

Role of Dufour's gland and mandibular gland secretion in Ant Colony organisation and defense mechanisms of *Camponotus compressus* and *Oecophylla smaragdina*

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ABSTRACT

Ants are eusocial insects and observed in 3 castes secreting a wide variety of pheromones for surviving in the colony. They can be sex pheromone, trail pheromone and alarm pheromone. These pheromones help in the recruitment of workers, mating, foraging food. These



pheromones contain a single compound from a single gland or from two glands. This study analyses the knowledge of these pheromones and their chemical structure. The mandibular gland and Dufour's gland of *Oecophylla smaragdina* and *Camponotus compressus* were extensively studied to provide a valuable resource in chemical ecology research. Limited research has been done on pheromones released by *Oecophylla smaragdina* and *Camponotus compressus*. The Dufour's gland is one of the most well-developed glands, playing vital roles in defense, foraging, information exchange, and reproduction. The chemical components were analyzed using gas chromatography-mass spectrometry. The secretions from the Dufour's gland and mandibular gland contained high concentration of n-undecane, which serves as an alarm pheromone, and compositions varied among different castes. This highlights a research gap and the need to investigate the differences in the chemical composition between these two ant species, we analyse the diverse chemicals released from the Dufour's gland and the mandibular gland.

Keywords: Ants, Mandibular gland, Dufour's gland, Pheromones, Defense mechanisms

INTRODUCTION

Ant colonies usually use multiple ways for communication signals to perform individual tasks such as looking for food, territorial defense and cooperative offspring care by trail pheromones.¹ The substances present inside the mandibular and Dufour's gland usually contain small molecules and volatile in nature which needs to be studied by Gas Chromatography. A few forager ants venture out from the nest in search of food. Upon finding a food source, they release trail pheromones, enabling their nestmates to follow the path to the same resource.² Ants in the Formicidae family typically release various chemical substances

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known as semiochemicals to communicate.³ Pheromones are semiochemicals that facilitate communication within the same species, while semiochemicals that enable communication between different species are known as allelochemicals.⁴ Pheromones are semiochemicals that facilitate communication within the same species, while semiochemicals that enable communication between different species are known as allelochemicals.

An allomone is typically released by insects to trigger a defensive response in potential predators, while a kairomone is generally secreted to help locate prey.⁵ In ant's trail pheromones, it has been observed that synomone is having a mutual beneficiary aspect to both the species. The mandibular gland is an exocrine gland present in Hymenoptera.⁶ It was first observed by Meinhert (1860) and then by Bordas (1895) that to be ubiquitous in hymenoptera. The gland secretion plays an important role in reproduction as well. Communication with the help of chemicals helps in functioning of ant colony. Predation by ponerine ants usually takes place through recruitment behaviour which depends

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on pheromones secreted by foraging workers⁷ and the queens of Pharaoh ants secrete pheromones that allows them to recognise by workers.⁸ It has been observed that worker ants belonging to Myrmica when targets a prey, it usually moves around in a circuit around the prey and secrete from its mandibular gland.⁹ The secretion comes out through the base of the mandible which forms a film or droplet across the mandibles and these secretions contain volatile chemicals which alters the behaviour of other workers in the nearby area from the source. Alarm behaviour is known as the most common behaviour and it has a broad range of response. The substances that make up the alarm pheromone have five to ten carbon atoms and a molecular weight between 100 and 200 Da.¹⁰ When leaf cutting ants are alarmed they do show a defensive response ¹¹ and secretes an alarm pheromone from their mandibular gland.^{12,13} Numerous studies on the chemistry of this gland have shown that citral is the primary component in Atta sexdens rubropilosa (Butenandt et al., 1959). It was observed In A. texana workers, 4-methyl-3-heptanone and 2-heptanone are the main components, occurring in a 4:1 ratio¹³ but after further studies it was found that the mandibular gland secretion contains 56 compounds and 4-methyl-3-heptanone is the major releaser of alarm behaviour in Atta.¹³

The Dufour's gland contents are mostly for trail pheromone,¹⁴ the gland also produces sex pheromone¹⁵ and queen control pheromone.⁸ The contents also function for making slave workers in the process¹⁶ or can be used as an aggregation stimulus and pheromone for recruitment.¹⁷

DUFOUR'S GLAND AND MANDIBULAR GLAND

The Dufour's gland, an important gland linked to the sting apparatus in female castes, primarily functions as a storage site for chemical secretions. This gland plays a vital role in trail marking, signaling alarms for colony defense, regulating slave-making behavior, and supporting reproduction. It contains a diverse range of volatile and semi-volatile compounds, including saturated and unsaturated hydrocarbons, terpenoids, alcohols, acids, fatty acids, quinones, and aromatic substances.¹⁸ The chemical compounds found in the Dufour's gland are specific to each species and can serve as chemotaxonomic indicators to distinguish between different ant species.¹⁹ In the family of Formicidae, Considering the size and population of their colonies, Camponotus is one of the largest genera. Females of the genus Camponotus typically show a division of labor based on their body size and the tasks they perform, with workers categorized as major, medium, or minor.²⁰ Over 20 species of Camponotus, the Dufour's gland have been studied.²¹ However this study focuses on the comparison o the chemical composition in two different species of ants Camponotus compressus and Oecophylla smaragdina.

An exocrine gland found in the head is the mandibular gland.²² The mandibular glands are situated in the head and are present very close to the mandibles and intramandibular glands are present within the mandibles. The mandibular glands are the most studied ones.²³ They usually produce pheromones for the recognition of nestmates,²² alarm and antibiotic substances.²⁴

The mandibular glands of Formicidae contains some major volatile organic compounds that initiate ant behaviours¹ such as

alarm behaviours which shows accelerated movements and attack for defense.²⁵ Alarm pheromones may not work in ants that dwell in disorganized colonies; instead, they serve as a signal to leave the nest.¹¹

In ant species from the Myrmicinae subfamily, such as *Atta* and *Acromyrmex*, the mandibular gland releases secretions primarily composed of highly volatile mixtures with low molecular weight, often in the form of alcohols or ketones, which aid in communication.²⁶

MATERIALS AND METHODS

A mature nest of *Camponotus compressus* and *Oecophylla smaragdina* were selected which comprised of minor workers, major workers, gyne and queen in Bagepalli range in Verlakonda region of Chikkaballapur forest of Karnataka, India. Which is situated at 13.6433 \Box N Latitude and 77.7845 \Box E longitude. They were selected at random from each caste (minor workers, major workers, gynes, queens, and original workers) and utilized to analyze the chemical makeup of their mandibular and Dufour's glands.

GC-MS ANALYSIS

Individual ant species were first placed at a temperature of $-20\Box C$ for 10 minutes to decrease their activity rate and then they were dissected by observing under the stereo microscope (LABOMED, America with \Box 110-240 V, 50/60 Hz, 35 watts, Bottom lamp 12 V 10W, Top lamp 12 V, 20W, 250 V F 2.5A) and kept in a 0.65% NaCl₂ solution. A glass capillary measuring 2 mm in diameter, 0.2 mm in wall thickness, and 100 mm in length was used to store the samples. The glands were removed for two hours at room temperature after being placed in a vial with one milliliter of chloroform.²⁷

A clean needle was used to separate the gland tissues after two hours, and 300 ng of n-tetradecane was added for use as an internal reference. For every caste, three duplicates were made; the blanks only had 300 ng of n-tetradecane and 1 ml of chloroform. Both the sample and the blanks were submitted for GC-MS analysis.

Gas Chromatography (GC) was utilized to examine volatile compounds extracted from the Dufour's gland in the abdomen and the mandibular gland in the head region. A SHIMADZU GCMS-QP2010 SE was employed, offering a mass range of m/z 1.5–1000 AMU. For the 1 pg octafluoronaphthalene molecular ion at m/z 272, the EI scan achieved a sensitivity of S/N > 200/1 RMS, with a resolution of R=2M (FWHM). Scanning was performed at intervals (event time) of 0.5 seconds within an m/z range of 200–300, using an Rtx-5 ms column with dimensions of 30 m x 0.25 mm I.D. and a 0.25 µm film thickness. The system can scan up to 10,000 u/sec (single scan). The GC-2010's single split/splitless injection port, equipped with Advanced Flow Control (AFC-2010), supports pressures and flow rates up to 970 kPa.²⁸

RESULTS AND DISCUSSION

The research identified alkanes as the main constituents in the Dufour's glands of *Camponotus compressus* and *Oecophylla smaragdina*. In *Camponotus compressus*, undecane emerged as the major compound (shown in Table 2), while hexadecane was predominant in *Oecophylla smaragdina* (Table 4). Although

species like C. herculeanus, C. sericeus, and C. lateralis rebeccae primarily contain n-tridecane, n-undecane generally serves as an active alarm pheromone across most Camponotus species. For C. gastroi and C. balzani, the primary secretions include octyl hexanoate and 5-methyl-tridecane. The Dufour's gland secretion of Camponotus compressus consists mainly of alkanes, with 97% of workers containing n-undecene²⁹. One of the main ingredients that gives ants their warning pheromone is N-undecane. As shown in Table 1, it has been noted that *Formica hugubris* functions as a sex attractant. This indicates that n-undecane may serve as an alarm pheromone for worker ants and as a sex attractant for gynes. Only trace levels of alkanes were detected in the Dufour's gland of Oecophylla smaragdina workers. In Camponotus compressus and Oecophylla smaragdina, most alkanes in the male mandibular glands functioned as sex pheromones, with tridecene acting as a sex pheromone specifically in Oecophylla smaragdina. In Camponotus compressus, the primary compounds in the mandibular gland were trail pheromones, specifically 2-methylnonane and nonadecane, as shown in Table 3.

Structure and location of mandibular gland in *Camponotus* compressus

In all ant species, the mandibular glands have a consistent structure, positioned on each side of the head and attached to the base of the mandibles. Each gland consists of secretory units and a reservoir, which is oval in males and round in females. An ant's body is segmented into the head, thorax, and abdomen, with primary exocrine glands in the head including the mandibular, propharyngeal (maxillary), and post-pharyngeal glands. The mandibular glands are formed by clusters of glandular cells that release their secretions into a reservoir linked to the main duct ⁶. It is situated below the clypeus. Many hypotheses talk about the mechanism of its opening and release of the secretion and two of the hypotheses states that due to an increased hemolymph pressure the mandible opens and releases secretion from the mandibular gland ³⁰. According to another theory, the gland content may be released by a cuticular lamella that joins the gland duct to the mandalus.31

Structure and location of Dufour's gland in *Camponotus* compressus

In all ant species, the mandibular glands have a similar structure, situated on either side of the head and connected to the bases of the mandibles. Each gland consists of secretory units and a reservoir, which is oval-shaped in males and round in females. The body of an ant is divided into three parts: the head, thorax, and abdomen. Important exocrine glands located in the head include the mandibular, propharyngeal (maxillary), and post-pharyngeal glands. The mandibular glands comprise clusters of glandular cells that release their secretions into a reservoir connected to the main duct. The nuclei of these secretory cells are usually round, and their cytoplasm contains structures such as smooth endoplasmic reticulum, Golgi apparatus, small electron-lucent vesicles, mitochondria, and lysosomes, with lipid compounds visible in histochemical slides.³²

Structure and location of mandibular gland in Oecophylla smaragdina

We noted that the mandibles of Oecophylla smaragdina are



Figure 1: Camponotus compressus



Figure 2: Oecophylla smaragdina

elongated and thin. There was a concave side that looked like a sickle-shaped structure. Compared to other teeth, the first tooth is noticeably longer. *Oecophylla smaragdina* has 11–13 teeth in its mandibles. There are several secretory cells with matching duct cells. A collection of glandular cells is visible. Adjacent to one another are the secretory units. A longitudinal section of the mandibles shows that the duct cells pass through the mandibular cuticle at an oblique angle, directing toward the tip of the mandible. *Oecophylla smaragdina*, also referred to as the weaver ant, has mandibular glands in its head. These glands, which are connected to the jaw (mandibles), are important

Structure and location of Dufour's gland in Oecophylla smaragdina

Oecophylla smaragdina has a long, tube-shaped Dufour's gland. Secretory epithelial cells typically form a single layer lining the gland. Pheromones were created by these cells. The gland's central lumen serves as a storage space for its secretions. Cuticular tissue lines the gland's duct, creating a channel for the secretion to travel from the gland to the outside.

Dufour's gland usually they open near the sting apparatus for helping in the release of pheromones during stinging or other defensive behaviours. There are muscle attachments that provides the support for the expulsion of the glandular secretions ¹. The epithelial cells shapes ranges from squamous to a cuboidal in shape appearance. A rounded nucleus is present at the centre of each cell and possess a huge amount of scattered heterochromatin. The cytoplasm consists of a smooth endoplasmic reticulum, huge number of ribosomes and a small number of mitochondria. The cells are usually linked by septate junction. The cell membrane at the base is closely linked to the thin basement membrane. The gland is surrounded by continuous muscle layer and nerve fibres and tracheoles observed in some cases.³³ Discovered and described by Dufour in 1841, the Dufour's gland is an ectodermal structure situated near the base of the abdomen and is believed to serve as an accessory reproductive gland. The spermatheca, poison gland, and Dufour's gland are generally observed to develop as invaginations from the sternum valves. In ants, the duct of the Dufour's gland usually opens into the sting. It is believed that the Dufour's gland evolved from the colleterial gland. In *Oecophylla smaragdina*, the Dufour's gland is positioned near the sting apparatus at the rear of the abdomen, specifically within the metasoma, and is closely linked to the reproductive system.³⁴

Sex Pheromone

In ant subfamilies, workers cannot produce fertile eggs while a living queen remains in the nest. A short annual rise in gyne production takes place when the peak sexualization potential coincides with a reduction in queen pheromone levels. These pheromones impact worker behavior and stimulate developmental changes in the larvae.

Sex pheromones, also known as assembling scents, derive from the Greek words 'Pherein' (meaning to carry) and hormone (to stimulate or excite). These chemicals act as sex attractants, drawing males for mating. The scents released by females typically stimulate sexual arousal in males, leading to copulation.

Male ants typically leave their nest to search for reproductive females. They can navigate against the wind, detecting female locations, and exhibit flitting and crawling behaviors along plant stems where female traces are present. Notably, crushed females do not attract males, as the scent differs; the pheromone remains inactive until the female actively stimulates it. When males were placed in a glass vial previously occupied by a female ant, they became excited and displayed sexual responses upon emerging. In *Camponotus compressus*, sex pheromones from the Dufour's gland include compounds like Nonadecane [C19H40] and Octadecane [C18H38]. Similarly, species like *Harpagoxenus sublaevis* release chemicals that attract males.³⁵

The queen of *Monomorium pharaonis* has been observed to release a sex pheromone from the Dufour's gland. In *Camponotus herculeanus*, both sexes are synchronized by a secretion during their nuptial flight. The females of Rhytidoponera metalica usually release sex pheromone from the pygidial gland.³⁶ *R. metallica* emerges from the nest and, positioned near the entrance, females lower their heads and thoraxes toward the ground while raising their gaster, extending the intersegmental membrane between the last two segments dorsally. This posture attracts males.

Trail Pheromone

Trail pheromones are chemical substances that is released by a single individual and then this chemical is followed by the other individuals. So ants use such chemicals when they find food source they release and that is the trail pheromone till they reach their nest so that other worker individuals can trace the food source by following the trail pheromone. Once the food source gets exhausted they no longer release the pheromone and the trail pheromone evaporates. According to Darwin (1871) had stated that one of the marvellous atoms of matter in the world is the ant's brain. The odour trail is the most established chemical communication.



Figure 3: Schematic outline of the life of a foraging trail

(A)A foraging ant randomly **(a)** finds food (b) It releases a chemical trail starting from the food source till it reaches its nest. (c) The workers present inside the nest gets stimulated in various ways and follow the trail. (d) Ants filled with the pheromones continues to release the trail pheromone so that they don't lose the track. (e) Once the food source gets exhausted completely, they no longer release pheromone. (f) The food gets exhausted and no trail pheromone is released. The intensity of the odour depends on the thickness of the line. Dotted lines show that the intensity of the odour is disappearing.

The trail pheromone of the ant species *Camponotus compressus* have been found to comprise 1-Iodo-2-Methylnonane $[C_{10}H_{21}]$, Nonadecane $[C_{19}H_{40}]$ which is stated in the table-1. In ants it has been observed that that the poison gland, Dufour's gland, metatibial gland, hind-gut gland, rectal gland, Pavan's gland, Pygidial gland or sternal gland are mostly the origin of trail pheromones.

It had been observed by (Bonnet,1779) that the trail pheromone contains an odour which allow the ants to follow the trail. In the *Myrmica* laboratory it was observed that when a worker ant had foraged a sugar solution, after sometime few ants travelled the same route taken by the first ant and when the floor was cleared out the ants wandered.

Alarm Pheromone

Communication by alarm pheromone is a diverse method in eusocial insects such as in ants as they contain a wide variety of compounds in it. A study shows that alarm pheromones are usually processed in their brain. The sources of the alarm pheromone is mostly Dufour's gland, poison gland and the the mandibular gland ⁴⁵. The reaction of ants towards alarm pheromone depends on the concentration of pheromone. There are two main behavioural responses that have been observed in alarm cues, the first one is panic The first response involves individuals reacting to a threat by attempting to flee the area or exhibiting a flight response to disperse from the danger zone. The second response is aggressive, where workers move toward the threat in reaction to it.

It has been noted that these threats depend on several factors, including the concentration of pheromones, the specific compounds within the pheromones, and the size of the colony ¹. It has been observed that many species show a specialises defensive caste, be it guarding the colony of honeybees or morphologically different soldiers in stingless bee, ant, termite and aphid ³⁸. These individuals are the first line of defence for the group and they respond efficiently to aggressive stimuli. It has been noted that these threats

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depend on several factors, including the concentration of pheromones, the specific compounds within the pheromones, and the size of the colony.

 Table 1: Chemical compounds from hexane extract from the mandibular gland of *Camponotus compressus*

| SL NO | Retenti on time | Compound name | Area % | Chemica 1 formula | Molecu lar weight | Bioactivity |
|----------|--------------------|---------------------------|------------|---------------------------------|-------------------------|-----------------------|
| 1. | 9.032 | Undecane-2- methyl | 9.032 | C12H26 | 170.34 | Aggregation pheromone |
| 2. | 10.637 | 1-Iodo-2- Methylnonane | 10.63 7 | C10H21 | 268.18 | Trail pheromone |
| 3. | 10.919 | 1-Undecene, 4-methyl | 10.91 9 | C12H24 | 168.32 | Alarm pheromone |
| 4. | 11.435 | Diethyl Phthalate | 11.43 5 | C12H26 | 170.33 | Repellent pheromone |
| 5. | 12.013 | Nonadecane | 12.01 3 | C19H40 | 268.52 | Trail pheromone |
| 6. | 13.760 | Octadecane | 13.76 0 | C ₁₈ H ₃₈ | 254.50 | Sex pheromone |



Figure 4: GC-MS chromatogram of the hexane extract from the mandibular gland of *Camponotus compressus*. The peak number corresponds to the number in table-1

Table 2: Chemical compounds from hexane extract from the Dufour's gland of *Camponotus compressus*

| SL N O | Retenti on time | Compound name | Are a % | Chemica l formula | Molecul ar weight | Bioactivity |
|--------------|--------------------|----------------------------|------------|-----------------------------------|-------------------------|--------------------------------|
| 1. | 11.906 | Tridecane, 4- methyl | 0.43 | C14H30 | 198.39 | Alarm pheromone |
| 2. | 13.807 | Heptadecane, 2- methyl | 0.52 | C ₁₈ H ₃₈ | 254.5 | Defense Pheromone |
| 3. | 14.302 | Undecane, 4,8- dimethyl | 0.84 | C13H28 | 184.36 | Alarm pheromone |
| 4. | 14.998 | 3-Tridecene | 0.94 | C13H26 | 182.35 | Sex Pheromone |
| 5. | 16.159 | 1-Iodo-2- methylnonane | 0.85 | $C_{10}H_{21}I$ | 268.18 | Trail pheromone |
| 6. | 16.758 | Decane, 2,9- dimethyl | 0.52 | C12H26 | 170.33 | Pacifying pheromone |
| 7. | 18.513 | Hexadecane | 0.61 | C ₁₆ H ₃₄ | 226.45 | Communicat ion pheromone |
| 8. | 18.871 | Tridecane, 2- methyl | 0.56 | C14H30 | 198.39 | Sex pheromone |
| 9. | 19.25 | 1-Tetradecene | 1.22 | C14H28 | 196.37 | Aggregation pheromone |
| 10 | 20.360 | 9-Octadecenal | 1.99 | C ₁₈ H ₃₄ O | 266.46 | Trail pheromone |

| 11 | 20.402 | Benzeneacetic acid | 0.71 | C ₈ H ₈ O | 136.15 | Defense pheromone |
|----|--------|---|-----------|---|---------|----------------------|
| 12 | 20.488 | Oxalic acid, 2- isopropylphenyl -pentyl ester | 3.75 | C ₁₃ H ₁₆ O 4 | 236.26 | Attractant pheromone |
| 13 | 20.584 | Pentafluropropi onic acid, 4- methoxybenzyl | 2.26 | C3HF5O | 164.03 | Alarm pheromone |
| 14 | 20.671 | 4- methoxybenzyl isothiocyanate | 5.84 | C9H9N OS | 179.24 | Repellent pheromone |
| 15 | 20.747 | Propanedinitrile | 5.88 | $C_3H_2N_2$ | 66.06 | Defense pheromone |
| 16 | 20.958 | 4-(4- Methoxyphenyl)-1-butanol | 15.7 6 | C ₁₁ H ₁₆ O 2 | 180.24 | Trail pheromone |
| 17 | 20.995 | 2-(4- Methoxyphenyl) ethanol | 7.98 | C ₉ H ₁₂ O ₂ | 152.193 | Trail Pheromone |



Figure 5: GC-MS chromatogram of the hexane extract from the Dufour's gland of *Camponotus compressus*. The peak number corresponds to the number in table-2.

| Table 3: Chemical compounds from hexane extract from the |
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| mandibular gland of Oecophylla smaragdina |

| SL NO | Retentio n time | Compound name | Area % | Chemical formula | Molecul ar weight | Bioactivity |
|----------|--------------------|--------------------------------------|-----------|--|-------------------------|--|
| 1. | 9.495 | Undecane, 2-methyl | 6.27 | C ₁₂ H ₂₆ | 170.34 | Aggregation pheromone |
| 2. | 9.566 | Undecane, 4,8- dimethyl | 1.84 | C13H28 | 184.36 | Alarm pheromone |
| 3. | 10.694 | 1-Iodo-2- methylnona ne | 7.51 | C ₁₀ H ₂₁ I | 268.18 | Communicati on pheromone |
| 4. | 10.977 | Nonadecane | 8.03 | C19H40 | 268.52 | Sex pheromone |
| 5. | 11.101 | Hexadecane | 1.26 | C ₁₆ H ₃₄ | 226.45 | Worker attractant |
| 6. | 11.424 | Diethyl Phthalate | 21.5 4 | C ₁₂ H ₁₄ O ₄ | 222.24 | Communicati on pheromone |
| 7. | | Pyridine 1- oxide,2,6- dibromo | 4.97 | C ₅ H ₃ Br ₂ N O | 252.89 | Aggregation pheromone |
| 8. | 12.731 | Octadecane | 6.63 | C18H38 | 254.49 | Sex pheromone |
| 9. | 12.904 | Heneicosan e | 7.74 | C ₂₁ H ₄₄ | 296.57 | Chemical communicati on within the nest |



Figure 6: GC-MS chromatogram of the hexane extract from the mandibular gland of *Oecophylla smaragdina*. The peak number corresponds to the number in table 3.

| Table 4: Chemical compounds from hexane extract from the |
|--|
| Dufour's gland of Oecophylla smaragdina. |

| SL NO. | Retenti on time | Compound name | Are a % | Chemical formula | Molecular weight | Bioactivity |
|-----------|--------------------|--------------------------------------|------------|--|---------------------|--------------------------------|
| 1. | 11.332 | Tridecane, 4- methyl | 2.49 | C14H30 | 198.39 | Alarm pheromone |
| 2. | 11.915 | Undecane, 2- methyl | 1.81 | C12H26 | 170.34 | Sex pheromone |
| 3. | 14.658 | Decane,2,3,5, 8-tetramethyl | 3.00 | C ₁₄ H ₃₀ | 198.39 | Pacifier pheromone |
| 4. | 15.015 | 3-Tridecene | 3.66 | C13H26 | 182.35 | Sex pheromone |
| 5. | 16.765 | Octane, 3- ethyl-2,7- dimethyl | | C10H26 | 170.33 | Sex pheromone |
| 6. | 17.475 | 1-Dodecane | 4.77 | C12H26 | 170.34 | Aggression pheromone |
| 7. | 18.871 | Tricosane, 2- methyl | 2.57 | C24H50 | 338.7 | Pacifying signal |
| 8. | 19.258 | 1- Hexadecaneth iol | 6.15 | C16H34S | 258.5 | Communicati on pheromone |
| 9. | 21.100 | Heptadecane,2 -methyl | 4.31 | C ₁₈ H ₃₈ | 254.50 | Alarm pheromone |
| 10. | 19.984 | Heptadecane,2 ,3-dimethyl | 2.17 | C19H40 | 268.52 | Alarm pheromone |
| 11. | 20.279 | Octadecane, 2- methyl | 0.69 | C19H40 | 268.6 | Alarm pheromone |
| 12. | 20.361 | Oleic acid | 4.97 | C ₁₈ H ₃₄ O 2 | 282.46 | Trail pheromone |
| 13. | 21.430 | 1-Hexene, 4,4- diethyl | 2.78 | C10H20 | 140.27 | Communication pheromone |



Figure 7: GC-MS chromatogram of the hexane extract from the Dufour's gland of *Oecophylla smaragdina*. The peak number corresponds to the number in table-4.





Figure 8: Images A and B (Oecophylla smaragdina) C and D (Camponotus compressus) showing the location of mandibular gland under Scanning electron microscope.



Figure 9: Images F, G and H (*Camponotus compressus*) showing the location of Dufour's gland under Scanning electron microscope.

DISCUSSION

The chemical compounds released from these two species of ants *Camponotus compressus* and *Oecophylla smaragdina* mostly play a big role in their defensive and food trailing behavior. A similar study was performed with *Oecophylla longinoda* and it showed that they release hexanal and 1-hexanol to alert the colony if in danger and aggregation responses too and 2-butyloct-2-enal and 3-undecanone for attacking. Research work mentioned in this paper worked with *Oecophylla smaragdina* and it was found that Undecane,2-methyl, Undecane, 4,8-dimethyl and Tridecane, 4-methyl functioned as alarm pheromones.

There are five male specific compounds which have been discovered from seven species of Camponotus (BRAND et al., 1973 a,b) but nothing is similar to those produced by *Camponotus compressus*. It has been observed that alkanes are the most abundant compound found in the Dufour's gland of *Camponotus* genus. It has been observed that N-undecane is the major active alarm pheromone in most of *Camponotus* species excluding *C. herculeanus, C. ligniperda, C. intrepidus, C. sericeus, C. lateralis rebeccae* and *C balzani* and our result showed that the main component secretion of *C. compressus* is similar with the majority

species of *Camponotus* with undecane being the main compound as alarm pheromone ³⁹. Further study needs to be done with Undecane to know whether Undecane are used in other types of pheromones.

CONCLUSION

The mandibular glands of both workers and reproductive of Camponotus compressus and Oecophylla smaragdina produces a variety of pheromones. A large number of pheromones have been collected from the mandibular glands from these two species. In Camponotus compressus the mandibles are used for various functions such as catching prey, fighting, digging to make tunnels, seed crashing or wood scrapping, grooming brood case and trophyllaxis. The mandibular gland of Camponotus compressus usually help them in social communication in the colony, it also secretes alarm pheromone when the colony is attacked. The secretion also contains sex pheromone. The present study also assessed various chemicals present in the gland of Oecophylla smaragdina. It was observed that secretion from the crude gland gave strong response from worker and species. Undecane was found to be in larger quantity in Dufour's gland. There were fatty acids and esters too in the secretion of Dufour's gland for stabilising the secretion.

Recognition among the nest mate was done by odours secreted by the mandibular glands. There is a possibility that the secretions could be used by the workers to differentiate in the caste. These chemicals are secreted from the ant's glands in a continuous basis in low quantities and they get absorbed by cuticular hydrocarbons present all over the body.

It is stated that there is a possibility that chemicals released from the mandibular gland were initially released as alarm pheromones but then slowly volatile formic acid was used as a defensive substance which eventually evolved as an alarm pheromone.

It has been observed that *Oecophylla smaragdina* are varied in color for instance in India it is red in color while green in South-East Asian countries and Australia. The taxonomic level shows smaller scale colony variation. The results of variables maybe because of heritable differences but also can show differences from nutrition as well.

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CONFLICT OF INTEREST STATEMENT

Authors declare that there is no conflict of interest for publication of this work.

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