

Essential Oil Composition of Three Compositae- *Aspillia africana*, *Chromolaena odorata*, *Syndrella nodiflora* and One Labiatae- *Hyptis suaveolens* Plants Commonly Utilized as Rabbit Feeds

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Abstract

Rabbits showed varied preferences in the order of their consumption of these four plants as meal: *Syndrella nodiflora* > *Hyptis suaveolens* > *Aspillia africana* > *Chromolaena odorata*. Chemical compositions with differences in levels of nutrients, palatability and odour may be responsible for the preferences observed. This study was targeted at examining volatile chemical constituents of both leaf and stem (parts utilized as meal) essential oils, which are likely responsible for the preferences observed. GC and GC-MS analyses conducted on eight essential oil samples revealed thus: Nine major compounds were responsible for 59.8 % of *Syndrella nodiflora* leaf essential oil. In its stem essential oil, twelve compounds which account for 40.5% of the oil were identified, these were dominated by terpenes (28.4%). Fifteen compounds were identified in leaf essential oil of the second preferred plant meal- *Hyptis suaveolens*, and was dominated by terpenes (61.3%), its most abundant compound being p-cineole (17.6%). Thirteen compounds make up 76.2% of its stem oil, two of which are yet to be identified; Seven (77.7%) and nine (75.7%) compounds were identified respectively in the leaf and stem volatile oils of *Aspillia africana* the third preferred plant meal. Its leaf oil is dominated by α -bisabolol (46.4%), while terpenes make-up 70.4%. The stem oil has its most abundant compound to be (+) D-nerolidol (17.5%). Other interesting classes of compounds in the stem oil include hydrocarbons (23.5%), acid (9.1%) and alcohol (6.3%); Thirteen compounds each were identified in leaf and stem oils of *Chromolaena odorata*, the least preferred plant as meal by the rabbits, these compounds account for 57.3% and 72.3% of the oils respectively. Caryophyllene oxide (11.4%) is the dominant compound in leaf oil, while the stem oil is 3Z-3-heptadecen-5-yne (13.6%). Both oils are characterized with the dominance of terpenoids (45.4% and 47.3% respectively). Our results fully agree with compositions of the oils that have been reported in literature. This study presents compositions of stem essential oils of *Syndrella nodiflora*, *Hyptis suaveolens* and *Chromolaena odorata* for the first time.

Key words: Compositae, Labiatae, essential oil, GC and GC-MS.

Introduction

Compositae is one of the outstanding five families with the largest number of species in Nigeria [1]. They are utilized as feeds, cure for ailments and are of great economic importance [2]. The four plant species in this study are readily available, they are comprised of three Compositae (*Aspillia*

africana; *Chromolaena odorata* and *Syndrella nodiflora*) and *Hyptis suaveolens* (a Labiatae); hence, the four are utilized as alternate non-conventional rabbit feeds. The four plants have many unique folklore uses, with strong biological activities. There are many reports on studies involving evaluating the effects of supplement concentrate in diets of animals like rabbits [3-6].

Aspillia africana [C.D. Adams] leaves are utilized in traditional medicine for reducing and stopping bleeding from wounds, as well as accelerating healing of wounds. They are used for curing of bees and scorpion stings; removing opacity from eyes; and to cure skin infections. This is the reason why it is called haemorrhage plant or wild sunflower, and in Yorubaland it is referred to as 'abamoda' and

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'ako yunyun [7]. In francophone territories, it is used in fumigation to remove guinea worm. *A. africana* can be classified among plants with low toxicity [8]. It contains high amounts of secondary metabolites which justifies its wide applications; these include terpenoids, sterols, saponins, flavonoids, alkaloids, resins and tannins. It also contains carbohydrates, minerals and vitamins [9-11]. Usman et al. (2010) reported that GC-MS analyses of leaf oil has abundance of sesquiterpenes, with α -cubebene as the major component [12]. Ogunwande et al. (2012) also reported the presence of germacrene D, α -pinene, β -caryophyllene and caryophyllene oxide as the major compounds of volatile oil of *A. africana* [13]. Ethanol extract of dried leaves of *A. africana* yielded white crystals which was identified by spectroscopy to be inositol [14].

Chromolaena odorata also known as Akintola or Awolowo plant by Yorubas in southwestern Nigeria is a widely spread weed [15, 16]. The plant has high therapeutic potentials for treating coughs, colds and skin diseases; it is known to have antibacterial, anti-oxidant and anti-inflammatory activities [17-21]. *C. odorata* is rich in secondary metabolites like flavonoids, steroids and alkaloids [17-19, 22]. The shrub inhibits growth of insects, fungi and plants [23, 24]. The leaf volatile oil has been reported to be dominated by terpenoids consisting of α - and β -pinene, (*E*)-caryophyllene, δ -cadinene, α -copaene, caryophyllene oxide, germacrene-D, δ -humulene, β -copaen-4-ol, geijerene and pregeijerene [20, 21]. Twenty-nine compounds were identified in the root volatile oil which was found to be of different chemotypes compared to the leaf oil. Main root constituents were himachalol, 7-isopropyl-1,4-dimethyl-2-azulenol, andro-encecalinol, and 2-methoxy-6-(1-methoxy-2-propenyl)naphthalene; phenyl derivatives, sesquiterpenoids, long-chain hydrocarbons and monoterpenoids [25]. *Chromolaena odorata* and *Syndrella nodiflora* were evaluated as

pesticidal and repellent plants for controlling pests [26].

Syndrella nodiflora (L.) Gaertn is one of the highly rich proteinous leafy plants which serve as grasshopper (*Zonocerus variegatus*) meal. Njidda and Isidahomen (2010) reported that grasshoppers which fed on leafy plants were found to serve as better replacement for the known fish meal offered to growing rabbits. It was also reported that it had no adverse effects on the haematological parameters, serum biochemistry and carcass characteristics of the rabbits.³ Vegetation survey conducted on plant species within feeding heights of giant tortoise (*Dipsochelys arnoldi*) in Grande Barbe by Pemberton and Gilchrist (2009) revealed that *Syndrella nodiflora* is one of the selectively foraged plant species by the tortoise [27].

Syndrella nodiflora extract is known to contain steroids, triterpenoids, tannins, phenolic compounds, alkaloids, sugars, flavonoids and saponins. It has also been reported to have anti-inflammatory and antimicrobial activities [28]. Classes of compounds detected in the leaf oil of *Syndrella nodiflora* were aliphatic alcohols, monoterpenes, sesquiterpene hydrocarbons and oxygenated sesquiterpenes. Major components of the leaf oil were β -caryophyllene, β -farnesene, germacrene-D and β -cubebene [29].

Hyptis suaveolens (L) Poit, a Labiatae, is commonly referred to as 'bush tea leaves, bush mint' [30]; it is an edible annual shrub which is consumed as vegetable and acts as appetizer because of the flavoured essential oils [31]. *H. suaveolens* is applied in traditional medicine because of its bioactivities such as antimicrobial, anti-oxidant, antifungal, insecticidal and anti-cancer activities [30-36]. Leaf ethanolic extract showed healing property with supportive role of antioxidant [37]. Phytochemical screening shows the presence of starch, proteins, tannins, saponins, fats, alkaloids and glycosides [34]. From the root of *Hyptis suaveolens*, were isolated two

triterpenoids with similar spectroscopic data to betulin and betulinic acid [38]. Phytochemicals earlier reported include β -sitosterol, oleanolic acid, urs-12-en-3 β -ol-27-oic acid (α -peltoboykinolic acid) [39]. Essential oil of *H. suaveolens* had its major and active compounds from GC-MS analyses, to be sabinene, α -terpinolene, 1,8-cineole and β -caryophyllene, eucalyptol, ellemene, β -pinene, germacrene, phellandrene, β -ocimene, terpineol [31, 33, 40-44]. Physico-chemical properties of the oil are documented [44]. A slight difference observed in essential oil content of *H. suaveolens* was suggested to be due to differences in environmental factors [31, 41, 43].

This study was targeted at examining chemical compositions of both leaf and stem essential oils of the four plants; *Aspilia Africana*, *Chromolaena odorata*, *Syndrella nodiflora* (Compositae) and *Hyptis suaveolens* (Labiatae) which are utilized as alternate feed for rabbits.

Materials and Methods

Plant Material

Leaf and stem samples of *Aspilia Africana*; *Chromolaena odorata* and *Syndrella nodiflora* (Compositae) and *Hyptis suaveolens* (Labiatae) were collected around the Faculty of Science, mini campus of Olabisi Onabanjo University, Ago-Iwoye, Ogun-State, Nigeria in 2009. The plants were authenticated by a botanist in the herbarium, Department of Botany and Microbiology, University of Ibadan, Ibadan.

Isolation of Essential Oils

Each of the four plants was separated into leafy aerial and stem parts. Each part was crushed and hydrodistilled for 2.5 hours in an all glass Clevenger-type apparatus designed to British Pharmacopoeia specifications, with a small quantity of distilled *n*-hexane (0.3 ml),

which are solvent peaks on the chromatograms between 3 to 5 mins retention time.

Gas Chromatography

Each of the eight essential oils was subjected to GC analyses on GC-2010[AOC-20i] gas chromatograph. Column oven temperature was 60°C, injection temperature of 250°C, split injection mode at 100.2 kPa; column flow of 1.61 ml/min and total flow of 6.2 ml/min; 1.0 split ratio; oven temperature programming was 60°C (for 5 mins) and at the rate of 5°C/min till 140°C, 15°C/min till 280°C.

Gas Chromatography-Mass Spectrometry

The GC-MS analyses were performed on GC-MS QP2010 Plus. Ion source temperature 200°C; interface temperature 250°C; solvent cut time 2.5 min; with relative detector gain mode and threshold 3000; scan MS ACQ mode; mass range of *m/z* 40-400.

Identification of Components

Identification of the essential oil components was based on their retention indices (determined with reference to a homologous series of *n*-alkanes), and by comparison of their mass spectral fragmentation patterns in computer matching against library data base as well as in-house "Başer Library of Essential Oil Constituents" built up by genuine compounds and components of known oils such as Joulain and Koenig (1998), Adams (1995), and Massada (1976) [45-47] Library.

Results and Discussion

The percentage yield of the essential oils ranged from 0.14% to 0.43%. The leaf oil of *H. suaveolens* had the highest yield (0.43%) while the stem oil of *S. nodiflora* had the lowest yield (0.14%). The oils possessed different characteristic odours (Table 1).

Table 1: Yields of Essential Oils procured from Leaf and Stem Parts of *Aspillia africana*; *Chromolaena odorata*, *Syndrella nodiflora*, and *Hyptis suaveolens*

Plant	Parts	Weight of sample (g)	Weight of volatile oil procured (g)	% Yield of essential oil procured	Physical Characteristics
<i>Aspillia africana</i>	Leaf	250	0.98	0.39	Whitish with leafy herbal smell
	Stem	300	0.87	0.29	
<i>Chromolaena odorata</i>	Leaf	200	0.55	0.28	Colourless plain oil with characteristic odour
	Stem	150	0.59	0.39	
<i>Syndrella nodiflora</i>	Leaf	300	0.95	0.32	Milky with characteristic herbal smell
	Stem	250	0.35	0.14	
<i>Hyptis suaveolens</i>	Leaf	120	0.51	0.43	Milky white with herbal odour
	Stem	100	0.15	0.15	

Each of the leaf and stem oils of the four plants were analyzed using GC and GC-MS. The chromatograms are presented in Figures 1-8. Constituents of the essential oils of the leaf and stem of each plant are presented in Tables 2-9.

It was observed that rabbits showed the following order in their consumption of the four plants as meal: *Syndrella nodiflora* > *Hyptis suaveolens* > *Aspillia africana* > *Chromolaena odorata*. Our study examined volatile chemical compositions of both leaf and stem essential oils. The two plant parts are utilized as meal, hence their chemical compositions are likely responsible for the preferences observed. GC and GC-MS analyses conducted on the eight essential oils will be discussed and presented in this order.

Literature review indicated that *Syndrella nodiflora* is rich in protein, the leaves gave high essential oil yield of 0.32%, which was whitish with characteristic herbal smell

(Table 1). The Nigerian *Syndrella nodiflora* leaf oil contained nine major compounds constituting 59.8% of the oil out of which six were identified (Table 2) (Fig. 1). Aalbersberg and Singh, 2006 reported that the major compounds in *S. nodiflora* leaf volatile oil were sesquiterpenoids [29]. The unidentified prominent compounds in chromatogram of this (Nigerian) sample were likely to be sesquiterpenoids. However, we were able to identify the presence of hydrocarbons and amide in this (Nigerian) sample.

Twelve identified compounds in stem essential oil of *S. nodiflora* (Table 3) were responsible for 40.5% of the constituents, and were dominated by terpenes (28.4%) (Fig. 2). Other compounds present are hydrocarbons (9.0%), alcohol (1.5%), ketone (0.8) and amide (0.8%). *S. nodiflora* stem essential oil has not been previously reported.

Table 2: Chemical Composition of Essential Oil obtained from the Leaves of *Syndrella nodiflora*

Retention time [mins] ^a	Peak No ^b	MS [Base peak+most abundant peaks] ^c	Identified compound ^d	%TIC ^e	Calculated RI ^f
4.9	-	-	u-i. ^g	13.3	
5.2	-	-	u-i. ^g	10.6	
6.1	-	-	u-i. ^g	15.9	
22.3	6	57,71,43,85,41,99,55,56,70,42,69,113,	n-tetradecane	1.3	1976
30.9	2	57,71,85,43,99,113,40,	n-pentadecane	4.0	2830
31.5	3	57,71,85,43,99,41,113,55,56,69,70,83,97,	2-ethylhexyl isohexyl sulfurate	2.7	3144
32.2	4	57,71,85,43,99,113,55,69,97,40,56,70,83,	Eicosane	4.0	3162
33.0	5	57,71,85,43,99,41,113,41,55,97,56,69,70,83,	Heneicosane	2.7	3183
34.1	1	59,72,55,43,69,97,98,60,	Z-13-docosenamide	5.3	3212

Key: ^a Retention time in minutes in increasing order of elution of compounds; ^b According to GC [Figure 1]; ^c [m/e] values of base peak 1st stated, and other prominent ions; ^d see identification of components; ^e Estimated total ion concentration in %; ^f Retention Index determined with reference to homologous series of n-alkanes; ^g u-i-unidentified compound present in significant amount.

Table 3: Chemical Composition of Essential oil Obtained from the Stem of *Syndrella nodiflora*

Retention time [mins] ^a	Peak No ^b	MS [Base peak+most abundant peaks] ^c	Identified compound ^d	%TI C ^e	Calculated RI ^f
7.7	11	82,67,81,55,83,96,41,53,54,66,68,95..109,	cis-pinane	1.5	724
9.1	12	105,57,43,71,41,69,85,120,	3-octanone	0.8	764
10.1	10	93,41,69,91,77,79,80,53,92,94,	myrcene	1.5	792
11.4	1	68,67,93,79,94,107,41,53,77,92,91,107,	D-limonene	14.9	1079
22.3	3	57,71,43,85,41,56,55,70,99,	n-tetradecane	3.0	1976
22.9	5	93,69,41,79,105,107,81,55,67,77,106,	caryophyllene	1.5	1990
23.4	2	69,93,41,79,81,67,55,91,53,77,105,120,133,	α -Santalene	9.0	2332
25.1	4	57,71,43,85,41,99,70,56,55,69,113,	n-hexadecane	3.0	2373
27.0	6	57,71,43,85,41,99,55,56,113,	n-heptadecane	1.5	2419
29.7	8	59,97,83,55,69,72,57,43,41,111,70,71,125,	3-hexadecanol	1.5	2799
30.9	9	57,71,85,43,99,56,98,84,113,	n-eicosane	1.5	2830
34.1	7	59,72,55,69,43,41,97,112,	Z-13-docosenamide	0.8	3212

^a Retention time in minutes in increasing order of elution of compounds; ^bAccording to GC [Figure 2]; ^c[m/e] values of base peak 1st stated, and other prominent ions; ^dsee identification of components; ^eEstimated total ion concentration in %; ^fRetention Index determined with reference to homologous series of n-alkanes.

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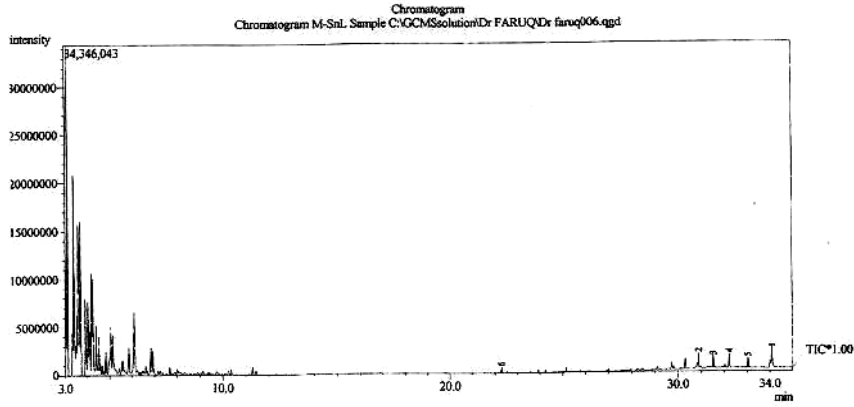


Fig. 1: Gas chromatogram of the leaf essential oil of *Syndrella nodiflora* (L) Gaertn.

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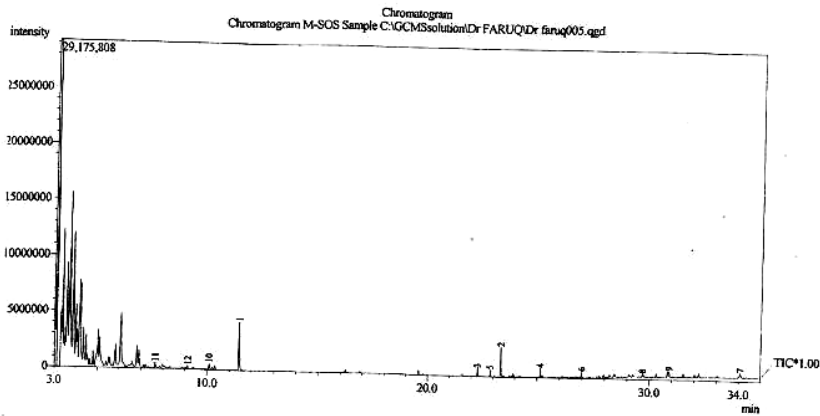


Fig. 2: Gas chromatogram of the stem essential oil of *Syndrella nodiflora* (L) Gaertn.

Fifteen compounds were identified in leaf essential oil of *Hyptis suaveolens*, which is dominated by terpenoids (61.3%) (Fig. 3). Its most abundant compound is p-cineole (17.6%) (Table 4). The other fourteen compounds are as listed in Table 4. Our result is in agreement with past reports [31, 33, 40-44], including that of Asekun et al. 1999 [42]. We also agreed with the suggestion that slight differences observed in essential oil content

of *H. suaveolens* may be due to differences in environmental factors.

Thirteen compounds make up 76.2% of *Hyptis suaveolens* stem oil. Two of these are yet to be identified (Fig. 4). The eleven that have been identified are listed as shown in Table 5. The stem oil does not have high terpenoids content, compared to the leaf oil, the two are of different chemo-types. There are no earlier reports on the stem oil.

Table 4: Chemical Composition of Leaf Essential Oil of *Hyptis suaveolens*

Retention time [mins] ^a	Peak No ^b	MS [Base peak+most abundant peaks] ^c	Identified compound ^d	%TIC ^e	Calculated RI ^f
8.2	8	93,91,92,77,79,41,105,94,80,67,	α -pinene	3.3	739
9.6	2	93,91,77,79,41,80,94,69,92,43	β -phellandrene	10.8	778
11.6	1	43,81,84,108,71,69,41,	p-cineole	17.6	1084
12.8	13	71,43,93,69,81,41,111,79,	5-isopropyl-2-methylbicyclo[3.1.0]hexan-2-ol	1.8	1113
13.8	15	71,43,93,81,69,41,79,111,55,	cis- β -terpineol	1.0	1138
16.4	14	71,93,43,111,69,41,	linalyl acetate	1.2	1509
21.0	10	93,121,107,79,81,91,41,94,105,67,	δ -elemene	2.0	1946
22.0	11	105,119,93,91,81,161,92,41,	Copaene	1.6	1969
22.3	12	93,81,68,107,67,79,57,41,43,55,71,53,	1-ethenyl-1methyl-2,4-bis-(1-methylethenyl)-1S-1 α ,2 β ,4 β -cyclohexane	2.3	1976
22.9	3	93,69,41,91,79,105,55,81,107,133,	4,11,11-trimethyl-8-methylenebicyclo[7.2.0]undec-4-ene	10.5	1990
23.9	5	105,91,81,161,79,41,119,55,93,77,67,69,	germacrene D	4.6	2344
24.1	4	93,121,107,79,41,105,91,81,94,67,77,119,	germacrene B	9.5	2349
25.2	6	43,79,93,41,69,91,95,67,81,96,55,107,	caryophyllene oxide	3.8	2376
25.4	7	93,119,91,79,43,107,41,55,68,105,77,	Z- α -trans bergamotol	3.6	2380
26.3	9	93,119,91,79,107,55,105,43,41,77,105,92,81,68,	Z- α -trans-bergamotolacetate	2.3	2402

^a Retention time in minutes in increasing order of elution of compounds; ^b According to GC [Figure 3]; ^c [m/e] values of base peak 1st stated, and other prominent ions; ^d see identification of components; ^e Estimated total ion concentration in %; ^f Retention Index determined with reference to homologous series of n-alkanes.

Table 5: Chemical Constituents of Stem Essential oil of *Hyptis suaveolens*

Retention time [mins] ^a	Peak No ^b	MS [Base peak+most abundant peaks] ^c	Identified compound ^d	%TIC ^e	Calculated RI ^f
3.5			u.i. ^g	31.4	
3.7			u.i. ^g	14.8	
6.1	10	91,106,105,77,78,79,51,63,40	Xylenes	3.7	507
6.9	11	43,57,41,85,56,	2,3-heptanedione	1.1	538
11.6	9	81,71,41,69, 40,43,	Neomenthol	0.4	1084
16.9	8	57,43,71,41,85,	n-dodecane	0.4	1521
22.3	7	57,43,71,85,41,55,42,56,70,99,	3,7-dimethyldecane	2.6	1976
24.1	6	93,41,71,107,121,91,43,67,77,81, 105,	thymoquinone	1.1	2349
25.2	5	57,43,71,41,85,55,56,69,70,	3,8-dimethylundecane	2.6	2376
27.0	4	57,71,85,43,41,55,99,	1-iododecane	1.5	2419
28.2	3	60,73,43,83,97,41,55,57,69,71,	undecanoic acid	1.1	2762
29.7	2	59,72,43,57,85,41,71,	dodecanamide	2.6	2799
30.8	1	59,72,55,41,43,	octadecenamide	12.9	2827

^a Retention time in minutes in increasing order of elution of compounds; ^bAccording to GC [Figure 4]; ^c[m/e] values of base peak 1st stated, and other prominent ions; ^dsee identification of components; ^eEstimated total ion concentration in %; ^fRetention Index determined with reference to homologous series of n-alkanes; ^gunidentified compound present in significant amount.

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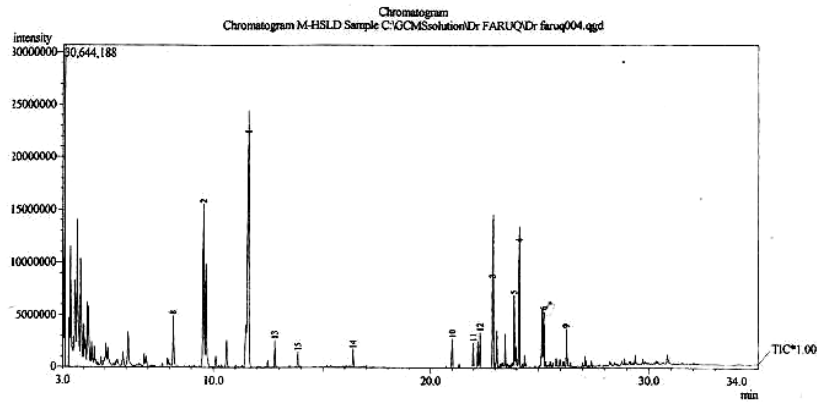


Fig. 3: Gas chromatogram of the leaf essential oil of *Hyptis suaveolens* (L.) Poit.

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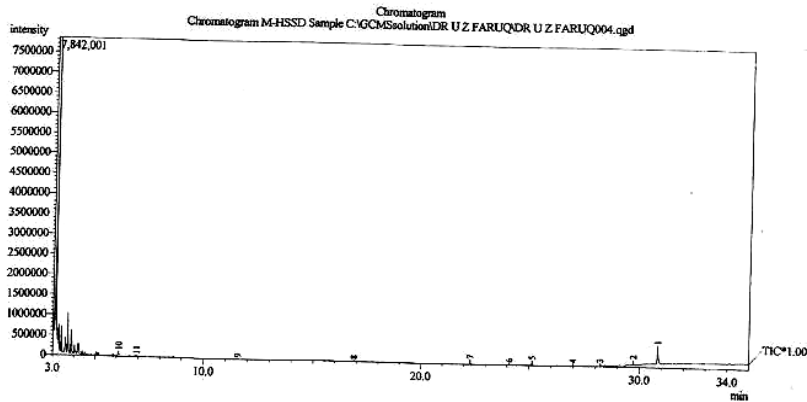


Fig. 4: Gas chromatogram of the stem essential oil of *Hyptis suaveolens* (L.) Poit.

In *Aspillia africana* seven compounds make-up 77.7% of its leaf oil (Table 6) (Fig. 5), while nine constituents listed are responsible for 75.7% of the stem oil (Table 7) (Fig. 6). The major component of the leaf oil is α -bisabolol (46.4%), with terpenes making up 70.4% of the oil; while the stem oil has its

most abundant compound to be (+) D-nerolidol (17.5%). The other interesting classes of compounds in the stem oil are hydrocarbons (23.5%), acid (9.1%) and alcohol (6.3%). (Tables 6 and 7) Our result is in agreement with earlier reported abundance of sesquiterpenoids in the leaf oil [12, 13].

Table 6: Chemical Composition of Leaf Essential oil of *Aspillia africana*

Retention time [mins] ^a	Peak No ^b	MS [Base peak+most abundant peaks] ^c	Identified compound ^d	%TIC ^e	Calculated RI ^f
6.1	7	91,106,105,77,65,51,79,78,52	xylene	2.1	508
22.3	6	57,43,71,85,41,99,55,56,	n-tetradecane	2.1	1976
23.4	4	69,41,93,79,67,55,81,	Z- β -farnesene	10.4	2332
23.9	2	105,91,81,79,41,93,119,55,161,67,	germacrene-D	7.3	2344
24.6	3	93,80,67,79,41,109,119,107,121,	α -caryophyllene	6.3	2361
26.2	1	69,43,109,119,93,41,71,	α -bisabolol	46.4	2400
30.8	5	59,72,55,41,43,69,95,112,126,	9-octadecenamide	3.1	2827

^a Retention time in minutes in increasing order of elution of compounds; ^b According to GC [Figure 5]; ^c m/e values of base peak 1st stated, and other prominent ions; ^d see identification of components; ^e Estimated total ion concentration in %; ^f Retention Index determined with reference to homologous series of n-alkanes.

Table 7: Stem essential oil composition of *Aspillia africana*

Retention time [mins] ^a	Peak No ^b	MS [Base peak+most abundant peaks] ^c	Identified compound ^d	%TIC ^e	Calculated RI ^f
16.9	9	57,43,71,41,85,56,55,	3,7-dimethylnonane	2.5	1521
22.3	7	57,71,43,85,41,55,56,99,	n-dodecane	9.1	1976
25.1	6	57,71,43,85,41,99,55,	2,7,10-trimethyldodecane	8.4	2373
26.2	1	69,43,109,93,41,119,95,67,	(+) D-nerolidol	17.5	2400
27.0	8	57,71,43,85,41,99.56,55,	2,6,11-trimethyldodecane	3.5	2419
28.2	3	73,60,43,55,41,57,256,	n-hexadecanoic	9.1	2762
29.3	4	71,57,43,55,68,69,41,81,95,	trans-phytol	6.3	2789
29.7	5	59,57,72,71,85,41,99,55,	nonadecanamide	6.3	2799
30.8	2	59,72,55,41,69,86,98,112,	9-octadecenamide	13.0	2827

^a Retention time in minutes in increasing order of elution of compounds; ^b According to GC [Figure 6]; ^c [m/e] values of base peak 1st stated, and other prominent ions; ^d see identification of components; ^e Estimated total ion concentration in %; ^f Retention Index determined with reference to homologous series of n-alkanes.

NARICT, ZARIA GCMS ANALYSIS

DR FARUQ (SAMPLE M-AaWL)

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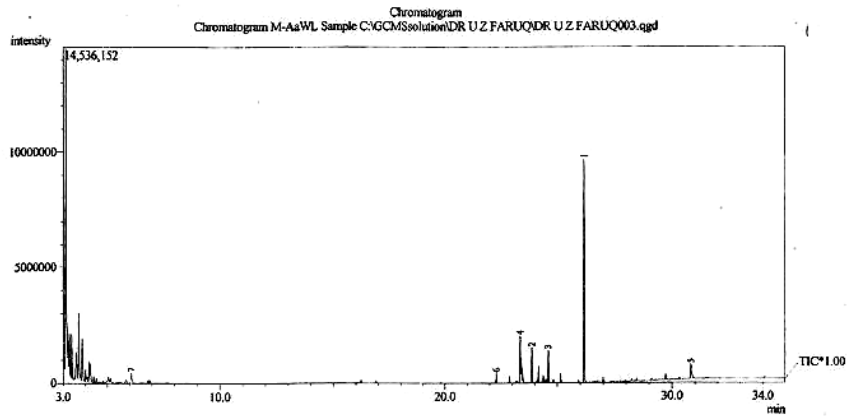


Fig. 5: Gas chromatogram of the leaf essential oil of *Aspillia africana* C.D. Adams.

NARICT, ZARIA GCMS ANALYSIS

DR FARUQ (SAMPLE M-AaWS)

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SHIMADZU, JAPAN

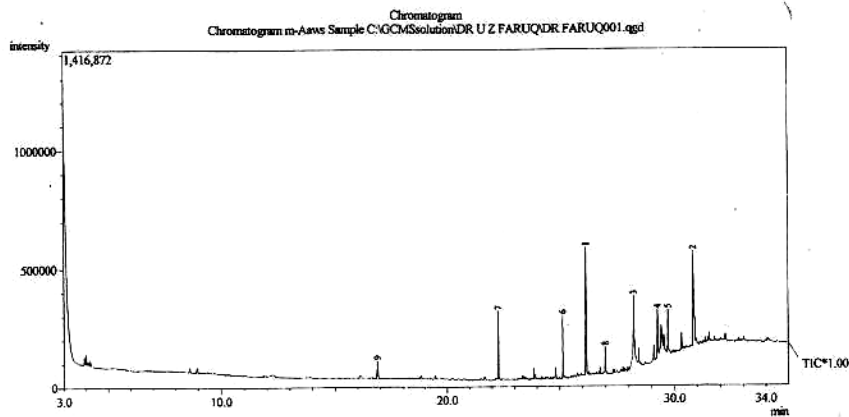


Fig. 6: Gas chromatogram of the stem essential oil of *Aspillia africana* C.D. Adams.

Chromolaena odorata is the least preferred plant of the four which rabbits fed on. Thirteen compounds each were identified in its leaf (57.3%) (Fig. 7) and stem (72.3%) (Fig. 8) oils. Caryophyllene oxide (11.4%) is the dominant compound, while terpenes account for 45.4% in leaf oil. Other groups of compounds present in leaf oil are hydrocarbons (6.7%), acid (2.5%) and alcohol (2.7%) (Table 8). Our result is in

agreement with earlier report that leaf volatile oil is dominated by terpenoids [20, 21].

The stem oil is characterized with the dominance of terpenoids (47.3%), though the most abundant compound is 3Z-3-heptadecen-5-yne (13.6%). Alkynes are 17.4% and ester is 7.6% of stem oil. There is presence of cis- and trans- p-menthan-8-ol (isomers) (Table 9). The root volatile oil has been reported earlier [25], but this is the first report of the stem oil compounds.

Table 8: Chemical Components of *Chromolaena odorata* Leaf Essential Oil

Retention time [mins] ^a	Peak No ^b	MS [Base peak+most abundant peaks] ^c	Identified compound ^d	%TIC ^e	Calculated RI ^f
8.2	12	93,92,91,77,79,67,80,94,105,	α -Pinene	1.7	739
14.0	11	57,41,43,56,44,55,70,69,98,	n-undecane	1.4	1142
15.2	4	79,94,77,91,93,80,41,53,65,78, ,95,105,106,120,	3,4-diethenyl-3-methylcyclohexene	4.2	1481
22.0	9	105,119,93,91,81,161,92,41,5 5,	α -copaene	2.7	1969
22.9	3	93,69,41,91,79,105,81,107,67, 55,133,77,106,120,	Z- α -bergamotene	8.3	1990
23.5	10	93,80,92,91,41,67,79,107,55,	α -humulene	4.0	2334
23.9	2	105,91,161,81,79,119,93,41,7 7,55,67,204	γ -muurolene	9.4	2344
25.3	1	79,93,69,43,41,95,109,91,96,6 7,55,81,106,107,	caryophyllene oxide	11.4	2378
26.3	8	109,91,79,41,81,43,93,67,	widdrol	3.4	2402
27.4	5	58,43,71,59,57,85,95,55,69,10 9,41,	hexahydrofarnesyl acetone	4.5	2429
28.3	6	73,60,57,55,41,71,69,85,	n-pentadecanoic acid	2.5	2764
29.3	7	71,57,81,69,55,95,43,	trans-phytol	2.7	2789
30.3	13	57,71,85,41,99,41,113,	n-heneicosane	1.1	2814

^a Retention time in minutes in increasing order of elution of compounds; ^b According to GC [Figure 7]; ^c [m/e] values of base peak 1st stated, and other prominent ions; ^d see identification of components; ^e Estimated total ion concentration in %; ^f Retention Index determined with reference to homologous series of n-alkanes.

Table 9: Chemical Composition of *Chromolaena odorata* Stem Essential Oil

Retention time [mins] ^a	Peak No ^b	MS [Base peak+most abundant peaks] ^c	Identified compound ^d	%TIC ^e	Calculated RI ^f
13.9	6	73, 45,59,154,87,108	Cis-linalool oxide	5.3	1140
18.3	7	109,79, 93,45,113,60	Cis-verbenol	3.0	1553
20.1	5	73,45,59,67,117,133,147	Tetrahydrolinalool	6.8	1925
23.8	3	73,45,59,87,117,147,131	Isopentyl isovalerate	7.6	2342
23.9	4	105,91,81,79,93,41,161,119,7,55,67,	germacrene D	5.7	2344
25.2	1	79,93,43,91,41,69,95,67,109,	3Z-3-heptadecen-5-yne	13.6	2376
25.5	9	67,96,43,109,41,55,81,93,	3-octadecyne	3.8	2383
25.7	8	73,45,59,87,117,131,147	Cis-p-menthan-8-ol	6.1	2388`
27.1	10	73,45,59,87,117,131,147	Trans-p-menthan-8-ol	3.4	2448
27.7	2	59,45,156,51,73,91,107	Hydroxyl citronellal	8.0	2749
28.2	13	65,94,147,221	Elemol	3.0	2762
28.6	12	197,135,73,43,59,91,107,149	Guaiol	3.0	2772
30.9	11	73,59,41,55,133,147	α -eudesmol	3.0	2830

^a Retention time in minutes in increasing order of elution of compounds; ^bAccording to GC [Figure 8]; ^c[m/e] values of base peak 1st stated, and other prominent ions; ^dsee identification of components; ^eEstimated total ion concentration in %; ^fRetention Index determined with reference to homologous series of n-alkanes.

NARICT, ZARIA GCMS ANALYSIS

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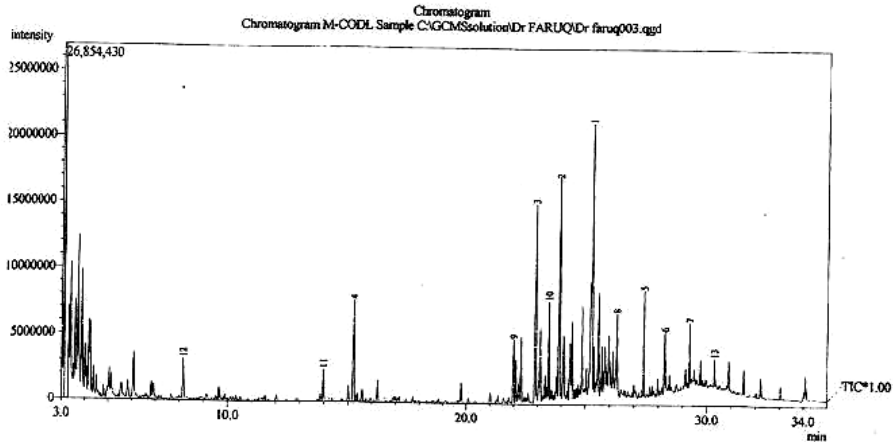


Fig. 7: Gas chromatogram of the leaf essential oil of *Chromolaena odorata* (L.) King and Robinson.

NARICT, ZARIA GCMS ANALYSIS

DR FARUQ (SAMPLE M-CoDS)

GCMS-QP2010 PLUS
SHIMADZU, JAPAN

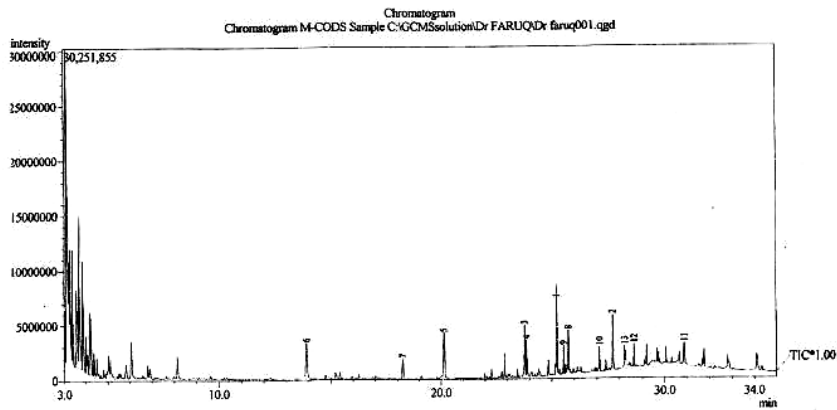


Fig. 8: Gas chromatogram of the stem essential oil of *Chromolaena odorata* (L.) King and Robinson.

There are differences in chemical composition and classes of compounds present in the eight essential oils. The leaf and stem oils of each plant also differ in their compositions, indicating differences in chemo-type. These differences are not unusual with respect to composition of plant essential oils [48, 49], but may not be unconnected with the observed differential preferences when administered as feed materials to rabbits.

Conclusion

Rabbits clearly showed their preferences when fed with *Aspillia africana*; *Chromolaena odorata* and *Syndrella nodiflora* (Compositae), and *Hyptis suaveolens* (Labiatae), which are utilized as alternate rabbit feeds. The most preferred plant is *Syndrella nodiflora* while, the least preferred is *Chromolaena odorata*. This may be due to differences in the chemical compositions of each plant, including their levels of nutrients, palatability and odour. The preferred plants contain high amounts of terpenes and their oxygenated derivatives, which are usually responsible for acceptable odours in essential oil. All isolated essential oils are highly flavoured with different characteristic odour which might have caused the preference. Our results fully agree with compositions of the oils that have been reported in literature. This study presents compositions of stem essential oils of *Syndrella nodiflora*, *Hyptis suaveolens* and *Chromolaena odorata* for the first time.

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