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Research Article

Chemical composition of essential oil of *Tithonia diversifolia* (Hemsl.) A. Gray from the Southern slopes of Mount Elgon in Western Kenya

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Abstract

Ethnopharmacologically, *Tithonia diversifolia* has a lot of applications in the history of human life. The current study aimed at characterizing the essential oil from fresh aerial parts of *T. diversifolia*. The plant materials were obtained from western Kenya and oil extracted by hydrodistillation and analyzed by gas chromatography (GC) and gas chromatography-mass spectrometry (GC-MS). Compounds were identified by comparison of their mass spectra with those in Wiley NBS and NIST databases and GC retention times to those of authentic samples. The percentage yield of the essential oil of *T. diversifolia* was 0.00015% w/w. The oil showed a complex composition of about 50 compounds, a mixture of monoterpenes and sesquiterpenes, 54% and 46%, respectively. Within the sub-classes of terpenes, hydrogen-carbon-containing terpenes (72%) occurred more than oxygen-containing terpenes (28%) with hydrogen-carbon-containing sesquiterpenes (38%) occurring in the highest proportion, followed by hydrogen-carbon-containing monoterpenes (34%), oxygenated monoterpenes (20%) and oxygenated sesquiterpenes (8%). Twenty-four percent of the 50 compounds and most of the monoterpenes were found in literature to have some repellence properties. Of these 50 compounds, α-pinene occurred in the largest amount (63.64%), followed by β-pinene (15.0%), *iso*-caryophyllene (7.62%), nerolidol (3.70%), 1-tridecanol (1.75%), limonene (1.52%), sabinene (1.00%), α-copaene (0.95%), α-gurjunene (0.56%) and cyclodecene (0.54%). With multipotential applications nature of *T. diversifolia* plant and its products, these compounds may in future be useful in pharmaceutical, agricultural, food and perfumery industries.

Key words: *Tithonia diversifolia*; Asteraceae; essential oil constituents; monoterpenes; sesquiterpenes; Western Kenya

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INTRODUCTION

1.1 Origin and morphology of *Tithonia diversifolia* (Hemsley) A. Gray

Tithonia diversifolia is a shrub and a member of the sunflower family, Asteraceae [1, 2]. *Tithonia* was named for '*Tithonus*', a legendary Trojan loved by the dawn goddess, Eos, who turned him into a grasshopper. Whereas the specific name '*diversifolia*' means 'separated leaves', from the Latin '*diversus*' (divergent) and '*folium*' (leaf) [1, 3]. The plant is commonly known by various names such as the tree marigold, Mexican arnica, Mexican tournesol, Mexican sunflower, Japanese sunflower and Nitobe chrysanthemum [1, 3]. *Tithonia diversifolia* is native to Eastern Mexico and Central America and is cultivated for its beautiful flowers and enormous size [1, 3]. The genus occurs throughout Middle America and the West Indies and has become naturalized around the tropics as an introduced species [2]. Depending on the geographical location of the plant, it may be either annual or perennial, 1.2 - 3 m in height with upright and sometimes ligneous stalks in the form of woody shrubs (Figure 1) [3]. The plant is mostly leafless on its lower parts[1]. The leaf arrangement is helically alternate. The typical leaf is fifteen to thirty centimeters long and has a minute roughness (scabridity) on the upper surface. The whitish petiole (acuminate) is fringed halfway with blade tissue at the terminus of which three whitish midveins vascularize a mitten-like blade with three or five prominent lobes [1]. Occasionally, new shoots possess unlobed leaves.

The plant's flowers are a favorite of bees and are formed by an orange-yellow inflorescence composed of many small flowers crowded together. Around the perimeter, 11-13 ray florets (ligulate flowers) frame 200-300 tubular disk florets, which smell like a daisy (Figure 1). After pollination, the inferior ovary of each disk floret develops as a hairy, gray, flattened, dry, one-seeded fruit (an achene) hidden by papery, brown-tipped bracts that, at maturity, are arranged into a hemispherical mound [1].

1.2 Ethnobotanic use of *Tithonia diversifolia*

By etymological description, the Kenyan common ethnic names include among others, *maruru*, *maua* and *amalulu* (for Luhya tribe), *maua makech* (for Luo tribe) and *amaua amaroro* (for Kisii tribe), all implying that the plant is bitter to the taste [3]. The African farmers have many uses for the plant, the most popular use being as an organic fertilizer for vegetable crops in either compost or a tea form [2]. Traditionally, Kenyans use it for ornamentals, livestock feeds, wind breaks, environmental conservation (for both soil and water conservation), honey production, curing of fire-cured tobacco, fuel wood (dry stumps), live fences, boundary demarcation and as a medicine in the form of leaves' infusion for constipation, stomach pains, indigestion, sore throat and liver pains and diarrhoea in livestock[3].

Adoyo and co-workers [4] reported that farmers working with the Kenya Woodfuel and Agroforestry Programme (KWAP) identified *T. diversifolia* as a potential insecticide to control termite infestations in farms and homes. One farmer's experiment with tea from either fresh leaves or the ash of *T. diversifolia*, *Cassia siamea* and *C. spectabilis* applied to affected trees, provided protection from termites for up to 45 days. Another farmer, who had a problem with underground termites, made a solution based on fermented extracts of *T. diversifolia* and *Melia azedarach*, which controlled the pests when the concoction was made and poured into the termite mound. After two years of research, farm results showed the most effective treatment to be a solution made from *T. diversifolia*, *Vernonia amygdalina* and *Agave sisalana* [4]. Not only did this solution control termites, but it also contributed to soil fertility [5, 6]. By using such local resources, one avoids the need to purchase the hazardous synthetic chemical pesticides. Moreover, farmers were reported as being enthusiastic about the use of *Tithonia* green manure and its effectiveness [4].

In Nigeria, there are oral reports among herbal medicine practitioners linking *T. diversifolia* with the treatment of menstrual pain [7]. In addition, *T. diversifolia* extracts are used in the treatment of wounds [8] and diabetes mellitus [9]. And recently, in western and central Kenya, *T. diversifolia* has been discovered as a very important organic fertilizer [10, 11], enhancing the availability of Phosphorus to crops, which led to its recommendation for biomass transfer technologies in Kenya [12, 23].

In Mexico, the place of origin of the plant, it is used to treat sprains, bone fractures, bruises and contusions. People in Mexico grow the plant in their gardens and use it for relieving dermatological problems [14]. In the Lowland Mixe, it is used orally to treat malaria and other forms of fever and topically to treat hematomas and muscular cramps [15, 16, 17]. It is also used as a liniment in Yucatan [18]. These medicinal uses may result from the similarity of the flower heads of this species to the ones of European arnica (*Arnica montana* L.). This assumption is corroborated by its popular names: *Arnica de la montana* and *arnica*. Berlin and Berlin [19] listed *T. diversifolia* as an important remedy for gastrointestinal complaints and it is cited as an anti-inflammatory and as treatment for wounds and skin eruptions. In southern China, *T. diversifolia* is used to treat skin diseases (such as athlete's foot), night sweats, as a diuretic, hepatitis, jaundice and cystitis. In Taiwan, the plant is sold in herbal medicine markets as an infusion to improve liver function while in Thailand and Japan, it is highly regarded as an ornamental plant [7].

1.3 Ecology of *Tithonia diversifolia*

In Kenya, *T. diversifolia* is found growing in Western and Central Provinces as well as coastal regions and parts of the Rift Valley, between latitude: 1° 0' N and longitude: 38° 0' E. It is a bushy perennial weed and a valuable green manure [6, 20]. It was introduced in Kenya from Central America as an ornamental and escaped from cultivation and now grows as a wild plant on the fields, in hedges, along roadsides and on wasteland/disturbed areas, not only in Kenya, but also elsewhere in the world [21, 22, 23]. Although sometimes cultivated [24], *T. diversifolia*, is now a pantropically distributed weed.

1.4 Chemotypes of *Tithonia diversifolia*

The chemical composition of essential oils from *T. diversifolia* has been previously described [25, 26, 27, 28]. For instance, Moronkola and co-workers [28] found leaf oil to comprise of an abundance of α-pinene (32.9%), β-caryophyllene (20.8%), germacrene D (12.6%), β-pinene (10.9%) and 1, 8-cineole (9.1%). Germacrene D (20.3%), β-caryophyllene (20.1%) and bicyclogermacrene (8.0%) characterized the oil of the flower while a number of aliphatic fatty acids and a diterpenoid compound, sandaracopimaradiene, that were present in the flower, could not be detected in the leaf oil [28].

1.5 The biological properties of the essential oil from *Tithonia diversifolia*

The validation of some of the folkloric claims have shown that *T. diversifolia* contains compounds that have a wide range of bioactive properties, namely, cytotoxic [29, 30, 31], anti-malarial [32, 33], anti-inflammatory[7, 8, 30] , potential cancer chemopreventive[8, 34, 35] , anti-amoebic [36], antiviral activity against human immunodeficiency virus type-1 [37], antifeedant activity[38], anti-diarrhoeal [39], anti-amoebic and spasmolytic activities [36, 40], and analgesic properties[7]. Tagitinin C, an anti-plasmodial sesquiterpene lactone, has been isolated from the aerial parts of *T. diversifolia* for development [41, 42]. The (-)-Germacrene D, a chemical constituent of the essential oil of *T. diversifolia*, increases attraction and oviposition by the tobacco budworm moth *Heliothis* virescens [43]. Compounds isolated from the aerial parts of *T. diversifolia* showed cytotoxic activity against HL-60 leukemia cells with IC_{50} values ranging from 0.13 to 13.0 μM[31]. *Tithonia diversifolia* has also been reported as a potential candidate for bioinsecticide preparations against *Callosobruchus maculatus* (Coleoptera: Bruchidae) [44].

From the foregoing evaluation therefore, the *T. diversifolia* plant and its products have a lot of multipotential applications in human life at various levels. Here in Kenya, there are no reports on the chemical components of the essential oil extracted from the aerial parts of *T. diversifolia*. We therefore report the characterization of the constituents of the essential oil of *T. diversifolia* with a view to exploiting it for future applications in pharmaceutical, agrochemical, food and perfumery industries.

MATERIALS AND METHODS

2.1 Study area and plant materials

Plant materials of the *Tithonia diversifolia* (Figure 1) were collected from Bungoma County, Western Kenya along the Southern slopes and foothills of Mount Elgon at altitudes ranging from about 1 300 m (thermal zone 3: 20.0-22.5 ^{o}C) in the south to about 3 500 m (thermal zone 8: 5.0-10.0 ^{o}C) in the north. The County is located between latitude 0^{o} 25'S and 0^{o} 53'N and longitude 34^{o} 21'W and 35^{o} 04'E. All aerial parts of the plants were collected. The plants were identified in the herbarium at the School of Biological Sciences, University of Nairobi, Kenya. A Voucher specimen of the plant species was deposited at the University of Nairobi Herbarium: *Tithonia diversifolia* (Hemsley) A. Gray (015-BGM-Muf/2015).

2.2 Extractions of essential oils

The collected plant materials were left in a well-ventilated room for 1-2 weeks before extraction of essential oil by hydrodistillation method. The materials were cut into small pieces and about 1 kg hydrodistilled using a Clavenger-type apparatus for 8 h [45]. After the distillation process was complete, the volatile essential oils were removed from the top of the hydrosol and dried over anhydrous sodium sulphate (Na_2SO_4). The oils were then filtered using Whatman filter paper (No. 1) and collected into 2 mL airtight glass vials and stored at –20 ^{o}C in a freezer until when required for chemical analysis.

1.3 Determination of the chemical composition of the essential oil

Both qualitative and quantitative characteristics of the essential oil were studied using gas-chromatography (GC) and gas-chromatography/Mass Spectrometry (GC-MS) techniques [46]. The constituents of the essential oils were identified by analysis of their mass spectra, direct comparison of their mass spectra to the Wiley NBS and NIST databases or library of mass spectra and co-injection with authentic standards on the GC.

The GC analyses were performed with a Hewlett Packard HP 5890A Gas Chromatography equipped with a flame ionization detector (at 230 °C). A fused silica capillary column (Hewlett Packard, 50 m x 0.22 mm x 0.33 mm CD) coated with methyl silicon (0.3 µm film thickness) was used with nitrogen as the carrier gas. All GC analyses were performed in the splitless mode with the injector temperature at 270 °C. The oven temperature was programmed from 60 °C isothermal for 7 min, to 120 °C at 5 °C per min, then to 180 °C at 10 °C per min and finally to 220 °C at 20 °C per min, where it was maintained for l0 min. Peak areas were calculated using a Hewlett Packard 3393 B series integrator and together with their GC retention times, compared to those of authentic samples.

The GC-MS analyses were performed with a VG Masslab 12-250 quadruple gas chromatography-mass spectrometer. Chromatographic separations were achieved using a fused silica capillary column (Hewlett Packard, 50 m x 0.32 mm ID) coated with Carbowax 20M (0.3 µm film thickness) with helium as the carrier gas. All the GC-MS analyses were made in the splitless mode with helium as the carrier gas. The GC column was temperature programmed as in the case of GC analysis. Compounds were identified by their electron impact (El) mass spectral data, order of elution and relative GC retention times, and by comparison of their mass spectra and GC retention times to those of authentic samples (where possible).

The computer on the GC-MS system records a mass spectrum for each scan and has a library of spectra that can be used to identify an unknown chemical in the sample. The library compares the mass spectrum from a sample component with mass spectra in the library. It then reports a list of likely identifications along with the statistical probability of the match. The Synthetic standard chemicals (authentic samples) used in GC co-injections were obtained from Sigma Chemical Company, Poole, UK and Aldrich Chemical Company, Gillingham, UK. All the authentic samples used were over 95% pure.

RESULTS AND DISCUSSION

3.1 Yield and properties of the essential oil of *Tithonia diversifolia*

The yield of the essential oil from fresh aerial parts of *T. diversifolia* was 0.00015% w/w. This percentage yield was lower than previously reported (0.019% w/w) [28]. The essential oil was observed to be pale yellow and soluble in dichloromethane (DCM), ether, ethanol and dimethyl sulfoxide (DMSO) and insoluble in water. The oil was liquid at room temperature and maintained this state even on storage at - 20 °C.

3.2 Chemical composition of the essential oil of *Tithonia diversifolia:*

The analysis of the essential oils of aerial parts of *Tithonia diversifolia* identified 50 chemical constituents occurring in different proportions and, some, reported to have insecticidal, acaricidal, pesticidal and/or repellant properties in literature (Table 1). In this essential oil, monoterpenes (54%) were more than sesquiterpenes (46%), whereas within the sub-classes of terpenes, hydrogen-carbon-containing terpenes (72%) were more than oxygen-containing terpenes (28%) with hydrogen-carbon-containing sesquiterpenes (38%) occurring in the highest proportion, followed by hydrogen-carbon-containing monoterpenes (34%), oxygenated monoterpenes (20%) and oxygenated sesquiterpenes (8%) in that order (Table 2). Twenty-four percent (12 compounds) of the 50 compounds and most of the monoterpenes were found in the literature to have some repellence properties. Of these 50 compounds, α-pinene occurred in the largest amount (63.64%), followed by β-pinene (15.0%), *iso*-caryophyllene (7.62%), nerolidol (3.70%), 1-tridecanol (1.75%), limonene (1.52%), sabinene (1.00%), α-copaene (0.95%), α-gurjunene (0.56%) and cyclodecene (0.54%) in that order (Table1). This composition compares favourably with the previous analysis made by Moronkola and coworkers in Nigeria[28] in which the essential oil from the leaf comprised an abundance of α-pinene (32.9%), β-caryophyllene (20.8%), germacrene D (12.6%), β-pinene (10.9%) and 1, 8-cineole (9.1%) while that from the flower comprised an abundance of Germacrene D (20.3%), β-caryophyllene (20.1%) and bicyclogermacrene (8.0%), respectively. In view of the biological properties of some of the essential oil constituents from literature shown in Table 1, the chemical profile represents a group of useful compounds with potential application in agriculture, medicine, pharmacy, nutrition, cosmetics etc. A number of monoterpenes have been previously reported to be repellent against insects such as *Phoebis sennae amphitrite* Feisthamel, *Pieris brassicae* L., *Tatochila autodice blanchardi* Butler, *T. mercedis mercedis* Eschscholtz, *Battus polydamas archidamas* Boisduval, *Cosmosatyrus chilensis chiliensis* Guérin, *Vanessa carye* Hübner, *Helephila venusta* Hayward, *Culex pipiens pallens* Rank and *Castnia psittachus* Molina [47, 48] as well as host-seeking nymphs of *Ixodes ricinus* [49, 50, 51]. Therefore, the repellent activity of the essential oil of *T. diversifolia* [33], its insect feeding deterrent activities and its reputed anti-leishmanial activity [52], and insecticidal properties [53], make it an attractive candidate for both laboratory and field evaluation as a pest control and management agent as shown by biological properties of some of the essential oil constituents from literature in Table 1.

Figure 1: *Tithonia diversifolia* (Hemsley) A. Gray plant showing the aerial parts used in the extraction of the essential oil for chemical composition evaluation.

Table 1: GC and GC-MS identified major constituents in the essential oil of *Tithonia diversifolia* plant from Bungoma County, western Kenya.

Peak No.	Name of the compound identified in the essential oil	Biological property from literature	Molecular formula	RT	M^+ g/mol	Base peak	Major peaks	Relative % abundance	GC-MS co
1	α-phellandrene	R	$C_{10}H_{16}$	18.400	136.24	93	39,77,136	0.045	
2	α-pinene	R^{PmAg} ,I,P,R	$C_{10}H_{16}$	18.975	136.24	93	39, 77, 79, 121, 136	63.64	✓
3	Camphene	R^{PmAg}	$C_{10}H_{16}$	19.350	136.24	93	39, 67, 79, 107, 121,	0.17	
4	Sabinene	I(Jaenson et al, 2005)	$C_{10}H_{16}$	20.150	136.24	93	39, 41, 77, 79, 136	1.00	✓
5	β-pinene	$R^{PmAg\ P}$,R,I (Jaenson et al, 2005)	$C_{10}H_{16}$	20.425	136.24	93	41, 121, 136	15.0	✓
6	β-myrcene	R^{PmAg},P,R, I	$C_{10}H_{16}$	20.650	136.24	69	41, 69, 136	0.02	✓
7	Linalyl acetate		$C_{12}H_2O_2$	21.250	196.29	93	53, 69, 79, 121,	0.02	
8	Terpinolene	R^{PmAg}	$C_{10}H_{16}$	21.725	136.24	93	39, 77, 105, 121, 136	0.08	✓
9	Paracymene	R^{tRa}	$C_{10}H_{16}$	21.850	136.24	119	39, 77, 91, 134	0.03	
10	Limonene	RtRa A,I,P,R	$C_{10}H_{16}$	22.200	136.24	68	39, 41, 53, 67, 93	1.52	✓
11	Carene		$C_{10}H_{16}$	23.200	136.24	93	39, 77, 121, 136	0.17	✓
12	*cis*-sabinenihydrate	R^{PmAg}	$C_{10}H_{18}O$	23.750	154.25	93	43, 71, 111, 121, 136	0.04	
13	α-Terpinene	R^{PmAg}	$C_{10}H_{16}$	24.326	136.24	93	77, 105, 121, 136	0.05	✓
14	Linalool	RtRa R^{PmAg}	$C_{10}H_{18}O$	24.601	154.25	71	41, 43, 55, 80, 93, 121	0.12	✓
15	(E)-4,8-Dimethyl-1,3,7-Nonatriene		$C_{10}H_{16}$	25.126	136.24	69	41,81, 107, 150	0.12	
16	4-Terpineol		$C_{10}H_{18}O$	27.426	154.25	71	43, 55, 86, 93, 111	0.19	
17	Linalyl propanoate		$C_{13}H_{22}O_2$	27.926	210.00	59	43, 68, 81, 93, 121, 136	0.07	
18	β-Angelicalactone			28.351		55	43, 70, 98	0.03	
19	2-undecene,6-methyl-.(z)		$C_{12}H_{24}$	31.101	168.00	55	29,,43,57,69,83,97,112	0.04	

20	Decane,2,6,8-Trimethyl		$C_{13}H_{28}$	31.401	184.00	57	41, 43, 71, 85, 99	0.15	
21	Megastigmatrienone		$C_{13}H_{18}O$	31.976	190.00	190	41,91,133,148, 175	0.01	
22	1,3,6-Heptatriene,2,5,6, Trimethyl		$C_{10}H_{16}$	32.501	136.24	121	41,71,93,107,136	0.0082	
23	Bi cycloelemene		$C_{15}H_{24}$	33.776	204.36	121	41, 93, 107, 136, 161	0.11	
24	α-Cubebene		$C_{15}H_{24}$	33.201	204.36	105	41,55,81,91,119,161	0.009	
25	α-Guaiene		$C_{15}H_{24}$	33.326	204.36	91	41,,81,105,133,147,189	0.08	
26	Damascenone C		$C_{13}H_{18}O$	33.651	190.00	69	41, 84, 105, 121	0.006	
27	γ-1-cadinene		$C_{15}H_{24}$	33.876	204.36	93	55,77,79,105,119,161	0.004	
28	α-Copaene		$C_{15}H_{24}$	34.101	204.36	105	41, 77, 81, 93, 119, 161	0.95	
29	α-Gurjunene		$C_{15}H_{24}$	34.376	204.36	189	41,55,91,105,119,161,161,204	0.56	
30	Bicyclo 2.2.2.octa-2,5 diene, 1,2,3,6-tetramethyl		$C_{15}H_{24}$	34.551	204.36	162	41,91,105,119, 162	0.20	
31	*trans*-Caryophyllene		$C_{15}H_{24}$	34.976	204.36	93	55, 69, 105, 133	0.007	
32	δ-Guaiene		$C_{15}H_{24}$	35.201	204.36	108	41, 79, 81, 93, 148, 189	0.20	
34	*iso*-caryophyllene		$C_{15}H_{24}$	35.476	204.36	93	41, 69, 105, 133, 161	7.62	
35	Nerolidol isomer		$C_{15}H_{26}O$	35.776	222.37	69	41, 55, 93, 107	0.004	✓
36	α-Humulene		$C_{15}H_{24}$	36.326	204.36	93	41, 80, 121, 147	0.28	
37	β-Ionane		$C_{12}H_{12}O$	36.551	172.00	43	77, 91, 177	0.01	
38	1-Tridecanol		$C_{14}H_{13}O$	36.876	197.00	55	43, 69, 83, 97	1.75	✓
39	Germacrene D	T^{PRL}	$C_{15}H_{24}$	37.001	204.36	161	81, 91, 105, 119, 204	0.04	
40	Bycyclogermacrene		$C_{15}H_{24}$	37.401	204.36	93	41, 79, 121, 136, 161	0.24	
41	Cyclamen Aldehyde		$C_{13}H_{18}O$	37.651	180.00	133	91, 105, 147, 175, 190	0.02	
42	δ-Cadinene		$C_{15}H_{24}$	37.876	204.36	161	91, 105, 119, 204	0.09	
43	*trans*-Sabinene		$C_{10}H_{16}$	38.251	136.00	43	79, 93, 121	0.008	

44	Cholesta,3,5-iene		$C_{15}H_{24}$	38.376	204.36	157	43,57,81,93,181	0.004	
45	Nerolidol		$C_{15}H_{26}O$	38.651	222.37	69	41, 43, 93, 107, 161	3.70	✓
46	Caryphyllene oxide		$C_{15}H_{24}$	39.676	204.36	43	41,55,79,93	0.12	
47	Cyclodecene		$C_{12}H_{22}$	41.251	166.00	67	41, 54, 81, 95	0.54	
48	Juniper camphor		$C_{15}H_{26}O$	41.451	222.37	43	41, 81, 161, 189, 204	0.10	
49	Pentadecanone		$C_{15}H_{30}O$	45.426	226.00	43	58, 71, 85	0.46	
50	**Cycloundecanone**		$\mathbf{C_{11}H_{16}}$	**46.076**	**158.00**	**55**	**41, 58, 98, 111**	**0.06**	

KEY

RT – Retention Time in Minutes
GC–MS co – Gas Chromatography – Mass Spectrometry Co-injection
R^{pmAg} – Repellent to mosquito –*Anopheles gambiae* [57].
R^{Ag} – Repellent to *A. gambiae* [55].
R^{tRa} – Repellent to tick *Rhipicephalus appendiculatus* [58, 59].
R^{PIr} – Part of repellent essential oil to *lxodes ricinus* nymphs [60].
R^{PmAa} – Part of repellent essential oil to mosquitoes, *Aedes aegypti* [49].
R^{tRa} – Repellent against ticks – *R. appendiculatus* [58, 59].
A – Acaricidal
I – Insecticidal
R – Insect repellant (insectifuge)
P – Pesticidal
} According to Duke [61] or other source(s) as referred to in table 1 or other references as indicated by the authors.

R^{Tm} – Repellent to *Tetranychus* mites [62].
T^{PRL} – Part of *Ageratum houstonianum* essential oil toxic to *Rhipicephalus appendiculatus* [63, 64].
✓ - Gas Chromatography – Mass Spectrometry Co-injection was conducted.

Table 2: Percentage group composition and classification of terpene constituents in the essential oil from the aerial parts of *Tithonia diversifolia.*

Group composition of the essential oil		**Classification of terpene constituents**	
Type of terpene in the composition	Proportion of the terpene in the composition	Hydrogen-carbon terpenes only	Oxygen-containing terpenes
Monoterpenes	27 (54%)	17 (34%)	10 (20%)
Sesquiterpenes	23 (46%)	19 (38%)	4 (8%)
Total	50 (100%)	36 (72%)	14 (28%)

These findings further present very important phenomenon as in the recent past, the monoterpenes have been previously identified and found to offer an attractive alternative to chlorofluorocarbons in many industrial applications [54, 55]. A number of these monoterpenes (α-pinene, limonene, γ-terpinene, terpinolene, arbanol, α-terpineol, linalool, and plinol) (some also found in this study (Table 1), were evaluated of their properties to predict their most likely fate in the environment in case they are found suitable for industrial application. The existence of a wide array of these potentially useful natural products therefore shades more light on the likelihood application of the essential oil of *T. diversifolia* in the industrial development. Nevertheless, the use of this plant is ancient in the pharmacopoeia and this may be widespread in the plant's family, Asteraceae (Compositae) [56] .

CONCLUSION

In this study, the essential oils from aerial parts (leaf, stem and flowers) of *Tithonia diversifolia* was found to comprise of mainly terpenes; a mixture of monoterpenes and sesquiterpenes, 54% and 46%, respectively. The oil was predominated by α-pinene which accounted for 63.64%. Within the different sub-classes of terpenes, hydrogen-carbon-containing terpenes (72%) occurred more than oxygen-containing terpenes (28%) with hydrogen-carbon-containing sesquiterpenes (38%) occurring in the highest proportion, followed by hydrogen-carbon-containing monoterpenes (34%), oxygenated monoterpenes (20%) and oxygenated sesquiterpenes (8%). The work lays down substantial groundwork for even more comprehensive studies on the potential applicability of the compounds from *Tithonia diversifolia* essential oils in pharmaceutical, agricultural, food and perfumery industries.

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COMPLIANCE WITH ETHICS GUIDELINES

Conflict of interest

The authors declare that they have no conflicts of interest.

Human and animal rights, informed consent

This article does not contain any studies with human or animal subjects.

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