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Zekeya, Never

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Never Zekeya
Nelson Mandela African Institute of Science and Technology, School of Life Science and Bio-engineering, P.O. Box 447, Arusha, Tanzania

Musa Chacha
Nelson Mandela African Institute of Science and Technology, School of Life Science and Bio-engineering, P.O. Box 447, Arusha, Tanzania

Francis Shahada
Nelson Mandela African Institute of Science and Technology, School of Life Science and Bio-engineering, P.O. Box 447, Arusha, Tanzania

Abdul Kidukuli
Nelson Mandela African Institute of Science and Technology, School of Life Science and Bio-engineering, P.O. Box 447, Arusha, Tanzania.

Correspondence:
Never Zekeya
Nelson Mandela African Institute of Science and Technology, School of Life Science and Bio-engineering, P.O. Box 447, Arusha, Tanzania.

Analysis of phytochemical composition of *Bersama abyssinica* by gas chromatography – mass spectrometry

Never Zekeya, Musa Chacha, Francis Shahada and Abdul Kidukuli

Abstract
Bersama abyssinica has been reported to possess a varied range of therapeutical and pharmacological applications due to presence of bioactive compounds. The present study was carried out to determine the phytochemical present in the Bersama *abyssinica* leaf, stem bark and root bark methanolic fraction using Gas chromatography coupled to mass spectrometer (GC-MS) analysis. A total of 24 phytocompounds were identified from leaves whereas 21 compounds from stem bark and 19 from root bark. The classes of compounds identified include; terpenes, vitamin, carotenoid (rhodopin), flavonoids, steroid, unsaturated and saturated fatty acids. Most of the identified compounds were previously reported to possess antimicrobial, antitumor, antiseptic, preservative, and insecticidal and antioxidant activities. *Bersama abyssinica* leaf methanolic fraction had higher amount of compounds. The most abundant metabolites to all fractions are 2-furancarboxaldehyde, 5-(hydroxymethyl)-, 1,2,3-benzenetriol, 2,3-dimethylfumaric acid, 4-pyridinecarboxylic acid, ethyl ester; levoglucosenone, 2,5-dimethoxythiophenol and D-Melezitose recorded in leaves , stem bark and root bark of *Bersama abyssinica*.

Keywords: Phytochemical, GS-MS analysis, Bersama abyssinica, therapeutical, pharmacological uses

1. Introduction
Bersama abyssinica (Melianthaceae) belongs to the genus *Bersama* which comprises four species [1]. Other species includes *B. engleriana*, *B. swynnertonii*, *B. swinnyi* and *B. yangambiensis*. In East Africa, there are two subspecies of *Bersama abyssinica* namely; *Bersama abyssinica* Fresen. subssp. *abyssinica and B. abyssinica subsp. paullinioides* [2]. Ethno medicinal information conveying this genus reveals that the plant species are used for various medicinal purposes. For example, in West Africa the *Bersama engleriana* bark, leaf and root decoctions are widely taken as a purgative to treat a range of stomach disorders, such as abdominal pain, colic, diarrhea, cholera, intestinal worms, amoebiasis and dysentery. Rabies, syphilis, gonorrhea and malaria are also treated with these decoctions [3]. *Bersama abyssinica* is used for treatment of rheumatism, aphrodisiac and snake bites by Babungo villagers in Cameroon [4]. Previous biological analysis of *Bersama abyssinica* revealed the presence of antimicrobial secondary metabolites [5]. Despite the activity displayed by *Bersama abyssinica*, only *Bersama engleriana* has been phytochemically studied where Xanthone glycosides, terpenoids and anthraquinones with antitumour, antibacterial and antifungal activities were reported from the stem bark, roots and leaves of *Bersama engleriana* [6]. Taking into account the importance of this medicinal plant, the methanolic fractions of leaves stem bark and root bark of *Bersama abyssinica* were analyzed for the phytochemical constituent for the first time using GC MS.

Fig 1: *Bersama abyssinica* seeds

Fig 2: *Bersama abyssinica* plant

2. Material and Methods

2.1 Preparation of plant materials and extraction

Leaves stem bark and root bark of *Bersama abyssinica* were collected from Ilolo village of Rungwe district in Mbeya, Tanzania. The plant materials were air dried under shade and then pulverized into fine particles and authenticated by Mr. Ahmed Mndolwa of Tanzania Forestry Research Institute (TAFORI). The voucher specimen number (BANZ 0114) was kept at Nelson Mandela African Institution of Science and Technology.

The plant materials were air dried under shade and then pulverized into fine particles. The pulverized leaves (1000 g), stem bark (1000 g) and root bark (1000 g) were sequentially macerated using petroleum ether, ethyl acetate, chloroform and ethanol for 48 h twice for each solvent. The respective extracts were filtered through muslin cloth on a plug of glass wool in a glass column and solvents were evaporated in vacuum using a rotary evaporator and stored in refrigerator at -20 ℃.

2.2 GC-MS Analysis

An Agilent 6890N GC was connected to an Agilent 5975 MS used (Agilent technologies, USA). The GC-MS was equipped with Agilent 7683 B autosampler and split/ splitless injector with electronic pressure control.

Capillary column (HP-5MS, 30 m, 0.25 mm i.d., 0.25 μm, Agilent J & W GC columns) was used. The temperature program was as follows: Initial temperature 70 °C, held for 1 min, 10 °C·min–1 ramp to 160 °C then held for 5 min, finally by 3 °C·min–1 to 240 °C and held for 18.5 min. The temperature of the injection port is 250 °C, helium used as carrier gas .The mass spectrometer operated in electron ionization mode with an ionizing energy of 70 eV, ion source temperature 230 °C, MS quadruple temperature 150 °C.

Table 1: Compounds detected in methanolic fractions of *Bersama abyssinica* Leaves

SN	Retention Time	Compound name	Molecular Formula	Molecular Weight
1	28	3,7,11,15-Tetramethyl-2-hexadecen-ol	$C_{20}H_{40}O$	296.531
2	28.612	Ethanol,2-(9-octadecenyloxy)-,(Z)-	$C_{20}H_{40}O_2$	312
3	32.240	Gibberellic acid	$C_{19}H_{22}O_6$	346.37
4	42.988	Hexa-t-butylselenatrisiletane	$C_{24}H_{54}SeSi_3$	506
5	45.561	Trimethyl-3-(3,8,12,16-tetramethyl-heptadeca-3,7,11	$C_{30}H_{52}O$	428.733
6	51.616	7,8-Epoxylanostan-11-ol,3-acetoxy	$C_{32}H_{54}O_4$	502
7	52.852	Vitamin E	$C_{29}H_{50}O_2$	430.7
8	18.25	2-Furancarboxaldehyde,5-(hydroxymethyl)-	$C_6H_6O_3$	126.11
9	19.626	1,1,3,3-Tetramethyl-1,3-disilaphenalane	$C_{15}H_{20}Si_2$	256.4903
10	21.308	1,2,3-Benzenetriol	$C_6H_6O_3$	126
11	25.838	Dasycarpidan-1-methanol,acetate (ester)	$C_{20}H_{26}N_2O_2$	326
12	5.739	Disiloxane 1,3-diethoxy-1,1,,3,3-tetramethyl	$C_8H_{22}O_3Si_2$	222.43
13	6.00	2H-q-Benzopyran,3,5,6,8a-tetrahydro-2,5,5,8a-tetramethyl	$C_{13}H_{20}O$	192.2973
14	6.244	Furfural	$C_5H_4O_2$	96.08
15	6.681	Decane	$C_{10}H_{22}$	142.28
16	7.001	Decane,4-methyl-	$C_{11}H_{24}$	156.308
17	8.01	1-Dodecanol,2-methyl-,(S)	$C_{13}H_{28}O$	200.36
18	7.861	Capric ether	C_7H_{15} COOH	144
19	8.986	Undecane	$C_{11}H_{24}$	156
20	9.629	Perhydrocyclopropa[e]azulene-4,5,6-triol,1,1,4,6-tetramethyl	$C_{15}H_{26}O_3$	254.365
21	11.581	Dodecane	$C_{12}H_{26}$	170.33
22	13.505	2,3-Dimethylfumaric acid	$C_6H_8O_4$	144.127
23	14.728	Levoglucosenone	$C_6H_6O_3$	126
24	15.543	4-Pyridinecarboxylic acid, ethyl ester	$C_8H_9NO_2$	151.1626

Table 2: Compounds detected in methanolic fractions of *Bersama abyssinica* stem bark

SN	Retention Time	Compound name	Molecular Formula	Molecular Weight
1	13.521	2,3-Dimethyfumaric acid	$C_6H_8O_4$	144.127
2	14.728	Levoglucosenone	$C_6H_6O_3$	126
3	15.208	2-t-Butyl-5-propyl-[1,3]dioxolan-4-one	$C_{11}H_{20}O_3$	200.27
4	18.5	2-Furancarboxaldehyde5-(hydroxymethyl)	$C_6H_6O_3$	126
5	19.638	1,1,3,3-Tetramethyl-1,3-disilaphenalane	$C_{15}H_{20}Si_2$	256.4903
6	21.308	1,2,3-Benzenetriol	$C_6H_6O_3$	126
7	22.42	1,4-Benzenediol,2-methoxy-	$C_7H_8O_3$	140.1366
8	25.832	D-Melezitose	$C_{18}H_{32}O_{16}$	504.44
9	26.962	Vanillic acid hydrazide	$C_8H_{10}N_2O_3$	182.1766
10	29.37	2,5-Dimethoxythiophenol	$C_8H_{10}O_2S$	170.23
11	30.635	Pentadecanoic acid,13-methyl-,methyl ester	$C_{17}H_{34}O_2$	270.4507
12	30.98	4-Hydroxy-2-methoxycinnamaldehyde	$C_{10}H_{10}O_3$	178.1846
13	32.622	Ethanone,2-(benzoyloxy)-1-[1,1'-biphenyl]-4-yl	$C_{21}H_{18}O_2$	302.38
14	34.422	8,11-Octadecadienoic acid, methyl ester	$C_{19}H_{34}O_2$	294.4721
15	36.332	5,8,11,14-Eicosatetraynoic acid	$C_{20}H_{24}O_2$	296.5
16	51.293	Rhodopin	$C_{40}H_{58}O$	554.89

17	5.81	Ethyl iso-allocholate	$C_{26}H_{44}O_5$	436
18	5.81	Pyrrolidine,2-butyl-1-methyl-	$C_5H_{11}N$	85.15
19	6.272	Furfural	$C_5H_4O_2$	96.08
20	8.983	Undecane	$CH_3(CH_2)_9CH$	156.31
21	9.41	2-furancarboxaldehyde,5-methyl	$C_6H_6O_2$	110.1106

Table 3: Compounds detected in methanolic fractions of *Bersama abyssinica* root bark

SN	Retention Time	Compound name	Molecular Formula	Molecular Weight
1	5.833	Pyrrolidine,2-butyl-1-methyl-	$C_5H_{11}N$	85.15
2	6.285	Furfural	$C_5H_4O_2$	96.08
3	8.983	Undecane	$CH_3(CH_2)_9CH$	156.31
4	9.414	2-Furandicarboxaldehyde,5-methyl-	$C_6H_4O_3$	124.0942
5	13.511	2,3-Dimethylfumaric acid	$C_6H_8O_4$	144.127
6	14.1	2,5-Furandicarboxaldehyde	$C_6H_4O_3$	61.68
7	14.737	Levoglucosenone	$C_6H_6O_3$	126
8	15.253	Pentanoic acid, heptyl ester	$C_{12}H_{24}O_2$	200.3178
9	18.404	2-Furancarboxaldehyde,5-(hydroxymethyl)-	$C_6H_6O_3$	126
10	19.638	1,1,3,3-Tetramethyl-1,3-disilaphenalane	$C_{15}H_{20}Si_2$	256.4903
11	21.34	1,2,3-Benzenetriol	$C_6H_6O_3$	126
12	45.56	2,2,4-Trimethyl-3-(3,8,12,16-tetramethyl-heptadecane-3,7,11,15-t	$C_{30}H_{52}O$	428
13	46.829	Ethyl iso-allocholate	$C_{26}H_{44}O_5$	436
14	52.85	Cholest-4-ene,3?-(methoxymethoxy)-	$C_{29}H_{50}O_2$	430.706
15	57.215	9,19-Cyclolanostane-3,7-diol	$C_{30}H_{52}O_2$	444
16	25.847	D(+)-Melezitose	$C_{18}H_{32}O_{16}.H_2O$	522.45
17	29.376	2,5-Dimethoxy thiophenol	$(CH_3O)_2C_6H_3SH$	170.23
18	30.637	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270.4507
19	34.419	9,12-Octadecadienoic acid, methyl ester, (E,E)-	$C_{19}H_{34}O_2$	294.4721

Table 4: Activity of phyto-compounds identified in the Methanolic Fractions of Bersama abyssinica Leaves, stem bark and root bark

S N	RT	Name of Compound	Nature of compound	Therapeutic Activity	References
1.	32	Gibberellic acid	Pentacyclic diterpene	Promoting growth and elongation of cells.	[7,8]
2.	28	3,7,11,15-Tetramethyl-2-hexadecen-ol	Terpene Alcohol	Antimicrobial, Anti-inflammatory	[9]
3.	51.616	7,8-Epoxylanostan-11-ol,3-acetoxy	Alcoholic compound	Antimicrobial, anti-inflammatory	[10,11]
4.	52.852	Vitamin E	Vitamin compound	Antidermatitic, Antileukemic, Antitumor, Antiageing, Analgesic, Antidiabatic, Anti-inflammatory, Antioxidant	[12,13]
5.	21.308	1,2,3-Benzenetriol	Pyrogallol	Antioxidant, Antiseptic, Antibacterial, Antidermatitic Fungicide, Pesticide, Antimutaginic Dye Candidicide	[14,15]
6.	7.001	Decane,4-methyl-	Alkane Compound	Antidermatitic	[16]
7.	8.01	Capric ether	Fatty acid	Decrease cholesterol, Antibacterial, antiprotozoal	[17,18]
8.	13.505	2,3-Dimethylfumaric acid	Fatty acid	immunomodulatory, Antitumor, sarcoidosis, antioxidant, antibacteria	[19]
9.	14.728	Levoglucosenone	Fatty acid	Chiral agent, catalyst	[20]
10.	26.962	Vanillic acid hydrazide	Phenolic compound	Antioxidant, flavor compound	[21]
11.	51.293	Rhodopin	Carotenoid.	Antioxidant	[22]
12.	9.414	2-furancarboxaldehyde,5-methyl	Aldehyde	antimicrobial, preservative, antioxidant	[23]
13.	46.829	Ethyl iso-allocholate	Steroid	anti-inflammatory, anticancer antimicrobial, antiasthma, diuretic	[24,25]
14.	58	9,19-Cyclolanostane-3,7-diol	Triterpene	anti-inflammatory	[26]

15.	30.637	Hexadecanoic acid, methyl ester	Palmitic acid methyl ester	Antioxidant, Hypocholesterolemic, Nematicide, Pesticide, Antiandrogenic flavor, Hemolytic, Alphareductase inhibitor	[27,28]
16.	34.419	9,12-Octadecadienoic acid, methyl ester, (E,E)-	Linoleic acid	Hypocholesterolemic, Nematicide, Antiarthritic, Hepatoprotective, Antiandrogenic, Hypocholesterolemic 5-Alpha reductase inhibitor, Antihistaminic, Anticoronary, Insectifuge, Antieczemic, Antiacne	[29,30]

3. Results

The results pertaining to GC-MS analysis led to the identification of number of compounds from the GC fractions of methanolic extract of *Bersama abyssinica.* These compounds were identified through mass spectrometry attached with GC and the results are tabulated in Table 1. The nature of active principles with their retention time (RT), molecular formula and molecular weight (MW)) in the methanolic fraction of *B. abyssinica* parts are presented in Table1, 2 and 3.

The results revealed the presence of 24, 22 and 21 various phytocompounds in leaves, stem bark and root bark respectively. The leaves presented high amount of phyto components where Gibberellic acid, Hexa-t-butylselenatrisiletane, 3,7,11,15-tetramethyl-2-hexadecen-ol, ethanol,2-(9-octadecenyloxy)-,(Z)-; trimethyl-3-(3,8,12,16-tetramethyl-heptadeca-3,7,11; 7,8-epoxylanostan-11-ol,3-acetoxy,vitaminE;2-furancarboxaldehyde,5-(hydroxymethyl),1,2,3-benzenetriol,dasycarpidan-1-methanol,acetate (ester),capric ether and dodecanol 2-methyl-,(S) are reported for the first time in Tanzanian *Bersama abyssinica* although similar compounds have been reported in other plants [31].

However, 2,3-dimethylfumaric acid, 1,1,3,3-tetramethyl-1,3-disilaphenalane,1,2,3-benzenetriol,fufural, 2-furancarboxaldehyde,5-(hydroxymethyl) and undecane were present in both leaves and stem bark and root bark showing that there are some compounds which are evenly distributed in all plant parts. On the other hand 4-pyridinecarboxaldehyde,5-(hydroxymethyl),2-furancarboxaldehyde,5-(hydroxymethyl)-,1,1,3,3-tetramethyl-1,3-disilaphenalane, 1,2,3-benzenetriol were identified in both root bark and stem bark showing that barks are potential source of bioactive compound as reported by other authors [25]

Among the detected compounds few some reported to be potential therapeutic agents. For instance,9,12-octadecadienoic acid, methyl ester is effective antihistaminic, anti-coronary, Insectifuge and antieczemic [32, 33]. These findings are also supported by the study done by Zekeya and others (2014) that revealed the insecticidal activity of *Bersama abyssinica* extracts.

On the other hand ethyl iso-allocholate was reported to exhibit anti-inflammatory, anticancer antimicrobial [35, 25], whereas hexadecanoic is effective antioxidant, hypocholesterolemic, nematicide and pesticide properties [36]. However, vitamin E, 9,19-cyclolanostane-3,7-diol, 3,7,11,15-tetramethyl-2-hexadecen-ol and 2-furancarboxaldehyde,5-methyl were also reported to possess antimicrobial, preservative, anti-inflammatory and antioxidant activity [26, 37, 38] whereas gibberellic acid was found to be effective in promoting growth and elongation of cells [39, 40].

Contrary, among the detected compounds only Levoglucosenone is known to be a chiral agent whereas 9,12-Octadecadienoic acid, methyl ester, (E,E)-was found to possess many activity than other compounds

Moreover, antimicrobial, antioxidant and anti-inflammatory activities were displayed by most compounds in this study indicating that different plant compounds can exhibit similar activity and this could be due to presence of similar functional groups

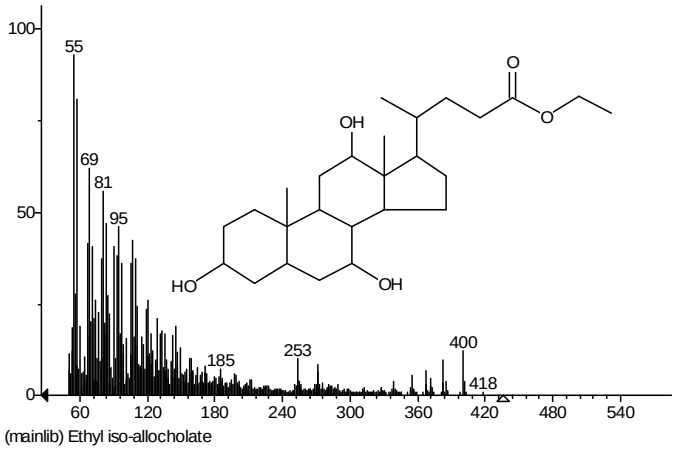

Fig 1: Mass Spectrum and structure of ethyl iso-allocholate

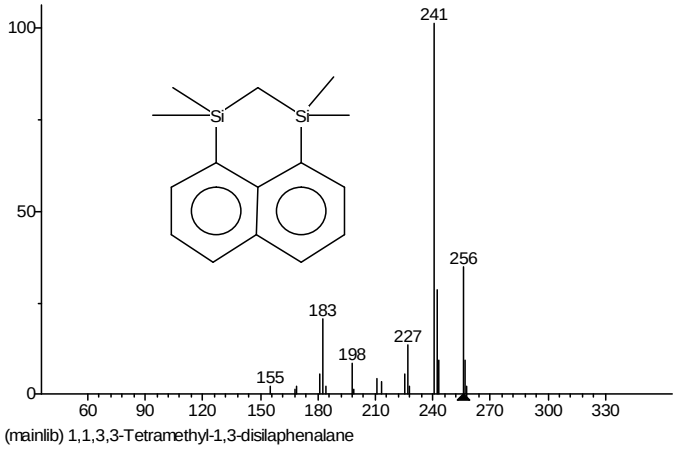

Fig 2: Mass Spectrum and structure of 1,1,3,3-tetramethyl-1,3-disilaphenalane

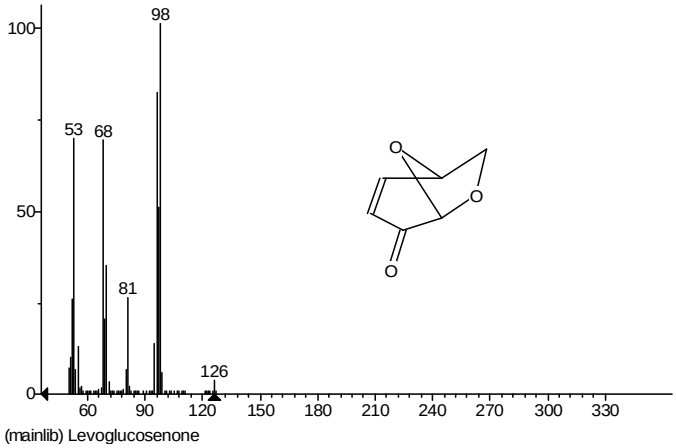

Fig 3: Mass Spectrum and structure of levoglucosenone

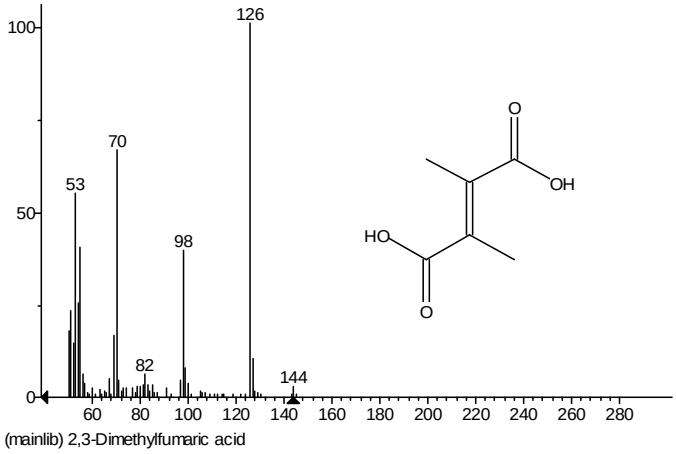

Fig 4: Mass Spectrum and structure of 2,3-dimethylfumaric acid

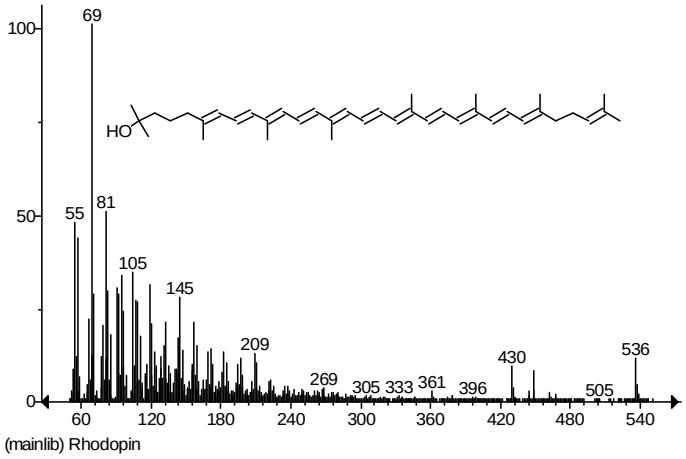

Fig 5: Mass Spectrum and structure of rhodopin

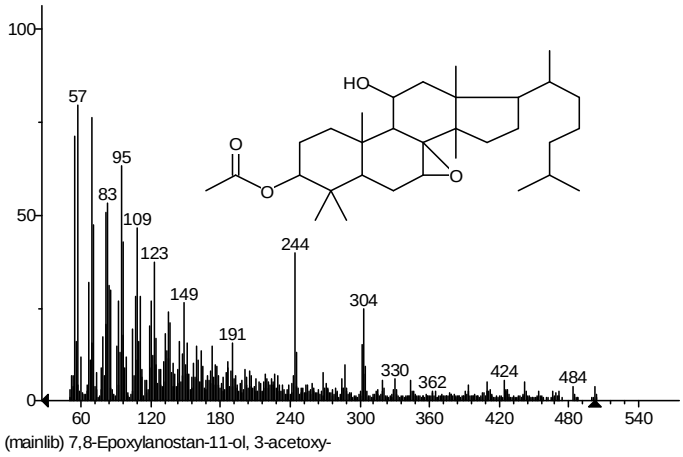

Fig 6: Mass Spectrum and structure of 7,8-epoxylanostan-11-ol,3-acetoxy

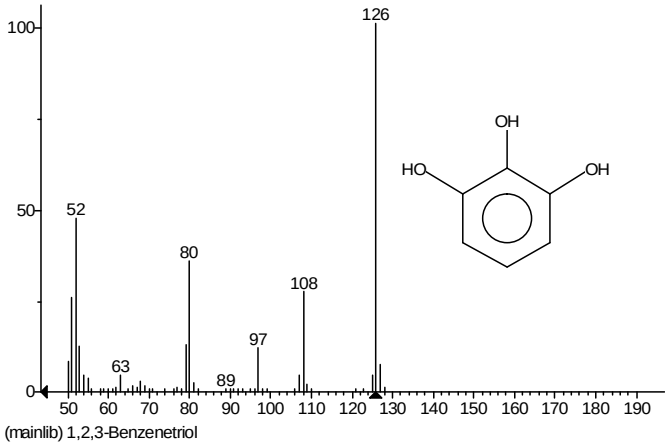

Fig 7: Mass Spectrum and structure of 1,2,3-benzenetriol

4. Acknowledgment

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5. References

1. Djemgou PC, Hussien TA, Hegazy M-E F, Ngandeu F, Neguim G, Tane P *et al.* C-Glucoside xanthone from the stem bark extract of *Bersama engleriana*. Pharmacognosy research 2010; 2:229.
2. Mikkelsen K, Seberg O. Morphometric analysis of the *Bersama abyssinica* Fresen. complex (Melianthaceae) in East Africa. Plant Systematics and Evolution 2001; 227:157-182.
3. Kuete V, Mbaveng AT, Tsaffack M, Beng VP, Etoa F-X, Nkengfack AE. Antitumor, antioxidant and antimicrobial activities of *Bersama engleriana* (Melianthaceae). Journal of ethnopharmacology 2008; 115:494-501.
4. Lather A, Gupta V, Tyagi V, Kumar V, Garg C. Phytochemistry and Pharmacological Activity of *Bersama abyssinica* Guerke-An overview. International Research Journal of Pharmacy 2010; 1(1):89-94
5. Zekeya N, Chacha M, Shahada F. Antibacterial and

antifungal activity of Tanzanian *Bersama abyssinica*, International Journal of Science and Research 2014; 3(7):1150-1154
6. Mbaveng AT, Kuete V, Mapunya BM, Beng VP, Nkengfack AE, Meyer JJM *et al*. Evaluation of four Cameroonian medicinal plants for anticancer, antigonorrheal and antireverse transcriptase activities. Environmental Toxicology and Pharmacology 2011; 32:162-167.
7. Filner P, Varner J. A test for de novo synthesis of enzymes: density labeling with H_2O18 of barley alpha-amylase induced by gibberellic acid. Proceedings of the National Academy of Sciences of the United States of America 1967; 58:1520.
8. Kaufman PB, Ghosheh N, Ikuma H. Promotion of growth and invertase activity by gibberellic acid in developing Avena internodes. Plant physiology 1968; 43:29-34.
9. Zhang L, Fang J, Joeckel R. Microbial biomass and community structure in alkaline lakes of the Nebraska Sand Hills, USA. Chemical Geology 2013; 356:171-180.
10. Hassan WH, El Gamal AA, El-Sheddy E, Al-Oquil M. Journal of Chemical and Pharmaceutical Research 2014; 6(2):604-615.
11. Lee H-S, Bilehal D, Lee G-S, Ryu D-S, Kim H-K, Suk D-H, Lee D-S. Anti-inflammatory effect of the hexane fraction from Orostachys japonicusin RAW 264.7 cells by suppression of NF-κB and PI3K-Akt signaling. Journal of Functional Foods 2013; 5:1217-1225.
12. Burton G, Le Page Y, Gabe E, Ingold K. Antioxidant activity of vitamin E and related phenols. Importance of stereoelectronic factors. Journal of the American Chemical Society 1980; 102:7791-7792
13. Acker VSA, Koymans LM, Bast A. Molecular pharmacology of vitamin E: structural aspects of antioxidant activity. Free Radical Biology and Medicine 1993; 15:311-328.
14. Song S, Lee H, Jin Y, Ha YM, Bae S, Chung HY *et al*. Syntheses of hydroxy substituted 2-phenyl-naphthalenes as inhibitors of tyrosinase. Bioorganic & medicinal chemistry letters 2007; 17:461-464.
15. Vadivel E, Gopalakrishnan S. Gc-Ms Analysis of some bioactive constituents of Mussaenda Frondosa Linn. Inter J Pharm Bio Sci 2011; 2.
16. Heintz A, Kulikov DV, Verevkin SP. Thermodynamic properties of mixtures containing ionic liquids. 1. Activity coefficients at infinite dilution of alkanes, alkenes, and alkylbenzenes in 4-methyl-n-butylpyridinium tetrafluoroborate using gas-liquid chromatography. Journal of Chemical & Engineering Data 2001; 46:1526-1529.
17. Bergsson G, Arnfinnsson J, Karlsson SM, Steingrímsson Ó, Thormar H. *In vitro* inactivation of Chlamydia trachomatis by fatty acids and monoglycerides. Antimicrobial agents and chemotherapy 1998; 42:2290-2294.
18. Goel G, Arvidsson K, Vlaeminck B, Bruggeman G, Deschepper K, Fievez V. Effects of capric acid on rumen methanogenesis and biohydrogenation of linoleic and α-linolenic acid. Animal 2009; 3:810-816.
19. Sangeetha J, Vijayalakshmi K. Determination of bioactive components of ethyl acetate fraction of Punica granatum rind extract. Int J Pharm Sci Drug Res 2011; 3:116-122.
20. Kudo S, Zhou Z, Yamasaki K, Norinaga K, Hayashi J-I. Sulfonate Ionic Liquid as a Stable and Active Catalyst for Levoglucosenone Production from Saccharides via Catalytic Pyrolysis. Catalysts 2013; 3:757-773.
21. Alissandrakis E, Kibaris AC, Tarantilis PA, Harizanis PC, Polissiou M. Flavour compounds of Greek cotton honey. Journal of the Science of Food and Agriculture 2005; 85:1444-1452.
22. Sheela D, Uthayakumari F. GC-MS Analysis Of Bioactive Constituents From Coastal Sand Dune Taxon–Sesuvium Portulacastrum (L.). Bioscience Discovery 2013; 4:47-53.
23. Park B-S, Lee K-G, Shibamoto T, Lee S-E, Takeoka GR. Antioxidant activity and characterization of volatile constituents of Taheebo (Tabebuia impetiginosa Martius ex DC). Journal of agricultural and food chemistry 2003; 51:295-300.
24. Daffodil E, Uthayakumari F, Mohan V. GC-MS determination of bioactive compounds of curculigo orchioides gaertn. Science Research Reporter 2012; 2:198-201.
25. Sarada K, Margret RJ, Mohan V. GC–MS Determination of Bioactive Components of Naringi crenulata (Roxb) Nicolson. Int J Chem Tech Research 2011; 3:1548-1555.
26. Priya KS, Satyavathi K, Bhojaraju P, Kumari YR, Prasad DV, Durga MSS *et al*. Gc-ms profiling and anthelmintic activity of antigonon leptopus leaves.
27. Ha YL, Storkson J, Pariza MW. Inhibition of benzo (a) pyrene-induced mouse forestomach neoplasia by conjugated dienoic derivatives of linoleic acid. Cancer Research 50, 1097-1101.
28. Zheng CJ, Yoo J-S, Lee T-G, Cho H-Y, Kim Y-H, Kim W-G. Fatty acid synthesis is a target for antibacterial activity of unsaturated fatty acids. FEBS letters 2005; 579:5157-5162.
29. Baumgard LH, Corl BA, Dwyer DA, Sæbø A, Bauman DE. Identification of the conjugated linoleic acid isomer that inhibits milk fat synthesis. American Journal of Physiology-Regulatory, Integrative and Comparative Physiology 2000; 278:R179-R184.
30. Reitz RC, Lands WE, Christie WW, Holman RT. Effects of ethylenic bond position upon acyltransferase activity with isomeric cis, cis-octadecadienoyl coenzyme A thiol esters. Journal of Biological Chemistry 1968; 243:2241-2246.
31. Yogeswari S, Ramalakshmi S, Neelavathy R, Muthumary J. Identification and Comparative Studies of Different Volatile Fractions from Monochaetia kansensis by GCMS. Global Journal of Pharmacology 2012; 6:65-71.
32. Gardner H, Weisleder D. Lipoxygenase fromZea mays: 9-d-hydroperoxy-trans-10, cis-12-octadecadienoic acid from linoleic acid. Lipids 1970; 5:678-683.
33. Griinari J, Corl B, Lacy S, Chouinard P, Nurmela K, Bauman D. Conjugated linoleic acid is synthesized endogenously in lactating dairy cows by Δ9-desaturase. The Journal of nutrition 2000; 130:2285-2291.
34. Zekeya N, Chacha M, Shahada F. Bioefficacy of *Bersama abyssinica* extracts against cowpea beetle; *Callosobruchus maculatu*s in storage International journal of Innovative Research and Development 2014; 3(8):337-341.
35. Muthulakshmi A, Margret RJ, Mohan V. GC-MS Analysis of Bioactive Components of Feronia

elephantum Correa (Rutaceae) 2012.
36. Al-Shammari LA, Hassan WH, Al-Youssef HM. Chemical composition and antimicrobial activity of the essential oil and lipid content of *Carduus pycnocephalus* L. growing in Saudi Arabia. Journal of Chemical and Pharmaceutical Research 2012; 4(2):1281-1287.
37. Porfirova S, Bergmüller E, Tropf S, Lemke R, Dörmann P. Isolation of an Arabidopsis mutant lacking vitamin E and identification of a cyclase essential for all tocopherol biosynthesis. Proceedings of the National Academy of Sciences 2002; 99:12495-12500.
38. Ramalakshmi S, Muthuchelian K. Analysis of Bioactive constituents from the Ethanolic leaf extract of Tabebuia rosea (Bertol.) DC by Gas Chromatography-Mass Spectrometry. International Journal of ChemTech Research 2011; 3.
39. Eriksson ME, Israelsson M, Olsson O, Moritz T. Increased gibberellin biosynthesis in transgenic trees promotes growth, biomass production and xylem fiber length. Nature biotechnology 2000; 18:784-788.
40. Fleet CM, Sun T-P. A DELLAcate balance: The role of gibberellin in plant morphogenesis. Current opinion in plant biology 2005; 8:77-85.