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CUTICULAR HYDROCARBONS OF WINGED AND WINGLESS MORPHS OF THE ECTOPARASITOID *CHOETOSPILA ELEGANS* WESTWOOD (HYMENOPTERA: PTEROMALIDAE) AND ITS HOST, LARVAL LESSER GRAIN BORER (*RHYZOPERTHA DOMINICA*) (COLEOPTERA: BOSTRICHIDAE)

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Abstract—1. The cuticular hydrocarbons from winged and wingless morphs of *Choetospila elegans*, a larval ectoparasite of several internal-feeding stored product beetle pests, were characterized.

2. All four morphs share the same cuticular hydrocarbons, with the major components being n-alkanes (C_{21}–C_{33}).

3. The minor components are 3-, 11- and 13-methyl branched alkanes and *Z*-10-monoenes.

4. Two-way analysis of variance (sex and wing morph) shows that males and females have the same profiles, whereas four components showed significant differences between wingtype morphs.

5. Only one of these four hydrocarbons (n-C_{31}) was a major component.

6. The cuticular hydrocarbons of larvae of the lesser grain borer, *Rhyzopertha dominica*, were also characterized.

7. Although the beetles' major components were the same n-alkanes as those found on the adult parasites, their minor components were different.

8. Thus, the beetle larvae have no alkenes, but instead have 3-, 11-, 13- and 15-methylbranched alkanes, as well as a series of 11,15-dimethylalkanes.

9. A cuticular hydrocarbon profile obtained on a population of *C. elegans* winged females that had been reared on rice weevil larvae was nearly identical to that from winged females reared on the lesser grain borer larvae.

INTRODUCTION

Cuticular hydrocarbons are increasingly being recognized as major semiochemicals for organisms in a diversity of ecological situations, functioning as species, gender, and colony recognition cues, and as pheromones, allomones and kairomones (Howard and Blomquist, 1982; Lockey, 1988; Stowe, 1988; Howard, 1993). Much of the early literature focussed on chemically surveying species in a wide range of higher order taxa for the presence of novel cuticular hydrocarbons (Jackson and Blomquist, 1976; Lockey, 1988). More recent studies have focussed on semiochemical roles and chemosystematic applications (Howard, 1993). Despite there now being well over 300 papers on the cuticular hydrocarbons of arthropods, major taxa are still virtually unknown. The order Hymenoptera is large and speciose, and has representatives in virtually every biome of this planet. Few studies on cuticular hydrocarbons have been conducted on members of this order, however, with most of those being on the ants, bees, and social wasps. Within the Hymenoptera, the parasitic taxa make up a large proportion of the known species, and yet the number of papers on their cuticular hydrocarbons is very small (Lockey, 1988; Howard, 1993).

The aculeate family Pteromalidae is an important one for biological control efforts in a diversity of pest species, including stored grain insects. Little is known of the biochemistry or chemical ecology of these important parasites, however. Indeed, we are aware of only one previous report on cuticular hydrocarbons in this family (Espelie *et al.*, 1990), and that paper deals with a chemotaxonomic separation of two apparent sibling species. As part of a larger study on the chemical ecology of stored grain insects and their parasites and predators, Howard (1992) recently reported on the cuticular hydrocarbon composition of two bethylid parasites (*Cephalonomia waterstoni* and *Laelius utilis*) associated with the rusty grain beetle, *Cryptolestes ferrugineus*, and the dermestid beetle *Trogoderma variable*, respectively. In that study, he also reported on the cuticular hydrocarbon

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composition of the host beetles. Each of these bethylid parasites was known to be relatively host-specific, whereas most pteromalid species are generally not known to be host-specific. We now report on the cuticular hydrocarbon composition of a pteromalid species found associated with a wide diversity of larval, internal-seed-feeding beetle species, especially species associated with stored grain (Gordh, 1979). This species, *Choetospila elegans*, is being studied in several laboratories for its potential as a biological control agent for major stored grain pests such as the rice and maize weevil, and the lesser grain borer. In addition, this species is known to exist as both winged and wingless morphs for both males and females (Gordh, 1979). Although other species of insects sometimes have gender-based differential hydrocarbon profiles (Howard, 1993), no reports exist on possible wing morph variability. We have therefore examined the various morphs of *C. elegans* (Fig. 1) for possible gender and wing morph differences in cuticular hydrocarbon composition. In addition, we have compared the hydrocarbon profiles of the adult parasites to the larval beetle hosts on which they were reared.

MATERIALS AND METHODS

Insects

Cultures of *Choetospila elegans* and *Rhyzopertha dominica* (F.) were collected from farm-stored wheat in Kansas and have been in laboratory culture on wheat for at least one year. Parasites were collected between one and three weeks after they emerged in stock culture jars, and killed by freezing at −40°C. They were then sexed and assorted by wing morphology into four classes (WF, winged females, WM, winged males, WLF, wingless females and WLM, wingless males). A second sample of parasites (137 WF) was obtained from J. Brower, USDA-ARS (Savannah, GA). These parasites had been reared from rice weevils [*Sitophilus oryzae* (L.)] cultured on whole wheat. *Rhyzopertha dominica* larvae were obtained by allowing mated adult female *R. dominica* to oviposit for 12 hr on wheat lightly dusted with flour. The wheat kernels were then gently shaken on a 30-mesh screen and beetle eggs that fell off the wheat kernels were collected in whole wheat flour enriched with 5% brewers yeast. These eggs were maintained in an incubator at 30°C and 70% R.H., and a 16:8 (L:D) photoperiod for three weeks. The flour was then sieved to separate out the lesser grain borer larvae for chemical analysis. These beetles were late-instar larvae of a size that the parasite would commonly oviposit onto.

Chemical analyses

Insects were killed by freezing at −40°C and cuticular lipids were extracted by immersing the insects in three successive 0.25-ml portions of hexane for 1 min each time. The combined portions from

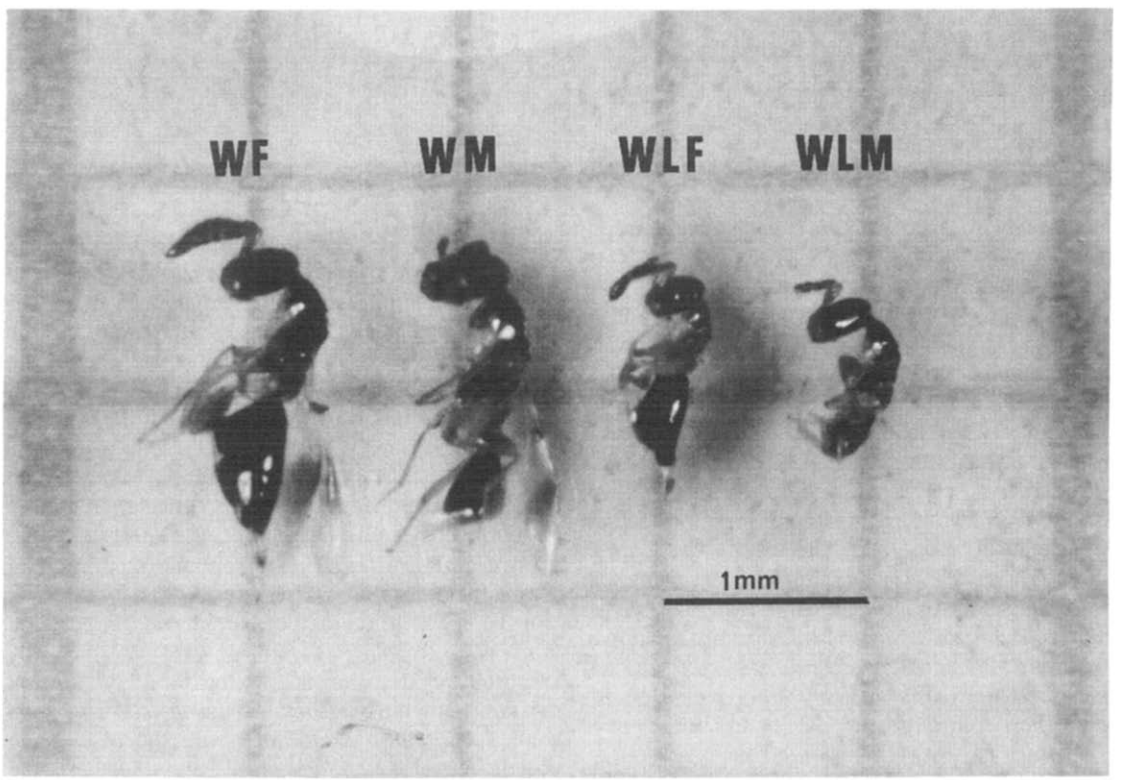

Fig. 1. Winged and wingless males and females of *Choetospila elegans*. WF (winged female), WM (winged male), WLF (wingless female), WLM (wingless male).

each sample were concentrated under a gentle stream of N_2, and hydrocarbons were isolated by chromatography on a 3-cm "mini-column" of Biosil A (Bio-Rad Laboratories, Richmond, CA) as described earlier (Howard *et al.*, 1978). A total of 3553 *C. elegans* (1445 WF, 1075 WM, 263 WLF and 770 WLM) was used for mass spectral chemical characterization. For compositional analysis, additional parasites were obtained from our stock cultures. These samples consisted of 10 wasps per replicate, and three replicates of each sex-morph category were analyzed.

Electron Impact Mass spectral analyses were conducted using a Hewlett–Packard 5790A GC (Hewlett–Packard, Inc., San Fernando, CA) containing a DB-5 bonded phase capillary column (10 m long, 0.19 mm inside diameter) (J and W Scientific, Folsom, CA) connected to a Hewlett–Packard 5970 mass selective detector and a Hewlett–Packard 9133 data system. Ultrapure helium was the carrier gas, with a column head pressure of 0.75 kg/cm^2. Mass spectra were obtained at 70 eV. Analyses were done using temperature programming, with an initial temperature of 150°C, a final temperature of 320°C, a program rate of 10°C, a 2 min initial hold period and a 20 min final hold period. The splitless Injector was set at 275°C and the GC/MSD interface was at 280°C. Retention times of each hydrocarbon component and equivalent chain length values (ECL) were obtained by comparison with known n-alkane standards (Howard *et al.*, 1978). Individual components were identified from their characteristic EI-MS fragmentation patterns (Jackson and Blomquist, 1976; Nelson, 1978).

Positive confirmation of molecular weights of all hydrocarbons was obtained by chemical ionization mass spectrometry (CI-MS). CI-MS were obtained using a Hewlett–Packard 5890 Series II gas chromatograph coupled to a Hewlett–Packard 5971A mass selective detector with a G1072A chemical ionization accessory. The GC was equipped with a DB-5 capillary column (30 m length; 0.32 mm inside diameter) and all analyses used temperature programming (150°C initial, 320°C final, 5.0°C per min, 2 min initial hold, 10 min final hold). The splitless injector port was set at 250°C and the GC/MSD interface was set at 280°C. Ultrapure helium was the carrier gas with a column head pressure of 0.75 kg/cm^2, and ultrapure methane was the ionizing gas. Mass spectra were scanned from m/z 50 to 650 and acquired data were collected and analyzed on a Hewlett–Packard 486/33T workstation using HP chemstation software.

Double-bond location in alkenes were obtained by preparing dithiomethyl ethers (DMS) (Francis and Veland, 1981) and examining their electron impact mass spectra. Stereochemistry of the parent alkene was inferred by comparison with the retention time of the DMS ethers of known standards.

Table 1. Identification of cuticular hydrocarbons from winged and wingless morphs of female and male *Choetospila elegans*

Hydrocarbon	ECL*	CN†	Diagnostic EI-MS fragments (m/z)
n-C_{21}	21.00	21	296
n-C_{22}	22.00	22	310
n-C_{23}	23.00	23	324
n-C_{24}	24.00	24	338
Z-10-$C_{25:1}$	24.72	25	350; 187, 257, 444‡
n-C_{25}	25.00	25	352
3-MeC_{25}	25.70	26	337; 309; 351
n-C_{26}	26.00	26	366
n-$C_{27:1}$	26.75	27	378
n-C_{27}	27.00	27	380
11-;13-MeC_{27}	27.29	28	169, 253; 197, 225
3-MeC_{27}	27.69	28	337; 351; 379
n-C_{28}	28.00	28	394
n-$C_{29:1}$	28.73	29	406
n-C_{29}	29.00	29	408
3-MeC_{29}	29.70	30	393; 365; 407
n-C_{30}	30.00	30	422
n-C_{31}	31.00	31	436
n-C_{33}	33.00	33	464
11-;13-MeC_{33}	33.35	34	169, 337; 197, 309
3-MeC_{33}	33.75	34	435; 421; 463

*Equivalent Chain Length.
†Carbon Number, determined by chemical ionization mass spectrometry.
‡Diagnostic dithiomethyl ether ion fragments.

Statistical analyses

Compositional analyses of hydrocarbons were conducted using the EI-MS system as described above. Area counts obtained from electronic integration were summed and converted into percent values. For statistical analysis of variance, these percent values (as proportions) were converted into arcsin square root transformations. Reported means and variances are on the untransformed values. Summary statistics and two-way analysis of variance were conducted using the personal computer software program Statgraphics Plus, Version 5 (Statistical Graphics Corp., Rockville, MD).

Voucher specimens

Voucher specimens (Lot No. 48) have been deposited in the Research Collection of Insects, Kansas State University, Manhattan, Kansas.

RESULTS

A variety of cuticular hydrocarbons were identified from *C. elegans* (Table 1). The major components (*ca* 95% of the total) were n-alkanes from C_{21} to C_{33}. The remaining 5% of the hydrocarbons were a homologous series of 3-, 11- and 13-methylalkanes and an apparent homologous series of *Z*-10-monoenes (interpretable mass spectra of the monoene DMS derivatives were obtained only for $C_{25:1}$, but the retention times of the other homologous monoenes are consistent with them also being members of the *Z*-10-series). No dimethylalkanes were detected, nor were any dienes detected. Both males and females, and winged and non-winged individuals all had the same cuticular hydrocarbon components, and some

apparent variation among the morphs was detected by visual inspection (Fig. 2, Table 2). A two-way analysis of variance was therefore conducted, using sex and wing morph as factors. The quantities of four hydrocarbons were shown to be significantly different between winged and wingless morphs: n-C_{22} ($P = 0.0264$), n-C_{23} ($P = 0.0160$), n-C_{26} ($P = 0.0320$) and n-C_{31} ($P = 0.0383$). These four components represent approximately 24% of the total in the winged morphs and approximately 18% in the non-winged morphs. No hydrocarbons were found to be significantly different between males and females ($P > 0.05$).

The cuticular hydrocarbons of the larvae of the lesser grain borer are also dominated by n-alkanes (*ca* 82% of the total mixture, ranging from n-C_{22} to n-C_{35}), but less so than in the parasites (Fig. 3, Table 3). The minor components also differ somewhat from that of the wasps. In particular, no alkenes are present in the beetle larvae, and the homologous series of methyl branched alkanes range from 3-Me to 15-Me. In addition, the beetles possess a homologous series of 11,15-dimethylalkanes that are missing from the wasps.

Since *C. elegans* is found associated with a variety of stored product beetle hosts, we deemed it wise to compare the cuticular hydrocarbon profile of our population, reared on the lesser grain borer, to the profile of *C. elegans* from a different population, reared on the rice weevil, *Sitophilus oryzae*. The hydrocabons of the winged female parasites reared on rice weevils were identical to the lesser grain borer parasite population, with only minor abundance differences (n-alkanes represent approximately 98% of the hydrocarbons in female rice weevils) (Fig. 4).

DISCUSSION

The cuticular hydrocarbons of parasitic Hymenoptera are very poorly known, with *C. elegans* being the seventh species examined for these phenotypic characters. To date, the taxa examined include

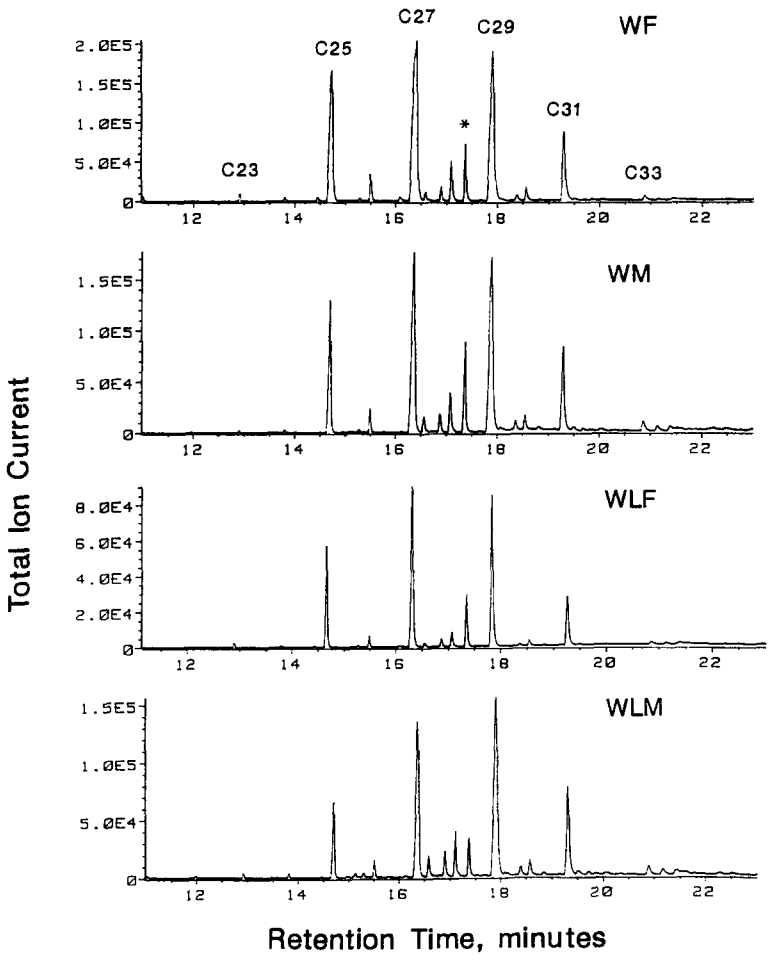

Fig. 2. Total ion chromatograms of the cuticular hydrocarbons from winged females (WF), winged males (WM), wingless females (WLF) and wingless males (WLM) of adult *Choetospila elegans*. These insects were reared on larvae of *Rhyzopertha dominica*, cultured on wheat. *Non-hydrocarbon contaminant.

Table 2. Mean (SE) per cent composition of the cuticular hydrocarbons of winged and wingless morphs of female and male *Choetospila elegans* ($N = 3$ replicates of 10 insects per treatment)

Compound	Winged female	Winged male	Nonwinged female	Nonwinged male
n-C_{21}	Tr*	Tr	Tr	Tr
n-C_{22}	0.20 (0.20)	1.42 (0.41)	1.44 (0.72)	3.08 (0.38)
n-C_{23}	1.19 (0.18)	1.46 (0.33)	1.83 (0.53)	3.26 (0.23)
n-C_{24}	5.50 (4.35)	1.51 (0.24)	1.86 (0.76)	3.58 (0.20)
Z-10-$C_{25:1}$	Tr	Tr	Tr	Tr
n-C_{25}	7.44 (1.44)	6.88 (0.96)	11.73 (3.14)	9.78 (1.39)
3-MeC_{25}	1.01 (0.31)	0.96 (0.26)	2.02 (1.30)	0.97 (0.17)
n-C_{26}	2.72 (0.25)	2.68 (0.19)	2.97 (0.80)	5.26 (0.38)
n-$C_{27:1}$	0.13 (0.12)	0.74 (0.26)	0.95 (0.46)	0.67 (0.39)
n-C_{27}	20.39 (2.10)	21.37 (1.04)	21.24 (6.59)	20.06 (1.27)
11-;13-MeC_{27}	1.00 (0.29)	2.49 (0.65)	1.98 (0.64)	2.04 (0.11)
3-MeC_{27}	1.06 (0.31)	2.43 (0.66)	1.99 (0.30)	1.90 (0.17)
n-C_{28}	3.92 (0.53)	5.04 (0.38)	4.06 (1.07)	5.74 (0.16)
n-$C_{29:1}$	Tr	Tr	Tr	Tr
n-C_{29}	28.80 (1.26)	32.37 (2.00)	28.18 (0.45)	28.16 (1.00)
3-MeC_{29}	0.30 (0.17)	1.53 (0.49)	1.17 (0.88)	1.15 (0.14)
n-C_{30}	2.42 (0.30)	2.80 (0.24)	3.18 (1.47)	2.76 (0.23)
n-C_{31}	19.93 (3.86)	12.67 (0.28)	11.96 (1.68)	10.53 (0.94)
n-C_{33}	2.58 (0.78)	1.61 (0.26)	1.57 (0.25)	0.92 (0.46)
11-;13-MeC_{33}	0.78 (0.77)	0.95 (0.16)	0.83 (0.19)	0.12 (0.12)
3-MeC_{33}	0.63 (0.36)	1.10 (0.15)	1.05 (0.66)	Tr

*Trace (less than 0.1 per cent).

the codling moth parasitoid *Ascogaster quadridentata* Wesmael (Braconidae) (Espelie and Brown, 1990), its hyperparasitoid *Perilampus fulvicornis* Ashmead (Perilampidae) (Espelie and Brown, 1990), an *Orsema* sp. (Eucharitidae), an inquiline of the red imported fire ant (Vander Meer *et al.*, 1989), two bethylids, *Cephalonomia waterstoni* Gahan and *Laelius utilis* Cockrell, both ectoparasites of stored product beetles (Howard, 1992), *Rhopalicus pulchripennis* (Crawford) (Pteromalidae), a parasitoid of bark beetles, and *C. elegans* (Pteromalidae), also an ectoparasitoid of stored product beetles. Given the diversity of the taxa involved (three superfamilies and distinctly different ecological niches) it is not surprising that the cuticular hydrocarbon profiles of these seven species are rather different.

The *A. quadridentata* profiles are dominated by n-alkanes (*ca* 40%) and monoenes (*ca* 60%) of undetermined double bond location and stereochemistry. No methyl-branched alkanes or dienes were reported. In contrast, the cuticular hydrocarbons of the perilampid are dominated by mono- and dimethylalkanes (*ca* 55 and 35%, respectively), very low proportions of n-alkanes (*ca* 6%) and female-only monoenes (*ca* 6%) of unspecified double bond location and stereochemistry. The *Orasema* sp. hydrocarbons represent a special case in that they are inquilines in ant nests, and appear to obtain at least part of their

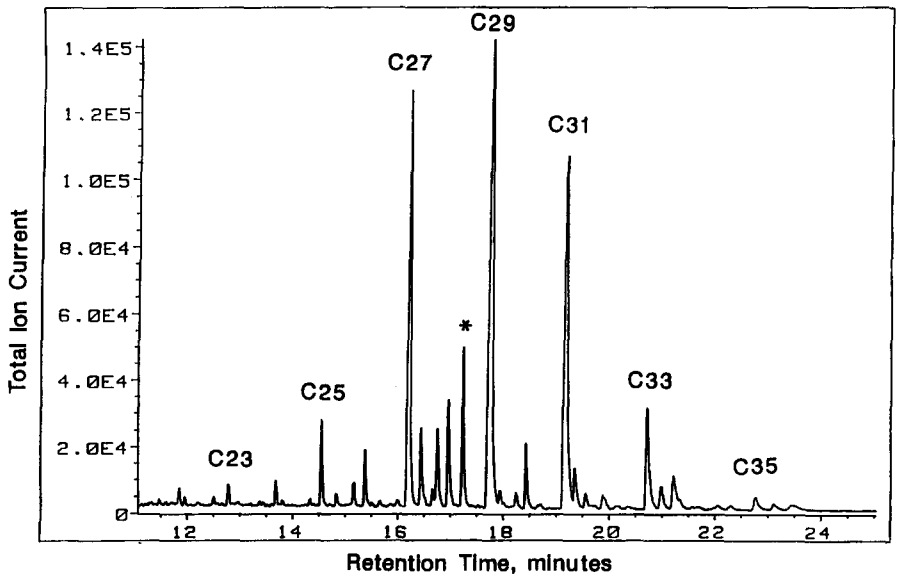

Fig. 3. Total ion chromatogram of the cuticular hydrocarbons from larvae of *Rhyzopertha dominica*. *Non-hydrocarbon contaminant.

Table 3. Identification and mean (SE) per cent composition of cuticular hydrocarbons of larval *Rhyzopertha dominica* ($N = 3$ replicates of 10 larvae each)

Hydrocarbon	ECL*	CN†	Mean % (SE)	Diagnostic EI-MS fragments (m/z)
n-C_{22}	22.00	22	1.74 (0.19)	310
11,15-DiMeC_{21}	22.11	23	Tr‡	99, 169, 239, 309
11-MeC_{22}	22.38	23	1.07 (0.22)	169, 183, 309
3-MeC_{22}	22.71	23	Tr	295, 267, 309
n-C_{23}	23.00	23	1.79 (0.34)	324
3-MeC_{23}	23.73	24	Tr	309, 281, 323
n-C_{24}	24.00	24	1.90 (0.13)	338
11,15-DiMeC_{23}	24.13	25	Tr	169, 211; 141, 239; 337
3-MeC_{24}	24.76	25	Tr	323, 295, 337
n-C_{25}	25.00	25	3.25 (0.28)	352
3-MeC_{25}	25.75	26	1.09 (0.18)	337, 309, 351
n-C_{26}	26.00	26	2.97 (0.20)	366
11,15-DiMeC_{27}	26.14	27	Tr	169, 239, 365
3-MeC_{26}	26.74	27	Tr	351, 323, 365
n-C_{27}	27.00	27	17.09 (1.13)	380
11-;13-MeC_{27}	27.30	28	4.53 (0.89)	169, 253; 197, 225; 379
11,15-DiMeC_{27}	27.59	29	0.74 (0.04)	169, 267, 197, 239; 393
3-MeC_{27}	27.71	28	3.33 (0.37)	337, 351, 379
n-C_{28}	28.00	28	3.83 (0.27)	394
n-C_{29}	29.00	29	23.49 (3.03)	408
11-;13-MeC_{29}	29.27	30	0.72 (0.14)	169, 281; 197, 253; 407
3-MeC_{29}	29.72	30	Tr	393, 365, 407
n-C_{30}	30.00	30	1.84 (0.07)	422
11-;13-MeC_{30}	30.36	31	Tr	169, 295; 197, 267; 421
n-C_{31}	31.00	31	18.65 (0.80)	436
15-MeC_{31}	31.26	32	1.97 (0.10)	225, 253, 435
11,15-DiMeC_{31}	31.52	33	1.04 (0.13)	169, 323, 239, 253; 449
n-C_{32}	32.00	32	0.58 (0.04)	450
n-C_{33}	33.00	33	5.06 (0.48)	464
13-MeC_{33}	33.25	34	1.25 (0.01)	197, 309, 463
11,15-DiMeC_{33}	33.48	35	2.07 (0.40)	169, 351, 239, 281; 477
n-C_{35}	35.00	35	Tr	492
13-MeC_{35}	35.26	36	Tr	197, 337, 477
11,15-DiMeC_{35}	35.52	37	Tr	169, 379, 239, 309; 505

*Equivalent Chain Length.
†Carbon Number, determined by chemical ionization mass spectrometry.
‡Trace (less than 0.1 per cent).

cuticular hydrocarbons from their host ants (*Solenopsis* spp.). Vander Meer *et al.* (1989) note, however, that adult *Orasema* captured outside the fire ant nests have a complex cuticular hydrocarbon profile with hydrocarbons of lower and higher carbon number than those of their host ants. The bark beetle pteromalid parasitoid, *R. pulchripennis*, has an extremely complex mixture of cuticular hydrocarbons, ranging in carbon numbers from C_{25} to C_{37}, and consisting of n-alkanes, 3-methylalkanes, internally branched monomethyl alkanes, 3,X-dimethylalkanes, and 11,15- and 13,17-dimethylalkanes (Espelie *et al.*, 1990). The species occurs in both the eastern and western United States on completely different hosts, and Espelie *et al.* (1990) argue from several considerations, including cuticular hydrocarbon differences, that the two populations represent different species.

One might anticipate that since the two bethylids previously examined (Howard, 1992) and *C. elegans* are all ectoparasitoids of stored product insects that they might share some similarity in their cuticular hydrocarbon profiles. Such is not the case. Adult *C. waterstoni* are characterized by n-alkanes (C_{23}–C_{27}), 2-, 3- and 5-methylalkanes, 5,15-, 5,17-, and 5,19-dimethylalkanes and a series of *Z*-11 monoenes. There are also sexual differences, with the males, but not the females having *Z*-7-monoenes. In addition, there are major qualitative abundance differences between the sexes with males having monoenes as their major components and females having the 5-methylalkanes and dimethylalkanes as their major components. Adult *L. utilis* are characterized by n-alkanes (C_{21}–C_{29}), 2-, 3-, 7-, 9- and 13-methylalkanes and *Z*-9-monoenes. Little sexual dimorphism was found in the cuticular hydrocarbons of adults of *L. utilis*.

All these bethylid profiles are vastly different from those of the pteromalid *C. elegans*, with its dominance of n-alkanes (C_{21}–C_{33}) (*ca* 95% or greater) (Table 2) and only low proportions of 3-, 11- and 13-methylalkanes and even smaller proportions of *Z*-10-alkenes. As with *L. utilis*, no sexual dimorphism was found in the cuticular hydrocarbon profile of *C. elegans*. The hydrocarbons of *C. elegans* are also completely different from those of the only other pteromalid examined, *R. pulchripennis* (Espelie *et al.*, 1990).

Many insects are known in which one or both sexes exist in two or more wing morphs (Wootton, 1992) and such is the case with *C. elegans*. We accordingly investigated the possibility that their might be wing-morph variability in the cuticular hydrocarbons of this species. Four of the n-alkanes (including n-C_{31}, one of the major components) showed significant

abundance differences between the wingless and winged forms. We do not know if these differences reflect wing-specific patterns or whether they reflect whole-body differences. Nothing is known of the differences (if any) in the biology or ecology of these various wing morphs, and in our judgment it is premature to speculate on the functional significance, if any, of these cuticular hydrocarbon wing-morph differences. Similar studies of the cuticular hydrocarbons from other species that show wing polymorphisms are clearly desirable.

Unlike our studies with *C. waterstoni* and *L. utilis*, where we could examine the cuticular hydrocarbon profiles of the immatures of these parasites, the lifestyle of *C. elegans* precludes facile access to the immatures. The hosts of this parasite are internal-feeding larvae of various stored product bettles (Van Den Assem and Keunen, 1958; Sharifi, 1972), and are very difficult to find without dissecting large numbers of grain kernels. Such dissections would likely result in many mutilated wasp larvae, and accordingly we made no efforts at this time to obtain larvae of *C. elegans* for hydrocarbon analysis. We did, however, characterize the cuticular hydrocarbons of the larvae of the lesser grain borer, the host on which we reared our parasites. We note, that although it is possible to induce the beetles to develop in flour, it is not possible to get the parasites to oviposit on these free larvae.

Although the cuticular hydrocarbons of larval *R. dominica* superficially resemble those of the adult pteromalids, they show several distinctive differences. Their n-alkanes make up a smaller proportion (*ca* 85% vs >95%) and they cover a slightly different range of carbon numbers (C_{22} to C_{35} vs C_{21} to C_{33}). In addition, their methylbranched alkanes contain additional points of branching, and more importantly, the beetles possess 11,15-dimethylalkanes and the pteromalid parasite contains none. Finally, the beetle contains no alkenes, whereas *C. elegans* does contain them. We note that the cuticular hydrocarbon profiles of the beetle hosts of the bethylids we earlier examined (Howard, 1992) differ in many ways from both *C. elegans* and *R. dominica*.

The remaining point that we address in this paper is the possibility of the parasites' cuticular hydrocarbons showing variability either from populational differences or from host-specific differences. This possibility was tested by examining the cuticular hydrocarbons from winged females of *C. elegans* from a culture obtained from Savannah, Georgia that had been maintained for several generations on larvae of the rice weevil cultured on whole wheat. The cuticular hydrocarbon compositions of these winged females and winged females from our cultures were nearly identical (Fig. 3). Although our sampling protocol confounds source population and host, the fact that we do not obtain any differences from the insects in our laboratories allows us to state with some confidence that the hydrocarbons we have described are an accurate reflection of the species, and not an artifact of rearing procedures. We note that Espelie *et al.* (1990) also examined the possibility of host-related differences in parasite cuticular hydrocarbon composition in the pteromalid species from Georgia that they studied, and like us, they found no association of host with hydrocarbon composition.

Given the enormous number of parasitic Hymenoptera in the world and the diversity of ecological interactions in which they play a major role, clearly our understanding of the manner in which cuticular

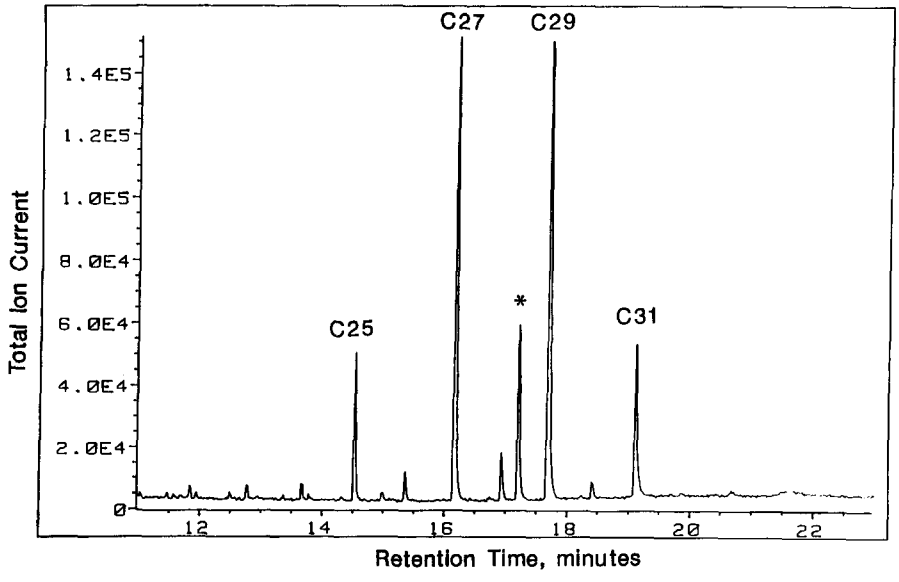

Fig. 4. Total ion chromatogram of the cuticular hydrocarbons of winged female *Choetospila elegans* reared on larvae of *Sitophilus oryzae*, cultured on whole wheat. *Non-hydrocarbon contaminant.

hydrocarbons are involved in their semiochemical interactions is woefully inadequate. Although most of these organisms are extremely small, the literature to date would certainly suggest that they possess sufficient quantities of cuticular hydrocarbons for chemical analysis. Given the known response of various parasitoids to kairomonal cuticular hydrocarbons from their hosts (Howard and Blomquist, 1982; Howard, 1993), it is to be expected that they could perceive and use hydrocarbons in a diversity of other semiochemical functions as do many other insects (Howard, 1993). Future studies will be needed to evaluate this prediction.

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