Research Article

Essential oil of *Elionurushensii* (Schum) from "Plateau des Cataractes" in Congo-Brazzaville: Variation of the Chemical Composition and Evaluation of the Antioxidant Activity

^{1, 2}Aubin Nestor Loumouamou, ^{1, 2}Kevin Bikindou, ^{1, 3}Thomas Silou, ^{4, 5}Pierre Chalard and ⁶Gilles Figueredo

¹Equipe Pluridisciplinaire en Alimentation et Nutrition (EPRAN), Faculté des Sciences et Techniques, Université Marien Ngouabi, BP 389, Brazzaville,

² Institut National de Recherche en Sciences Exactes et Naturelles (IRSERN), Cité Scientifique de Brazzaville, BP 2400, Congo

³Ecole Supérieure de Technologie (EST Cataractes), Brazzaville, Congo

⁴Université Clermont Auvergne, SIGMA Clermont, Institut de Chimie de Clermont-Ferrand, BP 10448, F-63000 Clermont-Ferrand,

⁵CNRS, UMR 6296, Institut de Chimie de Clermont-Ferrand, F-63178 Aubière,

⁶Laboratoire d'analyse des extraits végétaux et des arômes (LEXVA) Biopole, Clermont-Limagne, St.

Beauzire, France

Abstract: The aim of the present work is to study the seasonal variation of the chemical profile of the essential oil extracted from *Elionurushensii* and to evaluate its antioxidant activity. *Elionurushensii* is usually used by local people as theiform drink to relieve aches. The essential oil analyzed by GC-MS presents a different chemical composition in accordance with its origin (aerial part or root). The oil of the aerial part (stems, leaves and flowering tops) is rich in *para*-menthe dienolisomers (38-49%), the main ones are: *the trans-para*-mentha 1 (7), 8-dien 2-oi (10.45-20.83%), *cis-para*-mentha 1 (7), 8-dien-2-oi (10.30-18.81%), *trans-para*-mentha -2,8-dien-1-oi (5.32 and 16.22%) and *cis-para*-mentha-2,8-dien-1-oi (3.85-6.39%). The roots are rather rich in aristolone (14-48%) and her chemical structure was confirmed by the analyzes in ¹H and ¹³C NMR. The limonene is also a leading major both of the essential oil of the aerial part (2.21-19.28%) than that of the roots (9.73-30.44%). The antioxidant activity of the oil was evaluated. The essential oil of the aerial part distillates enriched in major compounds present an inhibiting power DPPH greater than that of complete essential oil, but low compared to that of vitamin C.

Keywords: Aristolone, antioxidant activity, elionurushensii (schum), para-menthadienols

INTRODUCTION

The *Elionurushensii* (Family: *Poaceae*) is distributed in tropical and subtropical regions of South America (Brazil and Argentina), Africa (Congo, Gabon, Democratic Republic of Congo, Angola) and Australia (Yang *et al.*, 2013). The species belongs to the *Elionurus* genus of which there are about twenty species. However, these species have not yet been the object of extensive scientific studies. Even so, we have found in the literature few studies concerning *Elionurusmuticus*, *Elionuruselegans* and *Elionurusviridulus*. The studies of the volatiles components of the aerial parts of *Elionurusmuticus* acclimated in Brazil and Zimbabwe have given respectively as majority compounds: β -caryophyllene and spathulenol (Hess *et al.*, 2007; Scramin *et al.*, 2000), the neral and geranial (Chagonda *et al.*, 2000). The essential oil of the roots of *Elionurusmuticus* is rich in aristolone (Chagonda *et al.*, 2012). The essential oil of the aerial parts and roots of *Elionoruselegans* contains mainly campherenone, *epi*- β -santalene oxide, caryophyllene and bisabolone (Mevy *et al.*, 2002). The β -acorenone was identified as a major compound of the essential oil of the aerial parts of the *Elionurusviridulus* acclimated in Argentina (Hefendehl and Fonseca, 1978). Moreover, the essential oil from aerial parts and roots of *Elionuruselegans* exhibits antibacterial, antifungal and antioxidant activity (Mevy *et al.*, 2002). A good antioxidant activity has also been reported

Corresponding Author: Aubin Nestor Loumouamou, Equipe Pluridisciplinaire en Alimentation et Nutrition (EPRAN), Faculté des Sciences et Techniques, Université Marien Ngouabi, BP 389, Brazzaville, Congo This work is licensed under a Creative Commons Attribution 4.0 International License (URL: http://creativecommons.org/licenses/by/4.0/).



Steam and leaves flowers



Roots

Fig. 1: Roots, steam, leaves and flowers of Elionurus hensii

in a study of phenolic compounds in methanol extracts of *Elionurusmuticus* (Dzingirai *et al.*, 2007; Muchuweti *et al.*, 2006). As we can see, some work on the species of the genus *Elionurus* shows that essential oils have a high variability in terms of chemical profile, which has the advantage of offering multiple possibilities for use in various fields of life, depending on the chemical or biological properties of oils.

In Congo-Brazzaville, Elionurushensii grows naturally in a very limited area of Congolese territory, the "plateau des cataracts". It is usually used by local people as theiform drink to relieve aches. Therefore, they attribute to Elionurushensii antalgic properties. This species has not been extensively studied, except those initiated as part of our team research program (Silou et al., 2006; Yang et al., 2013). The essential oil of the aerial parts (stems and flowering tops) and roots of this species revealed two distinct chemical profiles. The aerial parts are rich in monoterpene alcohols, menthadienol isomers: cis and trans-para-mentha-2, 8 (9) -dien-1-ol, cis and trans para-mentha-1 (7), 8-dien 2-ol. The roots are rather rich in sesquiterpene compounds, the main one is aristolone. These studies, which cover only the occasional analyzes of the oils derived from plant matter very localized (loufouloukari) and harvested at a specific time of the vegetative cycle of the plant, don't allow characterizing a significant potential variability of chemical profile and properties that result. Our present study aims to examine the stability of the chemical profile of any oils throughout the development of the growing season, since the species grows in an area that has significant climatic variations: from day to September, during which the plant is suffering from water stress because the rains are rare, and from October to April which is the rainy season. This study is also to confirm the chemical structure of major compound of the essential oil from the roots, supposedly to be of the aristolone.

MATERIALS AND METHODS

Plant material: The plant material consists of the aerial parts (stems, leaves and flowering tops) and roots was

harvested on the site of Loufoulakari (Congo-Brazzaville) at different times of the year, from February to December 2013 (Fig. 1). These harvest periods include periods of drastic reduction in rainfall (dry season) and periods of heavy rainfall (rainy season).

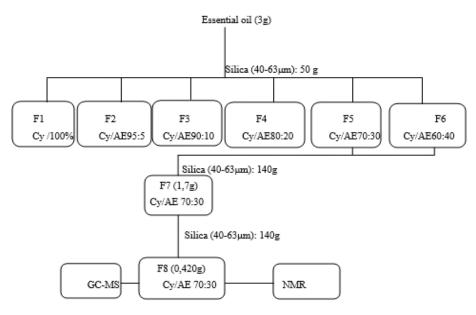
Method of production of the essential oil: After drying of the plant material, the essential oil has been obtained by vapohydrodistillation using Clevenger type apparatus (Clevenger, 1928). Each time, 300 g of vegetable material, consisting either only of roots or stems, leaves and young flowers are placed in a flask with 500 mL of water and subjected to distillation. The organic phase from the distillation is separated from the aqueous phase by extraction with diethyl ether. The organic phase thus obtained is dried over anhydrous sodium sulfate to remove traces of water and the essential oil is recovered after evaporation of the diethyl ether and then move on to the chromatographic analysis immediately.

Determination of the chemical composition:

Analysis by Gas Chromatography (GC): Quantitative analysis of the essential oil is made by means of a phase chromatograph Sparkling Agilent 6890 model, equipped with a DB5 column ($20m \times 0.18mm \times 0,18\mu m$). The oven temperature is 50°C for 3.2 min and then increased to 300°C at a rate of 10°C per min, the injector is 280°C. This device is equipped with a Flame Ionization Detector (FID) hydrogen (40mL/min)/Air (450 mL/min). The flow rate of carrier gas (hydrogen gas) is 1 mL/minute.

Analysis by coupling gas chromatography and mass spectrometry (GC-MS): Qualitative analysis is performed using a chromatograph phase Gaseous Agilent 7890 model coupled with an Agilent Mass Spectrophotometer, model 5975, equipped with a DB5 column (20m×0.18mm×0.18µm). The oven temperature is 50°C and remains constant for 3.2 min and then increased to 300°C at a rate of 8°C per minute, the injector is 280°C. Ionization is done by electron impact at 70 eV. The flow rate of carrier gas (helium) is set to 0.9 mL/min. The identification of the compounds has been done by comparison of their mass spectra and their Kovats Indices (KI) with those of databases Adams (2012), National Institute of Standards and Technology (NIST) (2008) and Konig et al. (2001) and those of laboratory.

Splitting of the essential oil: Three grams of essential oil has been fractionated on an open column containing 50 g of silica (40-63 μ m). The height of the silica in the column was 25cm. The eluent selected system is a mixture of increasing polarity solvents cyclohexane and ethylacetate (Fig. 2). Parallel chromatographic



Cy: cyclohexane; AE : ethyl acetate

Fig. 2: Fractionation diagram of the essential oil of the roots

monitoring on a silica plate (TLC) was performed to better identify the fractions containing the compound to be isolated.

F8 fraction (0.420 g) containing white crystal was analyzed by GC/MS and NMR (1 H and 13 C).

Identification of aristolone:

Determination of melting point: The fraction F8 has been consisted essentially of white crystals. We have determined the melting point of these crystals on a Buchi-5480 brand apparatus. A few milligrams of crystals had been introduced into a capillary tube and then heated until liquefied. The temperature has been measured and compared with that of the literature.

NMR analysis: The NMR spectra (¹H and ¹³ C) were recorded on a Brukeradvenced at 100 MHz to ¹³C and 400 MHz for ¹H. The solvent used was deuterated chloroform. The chemical shifts δ are given relative to the reference standard (TMS):

¹H NMR δ (ppm): 5.67 (1H, s, H-9); 2.39 (1H, dd, J = 16,6Hz and 5.6Hz, H-1 α); 2.21 (1H, J = 15,7Hz, H-1 β); 1.79 (1H, m, H-4); 1.78 (1H, m, H-2 α); 1.68 (1H, d, j = 7 Hz, H-7); 1.49 (1H, d, J = 10Hz, H-3 α); 1.37 (1H, m, H-3 β); 1.35 (1H, d, j = 6.7Hz, H-6); 1.34 (1H, m, H-2 β); 1.22 (3H, s, H-13); 1.16 (3H, s, H-12); 1.15 (3H, s, H-14); 1.02 (3H, d, j = 7.8Hz, H-15)

¹³C NMR δ (ppm): 24.31 (C-1); 39.13 (C-2); 39.53 (C-3); 38.64 (C-4); 30.52 (C-5); 26.12 (C-6); 33.13 (C-7); 167.60 (C-8); 124.22 (C-9); 196.25 (C-10); 35.49 (C-11); ; 29.69 (C12); 16.46 (C-13); 16.26 (C-14); 22.54 (C-15)

Evaluation of antioxidant activity: DPPH test (diphenylpicrylhydrazyl) is a method widely used for the evaluation of the antioxidant capacity of the essential oils include plant extracts. We used to evaluate the antioxidant activity of essential oil of *Elionurushensii* based on the method described by Brand-Williams *et al.* (1995) and Bourkhiss *et al.* (2010) with some modifications. The principle of this method consists in measuring the inhibition of free radicals of DPPH in methanol. Indeed, the addition of an antioxidant in a methanol solution of DPPH leads to discoloration of the latter which is directly proportional to the antioxidant capacity of added product. This bleaching may be followed by uv-visible spectrometry by measuring the decrease in absorbance at 517 nm.

We proceeded as follows: 1 mL an ethanol solution (the methanol was replaced by ethanol) of DPPH at the concentration of 100μ M was mixed with 0.1 mL of the ethanolic solution of the oil prepared in different concentrations (200, 400, 600, 800, 1000 µg/mL). The mixture is then incubated protected from light and at room temperature for 15 min. Absorbance is measured at 517 nm against a blank consisting of ethanol. The reference antioxidant used is ascorbic acid (vitamin C) prepared at the same concentrations and under the same operating conditions as the essential oil solutions. Percent inhibition is calculated using the following formula:

I% = (Abs C - Abs E)/Abs C

I%: Percentage inhibitionAbs C : Control absorbanceAbs E : Sample absorbance

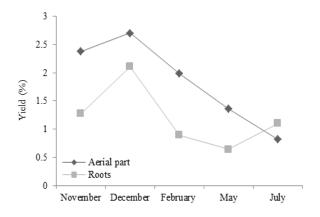


Fig. 3: Yield variation of the essential oil

RESULTS

Yield of essential oil: The yield of essential oil was determined for the different samples collected at different times of the year. The yield is more important in times of high rainfall, November and December, as shown in Fig. 3. The plant material harvested in

December provides yield of over 2.5% for the stems and 2% for the roots.

The lowest yield, 0.82% for the stems and 0.70% for roots, are obtained in May and July. These yields confirm those obtained by Silou *et al.* (2006) and Yang *et al.* (2013). Indeed, for the same periods of harvest, they have achieved the yield of 0.70 and 0.64% respectively, confirming that the plant is sensitive to water stress and this results a decrease in the yield of essential oil.

Moreover, *Elionurushensii* produces much higher yield than other species of the same genus. Indeed, with reference to available studies, the yield of essential oil of *Elionuruselegans* (Mevy *et al.*, 2002) and *Elionurusmuticus* (Hess *et al.*, 2007; Füller *et al.*, 2010) are less than 1%.

Chemical composition: Analysis by GC-MS of essential oil from samples of roots, stems, leaves and lowering tops of *Elionurushensii* harvested at different times of the year identified fifty compounds, or 80 to 98% of all compounds. The complete chemical composition is given in Table 1.

Table 1: Composition (%) of the essential oil of the aeria	l parts(stems and leaves) and roots	of Elionorushensii at different times of the year

		· · ·	Content (%)			
			February		May	
Components	KI lit	KI cal	Aerial part	Roots	Aerial part	Roots
Tricyclene	926	922	0.820	0.34	0.78	0.22
Thujene alpha	930	925	-	0.21	-	0.12
Pinene alpha	939	933	0.430	0.72	0.14	0.46
Camphene	954	950	2.480	1.09	2.41	0.91
Sabinene	975	973	0.070	0.19	-	0.16
Myrcene	990	989	0.290	0.54	-	0.85
Cymene <ortho-></ortho->	1024	1025	1.380	0.75	2.22	0.93
Limonene	1029	1029	16.99	9.73	2.21	22.80
Cineole <1.8->	1031	1032	-	3.83	0.25	3.70
Terpinolene	1088	1085	-	0.01	-	0.22
para -Cymenene	1091	1091	0.130	0.04	0.59	0.12
T- p -2.8- menthadien-1-ol	1122	1125	10.47	1.20	6.10	1.17
<i>Cis-p</i> -2.8- menthadien-1-ol	1137	1137	5.570	0.85	4.52	0.80
Pinocarveol <trans-></trans->	1139	1141	0.440	-	0.81	-
Pinocarvone	1164	1163	0.250	0.08	0.50	0.11
Terpinen-4-ol	1177	1182	0.080	0.25	-	0.62
T-p-1(7).8- menthadien-2-ol	1189	1192	10.45	2.24	20.83	3.77
Terpineol alpha	1190	1196	-	0.47	-	0.98
2-Butenal.3- methyl	1197	1201	2.690	-	5.48	-
Caranone< <i>cis</i> -4->	1200	1209	1.180	0.18	0.64	-
TransCarveol	1229	1221	4.040	0.07	7.46	1.34
<i>Cis-p-</i> 1(7).8- menthadien-2-ol	1232	1230	12.30	2.69	18.81	2.94
<i>Cis</i> -Carveol	1229	1233	0.69	0.82	-	-
Carvone	1243	1245	3.16	0.76	4.47	1.33
Piperitone	1245	1255	0.44	0.66	0.57	1.07
Perilla aldehyde	1232	1278	0.20	0.06	0.96	-
Phellandral	1271	1270	-	-	0.13	_
Bornyl acetate	1285	1287	0.97	0.29	1.66	0.63
Undecan-2-one	1294	1292	3.13	2.63	7.75	5.86
Elemene <beta-></beta->	1390	1391	5.15	0.30	-	0.08
Methyl eugenol	1403	1391	-	0.30	-	0.08
Gurjunene alpha	1403	1399	-	0.52	-	0.38
Cymene 2.5-dimethoxy-para	1409	1412	-	-0.47	-	0.04
Aristolene	1420	1413	-	0.47	-	0.39
	1423	1422	-	0.34	-	0.48
Aristola-1(10).8- diene	1429	1431	-	0.00	-	0.80

Table1: Continue						
Gurjunene <beta-></beta->	1433	1435	-	4.59	0.07	6.30
Selinene beta	1485	1488	-	-	-	1.13
Tridecan-2-one	1496	1495	1.32	1.88	4.53	2.71
Cadinene delta	1522	1521	-	0.07	-	0.08
7-epi-alpha- Selinene	1524	1522	-	0.63	-	0.89
Maaliol	1567	1575	-	1.69	-	2.14
Spathulenol	1578	1581	-	0.280	-	0.44
Intermedeol	1666	1671	1.29	2.660	3.35	5.04
Pentadecan-2- one	1697	1698	0.02	0.100	0.30	0.14
Aristolone	1763	1764	-	42.23	0.31	18.30
Hydrocarbon monoterpenes			23.09	13.79	8.63	26.73
Oxygenetedmonoterpenes			52.17	14.29	71.59	18.12
Hydrocarbon Sesquiterpenes			-	7.74	0.07	11.44
Oxygeneted Sesquiterpenes			6.810	53.61	18.09	37.23
Totals			82.08	88.84	98.39	92.20
	Content (%)				

	July		November		December	
Components	Aerial part	Roots	Aerial part	Roots	Aerial part	Roots
Tricyclene	0.470	0.100	0.350	0.35	0.89	0.30
Thujene alpha	-	0.020	-	0.22	-	0.17
Pinene alpha	0.090	0.190	0.250	0.78	0.42	0.61
Camphene	1.600	0.640	1.250	1.24	2.64	0.95
Sabinene	-	0.060	0.050	0.21	0.05	0.15
Myrcene	-	0.450	0.480	1.05	0.04	0.51
Cymene <ortho-></ortho->	1.890	0.590	0.950	1.11	1.36	0.37
Limonene	3.210	11.42	19.28	30.44	11.98	12.22
Cineole <1.8->	0.350	1.560	-	4.51	0.16	2.39
Terpinolene	-	-	0.060	0.27	-	-
para -Cymenene	0.600	-	0.360	0.26	0.21	-
T-p-2.8- menthadien-1-ol	5.320	0.650	16.22	2.15	9.06	1.16
<i>Cis-p-2.8-</i> menthadien-1-ol	3.850	0.290	6.390	1.63	6.12	0.76
Pinocarveol< <i>trans</i> ->	0.790	-	-	-	-	-
Pinocarvone	0.520	_	0.250	0.13	0.29	_
Terpinen-4-ol	-	0.200	-	0.15	-	_
T-p-1(7).8- menthadien-2-ol	18.67	1.730	12.89	3.49	10.93	1.48
Terpineol alpha	10.07	1.750	-	1.22	-	0.37
2-Butenal.3- methyl	- 4.740	-	-	1.22	-	0.57
Caranone< <i>cis</i> -4->	4.740	-	0.580	0.09	0.99	-
		-				-
TransCarveol	5.710	-	1.830	1.60	4.33	
<i>Cis-p</i> -1(7).8- menthadien-2-ol	16.46	1.310	13.52	3.23	12.66	1.60
CisCarveol		-	5.360		0.70	0.63
Carvone	4.080	0.710	3.450	1.48	3.02	0.44
Piperitone	0.550	0.370	0.410	1.02	1.45	0.19
Perilla aldehyde	0.860	-	0.070	-	0.29	-
Phellandral	0.110	-	0.080	-	-	-
Bornyl acetate	1.980	0.480	1.050	0.48	1.13	0.24
Undecan-2-one	10.14	4.330	3.520	6.20	3.44	1.97
Elemene <beta-></beta->	-	0.340	0.140	0.31	-	0.37
Methyl eugenol	-	0.310	-	0.42	-	0.19
Gurjunene alpha	-	0.180	-	0.03	-	-
Cymene 2.5-dimethoxy-para	-	0.210	-	0.52	-	0.29
Aristolene	-	0.140	-	0.35	-	-
Aristola-1(10).8- diene	-	0.450	-	0.65	-	0.54
Gurjunene <beta-></beta->	0.130	4.160	0.140	4.71	1.07	4.10
Selinene beta	-	-	-	0.75	-	-
Tridecan-2-one	7.120	2.110	1.970	2.60	3.10	1.67
Cadinene delta	-	-	-	-	-	-
7-epi-alpha- Selinene	0.100	-	-	0.68	0.09	0.57
Maaliol	-	1.740	-	1.83	-	1.23
Spathulenol	-	0.410	-	0.370	-	0.29
Intermedeol	5.300	9.630	3.190	3.030	0.01	4.50
Pentadecan-2-one	-	0.170	-	0.080	2.15	0.42
Aristolone	0.600	46.98	0.600	14.08	0.98	48.11
Hydrocarbon monoterpenes	8.24	13.50	23.07	35.97	17.60	15.53
Oxygenetedmonoterpenes	61.71	6.84	61.37	21.40	50.81	9.07
Hydrocarbon Sesquiterpenes	0.23	5.34	0.52	7.67	1.07	6.68
Oxygeneted Sesquiterpenes	25.50	66.39	10.70	29.65	10.94	58.94
Totals	95.70	92.09	95.67	94.71	80.44	90.23
Totais	95.70	92.09	93.07	94./1	80.44	90.23

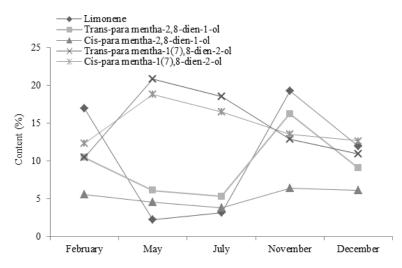


Fig. 4: Variation of main major components

Aerial part (stems, leaves and flowering tops): The essential oil of the aerial part is rich mainly in oxygenated monoterpenes whose contents range between 50.81 and 71.59% with as compounds major the isomers of para-menthadienols (trans-para-mentha-2,8-dien -1-ol, cis-para-mentha-2,8-dien-1-ol, transpara-mentha-1 (7), 8-dien-2-ol and cis-para-entha-1 (7), 8-dien-2-ol). These results confirm those obtained by Silou et al. (2006) and Yang et al. (2013). Depending on the time of sample collection, the contents of these four isomers vary between 38% and 50%. There are also some hydrocarbon monoterpenes, the most important is the limonene which is at a maximum content of around 19%, and oxygenated sesquiterpenes such as undecan-2-one and tridecan-2one. Oualitatively, the chemical profile varies very little because we found the main major compounds in the essential oil of the five samples. However, quantitatively, we find that the contents of major components vary with the harvest period. The limonene is found at low contents of around 2 at 3% for the samples collected in May and July corresponding to the period when the plant suffer from water stress (dry season period). During the same period, (i) the content of two isomers of mentha-1 (7), 8-dien-2-ol increase (20.83% and 18.57% for the *trans-para*-mentha 1 (7), 8-dien-2-ol, 18.81% and 16.46% for *cis-para*-mentha 1 (7), 8-dien-2-ol); (ii) the content of two isomers of mentha-2,8-dien-1-ol decrease (6.10% and 5.32% for the trans-para-mentha-2,8-dien-1-ol, 4.52 and 3.85% for *cis-para*-mentha-2,8-dien-1-ol) 4). (Fig. Furthermore, the contents of major sesquiterpenes also rise during the period of water stress: undecan-2-one (7.75 and 10.14%) and tridecan-2-one (4.53 and 7.12%).

Roots: The essential oil obtained from the roots is rather rich in oxygenated sesquiterpenes. These compounds represent between 37.23 and 66.39% of all

of the essential oil and the main sesquiterpene compound is aristolone (18-48%). Hydrocarbon monoterpenes are also in large quantities; they represent 13-35% of all of the essential oil and the main compound is the limonene of which content ranges between 9 and 30%. The contents of limonene and aristolone, which are the two most important major components, vary in the five samples. However, even if it seems difficult to establish a direct correlation between the biosynthesis of these compounds with the harvest period, the results show that when the content of aristolone is high that of limonene is low and vice versa. It therefore appears that the samples of May and November present a chemical profile rich in limonene, while the other three samples present a chemical profile rich in aristolone.

Identification of the major sesquiterpene of the roots: The distillation of the roots showed a mixture of oil and a few crystals glued usually on the refrigerant. We sought to know the physicochemical characteristics of the compound crystallized by taking a column fractionation of oil and a structural analysis of fractions (Fig. 2).

Melting point: The melting point obtained experimentally is 100.4 ± 1 °C. This value is the melting point given by the literature for the aristolone (99-101°C).

GC-MS Analysis: The analysis of fraction 8 (Fig. 2) by GC/MS showed a peak corresponding to a compound whose mass spectrum shows a molecular ion at m/z = 218.2 (EI-MS) corresponding to the molecular formula C_{15} H₂₂ O (Fig. 5).

NMR analysis: The chemical shifts in the ¹H and ¹³C NMR show the presence of a quaternary carbon deshielded (196.25 ppm), suggesting the existence of a

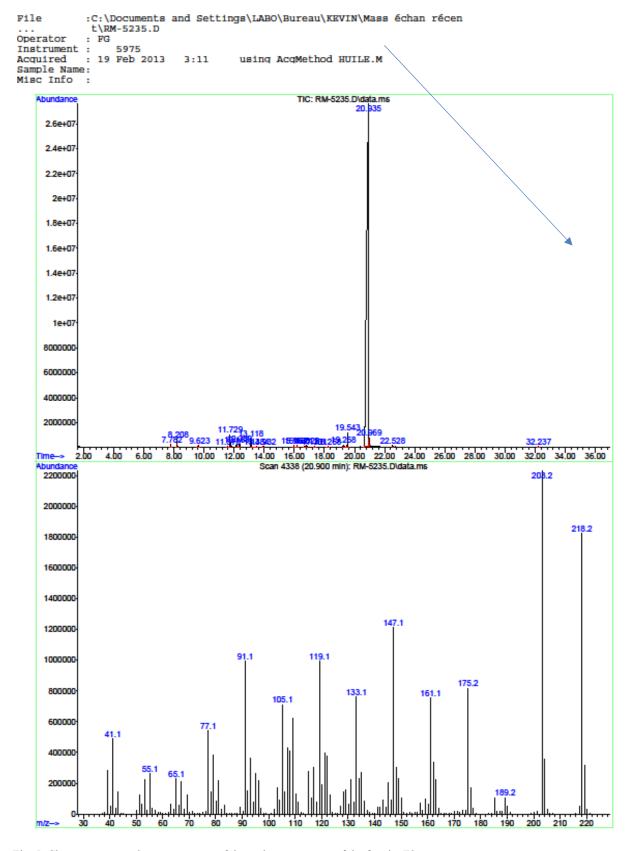


Fig. 5: Chromatogram and mass spectrum of the major component of the fractionF8

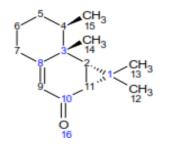


Fig. 6: Structure of aristolone isolated from the essential oil *Elionurushensii*

carbonyl moiety in the compound of the structure, two carbons deshielded (167.60 and 124.22 ppm) which correspond to sp2 carbons (signal emitting area between 170 and 140 ppm), four methyls (16.26. 16.46. 22.54 and 29.69 ppm), two armored quaternary carbons (24.31 and 39.53 ppm) three methines (35.49. 38.64 and 39.13 ppm) and three methylenes (26.12. 30.52 and 33.13 ppm).

The comparison of the NMR chemical shifts of ${}^{13}C$ observed with those described in the literature for the aristolone (Sun *et al.*, 2011; Shi *et al.*, 2009) confirms that the structure of the compound of molecular formula $C_{15}H_{22}$ O is the one proposed on Fig. 6.

Antioxidant activity: Initially, the antioxidant activity was evaluated on samples of essential oils from aerial parts and roots. The samples obtained after 3 hours of distillation (complete essential oil) have a fairly low antioxidant activity. The percentage inhibition (I%) of DPPH radical by the essential oil of the aerial part does not exceed 25% while that of vitamin C, the reference antioxidant used in our study, is around to 90% at concentrations of 800 and 1000 µg/mL. The essential oil of the roots exhibit antioxidant activity even lower (I≤15%). In all cases, we have not found a significant variation in antioxidant activity on various samples collected at different times of the year. These results confirm those reported in the literature (Yang *et al.*, 2013) for the antioxidant activity of the essential oil from the aerial part.

Furthermore, distillationkinetic of samples of the aerial part has produced successively three distillates corresponding to time 1, 2 and 3 hours of distillation (Table 2). (i) The distillate 1 is rather rich in limonene and menthadienol isomers (monoterpene alcohols); (2) the distillate 2 is rich in menthadienols isomers and intermedeol and distillate 3, low in menthadienols, is rich in ketone and alcohol sesquiterpene (2-tridecanone and intermedeol). The antioxidant activity of essential oils of three distillates is better than that of complete essential oil, although it is still low compared to that of vitamin C. The inhibition percentages of oil from distillates 2 and 3 are around40% at a concentration of 1000 mcg/mL. The essential oil of distillate3 presents

Table 2: Main major components of three distilla	ates of the aerial part
--	-------------------------

	Duration of the distillation				
Components	Distillate 1(1 h)	Distillate 2(2 h)	Distillate 3(3 h)		
Limonene	11.41	2.930	7.040		
Trans-para-mentha-2, 8-dien-1-ol	14.08	5.560	2.930		
Cis-para-mentha-2, 8-dien-1-ol	8.140	4.040	1.930		
Trans-para-mentha-1 (7), 8-dien-2-ol	16.60	10.24	4.230		
Carveoltrans	8.140	6.520	0.440		
Cis-para-mentha-1 (7), 8-dien-2-ol	17.69	19.41	5.150		
Carvone	4.390	2.670	1.090		
Undecan-2-one	4.040	4.300	4.030		
Tridecan-2-one	1.690	7.690	10.02		
Intermedeol	0.830	20.35	32.12		

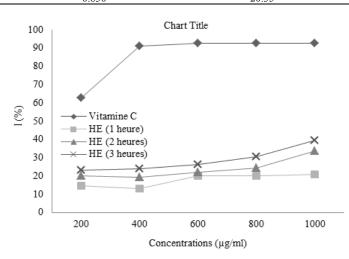


Fig. 7: Radical scavenging activity of DPPH by the essential oils of three distillates enriched in major components

an antioxidant activity more meaningful than those of the distillate 1 and 2 which are rather rich in monoterpene alcohols (isomers of menthadienol). It seems that the significant presence of the intermedeol improves antioxidant activity; however the menthadienol isomers have no significant effect on the antioxidant activity (Fig. 7).

CONCLUSION

The chemical profile of the essential oil of the roots is different from that of the oil obtained from the aerial part. The essential oil of roots appears to have two chemical profiles: a profile predominantly rich in limonene and another mainly rich in aristolone. The essential oil of the aerial part is rich mainly in isomers of para-menthadienols. However, at some periods of the year, the yield of limonene increases significantly and becomes the major constituent. This variation of the chemical profile suggests a variation of the essential oil properties. The evaluation of antalgic properties, which we considered studying, requires a differentiation of extracts according to the harvest period to better understand these effects. Furthermore, the evaluation of the antioxidant activity by the method of free radical trapping DPPH showed that the complete essential oil of *Elionurushensii* presents a moderate activity. The antioxidant activity of essential oil from the aerial part richest in monoterpene alcohols is higher than that of the roots but less than that of vitamin C. However, we have observed that the inhibiting power to the DPPH radical of the distillates rich in some components, such as the intermédeol, increases substantially. It is therefore likely that the essential oil of Elionurushensii contains the components that, once purified, may have a more significant activity.

ACKNOWLEDGMENT

The authors sincerely thank the authorities of University Marien NGOUABI (Congo) for promoted the collaboration with the school of chemistry of Clermont- Ferrand (France), this collaboration allowed to carry out the experimental work. Our thanks are also extended to the French cooperation in the Congo for the financial support we have been gratified.

REFERENCES

- Adams, R.P., 2012. Identification of Essential Oils by Capillary Gas Chromatography/mass Spectroscopy. Allured, Carol Stream, IL.
- Bourkhiss, M., M. Hnach, J. Paolini, J. Costa, A. Farah and B. Satrani, 2010. Propriétés antioxydantes et anti-inflammatoires des huiles essentielles des différentes parties de Tetraclinis articulata (Vahl) Masters du Maroc. B. Soc. Roy. Sci. Liège, 79: 141-154.

- Brand-Williams, W., M.E. Cuvelier and C. Berset, 1995. Use of a free radical method to evaluate antioxidant activity. LWT-Food Sci. Technol., 28(1): 25-30.
- Chagonda, L.S., C. Makanda and J.C. Chalchat, 2000. The essential oils of wild and cultivated *Cymbopogon validus* (Stapf) Stapf ex Burtt Davy and *Elionurus muticus* (Spreng.) Kunth from Zimbabwe. Flavour Frag. J., 15(2): 100-104.
- Chagonda, L.S., J.C. Chalchat and J.M. Bessière, 2012. Constituents of the root essential oil of cultivated *Elionurus muticus* (Spreng.) Kurth from Zimbabwe. Anal. Chem. Lett., 2(3): 177-181.
- Clevenger, J.F., 1928. Apparatus for the determination of volatile oil. J. Am. Pharm. Assoc., 17(4): 345-349.
- Dzingirai, B., M. Muchuweti, T. Murenje, C. Chidewe, M.A.N. Benhura and L.S. Chagonda, 2007.
 Phenolic content and phospholipids peroxidation inhibition by methanolic extracts of two medicinal plants: Elionurus muticus and Hypoxis hemerocallidea. Afr. J. Biochem. Res., 1(7): 137-141.
- Füller, T.N., C. Tessele, I.B.I. Barros and J.F. Barbosa Neto, 2010. Phenotypical, phytochemical and molecular characterization of "capim-carona" [*Elionurus muticus* (Spreng.) Kuntze] populations. Rev. Bras. PI. Med., Botucatu, 12(3): 261-268.
- Hefendehl, F.W. and L.R. Fonseca, 1978. Essential oil of *Elyonurus viridulus*. II. Composition of the oil of a second chemical race. Lloydia, 41: 283-285.
- Hess, S.C., M.T.L.P. Peres, A.L. Batista, J.P. Rodrigues, S.C. Tiviroli, L.G.L. Oliveira, C.W.C. Santos, L.E.S. Fedel, S.M.A. Crispim, A.S. Junior, E.F.A. Smania, A. Flach and S. Pantaroto, 2007. Evaluation of seasonal changes in chemical composition and antibacterial activity of *Elyonurus muticus* (Sprengel) O. Kuntze (Gramineae). Quim. Nova, 30(2): 370-373.
- Konig, W.A., D.H. Hochmuth and D. Joulain, 2001. Terpenoids and Related Constituents of Essential Oils. Library of Mass Finder 2.1. Institute of Organic Chemistry, Hamburg.
- Mevy, J.P., J.M. Bessiere, M. Dherbomez and J. Viano, 2002. Composition and some biological activities of the essential oils from an African pasture grass: *Elionurus elegans* Kunth. J. Agr. Food Chem., 50(15): 4240-4243.
- Muchuweti, M., L. Nyamukonda, L.S. Chagonda, A.R. Ndhlala, C. Mupure and M. Behura, 2006. Total phenolic content and antioxidant activity in selected medicinal plants of Zimbabwe. Int. J. Food Sci. Tech., 41(s1): 33-38.
- National Institute of Standards and Technology (NIST), 2008. Spectral Database for Organic Compounds. NIST WebBook: http://webbook.nist.gov/chemistry.

- Scramin, S., M.L. Saito, A. Pott and M.O.M. Marques, 2000. Essential oil of *Elyonurus muticus* (Sprengel) O.Kuntze (Gramineae). J. Essent. Oil Res., 12(3): 298-300.
- Shi, Y., H. Zhi, K. Shang, Y. Michael, A. Ali, F. Chang and Y. Chang, 2009. Chem. Biodivers., 6: 86.
- Silou, T., L. Loubaki, G. Figuérédo and J.C. Chalchat, 2006. Study of essential oil composition of *Elyonurus hensii* Schum from Congo. J. Essent. Oil Res., 18(5): 518-520.
- Sun, L.L., C.Y. Wang, H.F. Dai, C.L. Shao, W.L. Mei, T. Liu and Z.D. Ma, 2011. Chemical constituents of *Chondrophycus papillosus* and their cytotoxicity *in vitro*. Chem. Nat. Compd., 47: 650.
- Yang, Y., M.C. De Cian, S. Nsikabaka, P. Tomi, T. Silou, J. Costa and J. Paolini, 2013. Volatile fraction composition and total phenolic and flavonoid contents of *Elionurus hensii* -antioxidant activities of essential oils and solvent extracts. Nat. Prod. Commun., 8(5): 655-661.