Tanzania Journal of Health Research
Volume 21, Number 2, August 2019
Doi: https://dx.doi.org/10.4314/thrb.v21i2.6

Antimicrobial activities and phytochemical analysis of extracts from *Ormocarpum trichocarpum* (Taub.) and *Euclea divinorum* (Hiern) used as traditional medicines in Tanzania

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Abstract

Background: Medicinal plants have been of great value to human healthcare in most parts of the world for thousands of years. In Tanzania, over 12,000 species of higher plants have been reported, and about 10% are estimated to be used as medicines to treat different human health conditions. The present study aimed to determine *in vitro* antimicrobial activities and phytochemical analysis of *Ormocarpum trichocarpum* and *Euclea divinorum* which are commonly used as traditional medicine in Tanzania.

Methods: The Minimum Inhibitory Concentration (MIC) of plants extracts against tested bacterial and fungal species were determined using 96 wells microdilution method. In this method, 50 µL of nutrient and saboraud's dextrose broth for bacteria and fungus respectively were loaded in each well followed by 50 µL of extract to make final volume of 100 µL. Subsequently 50 µL were transferred from first rows of each well to the second rows and the process was repeated down the columns to the last wells from which 50 µL were discarded. Thereafter, 50 µL of the selected bacterial and fungal suspension were added to each well thus making final volume of 100µL. The lowest concentration which showed no microbe growth was considered as MIC. The study also evaluated phytochemical compounds present in the ethyl acetate extracts from *O. trichocarpum* stem bark and *E. divinorum* root bark extract using Gas Chromatography-Mass Spectrometry (GC-MS) technique.

Results: It was revealed that 66% of the tested microbes were susceptible to plant extracts at MIC value of 0.39 mg/mL whereas 83% being susceptible to extracts at MIC value of 0.781 mg/mL. Interestingly, four out of 18 tested plant extracts exhibited high antifungal activity below that of the standard antifungal drug, fluconazole. The GC-MS analysis revealed presence of various low molecular weight phytochemicals which belongs to six groups of secondary metabolites namely dieterpenes, alphatic hydrocarbons, tetraterpenes, sesquiterpenes, steroid and triterpenes.

Conclusion: The resence of various phytochemicals in the tested plant extracts were associated with pharmacological properties of *O. trichocarpum* and *E. divinorum* and therefore justifying ethnomedical usage of such plants.

Keywords: Antibacterial, antifungal, *Ormocarpum trichocarpum*, *Euclea divinorum*

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Doi: https://dx.doi.org/10.4314/thrb.v21i2.6

Introduction

Worldwide, consumers have a positive intention toward medicinal plants and traditional health systems in solving health care problems (Srivastava, 2018). Medicinal plants have been potential resources of health care services to the majority of people around the world for thousands of years (Mkangara et al., 2019). Globally, about 72,000-77,000 plant species are being utilized for medicinal purpose (Rao and Rajput, 2010). In Tanzania, over 12,000 species of higher plants have been reported, and about 10% are estimated to be used as medicines to treat different human health conditions (Mahunnah et al., 2012). Some of these reported plants which are commonly used for treatment of human diseases include *Ormocarpum trichocarpum* (Taub.) and *Euclea divinorum* (Hiern). The *O. trichocarpum* (Fabaceae) which is also known as "Esikilianjoi" in Maasai language is a shrub growing from 1 - 5 meters tall (Rufo et al., 2002). The plant is harvested from the wild as a local source of food, medicines and wood. In Africa, *O. trichocarpum* is distributed in Tanzania, Ethiopia, Somalia, Uganda, Kenya, eastern DR Congo, Rwanda, Burundi, Zambia, Zimbabwe, Mozambique and South Africa where it is mostly important in the management of various diseases such as tuberculosis, gastrointestinal problems, sexually transmitted diseases, stroke paralysis, bone setting and bilharzia (Johns et al., 1995; Ndubani and Hojer, 1999; Moshi et al., 2006; Tabuti et al., 2010). Traditionally, leaves are used by local people in Zimbabwe and Kenya for treating abdominal pains and various stomach ailments (Hutchings et al., 1996).

Regarding *Euclea divinorum,* (Ebenaceae) is a shrub or small tree up to about 6 m tall, often branching from the base or sometimes with a single stem. The barks are grey, fairly smooth in young trees but fissured in older specimens. Leaves are simple, coriaceus, lanceolate, margins wavy, sub-opposite or alternate with 3.5-9 cm long and 1-2.5 cm wide (Rufo et al., 2002). The upper surfaces of the leaves is light green or grey green, sometimes with a yellowish tinge while the lower surface is pale and smooth in texture. Flowers are small, cup-shaped and creamy in colour borne on a short dense head and inflorescence covered with tiny, rusty-brown dots. The plant is widely distributed in the northeastern parts of South Africa, through Mozambique, Zimbabwe, Botswana and northern Namibia into tropical Africa where it occurs naturally in thicket, thorn scrub, on hillsides, along riverbanks and in woodland (Hutchings et al., 1996). In Tanzania, the plant is locally known as "Mdaa" in Swahili language where it is commonly found in highlands of Arusha, Morogoro and Iringa. This plant is traditionally important in treating various human diseases. Fine powder prepared from roots and leave of this plant is reportedly used by many ethnic groups in Tanzania for treatment of chest pain, asthma, pneumonia, tonsillitis, sore throat, constipation, gout and snake bites (Kipkore et al., 2014).

Taking into consideration the contribution of *O. trichocarpum* and *E. divinorum* in the treatment of various human diseases, it is therefore justifiable that these plants are amongst the medicinal plants which are traditionally known for many remedial applications. Despite of their medicinal value, there is lack of scientific studies regarding the medicinal potential and phytochemicals responsible for therapeutic effects of such plants. This study therefore reports the antimicrobial activities and phytochemical analysis of these two herbal plants which are commonly used as traditional medicine in Tanzania.

Materials and methods

Acquisition of materials

Petroleum ether applied in this study to extract non polar compounds was purchased from Avantor Perfomance Materials Limited, Gujarat, India. Dimethyl sulphoxide (DMSO) and ethyl acetate applied in order to dissolve plant extracts and extracting non polar compounds from plant materials respectively, were bought from RFCL Limited, Haryana, India. The growth media used in this study was nutrient broth which supplied by HIMEDIA Laboratories Pvt. Limited, Mumbai, India. Four bacterial and two fungal strains namely *Staphylococcus aureus* (ATCC 25923), *Klebsiella pneumoniae*

Doi: https://dx.doi.org/10.4314/thrb.v21i2.6

(ATCC 708903), *Escherichia coli* (ATCC 25922), *Salmonella typhi* (NCTC 8385), *Candida albicans* (ATCC 90028) and *Cryptococcus neoformans* (Clinical isolate*)* respectively were obtained from the department of Microbiology and Immunology, Muhimbili University of Health and Allied Sciences (MUHAS) in Tanzania. Gentamycin and iodonitrotetrazolium chloride were supplied by Mediatech Incorporation, Manassas, USA and SIGMA® (Sigma- Aldrich®, St. Louis, USA) respectively.

Preparation of plant extracts and extraction

Plant materials of *O. trichocarpum* and *E. divinorum* collected from Esilalei and Imbibya villages in Monduli and Arumeru districts respectively. Plant species were identified by Mr. Gabriel Laizer, a botanist from Tropical Pesticide Research Institute (TPRI) and voucher specimens coded OT-0001 for *O. trichocarpum* and ED-0002 for *E. divinorum* are kept at the University of Dodoma. Plant materials were air dried and pulverized using electric blender. For non-polar and medium polar extraction, pulverized materials (250 g of leaves, stem and root barks) were sequentially successively macerated in petroleum ether and ethyl acetate. The respective extracts were filtered through Whatman No. 1 filter paper on a plug of glass wool in a glass column and solvents were evaporated through the vacuum using a rotary evaporator. For polar extraction, the same pulverized materials (250 g of leaves, stem and root barks) were added to a 1 L of distilled water at 70 °C and allowed to cool until reaching 40 °C in a water bath. The extracts were sieved and centrifuged at 5000 rpm for 10 min. The supernatant was collected and filtered using Whatman No. 1 filter paper and dried by freezing to eliminate water by sublimation. All extracts were stored in a deep freezer at -20 °C for further activities. To ensure complete extraction, each solution was shaken after every 30 minutes for six hours and allowed to stand for 48 hours as described by Olowa and Nuñeza, (2013). Extract yields (% yield = W_2/W_1*100, whereby, W_1 is the weight of pulverized sample before extraction, while W_2 is the weight of pulverized sample after extraction) of between 0.11- 4.65% were obtained. Respective extracts were filtered through Whatman No. 1 filter paper and solvents evaporated through vacuum using rotary evaporator and the final residues obtained were subjected to antimicrobial activity, preliminary phytochemical screening and GC-MS analysis.

Determination of antimicrobial activities

Minimum inhibitory concentrations (MICs) were determined by microdilution broth method using 96-well plates as described by Ellof, (1998). Plates were preloaded with 50 µL of nutrient broth for bacteria and Sabouraud's dextrose media for fungus in each well followed by addition of 50 µL of 100 mg/mL extract into the first wells of each row so as to make total volume of 100 µL in each of the first-row wells. The contents were mixed and 50 µL of the same drawn from each of the first-row wells and put into the next row wells. The process repeated down the columns to the last wells and the last 50 uL were discarded. Thereafter, 50 µL of microbes (0.5 McFarland standard turbidity) were added to each well thus making final volume of 100 µL per well. Gentamicin and fluconazole were used in two rows of each plate to serve as standard positive control drugs while nutrient broth and saboraud's dextrose was used as negative control and DMSO as solvent control. Plates were then incubated at 37°C for 24 hrs. Minimum Inhibition Concetrations for each extract were determined by adding 20 µL of 0.02% *p*-iodonitrotetrazolium chloride in each well followed by incubation at 32°C for 1 hr. Microbes growth was indicated by change of colour from clear to pink. Lowest concentration which showed no microbes growth considered as MIC.

Preliminary phytochemical screening

Preliminary phytochemical screening of the *O. trichocarpum* stem bark (OTSE) and *E. divinorum* root bark ethyl acetate (EDRE) extracts were tested for alkaloid, terpenoid, flavonoid, tannin and saponin. The choice of these extracts based on their highest potency against antimicrobial activity. The qualitative results are expressed as (+) for the presence and (−) for the absence of phytochemicals.

Test for alkaloid
A few drops of Dragendroff's reagent were added to a test tube containing 1 ml of plant extract and a colour change was observed. The appearance of an orange colour was an indication of the presence of alkaloids (Firdouse and Alam, 2011).

Test for terpanoid
In this test, about 5 ml of plant extract was added to a 2 ml of chloroform and 3 ml of concentrated sulphuric acid (H_2SO_4). The presence of terpenoids was indicated by reddish brown colour (Edeoga et al., 2005).

Test for flavonoids
About 2 ml of plant extract was treated with few drops of dilute sodium hydroxide (NaOH), followed by addition of dilute hydrochloric acid (HCl). A yellow solution with NaOH turned colorless with dilute HCl, which indicated presence of flavonoids (Onwukaeme et al., 2007).

Test for tannin
About 2 ml of plant extract was stirred with 2 ml of distilled water and few drops of ferric chloride ($FeCl_3$) solution were added. Formation of dark blue precipitate was indication of presence of tannins (Kumar et al., 2007).

Test for saponin
About 5 ml of plant extract was shaken vigorously with 5 ml of distilled water in a test tube. The formation of stable foam was taken as an indication of the presence of saponins (Parekh and Chanda, 2007).

GC-MS analysis
GC-MS analysis was carried out using Agilent 6890N GC connected to the Agilent 5975 MS (Agilent technologies, USA) with capillary column (HP-5) of 30 meter length, 0.25 mm diameter and 0.25 µm film thickness. Helium gas (99.999%) was used as carrier gas at a constant flow of 1mL/min and an injection volume of 1 µL was employed. The injector temperature was maintained at 250°C, the ion-source temperature was 280°C, the oven temperature was programmed from 110°C (isothermal for 2 min), with an increase of 10°C/min to 200°C, then 5°C/min to 280°C, ending with a 9 min isothermal at 280°C. The mass spectrometer operated in electron ionization mode with an ionizing energy of 70 eV and the ion source temperature was 230°C. The inlet line temperature was 200ºC and the total GC-MS running time was 36 minutes. Interpretation on mass spectrum of GC-MS was done using the database of National Institute Standard and Technology (NIST) having more than 62,000 patterns. Mass spectra of detected compounds from *O. trichocarpum* stem bark (OTSE) and *E. divinorum* root bark ethyl acetate (EDRE) extracts were compared with spectra of known compounds stored in the NIST library. As results, the name, molecular weight and structure of compounds contained in such extracts were determined.

Results
The antimicrobial activity of leaf, stem and root bark extracts from *O. trichocarpum* and *E. divinorum* were evaluated against four bacterial and two fungal species. The findings presented as minimum inhibition concentrations (MIC) indicated that plant extracts possess varying antimicrobial potencies as summarized in Tables 1 and 2. Out of six pathogenic microbes tested, only two bacteria and one fungus namely *S. aureus, S. typhi* and *C. neoformans* respectively were inhibited by plant extracts at MIC value of 0.048 mg/mL and 0.097 mg/mL respectively. Sixty six percent of tested microbes were susceptible to plant extracts at MIC value of 0.39 mg/mL while 83% being susceptible to extracts at

Doi: https://dx.doi.org/10.4314/thrb.v21i2.6

MIC value of 0.781 mg/mL. According to Rios and Recio, (2005) plant extracts that are recommended in the drug discovery initiatives are those with MIC values less than 1 mg/mL, and thus plant extracts that exhibited MIC values of 0.048 mg/mL, 0.097 mg/mL, 0.39 mg/mL and 0.781 mg/mL are potential source of drug templates. *Ormocarpum trichocarpum* stem aqueous (OTSA) and root bark ethyl acetate (OTRE) exhibited antimicrobial activity against all tested microbes with MIC range of 0.781-3.125 mg/mL and 0.097-0.781 mg/mL respectively suggesting that the two extracts are more active than other plant parts of *O. trichocarpum*. However, the highest antimicrobial activity was exhibited by *O. trichocarpum* stem bark ethyl acetate (OTSE) and *O. trichocarpum* root bark ethyl acetate (OTRE) extracts which displayed MIC value of 0.048 mg/mL and 0.097 mg/mL against *S. aureus, S. typhi* and *C. neoformans* respectively.

The root bark ethyl acetate (EDRE) extracts had high antimicrobial activity which is evidenced by the narrow MIC value range of 0.048-0.871 mg/mL followed by leaf aqueous (EDLA) extracts with MIC values range of 0.781-1.562 mg/mL. However, the highest antimicrobial activity was displayed by EDRE against *S. typhi* with MIC value of 0.048 mg/mL followed by *E. divinorum* stem bark aqueous (EDSA) and root bark petroleum ether (EDRP) extract against *S. aureus* with MIC values of 0.781 mg/mL. Additionally, the *E. divinorum* leaf ethyl acetate (EDLE) and *E. divinorum* leaf aqueous (EDLA) extracts displayed MIC value of 0.781 mg/mL against *S. typhi* and *C. albicans* respectively. Comparatively, the *E. divinorum* stem bark petroleum ether (EDSP) extract was the least active which showed MIC values of < 25 mg/mL against all tested pathogenic microbes.

Table 1: Antimicrobial activity of leaf, stem and root bark extracts from *O. trichocarpum*

Plant Extract	Minimum Inhibitory Concentrations (MICs) in mg/ml					
	S. aureus	***K. pneumoniae***	***E. coli***	***S. typhi***	***C. albicans***	***C. neoformans***
OTLP	3.125	>25	6.25	>25	>25	6.27
OTLE	1.562	3.125	6.25	3.125	>25	6.25
OTLA	1.562	6.25	1.562	6.25	3.125	1.562
OTSP	6.25	>25	6.25	>25	12.5	>25
OTSE	0.048	0.195	1.562	0.048	1.562	6.25
OTSA	0.781	1.562	1.562	3.125	3.125	0.781
OTRP	1.562	>25	>25	>25	>25	6.25
OTRE	0.097	0.39	0.781	0.39	0.781	0.097
OTRA	0.39	0.781	6.25	1.562	1.5625	0.39
Gentamycin	0.0625	0.0625	0.0625	0.0625	N/A	N/A
Fluconazole	N/A	N/A	N/A	N/A	1.56	0.78

Key: OTLP = *O. trichocarpum* leaf petroleum ether extract, OTLE = *O. trichocarpum* leaf ethyl acetate extract, OTLA = *O. trichocarpum* leaf aqueous extract, OTSP = *O. trichocarpum* stem bark petroleum ether extract, OTSE = *O. trichocarpum* stem bark ethyl acetate extract, OTSA = *O. trichocarpum* stem bark aqueous extract, OTRP = *O. trichocarpum* root bark petroleum ether extract, OTRE = *O. trichocarpum* root bark ethyl acetate extract, OTRA = *O. trichocarpum* root bark aqueous ether extract.

Table 2: Antimicrobial activity of leaf, stem and root bark extracts from *E. divinorum*

Plant Extract	Minimum Inhibitory Concentrations (MICs) in mg/ml					
	S. aureus	***K. pneumonia***	***E. coli***	***S. typhi***	***C. albicans***	***C. neoformans***
EDLP	>25	>25	6.25	>25	12.5	>25
EDLE	>25	>25	>25	0.781	>25	>25
EDLA	1.562	6.25	6.25	6.25	0.781	3.125
EDSP	>25	>25	>25	>25	>25	>25

Doi: https://dx.doi.org/10.4314/thrb.v21i2.6

Plant Extract	Minimum Inhibitory Concentrations (MICs) in mg/ml					
	S. aureus	***K. pneumonia***	***E. coli***	***S. typhi***	***C. albicans***	***C. neoformans***
EDSE	>25	>25	6.25	1.562	>25	>25
EDSA	0.781	>25	>25	>25	>25	>25
EDRP	0.781	>25	6.25	>25	>25	>25
EDRE	0.781	>25	6.25	0.048	6.25	>25
EDRA	3.125	>25	>25	>25	>25	6.25
Gentamycin	0.0625	0.0625	0.0625	0.0625	N/A	N/A
Fluconazole	N/A	N/A	N/A	N/A	1.56	0.78

Key: EDLP = *E. divinorum* leaf petroleum ether extract, EDLE = *E. divinorum* leaf ethyl acetate extract, EDLA = *E. divinorum* leaf aqueous extract, EDSP = *E. divinorum* stem bark petroleum ether extract, EDSE = *E. divinorum* stem bark ethyl acetate extract, EDSA = *E. divinorum* stem bark aqueous extract, EDRP = *E. divinorum* root bark petroleum ether extract, EDRE = *E. divinorum* root bark ethyl acetate extract, EDRA = *E. divinorum* root bark aqueous ether extract.

The preliminary phytochemical screening of *O. trichocarpum* stem bark (OTSE) and *E. divinorum* root bark ethyl acetate (EDRE) extracts revealed presence of major groups of phytochemical compounds namely alkaloids, terpenoid, flavonoid, tannin and saponin (Table 3). Furthermore, the volatile phytochemical compounds present in such extracts were identified by GC-MS technique. The retention time, peak areas, molecular formulas, molecular weights and biological activities of these compounds are presented in Table 4. These phytochemicals belong to various groups of secondary metabolites such as dieterpenes, alphatic hydrocarbons, tetraterpenes, sesquiterpenes, steroid and triterpenes. Dieterpene identified in the *O. trichocarpum* ethyl acetate stem bark extract was phytol whereas among aliphatic hydrocarbon group nonacosane was identified. Additionally, tetraterpene and sesquiterpene group of compounds identified in the same extract included tetracontane and τ-cadinol respectively (Figure 1). Furthermore, this study identified γ-sitosterol and germanicol from *E. divinorum* ethyl acetate root bark extract. These secondary metabolites belong to steroid and triterpenes group of compounds respectively as shown in Figure 2.

Table 3: Preliminary phytochemical screening of OTSE and EDRE extract

Phytochemical components	Plant extracts	
	OTSE	**EDRE**
Alkaloid	+	+
Terpenoid	+	+
Flavonoid	+	+
Tannin	-	+
Saponin	+	+

Key: + = Presence of compound, - = Absence of compound; OTSE= *O. trichocarpum* stem bark ethyl acetate extract bark extract; EDRE= *E. divinorum* root bark ethyl acetate extract

Doi: https://dx.doi.org/10.4314/thrb.v21i2.6

Table 4: Reported biological activities of volatile phytochemical compounds detected in *O. trichocarpum* stem bark (OTSE) and *E. divinorum* root bark ethyl acetate (EDRE) extracts

S/N	RT (min)	Peak area (%)	Name of compound	Molecular formula	Molecular weight (g/mol)	Reported bioactivity	References
1	29.69	9.40	Phytol	$C_{20}H_{40}O$	296.53	Antimicrobial, anticancer, diuretic, anti-inflammatory	Vivekanandadasan and Rajangam, (2015); Isaiah et al., (2016)
2	31.56	0.39	2-methylhexacosane	$C_{27}H_{56}$	380.73	Antimicrobial, hypocholesterolemic	Souti, (2016)
3	33.66	1.43	Nonacosane	$C_{29}H_{60}$	408.79	Tetany, anemia, pulmonary edema, antibacterial	Prabhadevi et al., 2012; Mihailovi et al., (2011)
4	39.51	1.30	Dotriacontane	$C_{32}H_{66}$	450.87	Antimicrobial, antioxidant, antispasmodic	Soosairaj and Dons, (2016)
5	44.76	43.31	Tetracontane	$C_{40}H_{82}$	563.08	Analgesic activity	Arora and Meena, (2018)
6	18.20	4.94	τ-cadinol	$C_{15}H_{26}O$	222.37	Antifungal	Ho et al., (2011)
7	51.47	4.09	γ-sitosterol	$C_{29}H_{50}O$	414.71	Antidiabetic, anticancer, antibacterial, antiviral, anti-inflammatory, anti-diarrhoeal, anti-infertility, antileukemic, antimutagenic, hepatoprotective, hypocholesterolemic, pesticide,	Elangovan et al., (2015)
8	51.72	88.84	Germanicol	$C_{30}H_{50}O$	426.73	Anticancer, anti-inflammatory	Padilha et al., 2010; Hu et al., (2016)
9	30.33	1.84	Oxacycloheptadec-8-en-2-one, (8Z)	$C_{16}H_{28}O_2$	252.39	Antiasthmatic activity	Soorya et al., (2017)

Doi: https://dx.doi.org/10.4314/thrb.v21i2.6

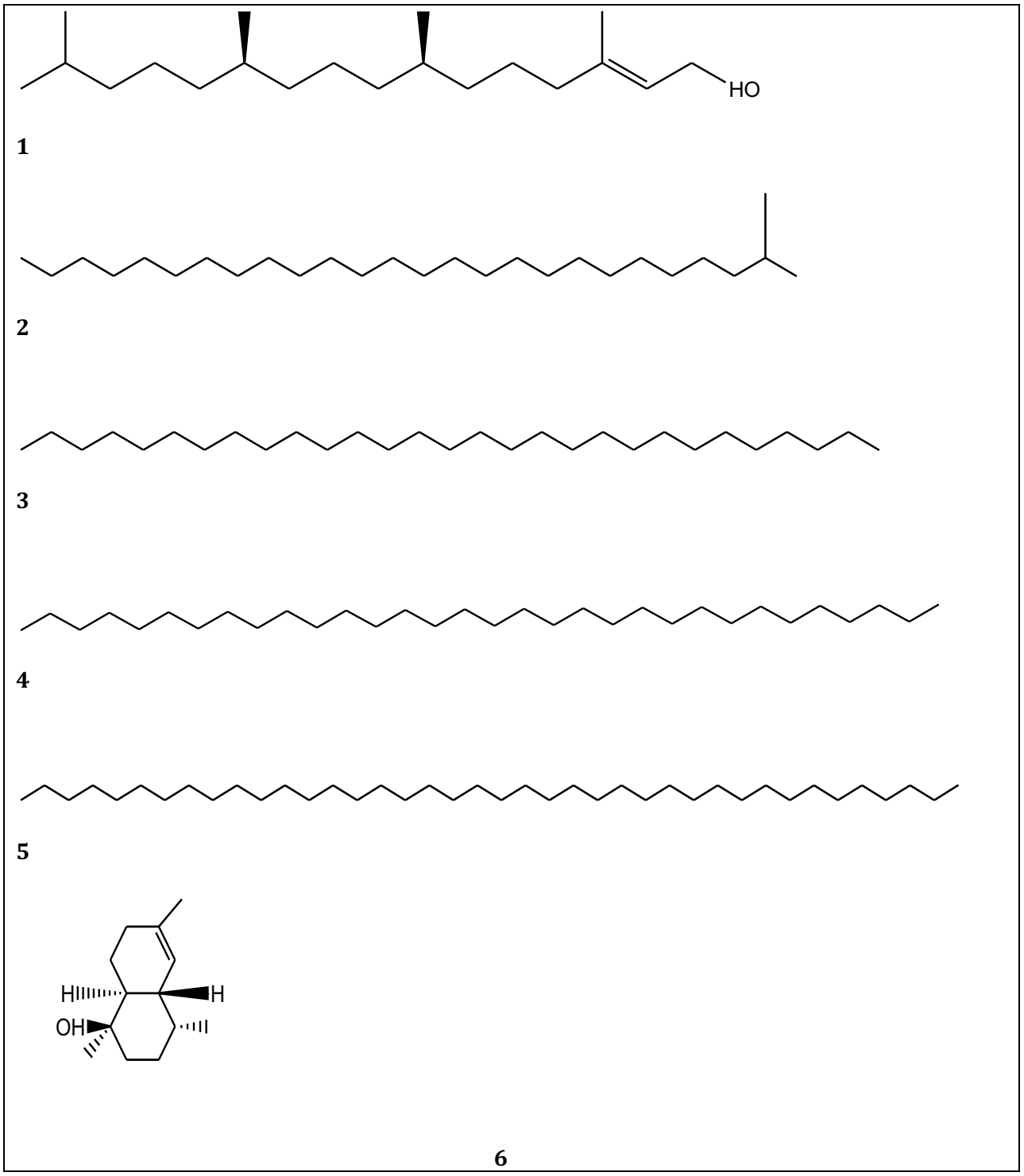

Figure 1: Structures of phytol (1), 2-methylhexacosane (2), nonacosane (3), dotriacontane (4), tetracontane (5) and τ-cadinol (6) from *O. trichocarpum* stem bark ethyl acetate extract

Doi: https://dx.doi.org/10.4314/thrb.v21i2.6

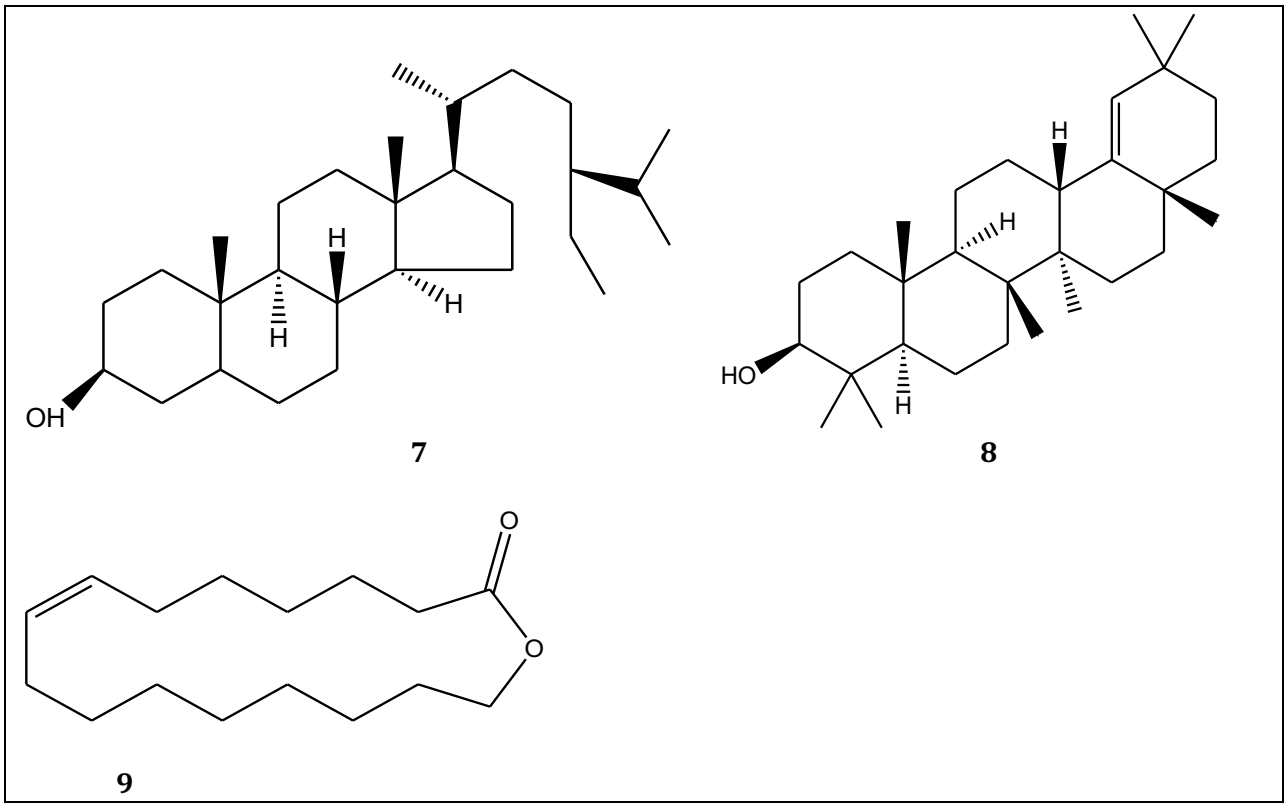

Figure 2: Structures of γ-sitosterol (7), germanicol (8) and oxacycloheptadec-8-en-2-one, (8Z) (9) from *E. divinorum* root bark ethyl acetate extract

Discussion

Validation of ethnomedical information of plants commonly used by ethnic groups has been a strategy in the discovery of novel bioactive secondary metabolites (Bonjar, 2004). Despite these efforts, there are still some plants that have never been screened for their biological potentials. That is why this study is reporting antimicrobial activity of *O. trichocarpum* and *E. divinorum* growing in Tanzania. According to Rios and Recio (2005), the maximum plant extract concentration of interest should be less than 1 mg/mL. Therefore, extracts reported in this study which displayed antimicrobial activity with minimum inhibition concentration (MIC) of 0.048 mg/mL, 0.097 mg/mL, 0.39 mg/mL and 0.781 mg/mL are therefore regarded as potential for drug sources. In this regard, the narrow MIC range of 0.097-0.781 mg/mL recorded by *O. trichocarpum* root bark ethyl acetate (OTRE) against all tested pathogenic microbes provide evidence that medium polar compounds in the root bark extract of this plant is a potent antimicrobial agent. Furthermore, the *O. trichocarpum* stem bark ethyl acetate (OTSE) and root bark aqueous (OTRA) extracts which demonstrated antimicrobial activity against *S. aureus, K. peumoniaea, S. typhi* and *C. neoformans* respectively are potential source of drug leads for the management of diseases caused by these pathogens. Findings from this study are in line with the previous study conducted by Chukwujekwu et al., (2013) which reported that leaves of *O. trichocarpum* growing in South Africa exhibited antibacterial activity against *Bacillus subtilis, Enterococcus faecalis, Escherichia coli* and *Staphylococcus aureus.*

The *Euclea divinorum* stem bark aqueous (EDSA) and root bark petroleum ether (EDRP) extracts selectivity inhibited the growth of *S. aureus* with an MIC value of 0.781 mg/mL whereas root bark petroleum ether (EDRE) inhibited growth of *S. typhi* at an MIC value of 0.048 mg/mL. It is therefore assumed that these extracts are potential source of drug leads for the management of infections caused by *S. aureus* and *S. typhi* respectively. Likewise, the *E. divinorum* leaf ethyl acetate (EDLE) and

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aqueous (EDLA) extracts which showed antimicrobial activity against *S. typhi* and *C. albicans* with MIC values of 0.781 mg/mL suggesting that the leaf of *E. divinorum* contain secondary metabolites that can be used in the management of infections caused by these pathogens. Results from this study collaborate with the previous antimicrobial investigation study of the same plant growing in Ethiopia and Yemen which found that aerial parts of *E. divinorum* possessed antimicrobial activity against *Staphylococcus aureus* and *Nesseiria gonorrhoea* (Geyid et al., 2005; Mothana et al., 2009).

In this study however, gas chromatography coupled to mass spectrometer was used to analyze ethyl acetate extracts from *O. trichocarpum* stem bark and *E. divinorum* root bark extract whereby secondary metabolites belonging to dieterpenes, alphatic hydrocarbons, tetraterpenes, sesquiterpenes, steroid and triterpenes were identified. Most of these phytochemicals have been reported to possess interesting biological activities against human infectious diseases and non-communicable diseases as shown in Table 3. Compounds that have been reported to exhibit antitumor activities are phytol and γ-sitosterol (Vivekanandadasan and Rajangam, 2015; Isaiah et al., 2016; Elangovan et al., 2015). Additionally, five compounds namely phytol, 2-methylhexacosane, nonacosane, dotriacontane and τ-cadinol from *O. trichocarpum* ethyl acetate stem bark extract have been reported to exhibit antibacterial and antifungal activities (Vivekanandadasan and Rajangam, 2015; Isaiah et al., 2016; Souti et al., 2016; Prabhadevi et al., 2012; Mihailovi et al., 2011; Soosairaj and Dons, 2016). Furthermore, the γ-sitosterol from *E. divinorum* ethyl acetate root bark extract also reported to possess the same biological activities (Elangovan et al., 2015). Compounds that have been reported to have anti-inflammatory effect include phytol and γ-sitosterol (Isaiah et al., 2016; Elangovan et al., 2015). These compounds have been identified from the *O. trichocarpum* stem bark and *E. divinorum* root bark extracts respectively. 2-methylhexacosane and γ-sitosterol from each of the tested extract have been reported to possess hypocholesterolemic activity while oxacycloheptadec-8-en-2-one, (8Z) from *E. divinorum* root bark extract reported to have antiasthmatic activity (Souti et al., 2016; Soorya et al., 2017). The reported biological activities of identified volatile fractions of ethyl acetate extracts from *O. trichocarpum* stem bark and *E. divinorum* root bark contribute to the drug discovery of such plants, despite of their traditional use by many communities in Africa to treat a number of ailments including infectious diseases (Boer et al., 2005; Iwu, 2012; Kariuki et al., 2014).

Conclusion

The extracts of *O. trichocarpum* and *E. divinorum* exhibit varying degrees of antimicrobial activities against four bacterial and two fungal species namely *S. aureus, K. pneumonia*, *E. coli, S. typhi*, *C. albicans* and *C. neoformans* respectively. A majority of the extracts confirmed to possess antimicrobial potency and thus the use of such plant materials as alternative to synthetic antimicrobial agents has shown promising results. Additionally, the presence of vast number of phytochemicals in these two botanicals, justifies its use for various ailments in Africa. However, characterization of secondary metabolites from these plants which could contribute in the drug discovery remained unveiled and therefore needs further investigations.

Competing interests: The authors declare that they have no competing interests.

Ethical approval: Not applicable

Consent for publication: Not applicable.

Availability of data and materials: The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Funding: Government of Tanzania through the University of Dodoma (UDOM)

Authors' contributions: This work was carried out in collaboration between all authors. Authors MK and UR designed the study. Authors CR and GS managed analyses of the study and literature searches. Author MK wrote first draft of the manuscript. All authors read and approved the final manuscript.

Acknowledgement: Authors are grateful to the Government of Tanzania through the University of Dodoma for financial support. Herbalists from Esilalei and Imbibya villages are appreciated for supply of ethnomedical information of medicinal plants.

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