Chemical composition of essential oils and floral waters of *Ocimum basilicum* L. from Dakar and Kaolack regions, Senegal

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Abstract: The chemical composition of essential oils and floral waters obtained by steam distillation from both fresh and dried plants of *Ocimum basilicum* L. from Dakar and Kaolack regions, Senegal were studied by GC and GC/MS. The main constituents identified in the oils were estragole and linalool. Estragole represented 73.3 and 70.2% (Dakar); 79.0 and 75.2% (Kaolack) and linalool constituted 12.8 and 11.7% (Dakar); 11.5 and 12.9% (Kaolack) in the oils from fresh and dried plants of *O. basilicum*, respectively. The most representative compounds identified in the floral waters was linalool. It was 50.5 and 51.3% in Dakar and was followed by camphor (15.4 and 17.0%), estragole (14.9 and 12.1%) and 1,8-cineole (5.9 and 6.4%). In the floral waters from Kaolack, linalool constituted 57.9 and 56.6%. Other

representative components were estragole (10.0 and 9.1%), 1,8-cineole (5.9 and 6.4%), geraniol (5.2 and 5.1%) and camphor (4.1 and 4.1%) in the floral waters from fresh and dried plants from Kaolack, respectively. This study showed that both essential oils and floral waters of *O. basilicum* from Dakar and Kaolack are characterized by the same constituents. However, oils and floral waters differ by their contents in estragole and linalool.

Keywords: *Ocimum basilicum*; Essential oils; Floral waters; Chemical composition; Estragole; Linalool.

INTRODUCTION

Ocimum genus contains several species such as *O. americanum* L., *O. canum* L., *O. basilicum* L. and *O. gratissimum* L. In Senegal, the most common species is *O. basilicum* L. It is one of the most important aroma plant that grows in the country. In west Africa, it is used for its nutritional and therapeutic properties.

Basil aerial parts are an important essential oils reserve. These oils are generally characterized by oxygenated monoterpenes (linalool, linalyl acetate, etc.) and phenolic components (estragole, eugenol, etc.). The essential oil composition can be affected by many factors like : drying methods [1,2], geographical origin [3], phenological state of harvested biomass [4], etc. Moreover different *O. basilicum* chemotypes have been previously identified: linalool, eugenol and *trans*-α-bergamotene [1]; estragole, limonene and α-phellandrene [5]; linalool, linalyl acetate, myrcene and α-terpineol [6]; linalool, methyl eugenol and 1, 8-cineole [7]; estragole, linalool, methyl eugenol and bergamotene [8]; linalool, geraniol, *p*-allylanisole, 1,8-cineole and *trans*-α-bergamotene [9].

Currently, a considerable interest is given to the use of essential oils for their biological properties. *O. basilicum* oils showed excellent antibacterial and antioxidant activities [1,9,10,11]. These antioxidant activities are undoubtly linked to the phenolic constituents [12,13]. Because they are more acceptable, safer to human health and environment than synthetic pesticides, essential oils are used in food products during storage. Indeed, *O. basilicum* is effective against *Callosobruchus maculatus* that causes a lot of damage on cowpea, one of the main sources of protein in sub-Saharan Africa [14]. It is also used against anopheles mosquitoes that transmits *Plasmodium* (malaria) parasite to humans [7,15].

Essential oils contents are generally small; their yields are less than 2.00% [16]. However floral waters that constituted their by-products are produced in high volume and can be valorized for different applications. According to the studies reported in the literature, essential oils and

floral waters are generally characterized by the same major compounds. Typical examples have been reported: *cis*, *cis*-*p*-menthenolide and pulegone for *Mentha suaveolens ssp. insularis* from Corsica [17]; pulegone, 1,8-cineole, menthone and isomenthone for *Mentha longifolia* L. from Senegal [18].

Nowadays, in this country, the study of local biodiversity of essential oil bearing plants and the chemical composition of essential oils are not sufficiently studied. The objective of this original study is to characterize essential oils and floral waters of *O. basilicum* L. from Dakar and Kaolack, two regions of Senegal where the plant is endemic. The results are part of a broad international program directed towards the valorization of essential oils in Senegal.

EXPERIMENTAL

Plant material and essential oils extraction

About 5 kg of *Ocimum basilicum* L. plants were collected in winter 2015 in two regions of Senegal: Dakar (14° 45' N, 17° 20' W) and Kaolack (14° 09' N, 14° 30' W). Flowering plants used were harvested before the sunrise (7h-8h) and divided into two portions. One portion was kept fresh and the another portion was dried in the shade at room temperature (18-27°C) for 14 days. A voucher specimens (KD_1 and KL_1) were deposited in the herbarium of the "Institut Fondamental d'Afrique Noire de l'Université Cheikh Anta Diop de Dakar". For essential oils extraction, 100 g of each both fresh (F) and dried (D) plants were separately subjected to steam distillation for 30 min using a steam generated outside the system and piped through biomass on a Clevenger-type apparatus. The essential oils (EOs) and floral waters (FWs) obtained were stored in amber vials at 4 °C until analysis.

Essential oils characterization

Essential oils and floral waters samples were subjected to gas chromatography. Essential oils solutions: 10 mg/10 ml (EOs/*n*-hexane) were prepared and diluted four times before analysis. Organic substances from floral waters were extracted by liquid-liquid with *n*-hexane (10/2,

v/v). 1 µl of these both solutions was injected by analysis. In GC/FID as in GC/MS, the chromatographic conditions were identical. The injector (Splitless mode) and detector temperatures were 280 °C and 290 °C, respectively. The oven temperature was programmed as follows: initial temperature 40 °C (5 min), ramp of 8 °C/min until final temperature 280 °C (5 min). The carrier gas was helium at a constant rate set at 1.5 ml/min. Air and hydrogen flows were 350 ml/min and 35 ml/min, respectively. The column used was a fused silica capillary, Optima-5-MS-Accent (Macherey-Nagel, Düren-Germany), 5% phenyl-95% methylsiloxane (30 m x 0.25 mm, 0.25 µm film thickness).

GC/FID-A Trace Ultra GC from Thermo Electron Corporation (Interscience Louvain-La-Neuve, Belgium) fitted with a flame ionization detector was used for the quantification of the constituents of essential oils. The percentage of each constituent was calculated as the ratio of peak area on the total of GC peak areas.

GC/MS-Identification of components was carried out on a mass spectrometer from Agilent 5973 Network Mass Selective Detector Quadrupole coupled to a gas chromatograph Agilent 6890N (G1530N), USA. Mass spectra were recorded at 70 eV and the mass scanned range was from 50 to 550 amu. Essential oils constituents were identified by comparing their retention indices and mass spectra with those from a computerized database (Wiley 275 L) and those given in the literature [19,20]. Identification of the major components were confirmed by comparing their GC data and mass spectra with those from pure compounds whenever commercially available. For that purpose, stock solutions (10 mg/10 ml *n*-hexane) of estragole (A29208), linalool (L2602), camphor (148075) and 1,8-cineole (C80601) from SIGMA ALDRICH (Boornem, Belgium) were prepared and diluted enough to obtain symmetrical peaks when using the afomentionned chromatographic conditions. The retention data of pure compounds were compared with those of essential oils constituents. The tolerance limit was ± 0.1 min.

RESULTS AND DISCUSSION

Essential oils

The yields and chemical compositions of the essential oils of *O. basilicum* obtained in the two regions showed some variations after drying. In Dakar as in Kaolack, the fresh plants yielded more essential oils than the dried plants. Essential oils yields were of 0.30 and 0.20% (Dakar), 0.27 and 0.24% (Kaolack) in the fresh and dried plants, respectively. Brada et al. (2011) [6] and Dabiré et al. (2011) [1] reported higher yields: (0.7%) and 1.10-0.79% with the same species from Algeria and Burkina Faso. The reduction of the yield in the Senegalese *O. basilicum* oils after drying can be explained by the logic loss of volatile constituents that may evaporate during the drying process [18]. Chromatographic study revealed 19 (F) and 21 (D) components in the oils from Dakar. For Kaolack, 21 (F) and 24 (D) components were identified. The prominent constituents of EOs were estragole and linalool in the two regions for both fresh and dried plants material (table 1). In the EOs from Dakar, estragole decreased from 73.3 (F) to 70.2% (D) and linalool represented 12.8 (F) and 11.7% (D). Other components which showed lower rates were camphor (2.7 and 2.3%), 1,8-cineole (2.4 and 2.9%), α-cadinol (2.0 and 1.9%) and *trans*-α-bergamotene (1.8 and 4.5%) in the fresh and dried plants, respectively. In the oils from Kaolack, estragole also decreased from 79.0 (F) to 75.2% (D) and linalool constituted 11.5 (F) and 12.9% (D). 1,8-Cineole represented 1.5 and 2.8%, α-cadinol 1.2 and 1.0% and *trans*-α-bergamotene 0.9 and 2.1% in the fresh and dried plants, respectively. The EOs from Dakar and Kaolack that were dominated by the same major compounds (estragole and linalool), differ mainly by their rate of estragole. This latter was higher in Kaolack than Dakar. Both estragole and linalool constituted 86.1 (F) and 81.7% (D) (Dakar) and 90.5 (F) and 88.1% (D) (Kaolack) of the total oils content. Similar compositions to that of the Senegalese oils have been reported in the literature. Indeed, Ngom et al. (2014) [8] obtained 38.8% estragole, 19.5% linalol, 10.0% methyl eugenol and 8.5% bergamotene in *O. basilicum*

oils. Chalchat et al. (2008) [5] identified in the Turkish EOs, estragole (52.6%), limonene (13.6%) and *exo*-fenchyle acetate (11.0%). In addition, it is noted that the proportions reported for estragole, 52.6 [5] and 38.8% [8] were lower than those obtained in the present EOs. Estragole proportions as high as those obtained in *O. basilicum* oils are usually identified in *Artemisia dracunculus* L. EOs (68.8-82.1%) [21]. For linalool, higher proportions have been indicated: 48.7% [1], 44.7% [6], 19.5% [8] and 69.9% [9]. The oils studied in this work could constitute a new chemotype. To validate this assumption a more comprehensive series of analyses on a lot of *O. basilicum* samples from all over the country is mandatory and ongoing research activities are performed in order to address this particular issue.

The use of estragole is submitted to restrictions [22]. Indeed, European Union fixed maximum levels in foodstuffs which naturally contain this compound: 10mg/kg in non-alcoholic beverages and 50mg/kg in dairy products, fish-based products, fruits, vegetables, nuts and transformed seeds [22]. *O. basilicum* essential oils may have very important biological properties because estragole belongs to the phenolic group which is correlated to the antioxydant activities [12,13]. Since EOs are considered as mixtures of natural products, *O. basilicum* extracts can be valorized in many fields such as phytotherapy, nutrition, crops protection, etc. [12,13,14]. Protection of stored products against insect pests is one of the major problems of agriculture in Africa [23]. To do this, the use of *O. basilicum* extracts as biopesticides is a valuable approach for the development of natural biopesticides (like fumigants) specially designed for stored product management in local granaries [23]. Ongoing studies on the antioxydant and insecticidal properties of EOs of *O. basilicum* will also provide useful information about their benefits and local valorization in Senegal.

Floral waters

In the floral waters of *O. basilicum*, 21 and 29 constituents were identified in the samples from Dakar, 28 and 31 constituents in Kaolack extracts from fresh and dried plants,

159 respectively (table 2). The most representative compound identified in the FWs was linalool.
160 Its proportions were of 50.5 (F) and 51.3% (D) in the samples of Dakar origin. It was
161 followed by camphor and 1,8-cineole that increased whereas estragole decreased. Camphor
162 constituted 15.4 and 17.0%, estragole represented 14.9 and 12.1% and 1,8-cineole was of 5.9
163 and 6.4%. In Kaolack, linalool was revealed for 57.9 (F) and 56.6% (D) in the FWs. Other
164 representative components identified in FWs from Kaolack were estragole (10.0 and 9.1%),
165 1,8-cineole (5.9 and 6.4%), geraniol (5.2 and 5.1%) and camphor (4.1 and 4.1%). The rate of
166 linalool was higher in FWs from Kaolack than those from Dakar and conversely for estragole
167 and camphor. The percentage of 1,8-cineole was identical in the two regions and geraniol was
168 only identified with a high content in FWs from Kaolack. FWs were mainly characterized by
169 the same compounds than EOs. But contrarily in the EOs where estragole was more important
170 than linalool, in the FWs, linalool was higher than estragole. Similar observations have been
171 previously reported by Sutour (2010) [17]. This author determined pulegone (44.4%) and *cis*,
172 *cis*-*p*-menthenolide (27.3%) as major components in the EOs of *Mentha suaveolens* ssp.
173 *insularis* from Corsica. Like EOs, FWs were also rich in *cis*, *cis*-*p*-menthenolide (67.3%) and
174 pulegone (14.8%) [17]. Diop et al. (2016) [18] also identified mainly the following constituents:
175 pulegone, 1,8-cineole, menthone and isomenthone in the EOs and FWs of *Mentha longifolia*
176 from Senegal. But for this latter, pulegone was major in both EOs and FWs [18]. Linalool, the
177 prominent constituent of FWs, has been widely identified as the main constituent of several
178 essential oils of *O. basilicum*: 48.7-80.7% [1], 44.7% [6] and 52.4% [7]. A similar composition to
179 that of the FWs has been described for *O. basilicum* oils from Kenya [3]. These latter contained
180 essentially 32.6% camphor and 28.2% linalool [3]. However, the particularity of the Senegalese
181 floral waters is the presence of linalool, estragole, camphor and 1,8-cineole in high rates; this
182 has never been reported in *O. basilicum* extracts to our knowledge. According to studies

reported in the literature, EOs as rich in linalool as the FWs from Senegal showed larvicidal activity against *Anopheles subpictus* [7].

CONCLUSION

This study undertaken on essential oils and floral waters of *Ocimum basilicum* L. from Senegal showed very interesting chemical compositions. According to the origin, little variations were noted in the chemical composition for both EOs and FWs of *O. basilicum*. However, between oils and floral waters, quantitative variations were observed in the content of the major constituents. Essential oils dominated by estragole (superior than 70.0%) with a proportion of linalool between 11.0 and 13.0% have been extracted from plants originating from Dakar and Kaolack; it is hypothesized that they could constitute a new chemotype. Floral waters composition was different of those from essential oils. They were dominated by linalool (50.0 to 58.0%) and also contained lower proportions of 1,8-cineole, camphor, estragole and geraniol. The study presented herein also supports further scientific and technical investigations to suggest optimal use of *O. basilicum* EOs and FWs in the more general context for the valorization of endemic Senegalese biodiversity.

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280 **Table 1: Chemical composition of essential oils of *Ocimum basilicum* L. from Senegal**

No.	Compounds	Retention times (min)	Retention indices	Dakar		Kaolack	
				Fresh plants	Dried plants	Fresh plants	Dried plants
1	β-Pinene	14.2	982	-	-	-	0.1
2	Myrcene	14.3	990	0.3	0.8	0.2	0.4
3	Limonene	15.3	1033	0.2	0.3	0.2	0.2
4	**1,8-Cineole**	15.4	1037	**2.4**	**2.9**	**1.5**	**2.8**
5	(*E*)-β-Ocimene	15.6	1047	-	0.9	0.5	0.6
6	Terpinolene	16.5	1090	0.1	-	-	-
7	**Linalool**	16.7	1100	**12.8**	**11.7**	**11.5**	**12.9**
8	**Camphor**	17.8	1157	**2.7**	**2.3**	0.5	0.6
9	*para*-Vinylanisole	18.2	1177	0.1	-	-	0.1
10	Borneol	18.3	1181	0.3	0.3	-	0.2
11	*trans*-Linalool oxide	18.3	1183	-	0.2	-	-
12	Terpinen-4-ol	18.4	1188	-	0.1	0.4	-
13	**Estragole**	18.7	1204	**73.3**	**70.2**	**79.0**	**75.2**
14	Neral	19.3	1241	-	-	0.1	0.1
15	Geraniol	19.5	1250	-	-	0.7	0.4
16	*para*-Anisaldehyde	19.8	1263	0.3	-	-	-
17	Geranial	19.8	1270	-	-	-	0.2
18	*trans*-Anethole	20.2	1291	-	-	0.3	0.3
19	Dihydroanethole	20.5	1304	-	-	0.1	-
20	β-Elemene	22.2	1407	0.2	0.8	0.8	0.7
21	α-Gurjunene	22.4	1423	-	0.1	-	-
22	***trans*-α-Bergamotene**	22.7	1442	**1.8**	**4.5**	**0.9**	**2.1**
23	α-Guaiene	22.8	1448	-	0.4	0.2	0.2
24	β-Funebrene	22.9	1454	-	0.1	-	-
25	α-Humulene	23.2	1473	-	0.1	-	0.1
26	Germacrene D	23.6	1496	0.1	0.3	-	0.2
27	δ-Guaiene	23.9	1515	0.2	-	0.3	-
28	**δ-Cadinene**	24.1	1529	0.7	**1.4**	0.5	0.9
29	**3-Methoxycinnamaldehyde**	24.8	1579	**2.0**	0.3	0.7	0.3
30	Spathulenol	25.1	1594	0.2	0.4	0.2	0.2
31	Not identified	25.6	1632	0.3	-	0.2	0.2
34	**α-Cadinol**	26.0	1656	**2.0**	**1.9**	**1.2**	**1.0**

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282 **Table 2: Chemical composition of floral waters of *Ocimum basilicum* L. from Senegal**

No.	Compounds	Retention times (min)	Retention indices	Dakar		Kaolack	
				Fresh plants	Dried plants	Fresh plants	Dried plants
1	***cis*-3-Hexen-1-ol**	11.0	854	**3.1**	0.2	**1.5**	-
2	**Benzaldehyde**	13.7	964	-	**1.1**	0.2	**2.3**
3	**1-Octen-3-ol**	14.1	979	**2.0**	0.9	0.4	0.3
4	Myrcene	14.3	990	0.4	0.3	0.8	0.6
5	**1,8-Cineole**	15.4	1037	**5.9**	**6.4**	**5.9**	**6.4**
6	(*E*)-β-Ocimene	15.6	1047	0.1	0.1	0.3	0.2
7	Not identified	16.1	1070	0.2	0.2	0.1	0.1
8	*cis*-Sabinene hydrate	16.1	1075	0.5	1.4	0.6	**1.2**
9	*cis*-Linalool oxide	16.5	1089	0.2	0.7	0.2	0.5
10	*trans*-Sabinene hydrate	16.6	1094	0.2	0.2	0.3	0.4
11	**Linalool**	16.7	1100	**50.5**	**51.3**	**57.9**	**56.6**
12	(*E*)-6-Methylhepta-3,5-dien-2-one	16.8	1105	0.4	0.7	0.4	0.8
13	*endo*-Fenchol	17.0	1115	-	0.1	-	0.1
14	*allo*-Ocimene	17.2	1128	0.2	0.1	-	0.1
15	**Camphor**	17.8	1157	**15.4**	**17.0**	**4.1**	**4.1**
16	Not identified	17.9	1164	0.1	0.2	-	-
17	*para*-Vinylanisole	18.2	1177	0.6	0.6	0.6	0.7
18	**Borneol**	18.3	1181	**3.2**	**3.2**	**2.7**	**2.5**
19	*trans*-Linalool oxide	18.3	1183	-	0.1	-	-
20	Terpinen-4-ol	18.4	1188	0.7	0.2	**2.0**	**1.9**
21	*para*-Cymen-8-ol	18.4	1189	-	0.3	-	0.2
22	**Estragole**	18.7	1204	**14.9**	**12.1**	**10.0**	**9.1**
23	Berbenone	18.9	1215	-	-	0.1	0.1
24	Nerol	19.1	1224	-	-	0.4	0.4
25	Neral	19.3	1241	-	-	0.1	0.2
26	Pulegone	19.4	1246	-	-	0.2	0.3
27	**Geraniol**	19.5	1250	0.4	0.5	**5.2**	**5.1**
28	***para*-Anisaldehyde**	19.8	1263	-	0.9	-	**2.2**
29	Geranial	19.8	1270	-	-	0.1	0.2
30	Piperitenone	21.2	1350	-	-	0.7	0.6
31	**Eugenol**	21.4	1358	0.7	0.3	**2.9**	**1.0**
34	**Methyl eugenol**	22.1	1400	0.3	0.2	**1.6**	**1.3**
35	*para*-Methoxypropiophenone	23.0	1458	-	-	0.2	-
36	α-Cadinene	24.4	1545	-	0.1	-	-
37	3-Methoxycinnamaldehyde	24.8	1579	-	0.4	-	0.3
38	α-Cadinol	26.0	1656	-	0.2	0.5	0.2

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