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Caste-specific cuticular lipids in the stingless bee *Friesella schrottkyi**

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Abstract – While a queen control pheromone complex that inhibits worker ovary development has been described for honey bees, no comparable control pheromones have been identified for their sister group, the stingless bees. The aim of the present work was to search for possible control pheromones in the stingless bee *Friesella schrottkyi*. No volatile substances were found in the heads of queens that might serve as queen control pheromones. On the other hand, distinct differences were found between the cuticular substances of queens and workers. The major hydrocarbons were different between the two castes, and while queens contained methyl-branched alkanes and no unsaturated hydrocarbons, workers contained alkenes and alkadienes but no methyl branched hydrocarbons. Colonies deprived of a queen produced laying workers. Differences were observed in the cuticular patterns of laying workers and workers from a queen controlled colony.

pheromone / queen control / laying workers / caste differences / hydrocarbons

1. INTRODUCTION

In social insects with small colonies, the oviposition dominance of the queen is sustained by physical intimidation which results in a dominance hierarchy between the queen and her subordinates or workers. However, when the colonies are composed of thousands of individuals, such as in honey bees, physical dominance becomes unlikely, or impossible. In these colonies, regulation of worker oviposition is mediated by substances produced by the queen (reviewed by Wilson, 1971; Michener, 1974; Hölldobler and Wilson, 1990). Thus, honey bee workers show atrophic or reduced ovaries in the queen's presence, while in her absence, worker ovaries develop and these workers rapidly start to lay eggs that will become males (Robinson et al., 1990). In honey bees, the pheromone complex that inhibits worker ovary development is produced by the queen's mandibular and cephalic glands, and is called QRP (queen retinue pheromone). The mixture is composed basically of five substances: (*E*)-9-keto-2-decenoic acid (9ODA), (*R*,*E*)-(-)- and (*S*,*E*)-(+)-9-hydroxy-2-decenoic acid (9HDA), methyl p-hydroxybenzoate (HOB) and 4-hydroxy-3-methoxyphenylethanol (also called homovanillyl alcohol or HVA), although four new substances (methyl (*Z*)-octadec-9-enoate (methyl oleate), (*E*)-3-(4-hydroxy-3-methoxyphenyl)-prop-2-en-1-ol (coniferyl alcohol), hexadecan-1-ol, and (*Z*9,*Z*12,*Z*15)-octadeca-9,12,15-trienoic acid) were recognized as important in eliciting retinue behavior from workers (Keeling et al., 2003). Although, there is more information concerning laying worker systems and social conflict in honey bees than in all

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other social insects (Ratnieks et al., 2006), the homogeneity of the *Apis* genus in this respect complicates approaches to answering socio-evolutionary questions.

Higher diversity in worker oviposition behavioral patterns and in physiological changes of workers can be found in stingless bees (e.g. Meliponini). In this group, some species have workers with developed ovaries even in queenright colonies (Sakagami et al., 1963). In other species, the workers show developed ovaries only in queenless colonies or in some special cases, such as overpopulation (Sakagami et al., 1973; Sakagami and Zucchi, 1974; Imperatriz-Fonseca and Kleinert, 1998). Also, there are some species in which the workers have permanently non-functional ovaries (Sakagami et al., 1963; Boleli et al., 2000).

Workers of the stingless bees *Leurotrigona muelleri* and *Friesella schrottkyi*, have non-functional ovaries in queenright colonies which become functional in queenless colonies (Sakagami et al., 1973; Sakagami and Zucchi, 1974), like honey bees. These species are considered intermediate between species in which workers have developed ovaries and those in which the workers do not show developed ovaries, even in queenless colonies (Zucchi, 1993).

Recent reports indicate that hydrocarbons that cover insects' cuticle are an important source of signals used in different levels of insects' communication (Howard and Blomquist, 2005). Studies in social insects showed that these substances can vary between individuals of different ages, gender, castes, and nest origin (Howard and Blomquist, 2005; Nunes et al., 2009a, b). Moreover, studies of different social insects have reported correlations of cuticle hydrocarbon variations with reproductive status (Bumble bee: Ayasse et al., 1995; Wasp: Sledge et al., 2001; Ant: Liebig et al., 2000; Cuvillier-Hot et al., 2001).

Some studies can also be found showing cuticle substances as prime fertility signals. In queenless colonies of the ant *Dinoponera quadriceps*, just one worker per colony is fertile. Studies showed that this dominant worker has significantly higher amount of 9-hentriacontene. In addition, Peeters et al. (1999) show that after the removal of this worker, a second worker mates and starts to reproduce and, correspondly, its level of 9-hentriacontene increases, reaching the amount found in dominant workers. Recently, Endler et al. (2004) showed that in colonies of *Camponotus floridanus* ants, the queen marks her eggs with cuticular hydrocarbons that act as indicator of queen presence, resulting in absence of reproduction by workers. Yet, D'Ettorre et al. (2004) showed that in the ant *Pachycondyla inversa*, the concentration of 3,11-dimethylheptacosane is direct correlated with ovary development and egg production. Finally, electroantennogram tests have shown that *P. inversa* ants can detect this substance and even variations in its concentration.

The aim of the present work was to analyze chemical secretions that could play a role in ovary development of workers in the stingless bee *Friesella schrottkyi*. We compared cephalic products and cuticular lipids of workers (laying and non laying) and queens, and found them to be significantly different.

2. MATERIALS AND METHODS

2.1. Study species

Friesella schrottkyi is a small stingless bee (3.0 mm) which produces colonies of about 300 workers, occurring in southeast and south areas in Brazil. This species nests in pre-existing cavities in wooden logs with a cryptic nest entrance that is closed during the night. The colonies present irregular brood shape and are not covered by involucres. Imprisonment chambers are usually found containing virgin queens (Norgueira-Neto, 1970; Roubik, 2006; Camargo and Pedro, 2007).

2.2. Colonies and individuals

For the tests, ten colonies of *Friesella schrottkyi* were used. The nests were kept in the University of São Paulo, in Ribeirão Preto, southeastern Brazil. The colonies were housed in wooden boxes ($15 \times 20 \times 10$ cm) which were covered by glass, and kept inside the laboratory and connected to the exterior by plastic tubes that allow foragers to freely exit and enter.

We collected 15 non-laying workers from four different queen-right colonies. Only old workers

were selected, avoiding newly-emerged individuals. The old workers were identified by their dark scutellum color and were collected directly from inside the nest. Also, seven physogastric queens were collected from inside the nests. Each queen was collected from a different colony. After the removal of the queens the colonies were kept orphaned, and all virgin queens that eventually appeared in these colonies were removed. After about two weeks of orphanage, some workers started laying eggs. Then, 15 laying workers were also collected. These workers were collected at the exact moment of their oviposition. All the collected bees were individually kept in small glass vials and killed by freezing in a conventional domestic freezer (temp. ca. –20 °C).

2.3. Chemical analysis

The chemical analysis was conducted at Keele University. For the analysis, comparisons were made between whole body cuticular hydrocarbon and ester profiles, head extracts of queens and workers, and laying and non-laying workers.

The samples were analyzed in a Hewlett-Packard 6890 gas chromatograph (equipped with a HP-5MS column; length, 30 m; ID, 0.25 mm; film thickness, 0.25 mm) directly coupled to a 5973 Mass Selective Detector (quadrupole mass spectrometer with 70 eV electron impact ionization). All the samples were injected in the splitless mode and the oven was programmed from 60 to 320 °C at 6 °C min^{-1}, then held at 320 °C for 10 min. Helium was used as carrier gas at a constant flow rate of 1.0 mL min^{-1}. The injector inlet temperature was held at 250 °C. The characterization of the compounds was conducted by the use of standard MS databases (NIST 2003), diagnostic ions, and the use of synthetic alkanes and long-chain esters. Pure oleic acid and methyl oleate were available for comparison. Triacontanyl acetate from the wax of *Melipona bicolor* (Koedam et al., 2002) was heated at 50 °C in hexane with a trace of p-toluene sulphonic acid and isobutyric acid to give triacontanyl isobutyrate. Similarly, triacontanyl butyrate was prepared for identification of high mass esters in worker cuticle. Octadecyl butyrate, hexanoate and decanoate were similary prepared with pure octadecanol (Sigma-Aldrich, Gillingham, UK). Identification of nonanal, geranial and neral (the last two known also as *trans* and *cis* citral) were confirmed by injection of the pure compounds and comparison of retention times and mass spectra. Substances with relative concentrations below 0.05% were treated as traces in the tables.

2.4. Cuticle hydrocarbon extracts

The heads of individual bees were separated from their bodies and heads and bodies were analyzed separately (see below). Compounds present in body cuticle were extracted by hexane for 15 min. Due to the very small size of these bees, five workers were analyzed in each extract. Three extracts were analyzed for each group of workers and each queen was analyzed separately (n = 7). Cuticle extracts were dried and then re-suspended in 20 μL of hexane. The extracts (2 μL) were injected in a Agilent 6890 Gas Chromatograph coupled to a Mass Selective Detector. Identification of the double bond positions of mono-alkenes was made through derivatization of a hexane extract of cuticle from five workers with dimethyl disulphide (DMDS) (Carlson et al., 1989). The extract was dried and re-suspended in 200 μL of hexane. Then, 50 μL of iodine solution (5%) and 100 μL DMDS were added. The vial was then purged with nitrogen, closed and maintained at 55 °C for 24 h. Thereafter, sodium thiosulfate solution was added to the mixture, and the organic phase was separated, dried, and analyzed by GC-MS. The derivatization with DMDS was effective to elucidate the double bond position for alkenes but not for alkadienes.

2.5. Head extracts

Workers heads were crushed in a small quantity of hexane. Crushed heads were kept in hexane for 15 min. As with the analysis of body cuticle, five heads were used in each sample. The hexane extracts were analyzed by GC-MS. Heads of queens were also analyzed by a different method from workers. Each queen head was analyzed separately by the solid injection method. One head was placed in small soft glass capillary tube sealed at one end. Then, the other end of the tube was sealed in a small flame. The capillaries were analyzed in the GC-MS system using the Keele solid sampler method (Morgan, 1990).

2.6. Hive wax

A sample of wax was collected from the pillars that sustain the brood inside the nest hive. A small

piece of wax was dissolved in 1 mL of hexane and filtered. A sample of the hexane solution (1 μL) was then injected in the GC-MS using the same gas chromatographic temperature programme used for analyses of cuticle substances.

3. RESULTS

The analysis revealed clear differences between workers and queens and between non laying workers and laying workers. The compounds found on the bodies of workers (laying and non-laying) and queens are listed in Table I. A total of fifty one substances were found in sufficient amount in the cuticle to quantify in the three analyzed groups. Comparatively more material was available on the queens, so a greater number of substances could be quantified there. The main compounds present are hydrocarbons, comprising of alkanes, alkenes, alkadienes, methyl and dimethyl alkanes from C_{21} to C_{35}. Small amounts of long chain esters was also identified. The worker cuticle contained a much simpler hydrocarbon profile when compared to queens (Fig. 1). Workers cuticle is mainly composed of linear alkanes, such as pentacosane, heptacosane, and nonacosane, and large amounts of very long chain alkanes and their corresponding alkenes and alkadienes, such as hentriacontane, 7-hentriacontene, 7-tritriacontene, tritriacontadiene and pentatriacontadiene. The most abundant compound on the workers' cuticle was nonacosane, followed by 7-hentriacontene, 7-tritriacontene and tritriacontadiene (Tab. I). Worker cuticle contained very-long-chain esters which were not found in queens. The main differences between laying and non-laying workers cuticle composition are in the relative concentration of the long chain alkenes and alkadienes. Non-laying workers showed higher concentrations of very long chain alkenes and alkadienes when compared to laying workers. Even though queen cuticle also contained hydrocarbons from C_{21} to C_{35}, their cuticle was rich in methyl- and dimethyl-branched hydrocarbons, which are completely absent from workers. The most abundant compound on the queen cuticle is pentacosane, followed by 11-methylnonacosane and 11-methylheptacosane. No volatile compounds were identified in the heads of queens.

A comparison was also made between volatile and cuticular substances between the heads and bodies of non-laying workers (Tab. II). This was done to identify any volatile candidate pheromones in the heads of workers, and to see if the high molecular weight esters were also present in the head cuticle. Three volatile aldehydes, nonanal, geranial and neral (the last two known collectively as citral) were present in nanogram quantities on the heads. Although the hydrocarbon pattern showed similarities between body and head, there were some notable differences. 7-Tricosene and 7-hentriacontene were significantly more abundant on the heads of workers, while nonacosane and pentatriacontadiene were more abundant on the body. Trace amounts of four high-mass butyrates were identified in the cuticle of laying and non-laying workers. These were identified by comparison of mass spectrum and retention times of authentic triacontanyl butyrate, prepared by transesterification from triacontanyl acetate. While three octadecyl esters were present on the heads and absent from the bodies, the C_{28} to C_{36} esters, present on the bodies, were absent from the heads. There were no differences between laying and non-laying workers' head profiles. Queens' heads presented only the same hydrocarbons found in its body cuticle. Their heads did not contain either the esters nor the volatile compounds identified in workers' heads.

Samples of wax from the hive were analyzed to see if the high molecular weight esters might be derived from the wax glands of the bees. The wax analysis showed nonacosane as the main compound. Small amounts of the long chain esters identified in the workers' extract were also identified in the wax samples.

4. DISCUSSION

A queen pheromone that regulates the ovary regulation of workers in stingless bees is still virtually unknown. In honey bees, the stingless bees' sister group, queen dominance is

Table I. Comparison between cuticular lipids from the bodies of workers (laying and non laying) and queens of the stingless bee *Friesella schrottkyi*. The four most abundant compounds in each column are printed bold.

			% of the total					
			Non laying Workers (n=3)		Laying Workers (n=3)		Queen (n=7)	
Peak	Ret. Time	Compounds	Mean	SD	Mean	SD	Mean	SD
1	23.06	Heneicosene	*t*		0.07	0.07	-	
2	23.45	Heneicosane	-		-		2.57	1.13
3	23.98	11-Methylheneicosane	-		-		0.26	0.13
4	24.17	γ-eicosanolactone *	-		-		0.11	0.10
5	24.55	Oleic acid	-		-		0.11	0.19
6	26.10	7-Tricosene	0.15	0.01	0.16	0.02	-	
7	26.38	Tricosane	0.06	0.03	0.06	0.01	1.42	0.61
8	26.88	11-Methyltricosane	-		-		0.26	0.16
9	27.37	5-Methyltricosane	-		-		0.22	0.10
10	27.81	Tetracosane	-		-		0.74	0.21
11	28.82	7-Pentacosene	0.15	0.11	0.07	0.03	-	
12	29.10	Pentacosane	0.33	0.10	0.33	0.03	**43.78**	**13.02**
13	29.63	11-Methylpentacosane	-		-		2.07	0.67
14	29.88	5-Methylpentacosane	-		-		1.12	0.39
15	30.16	9, 15-Dimethylpentacosane	-		-		0.64	0.29
16	30.42	Hexacosane	-		-		1.06	0.27
17	30.80	10, 15-Dimethylpentacosane	-		-		0.38	0.17
18	31.28	9-Heptacosene	0.12	0.03	0.07	0.03	0.21	0.09
19	31.60	Heptacosane	0.34	0.18	1.13	0.23	**5.26**	**2.87**
20	32.08	11-Methylheptacosane	-		-		**8.38**	**2.97**
21	32.37	11, 15-Dimethylheptacosane	-		-		1.23	0.57
22	32.61	5, 17-Dimethylheptacosane	-		-		2.50	1.05
23	32.78	*Not identified*	0.23	0.00	0.26	0.23	-	
24	32.89	??-Dimethylheptacosane	-		-		1.19	0.62
25	33.22	11-Methyloctacosane	-		-		1.56	0.62
26	33.74	Nonacosene	0.39	0.53	0.10	0.04	0.23	0.12
27	34.00	Nonacosane	**14.53**	**22.06**	**68.82**	**15.05**	2.91	2.72
28	34.40	11 and 13-Methylnonacosane	-		-		**16.66**	**6.45**
29	34.65	13,15-Dimethylnonacosane	-		-		1.09	0.87
30	34.93	5, 19-Dimethylnonacosane	-		-		1.08	0.48
31	35.14	??-Dimethylnonacosane	-		-		0.33	0.40
32	35.43	11, 17-Dimethylnonacosane	-		-		0.28	0.12
33	35.86	Hentriacontadiene	0.33	0.14	-		0.41	0.22
34	36.08	7-Hentriacontene	6.91	1.85	1.98	1.38	0.22	0.43
35	36.15	Hentriacontane	4.02	2.64	**9.28**	**3.29**	-	
36	36.49	11 and 13 and 15-Methylhentriacontane	-		-		0.50	0.21
37	36.95	5, 19-Dimethylhentriacontane	-		-		0.28	0.12
38	37.66	Octacosanyl acetate	*t*		*t*		-	
39	38.00	Tritriacontadiene	**36.79**	**14.51**	**4.08**	**1.51**	0.30	0.33
40	38.19	7-Tritriacontene	**23.56**	**4.27**	**6.20**	**6.00**	0.48	0.58
41	38.48	??-Methyltritriacontane	-		-		0.09	0.13
42	38.95	??-Methyltritriacontane	-		-		*t*	
43	39.40	Octacosanyl butyrate	*t*		*t*		-	
44	39.64	Triacontanyl acetate	*t*		0.31	0.08	-	
45	39.93	Pentatritriacontadiene	**11.50**	**5.06**	1.18	1.24	*t*	
46	41.31	Triacontanyl butyrate	*t*		*t*		-	
47	41.54	Dotriacontanyl acetate	0.15	0.26	1.46	0.53	-	
48	43.14	Dotriacontanyl butyrate	*t*		0.30	0.23	-	
49	43.32	Tetratriacontanyl acetate	0.17	0.29	3.02	1.16	-	
50	45.10	Tetratriacontanyl butyrate	*t*		0.41	0.19	-	
51	45.33	Hexatriacontanyl acetate	*t*		0.62	0.18	-	

* *unconfirmed identification*.

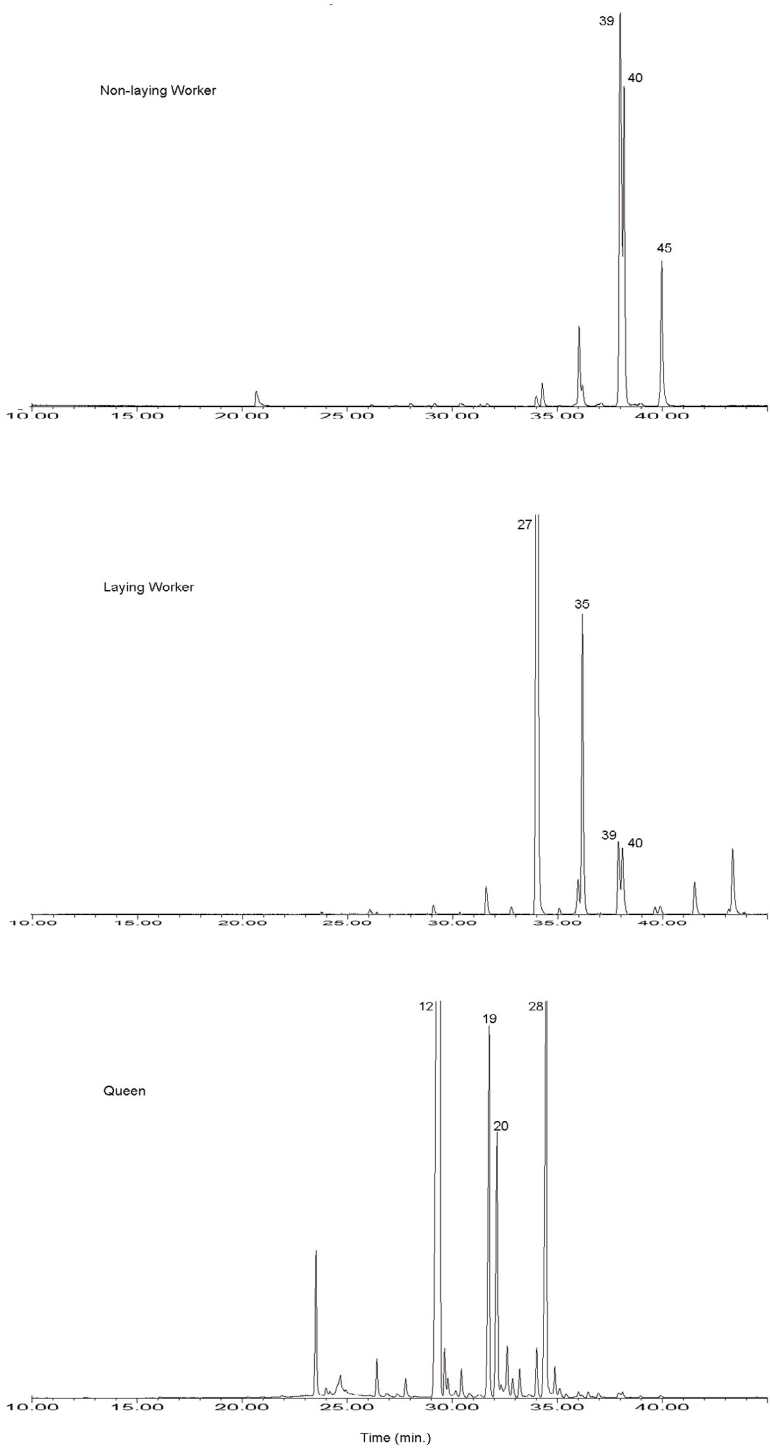

Figure 1. Ion chromatogram of worker and queen body cuticle. Numbered peaks correspond to the peaks in Table I.

Table II. Comparison between body lipids and head extracts of non-laying workers of the stingless bees *Friesella schrottkyi*. The four most abundant compounds in bodies and heads are printed bold.

			% of the total (n=3)			
			CH		Head	
Peak	Ret. Time	Compounds	Mean	SD	Mean	SD
1	5.84	Nonanal	-		0.30 ± 0.19	
2	9.11	Geranial	-		0.23 ± 0.08	
3	9.71	Neral	-		0.35 ± 0.06	
4	21.05	*Not identified*	-		0.11 ± 0.11	
5	23.06	Heneicosene	*t*		0.95 ± 0.24	
6	23.45	Heneicosane	-		0.09 ± 0.02	
7	24.71	Methyl oleate	-		0.16 ± 0.05	
8	26.10	7-Tricosene	0.15 ± 0.01		**10.57 ± 4.51**	
9	26.38	Tricosane	0.06 ± 0.03		0.24 ± 0.07	
10	27.07	*Not identified*	-		0.63 ± 0.11	
11	27.92	Octadecyl butyrate	-		0.38 ± 0.45	
12	28.82	7-Pentacosene	0.15 ± 0.11		1.30 ± 0.09	
13	29.10	Pentacosane	0.33 ± 0.09		0.69 ± 0.23	
14	30.45	Octadecyl hexanoate	-		0.69 ± 0.66	
15	31.28	9-Heptacosene	0.12 ± 0.03		0.25 ± 0.17	
16	31.60	Heptacosane	0.34 ± 0.18		0.39 ± 0.24	
17	32.78	*Not identified*	0.23 ± 0.00		0.67 ± 0.80	
18	33.74	Nonacosene	0.39 ± 0.53		0.29 ± 0.08	
19	34.00	Nonacosane	**14.60**	**±22.19**	4.78 ± 2.25	
20	35.05	Octadecyl decanoate	-		0.29 ± 0.26	
21	35.86	Hentriacontadiene	0.33 ± 0.14		1.40 ± 0.08	
22	36.08	7-Hentriacontene	**6.92**	**± 1.84**	**26.77**	**± 2.99**
23	36.15	Hentriacontane	4.04 ± 2.66		2.38 ± 0.55	
24	37.66	Octacosanyl acetate	*t*		-	
25	38.00	Tritriacontadiene	**36.82**	**± 14.45**	**30.88**	**± 2.34**
26	38.19	7-Tritriacontene	**23.59**	**± 4.21**	**13.97**	**± 4.07**
27	39.40	Octacosanyl butyrate	*t*		-	
28	39.64	Triacontanyl acetate	*t*		-	
29	39.93	Pentatriacontadiene	11.51 ± 5.04		1.21 ± 1.06	
30	41.31	Triacontanyl butyrate	*t*		-	
31	41.54	Dotriacontanyl acetate	0.15 ± 0.26		-	
32	43.14	Dotriacontanyl butyrate	*t*		-	
33	43.32	Tetratriacontanyl acetate	0.17 ± 0.29		-	
34	45.10	Tetratriacontanyl butyrate	*t*		-	
35	45.33	Hexatriacontanyl acetate	*t*		-	

maintained by mandibular secretions (Hoover et al., 2003). Our results showed the absence of compounds from the heads of queens that might correspond to the honey bee queen retinue pheromone. Hence, there is no evidence to indicate that queen dominance in this stingless bee occurs in the same way as in honey bees.

The results showed clear differences between workers and queens cuticle hydrocarbons (Fig. 1). The queens' cuticle contain high amounts of branched hydrocarbons which are completely absent in workers. The use of hydrocarbons as queen signals has already been described for others social insects (Endler et al., 2004). Likewise, the high levels

of branched hydrocarbons described for the queens of *F. schrottkyi* might be a signal of its presence and fertility for workers. Further bioassays with these substances is needed to examine their effect on workers sterility.

The results also showed differences between laying and non-laying workers in their cuticle profile. The variations found between the two groups of workers in the percentage of the compounds could reflect the development of workers ovaries. Modifications in cuticular pattern of workers after ovary activation were also described for ants and social wasps (Sledge et al., 2001; Liebig et al., 2000; Cuvillier-Hot et al., 2001; Howard and Blomquist, 2005). Another possibility for this variation might be due to differences in tasks or age of the collected workers. Although only workers with dark scutellum were choose for the tests, the collected workers could belong to a specific task group or could be of different age. Nunes et al. (2009a, b) described qualitative and quantitative differences for the cuticle compounds of young and old workers of the stingless bees *Schwarziana quadripunctata* and *Frieseomelitta varia*. Differences in cuticular pattern of individuals having different tasks were also described for some ant species (reviewed by Howard and Blomquist, 2005; Martin and Drijfhout, 2009).

The workers heads presented small amount of the volatile aldehydes, nonanal, neral and geranial. Neral and geranial (or *cis* and *trans* citral) were described as a trail pheromone in the stingless bee *Geotrigona mombuca* although recent studies disproved this possible function (Stangler et al., 2009). Citral is also the major volatile in heads of the robber bee *Lestrimelitta limao* (Wittmann et al., 1990), and citral and geraniol are part of the Nasanov pheromone complex of honey bees (Free et al., 1984). Behavioral studies showed that workers of different species of stingless bees are able to recognize this substance and react aggressively (Wittmann et al., 1990; Sakagami et al., 1993). Behavioral tests with very small amount of this material should elucidate the main function of these compounds in colonies of *Friesella schrottkyi*.

Small amounts of the volatile aldehyde nonanal were found in head extracts of workers. This compound was also identified as one of the main compounds in mandibular gland extracts of the stingless bee *Trigona spinipes*. Although the single synthetic aldehyde was not tested by the authors, the mandibular gland extracts incited aggressive behavior by workers (Schorkopf et al., 2009). Nonanal was also described as major volatile compound for the heads of *Trigona hyalinata* and *Trigona truculenta* and as a minor compound in the heads of *Nannotrigona testaceicornis* (W. Francke, unpubl. data).

Recently, the recruitment behavior in stingless bees has been the target of many detailed studies. For some species of these bees, trail pheromones were chemically identified and behaviorally testified. The known pheromones are secreted by the cephalic labial glands and are mainly esters, such as, hexyl decanoate (*Trigona recursa*: Jarau et al., 2006) and octyl octanoate (*Trigona spinipes*: Shorkopf et al., 2007). Our data showed that workers of *Friesella schrottkyi* produce some esters specifically in their heads. These compounds are completely absent in workers bodies and in queens extracts. The fact that these compounds are absent in queen extracts indicates that these substances should be related to workers activities. The chemical characteristics of the esters present in worker heads meets with the requirements of trail pheromones, since they are sufficiently volatile to be detected by odour but would not evaporate quickly. Further experiments are planned to investigate if these compounds could act as trail pheromones in this stingless bee species.

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Lipides de la cuticule spécifiques d'une caste chez l'abeille sans aiguillon *Friesella schrottkyi*.

phéromone / contrôle par la reine / ouvrière pondeuse / différence entre castes / hydrocarbones

Zusammenfassung – Kastenspezifische Lipide der Kutikula bei der stachellosen Biene *Friesella schrottkyi*. Bei Honigbienen (*Apis mellifera)* kontrollieren die Königinnen die Fertilität ihrer Arbeiterinnen, indem sie mit einer Substanzmischung aus den Kopfdrüsen die Entwicklung der Arbeiterinneneierstöcke unterdrücken. Bei den Völkern von stachellosen Bienen ist es hingegen nicht bekannt, wie die Königinnen die reproduktiven Neigungen ihrer Arbeiterinnen kontrollieren. Im Rahmen unserer Untersuchungen über ein mögliches Dominanzpheromon der Königin bei stachellosen Bienen haben wir auch die kutikulären Substanzen und Drüsensekrete von Arbeiterinnen und Königinnen bei *Friesella schrottkyi* untersucht. Bei dieser stachellosen Bienenart produzieren die Arbeiterinnen keine Eier, solange die Königin anwesend ist; bei Abwesenheit der Königin entwickeln sie aber ihre Ovarien und legen männlich determinierte Eier. Vergleichende Analysen der kutikulären Substanzen von Königinnen, legenden Arbeiterinnen aus weisellosen Völkern und nicht legenden Arbeiterinnen aus einem weiselrichtigen Volk ergaben eindeutige Unterschiede zwischen diesen Gruppen. Bei allen Bienen wurden unverzweigte kutikuläre Alkane gefunden. Königinnen hatten zusätzlich Methyl-verzweigte Alkane, die weder bei den legenden noch bei den nicht legenden Arbeiterinnen nachgewiesen wurden. Beide Arbeiterinnen-Gruppen hatten aber unverzweigte Alkene und Alkadiene, die nicht bei den Königinnen gefunden wurden. Zusätzlich fanden sich bei Arbeiterinnen kleine Mengen an langkettigen Estern, die nicht bei Königinnen vorkamen. Nicht legende Arbeiterinnen wiesen die Kohlenwasserstoffe mit den größten und Königinnen die mit den kleinsten Kettenlängen auf, während die Kettenlängen der legenden Arbeiterinnen dazwischen lagen. Das Kohlenwasserstoffmuster des Königinnenkopfes unterschied sich nicht von dem des Körpers. Bei Arbeiterinnen zeigten sich dagegen Unterschiede im Muster zwischen Kopf und Körper. So wurden kleine Mengen an volatilem Nonanal, Geranial und Neral sowie Octadecyl-Butyrat, Octadecyl-Hexanoat und Octadecyl–Decanoat ausschließlich in den Köpfen nachgewiesen. Es sind nun weitere Arbeiten geplant, um die Effekte von Einzelsubstanzen bzw. Substanzklassen auf das Verhalten von Arbeiterinnen zu überprüfen.

Pheromon / Königinnenkontrolle / legende Arbeiterinnen / Kastenunterschiede / Kohlenwasserstoffe

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