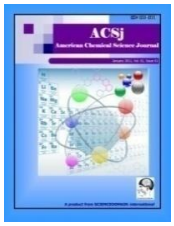

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Chemical Analyses of Volatile Compounds from Cuticular and Non-cuticular Abdominal Glands of African Weaver Ants (*Oecophylla longinoda*)

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Authors' contributions

This work is a collective contribution of two authors. Author OUI designed and supervised the research, interpreted the GC-MS spectra and drew the structures with Chemsketch software and also wrote the manuscript. Author PNE carried out the analyses and supplied some references. Both authors read and approved the final manuscript.

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ABSTRACT

Chemicals from the abdominal glands of African weaver ants (*Oecophylla longinoda*) were extracted with petroleum ether and fifteen pheromonal compounds were characterised using Gas Chromatography-Mass Spectrometry (GC/MS) Technique. The compounds analysed include decane (8.81%), undecane (5.97%), dodecane (4.52%), tridecane (1.75%), hexadecane (2.36%), methyl decanoate (3.31%), (3Z)-tetradec-3-ene (2.10 %), 2,6-dimethylheptadecane (13.29%), 11-octadecenoic acid methyl ester (17.01%), octanoic acid methyl ester (2.89%), 2-methyl nonadecane (11.92%), 1-fluorodecane (5.29%), (3E,11E)-tetradeca-3,11-dien-1-ol (12.02 %), cyclohexane-1,2-diol (3.71%) and 2-ethyl-1-decanol (5.07%). Fourier Transform-Infrared (FT-IR) analysis of the extract showed peaks at 1542.14, 1646.30, 2092.83, 2935.76 and 3427.62 cm^{-1} indicating the presence of alkene, alkyne, alkane and alcoholic compounds. These compounds consisted 50.72 % hydrocarbon, 23.21 % ester, 20.80 % alcohol and 5.29 % alkyl halide. The highest component was 11-octadecenoic acid methyl ester followed by 2,6-dimethylheptadecane. This investigation has

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shown that the cuticular abdominal chemicals derived from African weaver ants are mostly hydrocarbons, esters and alcohols which might be used by the insect as pheromone compounds.

Keywords: African weaver ants (Oecophylla longinoda); Chemicals; Pheromones; GC/MS analysis.

1. INTRODUCTION

Researchers have discovered that insect pheromones consist of mixtures of chemical compounds. In some cases, a single compound is produced but this is very rare. Insects produce pheromones from the chemical ingredients obtained from the breakdown of food that they digest [1]. Some insects acquire host plant chemicals to use them as sex pheromones or sex pheromone precursors. Other host plant volatiles can induce the production or release of pheromones in certain insects and often synergize or enhance insect responses to sex pheromones [2]. The pheromones are produced in the insect's gland cells. These gland cells are either in groups or individual cell forming tissue located on the insect's body which includes the antennae, head, thorax, abdomen or legs. There are exceptions but sex pheromones are only released by insects that are able to carry on reproduction. The age and development of the testes and ovaries as well as the food consumed by the insect determines the timing of release of pheromones [1].

In Africa, *Oecophylla longinoda* is one of the dominant ant species in forest canopies. Its colonies are exceptionally aggressive and territorial, tolerating almost no other insect ant species in the trees they occupy. They also exclude one another, in aggressive interactions so severe as to create narrow, unoccupied corridors that are in effect "no-ant's land" [3]. The weaver ants belong to the genus *Oecophylla* which contains two closely related living species: *O. longinoda* found in Sub-Saharan Africa and *O. smaragdina* found in southern India, Southeast Asia, and Australia [4]. Societies of these ants are monogynous, but they are usually highly polydomous (a single colony occupies numerous nests) [5]. Large societies of *O. longinoda* occupy hundreds of nests spread over an entire tree crown or even over a group of several trees [5,6].

In African weaver ants, exchange of information and modulation of worker behaviour that occur during worker-worker interactions are facilitated by the use of chemical and tactile communication signals. These signals are used primarily in the context of foraging and colony defence [7]. Successful foragers lay down pheromone trails that help recruit other workers to new food sources. Pheromone trails are also used by patrollers to recruit workers against territorial intruders. Along with chemical signals, workers also use tactile communication signals such as attention and body shaking to stimulate activity in signal recipients. Multimodal communication in *Oecophylla* weaver ants importantly contribute to colony self-organization [7,8]. It has been reported that the workers of African weaver ant use pheromones to advertise territories and deter invasion by alien workers and that the substances are effective even in the absence of the marking ants [3]. If other ant colonies expand their territories beyond these confrontational marks, then the colony will break into fighting action and defend their grounds [9]. *Oecophylla* ants produce visible droplets from rectal sac fluids (anal spots) and deposit them on the substrate where they forage. Initial studies on anal spots showed that they serve as territorial pheromones distributed throughout the home range of the colonies [10,11]. It has also been shown that these pheromones are very persistent [10]. Their persistence and cover of entire ant territories may present reliable cues of ant presence and predation risk and therefore warn potential prey [10,12]. It has been reported that the odour trails (pheromones) used by *O. longinoda* in recruiting nest-mates from the leaf nests to new food sources, new terrain, and territorial intruders are produced from the rectal gland [11].

African weaver ants are predatory insects which kill key pests of many tree crops in quest of their prey effectively decreasing the need to use pesticides [13]. They patrol trees continuously for prey. They effectively control a wide range of pests including beetles, sap-sucking bugs, caterpillars, fruit flies and thrips on many tree crops such as citrus, cashew, mango, coconut, oil palm, cocoa and lychee [13]. It has been reported that citrus farmers in Vietnam who did not have weaver ants present in their orchards spent double the amount on agrochemicals than their counterparts [9]. The consideration of the fact that African weaver ants play a significant role in the quality and quantity improvement of

agricultural produce but can only constitute hazards during harvesting made it a necessity to research into their abdominal chemicals probably used as pheromones which indeed would open up another research on behavioural bioassays in order to decipher if these chemicals could be engineered to lure them to crop farms during growing and also to repel them away from such farms during harvesting. Hence, chemical analyses of volatile compounds from cuticular and non-cuticular abdominal glands of African weaver ants (*Oecophylla longinoda*) are hereby reported.

2. MATERIALS AND METHODS

2.1 Insect Collection

Colonies of *O. longinoda* were collected from cocoa trees where they used larval silk to construct woven nests characteristic of the species. These trees are located in Umuariaga near Michael Okpara University of Agriculture, Umudike, Abia State, Nigeria. The insects were housed in a rectangular glass net. Identification and authentication of the organism was carried out in the Zoology Department of Michael Okpara University of Agriculture, Umudike. Only about 650 adult workers were used for the investigation. Hereafter, the word *O. longinoda* or African weaver ant refers only to adult workers unless otherwise stated.

2.2 Extraction of Insect's Abdominal Chemicals

The whole of the cuticular and non-cuticular (the rectal gland and anal spots) abdominal glands of *O. longinoda* were cut with fine brand new razor blade after anaesthetising the organisms by cleaning with chloroform which also removes cuticular surface contaminants. The tissue was extracted in petroleum ether for 20 min. at room temperature. Extract was placed in screw cap vials and stored at −15ºC until analysis.

2.3 Gas Chromatography/Mass Spectrometry (GC/MS) Analysis

GC analysis was carried out in SHIMADZU JAPAN gas chromatography 5890-11 with a fused GC column (OV-101) coated with polymethyl silicon (0.25 mm × 50 m) and the conditions were as follows: temperature programming from 80-280 ℃ held at 80℃ for 1 min, and at 200 ℃ for 4 min (rate 10℃/min), and finally at 280℃ for 5 min (rate 10℃/min). The injection temperature was 250 ℃ . GC/MS analysis was conducted using GCMS-QP 2010 Plus Shimazu Japan with column oven temperature of 80℃. The carrier gas was Helium with a pressure of 108.2 Kpa and linear velocity of 46.3 cm/s. Total flow was 6.2 mL/min, column flow was 1.58 mL/min, injection mode was split, flow control mode was linear velocity, purge flow was 3.0 mL/min and split ratio was 1.0. Also, ion source temperature was 230 ℃ , interface temperature was 250℃, solvent cut time was 2.5 min., detector gain was 0.00 KV, detector gain mode was relative and the threshold was 1000. For the mass spec., start time was 3.0 min., end time was 28.0 min, event time was 0.5 s, scan speed was 1250, and start m/z was 40 while end m/z was 600. The mass spectrum was also equipped with a computer fed mass spectra data bank. Hermle Z 233 M-Z centrifuge, Germany, was used. All solvents used were of analytical grade and were procured from Merck, Germany.

2.4 Components Identification

The components of the extract were identified by matching the peaks with computer Wiley MS libraries and confirmed by comparing mass spectra of the peaks and those from literature [14].

2.5 FT-IR Analysis

FT-IR measurement of the extract was performed using FTIR-8400S Fourier Transform Infrared Spectrophotometer, SHIMADZU, Japan, in a diffused reflectance mode at a resolution of 4 cm^{-1} in sodium chloride (NaCl) pellets in the range 4500-400 cm^{-1}.

3. RESULTS AND DISCUSSION

Cuticular and non-cuticular abdominal chemicals from African weaver ant were studied using GC/MS technique in combination with FT-IR Spectroscopy and fifteen compounds were analysed. From the chromatogram of the extract shown in Fig. 1, the peaks numbered from 1 to 15 represented the volatile chemical compounds. These compounds constituted 50.72% hydrocarbon, 23.21% ester, 20.80% alcohol and 5.29% alkyl halide. Fourier Transform-Infrared (FT-IR) analysis of the extract is shown in Fig. 2. FT-IR showed a peak at 1542.14 cm^{-1} which indicated the presence of C=C from an alkene. Another peak at 1646.30 cm^{-1} was also due to C=C vibration but monosubstituted. Vibrational

frequency at 2935.76 cm^{-1} was as a result of C−H from alkane molecules. A broad peak at 3427.62 cm^{-1} was due to O−H vibration from alcohols. The FT-IR results indicated the presence of alkene, alkane and alcoholic compounds. Table 1 shows the FT-IR results. The highest component was 11-octadecenoic acid methyl ester followed by 2,6-dimethylheptadecane. The mass spectra of these two most abundant compounds are shown in Figs. 3 and 4 while Fig. 5 shows the structures of the isolated fifteen compounds. Table 2 shows the nomenclatures, molecular formulae, molecular weights, retention times, peak areas and the nature of the fifteen volatile compounds analysed.

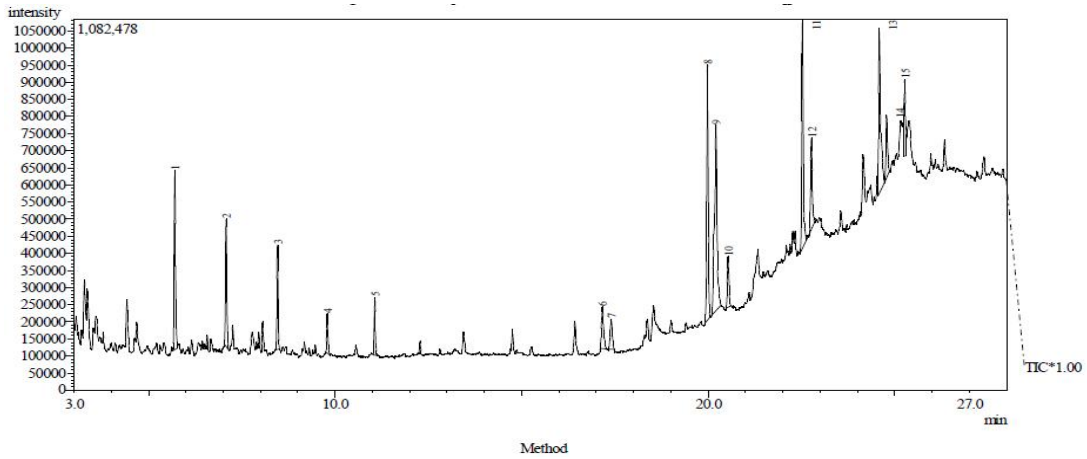

Fig. 1. GC-MS chromatogram of abdominal chemicals from *Oecophylla longinoda*

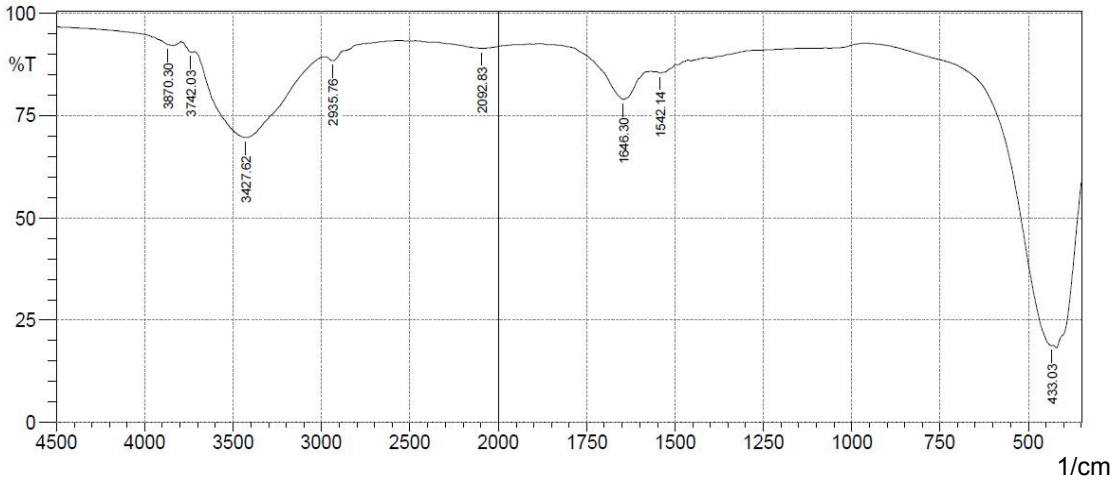

Fig. 2. FT-IR spectra of abdominal chemicals from *Oecophylla longinoda*

Table 1. FT-IR absorption of the extract from *Oecophylla longinoda*

S/N	FT-IR absorption (cm^{-1})	Functional group	Nature of compound
1	1542.14	C=C	Alkene
2	1646.30	C=C	Alkene (monosubstituted)
3	2092.83	C≡C	Alkyne
4	2935.76	C−H	Alkane
5	3427.62	O−H	Alcohol

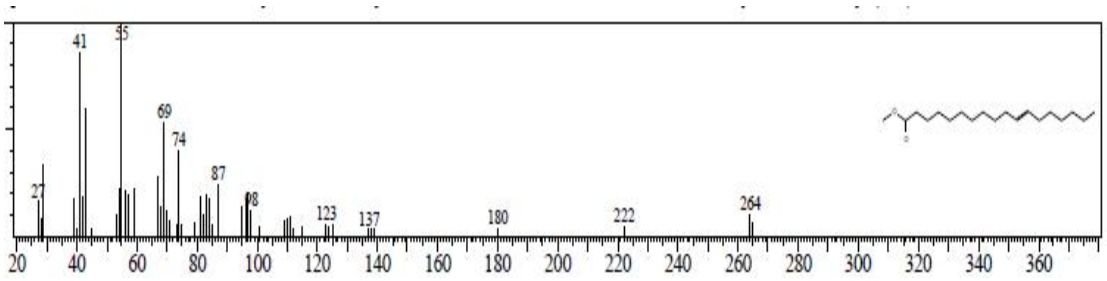

Fig. 3. Mass spectra of 11-Octadecenoic acid methyl ester

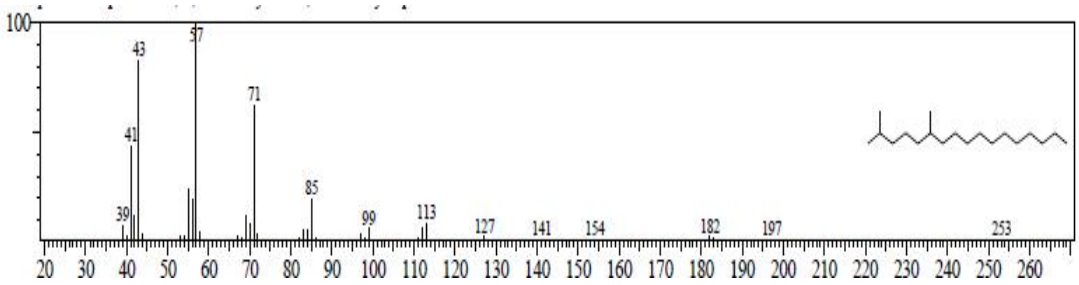

Fig. 4. Mass spectra of 2,6-dimethylheptadecane

The volatile chemical compounds derived from the abdominal glands of African weaver ants in Nigeria have close resemblance with those of other ants. For instance, caprylic acid has been reported to be isolated from the ant *Lasius fuliginosus* where it is used as trail pheromone [15]. This compound has certain similarity with compound 10, octanoic acid methyl ester also known as caprylic acid methyl ester that was isolated from African weaver ant. Compound 10 is suspected to be used as a trail pheromone in the ant. Methylated octadienyl dodecanoates have been isolated from *Gnamptogenys striatula*, another trail pheromone [16]. Meanwhile, compound 6, methyl decanoate was isolated in African weaver ant. The suspected phromonal alcoholic compounds isolated from African weaver ants in this current research were (3E,11E)-tetradeca-3,11-dien-1-ol (12.02 %), cyclohexane-1,2-diol (3.71 %) and 2-ethyl-1-decanol (5.07 %). So many alcoholic compounds have been reported as trail, sex, aggregating and defence pheromones in insects. 4-methyl-dodecan-7-ol, 4-methyl-tridecan-7-ol and 4-methyl-tetradecan-7-ol have been reported to be present as trail pheromones of the ant *Leptogenys peuqueti* [15]. Also, dodecatrienol, dodecadienol and dodecenol have been reported as trail and sex pheromones in many species of termites [16]. (Z)-9-tetradecenol and (Z)-11-hexadecenol have been reported as sex pheromones of *Helicoverpa zea* [17] and *Spodoptera exigua* [18] respectively. Host plants play a key role in the production and use of pheromones by herbivorous insects through larval or adult sequestration of chemically active compounds and pheromone precursors [19, 20]. This then follows that host plants could affect the nature of pheromonal semiochemicals secreted by insects suggesting the possibility of certain species secreting different or slightly different semiochemicals. The hydrocarbon molecules isolated from the African weaver ants were alkanes and alkenes. Hydrocarbons have been reported to be used as pheromones in other insects. It is noteworthy that these compounds that has been isolated from African weaver ants might be used as trail, sex, aggregating and alarm as well as defence pheromones by the insects. However, behavioural bioassays are required.

The African weaver ants have been testified as the farmers' friend since they serve as predators against insect pests on their farms. These volatile chemical compounds might be used to lure and aggregate the ants into farms where they help to control pest thereby improving agricultural produce. Further research is needed to know the effectiveness of these compounds. Gregarizing pheromones would cause individual ants to form low-density populations and consequently form a colony. These chemicals might artificially be used to manipulate the

presence or absence of the African weaver ants on crop farms and orchards as well as repel other insects (when used as territorial pheromones) which are prey to *O. longinoda* thereby reducing their negative effects on agricultural produce. This investigation has shown that the volatile compounds derived from African weaver ants are mostly hydrocarbons, esters and alcohols.

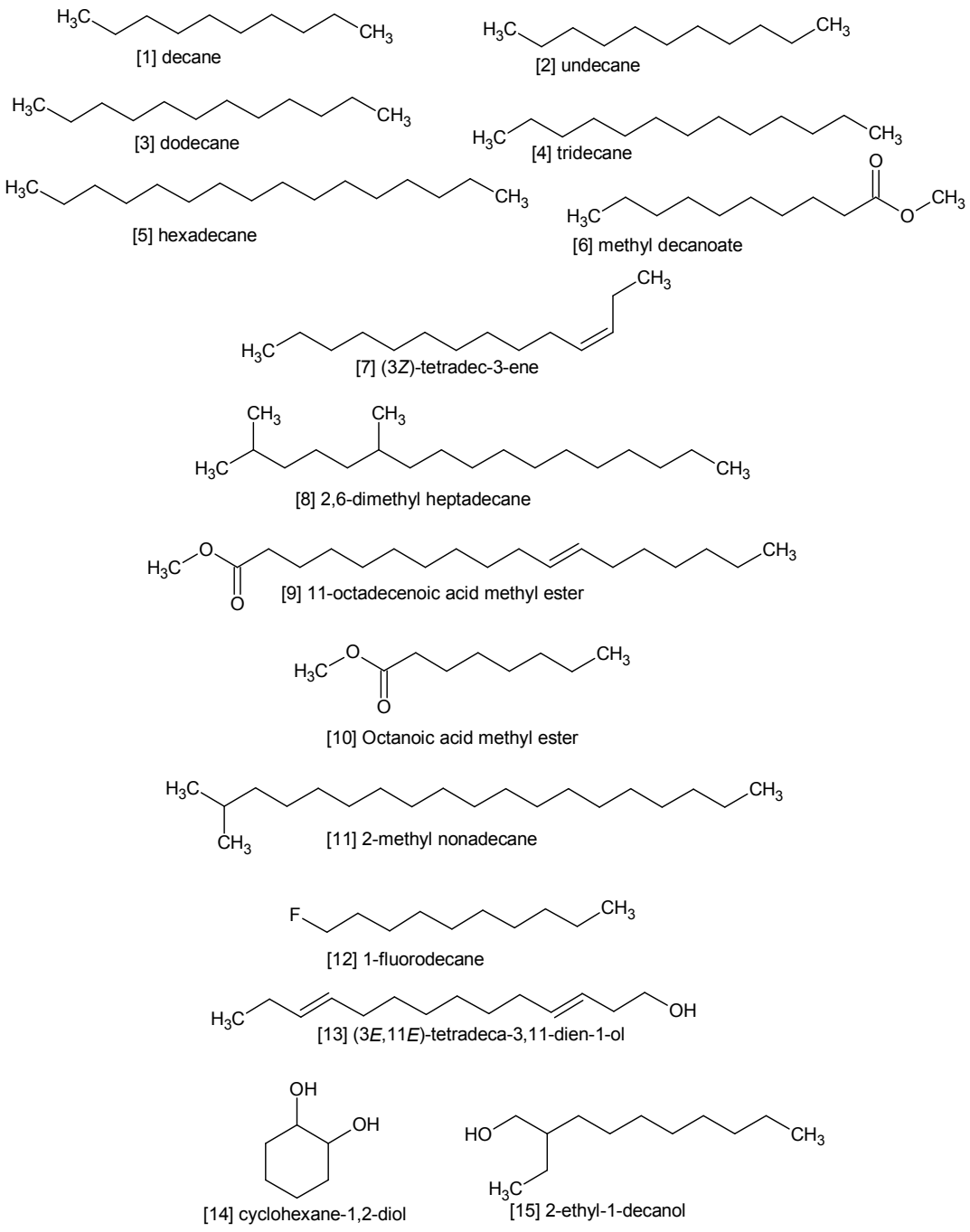

Fig. 5: Structures of volatile chemicals identified from the GC/MS result of *Oecophylla longinoda* extract

Table 2. Chemicals identified from the GC-MS analysis of the abdominal extract of *Oecophylla longinoda*

Chromatogra peak	Compound name	Molecular formula	Molecular weight	Retention time (min)	Peak area (%)	Nature of compound
1	Decane	$C_{10}H_{22}$	142	5.702	8.81	Hydrocarbon
2	Undecane	$C_{11}H_{24}$	156	7.077	5.97	Hydrocarbon
3	Dodecane	$C_{12}H_{26}$	170	8.455	4.52	Hydrocarbon
4	Tridecane	$C_{13}H_{28}$	184	9.788	1.75	Hydrocarbon
5	Hexadecane	$C_{16}H_{34}$	226	11.064	2.36	Hydrocarbon
6	Methyl decanoate	$C_{11}H_{22}O_2$	186	17.164	3.31	Ester
7	(3Z)-Tetradec-3-ene	$C_{14}H_{28}$	196	17.400	2.10	Hydrocarbon
8	2,6-Dimethylheptadecane	$C_{19}H_{40}$	268	19.980	13.29	Hydrocarbon
9	11-Octadecenoic acid methyl ester	$C_{19}H_{36}O_2$	296	20.204	17.01	Ester
10	Octanoic acid methyl ester (caprylic acid methyl ester)	$C_9H_{18}O_2$	158	22.530	2.89	Ester
11	2-Methylnonadecane	$C_{20}H_{42}$	282	22.526	11.92	Hydrocarbon
12	1-Fluorodecane	$C_{10}H_{21}F$	160	22.764	5.29	Alkyl halide
13	(3E,11E)-Tetradeca-3,11-dien-1-ol	$C_{14}H_{26}O$	210	24.584	12.02	Alcohol
14	Cyclohexane-1,2-diol	$C_6H_{12}O_2$	116	24.781	3.71	Alcohol
15	2-Ethyl-1-decanol	$C_{12}H_{26}O$	186	25.265	5.07	Alcohol

4. CONCLUSION

The volatile compounds from the abdominal glands of the African weaver ants were analysed with GC/MS and FT-IR techniques which revealed the presence of fifteen compounds. These compounds consisted 50.72% hydrocarbon, 23.21% ester, 20.80% alcohol and 5.29% alkyl halide. The highest component was 11-octadecenoic acid methyl ester followed by 2,6-dimethylheptadecane. This investigation has revealed that the cuticular and non-cuticular abdominal glands from African weaver ants are made up of a mixture of compounds. The synthetic forms of these compounds might be applied in luring and aggregating the ants to form a colony in farms where they are used in pest management and control. However, behavioural bioassays are required to decipher the pheromonal functionality of these compounds. This report has opened up another research involving the investigation and validation of the component with the greatest attractiveness to the ants. This method of pest control ensures environmental friendliness as the problems posed by dangerous chemicals are eliminated.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Braun T. Pheromones from Available: http://www.pcaapplicatorhours.com/pheromones.html viewed 18/08/2010.
2. Reddy GVP, Guerrero A. Interactions of insect pheromones and plant semiochemicals. TRENDS in Plant Science. 2004;9(5):253-261.
3. Holldobler B, Wilson EO. Colony-specific territorial pheromone in the African weaver ant, *Oecophylla longinoda* (Latreille). Ecology. 1977;74(5):2072-2075.
4. Tree of life web project. Oecophylla; 2004. Available:http://tolweb.org/Oecophylla/22908
5. Wojtusiaki J, Odziriska EJ. Factors influencing the responses to nest damage in the African weaver ant *Oecophylla longinoda* (Latreille). Acta Neurobiol. Exp. 1993;53:401-408.
6. Holldobler B, Wilson EO. The ants. Springer-Verlag, Berlin. 1990;732.
7. Anderson C, McShea DW. Individual versus social complexity, with particular reference to ant colonies; 2001.
8. Yulia R. The advantage of weaver Ants in Agriculture. SMA Negeri 1 Baleendah Bandung; 2010.
9. Nodwell E. Weaver ants, a photographic study of behaviour in Bijilo forest, The Gambia.
10. Offenberg J. The distribution of weaver ant pheromones on host trees. Insect, Soc. 2007;54:248-250.
11. Holldobler B, Wilson EO. The multiple recruitment systems of the African weaver ant *Oecophylla longinoda* (Latreille) (*Hymenoptera* Formicidae). Behav. Ecol. Sociobiol. 1978;3:19–60.
12. Offenberg J, Nielsen MG, Macintosh D, Havanon S, Aksornkoae S. Evidence that insect herbivores are deterred by ant pheromones. Proc. R. Soc. Lond. Ser. B: Biol. Sci. 2004;271:433-435.
13. Vayssieres JF, Van Mele P. Weaver ants help farmers to capture organic markets. Pesticides News. 2007;75. 15 December 2006. Available: http://www.juragricole.com
14. Igwe OU, Abii T. Characterization of bioactive sesquiterpenes, organic acids and their derivatives from the leaves of *Psidium guajava* Linn. International Research Journal of Pure and Applied Chemistry. 2014;4(4):456-467.
15. El-Sayed AM. The pherobase: database of insect pheromones and semiochemicals; 2008. Available: http://www.pherobase.net
16. Costa-leonardo AM, Casarin FA, Lima JT. Chemical communication in Isoptera. Neotropical Entomology. 2006;38(1):001-006.
17. Light DM, et al. Host-plant green volatiles synergize the synthetic sex pheromones of the corn earworm and codling moth (*Lepidoptera*). Chemoecology. 1993;4:145–152.

18. Dickens JC, et al. Detection and deactivation of pheromone and plant odor components by the beet armyworm, *Spodoptera exigua* (Hubner) (*Lepidoptera*: *Noctuidae*). J. Insect Physiol. 1993;39:503–516.
19. Nishida R. Sequestration of defensive substances from plants by *Lepidoptera*. Annu. Rev. Entomol. 2002;47:57–92.
20. Gadi VPR, Angel G. Interactions of insect pheromones and plant semiochemicals. TRENDS in Plant Science. 2004;9(5):253-261.

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