Progress in Essential Oils

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Bay Oil

The bay oil of commerce is obtained from the leaves of *Pimenta racemosa* (Miller) J. Moore var. *racemosa.* However, there are other varieties of *P. racemosa* that possess oils that are dissimilar to the bay oil of commerce. For the sake of thoroughness, these varieties are included in this review.

Tucker et al. (1991) examined the leaf oils of *P. racemosa* var. *grisea* (Kiaerskov) Fosb., *P. racemosa* var. *hispaniolensis* (Urban) Landrum and *P. racemosa* var. *ozua* (Urban et E. Ekman) Landrum, and compared them with a commercial bay oil produced from *P. racemosa* var. *racemosa.*

The composition of *P. racemosa* var. *grisea* was found to exist in three chemotypic forms: a geraniol-type, an (E)-methyl isoeugenol-type and a methyl eugenol-type, as shown in **T-1**. A trace amount (< 0.01%) of an isomer of (E)-2-hexenal, 1-octen-3-yl acetate and elemicin was also found in the methyl eugenol chemotype.

Four oils of *P. racemosa* var. *hispaniolensis* were all found to have different compositions, a summary of which can be seen in **T-2**.

Tucker et al. also determined that although the oils of *P. racemosa* var. *ozua* were all rich in 1,8-cineole, some variation in the other constituents was found to exist, as can be seen as follows:

α-thujene (0–0.10%)
α-pinene (1.36–2.17%)
sabinene (0–0.83%)
β-pinene (0.65–1.50%)
myrcene (0.45–0.86%)
3-octanol (0–0.20%)
α-phellandrene (0.05–0.48%)
α-terpinene (0–0.58%)
p-cymene (0.71–3.39%)
limonene (3.62–30.07%)
1,8-cineole (47.24–55.93%)
γ-terpinene (0.71–1.08%)
cis-linalool oxide[†] (0–0.01%)
trans-linalool oxide[†] (0–0.14%)
terpinolene (0.06–0.58%)
p-cymenene (0–0.19%)
linalool (0.51–1.81%)
nonanal (0–0.06%)
cis-rose oxide (0–0.04%)
cis-pinene hydrate[‡] (0–0.14%)
trans-rose oxide (0–0.14%)
trans-pinene hydrate[‡] (0–0.10%)
nonanol (0–0.05%)
terpinen-4-ol (4.00–15.67%)
p-cymen-8-ol (0.09–0.32%)
3-decanone (0.53–1.90%)
α-terpineol (6.68–15.12%)
methyl chavicol (0–0.24%)
trans-piperitol (0–0.06%)
citronellol (0–0.29%)
neral (0–0.06%)
geranial (0–0.08%)
2-undecanone (0–0.38%)
3-dodecanone (0–0.14%)
methyl eugenol (0–0.52%)
β-caryophyllene (0–0.12%)
(E)-methyl isoeugenol (0–0.56%)
caryophyllene oxide (0–0.09%)

Trace amounts (< 0.01%) of 3-octanone, (E)-β-ocimene, *cis*-sabinene hydrate, *trans*-sabinene hydrate, *cis*-piperitol and carvone were found in one of the oils. Also, the tentative identification of 9-hydroxy-2-nonanone was also reported for one of the oils.

Finally, Tucker et al. reported the analyses of two commercial samples of bay oil (*P. racemosa* var. *racemosa*). Although both oils were found to be rich in eugenol as expected, they differed in the composition of the other constituents, as shown in **T-3**.

[†] furanoid form; [‡] probably misidentification of p-menth-2-en-1-ol isomers

Percentage composition of chemotypes of the oil of *Pimenta racemosa* var. *grisea* **T-1**

Compound	(E)-Methyl isoeugenol-type	Methyl eugenol-type	Geraniol-type
benzaldehyde	0.19–0.35	< 0.01	0.03
1-octen-3-ol	0.14–0.86	0.30	0.31
3-octanone	2.15–3.02	0.92	1.23
myrcene	–	–	0.71
3-octanol	1.92–2.64	1.50	2.13
octanal	0.23–0.36	0.31	–
linalool	–	–	1.68
nonanal	–	0.07	–
3-octyl acetate	–	1.31	–
methyl chavicol	0.08–0.09	0.21	–
decanal	1.67–2.04	1.48	0.33
neral	–	–	1.06
geraniol	–	–	85.52
geranial	–	–	1.69
(E)-anethole	0.24–0.38	–	–
eugenol	–	0.08	1.47
geranyl acetate	–	–	1.23
methyl eugenol	2.46–2.47	92.60	0.30
β-caryophyllene	0.35–0.64	0.37	–
(Z)-isoeugenol	0.36–0.68	–	–
(Z)-methyl isoeugenol	0.86–1.18	–	–
germacrene D	0–0.16	–	0.46
(E)-methyl isoeugenol	85.08–86.32	0.16	–
caryophyllene oxide	–	0.06	–

Chemical composition (%) of four oils of *Pimenta racemosa* var. *hispaniolensis*

T-2

Compound	1	2	3	4
2-hexenal*	–	–	0.11	–
α-thujene	–	–	4.04	–
α-pinene	1.17	0.46	1.11	0.50
camphene	–	–	0.12	–
benzaldehyde	–	0.04	–	–
sabinene	0.19	–	1.08	–
β-pinene	0.35	0.06	0.55	0.14
3-octanone	0.06	0.37	0.12	–
myrcene	0.68	0.26	4.72	0.38
3-octanol	–	0.34	–	–
α-phellandrene	0.43	0.07	0.44	0.23
δ-3-carene	–	–	0.23	–
α-terpinene	1.18	0.04	3.66	0.57
p-cymene	6.81	1.44	8.59	2.09
limonene	4.55	2.24	0.65	2.58
β-phellandrene	–	–	0.70	–
1,8-cineole	37.96	17.57	0.05	27.50
(Z)-β-ocimene	–	–	0.51	–
(E)-β-ocimene	–	1.54	0.23	–
γ-terpinene	–	0.20	16.27	1.48
terpinolene	0.48	0.77	< 0.01	0.48
p-cymenene	–	–	1.06	–
linalool	0.89	0.30	0.26	0.43
p-mentha-1,3,8-triene	–	–	0.08	–
β-thujone	–	–	0.16	–
cis-pinene hydrate[†]	–	–	0.03	–
cis-sabinene hydrate	–	–	0.50	–
trans-sabinene hydrate	–	–	2.55	–
borneol	–	–	0.40	–
unbellulone	–	–	0.17	–
terpinen-4-ol	29.98	1.73	0.88	16.21
p-cymen-8-ol	0.19	0.13	0.27	0.10
α-terpineol	7.54	0.57	0.16	4.22
methyl chavicol	0.23	5.13	–	22.61
methyl thymol	–	–	0.32	–
chavicol	–	–	–	0.25
(E)-anethole	–	–	–	3.33
thymol	–	–	44.02	–
carvacrol	–	–	0.95	–
eugenol	–	0.23	0.02	–
α-copaene	–	–	0.10	–
methyl (E)-cinnamate	–	0.73	–	–
methyl eugenol	3.15	63.88	–	7.08
β-caryophyllene	–	–	1.54	–
trans-α-bergamotene	–	–	0.09	–
α-humulene	–	–	0.19	–
(Z)-methyl isoeugenol	–	–	–	0.15
germacrene D	–	–	0.35	–
α-selinene	–	–	0.50	–
(E)-methyl isoeugenol	–	0.75	–	8.11
δ-cadinene	–	–	0.02	–
caryophyllene oxide	–	–	0.14	–

[†] probably a misidentification of *cis*-p-menth-2-en-1-ol; * correct isomer not identified

Comparative percentage composition of two commercial samples of bay oil

T-3

Compound	Sample 1	Sample 2
α-pinene	0.47	0.46
camphene	–	0.14
1-octen-3-ol	1.74	–
β-pinene	–	0.09
3-octanone	1.10	–
myrcene	16.17	0.10
3-octanol	0.84	–
α-phellandrene	0.45	0.07
α-terpinene	0.19	0.02
p-cymenene[†]	–	0.30
limonene	3.90	2.07
1,8-cineole	1.38	1.39
(Z)-β-ocimene	0.10	0.08
(E)-β-ocimene	1.42	0.08
γ-terpinene	0.16	t
terpinolene	0.24	0.09
p-cymene	0.97	t
linalool	3.63	0.13
terpinen-4-ol	0.90	0.05
α-terpineol	0.52	0.14
methyl chavicol	0.03	0.05
chavicol	15.51	t
geraniol	0.12	–
geranial	0.22	–
eugenol	44.41	68.93
α-copaene	0.41	0.32
β-elemene	–	0.20
methyl eugenol	–	11.88
α-gurjunene	–	0.13
β-caryophyllene	0.64	7.24
α-amorphene	0.39	–
aromadendrene	–	0.09
α-humulene	0.15	1.31
allo-aromadendrene	–	0.28
β-chamigrene	–	0.17
β-selinene	–	0.33
α-selinene	–	0.63
α-muurolene	–	0.05
α-farnesene*	0.59	–
γ-cadinene	0.15	0.05
cis-calamenene	0.07	t
δ-cadinene	1.03	0.28
eugenyl acetate	–	0.51
spathulenol	–	0.12
globulol	–	0.36
α-cadinol	–	0.30

[†] incorrect identification based on GC elution order; t = trace (< 0.01%); * correct isomer not identified

Trace amounts (< 0.01%) of α-thujene, sabinene, camphor, p-cymen-8-ol, α-cubebene, β-cadinene and α-muurolol were also found in the chavicol-poor oil. It is worth noting that the sample one oil was very typical of commercial bay oil, whereas the sample two oil was atypical.

Ayedoun et al. (1996) analyzed the composition of an oil produced in the laboratory from trees grown in Benin. The oil composition was found to be as follows:

α-thujene (0.2%)
1-octen-3-ol (1.6%)
myrcene (26.6%)
α-phellandrene (0.7%)
δ-3-carene (0.2%)
limonene (3.1%)
1,8-cineole (1.3%)
(Z)-β-ocimene (0.3%)
(E)-β-ocimene (0.2%)
p-cymene (0.5%)
linalool (1.9%)
borneol (0.8%)
p-cymen-8-ol (0.6%)
α-terpineol (0.3%)
chavicol (6.3%)
eugenol (52.7%)
isoeugenol* (0.3%)
α-cadinene (0.5%)
eugenyl acetate (0.2%)
T-cadinol (0.2%)

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* correct isomer not identified

Aurore et al. (1998) examined the antimicrobial activity of the three chemical forms (neral/geranial, methyl chavicol/methyl eugenol and chavicol/eugenol) of *Pimenta racemosa* (P.Miller) J.W. Moore var. *racemosa*. They found that the bacterial strains tested were much less sensitive to the oils than the fungi screened. Furthermore, the authors noted that although the oils were both bacteriostatic and fungistatic, they were not microbiocidal. Also, there appeared to be no difference in activity amongst the three chemical forms.

An oil of *P. racemosa* that was screened by Delespaul et al. (2000) for its antifungal activity was determined to contain the following major components:

myrcene (6.3%)
p-cymene (0.3%)
3-octanone (0.4%)
limonene (1.4%)
1,8-cineole (0.1%)
linalool (2.2%)
methyl chavicol (1.3%)
terpinen-4-ol (0.6%)
methyl eugenol (5.4%)

α-copaene (1.3%)
bergamotene* (0.2%)
β-caryophyllene (4.0%)
δ-cadinene (0.5%)
chavicol (10.4%)
eugenol (60.0%)

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* correct isomer not identified

Bello et al. (2001) analyzed an oil produced from *P. racemosa* var. *racemosa* leaves collected in western Cuba. The oil, which was analyzed by GC/MS, was determined to contain the following components:

α-thujene (1.1%)
α-pinene (2.0%)
1-octen-3-ol (1.5%)
sabinene (0.3%)
β-pinene (0.3%)
octanol (0.3%)
myrcene (1.2%)
α-phellandrene (1.6%)
α-terpinene (0.1%)
p-cymene (8.0%)
1,8-cineole (20.4%)
γ-terpinene (4.6%)
p-cymenene (0.2%)
terpinolene (3.1%)
linalool (1.3%)
terpinen-4-ol (20.7%)
α-terpineol (10.0%)
methyl chavicol (0.8%)
chavicol (10.1%)
neral (0.2%)
geranial (0.2%)
(E)-anethole (0.1%)
eugenol (10.7%)
methyl cinnamate (0.1%)
methyl eugenol (0.3%)
4-allylsyringol (0.3%)

This is an unusual oil because bay oil normally contains eugenol, myrcene and chavicol as major components with eugenol being found in amounts greater than 50%.

It is of interest to note that the oils of two *P. racemosa* varieties were found to be dissimilar in composition to the normally encountered var. *racemosa* (Garcia et al. 2002). The oil of *P. racemosa* var. *terebinthina* (Burret) Landrum (known in the Dominican Republic as "canelilla") was found to contain the following components:

α-pinene (2.85%)
benzaldehyde (0.70%)
5-methyl-3-heptanone (0.70%)
dehydro-1,8-cineole (0.30%)
myrcene (t)
p-cymene (0.20%)
1,8-cineole (t)
δ-3-carene† (2.25%)
linalool (1.10%)
verbenol* (1.25%)
terpinen-4-ol (5.95%)
α-terpineol (20.00%)
neral† (2.50%)
geraniol (4.50%)
α-terpinyl acetate (27.00%)
methyl cinnamate (0.60%)

methyl eugenol (12.60%)
β-caryophyllene (2.40%)
methyl isoeugenol[*] (1.10%)
α-muurolol (5.20%)
spathulenol (0.60%)
caryophyllene oxide (0.40%)
manool (2.40%)

[†] incorrect identification based on GC elution order; [*] correct isomer not identified; t = trace (< 0.01%)

It should be noted that either α-terpinyl acetate or methyl cinnamate were incorrectly identified based on the retention indices found in the report.

In contrast, the composition of the oil of *P. racemosa* var. *grisea* (Kiarskou) Fosberg (known as "ozua" in the Dominican Republic) was found to be:

9-methyl-2-undecene[†] (0.49%)
3-octanone (2.10%)
3-octanol (0.35%)
isobutyl tiglate (0.34%)
m-cymene (0.24%)
p-cymene (0.23%)
1,8-cineole (2.33%)
linalool (t)
verbenol (t)
terpinen-4-ol (t)
octyl oxirane[†] (0.91%)
neral (0.20%)
geranial (0.30%)
(E)-anethole (t)
methyl eugenol (4.52%)
β-caryophyllene (3.59%)
methyl isoeugenol[*] (75.23%)
α-muurolene (0.49%)
γ-cadinene (t)
δ-cadinene (t)
caryophyllene oxide (0.32%)
γ-eudesmol (t)
α-muurolol (t)
γ-bulgarene[†] (t)
elemicin (t)

[†] incorrect identity based on GC elution order; [*] correct isomer not identified; t = trace (< 0.01%)

A.O. Tucker, M.J. Maciarello, R.P. Adams, L.R. Landrum and T.A. Zanoni, *Volatile leaf oils of Caribbean Myrtaceae. I. Three varieties of* Pimenta racemosa *(Miller) J. Moore of the Dominican Republic and the commercial bay oil.* J. Essent. Oil Res., **3**, 323–329 (1991).

A.M. Ayedoun, B.S. Adeoti, J. Setondji, C. Menut, G. Lamaty and J-M. Bessière, *Aromatic plants from tropical West Africa. IV. Chemical composition of leaf oil of* Pimenta racemosa *(Miller) J.W. Moore var.* racemosa *from Benin.* J. Essent. Oil Res., **8**, 207–209 (1996).

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A. Bello, J. Pino, R. Marbot, A. Urquiola and J. Aguero, *Components volátiles des plantas de la famila Myrtaceae de la región occidental de Cuba.* Revista CENIC, Cienc. Quim., **32**, 143–147 (2001).

D. García, A. Alvarez, P. Tornos, A. Fernandez and T. Sáenz, *Gas chromatographic-mass spectrometry study of the essential oils of* Pimenta racemosa *var.* terebinthina *and* P. racemosa *var.* grisea. Z. Naturforsch., **57c**, 449–451 (2002).

Western Red Cedar Leaf Oil

Limited quantities of western red cedar leaf oil (ex *Thuja plicata* D. Don) are available annually from Western Canada. Oils produced in the laboratory from leaves collected from the Prince Rupert area of British Columbia and the Saskatoon area of Saskatchewan (Canada) were subjected to analysis by von Rudloff (1962). The oils were found to contain the following components:

α-pinene (1.5–2.0%)
camphene (0.2–0.4%)
sabinene (6.3–6.4%)
δ-3-carene[†] (2.8%)
limonene (0.3–0.5%)
1,8-cineole (t–0.1%)
γ-terpinene (t–0.1%)
p-cymene (0.1–0.2%)
terpinolene (0.2–0.4%)
α-thujone (76.0–77.5%)
β-thujone (7.5–7.8%)
terpinen-4-ol (1.4–1.7%)

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t = trace (< 0.1%); [†] tentative identification

Von Rudloff and Lapp (1979) found that the oil composition of western red cedar leaf varied less than a normal sampling error, irrespective of the ecosystem within which the trees were found. They also compared the tree-to-tree variation in oil composition, a summary of which can be seen as follows:

α-thujene (0.3–0.4%)
α-pinene (1.2–1.7%)
sabinene (3.4–6.3%)
myrcene (1.0–2.3%)
δ-3-carene (0.2–0.4%)
α-terpinene (0.6–0.7%)
limonene (0.6–0.7%)
γ-terpinene (1.1–1.4%)
terpinolene (0.2–0.3%)
p-cymene (0.2–0.3%)
fenchone (0.2–0.4%)
α-thujone (72.8–77.2%)
β-thujone (7.6–8.8%)
terpinen-4-ol (2.8–3.5%)
α-terpineol (0.6–0.8%)
geranyl acetate (0.2–0.7%)
sesquiterpenes[†] (0.3–0.5%)
diterpenes[†] (0.3–0.6%)

[†] identification not determined

Von Rudloff et al. (1988) compared the main component compositions of western red cedar leaf oil obtained from ten 2 cm branchlets collected at random from eight- to 50-year-old trees grown along the costal, intermediate and interior regions of Western Canada. The results of the analyses can be seen in **T-4**.

The main components (those in amounts in excess of 0.5%) of two commercially produced oils of western red cedar leaves were found by Lawrence (1991) to be as follows:

α-pinene (1.46–2.06%)
sabinene (2.38–4.60%)
myrcene (0.52–1.43%)
limonene (0.82–0.85%)
p-cymene (1.06–2.35%)
α-thujone (79.49–82.48%)
β-thujone (6.83–7.51%)
terpinen-4-ol (1.99–2.20%)

Lopes and Kolodziejczyk (2003) analyzed a range of oils produced from the leaves of trees grown in Western Canada. Among the oils analyzed was a lab-distilled western red cedar leaf, the composition of which was determined to be as follows:

tricyclene (0.06%)
α-thujene (0.64%)
α-pinene (2.96%)
α-fenchene (1.31%)
camphene (1.39%)
sabinene (2.34%)
β-pinene (0.21%)
myrcene (1.31%)
α-phellandrene (0.08%)
α-terpinene (0.42%)
p-cymene (1.36%)
limonene (1.70%)
γ-terpinene (0.84%)
cis-sabinene hydrate (0.18%)
fenchone (13.04%)
trans-sabinene hydrate (0.43%)
α-thujone (47.81%)
α-fenchyl alcohol (0.08%)
β-thujone (8.76%)
camphor (2.35%)
borneol (0.26%)
terpinen-4-ol (2.23%)
p-cymen-8-ol (0.13%)
α-terpineol (0.37%)
α-fenchyl acetate (0.33%)
pulegone (0.10%)
methyl carvacrol (0.15%)
bornyl acetate (2.75%)
geranyl acetate (0.07%)
β-caryophyllene (0.12%)
δ-cadinene (0.14%)

In addition, trace amounts (< 0.01%) of (Z)-3-hexenol, hexanol, thuja-2,4(10)-diene, α-campholenal, *trans*-carveol, piperitone, p-cymen-7-ol, α-copaene, α-humulene, ethyl (E)-cinnamate, γ-muurolene and α-muurolene were also found in this same oil. It was a surprise that the authors identified fenchone as a constituent of western red cedar leaf oil, as it has never been previously characterized as a major constituent of the oil. Fenchone is commonly found in eastern white cedar leaf oil (ex *Thuja occidentalis*) so

Average main components (%) of western red cedar leaf oils from three different regions of Western Canada

T-4

Compound	Coastal zone oils	Intermediate zone oils	Interior zone oils
α-pinene	1.32	1.50	1.67
sabinene	4.26	4.40	3.90
myrcene	1.30	1.71	1.64
α-terpinene	0.63	0.71	0.66
limonene	0.74	0.79	0.84
γ-terpinene	1.14	1.31	1.21
linalool	0.24	0.19	0.19
α-thujone	72.05	73.27	74.31
β-thujone	8.91	7.91	7.69
terpinen-4-ol	3.19	3.48	3.10
α-terpineol	1.03	0.82	0.69
geranyl acetate	0.79	0.37	0.39
sesquiterpenes†	0.91	0.70	0.71
rimuene	0.31	0.25	0.20
beyerene	0.39	0.24	0.23

†identifications not made

Percentage composition of major constituents of ambrette seed volatiles isolated by different processes

T-5

Compound	1	2	3
decyl acetate	7.2	2.3	5.7
β-farnesene*	9.7	13.6	9.6
nerolidol*	2.2	–	–
dodecyl acetate	4.5	4.7	5.1
(Z)-5-tetradecen-14-olide	1.2	1.5	1.3
(Z)-5-tetradecenyl acetate	0.9	1.3	1.2
(Z,E)-farnesyl acetate	3.4	3.7	3.8
(E,E)-farnesyl acetate	47.6	43.7	50.4
(Z)-7-hexadecen-16-olide	9.0	9.3	9.3
(Z)-9-octadecen-18-olide	0.6	1.3	1.2

*correct isomer not identified; 1 = steam distillation; 2 = hexane extraction followed by steam distillation; 3 = methanol extraction followed by hexane extraction and fatty acid removal; all processes used whole seed

it is possible that there was some mix-up in plant material used for distillation. Otherwise, the stability of the oil composition of western red cedar leaf that was found by von Rudloff and coworkers is in question.

E. von Rudloff, *Gas-liquid chromatography of terpenes. VI. The volatile oil of* Thuja plicata *Donn.* Phytochemistry, **1**, 195–202 (1962).

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Ambrette Seed Oil

Two oils of ambrette seed (ex *Abelmoschus moschatus* Medik.) that were populated in the laboratory from seeds grown in Vietnam were subjected to analysis by Dung et al. (1999). The constituents identified in these oils were:

decyl acetate (0–0.6%)
(E)-β-farnesene (0–0.2%)
(E,E)-α-farnesene (0–0.2%)
(E)-nerolidol (0.2–0.3%)
spathulenol (0–0.3%)
caryophyllene oxide (t–0.4%)
(Z)-5-dodecenyl acetate (0.4–0.5%)
dodecyl acetate (1.7%)
(E)-2,3-dihydrofarnesol (t–0.7%)
(Z)-5-tridecenyl acetate (0–0.1%)
(Z)-oxacyclopentadec-6-en-2-one (1.4–1.5%)
(E,E)-farnesol (0–0.4%)
(E,E)-farnesal (t–0.1%)
tetradecenyl acetate* (0–1.4%)
(Z)-5-tetradecenyl acetate (0–1.2%)
(E)-2,3-dihydrofarnesyl acetate (32.9–67.3%)
(Z,E)-farnesyl acetate (t–5.1%)
(E,E)-farnesyl acetate (14.9–35.5%)
(E)-2,3-dihydrofarnesyl propionate (0.4–0.8%)
(E,E)-farnesyl propionate (0–0.4%)
Z-oxacycloheptadec-8-en-2-one‡ (3.0–5.5%)
hexadecanoic acid (t–0.4%)
(Z)-oxacyclononadec-10-en-2-one (1.0–2.5%)

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*correct isomer not identified; t = trace (< 0.1%); ‡also known as ambrettolide

Trace amounts (< 0.1%) of 2-methyl-2-propenal, 2,3-dihydro-4-methylfuran, hexanal, butyl acetate, isoamyl acetate, geranial, selin-11-en-4α-ol, (E)-2,3-dihydro-farnesyl butyrate and linoleic acid were also characterized in one of the oils.

As the essential oil is located mainly in the seed coat of *Abelmoschus moschatus* that is difficult to separate from the endosperm and embryo (kernel), the process for essential oil production by the routine steam distillation can be both time and energy consuming and not highly efficient. Because of this, Rout et al. (2003) examined the method of seed preparation and the various processes used to isolate the volatiles such as steam distillation, hexane extraction followed by steam distillation and methanol extraction. The methods examined to separate the seed coat from the embryo were (a) soaking in water followed by roller milling, (b) pounding, (c) hammer mill grinding,

(d) milling with a head-operated roller mill and (e) grinding using a domestic grinder that was fitted with a vegetable cutter. The authors found that the latter method resulted in shredding the seed coat into larger pieces while pulverizing the embryo. This was determined to be the most ideal seed preparation prior to volatile isolation. For isolation of the volatiles, Rout et al. compared steam distillation of whole seed (0.15–0.20% yield), extraction with hexane followed by steam distillation of concrete (0.20–0.25% yield), and extraction of whole seeds with methanol, followed by liquid-liquid extraction with either pentane or hexane and short path silica gel column removal of fatty acids and other higher molecular weight polar compounds (0.24–0.35% yield). The main constituents of the three volatile isolation processes can be seen in **T-5**.

Rout et al. (2004) compared the composition of a water-distilled oil with the volatile concentrate produced by the methanol, extraction/liquid-liquid extraction (pentane), followed by short column silica gel chromatography. The composition of the volatiles of the two processes are shown in **T-6**.

Comparative percentage composition of ambrette seed oil and its volatile concentrate **T-6**

Compound	Oil	Volatile concentrate
octyl acetate	–	t
octyl butyrate	–	0.1
tetradecene*	t	t
decyl acetate	7.2	5.4–5.7
(E)-β-farnesene	9.6	9.6–10.1
curcumene*	t	t
(Z,E)-α-farnesene	2.0	2.0–2.2
(E,E)-α-farnesene	4.9	5.0–5.4
(E)-γ-bisabolene	t	t–0.1
cis-sesquisabinene hydrate	t	t
(Z)-nerolidol	2.2	–
dodecenyl acetate*	0.9	0.9–1.0
(E)-β-elemenone	0.6	0.6
dodecyl acetate	4.5	4.7–5.1
tetradecanol	t	t
(E)-nerolidol acetate	0.3	0.3
(E,E)-farnesol	1.7	–
(Z)-5-tetradecenolide	1.2	1.3
2-pentadecynol	–	0.1
tetradecenyl acetate*	0.9	1.2
tetradecyl acetate	0.1	0.3
(Z,E)-farnesyl acetate	3.5	3.8–4.0
(E,E)-farnesyl acetate	47.6	50.3–50.4
methyl palmitate	0.3	0.8
(Z)-7-hexadecenolide	9.0	9.2–9.3
methyl linoleate	0.1	0.2
methyl oleate	0.2	0.3
(Z)-9-octadecenolide	0.6	1.2–1.3

*correct isomer not identified; t = trace (< 0.1%)

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