SEMIOCHEMICAL-MEDIATED LOCATION OF HOST-HABITAT BY *Apanteles carpatus* (SAY), A PARASITOID OF TINEID LARVAE

by

Stephen J. Takács

B.Sc., Simon Fraser University, 1994

THESIS SUBMITTED IN PARTIAL FULFILLMENT OF
THE REQUIREMENTS FOR THE DEGREE OF
MASTER OF PEST MANAGEMENT

in the Department
of
Biological Sciences

Simon Fraser University

August 1996

Frontispiece: Female *Apanteles carpatus* (Say) (Hymenoptera: Braconidae) ovipositing in a larva of the casemaking clothes moth, *Tinea pellionella* L. (Lepidoptera: Tineidae).

APPROVAL

Name: STEPHEN J. TAKACS

Degree: Master of Pest Management

Title of Thesis:

SEMIOCHEMICAL-MEDIATED LOCATION OF HOST-HABITAT BY *APANTELES CARPATUS* (SAY), A PARASITOID OF TINEID LARVAE.

Examining Committee:

Chair: Dr. M. Mackauer, Professor

Dr. G. Gries, Assistant Professor, Senior Supervisor
Department of Biological Sciences, SFU

Dr. J. Borden, Professor
Department of Biological Sciences, SFU

Dr. B. Roitberg, Professor
Department of Biological Sciences, SFU
Public Examiner

Date Approved: August 1, 1996

PARTIAL COPYRIGHT LICENSE

I hereby grant to Simon Fraser University the right to lend my thesis, project or extended essay (the title of which is shown below) to users of the Simon Fraser University Library, and to make partial or single copies only for such users or in response to a request from the library of any other university, or other educational institution, on its own behalf or for one of its users. I further agree that permission for multiple copying of this work for scholarly purposes may be granted by me or the Dean of Graduate Studies. It is understood that copying or publication of this work for financial gain shall not be allowed without my written permission.

Title of Thesis/Project/Extended Essay

SEMIO CHEMICAL-MEDIATED LOCATION OF HOST-HABITAT BY APANTELES CARPATUS (SAY), A PARASITOID OF TINEID LARVAE.

Author:
(signature)

STEPHEN J. TAKACS
(name)

1 August 1996
(date)

ABSTRACT

In Y-tube olfactometer bioassays, adult *Apanteles carpatus* (Say), were attracted to beaver or rabbit pelts infested with larvae of the casemaking clothes moth (CCM), *Tinea pellionella* L. Porapak Q-captured volatiles from a CCM-infested beaver pelt were also very attractive, whereas isolated CCM larvae or larval faeces were not. Coupled gas chromatographic-electroantennographic detection (GC-EAD) analysis of the Porapak Q volatile extract revealed two compounds that elicited responses by *A. carpatus* antennae. Coupled GC- mass spectrometry (MS) in electron impact and chemical ionization modes of these compounds indicated, and GC-MS and GC-EAD of authentic standards confirmed, that they were nonanal and geranylacetone. While each compound singly did not attract *A. carpatus,* a 1:1 blend of both compounds was as attractive as the volatile extract. Because these compounds are host-habitat derived, *A. carpatus* is a habitat- rather than host-specialist responding to kairomonal indicators of localized and specific habitats such as animal hair or feather. The tritrophic interaction between *A. carpatus*, its clothes moth hosts and their animal-derived habitats is similar to the well-studied relationship between parasitoids of insect herbivores and their host-plant habitats.

ACKNOWLEDGMENTS

I wish to express my appreciation to my senior supervisor Gerhard Gries for generous assistance and encouragement during the course of study and preparation of this thesis. I am grateful to the following members of the Simon Fraser University community: Regine Gries for sharing her technical expertise in electrophysiology, Manfred Mackauer and Steve Halford for valuable advice, Harold D. Pierce Jr. for assistance in volatile collections, Greg Owen for mass spectrometry, Les Wakida for glassware construction, Ron Long for photography and Elizabeth K. Carefoot for preparation of figures. M. Sharkey (Biosystematics Research Centre) confirmed the identity of *Apanteles carpatus* and Robin Todd, Insect Control and Research Inc., provided *Tineola bisselliella* when it was most needed. Finally, I thank my committee member, John H. Borden for encouragement and constructive criticism during the course of this study.

TABLE OF CONTENTS

LIST OF FIGURES

FIGURES **PAGES**

TABLE

PAGES

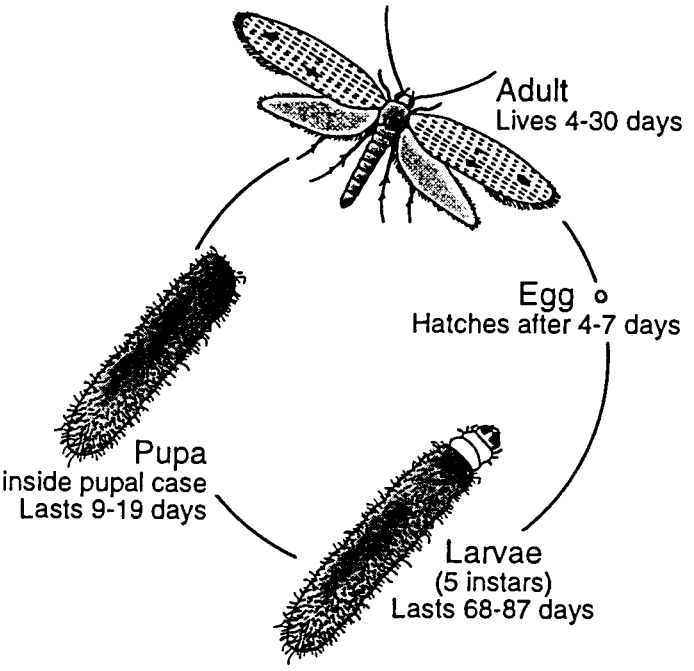
1.0. INTRODUCTION

Apanteles carpatus (Say) (Fallis, 1942; Rutz and Scoles, 1989) is a parthenogenetic parasitoid of larval clothes moths, such as the webbing clothes moth (WCM), *Tineola bisselliella* (Hum.), casemaking clothes moth (CCM), *Tinea pellionella* L., tapestry moth, *Trichophaga tapetzela* L., *Tinea fuscipunctella* (Haw.), *Phercoeca utrella* (Wlsm.), *Tinea despecta* Meyr., and *Tinea columbariella* (Wocke) (Krombein et al. 1979). Emergent *A. carpatus* adults are highly phototropic, resulting in emigration from the habitat and reduced likelihood of superparasitism. Foraging activities of parasitoids peak at an "optimal" temperature rather than light intensity (Key and Common, 1959). The parasitoids oviposit 1-100 eggs in a single host larva (Fig. 1). All eggs hatch but only one larva survives to the pupal stage (Fallis, 1942). Development of the endoparasitic larvae at constant 24°C and optimal 30% R. H. takes 20-154 days (Fallis, 1942). To pupate, a larva emerges from its host and spins a white silken cocoon within the larval casing. After 15 days at 24°C the parasitoid imago emerges from her cocoon by cutting a symmetrical opening, pushing the cap out of the way and finally cutting a hole through the side of the host larval casing.

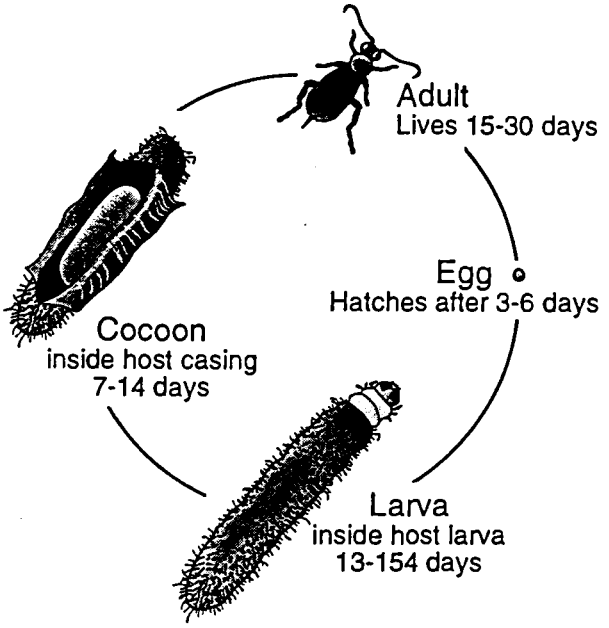
The natural environment of the CCM includes well-sheltered nests (Woodroffe, 1953), dry corpses (Bornemissza, 1957) or animal lairs (Mallis, 1969; Hill, 1990) that are not exposed to direct light. CCM larvae feed on and digest animal-derived fabric, feather, hide, horn, hair and fur, causing substantial damage in textile and fur warehouses, museums, and households (Mallis, 1969; Hill, 1990).

FIG. 1. Life cycle of the casemaking clothes moth, *Tinea pellionella* and its larval parasitoid, *Apanteles carpatus*.

Casemaking Clothes Moth, *Tinea pellionella*



Parasitoid, *Apanteles carpatus*



Wild CCM populations in northern and southern North American have one and two generations per year, respectively, but may have up to four generations in protected environments like households (Mallis, 1969). Males and females are photophobic and males are the strongest fliers (Key and Common, 1959; Mallis, 1969). Adults eclose in May through July, can mate within a few hours (Mallis, 1969), and live up to 30 days. Within a day of eclosion (or mating) females begin to oviposit 30 -160 eggs singly or in small groups on suitable host material (Fig. 1). Larvae hatch in 4 -10 days (24°C), begin feeding immediately, and construct a portable case of silk, food fibers, and other detritus which protects against moisture loss (Chauvin et al. 1979). Depending on food availability, larvae undergo ≥5 instars over 68-87 days of development, and then search for a pupation site, close both ends of the casing with silk, and pupate within the larval casing (Mallis, 1969). Following a 10-54 day pupal stage, the pupa partially emerges from the case before the imago ecloses.

Host location by hymenopterous parasitoids is commonly mediated by semiochemical stimuli (Vinson, 1984). These are often produced by the insect host itself or its products, e. g. larval frass or silk (Lewis and Jones, 1971; Wesoloh, 1976). In well-studied tritrophic communication systems, host-finding by parasitoids is mediated by kairomones emitted by the host-habitat (Dicke et al. 1990; Turlings et al. 1990; Steinberg et al. 1992), such as host plants (Takabayashi et. al. 1991; Turlings et al. 1991) or fungi (Vet, 1983; Vet and Van Opzeeland, 1985) and yeasts (Dicke et al., 1984) growing on (dead) plants.

My objectives were to: 1) determine if host location by *A. carpatus* is semiochemical-mediated; 2) identify the source of the semiochemicals; 3) identify the bioactive semiochemicals; and 4) to characterize the attractiveness of these semiochemicals.

2.0. METHODS AND MATERIALS

2.1. Insect Cultures

Adult *A. carpatus* (identified by M. Sharkey; voucher deposited at the Simon Fraser University Natural History Collection) emerged from CCM larvae infesting a 20 month-old, untanned beaver pelt originally from the British Columbia interior in a museum storage area. The pelt was kept at 20-25°C, 40-70% R. H. and a 10L:14D photoperiod in a 40 L glass container with mesh lid. Second and third generation parasitoids were reared on larvae of WCM (Insect Control and Research, Baltimore MD 21228) and larvae of CCM, respectively, feeding on discarded wool clothing (WCM) and beaver pelt (CCM), both supplemented with brewer's yeast. When beaver pelts became unavailable they were replaced by alum-tanned rabbit pelts (Pacific Leather & Fur Dressers, Vancouver B. C).

For two experiments (Table 1; Exps. 2, 3), adult *A. carpatus* eclosed from cocoons removed from CCM casings and placed individually in 35 mm Petri dishes. Parasitoids for all other experiments (Exps. 1, 4-14) emerged naturally from CCM larvae on rabbit (Exps. 4, 5, 7) or beaver pelts (Exps. 3, 6, 8-14). Groups of about 20 *A. carpatus* adults were maintained in wooden cages (15 X 15 X 12 cm) with mesh backs and Plexiglas™ fronts, and were provided daily with fresh sliced apple, sucrose, and a 10% honey-water solution.

2.2. Capture and Extraction of Volatiles

A CCM-infested beaver pelt (900 cm^2) was aerated for one week in a cylindrical Pyrex® glass chamber. A water-aspirator drew charcoal-filtered, humidified air at 2

L/min through the chamber and a glass column (14 cm X 13 mm O. D.) filled with Porapak Q (50-80 mesh, Waters Associates, Inc. Milford, Mass. 01757). Volatiles were eluted from the Porapak Q with 5 ml of redistilled pentane and the eluent concentrated to 2 ml by distillation in a 30 cm Dufton column. The concentration of volatiles was adjusted so that 2 µl equaled 5 pelt-min of volatile collection. Uninfested, salted, air-dried deer pelt (500 cm^2), and 25 pieces (2.3 cm^2 each) of unifested 'cut' and 'intact' rabbit pelt were also aerated, and volatiles eluted and concentrated as above. Fur of 'cut' rabbit pelt was shortened to 5 mm with blunt scissors and the cut hairs were included in the aerated material. Separated casings and larvae from 150 CCM third to fifth instars and 150 WCM fourth and fifth instars were extracted for 5 min in hexane. The extracts were concentrated by a gentle nitrogen stream so that 1 µl equaled 10 larval equivalents.

2.3. Volatile Analyses

Aliquots of 2.5 pelt-min equivalents of Porapak Q-captured volatiles or 10 larvae equivalents of hexane extracts were subjected to analysis by coupled gas chromatographic-electroantennographic detection (GC-EAD) (Arn et al., 1975), employing a Hewlett Packard (HP) 5890A gas chromatograph equipped with a fused silica column (30 m X 0.25 or 0.32 mm ID) coated with DB-5, DB-210 or DB-23 (J & W Scientific, Folsom, CA 95630). Full scan electron impact (EI) and chemical ionization (isobutane) (CI) mass spectra of EAD-active compounds were obtained by coupled GC-mass spectrometry (MS), using a Varian Saturn II Ion Trap GC-MS and a HP 5985B, respectively, each fitted with the DB-210 or DB-5 column, respectively, referred to above.

2.4. Laboratory Bioassay

2.4.1. Y-tube Olfactometer Bioassay

Anemotactic responses of 20-50 walking *A. carpatus* to odour sources were assessed per experiment in a Y-shaped Pyrex® glass olfactometer (ID 23 mm; stem 25 cm; arms 20 cm) at 22-27°C and 40-70% R. H. Air drawn through the apparatus at 5 L/min with a water aspirator carried volatiles from odour sources inside arms toward parasitoids released individually into the stem of the Y-tube. A parasitoid that penetrated ≥ 10 cm into a side arm within 15 min was classified as a responder. All others were classified non-responders and were not included in statistical analysis. For each replicate, odour sources were randomly assigned to, and placed near the orifice of side arms. During bioassays, a spot light at the Y-junction served as a light source, and filter paper barriers placed in each arm downwind of test stimuli standardized visual cues. Y-tubes were washed between replicates with Sparkleen™ and dried at 125°C >15 min.

2.4.2. Windtunnel Bioassay

Upwind flight by *A. carpatus* in response to volatile stimuli was assessed in a Plexiglas™ windtunnel (0.5 X 0.5 X 1.0 m) illuminated with a diffused, broadspectrum 40 W fluorescent light, through which air was drawn at 8cm/sec in a laminar flow (McDonald, 1995). Upwind, paired glass tubings (11 cm O. D.; 25 cm apart; 3 cm above windtunnel floor) were baited with Whatman® #1 filter paper impregnated with volatile stimuli. Five parasitoids per each of 12 replicates were introduced 1 m downwind of the volatile sources and their flight behaviour was observed for up to 30 minutes.
A parasitoid entering a glass tubing was recorded as a responder and was immediately

removed. All others were classed as non-responders and were not included in statistical analyses.

2.4.3. Bioassay Experiments

Fourteen binary choice experiments were conducted (Table 1). The first experiment tested the response of host-experienced parasitoids to odours of CCM-infested beaver pelt and a blank control. The second and third experiment tested the response of host-naive (Exp. 2) and host-experienced (Exp. 3) parasitoids to uninfested and CCM-infested rabbit pelt. To locate the source of semiochemicals, Exps. 4-7 tested the response of experienced parasitoids (first generation) to various odour sources, as follows: CCM larval faeces *versus* blank (Exp. 4); rabbit pelt with CCM larvae *versus* CCM larvae (Exp. 5); beaver pelt with or without CCM larvae (Exp. 6); and intact *versus* scissors-cut rabbit pelt (Exp. 7).

Experiments 8-13 tested the response of parasitoids (third generation) to volatile extracts and to EAD-active, synthetic volatiles, as follows: volatiles from CCM-infested beaver pelt *versus* control (Exp. 8); geranylacetone (Exp. 9) and nonanal (Exp. 10) (both Aldrich Chem. Company, Inc., Milwaukee, Wisconsin 53233) *versus* control; volatiles from CCM-infested beaver pelt *versus* geranylacetone (Exp. 11), and *versus* geranyl-acetone plus nonanal (Exp. 12); geranylacetone *versus* geranylacetone plus nonanal (Exp. 13). A final experiment (Exp. 14) tested upwind flight of host-experienced *A. carpatus* (third generation) in response to geranylacetone plus nonanal or to a solvent control.

Table 1. Numbers of host-naive or host-experienced *Apanteles carpatus* and summary of natural or synthetic stimuli in Y-tube olfactometer or windtunnel bioassay experiments.

Exp. No.	No. Parasitoids Tested	Disposition of Parasitoids*	Experimental Treatment: Y-tube Olfactometer Arm 1	Experimental Treatment: Y-tube Olfactometer Arm 2
1	20	Experienced	2.3 cm^2 piece of beaver pelt infested ≥5 days with 5 CCM larvae (4th instar)	Blank
2	50	Naive	2.3 cm^2 piece of rabbit pelt infested ≥3 days with 5 CCM larvae (4th instar)	2.3 cm^2 piece of uninfested rabbit pelt
3	20	Experienced	2.3 cm^2 piece of rabbit pelt infested ≥3 days with 5 CCM larvae (4th instar)	2.3 cm^2 piece of uninfested rabbit pelt
4	20	Experienced	Larval faeces (10 mg) collected within 24 hr of production by 200 CCM larvae (3-4th instar) feeding on rabbit pelt; stored at -2°C until 1 hr before testing	Blank
5	40	Experienced	5 CCM free-moving larvae (4th instar) with casing	2.3 cm^2 piece of rabbit pelt infested 5 days with 5 CCM larvae (4th instar)
6	30	Experienced	2.3 cm^2 piece of beaver pelt infested ≥5 days with 5 CCM larvae (4th instar)	2.3 cm^2 piece of beaver pelt infested ≥5 days with 5 CCM larvae (4th instar) which were removed immediately prior to bioassay
7	40	Experienced	2.3 cm^2 piece of rabbit pelt with scissors-cut hair	2.3 cm^2 piece of rabbit pelt with intact hair
8	20	Experienced	Porapak Q-captured volatiles from beaver pelt infested ≥5 days with CCM larvae (5 pelt-min in 2 µl of pentane on Whatman® #1 filter paper)	Pentane (2 µl) control on Whatman® #1 filter paper

Table 1 cont.

Exp. No.	No. Parasitoids Tested	Disposition of Parasitoids	Experimental Treatment	
			Y-tube Olfactometer	
			Arm 1	Arm 2
9	20	Experienced	Nonanal (16ng) in hexane (2µl) on Whatman® #1 filter paper	Hexane (2 µl) control on Whatman® #1 filter paper
10	20	Experienced	Geranylacetone (16ng) in hexane (2µl) on Whatman® #1 filter paper	Hexane (2 µl) control on Whatman® #1 filter paper
11	30	Experienced	Geranylacetone (16ng) in pentane (2µl) on Whatman® #1 filter paper	Porapak Q-captured volatiles from beaver pelt infested ≥5 days with CCM larvae (5 pelt-min in 2 µl of pentane on Whatman® #1 filter paper)
12	30	Experienced	Geranylacetone (16ng) and nonanal (16ng) in pentane (2µl) on Whatman® #1 filter paper	Porapak Q-captured volatiles from beaver pelt infested ≥5 days with CCM larvae (5 pelt-min in 2 µl of pentane on Whatman® #1 filter paper)
13	30	Experienced	Geranylacetone (8ng) and nonanal (8ng) in hexane (2µl) on Whatman® #1 filter paper	Geranylacetone (8ng) in hexane (2µl) on Whatman® #1 filter paper
			Windtunnel	
			Tube 1	Tube 2
14	60	Experienced	Geranylacetone (8ng) and nonanal (8ng) in hexane (2µl) on Whatman® #1 filter paper	Hexane (2µl) control on Whatman® #1 filter paper

* For Exps. 2-5, 7, exposure (>24 hrs) of emergent *A. carpatus* to rabbit pelt (2.3 cm^2) infested with two fourth instar CCM feeding >3 days produced host-experienced parasitoids. Host-naive parasitoids had no exposure to host infested pelt. Host-experienced *A. carpatus* for Exps. 1, 6, 8-14 were produced by exposing (>24 hrs) emergent adults to fourth instar CCM feeding >5 days on beaver pelt.

2.5. Statistical Analysis

The numbers of parasitoids responding to stimuli in various bioassays were compared with the χ^2 Goodness of Fit test, using Yate's correction for continuity ($\alpha = 0.05$) (Zar, 1984).

3.0. RESULTS

Experienced *A. carpatus* were strongly attracted to CCM larvae feeding on beaver pelt (Fig. 2; Exp. 1). Similarly, both experienced and naive parasitoids were strongly attracted to CCM larvae feeding on rabbit pelt (Fig. 2; Exps. 2, 3). In various experiments designed to identify the source of the attraction of *A. carpatus*, faeces from CCM larvae did not elicit a response (Fig. 3; Exp. 4); rabbit pelt with feeding CCM larvae was preferred over isolated CCM larvae and casings (Fig. 3; Exp. 5); and pieces of infested beaver pelt with or without feeding larvae were equally attractive (χ^2 test, P=0.36) (Fig. 3; Exp. 6). Scissors-cut rabbit pelt was more attractive than intact rabbit pelt (Fig. 3; Exp. 7).

Extracts of Porapak Q-captured volatiles from infested beaver pelt were highly attractive (Fig. 4; Exp. 8). Analysis of these volatiles by GC-EAD revealed two antennally active compounds (Fig. 5) with both EI (Figs. 6, 7) and CI (Figs. 8, 9) mass spectra very similar to those previously reported for nonanal and geranylacetone (Adams, 1989). Identical retention and mass spectral characteristics, and comparable EAD-activity of authentic nonanal and geranylacetone and corresponding compounds in volatile extracts confirmed these assignments. GC-EAD analyses also confirmed the presence of these two compounds in volatiles emanating from deer and rabbit pelts, but failed to detect them in hexane extracts of CCM larvae, or their casings, or in WCM larvae. Geranylacetone, but not nonanal (χ^2 test, $P = 0.17$) attracted *A. carpatus* (Fig. 4; Exps. 9, 10).

FIG. 2. Responses in Exp. 1-3 (Table 1) of walking host-experienced *Apanteles carpatus* in a Y-tube olfactometer to beaver pelt infested with instar casemaking clothes moth, *Tinea pellionella* or to a blank, and responses of host-naive and host-experienced *A. carpatus* to uninfested rabbit pelt or to pelt infested with third-fifth instar *T. pellionella*. Numbers of individuals responding to each stimulus are given in parentheses within bars. Bars with asterisks indicate a significant preference for a particular treatment (χ^2 test; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$).

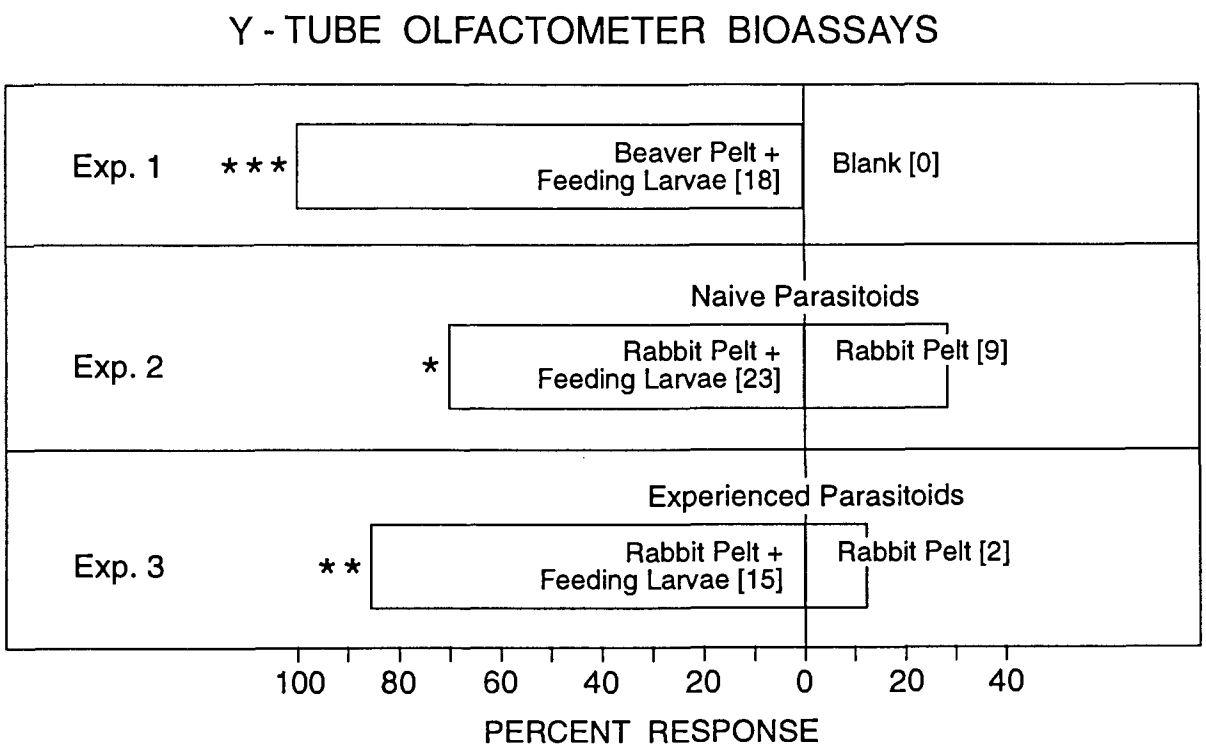

Y - TUBE OLFACTOMETER BIOASSAYS
Exp. 1
***
Beaver Pelt +
Feeding Larvae [18]
Blank [0]
Naive Parasitoids
Exp. 2
*
Rabbit Pelt +
Feeding Larvae [23]
Rabbit Pelt [9]
Experienced Parasitoids
Exp. 3
**
Rabbit Pelt +
Feeding Larvae [15]
Rabbit Pelt [2]
100
80
60
40
20
0
20
40
PERCENT RESPONSE

FIG. 3. Responses in Exp. 4-7 (Table 1) of walking host-experienced *Apanteles carpatus* in a Y-tube olfactometer to various stimuli from *Tinea pellionella* host-larvae or from host-habitat. Numbers of individuals responding to each stimulus are given in parentheses within bars. Bars with asterisks indicate a significant preference for a particular treatment (χ^2 test; * $P < 0.05$; ** $P < 0.01$).

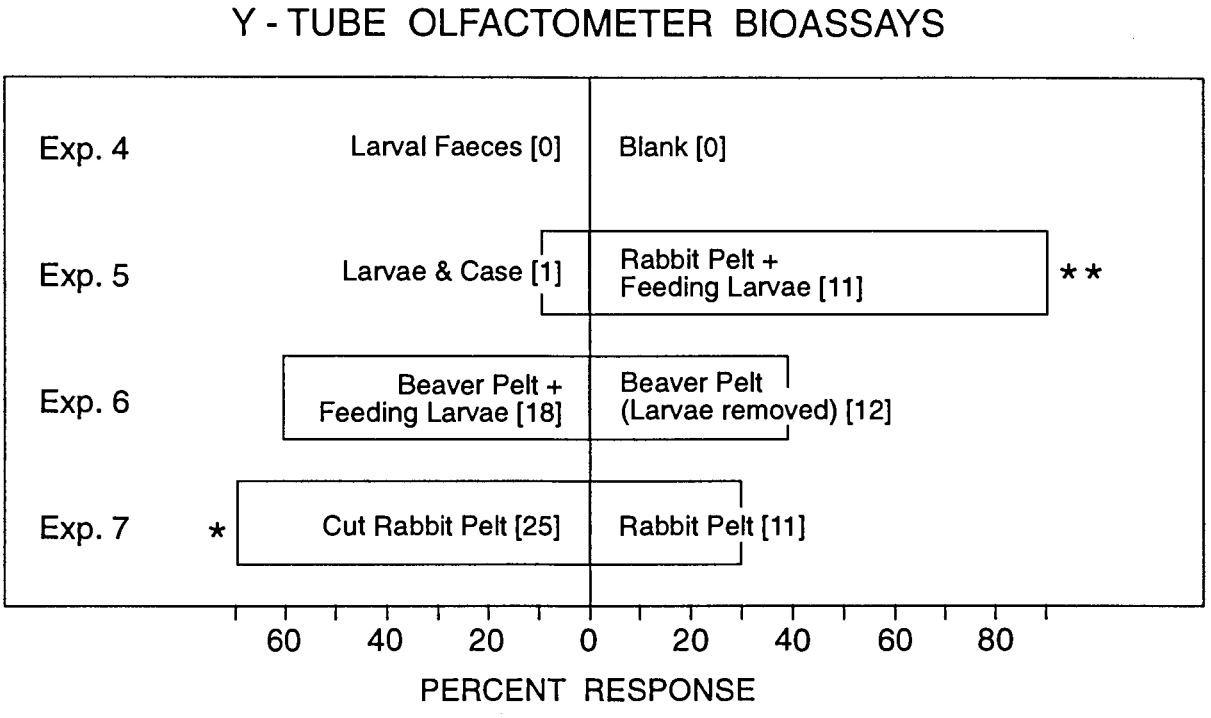
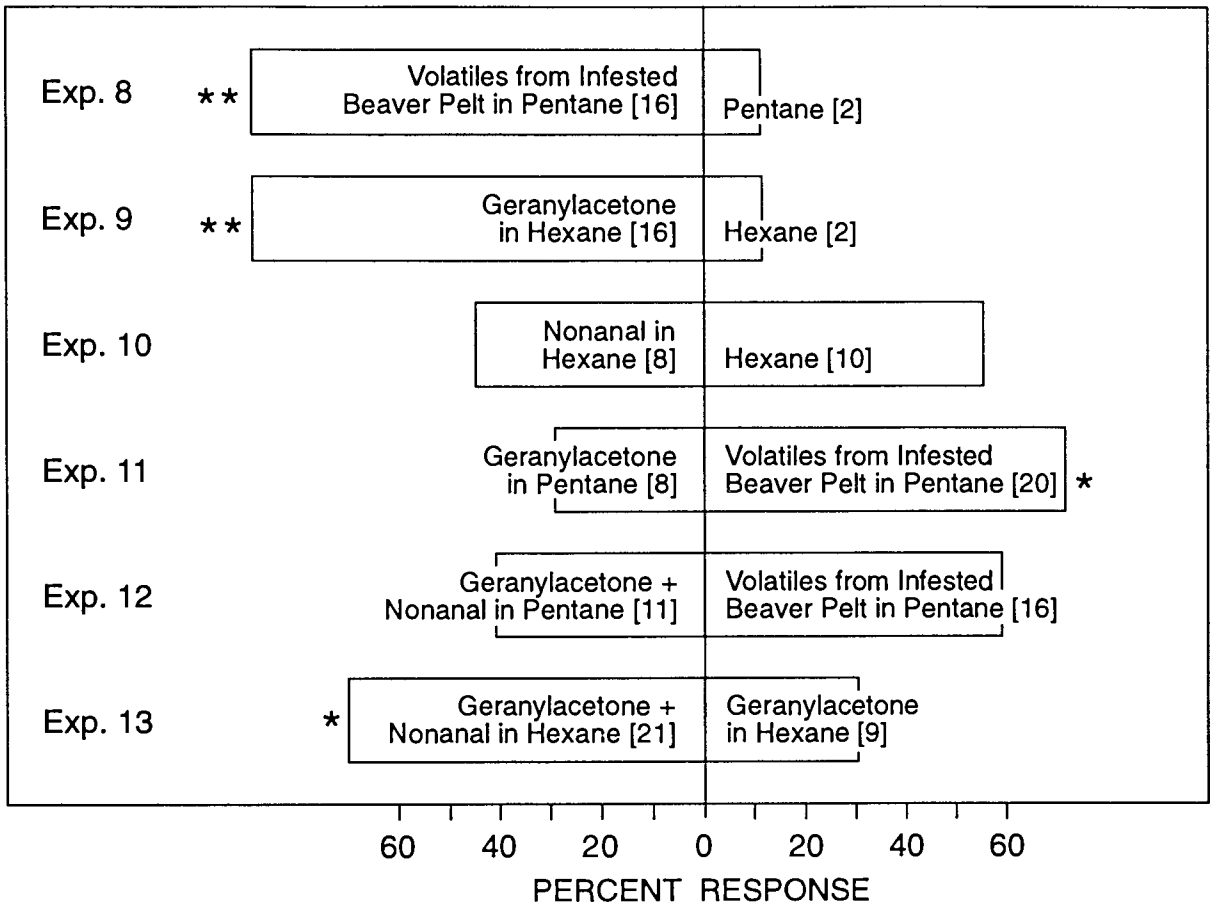

Y - TUBE OLFACTOMETER BIOASSAYS
Exp. 4
Larval Faeces [0]
Blank [0]
Exp. 5
Larvae & Case [1]
Rabbit Pelt +
Feeding Larvae [11]
* *
Exp. 6
Beaver Pelt +
Feeding Larvae [18]
Beaver Pelt
(Larvae removed) [12]
Exp. 7
*
Cut Rabbit Pelt [25]
Rabbit Pelt [11]
60
40
20
0
20
40
60
80
PERCENT RESPONSE

FIG. 4. Responses in Exp. 8-13 (Table 1) of walking host-experienced *Apanteles carpatus* in a Y-tube olfactometer to volatiles from beaver pelt infested with *Tinea pellionella* larvae or to synthetic candidate semiochemicals; and responses in Exp. 14 of flying host-experienced parasitoids in a windtunnel to synthetic semiochemicals and to a solvent control . Numbers of individuals responding to each stimulus are given in parentheses within bars. Bars with asterisks indicate a significant preference for a particular treatment (χ^2 test; * $P < 0.05$; ** $P < 0.01$).

Y - TUBE OLFACTOMETER BIOASSAYS



WINDTUNNEL BIOASSAY

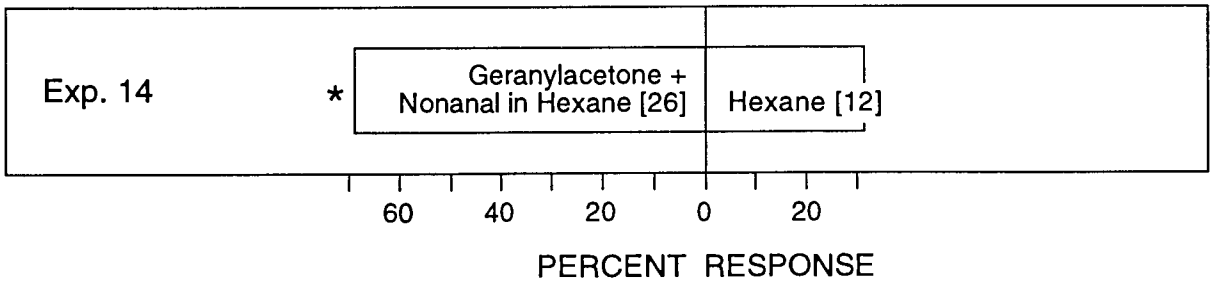


FIG. 5. Flame ionization detector (FID) and electroantennographic detector (EAD: *Apanteles carpatus* antenna) responses to aliquots of 2.5 pelt-min equivalents of volatiles from beaver pelt infested with *Tinea pellionella* larvae. Chromatography: Hewlett Packard 5890A equipped with DB-5 coated column; linear flow velocity of carrier gas: 35cm/sec; injector and FID detector temperature: 240 °C; temperature program: 1 min at 50°C, 10°C/min to 240°C.

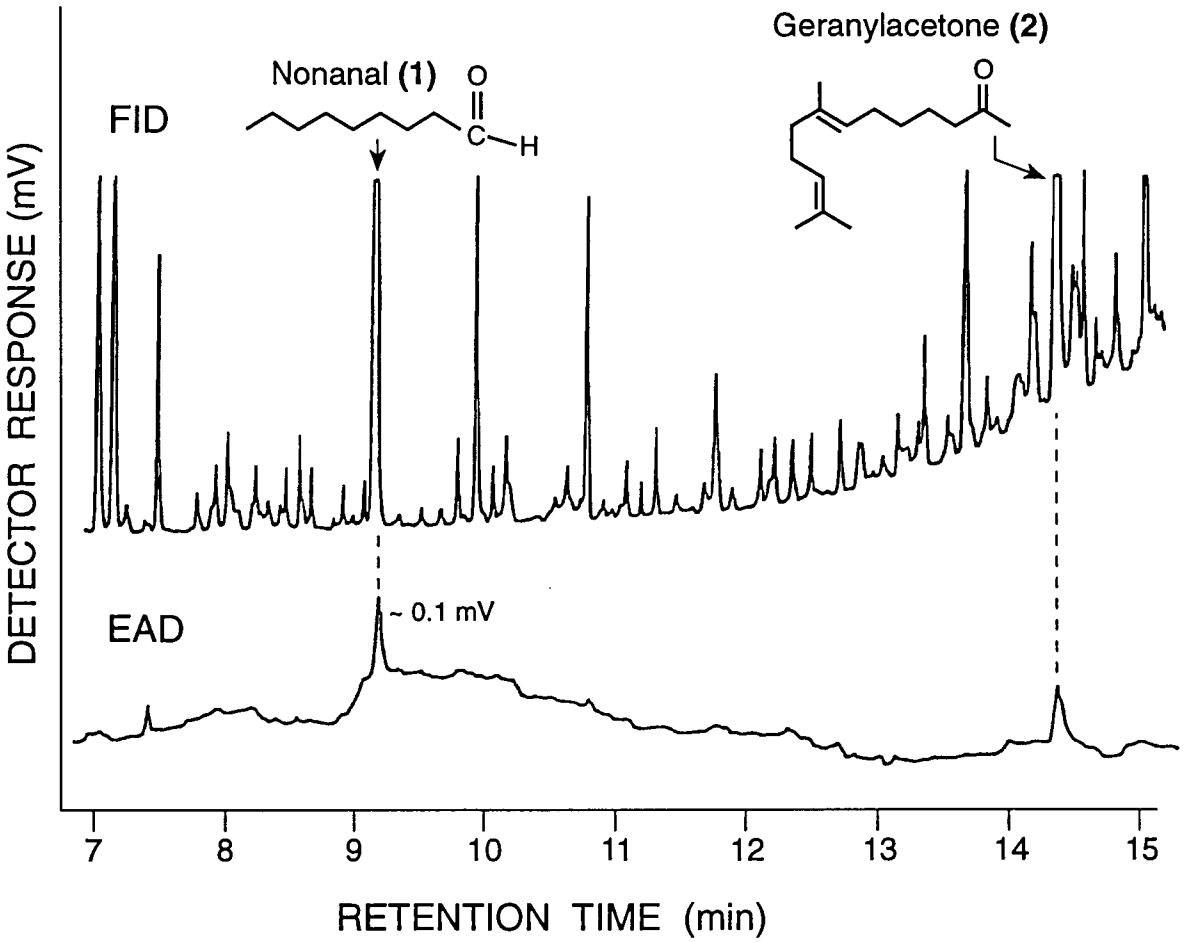

Geranylacetone (2)
Nonanal (1)
O
C
H
FID
EAD
~ 0.1 mV
DETECTOR RESPONSE (mV)
7
8
9
10
11
12
13
14
15
RETENTION TIME (min)

FIG. 6. Electron impact (70 eV) mass spectra of the EAD-active compound **1** (Fig. 5) and of authentic nonanal. Chromatography: Varian Saturn II Ion Trap GC-MS equipped with a DB-210 coated column; linear flow velocity of carrier gas: 35cm/sec; injector and FID detector temperature: 240°C; temperature program: 1 min at 50°C, 15°C/min to 220°C.

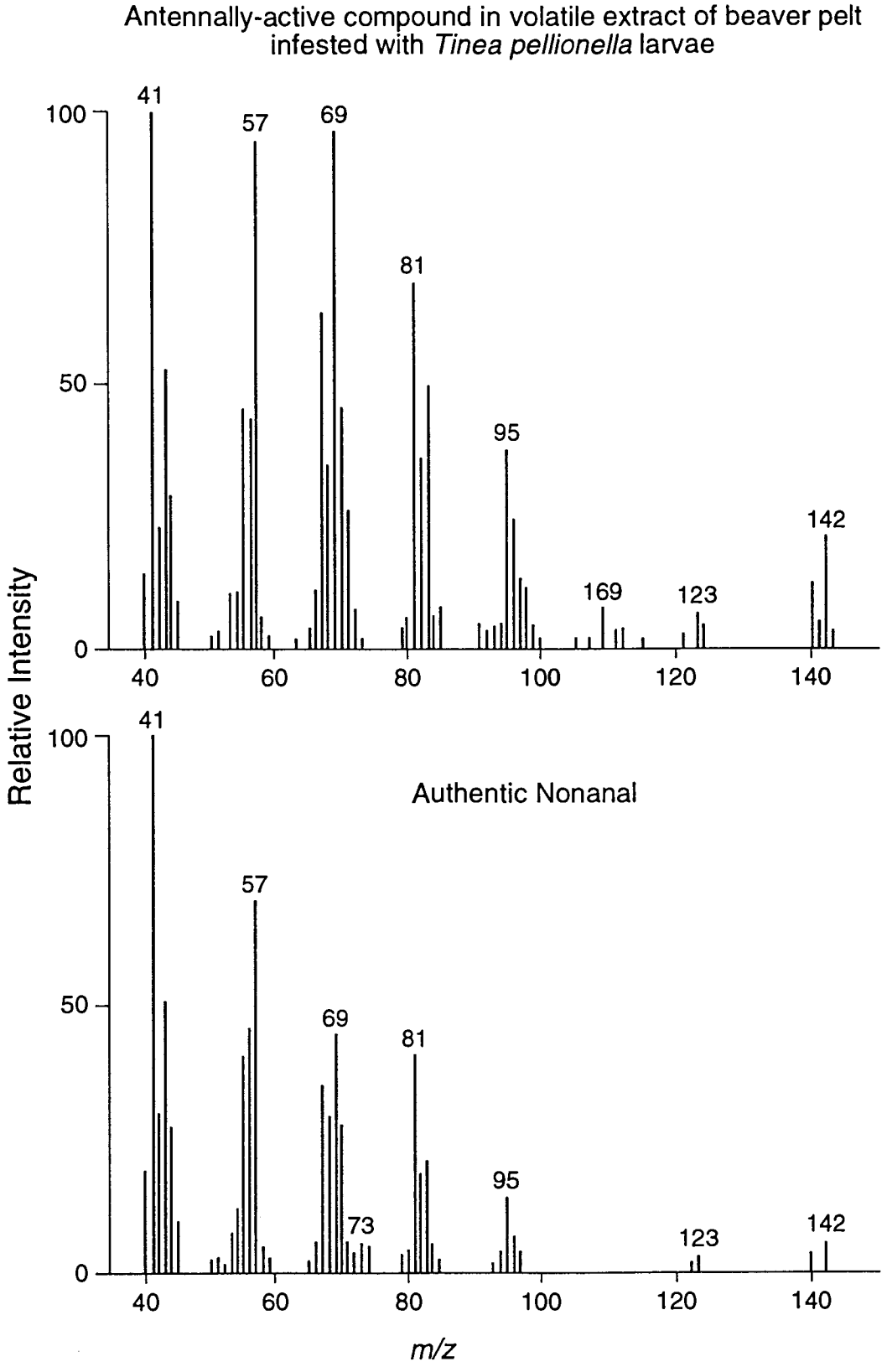

Antennally-active compound in volatile extract of beaver pelt infested with *Tinea pellionella* larvae
Relative Intensity
100
50
0
41
57
69
81
95
142
169
123
40
60
80
100
120
140
Authentic Nonanal
73
m/z

FIG. 7. Electron impact (70 eV) mass spectra of the EAD-active compound **2** (Fig. 5) and of authentic geranylacetone. Chromatography: Varian Saturn II Ion Trap GC-MS equipped with a DB-210 coated column; linear flow velocity of carrier gas: 35cm/sec; injector and FID detector temperature: 240°C; temperature program: 1 min at 50°C, 15°C/min to 220°C.

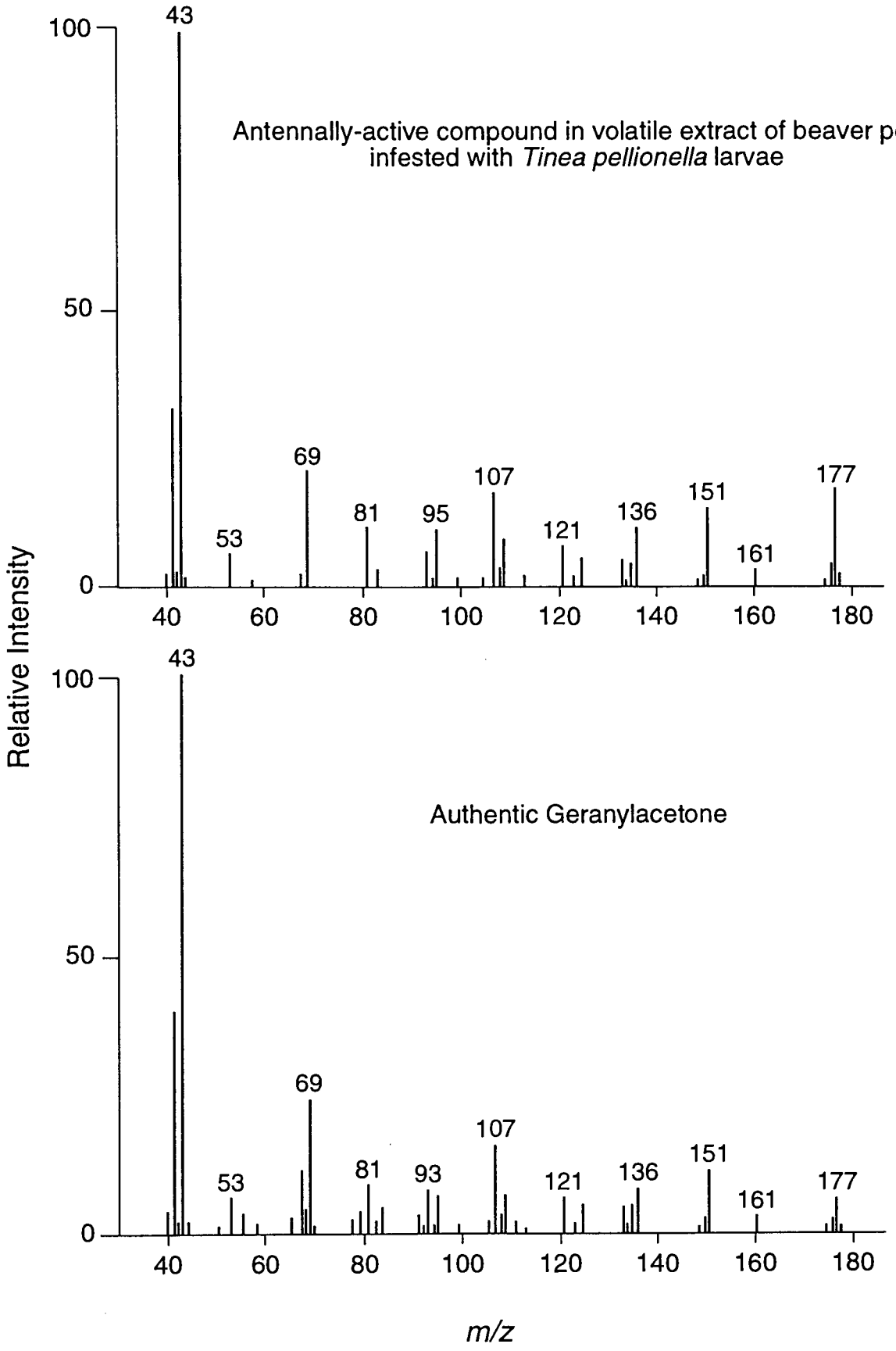

Antennally-active compound in volatile extract of beaver pelt infested with *Tinea pellionella* larvae
Authentic Geranylacetone
Relative Intensity
m/z
100
50
0
40
60
80
100
120
140
160
180
43
69
53
81
95
107
121
136
151
161
177
93

FIG. 8. Chemical ionization (isobutane) mass spectra of EAD-active compound **1** (Fig. 5) and of authentic nonanal. Chromatography: Hewlett Packard 5985B equipped with DB-5 coated column; linear flow velocity of carrier gas: 35cm/sec; injector and FID detector temperature: 240°C; temperature program: 1 min at 50°C, 10°C/min to 240°C.

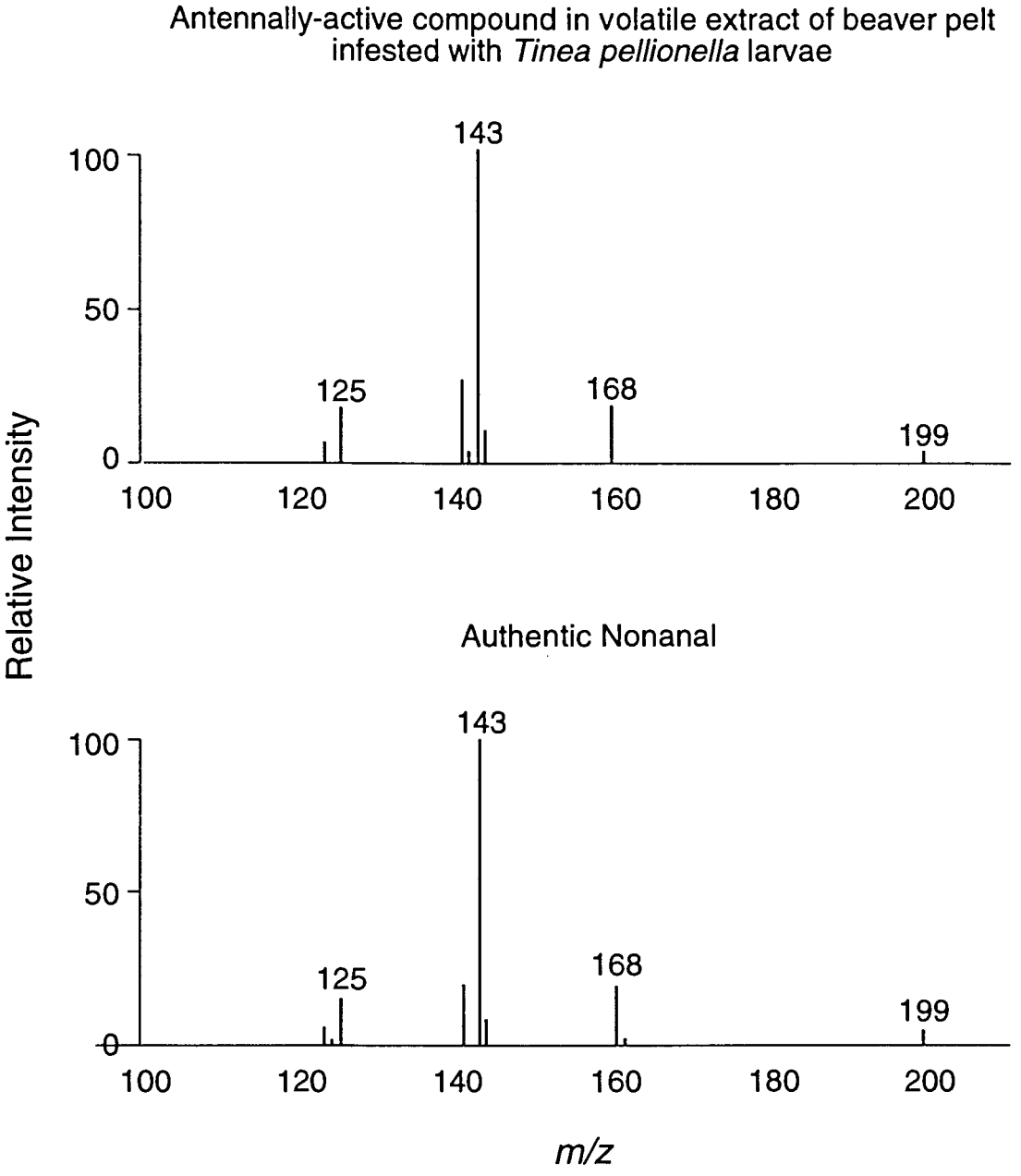
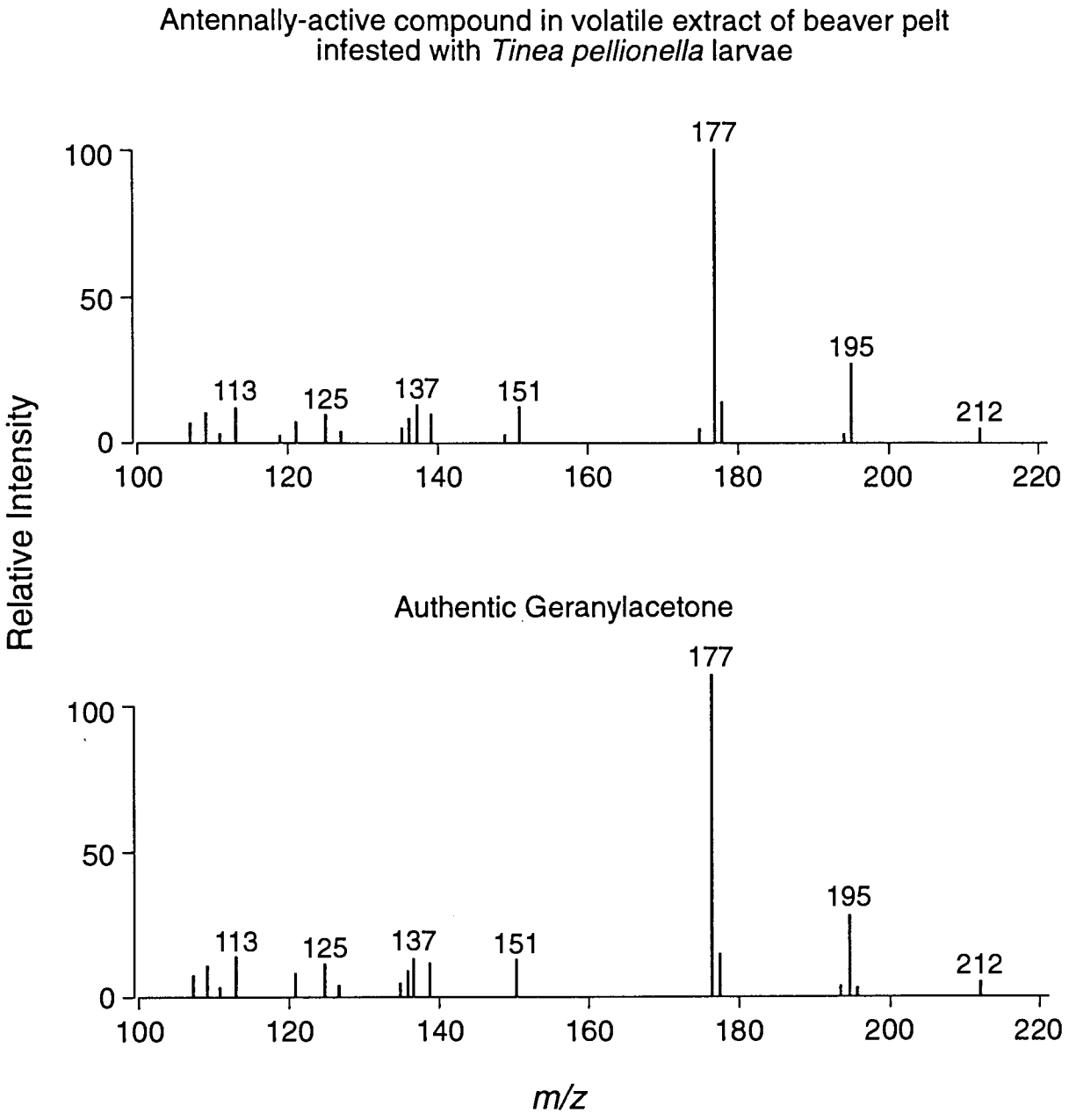

Antennally-active compound in volatile extract of beaver pelt infested with *Tinea pellionella* larvae
Relative Intensity
100
50
0
143
125
168
199
100
120
140
160
180
200
Authentic Nonanal
100
50
0
143
125
168
199
100
120
140
160
180
200
m/z

FIG. 9. Chemical ionization (isobutane) mass spectra of EAD-active compound **2** (Fig. 5) and of authentic geranylacetone. Chromatography: Hewlett Packard 5985B equipped with DB-5 coated column; linear flow velocity of carrier gas: 35cm/sec; injector and FID detector temperature: 240°C; temperature program: 1 min at 50°C, 10°C/min to 240°C.


Antennally-active compound in volatile extract of beaver pelt infested with *Tinea pellionella* larvae
Relative Intensity
100
50
0
177
195
113
125
137
151
212
100
120
140
160
180
200
220
Authentic Geranylacetone
177
100
50
195
113
125
137
151
212
0
100
120
140
160
180
200
220
m/z

Volatiles from infested beaver pelt were more attractive than geranylacetone alone (Fig. 4; Exp. 11) and as attractive (χ^2 test, $P = 0.09$) as geranylacetone plus nonanal (Fig. 4; Exp. 12). These two compounds combined were significantly more attractive than geranylacetone alone (Fig. 4; Exp. 13). In the windtunnel bioassay, attractiveness of these two compounds exceeded that of a solvent control (Fig. 4; Exp. 14).

4.0. DISCUSSION

Orientation of *A. carpatus* to CCM-infested beaver pelt (Fig. 2; Exp. 1) or to extracts of Porapak Q-captured volatiles from infested beaver pelt (Fig. 4; Exp. 8), and antennal responses to compounds in the captured volatiles (Fig. 5) indicate olfactory recognition of, and long range attraction to, the host and/or host-habitat. Similar attraction of host-naive and host-experienced parasitoids to CCM-infested rabbit pelt (Fig. 2; Exps. 2, 3) suggests that the foraging response is unconditioned. However, furthur experiments are required to thoroughly test this hypothesis (Steinberg et al, 1993). Lack of parasitoid attraction to isolated host larvae plus casing (Fig. 3; Exp. 5), or to larval faeces (Fig. 3; Exp. 4), and equal attraction to beaver pelt with or without host larvae (Fig. 3; Exp. 6) suggests that long range foraging behaviour is mediated by semiochemicals originating from the host-habitat but not the host larva. Because synthetic nonanal and geranylacetone at equivalent quantities were as attractive as volatiles from infested beaver pelt (Fig 4; Exp. 12), foraging response of parasitoids is obviously associated with these two compounds. In a synergistic manner, both compounds attract walking and flying (Fig. 4; Exps. 9, 12-14) parasitoids.

While nonanal and geranylacetone mediate long-range attraction of *A. carpatus* to host-habitat, tactile and vibrational cues may facilitate detection of host larvae. In the presence of host larvae, *A. carpatus* probes any fibrous substrate such as cotton balls and wool fabric (personal observation). Although CCM faeces do not contribute to long-range attraction (Fig. 3; Exp. 4), their presence in or on larval tubes of WCM increase oviposition rates of the parasitoids (Fallis, 1942).

Well-studied tritrophic communication systems involve plants, herbivores and their parasitoids (Stand and Vinson, 1982; Dicke et al. 1990; Turlings et al. 1990; Steinberg et al. 1992; McCall et al. 1993). Feeding by a herbivore changes the quantity and quality of the plant volatile blend (Turlings et al. 1991; Steinberg et al. 1993), which in turn induces attraction of parasitoids or predators to the herbivore habitat. Because the keratinaceous food source of clothes moth larvae in the *A. carpatus* tritrophic system is no longer alive, it is questionable whether the communication between the first and third trophic level can be classed as semiochemical-based. Superior attractiveness of scissors-cut rather than intact rabbit pelt (Fig. 3; Exp. 7), however, suggests that mechanical damage to fur induced a foraging response analogous to that of a caterpillar feeding on a plant leaf (Mattiacci et. al. 1994). While scissors-cutting of fur resulted in only slight quantitative increase of released nonanal and geranylacetone, multiple hair cuttings by larval mouthparts may effectively enhance release of these compounds. If so, *A. carpatus*, like herbivore parasitoids, would respond to semiochemicals provided by the first but induced and/or enhanced by organisms in the second trophic level.

Hosts of *A. carpatus* are invariably keratinophagous caterpillars. Natural host habitats, such as animal cadavers, animal lairs, or bird nests (Mallis, 1969; Hill, 1990), are highly specific and localized. Both olfactory recognition of (Fig. 5), and orientation toward host habitat volatiles (Fig. 4; Exps. 8, 12-14) suggest that *A. carpatus* is a habitat- rather than host-specialist (Vet and Dicke, 1992).

Because geranylacetone and nonanal are common in volatile blends from plants (Borg-Karlson, 1987; Chinta et al. 1994), animals (Chung and Cadwaller, 1993), and stored products (Pierce et al. 1990; Mushobozy et al. 1993), one might doubt whether they are specific enough to serve as reliable indicators of host-infested habitat. However, relative concentrations of geranylacetone in plant volatiles are low (Borg-Karlson, 1987) and although nonanal may be abundant (Borg-Karlson, 1987; Chinta et al. 1994) it alone is not attractive to foraging parasitoids (Fig. 4; Exp. 9).

High concentrations both of nonanal and geranylacetone in volatile extracts of infested beaver pelt (Fig. 5), accounting for 15% of all compounds, may be sufficient to provide reliable host-habitat stimuli, analogous to herbivore-induced synomones that guide certain parasitoids to their herbivorous hosts. Because habitat- but not host-volatiles mediate foraging behaviour by *A. carpatus* (Fig. 3; Exps. 4-7), periodic departure of third to fifth instar CCM from their feeding-habitat (personal observation) may represent a strategy of parasitoid avoidance (Vet and Dicke, 1992) as reported for caterpillars of the cabbageworm, *Pieris rapae* (L.) (Mauricio and Bowers, 1990).

REFERENCES

ADAMS, R. P. 1989. Identification of Essential Oils by Ion Trap Mass Spectroscopy. *Academic Press, Inc.*: San Diego, California.

ARN, H., STÄDLER, E., and RAUSCHER, S. 1975. The electroantennographic detector - a selective and sensitive tool in the gas chromatographic analysis of insect pheromones. *Z. Naturforsch.* 30c: 722-725.

BORG-KARLSON, A. K. 1987. Chemical basis for the relationship between *Ophrys* orchids and their pollinators. III. *Chem. Scr.* 27: 313-325.

BORNEMISSZA, G. F. 1957. An analysis of arthropod succession in carrion and the effect of its decomposition on the soil fauna. *Aust. J. Zool.* 5: 1-12.

CHAUVIN, G., VANNIER, G., and GUEGUEN, A. 1979. Larval case and water balance in *Tinea pellionella*. *J. Insect Physiol.* 25: 615-619.

CHINTA, S., DICKENS J. C., and ALDRICH, J. R. 1994. Olfactory reception of potential pheromones and plant odors by tarnished plant bug, *Lygus lineolaris* (Hemiptera: Miridae). *J. Chem. Ecol.* 20: 3251-3267.

CHUNG, H. Y., and CADWALLER, K. R. 1993. Volatile compounds in blue crab (*Callinectes sapidus*) meat and processing byproduct. *J. Food Sci.* 58: 1203-1207, 1211.

DICKE, M., VAN LENTEREN, J. C., BOSKAMP, G. J. F, and VAN DONGEN-VANLEEUWEN, E. 1984. Chemical stimuli in host-habitat location by *Leptopilina heterotoma* (Thomson) (Hymenoptera: Eucoilidae) a parasite of Drosophila. *J. Chem. Ecol.* 10: 695-712.

DICKE, M., SABELIS, M. W., TAKABAYASHI, J., BRUIN, J., and POSTHUMUS, M. A. 1990. Plant strategies of manipulating predator-prey interactions through allelochemicals: prospects for application in pest control. *J. Chem. Ecol.* 16: 3091-3118.

FALLIS, A. M. 1942. The life cycle of *Apanteles carpatus* (Say) (Hymenoptera: Braconidae), a parasite of the webbing clothes moth, *Tineola Bisselliella* Hum. *Can. J. Res. Sec. D.* 20: 13-19.

HILL, D. S. 1990. Pests of Stored Products and their Control. Belhaven Press, London.

KEY, K. H. L. and COMMON, I. F. B. 1959. Observations on the ecology of the clothes moths *Tineola bisselliella* (Humm.) and *Tinea pellionella* L. in a bulk wool store. *Aust. J. Zool.* 7: 39-77.

KROMBEIN, K. V., HURD Jr, P. D., SMITH, D. R., and BURKS, B.D. 1979. Catalog of Hymenoptera in America North of Mexico. *Smithsonian Institution Press.* Washington, D.C.

LEWIS, W. J. and JONES, R. L. 1971. Substance that stimulates host seeking by *Microplitis croceipes* (Hymenoptera: Braconidae), a parasite of *Heliothis* species. *Ann. Ent. Soc. Amer.* 64: 471-473.

MATTIACCI, L., DICKE, M. and POSTHUMUS, M. A. 1994. Induction of parasitoid attracting synomone in brussels sprouts plants by feeding of *Pieris brassicae* larvae: role of mechanical damage and herbivore elicitor. *J. Chem. Ecol.* 20: 2229-2247.

MALLIS, A. 1969. Handbook of Pest Control: The Behavior, Life History, and Control of Household Pests. Mac Nair-Dorland Company, New York.

MAURICIO, R and BOWERS, M. D. 1990. Do caterpillars disperse their damage?: larval foraging behaviour of two specialist herbivores, *Euphydryas phaeton* (Nymphalidae) and *Pieris rapae* (Pieridae). *Ecol. Entomol.* 15: 153-161.

MCCALL, P. J, TURLINGS, T. C. J., LEWIS, W. J., and TUMLINSON, J. H. 1993. Role of plant volatiles in host location by the specialist parasitoid *Microplitis croceipes* Cresson (Braconidae: Hymenoptera). *J. Insect Behav.* 6: 625-639.

MCDONALD, R. S. 1995. Ecological and physiological factors affecting mate-finding and mating behaviour of *Delia antiqua* (Meigen) (Diptera: Anthomyiidae). Ph.D. Thesis, Simon Fraser University, Burnaby, B.C.

MUSHOBOZY, D. K., PIERCE, H. D., JR., and BORDEN, J. H. 1993. Evaluation of 1-octen-3-ol and nonanal as adjuvants for aggregation pheromones for three species of cucujid beetles (Coleoptera: Cucujidae). *J. Econ. Entomol.* 86: 1835-1845.

PIERCE, A. M., PIERCE, H. D., JR., OEHLSCHLAGER, A. C, and BORDEN, J. H. 1990. Attraction of *Oryzaephilus surinamensis* (L.) and *Oryzaephilus mercanter* (Fauvel) (Coleoptera: Cucujidae) to some common volatiles of food. *J. Chem. Ecol.* 16: 465-475.

RUTZ, D. A. and SCOLES, G. A. 1989. Occurrence and seasonal abundance of parasitoids attacking muscoid flies (Diptera: Muscidae) in caged-layer poultry facilities in New York. *Environ. Entomol.* 18: 51-55.

STRAND, M. R. and VINSON, S. B. 1982. Behavioral response of the parasitoid *Cardiochiles nigriceps* to a kairomone. *Entomol. Exp. Appl.* 31: 308-315.

STEINBERG, S., DICKE, M., and VET, L. E. M. 1993. Relative importance of infochemicals from first and second trophic level in long range host location by the larval parasitoid *Cotesia glomerata. J. Chem. Ecol.* 19: 47-59.

STEINBERG, S., DICKE, M., VET, L. E. M., and WANNINGEN, R. 1992. Response of the braconid parasitoid *Cotesia*(=*Apanteles*) *glomerata* to volatile infochemicals: effects of bioassay set-up, parasitoid age and experience and barometric flux. *Entomol. Exp. Appl.* 63: 163-175.

TAKABAYASHI, J., NODA, T. and TAKAHASI, S. 1991. Plants produce attractants for *Apanteles kariyai*, a parasitoid of *Pseudaletia separata*; Cases of 'communication' and 'misunderstanding' in parasitoid-plant interactions. *Appl. Ent. Zool.* 26: 237-243.

TURLINGS, T. C. J., TUMLINSON, J. H., and LEWIS, W. J. 1990. Exploitation of herbivore induced plant odors by host seeking parasitic wasps. *Science.* 250: 1251-1253.

TURLINGS, T. C. J., TUMLINSON, J. H., and LEWIS, W. J. 1991. Larval damaged plants: source of volatile synomones that guide the parasitoid *Cotesia marginiventris* to the microhabitat of its hosts. *Entomol. Exp. Appl.* 58: 75-82.

VET, L. E. M. 1983. Host-habitat location through olfactory cues by *Leptopilina clavipes* (Hartig) (Hymenoptera: Eucoilidae), a parasitoid of fungivorous Drosophila: the influence of conditioning. *Neth. J. Zool.* 33: 225-248.

VET, L. E. M. and DICKE, M. 1992. Ecology of infochemical use by natural enemies in a tritrophic context. *Annu. Rev. Entomol.* 37: 141-172.

VET, L. E. M. and VAN OPZEELAND, K. 1985. Olfactory microhabitat location in *Leptopilina heterotoma* (Thomson) (Hymenoptera: Eucoilid), a parasitoid of Drosophilidae. *Neth. J. Zool.* 35: 497-504.

VINSON, S. B. 1984. Parasitoid-host relationship. *in* Chemical Ecology of Insects. Bell. W. J. and Cardé, R. T. Sinaur Associates, Inc: Massachusetts.

WESELOH, R. M. 1976. Behavioral responses of the parasite, *Apanteles melanoscelus*, to gypsy moth silk. *Environ. Entomol.* 5: 1128-1132.

WOODROFFE, G. E. 1953. An ecological study of the insects and mites in the nests of certain birds in Britain. *Bull. ent. Res.* 44: 739-772.

ZAR, J. H. 1984. Biostatistical Analysis. Prentice-Hall, Englewood Cliffs, New Jersey.