GEOGRAPHIC VARIATION IN LEAF ESSENTIAL OILS OF DOUGLAS FIR (*PSEUDOTSUGA MENZIESII*)

Robert P. Adams
Biology Department, Baylor University, Box 97388, Waco, TX 76798, USA Robert_Adams@baylor.edu

J. Jesús Vargas-Hernández
Programa Forestal - Campus Montecillo, Colegio de Postgraduados km 65.5 Carretara México - Texcoco, México C.P. 56230 México

M. Socorro González Elizondo
CIIDIR Unidad Durango, Instituto Politecnico Nacional, Sigma 119, Fracc. 20 de Noviembre II, Durango, Dgo., 34220 Mexico

Gay Hunter
Museum Curator, Olympic National Park
600 East Park Ave., Port Angeles, WA 98362

Thomas A. Fairhall
1136 Water St., Suite 101, Port Townsend, WA 98368

David Thornburg
220 W. Winchester Lane, Cottonwood AZ

and

Frank Callahan
Callahan Seeds, P. O. Box 5531, Central Point, OR 97502

ABSTRACT

The volatile leaf oils of Douglas fir (*Pseudotsuga menziesii*) were analyzed from throughout its range. The major differentiation found was the divergence of the inland populations (var. *glauca*) from the coastal and Sierra Nevada populations (var. *menziesii*). The oils of var. *menziesii* differed from var. *glauca* in their major components: camphene (0.4-17, 25-30%), β-pinene (25-38, 2.5-12%), sabinene (4-12, trace-0.5%), α-terpinene (2.1-3.3, 0.2-0.0%), γ-terpinene (3.4-5.5, 0.1-0.3%),

terpinolene (9.5-14.6, 1.2-1.6%), cis-p-menth-2-en-1-ol (0.7-0.8, 0.0-trace), terpinen-4-ol (10-12.1, trace-0.5%), and bornyl acetate (0.2-1.3, 14.6-44.7%). The oil of the Sierra Nevada population (Sierra Nevada race of Snajberk and Zavarin, 1976) was quite similar to typical coastal Doug Fir from WA and OR and not as distinct as found in the oleoresin oils of Snajberk and Zavarin. Overall the oils of the inland variety (var. *glauca*) varied from Yellowstone southward with the AZ - NM oils forming a group along with the southern Mexico oils except for the population at Cerro Potosi that had a different oil. The Cerro Potosi oil differed from other inland (var. *glauca*) oils in having larger amounts of α-pinene, β-pinene, δ-3-carene and smaller amounts of camphene, limonene and bornyl acetate. The leaf oil of var. *oaxacana* did not differ from var. *glauca* from nearby populations in southern Mexico. The leaf essential oils of *P. macrocarpa* are also reported. *Phytologia 94(2):199-218 (August 1, 2012).*

KEY WORDS: *Pseudotsuga menziesii,* var. *menziesii,* var. *glauca,* var. *oaxacana,* leaf essential oils, geographic variation, Douglas Fir.

Douglas Fir [*Pseudotsuga menziesii* (Mirb.) Franco] is a wide ranging, common forest tree in North America (Fig. 1). The nomenclatural history of the name is a morass (see http://www.plantsystematics.org/reveal/pbio/LnC/dougfir.html), but seems to be settled by James Reveal. In a recent treatment, Eckenwalder (p. 572, 2009) recognizes two varieties: var. *menziesii* and var. *glauca* (Mayr) Franco [cited as (Beissn.) Franco in Eckenwalder, 2009]. Eckenwalder (2009) did not recognize var. *oaxacana* Debreczy & Racz, described from Oaxaca (Debreczy and Racz, 1995).

The leaf essential oils of *P. menziesii* have been exhaustively studied by von Rudloff (von Rudloff, 1972, 1973, 1984, von Rudloff and Rehfeldt, 1980) who carefully documented the large differences in oils between coastal (var. *menziesii*) and inland (var. *glauca*) in many compounds including camphene (0.3-8, 20-30%), β-pinene (15-30, 5-10%), sabinene (2-12, 0.1-0.5%), α-terpinene 1-3, 0-0.3%), γ-terpinene (2-8, 0.1-1%), terpinolene (5-15, 0.5-3%), terpinen-4-ol (5-15, 0.5-3%), and bornyl acetate (0.5-5, 20-30%). However, von Rudloff's studies

were limited to the Pacific Northwest and northern US-Canada (south to Wyoming).

A second team from USDA, Forest Products, Richmond, CA (Snajberk and Zavarin), made extensive studies of the terpenoids from the oleoresin of Douglas Fir (Snajberk, Lee and Zavarin, 1974, Snajberk and Zavarin, 1976, Zavarin and Snajberk, 1973, 1975). In their most comprehensive study (Snajberk and Zavarin, 1976), they found four chemical races: coastal, northern inland, southern inland and Sierra Nevada. These are shown in Fig. 1, along with populations used in the present study. Notice the populations sampled in the present study are from each race: coastal (2), northern inland (1), Sierra Nevada (1), southern inland (2), plus Mexico (6) as well as *P. macrocarpa*. Douglas Fir in Mexico is principally found in small restricted populations, except for a larger area in Chihuahua in northern Mexico (Fig. 1). In spite of the very exhaustive studies of leaf and wood oils in the United States and Canada, nothing has been reported about variation in Douglas Fir oils in Mexico.

However, there has been work at the molecular level on Douglas Fir in Mexico. Li and Adams (1989) reported that allozymes divided the Douglas Fir into northern coastal (var. *menziesii*) and inland (var. *glauca*) with two subgroups (northern and southern inland). They did not find evidence of a subgroup of Sierra Nevada Douglas Fir as Snajberk and Zavarin (1976) found, based on the oleoresin oils. In addition, Li and Adams (1989) found a distinct pattern in the allozymes from population 103 at General Cepeda, Coah., MX and speculated that it might be *P. flahaultii* Flous (also recognized by Martinez, 1963). However, a nearby collection (104, La Encantada, near Zaragoza, NL) clustered closely with *P. menziesii* from New Mexico. So that case seems unresolved.

Gugger et al. (2010) examined mtDNA and cpDNA sequences and found support for coastal (var. *menziesii*) and inland (var. *glauca*) in the United States and Canada. No evidence was found for a Sierra Nevada taxon, but mtDNA suggested the inland (var. *glauca*) might be divided into northern and southern groups. In a subsequent study, Gugger et al. (2011) examined Douglas Fir from Mexico. They found considerable divergence in cpDNA from Cerro Potosi, NL and Jamé,

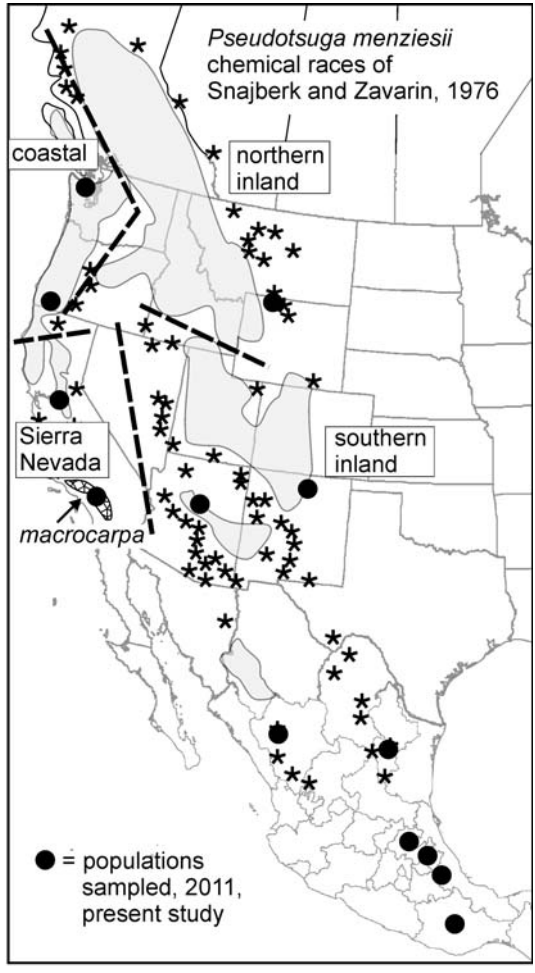

Figure 1. Distribution of *P. menziesii* with chemical races of Snajberk and Zavarin, 1976 and populations sampled in the present study.

Coah. from other Mexico populations. CpSSRs supported two clades in Mexico (Gugger, Fig. 4c), but that pattern was not supported in mtDNA (Gugger, Fig. 4a) or cpDNA (Gugger, Fig. 4b) data; in summary, they concluded that "Mexican populations were genetically

distinct from USA and Canadian populations, but more closely related to the Rocky Mountain variety than the coastal variety". As Gugger et al. (2010) did not show data from Mexico, and Gugger et al. (2011) showed only data from that country, it is difficult to ascertain the relationship of Mexican populations to those of the USA.

Phenotypic analyses (Reyes Hernández et al. 2006) revealed that *Pseudotsuga* populations of northern Mexico are morphologically similar to *P. menziesii* var. *glauca* from southwestern USA, but the populations from central Mexico differed. They also found a population of NE Mexico (San Francisco) morphologically separated from the rest, even from those of the same geographical region, suggesting an effect of isolation. This population is just 15 km NW from the one from El Potosi analyzed here.

The purpose this paper is to report on geographic variation in the leaf essential oils of *Pseudotsuga menziesii* from the USA and Mexico.

MATERIALS AND METHODS

Plant material: *P. menziesii* var. *menziesii* (coastal/ Sierra Nevada): *Adams 12918-12922*, Olympic National Forest, Port Angeles, WA, 48º 02' 48.1" N, 123º 25' 04.08" W, 525 m; *Adams 12745-12757*, on serpentine, Oregon Mtn., OR, 41º 59' 59.1" N, 123º 47' 10.2" W, 895 m; *Adams 12779-12783*, 6 km e of Buck Meadows, CA, 21 km w of Yosemite NP on US 120, 37º 49.579' N, 119º 58.421' W, 1150 m. var. *glauca*: *Adams 12556-12560*, 13 km w of Cimarron, NM on US 64, 36.54684º N, 105.03321º W, 2125 m; *Adams 12744-12748* (ex *D. Thornburg, 1-5*), 9 km ne of Pine, AZ on Hwy 87, 34º 27.422' N, 111º 24.115' W, 2250 m; *Adams 12818-12822*, 20 km e of Yellowstone NP, on US 14 at the Palisades, 44.45448º N, 109.78182º W, 1910 m; *Adams 13056-13060*, (ex *M. Socorro González Elizondo 7777a-e*), Cerro Potosi, NL, 24º 53' 9" N, 100º 13'14" W, 3141 m.; *Adams 13061-13066*, (ex *Martha González Elizondo 4408-4409, 4413-4416*) Los Altares, Dur., 25º2'56" N, 105º59'48" W, 2310 m; *Adams 13082-13087* (ex *Vargas-Hernandez J1-J6*), El Chico Natl. Park, Mineral del Chico, Hgo., 20º 10' 16" N, 98º 43' 55" W, 2,765m, Nov. 4, 2011; *Adams 13088-13094* (ex *Vargas-Hernandez C1-C7*), Cuatexmola,

Ixtacamaxtitlan, Puebla, 19º 30' 22" N, 97º 50' 21" W, 2,980m, Oct. 30, 2011; *Adams 13095-13100* (ex *Vargas-Hernandez* T1-T6), Ejido Paso National, Tlachichuca, Puebla, 19º 17' 47" N, 97º 19' 45" W, 3,000m, Sep. 30, 2011; var. *oaxacana*: *Adams 13101-13103, 13105-13106* (ex *Vargas-Hernandez I1-I6*, Paraje Peña Prieta, Oaxaca, 17º 09' 38" N, 96º 38' 07" W, 2,700m, Oct. 21, 2011. *P. macrocarpa*: *Adams 12776-12778*, USFS Eddy Arboretum, Placerville, CA. Voucher specimens are deposited in the Herbarium, Baylor University (BAYLU, CIIDIR and CHAPA).

Isolation of Oils - Fresh leaves (200 g) were steam distilled for 2 h using a circulatory Clevenger-type apparatus (Adams, 1991). The oil samples were concentrated (ether trap removed) with nitrogen and the samples stored at -20ºC until analyzed. The extracted leaves were oven dried (100ºC, 48 h) for determination of oil yields.

Chemical Analyses - Oils from 10-15 trees of each of the taxa were analyzed and average values reported. The oils were analyzed on a HP5971 MSD mass spectrometer, scan time 1/ sec., directly coupled to a HP 5890 gas chromatograph, using a J & W DB-5, 0.26 mm x 30 m, 0.25 micron coating thickness, fused silica capillary column (see Adams, 2007 for operating details). Identifications were made by library searches of the Adams Essential Oil library (Adams, 2007), using the HP Chemstation library search routines, coupled with retention time data of authentic reference compounds. Quantitation was by FID on an HP 5890 gas chromatograph using a J & W DB-5, 0.26 mm x 30 m, 0.25 micron coating thickness, fused silica capillary column using the HP Chemstation software.

Data Analysis - Terpenoids (as per cent total oil) were coded and compared among the species by the Gower metric (1971). Principal coordinate analysis was performed by factoring the associational matrix using the formulation of Gower (1966) and Veldman (1967).

RESULTS AND DISCUSSION

The leaf essential oils of *P. menziesii* var. *menziesii* (coastal) were found (Table 1) to be nearly identical to the reports of von

Rudloff (1972, 1973). This is remarkable considering the changes in gas chromatography from packed to fused capillary columns. The oils of var. *menziesii* differed (Table 1) from var. *glauca* in their major components: camphene (0.4-17, 25-30%), β-pinene (25-38, 2.5-12%), sabinene (4-12, trace-0.5%), α-terpinene (2.1-3.3, 0.2-0.0%), γ-terpinene (3.4-5.5, 0.1-0.3%), terpinolene (9.5-14.6, 1.2-1.6%), cis-p-menth-2-en-1-ol (0.7-0.8, 0.0-trace), terpinen-4-ol (10-12.1, trace-0.5%), and bornyl acetate (0.2-1.3, 14.6-44.7%). The oil from the Yosemite NP, Sierra Nevada (Yose in Table 1) population (cf. Sierra Nevada race of Snajberk and Zavarin, 1976) was quite similar to typical coastal Doug Fir from WA (ONF, Table 1) and not as distinct as found in the oleoresin oil of Snajberk and Zavarin. However, comparing coastal (ONF) and Sierra Nevada (Yose, Table 1) oils reveals the coastal oil is higher in sabinene (12.9, 4.0%), δ-3-carene (1.8, 0.5%), γ-terpinene (5.4, 3.4%), terpinolene (14.6, 9.5%) and geranyl acetate (2.1, 0.9%), but lower in β-pinene (25.5, 38.0%), bornyl acetate (0.2, 1.3%) and citronellyl acetate (1.2, 2.8%). The inland group (var. *glauca*, Yell, AZ, NM, A Du, El C, Oax, Table 1) shows some differences between the northern inland (Yell) and southern inland oils (excluding Cerro Potosi) for camphene (17.0 vs. 24-30%), α-pinene (2.5 vs 3.2-9.1%) and bornyl acetate (44.7 vs. 16-32%).

The Cerro Potosi oil (C Po, Table 1) differed from other inland (var. *glauca*) oils in having larger amounts of α-pinene, β-pinene, δ-3-carene and smaller amounts of camphene, limonene and bornyl acetate. The leaf oil of var. *oaxacana* (Oax, Table 1) did not differ from var. *glauca* oils in nearby populations in southern Mexico.

Principal Coordinate (PCO) analysis of 24 terpenoids (boldface, Table 1) excluded one (Z-β-ocimene) whose maximum value was too small (0.4%). PCO of the resulting 23 terpenoids gave eigenroots that accounted for 48.7, 15.4, 7.9, 7.5 and 5.0% of the variation before the eigenroots asymptoted. The overall trends among the oils are seen in Fig. 2 where the major Principal Coordinate (PCO) accounted 49% of the variance among the populations. The first axis separated coastal (var. *menziesii*) and inland (var. *glauca*) populations. Notice that *Pseudotsuga macrocarpa* is well resolved (Fig. 2).

To examine variation among the *P. menziesii* populations, *P. macrocarpa* was removed form the data and a new PCO performed. PCO analysis of 24 terpenoids (boldface, Table 1) excluded Z-β-ocimene (max. value 0.4%) and humulene epoxide II (max. value 0.0). PCO of the resulting 22 terpenoids gave eigenroots that accounted for 57.6, 9.4, 8.4, 5.9 and 4.2% of the variation before the eigenroots asymptoted.

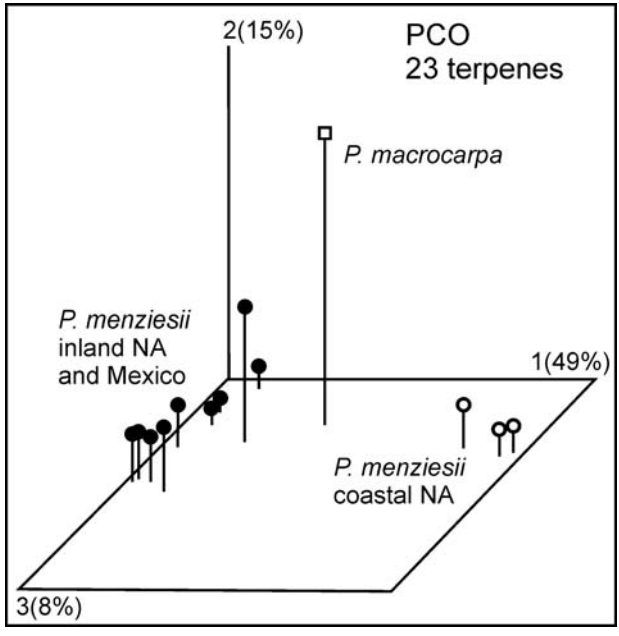

Figure 2. PCO of *P. menziesii* populations and *P. macrocarpa* based on 23 terpenes.

This resulted in an even greater amount of variance removed by PCO axis 1 (58%, Fig. 3). One can now see some differentiation among the inland populations. Note particularly the much lower similarity of Cerro Potosi oil to other inland populations (0.774), than seen between other inland populations (0.892, 0.888). There appears to be a slight north - south cline from Yellowstone - AZ, NM - Mexico populations (see tricyclene, α-pinene, camphene, β-pinene and bornyl acetate, Table 1).

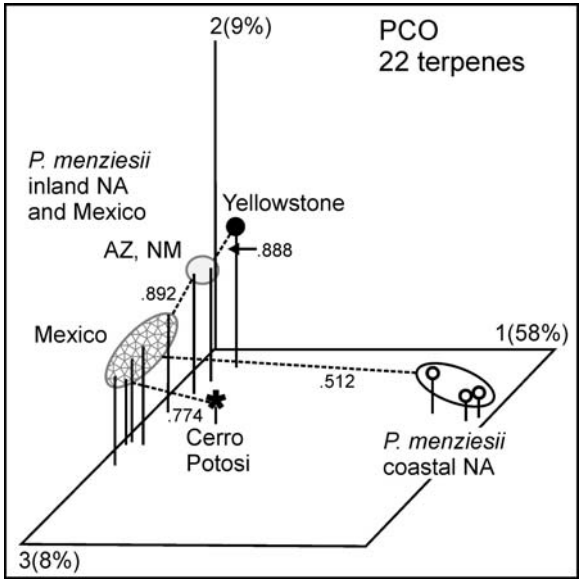

Figure 3. PCO of 12 populations of *P. menziesii* using 22 terpenes. Dashed lines are minimum spanning links. Numbers next to the lines are similarities.

Another way to visualize the clustering and similarities is by use of a phenogram. Figure 4 shows the clustering of populations based on 22 terpenes. The coastal - inland split is the major trend.

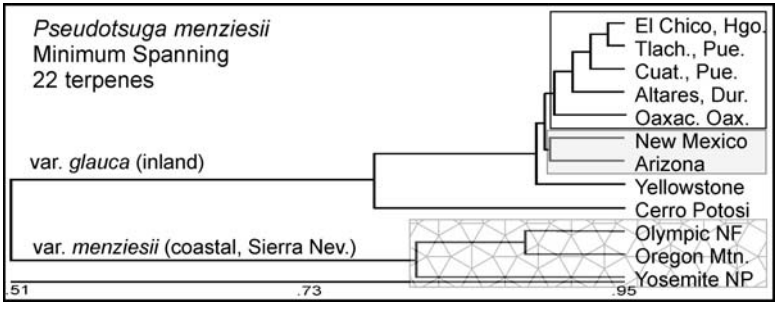

Figure 4. Minimum spanning diagram based on 22 terpenes for populations of Douglas Fir.

One can also see that the oil from the Cerro Potosi population is the most unusual oil in the inland (var. *glauca*) group (Fig. 4). In addition, the divergence of the Yosemite NP (Sierra Nevada) population is clearly seen. The Mexico populations (excluding Cerro Potosi) are very uniform in their oils, including var. *oaxacana* (Oaxac. in Fig. 4). There is no support for the recognition of var. *oaxacana*, but it may be divergent in molecular characters.

To better understand the variation, geographic clustering was preformed. The resulting diagram (Fig. 5) clearly shows the uniformity of the oils in the inland group and the divergence of the oil of Cerro Potosi from other Mexico populations. Again, the oil of var. *oaxacana* from Oaxaca is shown to be very similar to nearby populations of var. *glauca*.

These oils data are similar to the cpDNA sequencing data of Gugger et al. (2011), showing Cerro Potosi population to be different from other Mexico populations. A recent study (Wei et al. 2011) found a similar pattern in mtDNA (coastal and inland groups), but they found the cpDNA of inland Mexico populations to differ from inland USA populations. Additional molecular studies (in progress) are needed to clarify the taxonomic relationship of Cerro Potosi and inland USA populations.

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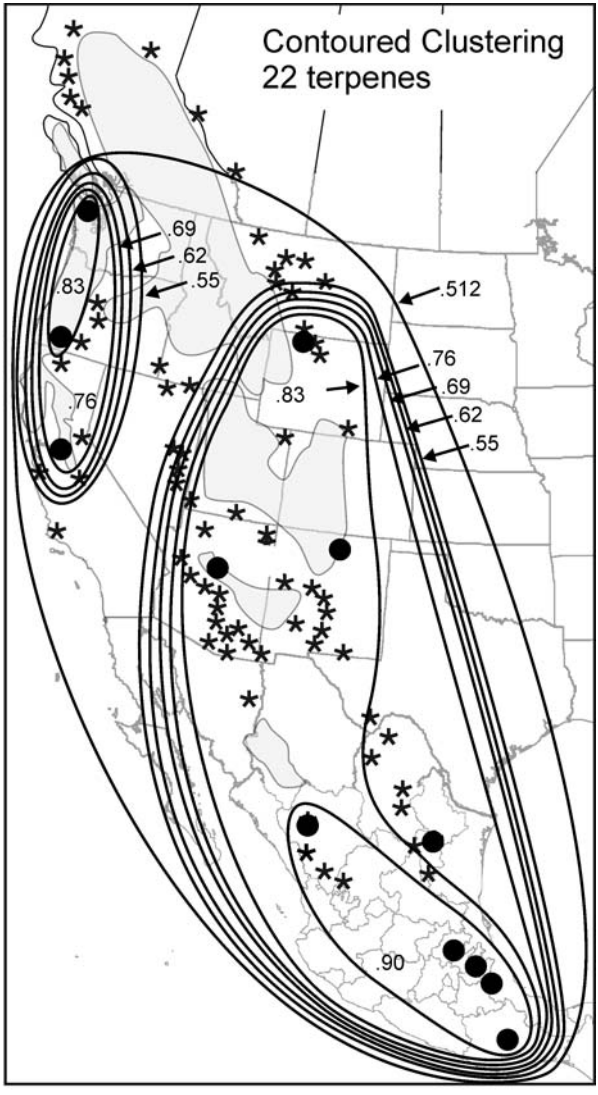

Figure 5. Contoured clustering (minimum spanning) based on equal interval clustering (0.07 similarity step intervals). Based on Fig. 4. data.

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Table 1. Comparison of leaf oil compositions for *Pseudotsuga menziesii* oils. var. *menziesii*: ONF = Olympic National Forest, Yose = Yosemite NP; var. *menziesii* var. *glauca*: Yell = Yellowstone NP, AZ = Mogolllon Rim, AZ, NM = Cimarron, NM, C Po = Cerro Potosi, NL, A Du = Altares, Dur.; var. *oaxacana*: Oax = Paraje Pena Prieta, Oax., El C= El Chico Nat. Pk., Hgo., Oax., Mac = *P. macrocarpa*. Compounds in bold face appear to separate the taxa and were used in numerical analyses.

KI	cpd	ONF	Yose	Yell	AZ	NM	C Po	A Du	El C	Oax	Mac
884	**santene**	**t**	**-**	**1.8**	**2.1**	**1.7**	**1.2**	**3.1**	**4.1**	**4.8**	**-**
900	n-nonane	-	-	-	-	-	t	-	-	-	-
921	**tricyclene**	**t**	**0.1**	**1.0**	**2.2**	**2.5**	**1.3**	**3.1**	**2.8**	**3.0**	**0.1**
924	α-thujene	0.4	0.6	-	-	-	0.1	-	-	-	-
932	**α-pinene**	**6.4**	**7.9**	**7.0**	**9.7**	**4.5**	**18.0**	**13.5**	**10.6**	**11.6**	**14.0**
946	**camphene**	**0.4**	**1.3**	**17.0**	**24.6**	**26.4**	**13.8**	**30.3**	**30.0**	**28.2**	**1.0**
969	**sabinene**	**12.9**	**4.0**	**0.1**	**t**	**t**	**0.5**	**0.3**	**0.1**	**0.4**	**t**
974	**β-pinene**	**25.5**	**38.0**	**2.5**	**8.1**	**3.2**	**12.3**	**8.1**	**9.1**	**7.8**	**16.2**
988	myrcene	1.6	1.8	1.9	1.5	2.0	7.8	3.0	7.0	1.6	9.2
998	n-octanal	-	-	t	-	-	-	-	-	-	-
1002	α-phellandrene	0.2	0.2	0.1	0.1	t	0.1	0.1	0.1	t	t
1008	**δ-3-carene**	**1.8**	**0.5**	**0.6**	**0.7**	**0.8**	**3.9**	**1.9**	**0.1**	**0.3**	**0.2**
1014	**α-terpinene**	**3.3**	**2.1**	**-**	**t**	**t**	**0.2**	**0.1**	**t**	**t**	**t**
1020	p-cymene	0.7	0.9	0.1	t	t	0.2	0.1	t	0.1	t
1024	**limonene**	**1.6**	**2.0**	**6.6**	**5.0**	**6.0**	**2.8**	**6.0**	**6.0**	**5.4**	**20.6**

KI	cpd	ONF	Yose	Yell	AZ	NM	C Po	A Du	El C	Oax	Mac
1025	**β-phellandrene**	**2.3**	**2.4**	**5.1**	**3.4**	**4.3**	**5.4**	**5.5**	**4.9**	**3.9**	**14.0**
1032	**(Z)-β-ocimene**	**t**	**-**	**-**	**-**	**-**	**0.3**	**t**	**t**	**t**	**2.9**
1044	(E)-β-ocimene	0.1	0.2	-	t	0.5	1.3	2.3	0.9	0.1	0.2
1054	**γ-terpinene**	**5.4**	**3.4**	**0.1**	**0.1**	**0.2**	**0.3**	**0.2**	**0.1**	**0.1**	**0.1**
1065	cis-sabinene hydrate	0.3	0.4	-	-	-	-	-	-	-	-
1086	**terpinolene**	**14.6**	**9.5**	**1.2**	**1.0**	**1.2**	**1.6**	**1.5**	**1.2**	**1.3**	**0.6**
1089	6-camphenone, isomer	-	-	0.3	0.1	t	t	t	0.2	t	-
1095	trans-sabinene hydrate	0.2	t	-	-	-	-	-	-	-	-
1095	linalool	0.2	1.7	1.3	1.5	3.2	t	0.7	0.4	-	0.4
1100	undecane	-	t	-	-	-	1.2	-	-	-	-
1100	n-nonanal	-	-	-	-	-	t	-	-	-	-
1118	**cis-p-menth-2-en-1-ol**	**0.8**	**0.7**	**-**	**t**	**t**	**t**	**-**	**-**	**-**	**-**
1118	endo-fenchol	t	0.1	t	0.1	t	t	t	t	-	t
1122	α-campholenal	0.5	-	0.1	-	-	0.1	-	-	t	0.1
1123	methyl octanoate	-	0.1	-	t	-	-	-	-	-	-
1131	myroxide	-	0.2	-	-	-	-	-	-	-	-
1135	trans-pinocarveol	-	-	-	-	-	t	-	-	t	-
1136	trans-p-menth-2-en-1-ol	-	0.4	-	t	t	-	-	-	-	-

KI	cpd	ONF	Yose	Yell	AZ	NM	C Po	A Du	El C	Oax	Mac
1141	camphor	t	t	0.2	0.5	0.2	t	0.4	0.5	0.3	t
1145	**camphene hydrate**	**t**	**0.1**	**2.0**	**1.7**	**1.2**	**0.3**	**1.2**	**0.6**	**0.1**	**0.1**
1148	**citronellal**	**0.5**	**0.4**	**0.3**	**0.3**	**0.2**	**0.1**	**-**	**-**	**-**	**-**
1155	isoborneol	t	-	t	-	-	-	-	0.2	-	-
1165	**borneol**	**-**	**-**	**1.3**	**2.2**	**1.5**	**0.4**	**1.3**	**0.8**	**0.4**	**-**
1169	ethyl benzoate	-	-	-	-	-	t	-	-	-	-
1174	**terpinen-4-ol**	**12.1**	**10.1**	**0.5**	**0.2**	**0.2**	**0.4**	**0.3**	**0.2**	**t**	**0.3**
1179	p-cymen-8-ol	t	0.1	t	-	-	t	t	t	t	t
1186	**α-terpineol**	**2.4**	**2.3**	**0.3**	**0.2**	**0.8**	**0.4**	**0.5**	**0.3**	**0.2**	**2.9**
1190	methyl salicylate	-	0.4	0.1	t	t	-	-	-	-	-
1195	methyl chavicol	-	-	-	-	-	t	-	-	-	-
1196	cis-piperitol	t	-	-	-	-	-	-	-	-	-
1201	n-decanal	-	-	-	-	-	t	-	-	-	t
1204	verbenone	-	-	-	-	-	t	-	-	-	-
1207	trans-piperitol	t	-	-	-	-	-	-	-	-	-
1218	endo-fenchyl acetate	-	-	0.2	-	t	0.3	-	-	-	-
1223	citronellol	0.2	0.9	0.3	1.5	0.2	0.3	t	0.5	t	-
1232	thymol, methyl ether	-	-	-	-	-	-	-	-	-	0.2

KI	cpd	ONF	Yose	Yell	AZ	NM	C Po	A Du	El C	Oax	Mac
1249	piperitone	t	t	-	t	0.2	0.1	-	-	t	-
1274	neo-isopulegol acetate	-	t	-	-	-	-	-	-	-	-
1287	**bornyl acetate**	**0.2**	**1.3**	**44.7**	**32.4**	**28.9**	**14.6**	**16.0**	**20.7**	**25.2**	**0.1**
1298	trans-pinocarvyl acetate	-	t	-	-	-	-	-	-	-	-
1300	tridecane	-	-	-	-	-	0.1	-	-	-	-
1324	myrtenyl acetate	-	t	-	-	-	0.1	-	-	-	t
1342	43,93,121,194	-	-	0.3	0.1	0.1	-	-	-	-	-
1345	α-cubebene	-	-	-	-	-	0.3	-	-	-	-
1350	α-longipinene	-	-	-	-	-	-	-	-	-	0.2
1350	**citronellyl acetate**	**1.2**	**2.8**	**0.2**	**0.1**	**0.4**	**0.2**	**t**	**t**	**t**	**t**
1359	neryl acetate	-	-	-	-	-	-	-	-	-	t
1374	α-copaene	-	-	-	-	t	0.4	-	-	t	-
1379	**geranyl acetate**	**2.1**	**0.9**	**0.2**	**-**	**0.3**	**0.1**	**t**	**0.1**	**t**	**6.1**
1387	β-bourbonene	-	-	-	-	-	t	-	-	-	-
1387	β-cubebene	-	-	-	-	-	0.1	-	-	-	-
1389	longifolene	-	0.2	-	t	t	0.2	-	-	t	0.6
1417	**(E)-caryophyllene**	**-**	**-**	**-**	**-**	**-**	**0.5**	**-**	**-**	**-**	**0.2**

KI	cpd	ONF	Yose	Yell	AZ	NM	C Po	A Du	El C	Oax	Mac
1430	β-copaene	-	-	-	-	-	t	-	-	-	-
1432	trans-α-bergamotene	-	-	-	-	-	-	-	-	t	-
1451	trans-muurola-3,5-diene	-	-	-	-	-	0.2	-	-	-	-
1452	α-humulene	t	-	-	-	-	0.1	-	-	-	0.8
1465	ethyl cinnamate	-	-	-	-	-	t	-	-	-	-
1471	massoia lactone	-	-	-	-	-	t	-	-	-	-
1475	trans-cadina-1(6).4-diene	-	-	-	-	-	0.4	-	-	-	-
1478	γ-muurolene	-	-	-	-	t	0.1	t	t	t	-
1480	germacrene D	t	0.2	-	t	0.3	0.4	0.1	0.3	0.7	0.7
1493	trans-muurola-4,5-diene	-	-	-	-	-	0.4	-	-	-	-
1493	epi-cubebol	-	-	-	-	-	0.4	-	-	-	-
1500	α-muurolene	-	-	-	-	t	0.5	0.1	-	0.2	-
1505	β-bisabolene	t	-	-	-	-	-	-	-	-	-
1513	γ-cadinene	-	-	-	-	-	-	t	-	0.2	-
1514	cubebol	-	-	-	-	-	1.0	-	-	-	-
1522	δ-cadinene	t	t	t	-	t	1.5	0.2	0.3	0.5	0.3
1533	trans-cadina-1,4-diene	-	-	-	-	-	0.2	-	-	-	-

KI	cpd	ONF	Yose	Yell	AZ	NM	C Po	A Du	El C	Oax	Mac
1541	(E)-α-bisabolene	-	-	-	-	-	-	-	t	0.1	0.2
1561	(E)-nerolidol	-	-	-	-	-	0.1	-	-	-	-
1574	germacrene D- 4-ol	-	-	-	-	-	-	-	-	-	-
1582	caryophyllene oxide	-	-	-	-	-	0.1	-	-	-	-
1608	β-atlantone	-	-	-	-	-	-	-	-	-	0.1
1608	**humulene epoxide II**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**0.6**
1616	43,81,161,222	-	0.2	-	-	-	-	-	-	-	-
1627	1-epi-cubenol	-	-	-	-	-	0.6	-	-	-	-
1632	α-acorenol	-	t	-	t	t	-	-	-	-	0.8
1638	epi-α-cadinol	-	t	t	t	t	0.2	0.1	0.1	0.1	0.2
1638	epi-α-muurolol	-	t	t	t	t	0.2	0.1	0.1	0.1	0.2
1644	α-muurolol	-	-	-	-	t	t	t	t	t	-
1646	119,107,91,202	-	-	-	-	-	-	-	-	-	0.8
1652	α-cadinol	0.1	t	0.3	0.2	0.4	0.2	0.2	0.2	0.3	0.7
1711	pentadecanal*	-	-	-	-	-	0.2	t	t	t	-
1759	benzyl benzoate	-	t	-	t	t	-	-	-	-	-
1814	hexadecanal	-	-	-	-	-	-	t	-	t	-
1864	benzyl salicylate	-	t	-	-	-	-	-	-	-	-
1887	octadecadiene*	-	-	-	-	-	t	-	-	-	-

KI	cpd	ONF	Yose	Yell	AZ	NM	C Po	A Du	El C	Oax	Mac
1889	heptadecatrienal*	-	-	-	-	-	0.1	-	t	-	-
1937	cembrene	-	-	-	t	t	0.1	-	-	-	0.2
1943	iso-cembrene	-	-	-	-	t	-	-	-	-	t
1987	manoyl oxide	-	-	-	-	t	-	-	-	-	-
1992	ethyl hexadecanoate	-	-	-	-	-	0.3	-	t	-	-
2014	palustradiene	-	-	-	t	t	t	-	-	-	-
2048	thunbergol	-	-	-	t	t	0.1	-	-	-	0.5
2055	abietatriene	-	-	-	-	-	t	-	-	-	-
2056	manool	-	-	t	t	t	1.0	-	-	t	t
2087	abietadiene	-	-	-	-	t	t	-	-	-	-
2149	abienol	-	-	-	-	-	t	-	-	-	-
2165	9Z,12Z,15Z-octadeca-trienoic acid, ethyl ester*	-	-	-	-	-	0.2	-	-	-	-
2300	tricosane(C23)	0.1	t	-	t	t	-	-	-	-	0.3
2313	abietal	-	-	-	-	t	-	-	-	-	-

KI = Kovats Index (linear) on DB-5 column. Compositional values less than 0.1% are denoted as traces (t). Unidentified components less than 0.5% are not reported. For unknown compounds, four ions are listed, with the largest ion underlined. *tentatively identified.