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Chemical composition and antibacterial activity of essential oils from the rhizomes of *Cyperus papyrus* L. grown in South Africa

[Composición química y actividad antibacterial de los aceites esenciales de los rizomas de *Cyperus papurus* L., crecidos en Sudafrica]

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Abstract: Essential oils hydrodistilled from the rhizomes of *Cyperus papyrus* L. growing wild in two localities (KwaDlangezwa and Richard's Bay) of uMhlathuze City, KwaZulu-Natal Province, South Africa has been studied. The major components of KwaDlangezwa oil were caryophyllene oxide (12.7%), cyperene (10.2%) and 1,8-cineole (8.4%). The oil of Richard's Bay comprised mainly of caryophyllene oxide (24.4%), humulene epoxide II (13.2%), aristolene (9.1%) and aromadendrene epoxide II (7.3%). The antibacterial activity of the oils was assayed using agar-disc diffusion and broth-microdilution methods. The minimum inhibitory concentration (MIC) revealed that the oil samples inhibited the growth of *Staphylococcus aureus* (ATCC 3983 and ATCC 6538), with MIC of 1.25 and 0.31 mg/mL for each oil. *Streptococcus faecalis* (ATCC 29212; MIC of 1.25 and 0.6 mg/mL, respectively) and *Escherichia coli* (ATCC 4983; MIC of 1.25 mg/mL for both oils). Only the Richard Bay oil showed activity against *Bacillus cereus* and *Bacillus pumilus* with MIC of 1.25mg/mL, respectively.

Keywords: *Cyperus papyrus*, Cyperaceae, essential oil composition, caryophyllene oxide, antibacterial activity.

Resumen: Los aceites esenciales hidrodestilados de los rizomas de *Cyperus papyrus* L., que crecen en dos localidades (KwaDlangezwa y Bahía Richard) de la ciudad de uMhlathuze, la provincia KwaZulu-Natal, de Sudafrica han sido estudiados. Los mayores componentes del aceite de KwaDlangezwa fueron óxido de cariofileno (12,7 %), cipereno (10,2 %) y 1,8-cineol (8,4%). El aceite de la bahía de Richard consistió principalmente cariofileno (24,4 %), epóxido II de humuleno (13,2%), aristoleno (9,1%) y epóxido II de aromandreno (7,3%). La actividad antibacterial de los aceites fueron ensayados utilizando la difusión en discos de agar y el método de microdilución en caldo. La concentración mínima inhibitoria (CMI) reveló que las muestras inhibieron el crecimiento de *Staphylococcus aureus* (ATCC 3983 y ATCC 6538), con una MIC de 1,25 y 0,31 mg/ml de cada aceite. *Streptococcus faecalis* (ATCC 29212; CMI de 1,25 y 0.6 mg/mL, respectivamente) y *Escherichia coli* (ATCC 4983; CMI de 1,25 mg/mL para ambos aceites). Solo el aceite de la bahía Richard mostró actividad contra *Bacillus cereus* y *Bacillus pumilis* con CMI de 1,25 mg/mL, respectivamente.

Palabras clave: *Cyperus papyrus*, Ciperaceae, composición de sus aceites esencailes, óxido de cariofileno, actividad antibacterial

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Abbreviation List
v/w- volume by weight, GC-Gas Chromatography, GC-MS-Gas Chromatography coupled with Mass spectrometry, IZ-zone of inhibition, MIC-minimum inhibitory concentration, ATT-American type culture collection, USA, CSIR-Council for scientific and industrial research, South Africa.

INTRODUCTION

Cyperus papyrus L. (Cyperaceae) is a monocotyledon and perennial plant with loutish rhizomes growing up to 2.5 m height (Pooley, 1998). It is native to Africa, Madagascar and the Mediterranean countries (Goetghebeur, 1998). In Southern Africa, it is limited to the lower altitude and warmer parts of Namibia, Botswana, Limpopo, Mpumalanga and KwaZulu-Natal (Pooley, 1998). *Cyperus papyrus* has been reportedly used for various purposes; the most famous is the papyrus paper (Roberts, 1963). In addition, the plant has been the subject of intense ecological studies centered on its prodigious growth rate and ability to recycle nutrients (Hamed *et al*., 2012). The ethanol extract from the tubers of the plant have also been reported to show both antioxidant and cytoprotective properties (Hamed *et al*., 2012). Today, *C. papyrus* is widely cultivated as an aquatic ornamental plant (Oakes, 1990).

Although, the chemical compositions of essential oils of *Cyperus* species of different origins have been published, and the major compounds identified in the oil samples comprised mainly of ubiquitous monoterpenes and sesquiterpenes (Kilani *et al*., 2005; Olawore *et al*., 2006; Lawal & Oyedeji, 2009a; Lawal & Oyedeji, 2009b; Lazarević *et al*., 2010; Rameshkumar *et al*., 2011; Feizbaksh *et al*., 2012; Aghassi *et al.*, 2013; Nassar *et al*., 2015). Also, there are few reports on the antibacterial activity of essential oils of some species of the genus *Cyperus* (Kilani *et al*., 2005; Oladosu *et al*., 2011; Bisht *et al*., 2011). However, literature information on the essential oil composition of *C. papyrus* is scant; expect that of Cameroonian and Egyptian species (Sonwa, 1999; Hassanein *et. al*., 2014) and no previous study has reported any biological activity of *C. papyrus* essential oil, to the best of our knowledge.

In continuation of our studies on the chemical composition and biological activities of essential oils of *Cyperus* species from South Africa (Lawal & Oyedeji, 2009a; Lawal & Oyedeji, 2009b; Lawal *et al*., 2015), this paper aims at investigating the *in vitro* antibacterial activity against some common bacteria pathogens and to determine the chemical composition of essential oils of *C. papyrus* growing wild in two different locations in the city of uMhlathuze, KwaZulu-Natal Province, South Africa.

MATERIALS AND METHODS

Plant materials

Fresh materials of *C. papyrus* were randomly collected from wild plants growing along stream banks at KwaDlangezwa village, a local settlement with little farmland and Richard's Bay, an industrial area. The plant sample was identified by Mrs. N. R. Ntuli, Department of Botany, University of Zululand. Voucher specimens, LAWAL, OA 04 (ZULU) and LAWAL 07 (ZULU) respectively for KwaDlangezwa and Richard's Bay samples, were deposited in the University Herbarium.

Isolation of essential oils

The air-dried and pulverized rhizomes (800 g each) of *C. papyrus* material were subjected to separate hydrodistillation for 8 h in a Clevenger-type apparatus according to the British Pharmacopoeia specification (1980). The essential oil isolated was collected in a sealed sample tube and stored under refrigeration until analysis.

Gas Chromatography (GC) *analysis*

GC analysis was carried out on a Hewlett Packard Gas Chromatography HP 6820 equipped with FID detector and DB-5MS column (60 m x 0.25 mm i.d., film thickness was 0.25 µm) and the split ratio was 1:25. The oven temperature was programmed from 50º C (after 2 min) to 240º C at 5º C/min and the final temperature was held for 10 min. Injection and detector temperatures were maintained at 200º C and 240º C respectively. Hydrogen was the carrier gas at a flow rate of 1 mL/min. 0.5 µL of the diluted oil was injected into the GC. Peaks were measured by electronic integration. *n*-Alkane was run at the same condition for retention indices determination.

Gas Chromatography-Mass Spectrometry (GC/MS) *analysis*

GC/MS analyses of the essential oils were performed using a Hewlett Packard Gas Chromatography HP 6890 equipped with a DB-5MS capillary column (30 m x 0.25 mm i.d., film thickness 0.25 µm) interfaced with Hewlett Packard 5973 mass spectrometer system. The oven temperature was programmed from 70 – 240º C at the rate of 5º C/min. The ion source was set at 240º C and electron ionization at 70 eV. Helium was used as the carrier gas at a flow rate of 1

mL/min, with split ratio of 1:25. Scanning range was 35 to 425 amu. 1.0 µL of diluted oil in hexane was manually injected into the GC/MS.

Identification of constituents

The identification of constituents was performed on the basis of retention indices (RI) determined by co-injection with reference to a homologous series of *n*-alkanes, under identical experimental conditions. Further identification was performed by comparison of their mass spectra with those from home-made MS library built up from pure substances and components of known essential oils, as well as by comparison of their retention indices with literature values (Joulain & König, 1998; Adams, 2007).

Test bacterial strains

Cyperus papyrus essential oils were tested against twelve reference bacterial strains obtained from Applied and Environmental Microbiology Research Group (AEMREG), Department of Biochemistry and Microbiology, University of Fort Hare, Alice, South Africa. These microbes were Gram-positive bacteria: *Bacillus cereus* (ATCC 10702), *Bacillus pumilus* (ATCC 14884), *Staphylococcus aureus* (ATCC 3983), *Staphylococcus aureus* (ATCC 6538) and *Streptococcus faecalis* (ATCC 29212) and Gram-negative strains: *Enterobacter cloacae* (ATCC 13047), *Escherichia coli* (ATCC 4983), *Kiebsiella pneumoniae* (ATCC 2983), *Proteus vulgaris* (ATCC 6830), *Proteus vulgaris* (CSIR 0030), *Pseudomonas aeruginosa* (ATCC19582) and *Serratia marcescena* (ATCC 9986). The stock cultures were maintained at 4º C in Müeller-Hinton agar (Oxoid, UK).

Determination of Antibacterial Assay

The antibacterial activity of *C. papyrus* essential oils was measured by the disc-diffusion method (Viljoen *et al*., 2006). The microorganisms were grown overnight at 37º C in 10 mL of Müeller Hinton Broth (Oxoid, UK) for 24 h. The cultures were adjusted with sterile saline solution to obtain turbidity comparable to that of McFarland Nº 0.5 standard (1.0 x 10^8 CFU/mL). Petri dishes containing Müeller Hinton agar were inoculated with these microbial suspensions. Concentrations of 40 mg/mL of each extract were prepared. Sterile Whatman Nº 1 (6 mm) discs paper was placed on the surface of the seeded agar plates and 10 µL of the extract in (1% DMSO solution) was applied to the filter paper disk. The plates were incubated overnight at 37º C for 24 h and the diameter of resulting zones of inhibition (mm) was measured. Each experiment was performed in triplicate. Standard antibiotic agent, chloramphenicol (25 µg) and 1% DMSO solution were used as positive and negative control respectively.

Determination of the minimal inhibitory concentrations (MIC)

Broth microdilution method was used to determine the minimal inhibitory concentrations (MIC) of the oils (Eloff, 1998). Bacterial cultures were incubated in Müller-Hinton (MH) broth overnight at 37 ºC and a 1:1 dilution of each culture in fresh Müller-Hinton broth was prepared prior to use in the micro dilution assay. 100 µL of bacterial culture of an approximate inoculum size of 1.0 x 10^8 CFU/mL was added to all well and incubated at 37º C for 24h. After incubation, 40 µL of 0.2 mg/mL *p*-iodonitotetrazolium violet (INT) solution was added to each well and incubated at 37º C. Plates were examined after about 30 min. of incubation. Microbial growth is indicated by the presence of a reddish colour, which is produced when INT, a dehydrogenase activity detecting reagent, is reduced by metabolically active microorganisms to the corresponding intensely coloured formazan. MIC is defined as the lowest concentration that produces an almost complete inhibition of visible microorganism growth in liquid medium. Solvent control (1% DMSO solution) and chloramphenicol were included in the assay.

Statistical analysis

Data were subjected to One-Way Analysis of Variance (ANOVA) followed by test of significance using Graph Pad Prism. Data are presented as mean ± standard error of the mean (SEM).

RESULTS AND DISCUSSION

The yields of the oils obtained from the hydrodistillation procedure were 0.10% (v/w) and 0.08% (v/w) respectively for KwaDlangezwa and Richard's Bay smaples, calculated on a dry weight basis. Both oils were pale yellow in colouration. Forty-eight and forty compounds, accounting for 95.5% and 90.4% of the total oil contents, were identified from KwaDlangezwa and Richard's Bay oils, respectively. The percentage of each constituent and the retention indices are summarized in Table 1, according to their elution order on a DB-5MS column. The classes of compounds identified in oils are sesquiterpene hydrocarbons (31.7% and 24.6%), oxygenated sesquiterpenes (26.5% and 57.8%) and monoterpenes (19.1% and 6.0%).

The main constituents of KwaDlangezwa and Richard's Bay oils were caryophyllene oxide (12.7% and 24.4% respectively), cyperene (10.2% and 3.7% respectively), humulene epoxide II (5.4% and 13.2% respectively) and rotundene (5.0% and 2.5% respectively). Quantitative and qualitative variations were observed between the oil compositions. For example, 1,8-cineole (8.4%) and di-isobutyl phthalate (6.4%) were identified only in KwaDlangezwa oil while aristolene (9.1%) and aromadendrene epoxide II (7.3%) were the other compounds present only in Richard's Bay oil. The variation in the chemical composition of the essential oil samples could be due to environmental conditions, geographical location, chemotypes and other factors which can influence essential oil composition (Loziene & Venskutonis, 2005).

In a previous report on the essential oils of *C. papyrus* from Egypt, myrtenol, cyperene and copaene were found to be the major compounds identified in higher concentrations, along with other constituents such as, *cis*-carveol, α-pinene, β-pinene, eucalyptol, caryophyllene, rotundene, germacrene D, *trans*-calamenene and cyperone (Hassanein *et al.,* 2014). In addition, cyclosativene, α-copaene, sativene, cyperene and rotundene were reported as the major constituents of the rhizome oil of *C. papyrus* from Cameroon (Sonwa, 1999). However, in our result (Tables 1), myrtenol was not detected in our samples, while, copaene was present only in small concentration. Also, comparing the present data with the compositional studies from the essential oils of several species of the genus *Cyperus* that has been previously reported (Lawal & Oyedeji, 2009a; Lawal & Oyedeji, 2009b; Lazarević *et al*., 2010; Rameshkumar *et al*., 2011; Feizbaksh *et al*., 2012; Aghassi *et al.,* 2013), although, some of our major constituents such as aristolene, di-isobutyl phthalate and aromadendrene epoxide II has not been previously reported as constituents of *Cyperus* essential oils. However, the abundance of caryophyllene oxide and cyprene in KwaDlangezwa oil makes it similar to *C. comprsessus* (Rameshkumar *et al.,* 2011) and *C. scarious* (Pandey & Chowdhury, 2002) from India. Similarly, the caryophyllene/humulene epoxide II combination as seen in Richard's Bay was observed earlier in *C. bulbosus* (Kilani *et al.,* 2005) from Thailand and *C. glomeratus* from Serbia (Lazarević *et al*., 2010). Finally, the composition of the essential oils of *C. papyrus* from South Africa shows that it is sesquiterpenoids rich, which makes it similar to most of other reported species of *Cyperus* growing in different parts of the world (Lawal & Oyedeji, 2009a; Lawal & Oyedeji, 2009b; Lazarević *et al*., 2010; Rameshkumar *et al*., 2011; Feizbaksh *et al*., 2012; Hassanein *et al.,* 2014).

The oil of *C. papyrus* from Richard's Bay displayed higher activities than the KwaDlangezwa oil sample, against most of the tested microorganisms (Table 2). The mean inhibition zones (IZ) and minimum inhibitory concentrations ranging from 7.7 ± 0.5 to 27.3 ± 0.9 mm and 0.31 to 10.0 mg/mL were observed in Richard's Bay oil while 6.0 ± 0.0 to 16.7 ± 1.3 mm and 1.25 to 10.0 mg/mL were obtained KwaDlangezwa oil. The MIC results revealed that the studied oils have broad spectrum of activity against the tested organisms being pronounced with *S. aureus* (ATCC 3983; 1.25 and 0.31 mg/mL respectively), *S. aureus* ATCC 6538; 1.25 and 0.31 mg/mL respectively), *S. faecalis* (ATCC 29212; 1.25 and 0.53 mg/mL respectively) and *E. coli* (ATCC 4983; both 1.25 mg/mL). Only the oil of Richard Bay exhibited moderate activity against *B. cereus* (ATCC10702) and *B. pumilus* (ATCC 14884) with MIC of 1.25 mg/mL. These findings are in agreement with previous studies on the variability of antibacterial activities of essential oils from the same plant species collected from different locations (Boira & Blanquer, 1998; Loziene & Venskutonis, 2005), and that Gram-positive bacteria are more prone to essential oils of *Cyperus* species than Gram-negative bacteria (Kilani *et al*., 2005).

In conclusion, the observed differences between the essential oils of *C. papyrus* from South Africa, Egypt and Cameroon, and of other species of the genus *Cyperus* growing in different parts of the world could be due to environmental, climatic, water stress, nutritional status and other factors which can influence essential oil composition (Medina-Holguin *et al*., 2007; Sarah *et al*., 2011; Masarovicova & Kralova, 2013). In addition, the antibacterial activity of *C. papyrus* is comparable with those from essential oils of other *Cyperus* species (Outtara *et al*., 1997; Kilani *et al*., 2005). This may be attributed to the presence of some major components such as caryophyllene oxide and 1,8-cineole and/or synergy with other components such as, β-pinene and linalool which are known to possessed antimicrobial and bacteriostatic activities (Lahlou, 2004; Kilani *et al*., 2005; Srinivasan *et al*., 2009). Furthermore, the essential oils of *C. papyrus* which showed antibacterial activity against most of the pathogens may have potential applications on nosocomial and urinary tract infections.

Table 1
Chemical composition of *Cyperus papyrus* oils from South Africa

Compounds[a]	RI (Cal.)	RI (Lit.)	% composition	
			KwaD	RichB
α-Pinene	935	932	2.6	0.5
β-Pinene	977	974	2.7	0.5
Limonene	1030	1024	-	0.9
1,8-Cineole	1033	1026	8.4	-
cis-Linalool oxide (furanoid)	1065	1067	-	0.4
trans-Llinalool oxide (furanoid)	1082	1084	-	0.2
Terpinolene	1089	1086	0.8	0.3
n-Nonanal	1102	1100	0.7	0.2
(*E*)-Pinocarveol	1139	1135	0.8	1.0
Pinocarvone	1162	1160	0.7	0.4
α-Terpineol	1187	1186	0.8	0.5
Myrtenal	1199	1195	0.7	0.9
Verbenone	1208	1204	Tr	0.4
Citronellol	1226	1223	0.9	-
Citronellyl formate	1275	1271	0.7	-
Cyprotene	1322	1322	2.2	0.5
Isobutyl benzoate	1327	1327	0.4	-
α-Longipinene	1345	1350	0.5	-
Decanoic acid	1363	1364	2.0	0.6
Cyclosativene	1363	1369	0.2	-
α-Ylangene	1371	1373	0.7	-
α-Copaene	1376	1374	T	0.8
Cyperene	1393	1398	10.2	3.7
β-Caryophyllene	1421	1417	1.1	1.4
cis-Thujopsene	1430	1429	0.3	0.9
isoamyl benzoate	1437	1433	1.8	-
α-Humulene	1459	1452	1.1	-
Rotundene	1462	1457	5.0	2.5
α-acoradiene	1465	1464	1.3	0.6
β-Cadinene	1473	1472	-	0.9
Germacrene D	1481	1483	-	1.2
ar-Curcumene	1484	1485	0.9	1.4
β-Selinene	1489	1489	0.3	0.4
α-Selinene	1493	1498	1.0	-
δ-Amorphene	1507	1511	1.4	Tr
δ-Cadinene	1522	1522	0.8	-
cis-Calamenene	1531	1528	1.9	-
α-Calacorene	1537	1544	0.8	-
Elemol	1548	1549	0.8	2.1
(*E*)-Nerolidol	1556	1561	1.2	-
Caryophyllene oxide	1581	1582	12.7	24.4
(*Z*)-β-Elemenone	1591	1589	-	1.2
Cedrol	1596	1600	1.9	2.6

β-Oplopenone	**1605**	**1607**	**-**	**1.9**
Humulene epoxide II	**1608**	**1608**	**5.4**	**13.2**
Aromadendrene epoxide II	**1623**	**1639**	**-**	**7.3**
Elema-1,3,11 (13)-trien-12-al[b]	**1629**	**1639**	**0.6**	**2.2**
Caryophylla-2(12)-6-dien-5β-ol	**1640**	**1640**	**0.8**	**0.7**
Caryophylla-3, 8 (13)-dien-5, β -ol	**1647**	**1644**	**1.4**	**-**
***n*-Heptadecane**	**1694**	**1700**	**0.6**	**-**
α-Cyperone	**1726**	**1727**	**0.7**	**-**
Isobicyclogermacrenal	**1730**	**1733**	**1.0**	**2.2**
Aristolone	**1760**	**1762**	**-**	**9.1**
***n*-Octadecane**	**1792**	**1800**	**0.5**	**0.1**
di-Isobutyl phthalate	**1864**	**1864**	**6.4**	**-**
***n*-Nonadecane**	**1893**	**1900**	**1.7**	**1.1**
di-Butyl phthalate	**1960**	**1970**	**1.9**	**-**
Total			**95.5**	**90.4**
Monoterpene hydrocarbons			**6.1**	**2.2**
Oxygenated monoterpenes			**13.0**	**3.8**
Sesquiterpene hydrocarbons			**31.7**	**24.6**
Oxygenated sesquiterpenes			**26.5**	**57.8**
Aromatic compounds			**10.5**	**-**
Fatty acids			**3.8**	**1.2**
Others			**2.9**	**0.8**

[a]Elution order on DB-5MS column; [b] Correct isomer not identified; RI (Cal.) Experimental retention indices on DB-5 MS column, relative to C_9-C_{24} *n*-alkanes; RI (Lit.) Literature retention indices; Tr, Trace amount (< 0.05%)-Not identified; KwaD = KwaDlangezwa; RichB = Richard Bay

Table 2
Antibacterial activity of essential oils of *Cyperus papyrus*[a]

Microorganisms	KwaDlangezwa		Richard's bay		Chloramphenicol	
	IZ[b]	MIC[c]	IZ	MIC	IZ	MIC
B. cereus	8.0 ± 0.8	10.0	20.0 ± 1.3	1.25	23.7 ± 1.3	0.08
B. pumilus	8.3 ± 1.3	10.0	18.3 ± 1.3	1.25	16.3 ± 1.3	0.63
S. aureus [d]	14.7 ± 1.3	1.25	27.3 ± 0.9	0.31	16.7 ± 1.3	0.31
S. aureus [e]	15.0 ± 0.8	1.25	26.3 ± 1.3	0.31	13.7 ± 1.3	0.31
S. faecalis	16.7 ± 1.3	1.25	23.3 ± 1.3	0.63	20.3 ± 1.3	0.16
E. cloacae	7.0 ± 0.8	10.0	7.7 ± 0.5	10.0	13.3 ± 1.3	5.0
E. coli	16.7 ± 0.9	1.25	18.0 ± 0.8	1.25	23.7 ± 1.3	0.08
P. vulgaris	10.7 ± 1.2	ND	8.7 ± 0.9	5.0	21.0 ± 2.0	0.63
P. vulgaris **(CSIR)**	6.0 ± 0.0	10.0	12.0 ± 0.8	10.0	6.0 ± 0.0	ND
K. pneumoniae	ND	10.0	10.0 ± 0.8	10.0	20.0 ± 1.4	0.63
P. aeruginosa	10.3 ± 1.2	10.0	ND	ND	22.7 ± 1.7	0.31
S. marcescena	9.0 ± 1.6	10.0	11.0 ± 0.8	10.0	6.0 ± 0.0	ND

[a]*C. papyrus* essential oil - 10 µg/mL; [b] IZ: Inhibition zones diameter (mm) including diameter of sterile disc (6mm), values are given as mean ± SD (3 replicates); [c] MIC - minimum inhibitory concentration (mg/mL); [d]ATCC 3983; [e]ATCC 6538; ATTC: American type culture collection, U.S.A; CSIR - Council for scientific and industrial research, South Africa. ND- Not determined.

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REFERENCES

Adams RP. 2007. **Identification of Essential Oil Components by Gas Chromatography/Quadrupole Mass Spectrometry**. 4th ed. Allured Publishing Corporation, Carol Stream IL, USA.

Aghassi A, Naeemy A, Feizbakhsh A. 2013. Chemical composition of the essential oil of *Cyperus rotundus* L. from Iran. **J Essent Oil Bear Pl** 16: 382 - 386.

Bisht A, Bisht G.R.S, Singh M, Gupta R, Singh V. 2011. Chemical composition and antimicrobial activity of essential oil of tubers of *Cyperus rotundus* Linn. collected from Dehradun (Uttarakhand). **Int J Pharm Biomed Res** 2: 661 - 665.

Boira A, Blanquer A. 1998. Environmental factors affecting chemical variability of essential oils in *Thymus piperella* L. **Biochem Syst Ecol** 26: 811 - 822.

British Pharmacopoeia 1980. H.M. Stationary Office, London, UK.

Eloff JN. 1998. A sensitive and quick microplate method to determine the minimal inhibitory concentration of plant extracts for bacteria. **Planta Med** 64: 711 - 713.

Feizbakhsh A, Aghassi A, Naeemy A. 2012. Chemical constituents of the essential oils of *Cyperus difformis* L. and *Cyperus arenarius* Retz from Iran. **J Essent Oil Bear Pl** 15: 48 - 52.

Goetghebeur P. 1998. **Cyperaceae.** In: The families and genera of vascular plants IV: *Alismataneae* and *Commelinanae* (except Gramineae). K. Kubitzki (Ed), Springer Verlag, Berlin, Germany.

Hamed A, Soltan M, Fry J, Hammouda F, Zaki A. 2012. Antioxidant and cytoprotective properties of three Egyptian *Cyperus* species using cell-free and cell- based assays. **Pharm Crops** 3: 88 - 93.

Hassanein HD, Nazif NM, Shahat AA, Hammouda FM, Aboutable EA, Saleh MA. 2014. Chemical diversity of essential oils from *Cyperus articulatus*, *Cyperus esculentus* and *Cyperus papyrus*. **J Essent Bear Pl** 17: 251 - 264.

Joulain D, König WA. 1998. **The Atlas of Spectral Data of Sesquiterpene Hydrocarbons**. EB Verlag, Hamburg, Germany.

Kilani S, Abdelwahed A, Chraief I, Ammar RB, Hayder N, Hammami M, Ghedira K, Chekir Ghedira L. 2005. Chemical composition, antibacterial and antimutagenic activities of essential oil from (Tunisian) *Cyperus rotundus*. **J Essent Oil Res** 17: 695 - 700.

Lahlou M. 2004. Methods to study the phytochemistry and bioactivity of essential oils. **Phytother Res** 18: 435 - 448.

Lawal OA, Oyedeji AO. 2009a. The composition of the essential oil from *Cyperus distans* rhizome. **Nat Prod Comm** 4: 1099 - 1102.

Lawal OA, Oyedeji AO. 2009b. Chemical composition of the essential oils of *Cyperus rotundus* L. from South Africa. **Molecules** 14: 2909 - 2917.

Lawal OA, Ojekale AB, Oladimeji OS, Osinaike TS, Sanni AA, Simelane MBC, Mosa RA, Opoku AR. 2015. *In vitro* antioxidant activity, total phenolic and total flavonoid contents of essential oils of three *Cyperus* species (Cyperaceae). **Br J Pharm Res** 7: 52 - 62.

Lazarević J, Radulović N, Palić R, Zlatković B. 2010. Chemical composition of the essential oil of *Cyperus glomeratus* L. (Cyperaceae) from Serbia. **J Essent Oil Res** 22: 578 - 581.

Loziene K, Venskutonis PR. 2005. Influence of environmental and genetic factors on the stability of essential oil composition of *Thymus pulegioides*. **Biochem Syst Ecol** 33: 517 - 525.

Masarovicova E, Kralova K. 2013. **Plant-heavy metal interaction: Phytoremediation, biofortification and nanoparticles**: In Advances in selected plant physiology aspects. Montanaro G, Dichio B (Ed), In Tech, Rijeka, Croatia.

Medina-Holguin AL, Micheletto S, Holguin FO, Rodriguez J, O'Connell MA. 2007. Environmental influences on essential oils in roots of *Anemopsis californica*. **Hort Science** 42: 1578 - 1583.

Nassar MI, Yassine YM, Elshamy AI, El-Beih AA, El-Shazly M and Singab ANB. 2015.

Essential oil and antimicrobial activity of aerial parts of *Cyperus leavigatus* L. (Family: Cyperaceae). **J Essent Oil Bear Pl** 18: 416 - 422.

Oakes AJ. 1990. **Ornamental grasses and grasslike plants**. 1st ed. Van Nostrand Reinhold, New York, USA.

Oladosu IA, Usman LA, Olawore NO, Atata RF. 2011. Antibacterial activity of rhizomes essential oils of two types of *Cyperus articulatus* growing in Nigeria. **Adv Biol Res** 5: 179 - 183.

Olawore NO, Usman A, Ogunwande IA, Adeleke KA. 2006. Constituents of rhizome essential oils of two types of *Cyperus articulatus* L. grown in Nigeria. **J Essent Oil Res** 18: 604 - 606.

Outtara B, Simard RE, Holley RA, Piettte GJP, Begin A. 1997. Antimicrobial activity of selected essential oils against six meat spoilage organisms. **Int J Food Microbiol** 37: 155 - 162.

Pandey AK, Chowdhury AR. 2002. Essential oil of *Cyperus scariosus* R. Br. tubers from Central India. **Indian Perfum** 46: 325 - 328.

Pooley E. 1998. **A field guide to wild flowers in KwaZulu-Natal and Eastern Region**. Natal Flora Publications Trust, Durban, South Africa.

Rameshkumar KB, Sudheesh N, George V, Mohanan N. 2011. Volatile constituents of the roots of *Cyperus compressus* Linn. **J Essent Oil Res** 23: 39 - 41.

Roberts CH. 1963. **The Greek Papyri.** In: The Legacy of Egypt. Edited by Glanville, S.R.K Clarendon Press, Oxford, UK.

Sarah K, Amir M, Hassan S, Khodayar H, Ahmad K. 2011. The effect of drought stress on growth parameters, essential oil yield and constituent of Peppermint (*Mentha piperita* L.). **J Med Plants Res** 5: 5360 - 5365.

Sonwa MM. 1999. **Isolation and structure elucidation of essential oil constituents: Comparative study of the oils of *Cyperus alopecuroide, Cyperus papyrus,* and *Cyperus rotundus***. Ph.D Dissertation, University of Hamburg, Germany.

Srinivasan GV, Sharanappa P, Leela NK, Sadashiva CT, Vijayan KK. 2009. Chemical composition and antimicrobial activity of the essential oil of *Leea indica* (Burm. f.) Merr. Flowers. **Nat Prod Rad** 8: 488 - 493.

Viljoen AM, van Vuuren SF, Gwebu T, Demirci B, Baser KHC. 2006. The geographical variation and antimicrobial activity of African wormwood (*Artemisia afra* Jacq.) essential oil. **J Essent Oil Res 18:** 19 - 25.