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Chemical characterization of *Tecoma stans* (L.) Juss. ex Kunth volatile oils

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Abstract
Gas chromatography (GC) and gas chromatography coupled with Mass spectrometry (GC-MS) analysis of the essential oils from leaf, stem, seed and flower of *Tecoma stans* (L.) Juss exKunth (family Bignoniaceae) obtained by hydrodistillation showed the presence of 64 chemical compounds. A total of 31 compounds in the leaf, 22 in the stem, 31 in the seed and 35 in the flower were identified representing 91.5%, 81.4%, 88.5% and 95.8% of the whole volatile oil respectively. The most abundant constituents were 1-octen-3-ol (24.8%) and 2,6,10-trimethylpentadecane (10.4%) in the leaf, linalool (11.4%) and 2,6,10-trimethylpentadecane (10.7%) in the stem, α-terpineol (11.7%) and β-selinene (9.3%) in the seed and (*E*)-β-ionone (11.2%) and linalool (10.8%) in the flower.

Keywords: *Tecoma stans*, bignoniaceae, GC and GC-MS

1. Introduction
Tecoma stans (L.) Juss ex Kunth (Bignoniaceae) also known as the yellow trumpet tree is an important ornamental medicinal plants used in the treatment of various diseases e.g diabetes, syphilis, stomach pain, vermifuge, diuretic, rheumatism and tonic in traditional medicine [1-3]. Much work has been done on the extract from different parts of the plant. Some of the work include phytochemical screening [4-6], antispasmodic [7], CNS depressant [8, 9], wound healing [10, 11], antimicrobial [12], antioxidant [13], cytotoxicity [14], antinociceptive and anti-inflammatory analysis [15]. Boschniakine, tecomine, 5β-hydroxyskitanthine, tecomamine, 4- noractinidine, 4-norskytanthine and tecostanine [1, 16] are some of the compounds isolated from the leaf of *Tecoma stans*. There is no work on the volatile component of the plant as at the time this research work commenced. Information on the medicinal, pharmacological and biological activities of essential oils has little value if the chemotype has not been discovered. Thus, the aim of this research work is to carry out qualitative and quantitative analyses on the essential oils hydrodistilled from different part of *Tecoma stans* growing in Nigeria, using gas chromatography and gas chromatography coupled with mass spectrometry.

2. Materials and methods
2.1 Preparation of Plant Materials
Tecoma stans leaf, stem, seed and flower were collected at Saunders road, University of Ibadan, Ibadan, Oyo state, Nigeria. The plant was identified at the herbarium in the Botany Department University of Ibadan. Identification was also done by experts at the Forest Research Institute of Nigeria (FRIN) where Voucher specimen was deposited in the Herbarium. The fresh plant materials were washed and air-dried for two weeks to remove dirt and moisture respectively. They were pulverized after drying to increase the surface area.

2.2 Isolation of Essential oil
Pulverized dried plant materials (500g each) of *Tecoma stans* (leaves, stem, seed) were hydrodistilled for 3h in an all-glass Clevenger apparatus, according to the British Pharmacopeia specification [17]. The receiver arm of the Clevenger contains water and a known amount of *n*-hexane which helps to retain and separate the essential oil from the water. The volatile oil obtained was dried over anhydrous sodium sulfate and stored in sealed glass vials in a refrigerator prior to analyses. Volatile oil was extracted from fresh flower sample.

2.3 Gas chromatography (GC)

Gas chromatographic analyses were carried out on a HP-5890 Gas chromatograph fitted with an HP-5 capillary column (30 m × 0.25 mm, 0.25 µm film thickness). The GC oven temperature was programmed at 60 ºC (held for 10 min), heated to 220 ºC at 5 ºC/min. The injector and detector temperatures were maintained at 250 ºC. Helium was used as carrier gas at a flow rate of 2 mL/min. The identification of components was done by comparing the retention time of the constituents with those of reference samples and by means of their linear retention indices (LRI) relative to the series hydrocarbons.

2.4 Gas chromatography/mass spectrometry (GC-MS)

GC-MS analyses were carried out on a Varian CP-3800 gas chromatograph equipped with a HP-5 capillary column (30 m x 0.25 mm; 0.25 µm coating thickness) interfaced to a Varian Saturn 2000 ion trap Mass Detector. The mass spectrometry was operated in EI mode with ionization voltage 70 eV. The injector and transfer line temperatures were 220 ºC and 240 ºC respectively. The GC oven temperature was programmed from 60 to 240 ºC at 3 ºC/min. Helium was used as a carrier gas at the flow rate of 1 mL/min. Identification of the constituents was based on comparing their retention indices relative to the series of n-hydrocarbons, and comparison of their mass spectra with literature [18, 19] and those of reference compounds from NIST 2012 library. The relative percentage of each constituent is calculated by integration of GC peak areas.

3. Results & Discussion

The yields of essential oils from *T. stans* leaf, stem, seed and flower are 0.79%, 0.46%, 0.52% and 0.59% respectively. From the GC and GC-MS analyses, the total numbers of compounds identified in each essential oil are 31, 22, 31 and 35 amounting to 91.5%, 81.4%, 88.5% and 95.8% of the whole oil in leaf, stem, seed and flower volatile oils respectively. The GC chromatographs of the oils were presented in Figures. 1-4. The leaf oil was dominated by 1-octen-3-ol (24.8%), 2,6,10-trimethylpentadecane (10.4%), pentadecanal (8.9%), linalool (7.7%), 3-octanol (6.0%) and citronellol (5.1%). The previous works from the literatures on the leaf essential oils from Bignoniaceae family have shown linalool and 1-octen -3-ol as their characteristics constituents [20, 21]. *Jacaranda acutifolia* essential oil and hexane extract contain methyl linolenate, 1- octen-3-ol, methyl pnenyl acetate, linalool and palmitic acid has major constituents. GC (FID) and GC/MS analysis of *Memora nodosa* leaf volatile oil gave benzaldehyde, 1- octen-3-ol, linalool and mandelonitrile as the most prominent components. Essential oil from air-dried leaves of *Mansoa alliacea* of Brazil origin showed the presence of sulfur compounds and 1-octen-3-ol [22]. Other notable representative compounds found in the *Tecoma stans* leaf oil are patchouli alcohol (4.3%), acorenone (3.8%) and (*E*)-β-ionone (2.7%). Two uncommon essential oil constituents were also identified only in the leaf oil, they are (*Z*) and (*E*)- rose oxide (0.7% and 0.8% respectively). *Z*-rose oxide (isobutenyl-4-methyl tetrahydropyran) is a perfumery ingredients [23]. Other chemical compounds present in the leaf oil alone are (*Z*)-3-octen-1-ol (0.5%), (*E*)-4-dodecene (0.9%), tetradecanal (0.5%), isocedranol (1.1%), 1-tetradecanol (0.7%) and patchouli alcohol. Monoterpene and sesquiterpene hydrocarbons are absent in the leaf essential oil. There are 14 non-terpene derivatives amounting to 58.1% of the leaf oil.

Of the twenty two components identified in the stem volatile oil linalool (11.4%) was the most abundant constituents. Two other major compounds are 2,6,10-trimethylpentadecane (10.7%) and 3- octanol (7.0%). Some important terpenes were also present in notable quantity like β-eudesmol (5.9%), β-selinene (5.4%) and α-terpineol (5.0%), (*E*)-nerolidol (3.7%), *epi*-α-bisabolol (2.4%), acorenone (2.4%) and spathulenol (2.0%). Among the non-terpene derivatives (40.1%), there were two aldehydes (3.8%), five aliphatic hydrocarbons (18.9%), two aliphatic alcohols (10.7%) and one aromatic hydrocarbon. The only aromatic compound was methyl salicylate (5.7%) while limonene (0.7%) which is one of the odor imparting constituents [24], was the also only monoterpene hydrocarbon. No apocarotenoid was detected in the essential oil. The compounds found only in the stem oil were spathulenol and β-eudesmol (oxygenated sesquiterpenes).

Monoterpenoid compounds (30.6%) were the dominating constituents in the seed essential oil. The major monoterpenoids were linalool (7.4%) and α-terpineol (11.7%). 1-octen-3-ol (5.5%) and 3-octanol (6.0%) were the only non-terpene aliphatic alcohols present in the seed oil. β-Selinene (9.3%) was the major component in the sesquiterpenoid class. There are also five aromatic compounds (10.1%) which are ethylbenzene (2.0%), *p*-xylene (3.2%), *o*-xylene (0.9%), 1-ethyl-4-methylbenzene (1.8%) and methylsalicylate (3.4%) known to be odor imparting constituents in the essential oil. Other classes of compounds identified were non-terpene aliphatic hydrocarbons (7.0%), aldehydes (7.8%) and one apocarotenoid, i.e., (*E*)-geranylacetone (1.4%). Constituents that were present only in the seed volatile oil include γ-terpinene (1.4%), terpinolene (0.8%), (*E*)-2-decenal (0.9%), β-elemene (1.5%), dodecanal (1.0%) and dendrolasin (1.8%).

On the other hand, the floral essential oil constitutes higher percentage of non-terpene derivatives (56.2%), low sesquiterpenoids (7.4%) and appreciable quantity of apocarotenoids (20.1%). There was no monoterpene hydrocarbon in the floral oil but two oxygenated monoterpenes amounting to 12.1% of the oil were detected. The flower volatile oil also has more chemical constituents than the other oil samples investigated in this work. The most prominent components were (*E*)-β-ionone (11.2%), linalool (10.8%), 2,6,10-trimethylpentadecane (8.4%), 1-octen-3-ol (7.9%), (*E*)-β-damascenone (4.1%) and *p*-xylene (4.0%). The terpenoids detected only in the flower oil were α-cardinol, germacrene-D-4-ol and (*E*, *E)*-α-farnesene. Higher percentage of apocarotenoids (product of oxidative cleavage of carotenoid double bonds) in the flower volatile oil agreed to the presence of β-carotene in the *T. stans* flower extract reported in the literature [1]. In contrast, there was much difference in the Bignoniaceae flower essential oils from the literature and the present work [25, 26].

Table 1: Chemical constituents of *Tecoma stans* volatile oils

Constituents [a,b]	l.r.i.	Percentage composition (%)			
		Leaf	Stem	Seed	Flower
Ethylbenzene	864	-	-	2.0	3.4
p-xylene	867	-	-	3.2	4.0
o-xylene	895	-	-	0.9	1.8
1-ethyl-4-methylbenzene	965	-	-	1.8	2.2
1-ethyl-3-methylbenzene	967	-	-	-	0.8
1-octen-3-ol	981	24.8	3.7	5.5	7.9
3-octanol	994	6.0	7.0	6.0	-
Mesitylene	996	-	-	-	2.7
n-decane	1000	-	-	-	1.1
Limonene	1032	-	0.7	2.1	-
(*Z*)-3-octen-1-ol	1051	0.5	-	-	-
4-methyldecane	1059	-	-	-	1.5
γ-terpinene	1063	-	-	1.4	-
Terpinolene	1090	-	-	0.8	-
6-camphenone	1092	0.5	-	1.0	-
Linalool	1101	7.7	11.4	7.4	10.8
Nonanal	1104	0.4	1.6	2.1	1.4
(*Z*)-rose oxide	1111	0.8	-	-	-
(*E*)-rose oxide	1128	0.7	-	-	-
Citronellal	1155	0.8	-	3.0	-
2,4-dimethylbenzaldehyde	1179	-	-	-	1.9
4-terpineol	1179	-	0.9	1.1	-
α-terpineol	1191	0.5	5.0	11.7	1.3
methyl salicylate	1192	2.2	5.7	3.4	-
(*E*)-4-dodecene	1193	0.9	-	-	-
n-dodecane	1200	1.1	2.1	1.2	1.5
Decanal	1206	0.4	2.2	2.6	-
β-cyclocitral	1222	0.6	-	-	1.6
Citronellol	1230	5.1	-	2.1	-
β-cyclohomocitral	1256	0.8	-	-	1.7
(*E*)-2-decenal	1263	-	-	0.9	-
3-methyldodecane	1273	-	-	-	1.0
(*E*,*E*)-2,4-decadienal	1316	-	-	1.2	1.0
3-methylundecanol	1326	-	-	-	1.0
(*E*)-β-damascenone	1382	0.9	-	-	4.1
β-elemene	1392	-	-	1.5	-
n-tetradecane	1400	-	1.2	1.6	1.6
Dodecanal	1409	-	-	1.0	-
(*E*)-α-bergamotene	1437	-	0.8	2.7	-
(*E*)-geranylacetone	1457	0.5	-	1.4	1.5
2-methyltetradecane	1462	-	-	-	1.0
β-selinene	1487	-	5.4	9.3	-
(*E*)-β-ionone	1487	2.7	-	-	11.2
n-pentadecane	1500	0.5	-	1.1	0.9
2-tridecanol	1506	-	-	-	2.3
(*E*,*E*)-α-farnesene	1508	-	-	-	2.2
δ-cadinene	1524	-	0.7	-	1.5
5-methylpentadecane	1551	-	0.8	-	0.9
(*E*)-nerolidol	1564	0.6	3.7	3.6	-
germacrene D-4-ol	1575	-	-	-	2.3
Spathulenol	1577	-	2.0	-	-
Dendrolasin	1580	-	-	1.8	-
n-hexadecane	1600	0.8	5.1	3.1	2.2
Tetradecanal	1614	0.5	-	-	-
2,6,10-trimethylpentadecane	1642	10.4	10.7	-	8.4
β-eudesmol	1650	-	5.9	-	-
α-cadinol	1654	-	-	-	1.4
patchouli alcohol	1660	4.3	-	-	-
Isocedranol	1668	1.1	-	-	-
1-tetradecanol	1675	0.7	-	-	-
epi-α-bisabolol	1687	2.0	2.4	-	-
Acorenone	1688	3.8	2.4	-	-
2-pentadecanone	1699	-	-	-	2.6
Pentadecanal	1716	8.9	-	-	3.1
Total identified		91.5	81.4	88.5	95.8

Monoterpene hydrocarbon		0.0	0.7	4.3	0.0
Oxygenated monoterpene		16.1	17.3	26.3	12.1
Sesquiterpene hydrocarbon		0.0	6.9	13.5	3.7
Oxygenated sesquiterpenes		11.8	16.4	5.4	3.7
Apocarotenoids		5.5	0.0	1.4	20.1
Other non terpenes		58.1	40.1	37.6	56.2

a- elution Order on HP-5MS column. Lri- Linear retention indices from analyses value

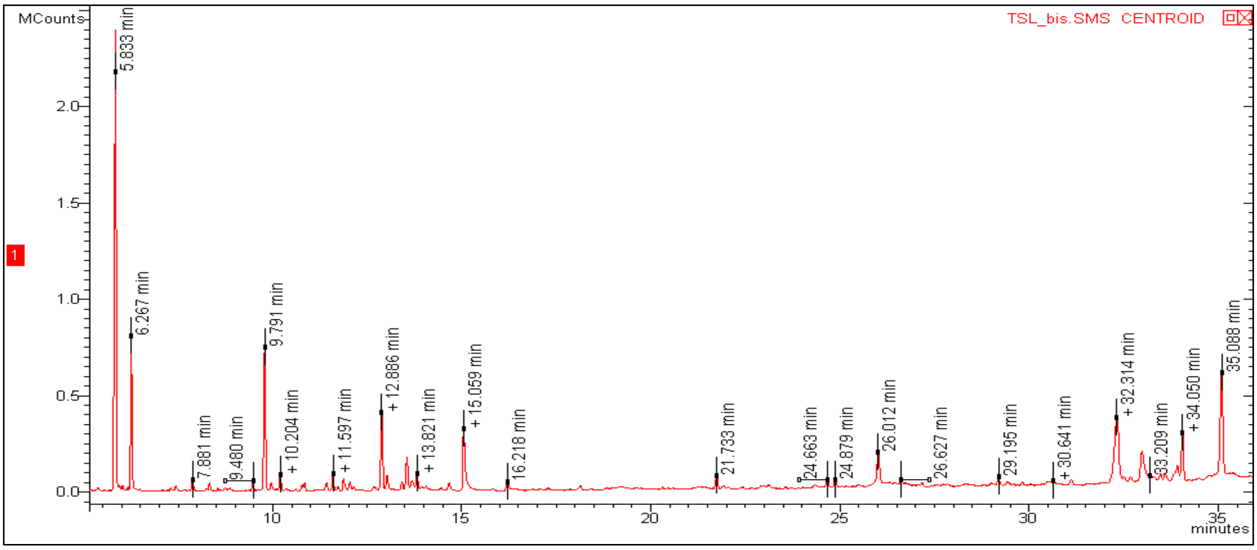

Fig 1: GC Chromatograph of Leaf Essential Oil

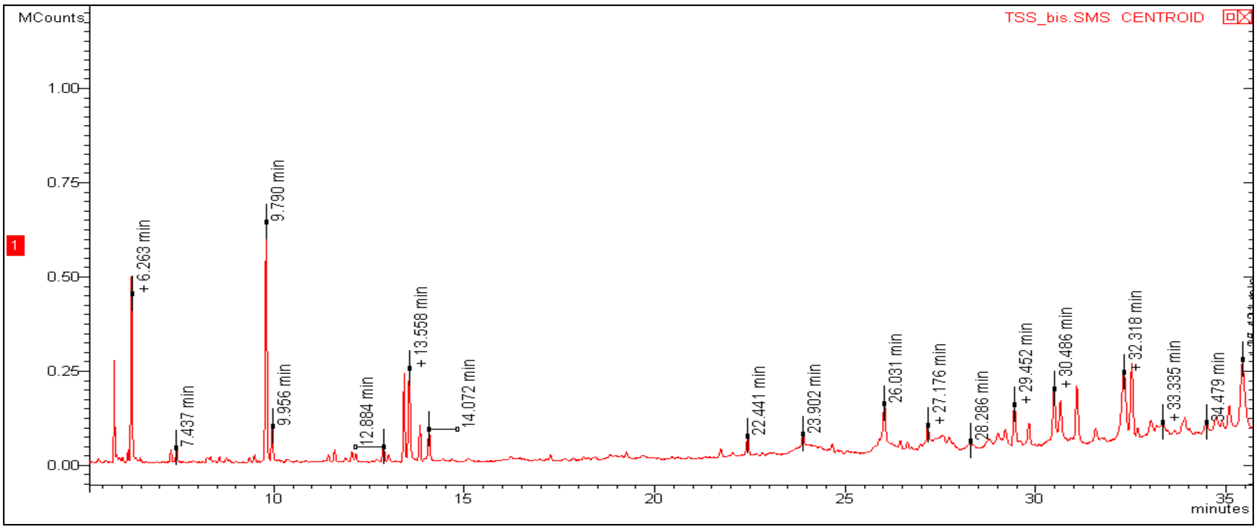

Fig 2: GC Chromatograph of Stem Essential Oils

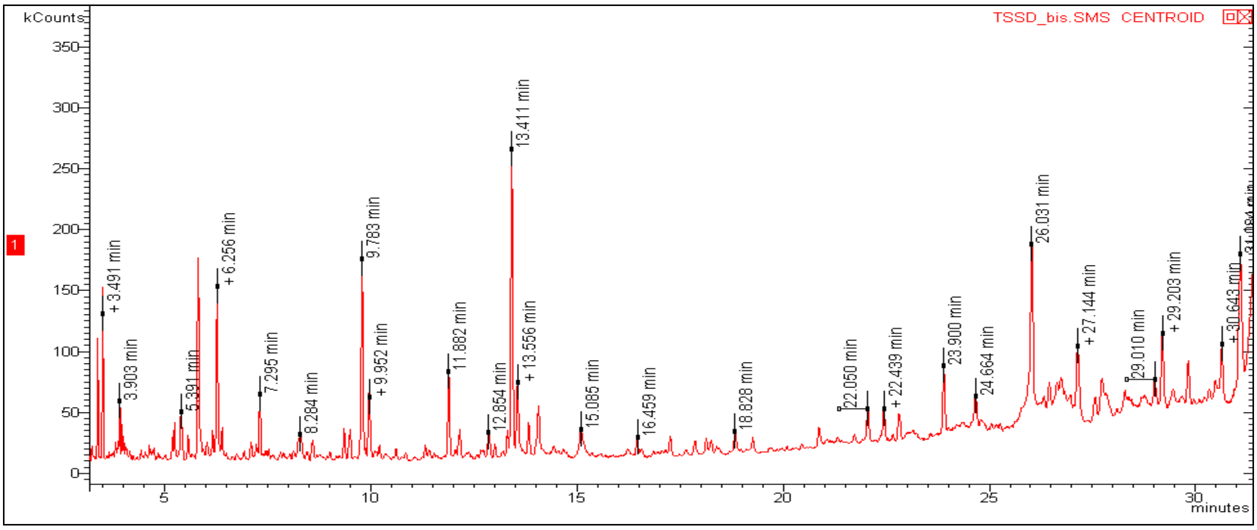

Fig 3: GC Chromatograph of Seed Essential Oils

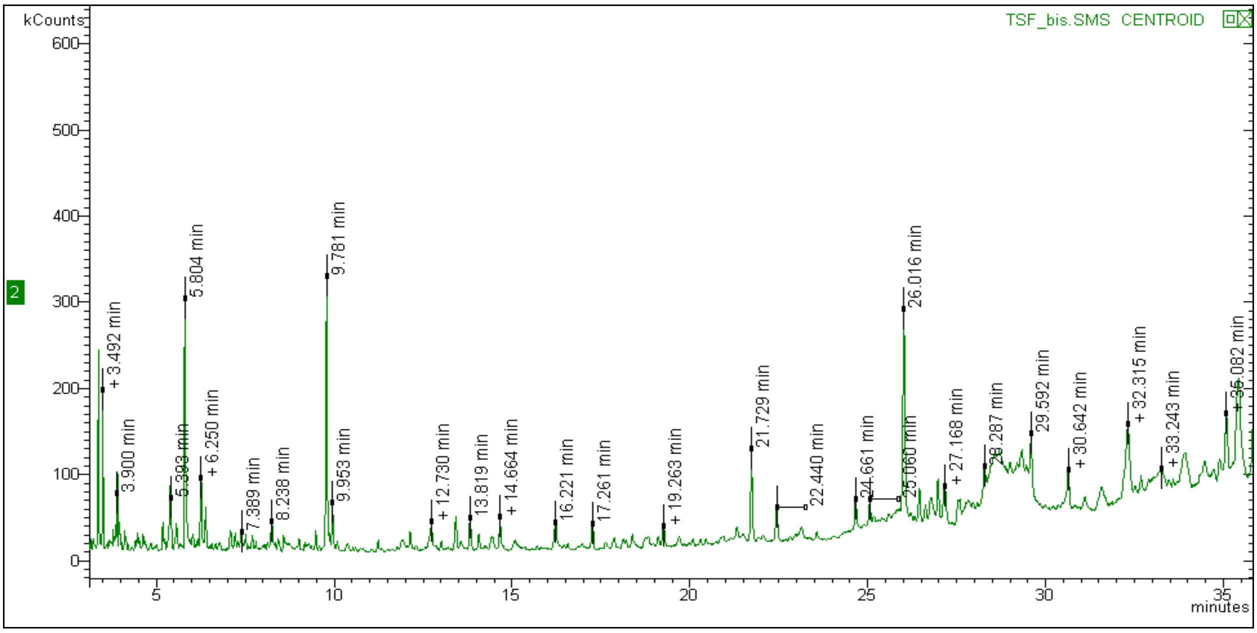

Fig 4: GC Chromatograph of Flower Essential Oils

4. Conclusions

The chemical constituents of essential oil of *Tecoma stans* leaf, stem, seed and flower are being reported. Comparing the volatile oils, six compounds were present in all the oil samples some of which have been listed above as dominant compounds, thirty-four components were found in at least two of the oil samples while twenty-four appear in only one of the essential oils making the total of sixty-four constituents identified altogether. Most of the compounds that are present in appreciable quantity are odor imparting constituents.

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