and acclimated 24 hours	4.5- 3.8	0.7-1.2	0-0
Emerged outside	15.0- 3.3	1.1-0	0.4-0

^{a/} ♂ - ♀. No insects were captured on the 4th and 5th days after release.



Optimization of Parasitoid-Host Densities for
Biosteres (Opius) longicaudatus and its Host Anastrepha suspensa

T. R. Ashley, P. D. Greany, and D. L. Chambers

Objectives: To determine the optimal ratio of parasitoids to hosts in a standard ovipositional cage so as to obtain maximum progeny production and longevity.

Methods: Two experimental designs were used. In both designs the parasitoid densities were 25, 125, and 250 pairs per 25 cm³ Plexiglas® cage. In the first design 5-day-old parasitoids were exposed to 500 hosts for 24 hr. Fifty of these hosts were examined for ovipositional scars, and the remainder were permitted to complete development. In the second design each of the 3 parasitoid densities received ad libitum hosts. Every 24 hr host larvae were changed and parasitoid mortality recorded. This procedure continued for 14 days.

Results: The data from both experimental designs indicate that 25 pairs are too few parasitoids in the ovipositional cage. In the first experimental design, doubling the number of parasitoids from 125 to 250 increased effective parasitization by only 7%. Therefore, an exposure period of 24 hr for 5-day-old parasitoids at a density of 125 pairs/cage resulted in the greatest utilization of parasitoids and hosts. Preliminary analysis of the data from the second experimental design also indicates achieving maximum efficiency at 125 pairs/cage.

Plans: No further studies in this area are contemplated. A manuscript is being prepared.



Simple Devices for Transferring Moths

W. K. Turner

Objective: To design, fabricate, and test simple devices for transferring moths to solid containers, or containers with perforated bottoms.

Methods: The devices can best be visualized from the figures. The device shown in Fig. 1 is for conveying moths to airtight containers. Materials in one of our units includes plexiglass tubing for the inner tube (9" L, 2" OD, 1-1/2" ID), outer tube (4" L, 3" OD, 2-1/2" ID), and side port (2" L, 1-3/4" OD, 1-1/4" ID). The small holes drilled in the inner tube should have a combined area approximately equal to the inner tube cross-sectional area, and must be much smaller than the overall dimensions of the insect transferred. With the vacuum hose connected to the side port, and the receiving container attached, air is drawn through the inlet port and passes through the perforations to the outer tube. Insects are drawn through the inlet hose and because of their momentum pass through to the receiving container, while the scales and other small particles are drawn into the vacuum cleaner.

Vacuum cleaners usually have universal type motors, and thus can be operated at reduced voltages to obtain the desired speed. We use a home vacuum at 50 volts. For the reduced voltages variable transformers may be used.

The device shown in Fig. 2 has an 8" x 8" x 4" plenum with a vacuum hose port on one side and 1/4" holes drilled in the top. Guide studs are attached to the upper plenum surface to facilitate placement of the container. The lid (cover for container) section is hinged and adjustable in height. The metal pipe stand is held in place with setscrews. With the container in place and vacuum applied, suction is created at the port in the lid section. Insects are drawn through the pickup hose and gently lose speed in the large volume of the container. Dimensions of components will depend on the insect being conveyed. Smaller containers can be used by inserting adapter plates on top of the plenum and/or adjusting the height of the lid section.

Results: The devices have been used extensively for transferring moths to be used in experiments with no apparent injury to the moths.

Plans: A technical note has been prepared for review.



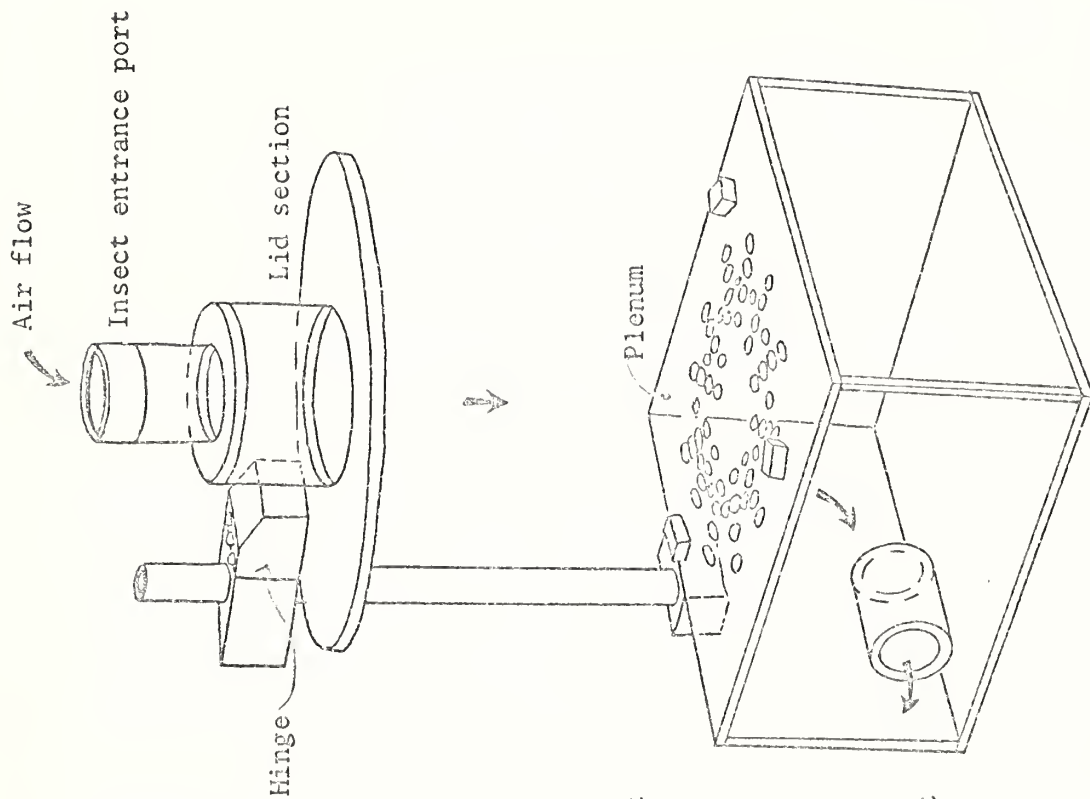


Fig. 2. Device to transfer moths to containers with perforated bottoms (container and hoses not shown).

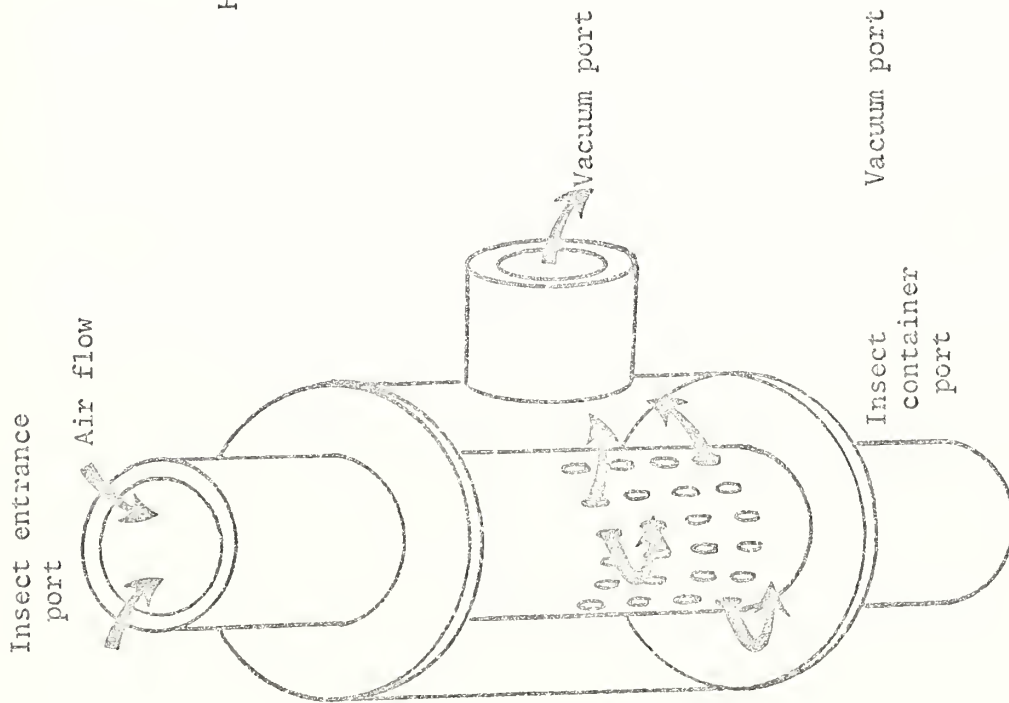


Fig. 1. Device to transfer moths to air-tight containers (container and hoses not shown).



Hibernation of the Plum Curculio,
Conotrachelus nenuphar (Herbst)

C. C. Calkins and M. McKoy

(Continuation of report 53 74(7-12))

Objective: To determine the overwintering mortality and the date of active emergence from hibernation quarters for plum curculios in Florida.

Methods: Virgin female plum curculios, reared under semi-outdoor conditions for 2 generations, were placed in 5-gal screened containers at the Tall Timbers Research Station and at Gainesville during November. Cans in each location were placed in well-protected wooded areas, beneath host trees, and in open areas. Equal numbers of males were added in December to determine if mating occurs during hibernation. Attempts to recover beetles and determine mortality were made from different containers at each location on December 12 (females only), January 16, February 6, and February 26.

Results: Mortality of adults was significantly higher at Tall Timbers Research Station than at Gainesville (Table 1). Weather conditions during December and January were considerably more moist at Tall Timbers and these wet conditions could have resulted in the high mortality. Several dead beetles were infected with what appeared to be a Beauvaria fungus. We were not able to determine if mating occurred during hibernation.

Plans: The occurrence of mating during hibernation will be determined using somewhat different techniques. Additional data on winter mortality also will be obtained.



Table 1. Percentage of beetles recovered alive from hibernation containers at Gainesville and Tall Timbers Research Station, Florida.

Date		Percentage of beetles alive	
		Gainesville	Tall Timbers
12-12-74	♀	44	32
1-16-75	♀	--	8
	♂	--	8
2-6-75	♀	56	32
	♂	20	12
2-26-75	♀	12	0
	♂	80	0
3-14-75	♀	16	--
	♂	88	--



Reciprocal Negative Interactions in Competitive Populations
of Cabbage and Soybean Loopers

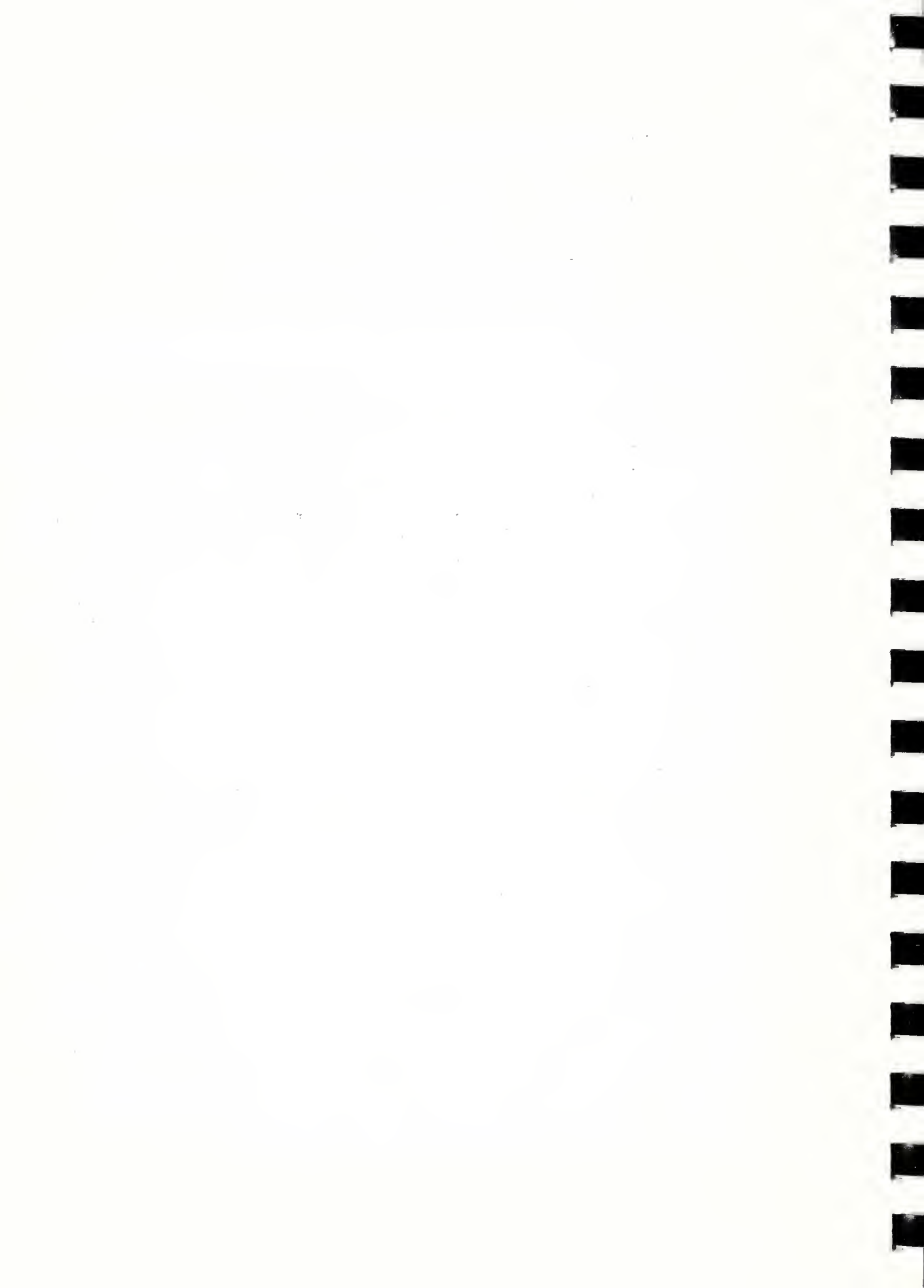
N. C. Leppla, W. K. Turner, E. R. Mitchell, W. J. Pons,
and F. L. Lee

(Continuation of report 60 74(7-12))

Objective: To determine the influences on cabbage looper (CL) and soybean looper (SBL) population dynamics of semiochemicals emitted by combinations of the two species maintained in the same environment but prevented from effecting direct contact.

Methods: Combinations of 30 pairs of CL or SBL moths per 46 x 38 x 31 cm plexiglass cage, or 15 pairs of each species on each side of a similar partitioned cage, were maintained at 27+1°C and 65+5% RH with a 14-hr photophase (complete visible spectrum, 17-29 ft-c, 1700-0700 hr). At 3-8 days of age, the insects were observed every 30 min from 0800 to 1500 hr, and the occurrence of mating, duration of copulation, and number of transferred spermatophores were recorded. In the same environment, 5094 cc cylindrical wire mesh oviposition cages, each containing 40 newly emerged pairs of a single species, were arranged in adjacent combinations of 4 CL, 4 SBL, or 2 CL + 2 SBL. The number of eggs deposited by these populations, and the percentage of those eggs that hatched, were recorded. Subsequently, 28 combinations of caged CL and SBL moths (10 insects of one sex, or 5 ♂ and 5 ♀/2344 cc wire actograph cage) were held under the same conditions, except for an 0800 to 2200 hr photophase. At 3-4 days of age, each species and sex was tested in isolation, arranged in 2:1 ratios of competitive congeneric populations combined in equal numbers of cages per treatment. Circadian rhythms of locomotor activity, total daily activity, and mating frequency were monitored.

Results: Exposure to congeneric populations produced similar effects for both species (data presented = CL ♂♀ isolated, CL ♂♀ + SBL ♂♀; SBL ♂♀ isolated, and SBL ♂♀ + CL ♂♀). Significant reductions occurred in the \bar{X} number of observed matings (61.50 + 0.50a, 32.67+4.44b; 58.00 +3.00a, 22.33+3.82b) and spermatophores transferred/♀ (73.00+15.00a, 40.50+4.49b; 70.00+2.30a, 26.17+3.53b). However, the \bar{X} duration of copulation (0.67+0.00, 0.67+0.03; 0.87+0.05, 0.79+0.05) and the number of spermatophores transferred/mated female (2.45+0.22, 2.67+0.29; 2.51+0.07, 2.13+0.24) were statistically equivalent (Fig. 1). Also, in smaller wire oviposition cages, there were comparable declines in the \bar{X} number of eggs deposited/female (340.27+29.48a, 173.53+13.59b; 397.10+28.64a, 21.20+19.56b), but mating was unimpaired and reductions in the % of eggs that hatched (82.00+3.66, 67.55+6.77; 50.37+7.15a, 9.85+3.33b) were essentially unilateral. Actographic analysis revealed significant, inhibitory influences on the average 24-hr locomotor activity of SBL ♂ by CL ♀, CL ♂♀ by SBL ♂, and of CL ♀ by SBL ♂♀ (Table 1).



The presence of CL ♂ or SBL ♂ also caused changed in the periodicity of activity recorded from congeneric females and pairs.

Plans: More definitive experiments will be conducted on the inhibitory influences of CL ♂ and SBL ♂ on congeneric females. The mechanism and target tissue must be identified in order to explain the observed reduction in fecundity and fertility of SBL populations exposed to CL populations.

Table 1. Relative 24-hr locomotor activity produced by various combinations of 3-4-day-old adult cabbage (CL) and soybean (SBL) loopers (n = 4 cages/treatment; 10 ♂, 10 ♀, or 5 pair/cage; AOV $P = 0.05^*$, 0.10^{**}).

Combination TMT X Standard	Relative activity	
	Combined $\bar{X} \pm SE$	Isolated $\bar{X} \pm SE$
CL ♂ X SBL ♀	138.75 \pm 63.19	645.00 \pm 175.98*
SBL ♂♀	537.50 \pm 331.15	648.75 \pm 262.31
SBL ♂	85.00 \pm 28.65	587.50 \pm 290.79**
SBL ♂ X CL ♂	1348.75 \pm 910.25	1481.25 \pm 622.73
CL ♀	698.75 \pm 120.53	1013.75 \pm 280.11
CL ♂♀	472.50 \pm 84.99	806.25 \pm 143.61**
CL ♂♀ X SBL ♀	448.75 \pm 171.85	645.00 \pm 174.98
SBL ♂♀	643.75 \pm 319.14	648.75 \pm 262.31
SBL ♂♀ X CL ♀	368.00 \pm 180.29	1013.75 \pm 280.11**

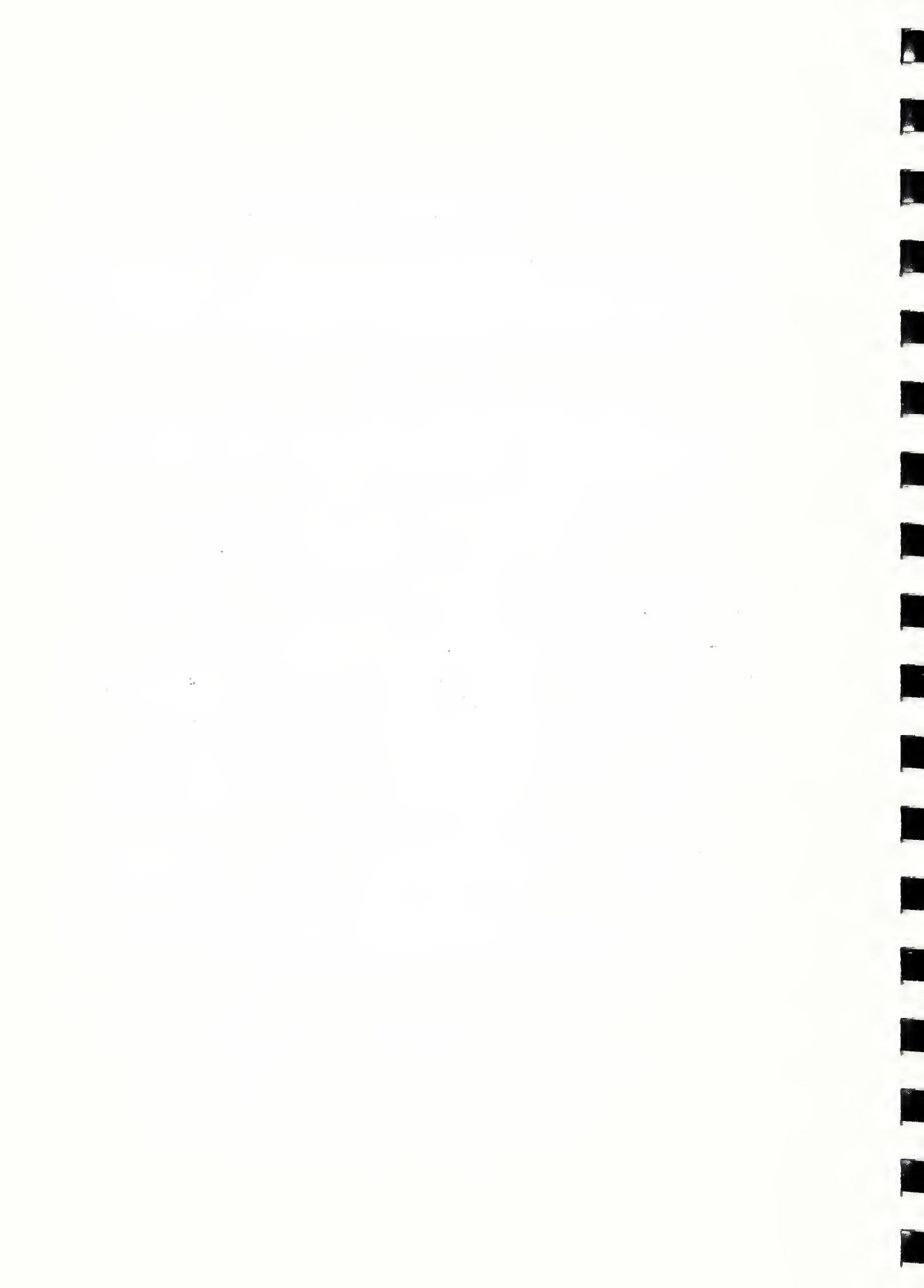
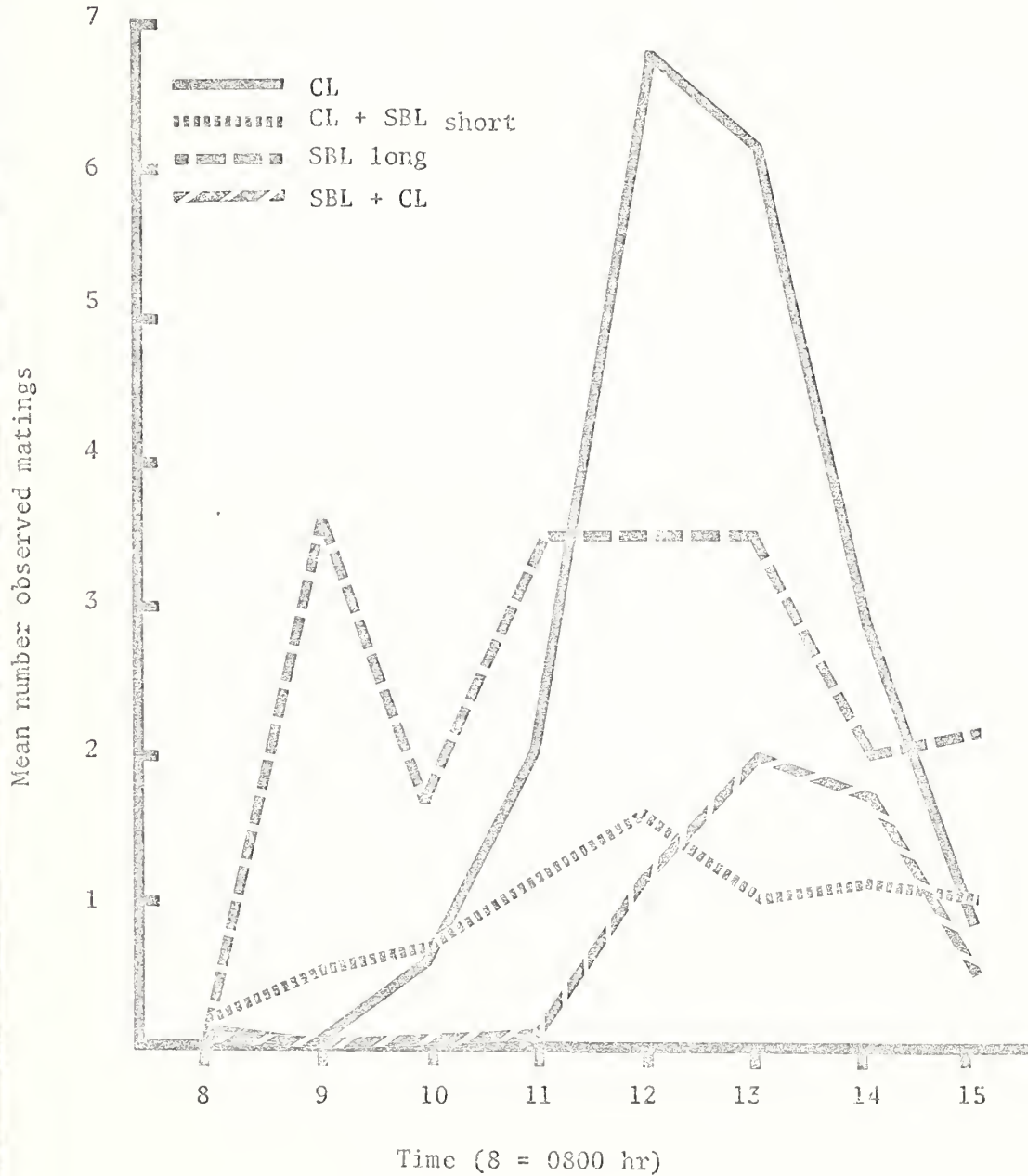


Figure 1. Matings of various combinations of 3-8 day old cabbage (CL) and soybean (SBL) loopers, observed during their peak 5-day reproductive period (n=6 cages/treatment; 30 pair CL or SBL, 15 pair CL + SBL/cage).





Trapping Equipment for the Almond Moth, Cadra cautella:
Evaluation of Trap Design

J. M. Stanley and D. W. Hagstrum

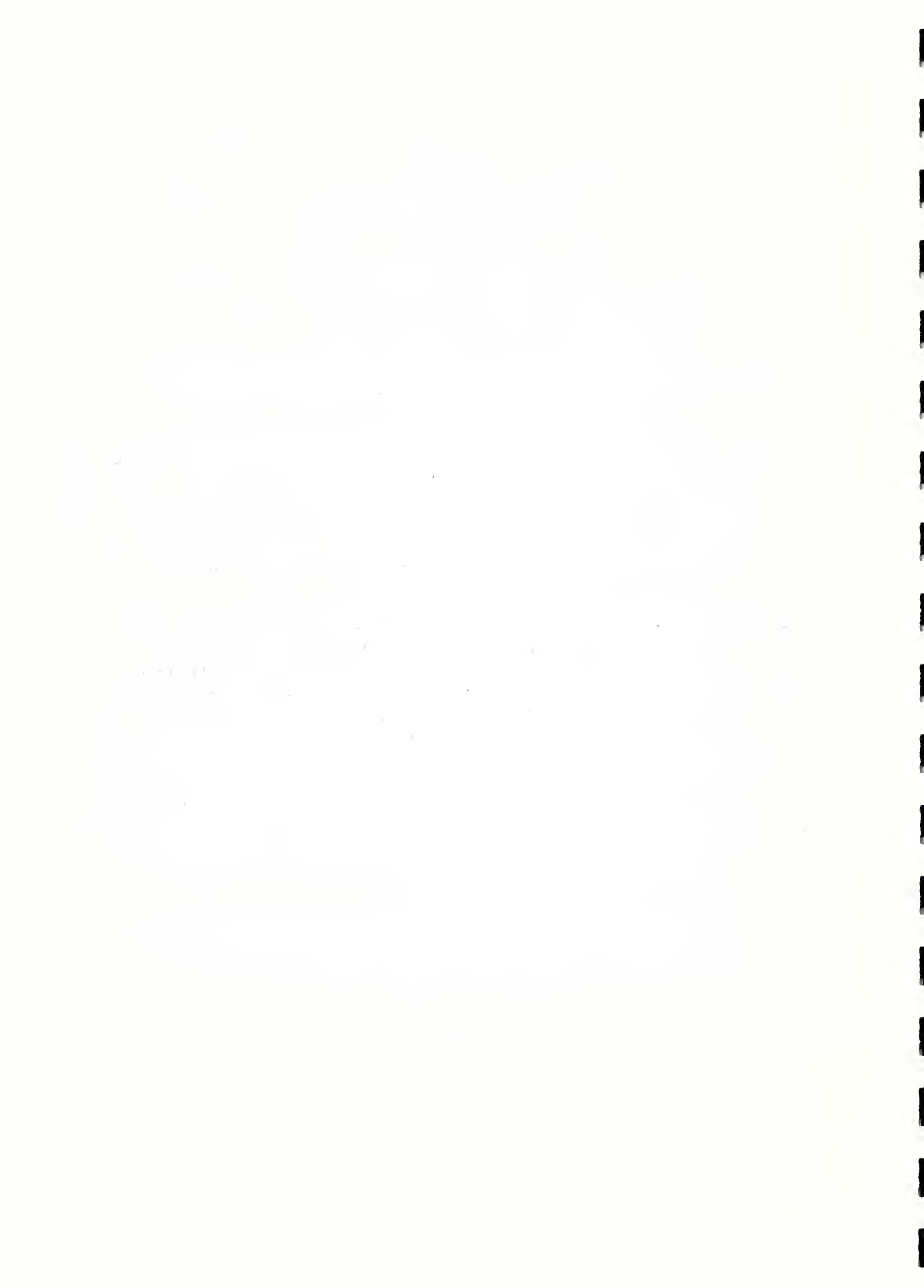
(Continuation of reports 64 74(1-6) and 58 74(7-12))

Objective: This study has been initiated to design an effective trap for collecting the adult of this insect in stored-product warehouses and to evaluate the potential of such equipment in insect management.

Methods: In 20' x 20' laboratory rooms fan traps were operated, one per room, with an energized 6-W BL lamp on the trap, an energized 6-W BL lamp in a remote corner of the room, or without energized BL lamps. These conditions were rotated and collections of known numbers of released almond moths tabulated. This same arrangement of traps with and without BL lamps was repeated in the 3'-wide aisles between stacks of citrus pulp in a storage warehouse. Traps were placed at ends of aisles against a wall.

Results: In the laboratory rooms the collections of male and female moths were greatest and similar when lamp was energized on the trap. The collection of male moths in the room with lamp in the remote corner was almost as great as with lamp on the trap. However, the collection of female moths lagged behind that of males when the trap was operated with lamp remote or without the lamp energized. The collection of moths in each instance correlated very well with temperature in the rooms, which ranged from 50 to 65°F. In the citrus pulp warehouse the collections of male and female moths were fairly similar under each of the three conditions. The total collections of moths in the trap with remote lamp position were similar to the collections in trap without lamp while the trap with BL lamp collected about half as many moths. In the laboratory tests using a TV camera to study moth activity the activity has been greater when moths are exposed to BL radiation.

Plans: Further study the conditions under which moths are trapped and the influence of the BL lamps on collections. Studies will be made under a higher temperature range in the laboratory rooms.



Solid State Photoswitch and Photoswitch Timer for Insect Survey Units

E. W. Hamilton

Battery-powered field insect survey units such as electrocutor grid traps or blacklight trans should have an automatic "off-on" mechanism to conserve battery power. The photoswitches described here require negligible battery power in the "off" state and perform the switching automatically to conserve energy.

Description.--The basic photoswitch (Fig. 1) requires less than 25 mamps in the "off" state with a 12V-DC power supply. Ca. 10 ma are needed to operate the relay when the photoswitch is on. Of course, additional power is required when the circuit is operated.

The circuit for night "turn-on" (Fig. 1a) operates as follows; In the daytime, the TIL 64 phototransistor has a resistance less than $R_1 + R_2$ resistors combined. Transistor Q_1 , which requires a positive input to operate, is biased off by the negative input. At dusk, the input to Q_1 becomes more positive until Q_1 is turned on and the relay is operated. The relay contacts in turn switch on a higher current to the equipment. The current-carrying capacity of the relay is relatively low (about 1 amp) so an output power transistor may be required for heavier power loads (see Fig. 2). Day "on", night "off" operation is similar (Fig. 1b) except for the placement of the phototransistor and resistor input combination.

A timing system has been included in the circuit shown in Fig. 2. In daylight, the 250- μ f capacitor is slowly charged to full power supply voltage because Q_1 is "on" and Q_2 is "off". In darkness, Q_1 is "off" and Q_2 is "on" effectively switches the capacitor to ground. Thus, Q_4 turns off, Q_3 is turned on; the relay is actuated; and power is supplied to the equipment being used. The value of R_1 must be higher than the grain to source resistance of Q_3 .

Timing operation begins as soon as Q_2 is on. As long as the 250- μ f capacitor maintains a charge sufficient to maintain Q_3 at a somewhat higher resistance than R_1 , the relay stays on. If no value of R_2 is inserted into the circuit, the relay will remain on ca. 15 h or until light is present at dawn. However, if one 5.1-megohm resistor is inserted at R_2 for each hour of operation desired, an on-time of between 1 and 15 h can be selected beginning at dusk. Several 5.1-megohm resistors can be wired in series at R_2 , and the number of hours of operation can be selected by switching in the number of resistors required.

Use.--With an electrocutor grid trap if R_2 is set for 3 hr, then the relay will turn on at dusk, and the electrocutor grid will operate for 3 hr and then be turned off. Battery power is thus conserved when all-night operation of the trap is not required. Transistor Q_5 is needed in this case to switch the high current requirements of the present design of electrocutor grid traps being used. The photo-timer will not reset and turn on until after it is exposed to daylight again.

Plans: A manuscript has been submitted to the journal for publication.

Figure 1. Photoswitch. (a) Night on (b) Day on.

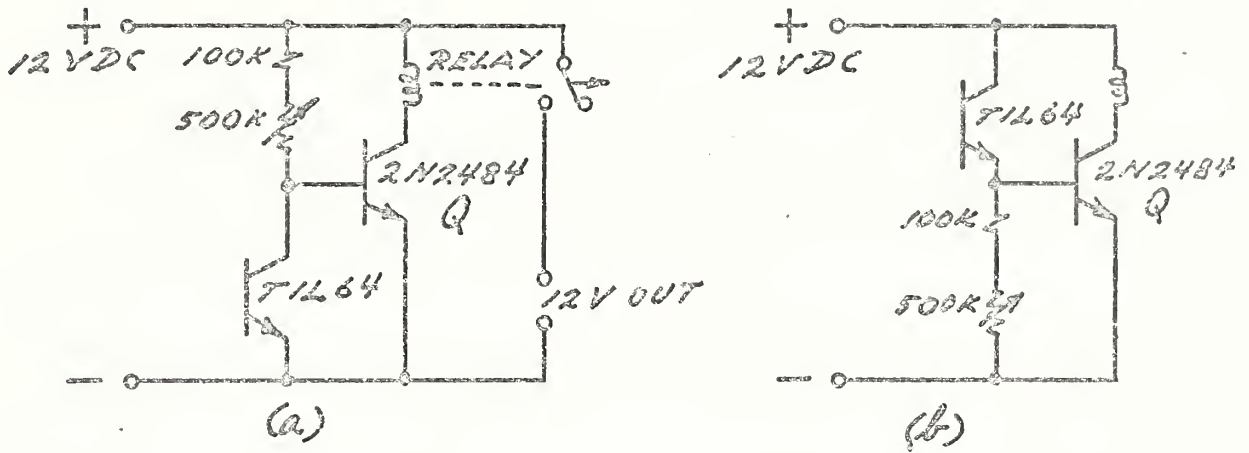
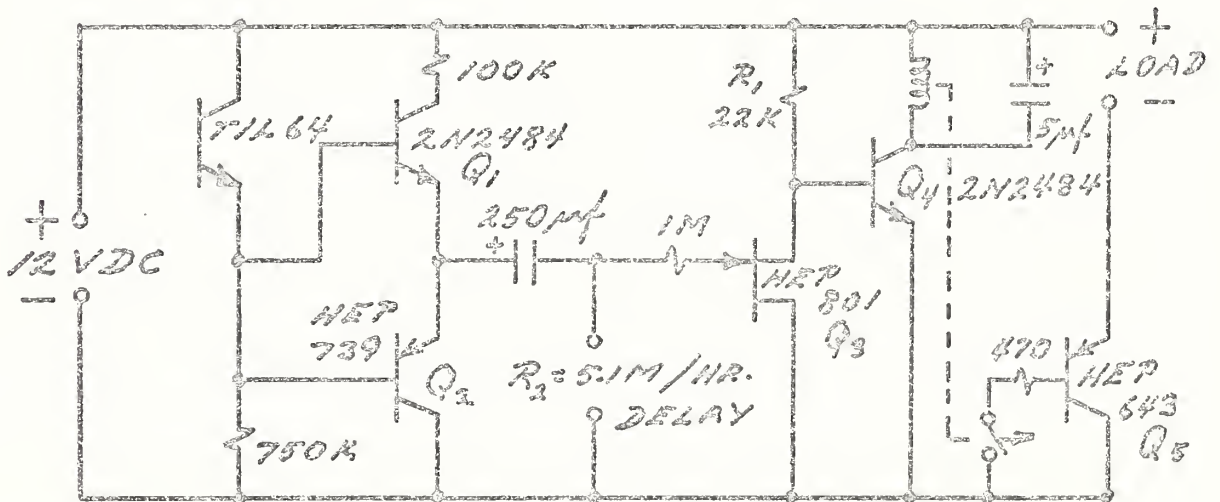


Figure 2. Solid state photoswitch-timer (night turn-on operation).





Collection of Beet Armyworm and
Southern Armyworm Larvae from Wild Hosts

F. C. Tingle, E. R. Mitchell, and T. P. Ashley

Objectives: To determine hosts of the beet armyworm, Spodoptera exigua, and the southern armyworm, S. eridania; also, to identify parasites emerging from collected larvae.

Methods: Catches of beet armyworm moths in survey traps baited with beet armyworm females and located in the Hastings, Fla., area indicated that this species was present in considerable numbers. Some southern armyworm moths were also captured in these traps. However, we had not observed any beet armyworm (or southern armyworm) larvae in the area, although we looked for these larvae while conducting other research.

On May 6, 1975, we observed Spodoptera spp. larvae feeding on pigweed (family: Amaranthaceae), a weed common in cultivated fields. Thereafter, weekly or biweekly collections of any larvae feeding on these plants were brought to the lab where larvae were reared to adults on artificial diet in individual 1-oz. cups. Head capsule measurements were made, and emerging parasites were kept for identification.

Results: Although almost all larvae collected from pigweed in May were beet armyworms, by early June almost 90% were southern armyworms. Larvae were difficult to find on June 30; however, of the Spodoptera spp. collected, all were southern armyworms. No Spodoptera larvae were found on pigweed on July 24. Only 1 beet armyworm larva and no southern armyworms were found on corn in the same field. Southern armyworm larvae were collected from 2 additional weed hosts.

A number of parasite species (at least 5) were obtained from both Spodoptera species and are in the process of being identified. Percent parasitism and number of larvae and adults collected or trapped are shown in Figures 1 and 2. We are waiting on identification of parasites to determine percent parasitism by each species.

Plans: Host relationships of Spodoptera spp. and their parasites will be investigated further. Efforts will be made to determine which parasite species would be best suited for use in population reduction of Spodoptera hosts.



NO. MOTHS / NIGHT

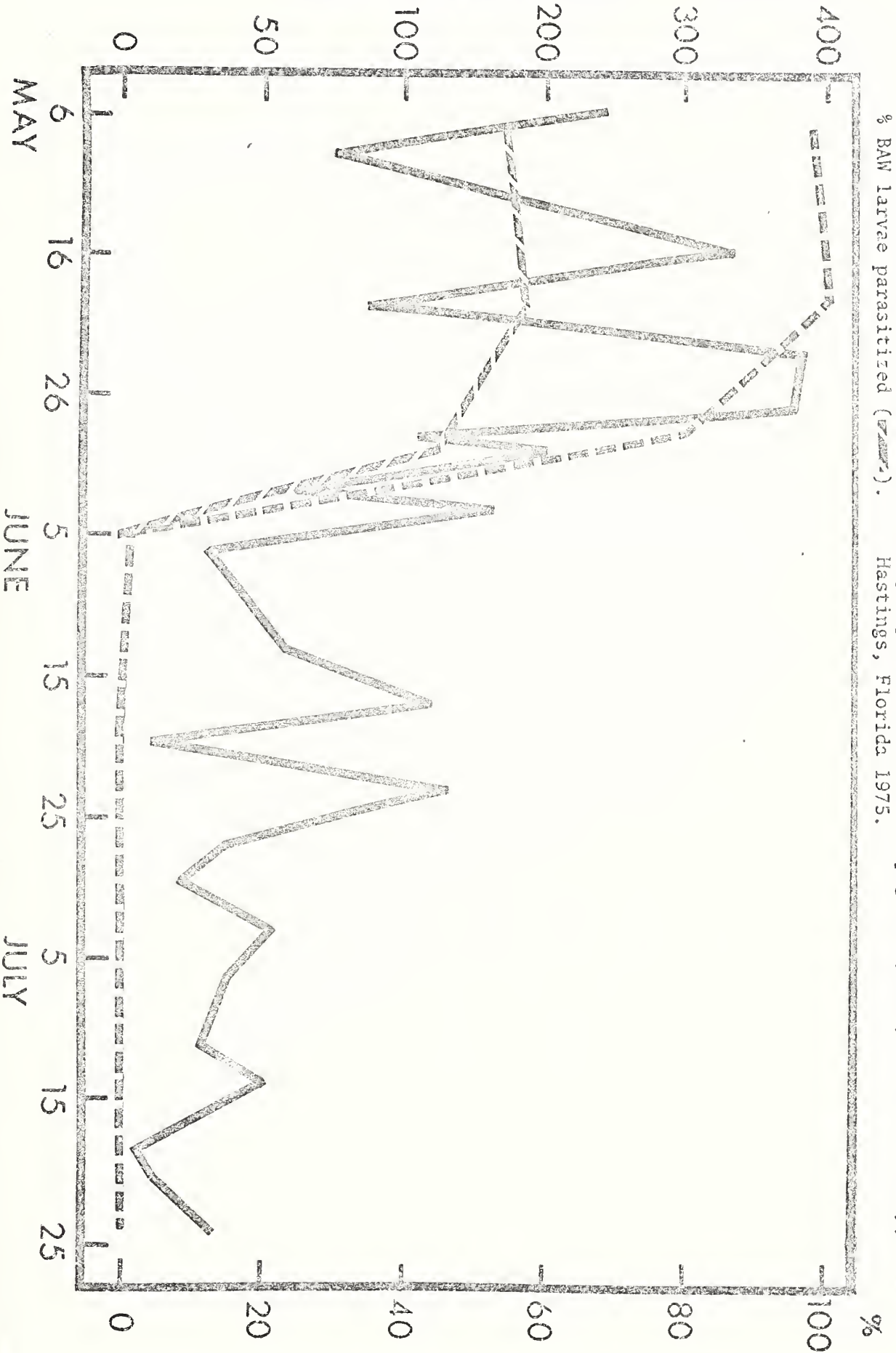
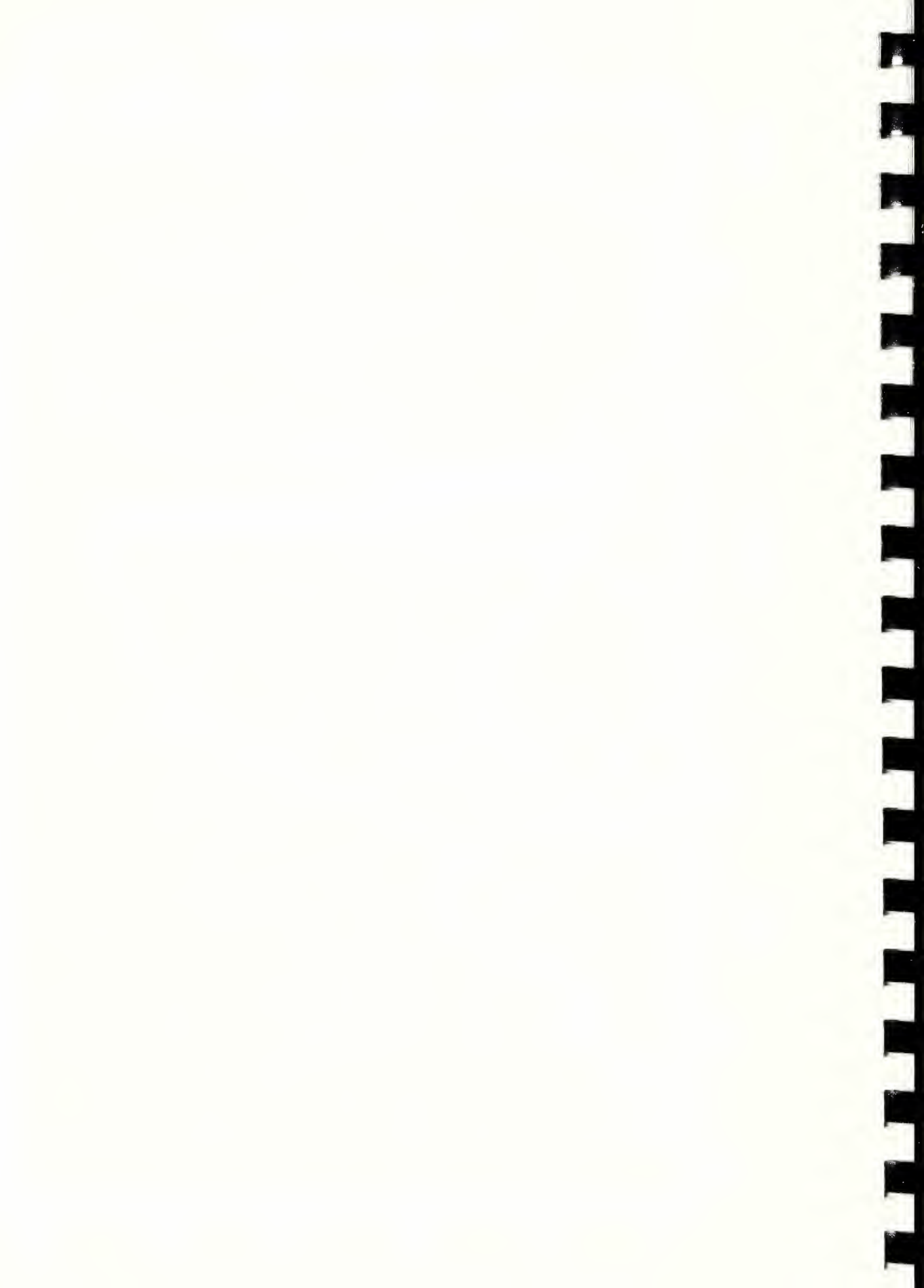


Figure 1. Number of beet armyworm (BAW) males captured in BAW female-baited electric grid trap (_____); % BAW larvae (-----) in collection of lepidopterous larvae from pigweed (family: Amaranthaceae); and % BAW larvae parasitized (-----). Hastings, Florida 1975.





Figure 2. Number of southern armyworm (SAW) males captured in beet armyworm female-baited electric grid trap (-----); % SAW larvae (dashed) in collection of lepidopterous larvae from pigweed (family: Amaranthaceae); and % SAW larvae parasitized (dash-dot). Hastings, Florida 1975.



Inheritance of Alleles at Four Allozyme Loci
in the Plum Curculio

H. D. Euettel and C. O. Calkins

Objective: To determine the mode of inheritance of alleles at the alcohol dehydrogenase (ADH), isocitrate dehydrogenase (IDH), phosphoglucomutase (PGM) and adenylate kinase (AdK) loci in the plum curculio.

Methods: Individual pair matings of the plum curculio were established. Green thinning apples were used as the oviposition and larval feeding substrate. Progeny were assayed by horizontal starch gel electrophoresis and enzyme-specific histochemical staining. Observed ratios of genotypes were compared statistically to ratios expected on the basis of simple condominant mendelian inheritance.

Results: The mode of inheritance has been determined for 2 alleles at the AdK and PGM loci and 3 alleles at the IDH and ADH loci. All are inherited as simple mendelian alleles, condominant in their phenotypes.

Plans: A manuscript describing the details of this study is being prepared.



Sperm Precedence in the Plum Curculio,
Conotrachelus nenuphar (Herbst)

H. Muettel, C. Calkins, and A. Hill

Objective: The rapid attainment of Hardy-Weinberg equilibrium in samples of progeny produced by mated and released plum curculios in the field (see 61 74(7-12)) indicated the operation of sperm precedence. This study was designed to determine the type of sperm precedence, if present.

Methods: Individual crosses were established using adults of two strains having different frequencies of the Idh-S allele. Females from these strains were mated sequentially to single males of the strains in appropriate combinations. Larval progeny were reared from green thinning apples. These larvae (24 crosses; >3000 larvae) were assayed and scored for genotype at the IDH, ADH, PGM and AdK loci using horizontal starch gel electrophoresis combined with staining.

Results: Sperm precedence in the plum curculio is of the type in which the second mating takes precedence over the first. It was detected in 74% of the crosses. The percentage of sperm from first matings used to fertilize eggs after sperm precedence occurred varied from 0 to about 15% in individual crosses.

Plans: Study will be terminated after completion of larval production from crosses.



Effect of Killing Agents on Capture of Heliothis virescens,
H. zea, and Spodoptera exigua in Pheromone Traps

F. C. Tingle and E. R. Mitchell

Objectives: To determine the effect of adding a killing agent to pheromone traps.

Methods: Male Heliothis virescens, H. zea, and Spodoptera exigua were confined in 3.8-liter paper cans lined with Pherocon[®] controlled release plastic strips containing 20% chlorpyrifos, diazinon, malathion, or naled. After a 1-, 2-, 5- or 10-min exposure, moths were transferred to untreated 0.5-liter paper cans. Mortality was recorded after 1, 2, and 24 hr. The effect of vapor action was also evaluated.

In a 2nd series of tests, Pherocon[®] 1C sticky traps treated with 25% diazinon, or lined with plastic strips containing diazinon, were tested against each species in 1.4 x 0.8 x 0.9 m screened cages located in the greenhouse. Virgin females were used as bait, and 50-140 male moths were released into each cage in the afternoon. Overnight capture of males was recorded from the untreated and treated traps, as well as those which had made contact with the trap and died elsewhere in the cage. Naled was not used because of possible mortality resulting from vapor action from overnight exposure.

Results: Higher mortality was obtained from contact with plastic strips containing diazinon than with the other chemicals tested (Table 1). Diazinon was tested for vapor action against all 3 species, and naled against H. virescens; no mortality resulted from 1-, 2-, 5-, or 10-min exposures to these chemicals.

Sticky traps treated with diazinon captured or killed considerably more moths than the untreated trap (Fig. 1). Few moths were killed when the trap surface was covered with the plastic strip containing diazinon.

Plans: These data are being reviewed for publication. Evaluation of pheromone traps containing killing agents is difficult in the field since most of the moths destroyed do not die on the traps. Additional research is planned using these methods to evaluate male annihilation and to determine trap efficiency with these and other insect species.

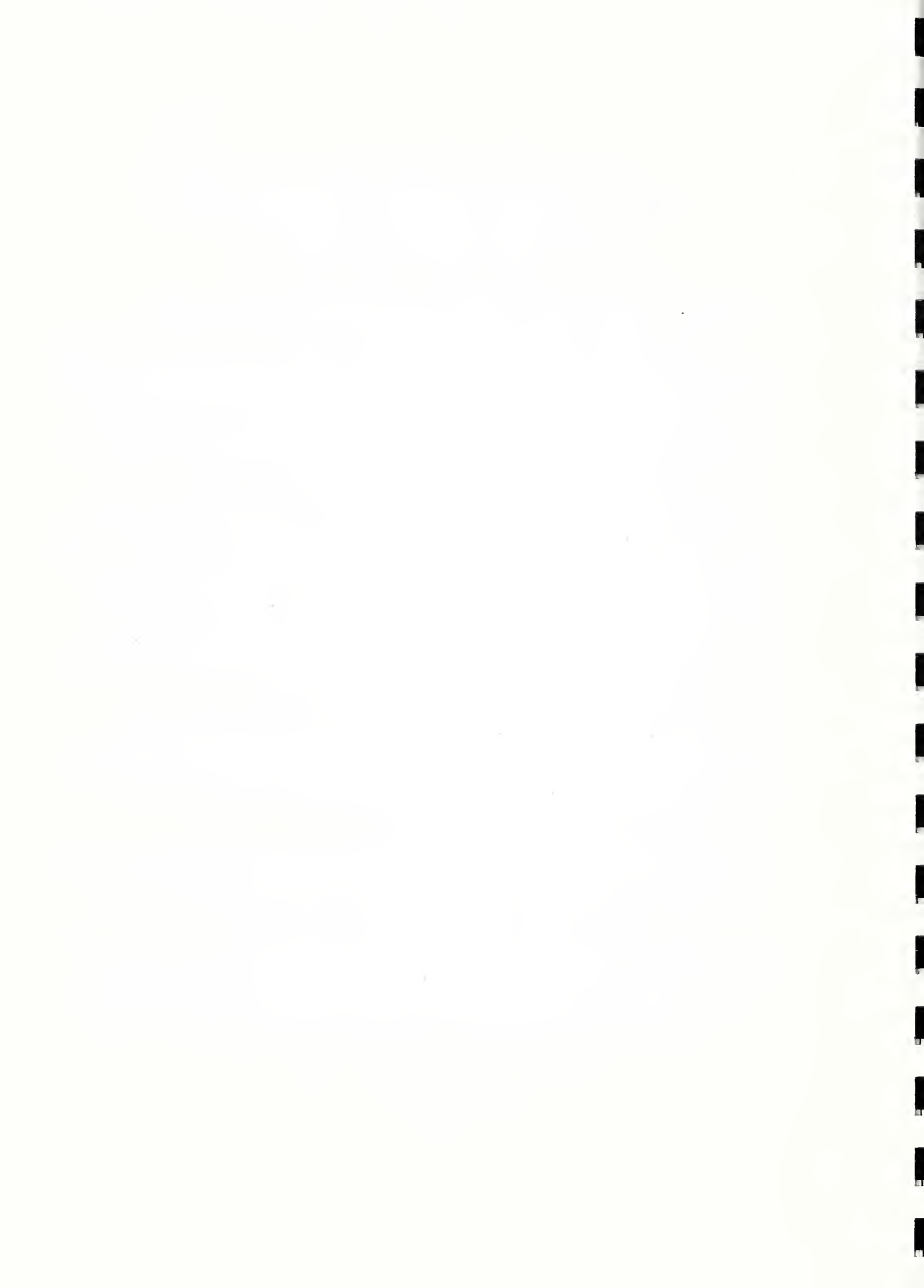


Table 1. Mean percent mortality of male tobacco budworms (TBW), corn earworms (CEW), and beet armyworms (BAW) after contact with indicated killing agent confined in 3.9-liter paper cans. 1975.

Killing agent ^{a/}	Exposure time (min)	% mortality 24 hr after exposure ^{b/}		
		TBW	CEW	BAW
Diazinon ^{c/}	1	37 c	96 a	
	2	22 cd	100 a	22 c
	5	83 a	100 a	37 bc
	10	100 a	100 a	80 a
Chlorpyrifos ^{c/}	2	0 d	30 bc	11 c
	5	0 d	40 b	10 c
	10	0 d	30 bc	53 b
Nalde ^{d/} (3 hr)	1	6 d	10 cd	5 c
	2	0 d	4 d	0 c
	5	75 ab	0 d	0 c
	10	100 a	7 d	0 c
Naled ^{d/} (24 hr)	1	0 d		
	2	10 d		
	5	61 b		
	10	100 a		
Naled ^{d/} (120 hr)	1	0 d		
	2	3 d		
	5	10 d	10 cd	7 c
	10	90 a	7 d	3 c
Control		0 d	0 d	0 c

a/ Each treatment replicated 3 times.

b/ Means in the same column followed by the same letter are homogeneous ($P = 0.05$ Duncan's multiple range test).

c/ Formulated in controlled release plastic sheets at 20% technical grade material by weight.

d/ Chemical (25.4% commercial grade by weight) brushed on cardboard; hours indicate elapsed time from treatment of cardboard until insects were exposed to treated surface.



\bar{x} % MORTALITY

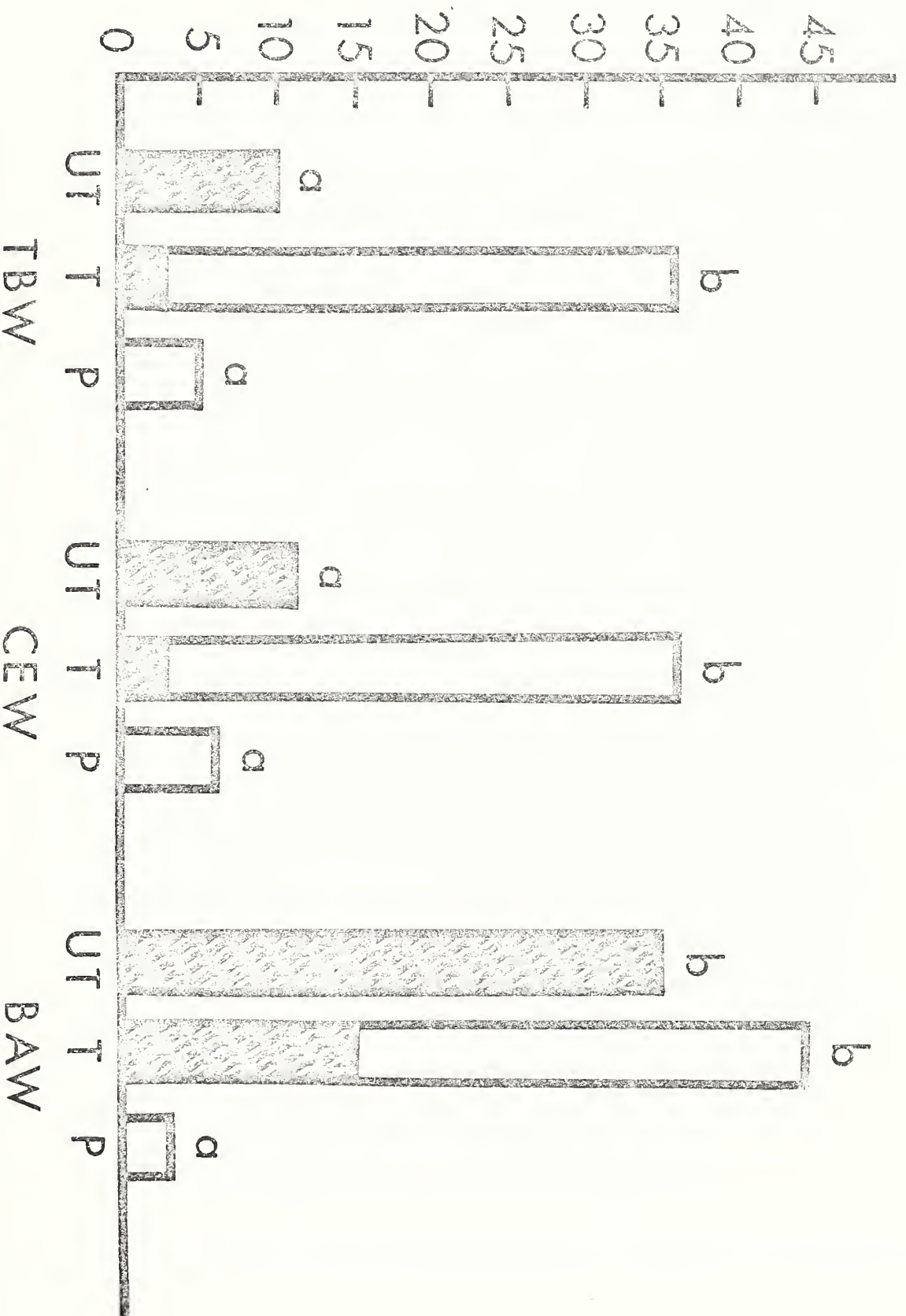


Figure 1. Mean percent of released (shaded area) or killed (unshaded area) tobacco budworm (TBW), corn earworm (CEW), and beet armyworm (BAW) moths captured in 1.4 X 0.8 X 0.9 m screened cages: Untreated (UT), treated with 25% diazinon (T), or sticky surface traps with controlled release plastic strip (P) containing 20% diazinon. Columns with the same letter are homogeneous (P=0.05 Duncan's multiple range test).

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- E. S. Miller, Clerk Steno.
- E. S. Turner, Clerk Steno.

MANUSCRIPT STATUS REPORT

September 16, 1975

NATIONAL TECHNICAL EDITOR REVIEW*

- Calkins, C. O., M. D. Huettel, and H. McKoy. Spatial and temporal distribution of oviposition by female plum curculios.
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