Volatile chemicals in glands of the carpenter ant, *Camponotus arminius*

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Camponotus arminius is a large black carpenter ant that occurs in tropical and sub-tropical Africa and has extensive foraging trails both in trees and on the ground. Analysis of excised mandibular glands has confirmed the presence of pentane-2,4-dione, *n*-hexanal, 4-methyl-3-heptanone, and mellein. Pentane-2,4-dione has not been reported from insects. The postpharyngeal gland contains *n*-heneicosane, *n*-tricosene, *n*-tricosane, *n*-pentacosene, *n*-pentacosane, and a significant amount of 11-methyl-*n*-tricosane and 11-methyl-*n*-pentacosane. Dufours glands contain mainly *n*-undecane, *n*-tridecane, and two terpenoid compounds that are unknown in the genus *Camponotus*. These latter two compounds are geranyllinalool and probably geranylfarnesol.

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Introduction

The subfamily Formicinae (formicine ants) has a large number of genera with the genus *Camponotus* being one of the largest. Many species of this genus are large, dark coloured or black ants that live in galleries in decaying wood, and are therefore commonly known as carpenter ants. Worker ants have no sting and spray a solution of formic acid and Dufours gland contents at attackers.

The species *Camponotus arminius* occurs in tropical and sub-tropical Africa, where it is found as far south as the south coast of Natal, South Africa. It makes large nests in galleries in trees with major workers often protecting the entrance holes. Workers can be found following long trails both in the trees and on the ground to sources of sugary plant exudates. This species is rather unique amongst formicines in that it has naked pupae, whereas, in general, the pupae of formicines are enclosed in cocoons.

As ants are social insects, they use volatile chemicals in a variety of ways for the communication necessary for social existence. Billen (1994) has counted 39 exocrine glands (i.e. glands secreting externally) in ants that are potential sources of pheromones. We have analysed the compounds present in the mandibular gland, the postpharyngeal gland, and the Dufours gland of *C. arminius*. Certain of the compounds have been identified in many other ant species. However, a compound (pentane-2,4-dione) previously unreported in insects, has been identified in the mandibular gland and the Dufours gland contains two terpenoid compounds (geranyllinalool and probably geranylfarnesol) that have not been reported from this genus.

Materials and methods

Worker ants were collected from a nest in an old *Syzygium cordatum* (umdoni) tree in the Margate district of Natal, South Africa. Glands were dissected under a microscope, placed in a cup made at the end of a 100 µl glass pipette, and inserted directly into the modified inlet of a Hewlett Packard 5890 GC as described by Burger, Munro, Smit, Schmidt, Wu & Tien (1990) and modified by Brand & Mpuru (1993).

An HP Ultra 1 column (50 m x 0.2 mm i.d. and 0.33 mm film thickness) was temperature programmed from 50°C to 260°C at 5°C min^{-1}. Mass spectra were recorded by a HP 5970 MSD at 70 eV ionisation potential.

Chemical standards used were: pentane-2,4-dione (Aldrich Chemical Co., Milwaukee, WI), *n*-hexanal (Aldrich Chemical Co., Milwaukee, WI), C-10 to C-21 *n*-alkanes (Alltech Associates, Deerfield, IL) and geranyllinalool (Akros, Loughborough, UK).

Results

The direct insertion into the GC inlet of excised mandibular glands gave chromatograms that always exhibited three peaks corresponding to pentane-2,4-dione, *n*-hexanal and mellein (Table 1). Peak 4, while often present in chromatograms, remains unidentified. The compounds identified from the chromatographic separation of the volatile components in the

Table 1 Identified compounds in an excised mandibular gland of *C. arminius*

Peak no	Ret. time[a]	Area %[b]	Identity	Evidence[c]
1	8.1	21.5	pentane-2,4-dione	GC, MS
2	8.2	9.8	*n*-hexanal	GC, MS
3	12.4	trace	4-methyl-3-heptanone	GC, MS
4	17.4	11.8	unknown	MS
5	24.6	trace	substituted pyrazine ?	MS
6	30.7	37.6	mellein	MS

a. Minutes

b. Typical values for one gland. Trace means present in some chromatograms

c. GC means that an authentic standard was available for comparison, otherwise identification relied on interpretation of the mass spectrum.

postpharyngeal gland and the Dufours gland are listed in Tables 2 and 3. Certain compounds were available as authentic standards for confirmation of identity, while others in the chromatograms were identified only from their mass spectra. As single glands were analysed, the reported area percent is typical of that observed for a single gland, but varies from one gland to another. Therefore, these values provide an indication of those compounds that are major components and those that are minor or trace components in the three glands.

Discussion

Old *Syzygium cordatum* trees seem to be a favoured tree for *C. arminius* nests in the area where they were collected. The trees secrete sugary secretions and old trees often have decayed regions where gallery construction is easy. The nests can be very extensive in the trunk and larger branches and an old established nest will contain many thousands of workers.

The mandibular gland of *C. arminius* contains an interesting variety of compounds (Table 1). Pentane-2,4-dione has been observed in every chromatogram obtained, both of whole heads and of excised mandibular gland tissue. The retention time and mass spectrum of an authentic standard has confirmed this structure and it is reported from an insect source for the first time. *n*-Hexanal, also confirmed by comparison with an authentic standard, has been found in the mandibular gland of another formicine ant, *Oecophylla longinoda*, the African weaver ant (Bradshaw, Baker & Howse 1975). Only trace amounts of 4-methyl-3-heptanone were detected. While this ketone occurs in a number of ant species, it is more commonly found in myrmicine and doryline secretions (Oldham, Morgan, Goblin, Schoeters & Billen 1994) than in formicine secretions. Certain chromatograms of mandibular glands showed peak 4, which remains unidentified, and a very minor peak 5, the mass spectrum of which suggested a substituted pyrazine. Mellein occurs in the mandibular gland of males of *Camponotus pennsylvanicus*, *C. herculeanus* and *C. novaeboracensis* (Brand, Fales, Sokoloski, Mac Connell, Blum & Duffield 1973) and in the hindgut of a number of *Camponotus*, *Lasius* and *Formica* species where it acts as a trail pheromone for certain species (Bestmann, Kern, Schäfer & Witschel 1992; Übler, Kern, Bestmann, Hölldobler & Attygalle 1995; Bestmann, Übler & Hölldobler 1997).

The postpharyngeal gland of *C. arminius* workers contains hydrocarbons in the C-21 to C-26 range with two 11-methyl branched alkanes being major components (Table 2). The only authentic standard available for GC and MS identification was *n*-heneicosane. Structural identifications of the other hydrocarbons in this gland are based on the interpretation of their mass spectra. This gland is reported to have aliphatic hydrocarbons similar to those found on the cuticle (Bagneres & Morgan 1991) but we have been unable to obtain chromatograms showing any significant peaks from the direct insertion technique of the cuticle from one complete abdomen. One must therefore conclude that the cuticle contains very little in the way of hydrocarbons of any kind.

The Dufours gland of ants is one of the best studied insect glands and the identified alkanes, especially C-11 and C-13 (Table 3), are commonly found in this gland of many formicines. While many ants have appreciable amounts of alkenes, often corresponding in chain length to the alkanes present, this species only has barely detectable trace amounts of two tridecenes. However, the presence of geranyllinalool, confirmed by comparison with an authentic standard, is unique among formicines, as is the likely presence of geranylfarnesol. In chromatograms of some glands these two compounds accounted for more than 20% of the total area. Other structurally related terpenoids have been reported in the Dufours gland of *Formica* species (Bergstrom & Lofqvist 1973) and geranyllinalool has been identified as a minor component in the Dufours gland of New World army ants (Keegans, Billen, Morgan & Gokcen 1993).

The identification of the major components of the three glands of *C. arminius* offers an opportunity to study the behavioural roles of their secretions. As worker ants follow long trails both in trees and along the ground, and also exhibit some alarm behaviour when provoked, these two behavioural characteristics are probably the most likely to yield results from bioassays of glandular contents. Complex behaviour

Table 2 Identified compounds in an excised postpharyngeal gland of *C. arminius*

Peak no	Ret. time[a]	Area %[b]	Identity	Evidence[c]
1	43.7	7	*n*-heneicosane	GC, MS
2	47.8	8	*n*-tricosene	MS
3	48.5	16	*n*-tricosane	MS
4	49.5	18	11-methyl-*n*-tricosane	MS
5	4.2	13	*n*-pentacosene	MS
6	55.2	trace	*n*-pentacosane	MS
7	56.7	30	11-methyl-*n*-pentacosane	MS

a. Minutes

b. Typical values for one gland. Trace means present in some chromatograms.

c. GC means that an authentic standard was available for comparison, otherwise identification relied on interpretation of the mass spectrum.

Table 3 Identified compounds in an excised Dufours gland of *C. arminius*

Peak no.	Ret. time[a]	Area %[b]	Identity	Evidence[c]
1	15.1	trace	*n*-decane	GC, MS
2	18.7	67	*n*-undecane	GC, MS
3	21.6	1.3	*n*-dodecane	GC, MS
4	24.35	trace	*n*-tridecene	MS
5	24.45	trace	*n*-tridecene	MS
6	24.8	19	*n*-tridecane	GC, MS
7	30.2	1.5	*n*-pentadecane	GC, MS
8	35.2	trace	*n*-heptadecane	GC, MS
9	42.2	7	geranyllinalool	GC, MS
10	54.6	3	geranylfarnesol?	MS

a. Minutes

b. Typical values for one gland. Trace means present in some chromatograms.

c. GC means that an authentic standard was available for comparison, otherwise identification relied on interpretation of the mass spectrum.

often relies on complex mixtures, but as some of the identified compounds will not be obtained readily, it will be difficult to test mixtures made up of pure compounds.

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