Cuticular Hydrocarbons of Adult *Pteromalus cerealellae* (Hymenoptera: Pteromalidae) and Two Larval Hosts, Angoumois Grain Moth (Lepidoptera: Gelechiidae) and Cowpea weevil (Coleptera: Bruchidae)

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ABSTRACT The pteromalid parasitoid *Pteromalus cerealellae* (Ashmead) is nominally a host-specific ectoparasitoid of the stored-product pest the Angoumois grain moth. It is readily cultured, however, on a number of other stored-product pests, including the cowpea weevil. The parasitoids resulting from these different hosts are disparate in size, leading me to ask if the cuticular hydrocarbons of *P. cerealellae* reared on different hosts would be different. This question is of importance because many parasitoids of stored-product pests use cuticular hydrocarbons as major species- and gender-recognition cues, and moth and beetle hosts differ greatly in their hydrocarbon profiles. The parasitoid hydrocarbon profile reported here was much more complex than the profiles of either host and showed both gender and host effects. Parasitoids reared on the larger cowpea weevils were larger than those reared on moths and contained substantially more hydrocarbon on their cuticle. Regardless of host, female wasps were always larger than males and contained more hydrocarbon than males. The hydrocarbon chemistry of wasps reared on different hosts were qualitatively the same, but quantitatively different. Both gender and host are important in the relative abundance of individual hydrocarbons. There are four major hydrocarbons of females constituting 49% of the total female profile: 3-MeC_{29}, 11-MeC_{31}, 3, 7-$DiMeC_{31}$ and 11, 17-, 11, 19- and 11, 21–$DiMeC_{33}$. There is one major male-specific hydrocarbon comprising 18% of the total hydrocarbons, 3, 7- and 3, 9–$DiMeC_{33}$. Although major gender differences occur between hydrocarbon classes, these differences are not host-related.

KEY WORDS *Pteromalus cerealellae*, Angoumois grain moth, cowpea weevil, parasitoid, chemical ecology, stored-product pest

THE ACULEATE FAMILY Pteromalidae is important 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. I am aware of only two previous reports on cuticular hydrocarbons in this family. Espelie et al. (1990) reported on the chemotaxonomic separation of two apparent sibling species that are parasitoids of bark beetles, and Howard and Liang (1993) reported on the winged and wingless morphs of *Choetospila elegans* Westwood [now *Theocolax elegans* (Westwood)], an ectoparasitoid of several stored-product insect pests. As part of a larger study on the chemical ecology of stored grain pests and their predators and parasites, Howard (1992) reported on the cuticular hydrocarbon composition of *Cephalonomia waterstoni* Gahan and *Laelius utilis* Cockerell (Bethylidae) and their respective beetle hosts, *Cryptolestes ferrugineus* (Stephens) (Cucujidae) and *Trogoderma variable* Ballion (Dermestidae); Howard (1998) reported on the hydrocarbons of *Cephalonomia tarsalis* (Ashmead) and its cucujid host, the sawtoothed grain beetle; and Howard and Infante (1996) described the cuticular hydrocarbon composition of *Cephalonomia stephanoderis* Betrem and its host, the scolytid beetle *Hypothenemus hampei* (Ferrari).

In this article I report on the cuticular hydrocarbon composition of *Pteromalus cerealellae* (Ashmead), a nominally host-specific parasitoid of the Angoumois grain moth, *Sitotroga cerealellae* (Olivier), but in actuality, a parasitoid that readily accepts numerous stored-product insects, including the cowpea weevil, as hosts (Brower 1991).

Materials and Methods

Insects. Cultures of *P. cerealellae* (the parasite) and *S. cerealellae* and *Callosobruchus maculatus* (F.) (the hosts) were obtained from the former USDA-ARS Stored-Product Insects Research and Development Laboratory, Savannah, GA, and have been in laboratory culture for >25 yr. Parasites to be used for experiments were collected from stock cultures as newly emerged adults, sexed and placed in vials with a cotton ball soaked with 50% honey-water solution which was rewetted daily. Parasitoids were held for ≈3 to 7 d before being sacrificed for analysis. Larval moths and weevils were wandering phase individuals taken from

cultures, transferred to vials, and were sacrificed within 24 h. Vials with insects were held in a rearing chamber at 30 ± 1°C and 50 ± 10% RH and a photoperiod of 16:8 (L:D) h.

Chemical Analyses. Insects were frozen at -5°C for 24 h. 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 each sample were concentrated under a stream of N_2, and hydrocarbons were isolated by chromatography on a 3-cm "mini-column" of BioSil A (Bio-Rad, Richmond, CA) as described earlier (Howard et al. 1978). The parasitoids are very small insects, containing insufficient hydrocarbon for single insect analyses, so samples consisted of five insects per replicate, and two to four replicates of each experimental treatment were analyzed. The larval moth and beetle hosts were larger, so samples were three insects per replicate, and three replicates of each were analyzed.

Electron impact mass spectral analyses were conducted using a Hewlett–Packard 5790A gas chromatograph (GC) (Hewlett–Packard, San Fernando, CA) containing a DB-5 bonded phase capillary column (15 m long, 0.25 mm i.d.) (J and W Scientific, Folsom, CA) connected to a Hewlett–Packard 5970 mass selective detector (MSD) 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 80°C, a final temperature of 320°C, a program rate of 5°C/min, 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 in the total ion scanning mode were identified from their characteristic EI-MS fragmentation patterns (Jackson and Blomquist 1976, Nelson 1978). Quantitative analyses were conducted using the selective ion scanning mode (SIM), m/z 57 being the target ion and an internal standard of 50.2 ng/μl of heptadecane.

Effects of Host and Gender on *P. cerealellae* Cuticular Hydrocarbons. The 2 × 2 factorial design compared male and female wasps reared on either larvae of the Angoumois grain moth or the cowpea weevil. All wasps were virgin and were killed at 1 wk of age.

Weight Measurements of P. cerealellae, *S. cerealellae*, and *C. maculatus*. Female and male *P. cerealellae* reared from Angoumois grain moth and cowpea weevil hosts were killed by freezing, dried 24 h at 105°C, and weighed on a Mettler UMT2 Microbalance (Mettler Scientific, Toledo, OH). Larval hosts were similarly killed, oven-dried, and weighed.

Statistical Analyses. Compositional analyses of hydrocarbons were conducted using the EI-MS system as described above. Area counts obtained from electronic integration in the SIM mode were summed and used as ng per insect values or they were converted into percent values. Each run had an internal standard of 50.2 ng/μl each of heptadecane. For multifactorial analysis of variance (ANOVA), nanogram values were used. Significantly different means were separated using the least significant difference test at the 95% confidence level. Reported means are on both absolute and percent values. One way ANOVA was conducted on the oven-dry weights of the parasitoids and their hosts. All statistical analyses were conducted using the personal computer software program Statgraphics Plus (Statgraphics 1998).

Voucher Specimens. Voucher specimens of *P. cerealellae, S. cerealellae,* and *C. maculatus* (Lot Nos. 111 and 112) have been deposited in the Kansas State University Museum of Entomological and Prairie Arthropod Research, Manhattan, KS.

Results

Male and female *P. cerealellae* yielded 62 identified cuticular hydrocarbon components and seven unidentified minor components. These included n-alkanes (C_{27}–C_{32}), a homologous series of 3-, 4-, 5-, 7-, 9-, 11-, 13-, 15- and 17-methyl alkanes, a homologous series of 3,7- and 3,9-dimethyl alkanes, a series of internal dimethyl alkanes (7, 11-; 9, 13-; 9, 15-; 11, 15-; 11, 16-; 11, 17-; 11, 18-; 11, 19-; 11, 20-; 13, 17-; 13, 19-; 13, 21-), and one trimethyl alkane (Table 1). In contrast, one larval host, the Angoumois grain moth, yielded only 31 hydrocarbon components. These included n-alkanes (C_{25}–C_{31}), a homologous series of monomethyl alkanes (3-, 4-, 7-, 9-, 11-, 13-, 15-) and a series of internally branched dimethyl alkanes (8, 12-; 10, 14-; and 11, 15-) (Table 2). The other larval host, the cowpea weevil, contained 57 identified hydrocarbon components and one unidentified component. These included n-alkanes (C_{25} - C_{32}), homologous monomethyl alkanes (3-, 4-, 5-, 6-, 7-, 9-, 11-, 13-, 14-, 15-), a series of terminal dimethyl alkanes (3, 7-; 3, 9-; 3, 11-; 3, 13-), and a series of internal dimethyl alkanes (7, 11-; 8, 12-; 9, 13-; 10, 14-; 11, 15-) (Table 3). Neither the wasps nor hosts contained any unsaturated hydrocarbons. Only 12 hydrocarbons were shared by all three species, and for the wasp these 12 components ranged in abundance from trace to ≈4% of their total hydrocarbons. The 12 hydrocarbons held in common are five n-alkanes (C_{27} - C_{31}), 9-Me C_{27}, 3-, 11-, and 13-Me C_{29}, 11-, and 13-Me C_{31} and 11, 15-DiMe C_{31}. The parasitoid and the moth larval host shared 13 hydrocarbon components, the parasitoid and beetle larvae shared 24 hydrocarbon components, and the moth and beetle shared 22 hydrocarbon components.

Although the parasitoid had the same cuticular hydrocarbons regardless of host, the relative abundance of each component varied as a function of both gender and host (Table 4). Forty-seven hydrocarbon peaks contained the 69 hydrocarbon components identified by mass spectrometry, and statistical comparisons were made accordingly. Thirty-one of the 47 peaks differed between males and females, 35 differed as a function of host, and 10 of the peaks showed gender by host interactions. In contrast, the absolute total quantity of hydrocarbons on the wasps did not differ

Table 1. Cuticular hydrocarbons of *Pteromalus cerealellae* adults

Hydrocarbon	ECL	CN	Diagnostic EI-MS ions
n-C_{27}	27.00	27	380
9-MeC_{27}	27.32	28	141, 280; 379 (M-15)
7-MeC_{27}	27.40	28	113, 309; 379 (M-15)
5-MeC_{27}	27.46	28	85, 337; 379 (M-15)
3-MeC_{27}	27.69	28	365, 337; 379 (M-15)
n-C_{28}	28.00	28	394
3,7-DiMeC_{27}	28.01	29	127, 309, 379; 393 (M-15)
10-MeC_{28}	28.30	29	155, 280; 393 (M-15)
7-MeC_{28}	28.40	29	113, 323; 393 (M-15)
5-MeC_{28}	28.47	29	85, 351; 393 (M-15)
3-MeC_{28}	28.70	29	379, 351; 393 (M-15)
n-C_{29}	29.00	29	408
11-MeC_{29}	29.30	30	169, 280; 407 (M-15)
13-MeC_{29}	29.30	30	197, 253; 407 (M-15)
9,13-DiMeC_{29}	29.61	31	141, 323, 211, 253; 421 (M-15)
7,11-DiMeC_{29}	29.61	31	113, 351, 183, 280; 421 (M-15)
3-MeC_{29}	29.73	30	393, 365; 407 (M-15)
n-C_{30}	30.00	30	422
3,7-DiMeC_{29}	30.01	31	113, 337, 407; 421 (M-15)
9-MeC_{30}	30.29	31	141, 323; 421 (M-15)
11-MeC_{30}	30.29	31	169, 295; 421 (M-15)
4-MeC_{30}	30.61	31	71, 393; 421 (M-15)
3-MeC_{30}	30.70	31	407, 379; 421 (M-15)
n-C_{31}	31.00	31	436
11-MeC_{31}	31.29	32	169, 309; 197, 280; 435 (M-15)
13-MeC_{31}	31.29	32	197, 280; 435 (M-15)
11,15-DiMeC_{31}	31.61	33	169, 323, 239, 253; 449 (M-15)
11,17-DiMeC_{31}	31.61	33	169, 323, 225, 267; 449 (M-15)
11,19-DiMeC_{31}	31.61	33	169, 323, 197, 295; 449 (M-15)
9,13-DiMeC_{31}	31.72	33	141, 351, 211, 280; 449 (M-15)
9,15-DiMeC_{31}	31.72	33	141, 351, 239, 253; 449 (M-15)
3-MeC_{31}	31.82	32	421, 393; 435 (M-15)
3,7-DiMeC_{31}	32.06	33	127, 365, 435; 449 (M-15)
3,9-DiMeC_{31}	32.06	33	155, 337, 435; 449 (M-15)
n-C_{32}	32.00	32	450
9-MeC_{32}	32.29	33	141, 351; 169, 323; 449 (M-15)
11-MeC_{32}	32.29	33	169, 323; 449 (M-15)
9,13-DiMeC_{32}	32.72	34	141, 365, 211, 295; 463 (M-15)
9,17-DiMeC_{32}	32.72	34	141, 365, 239, 267; 463 (M-15)
11,16-DiMeC_{32}	32.61	34	169, 337, 253; 463 (M-15)
11,18-DiMeC_{32}	32.61	34	169, 337, 225, 280; 463 (M-15)
11,20-DiMeC_{32}	32.61	34	169, 337, 197, 309; 463 (M-15)
Unknown 1	32.81	—	—
Unknown 2	33.00	—	—
Unknown 3	33.10	—	—
Unknown 4	33.15	—	—
11-MeC_{33}	33.30	34	169, 337; 463 (M-15)
13-MeC_{33}	33.30	34	197, 309; 463 (M-15)
15-MeC_{33}	33.30	34	225, 280; 463 (M-15)
17-MeC_{33}	33.30	34	253; 463 (M-15)
11,17-DiMeC_{33}	33.61	35	169, 351, 253, 267; 477 (M-15)
11,19-DiMeC_{33}	33.61	35	169, 351, 225, 295; 477 (M-15)
11,21-DiMeC_{33}	33.61	35	169, 351, 197, 323; 477 (M-15)
3-MeC_{33}	33.75	34	449, 421; 463 (M-15)
3,7-DiMeC_{33}	34.05	35	463, 127, 393; 477 (M-15)
3,9-DiMeC_{33}	34.05	35	463, 155, 365; 477 (M-15)
3,7,11-TriMeC_{33}	34.29	36	477, 127, 407, 197, 337; 491 (M-15)
Unknown 5	34.61	—	—
Unknown 6	34.90	—	—
11-MeC_{35}	35.30	36	169, 365; 491 (M-15)
13-MeC_{35}	35.30	36	197, 337; 491 (M-15)
15-MeC_{35}	35.30	36	225, 309; 491 (M-15)
13,17-DiMeC_{35}	35.62	37	197, 351, 267, 280; 505 (M-15)
13,19-DiMeC_{35}	35.62	37	197, 351, 253, 295; 505 (M-15)
13,21-DiMeC_{35}	35.62	37	197, 351, 225, 323; 505 (M-15)
3-MeC_{35}	35.72	36	477, 449; 491 (M-15)
3,7-DiMeC_{35}	36.05	37	491, 127, 421; 505 (M-15)
3,9-DiMeC_{35}	36.05	37	491, 155, 393; 505 (M-15)
Unknown 7	37.49	—	—

ECL, equivalent chain length; CN, carbon number.

significantly as a function of gender or host. The mean quantity of hydrocarbon on female and male wasps reared on the Angoumois grain moth was 575 and 260 ng, respectively ($n = 2$), and for wasps reared on the cowpea weevil, the values are 808 ng for females and 377 ng for males ($n = 4$). Comparison of these means by ANOVA indicated no significant differences for either gender ($F = 4.07$, $P = 0.0785$) or host ($F = 0.89$, $P = 0.3724$), and no significant interactions ($F = 0.10$, $P = 0.7604$). A summary of these absolute values is

Table 2. Cuticular hydrocarbons of Angoumois grain moth larvae

Hydrocarbon	ECL	CN	Diagnostic EI-MS ions
n-C_{25}	25.00	25	352
n-C_{26}	26.00	26	366
4-MeC_{26}	26.51	27	71, 337; 365 (M-15)
n-C_{27}	27.00	27	380
13-MeC_{27}	27.28	28	197, 225; 379 (M-15)
11-MeC_{27}	27.28	28	169, 253; 379 (M-15)
9-MeC_{27}	27.30	28	141, 281; 379 (M-15)
11,15-DiMeC_{27}	27.58	29	169, 267, 197, 239; 393 (M-15)
n-C_{28}	28.00	28	394
14-MeC_{28}	28.28	29	211, 225; 393 (M-15)
13-MeC_{28}	28.28	29	197, 239; 393 (M-15)
12-MeC_{28}	28.28	29	183, 253; 393 (M-15)
4-MeC_{28}	28.51	29	71, 365; 393 (M-15)
n-C_{29}	29.00	29	408
15-MeC_{29}	29.28	30	225; 407 (M-15)
13-MeC_{29}	29.28	30	197, 253; 407 (M-15)
11-MeC_{29}	29.28	30	169, 281; 407 (M-15)
9-MeC_{29}	29.30	30	141, 309; 407 (M-15)
7-MeC_{29}	29.32	30	113, 337; 407 (M-15)
13,17-DiMeC_{29}	29.58	31	197, 267; 421 (M-15)
3-MeC_{29}	29.67	30	393, 365; 407 (M-15)
n-C_{30}	30.00	30	422
10,14-DiMeC_{30}	30.46	32	155, 323, 225, 253; 435 (M-15)
8,12-DiMeC_{30}	30.46	32	127, 351, 197, 281; 435 (M-15)
4-MeC_{30}	30.51	31	71, 393; 421 (M-15)
n-C_{31}	31.00	31	436
15-MeC_{31}	31.28	32	225, 253; 435 (M-15)
13-MeC_{31}	31.28	32	197, 281; 435 (M-15)
11-MeC_{31}	31.28	32	169, 309; 435 (M-15)
9-MeC_{31}	31.30	32	141, 337; 435 (M-15)
11,15-DiMeC_{31}	31.60	33	169, 323, 239, 253; 449 (M-15)

ECL, equivalent chain length; CN, carbon number.

Table 3. Cuticular hydrocarbons of larval cowpea weevils

Hydrocarbon	ECL	CN	Diagnostic ion fragments m/z
C_{25}	25.00	25	352
3-MeC_{25}	25.68	26	337, 309; 351 (M-15)
C_{26}	26.00	26	366
4-MeC_{26}	26.51	27	71, 337; 365 (M-15)
3-MeC_{26}	26.67	27	351, 323; 365 (M-15)
C_{27}	27.00	27	380
9-MeC_{27}	27.30	28	141, 281; 379 (M-15)
7-MeC_{27}	27.32	28	113, 309; 379 (M-15)
5-MeC_{27}	27.43	28	85, 337; 379 (M-15)
9,15-DiMeC_{27}	27.59	29	141, 295, 197, 239; 393 (M-15)
3-MeC_{27}	27.69	28	365, 337; 379 (M-15)
C_{28}	28.00	28	380
3,7-DiMeC_{27}	28.05	29	379, 127, 309; 393 (M-15)
3,9-DiMeC_{27}	28.05	29	379, 155, 281; 393 (M-15)
10-MeC_{28}	28.26	29	155, 281; 393 (M-15)
6-MeC28	28.30	29	99, 337; 393 (M-15)
4-MeC_{28}	28.50	29	71, 365; 393 (M-15)
9,13-DiMeC_{28}	28.58	30	141, 309, 211, 239; 407 (M-15)
7,11-DiMeC_{28}	28.61	30	113, 337, 169, 281; 407 (M-15)
3-MeC_{28}	28.64	29	379, 351; 393 (M-15)
C_{29}	29.00	29	408
15-MeC_{29}	29.28	30	225; 407 (M-15)
13-MeC_{29}	29.28	30	197, 253; 407 (M-15)
11-MeC_{29}	29.28	30	169, 281; 407 (M-15)
9-MeC_{29}	29.30	30	141, 309; 407 (M-15)
7-MeC_{29}	29.32	30	113, 337; 407 (M-15)
5-MeC_{29}	29.39	30	85, 365; 407 (M-15)
9,13-DiMeC_{29}	29.49	31	141, 323, 211, 253; 421 (M-15)
7,11-DiMeC_{29}	29.60	31	113, 351, 169, 281; 421 (M-15)
3-MeC_{29}	29.68	30	393, 365; 407 (M-15)
C_{30}	30.00	30	422
3,9-DiMeC_{29}	30.05	31	407, 155, 309; 421 (M-15)
3,11-DiMeC_{29}	30.05	31	407, 183, 281; 421 (M-15)
3,13-DiMeC_{29}	30.05	31	407, 211, 253; 421 (M-15)
14-MeC_{30}	30.28	31	211, 253; 421 (M-15)
12-MeC_{30}	30.28	31	183, 281; 421 (M-15)
8,12-DiMeC_{30}	30.48	32	127, 351, 197, 281; 435 (M-15)
9,13-DiMeC_{30}	30.48	32	141, 337, 211, 267; 435 (M-15)
10,14-DiMeC_{30}	30.48	32	155, 323, 225, 253; 435 (M-15)
C_{31}	31.00	31	436
15-MeC_{31}	31.28	32	225, 253; 435 (M-15)
13-MeC_{31}	31.28	32	197, 281; 435 (M-15)
11-MeC_{31}	31.28	32	169, 309; 435 (M-15)
9,13-DiMeC_{31}	31.46	33	141, 351, 211, 281; 449 (M-15)
11,15-DiMeC_{31}	31.46	33	169, 323, 239, 253; 449 (M-15)
7,11-DiMeC_{31}	31.59	33	113, 379, 183, 309; 449 (M-15)
3-MeC_{31}	31.66	32	421, 393; 435 (M-15)
C_{32}	32.00	32	450
12-MeC_{32}	32.28	33	183, 309; 449 (M-15)
10,14-DiMeC_{32}	32.46	34	155, 351, 225, 281; 463 (M-15)
unknown	32.88		
13-MeC_{33}	33.28	34	197, 309; 463 (M-15)
9,13-DiMeC_{33}	33.48	35	141, 379, 211, 309; 477 (M-15)
11,15-DiMeC_{33}	33.48	35	169, 351, 239, 281; 477 (M-15)
7,11-DiMeC_{33}	33.60	35	113, 407, 183, 337; 477 (M-15)
13-MeC_{35}	35.29	36	197, 337; 491 (M-15)
9,13-DiMeC_{35}	35.48	37	141, 407, 211, 337; 505 (M-15)
11,15-DiMeC_{35}	35.48	37	169, 379, 239, 309; 505 (M-15)

ECL, equivalent chain length; CN, carbon number.

Table 4. Percent composition of the cuticular hydrocarbons of *Pteromalus cerealellae* as a function of gender (F is female and M is male) and larval host (AGM is the Angoumois grain moth and CPW is the cowpea weevil)

Hydrocarbon	AGM-F	CPW-F	AGM-M	CPW-(M-15)
n-C_{27}	0.54	1.30	0.39	1.23
9-MeC_{27}	0.05	0.27	Tr[1]	0.21
7-MeC_{27}	Tr	Tr	Tr	Tr
5-MeC_{27}	Tr	0.14	Tr	0.12
3-MeC_{27}	0.28	1.22	0.06	1.02
n-C_{28}	0.24	0.22	Tr	0.19
3,7-DiMeC_{27}	0.02	0.22	Tr	0.15
10-MeC_{28}	Tr	0.13	Tr	0.11
7-MeC_{28}	Tr	Tr	Tr	0.04
5-MeC_{28}	0.12	Tr	Tr	Tr
3-MeC_{28}	0.09	0.13	Tr	0.04
n-C_{29}	3.67	3.23	1.15	1.68
11-;13-MeC_{29}	0.92	3.00	0.28	1.71
9,13-DiMeC_{29}	Tr	2.83	Tr	2.11
7,11-DiMeC_{29}	0.06	1.15	Tr	0.85
3-MeC_{29}	12.34	11.24	0.64	1.54
n-C_{30}	0.25	0.44	Tr	0.19
3,7-DiMeC_{29}	0.95	1.10	Tr	0.31
9-;11-MeC_{30}	0.32	0.51	Tr	0.28
4-MeC_{30}	0.53	0.66	Tr	0.26
3-MeC_{30}	0.32	0.29	Tr	Tr
n-C_{31}	1.20	1.32	1.04	1.11
11-;13-MeC_{31}	6.14	5.33	3.03	2.42
11,15-;11,17-;11,19-DiMeC_{31}	3.78	2.38	1.56	1.02
9,13-;9,15-DiMeC_{31}	Tr	1.75	0.45	1.15
3-MeC_{31}	0.22	0.46	Tr	0.20
3,7-;3,9-DiMeC_{31}	6.69	6.45	0.93	0.69
n-C_{32}	0.10	0.16	0.18	0.21
9-;11-MeC_{32}	1.57	1.21	2.53	2.24
9,13-;9,17-DiMeC_{32}	1.26	1.14	1.15	0.96
11,16-;11,18-;11,20-DiMeC_{32}	2.51	2.16	1.31	1.07
unknown 1	0.12	0.10	0.20	0.18
unknown 2	0.04	Tr	0.49	0.57
unknown 3	0.20	0.26	1.48	1.68
unknown 4	Tr	Tr	0.54	0.52
11-;13-;15-;17-MeC_{33}	10.87	9.70	12.71	11.45
11,17-;11,19-;11,21-DiMeC_{33}	27.23	23.86	24.67	20.88
3-MeC_{33}	2.82	2.59	6.51	6.44
3,7-;3,9-DiMeC_{33}	2.29	1.80	18.39	17.66
3,7,11-TriMeC_{33}	1.15	0.91	3.18	2.87
unknown 5	1.52	1.36	1.57	1.40
unknown 6	Tr	Tr	0.60	0.59
11-;13-;15-MeC_{35}	1.23	1.29	2.11	1.74
13,17-;13,19-;13,21-DiMeC_{35}	6.91	6.38	10.34	8.73
3-MeC_{35}	0.38	0.34	0.78	0.70
3,7-;3,9-DiMeC_{35}	0.23	0.14	0.72	0.61
Unknown 7	0.84	0.84	1.03	0.86

All values are means for four replicates.

found in Table 5, with hydrocarbons grouped by class. A comparative proportional analysis is found in Table 6.

In many insects, males and females have one or more gender specific cuticular hydrocarbons. *Pteromalus cerealellae* is no exception. Both absolute and proportional values for several hydrocarbons that differ significantly between the male and female parasitoids are shown in Table 7. There are four major female derived hydrocarbons constituting ≈49% of their total hydrocarbons: 3-Me C_{29}, 11-Me C_{31}, 3,7-DiMe C_{31}, and 11, 15- DiMe C_{33}. There is one major group of male specific hydrocarbons, representing ≈18% of its total hydrocarbons: 3,7- and 3, 9-Di Me C_{33}. These sex-specific differences are independent of source host.

The mean weights for wasps reared on the Angoumois grain moth were 0.335 mg for females (n = 25) and 0.150 mg for males (n = 25); the mean weights for wasps reared on the cowpea weevils were 0.422 mg

Table 5. Mean absolute hydrocarbon composition (ng) of *P. cerealellae* by hydrocarbon class as a function of gender and larval host

Hydrocarbon class	Gender and larval host			
	♀ AGM	♀ CPW	♂ AGM	♂ CPW
n-alkanes	26.2	53.5	5.2	17.5
monomethyl alkanes	174.8	312.0	58.3	115.6
Dimethyl alkanes	239.8	414.4	121.6	220.5
Trimethyl alkanes	5.3	7.5	6.6	11.0
Unknowns	12.7	20.1	12.0	21.8
Total HC	458.8	807.5	203.7	386.4

AGM, Angoumois grain moth; CPW, cowpea weevil.

for females ($n = 24$) and 0.218 mg for males ($n = 25$). Analysis of these distributions by ANOVA indicated that the wasp oven-dry weights differed significantly by both gender ($F = 249.91, P < 0.0001$) and host ($F = 39.61$, ($P < 0.0001$), with no significant interaction term ($F = 0.48, P = 0.4879$). The oven-dry weights for the two host larvae were 2.489 mg for the Angoumois moth ($n = 50$) and 3.869 mg for the cowpea weevil ($n = 50$). Analysis of these distributions by ANOVA indicated that the two species differed significantly ($F = 92.48, P < 0.0001$).

Discussion

Cuticular hydrocarbons are increasingly 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, Blomquist et al. 1998). Much of the early literature focused 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 focused on semiochemical roles and chemosystematic applications (Howard 1993). Although well over 400 papers on the cuticular hydrocarbons of arthropods have appeared, many important taxa remain unstudied. The order Hymenoptera is large and speciose, and has representatives in virtually every biome. Few studies have been conducted on members of this order, however, with most of those being on the ants, bees, and social wasps.

Table 6. Mean percent hydrocarbon composition of *P. cerealellae* by hydrocarbon class as a function of gender and larval host

Hydrocarbon class	Gender and larval host			
	♀ AGM	♀ CPW	♂ AGM	♂ CPW
n-alkanes	5.7	6.6	2.6	4.5
Monomethyl alkanes	38.1	38.6	28.6	29.9
Dimethyl alkanes	52.3	51.3	59.7	57.1
Trimethyl alkanes	1.2	0.9	3.2	2.8
Unknowns	2.8	2.5	6.4	5.6

AGM, Angoumois grain moth; CPW, cowpea weevil.

Table 7. Major gender-specific cuticular hydrocarbons of *P. cerelellae*

Compound	%♀	%♂	ng ♀	ng ♂
3-MeC_{29}	12	1	75	3
11-MeC_{31}	5.5	2.8	35	7
3,7-$DiMeC_{31}$	6.7	0.8	40	2
11,X-$DiMeC_{33}$	25	22	150	60
3,X-$DiMeC_{33}$	2	18	12	50

Within the Hymenoptera, the parasitic taxa make up a large proportion of the known species, yet the number of papers on their cuticular hydrocarbons is very small (Lockey 1988, Howard 1993). Indeed, *P. cerealellae* is only the eleventh species examined for these phenotypic characters. Other species for which cuticular hydrocarbons have been reported include *Cardiochiles nigriceps* (Viereck) (Braconidae) (Syvertsen et al. 1995), the codling moth parasitoid *Ascogaster quadridentata* Wesmael (Braconidae) (Espelie and Brown 1990) and its hyperparasitoid *Perilampus fulvicornis* Ashmead (Perilampidae) (Espelie and Brown 1990); an *Orasema* sp. (Eucharitidae), an inquiline of the red imported fire ant (Vander Meer et al. 1989); four bethylids, *C. waterstoni*, *C. tarsalis*, *C. stephanoderis* and *L. utilis*, all ectoparasitoids of stored-product beetles (Howard 1992, 1998; Howard and Infante 1996); *Rhopalicus pulchripennis* (Crawford) (Pteromalidae), a parasitoid of bark beetles; and *T. (Choetospila) elegans*, also an ectoparasitoid of stored-product beetles (Howard and Liang 1993). In addition, I have conducted a preliminarily analysis of the hydrocarbons of another stored-product pteromalid, *Anisopteramalus calandrae* (Howard) (unpublished data). 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 species are all rather different. A comparison of the similarities and differences for all species except *C. tarsalis*, *C. stephanoderis* and *P. cerealellae* is to be found in Howard and Liang (1993) and is not repeated here.

The hydrocarbon compositions of the two previously examined pteromalid spp. differ substantially from each other and from *P. cerealellae.* The bark beetle parasitoid, *R. pulchripennis*, has a complex mixture of cuticular hydrocarbons, ranging in carbon numbers from C_{25} to C_{37}, and consisting of n-alkanes, 3-methyl alkanes, internally branched monomethyl alkanes, 3,X-dimethyl alkanes, and 11,15- and 13,17-dimethyl alkanes (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. In contrast, *T. (Choetospila) elegans* has a much simpler hydrocarbon composition, with its dominance of n-alkanes (C_{21}–C_{33}) (≈95% or greater), and only low proportions of 3-, 11-, and 13-methyl alkanes and even smaller proportions of Z-10-alkenes (Howard and Liang 1993). The hydrocarbons of *P. cerealellae* resemble those of *R. pulchrip-*

ennis much more than they do *T. (Choetospila) elegans.* The hydrocarbons of *P. cerealellae* comprise a slightly smaller range of carbon numbers (C_{27}–C_{37}) than the bark beetle parasitoid, but have a greater diversity of mono-, di-, and trimethyl alkanes (Table 1). Like the bark beetle parasitoid, they also contain no unsaturated components.

Like *T.* (*Choetospila*) *elegans,* the parasitoid *P. cerealellae* can be successfully cultured on several different hosts. In our earlier study (Howard and Liang 1993) we found that the hydrocarbon profiles of the parasitoid were essentially identical regardless of whether the wasp had been reared on the bostrichid *Rhyzopertha dominica* (F.) or on the curculionid *Sitophilus oryzae* (L.). Similarly, Espelie et al. (1990) also examined the possibility of host-related differences in the pteromalid from Georgia that they studied, and found no association of host with hydrocarbon composition. Here, although the chemicals present on the parasitoid were the same regardless of host, the absolute quantity of hydrocarbon and the relative proportions of individual hydrocarbons were host-related (Tables 5 and 6). The hydrocarbon profiles of the two hosts as noted earlier were very different from each other (Table 2 and 3), and both differed from that of the parasitoid. A clear trend is emerging in studies correlating the hydrocarbon composition of insects that are reared on other insects: there is little or at most limited correspondence between the cuticular hydrocarbon profiles of the hosts and that of the predator or parasitoid. Although few biosynthetic experiments have been conducted to directly show that the predator or parasitoid makes all of their own hydrocarbons rather than acquiring them from host tissues, there is an overwhelming preponderance of circumstantial evidence to suggest that they do. A similar situation exists for herbivorous insects. The only exception to this trend is where the predator or parasitoid is an inquiline of social insects, and in this case often a very high correspondence is found in the hydrocarbon profiles (Vander Meer et al. 1989; Howard et al. 1990a, 1990b; Howard 1993; Howard and Akre 1995).

In many insect species, there are gender-related differences in the cuticular hydrocarbon composition (Howard 1993), including several of the parasitoids associated with the stored-product environment (Howard 1992, 1998). Espelie et al. (1990) did not report their data as a function of gender, but Howard and Liang (1993) found no gender differences for the hydrocarbons of the stored-product pteromalid *T.* (*Choetospilai*) *elegans.* In contrast, *P. cerealellae* males and females have several gender-based differences that might serve as gender recognition cues, and these components are not affected by the host on which the parasitoid was reared (Table 7). As noted earlier, male and female wasps differ substantially in size, and although the absolute differences are clearly related to the size of the host that they were reared on, the total amount of hydrocarbon on their cuticle was not strongly related to host size. This consistency of cuticular hydrocarbon profile has important implications for semiochemical function. In preliminary experiments, I have found no evidence of different courtship behavior by wasps reared from either host; nor have I found any evidence of sexual preference among wasps as a function of host (unpublished data). Although the relevant experiments have yet to be conducted, I predict that as I found earlier for *C. tarsalis* (Howard 1998), the gender-specific hydrocarbons found on *P. cerealellae* will prove to be both species- and gender-recognition cues used in the final stages of courtship.

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