TAXONOMY OF INFRASPECIFC TAXA OF *ABIES CONCOLOR*: LEAF ESSENTIAL OILS OF *VAR. CONCOLOR AND* VAR. *LOWIANA*

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

R. M. Lanner
2651 Bedford Ave., Placerville, CA 95667

Michael Kauffmann
Biology Department, Humboldt State University
4312 Lentell Road, Eureka, CA 95503

and

David Thornburg
2200 W. Winchester Lane, Cottonwood AZ 86326

ABSTRACT

The leaf essential oils of *Abies concolor* var. *concolor* and var. *lowiana* had large amounts of β-pinene (41-52%), except the Cimarron, NM population had 3.1% β-pinene. The oils from central and northern California were very similar and were devoid of (E)-β-ocimene and 6-methyl-5-octen-one. The New Mexico oil was the most unusual being high in camphene (25.9%), bornyl acetate (28.7) and α-pinene (11.2) with several unique components. Populations of *Abies c.* var. *lowiana* in central and northwest California were uniform in their leaf oil compositions. In var. *concolor*, considerable differentiation was found, confirming the work of Zavarin et al. (1975) of the Cuyamaca Race, and three sub-types of *A. c.* var. *concolor* oils: group A, B1 and B2. The B1 oil (Cimarron, NM population) was very different from any other populations, warranting additional studies. *Phytologia 93(1): 107-117 (April 1, 2011).*

KEY WORDS: *Abies concolor* var. *concolor, A. c.* var. *lowiana, leaf* essential oils composition, geographic variation.

Abies concolor (Gord. and Glend.) Hilde. is a forest tree of western North America (Fig. 1) ranging from Oregon to northern Mexico (Zavarin et al. 1975). Eckenwalder (2009) recognized two varieties: var. *concolor* and var. *lowiana* (Gord.) Lemm. and noted that these have been treated as species by some authors. He also indicated that var. *lowiana* hybridizes with *A. grandis* (D. Doug. ex D. Don in Lamb.) Lindl. but not with *A. lasiocarpa* (Hook.) Nutt. Recently, Xiang et al. (2009) examined nrDNA sequence data and found *A. concolor* most closely related to *A. grandis*, so hybridization seems possible. Zavarin et al. (1975) analyzed wood monoterpenes of *A. concolor* from 43 populations and found evidence that var. *lowiana* (n. and c. California) was a group (Fig. 1), but called var. *lowiana* from s. California, the Cuyamaca race. In addition, Zavarin et al. (1975) subdivided var. *concolor* into three groups (A, B1 and B2, Fig. 1).

There appears to be only one paper reporting on the leaf essential oil of *A. concolor* (Wagner et al. 1989) from a population in the North Kaibab Ranger District, AZ, and those data were reported on a ppm basis instead of the normal percent total oil data.

The purpose of this research was to examine the leaf essential oils from *A. concolor* from major chemical types found by Zavarin et al. (1975) and report on their compositions.

MATERIALS AND METHODS

Plant specimens: *Abies concolor* var. *concolor*: *Adams 12405-12407*, Mill B trailhead, Big Cottonwood Canyon, Salt Lake City, UT, 40º 37.996' N, 111º 43.418' W, 6242 ft., *Adams 12481-12485*, (by D. Thornburg) 7 mi. nw of Pine, AZ along Rim Rd., 34º 26.844'N, 111º 21.520'W, 7597 ft., *Adams 12556-12560*, 7 mi. w of Cimarron, NM on US 64, 36º 32.81' N, 105º 02.0' W, 6980 ft.,
Abies concolor var. *lowiana*: *Adams 12427-12431* (by R. Lanner) 2 mi. n of jct. US50 on White Meadows Rd., ca. 22 mi e of Placerville, CA, 38º 47' 00" N, 120º 29' 20" W, 3450 ft., *Adams 12432-12436* (by R. Lanner) Mormon Emigrant Trail at jct. with Park Creek Rd., ca. 24 mi ese of Placerville, CA, 38º 43' 30" N, 120º 28' 20" W, 4000 ft., *Adams 12438-12442* (by M. Kauffmann) Klamath Mtns., CA, 40º 50' 21.4" N, 123º 43' 11.09" W, 4820 ft., *Adams 12464-12468* (by B. Miller) Lee

Summit, CA on Hwy 70/89, 39º 52.674' N, 120º 45.736' W, 4414 ft., *Abies concolor* var. *concolor / lowiana*: *Adams 12522-12526*, on CA Hwy 38 north side of Onyx Summit, CA, 34º 12.037' N, 116º 43.520' W, 8490 ft. All specimens are deposited in the BAYLU herbarium.

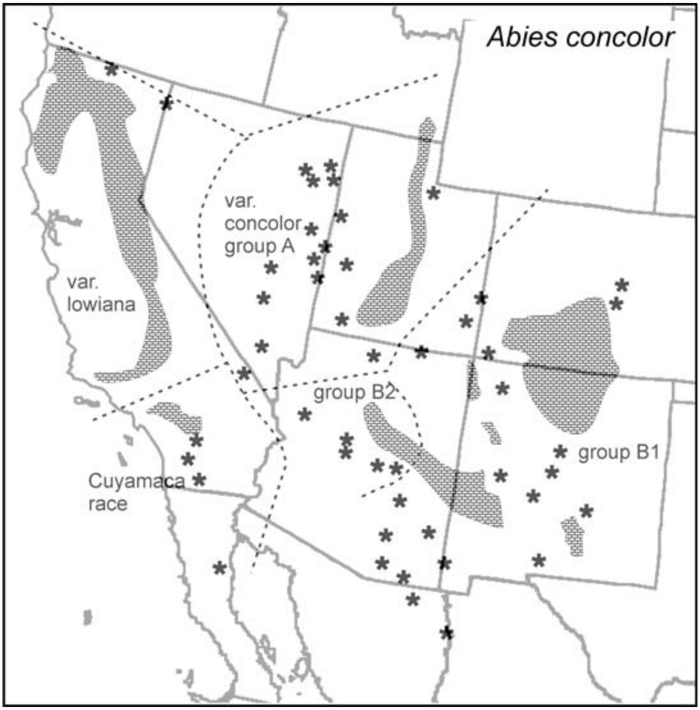

Figure 1. Distribution of *Abies concolor* (modified from Zavarin et al. 1975) with subgroups based on wood monoterpene data.

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 5 for operating details). Identifications were made by library searches of our volatile 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 percent total oil) were coded and compared among the species by the Gower (1971) metric. Principal coordinate analysis was performed by factoring the associational matrix using the formulation of Gower (1966) and Veldman (1967). Associational measures were computed using absolute compound value differences (Manhattan metric), divided by the maximum observed value for that compound over all taxa (= Gower metric, Gower, 1971; Adams, 1975). Principal coordinate analysis was performed by factoring the associational matrix based on the formulation of Gower (1966) and Veldman (1967).

RESULTS AND DISCUSSION

In general, the leaf oils of *A. concolor* are dominated by monoterpenes with only small amounts of sesquiterpenes and diterpenes (Table 1). Each of the populations are high in β-pinene, except the Cimarron, NM site where β-pinene is 3.1% (Table 1). The populations from central and northern California (Lee S, Plac, Klam, Table 1) share several components at similar levels: linalool, α-terpineol, geranyl acetate, RI 1617 sesquiterpene alcohol, and eudesm-7(11)-en-4-ol and all are devoid of (E)-β-ocimene and 6-methyl-5-octen-one (Table 1). The New Mexico oil is the most unusual oil being high in camphene (25.9%), bornyl acetate (28.7) and α-pinene (11.2) with several unique components: germacrene D, α-muurolene, α-acorenol, germacra-4(15),5,10(14)-trien-1-al, benzyl benzoate and cembrene (Table 1).

The overall similarities of the oils are shown in figure 2. Notice *A. c.* var. *lowiana* from central and northwestern California have very similar oils (0.833, 0.912, Fig. 2). The major difference in the

Klamath Mtns. oil is the presence of intermedeol (6.45, Table 1) that was only found in this oil. The oils from Utah and Onyx Summit are the next most similar (0.764, Fig. 2), with the oil from New Mexico being the least similar (0.670, Fig. 2).

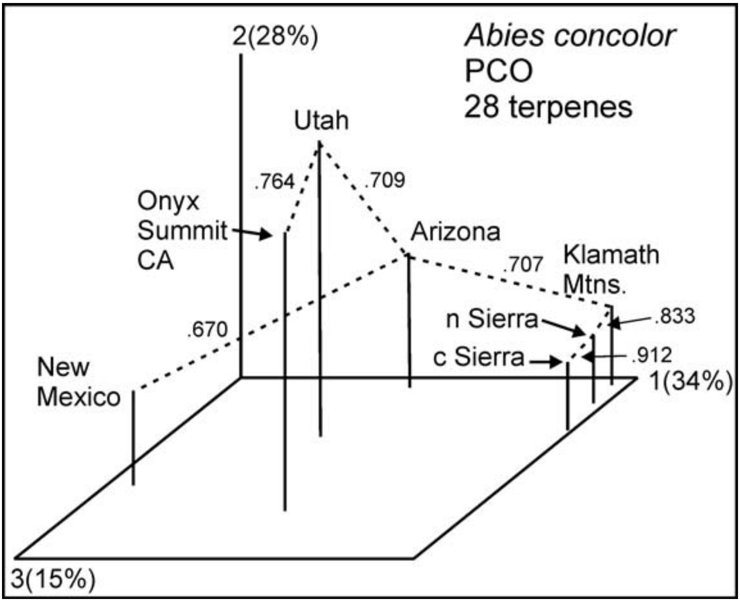

Figure 2. PCO based on 28 terpenes. The dotted lines are the minimum spanning network and the numbers next to the lines are the similarities.

Mapping the minimum spanning network onto the distribution of Abies concolor (Fig. 3) clearly shows the geographic affinities. The unity of the central and northwestern California *A. c.* var. *lowiana* populations is clear. Zavarin et al. (1975) designated the southern California populations as the Cuyamaca race and this analysis confirms their observation. Zavarin et al. (1975) did not indicate a strong affinity (in the wood monoterpenes) of the Cuyamaca Race to var. *concolor*, group A, but there is a high similarity in their oils (Figs. 2, 3).

Zavarin et al. (1975) divided var. *concolor* into 3 sub-groups: A, B1 and B2. The present analysis (based on leaf essential oils) confirms the same pattern of differentiation (Figs. 2, 3).

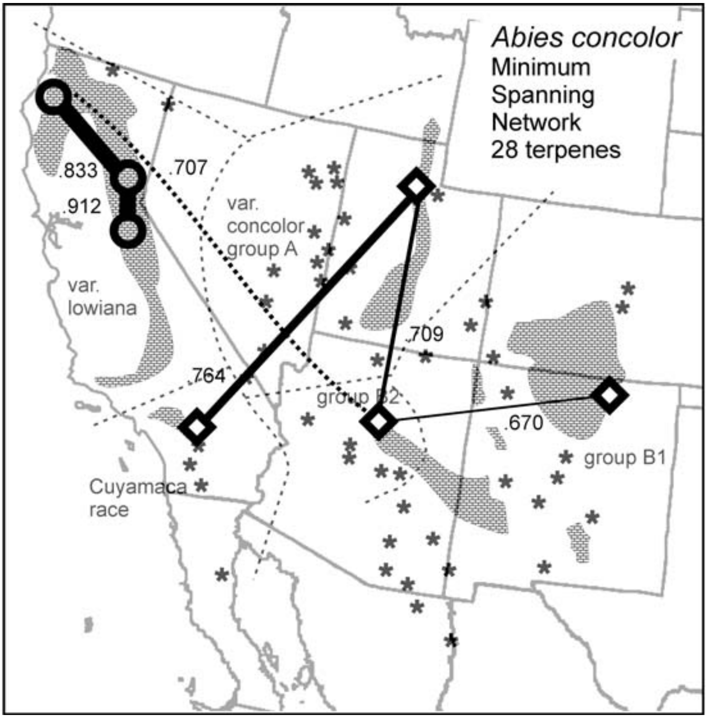

Figure 3. Minimum spanning network based on 20 terpenes. The open circles are *A. c.* var. *lowiana*, the open squares are generally treated as *A. c*. var. *concolor*. The numbers next to the lines are similarities.

CONCLUSIONS

In general there was very good agreement in the pattern of differentiation with Zavarin et al. (1975). The differentiation of the New Mexico oils was much greater than Zavarin el al. (1975) found in the wood monoterpenes. Additional sampling is needed (in progress, RPA) of *A. concolor* from Colorado, Utah and eastern Arizona to more clearly demarcate this differentiation.

It is premature to make taxonomic decisions at this time. Molecular research is needed (RPA, in progress) to clarify the taxonomic relationships of the varieties, the Cuyamaca Race and the unusual New Mexico plants.

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Table 1. Comparison of leaf oil compositions of *Abies concolor*. Klam = Klamath Mtns., NW CA, Lee S = Lee Summit, CA, Plac = Placerville, CA, AZ = Pine, AZ, NM = Cimarron, New Mexico, UT = Wasatch Mtns., UT, Onyx = Onyx Summit, CA. Compounds in bold face appear to separate the taxa. Compositional values less than 0.1% are denoted as traces (t). Unidentified components less than 0.5% are not reported. RI is the Kovat's Index using a linear approximation on DB-5 column. *= cpds used for PCO (28 cpds.)

RI	compound	Lee S	Plac	Klam	AZ	UT	Onyx	NM
884	santene*	0.2	0.2	0.3	-	t	t	1.6
921	tricyclene*	0.2	0.1	0.2	0.3	0.2	t	2.5
924	α-thujene	t	t	t	t	t	t	-
932	**α-pinene***	**5.1**	**4.4**	**4.7**	**7.9**	**8.9**	**7.8**	**11.2**
946	**camphene***	**2.5**	**1.4**	**2.7**	**4.3**	**2.9**	**0.5**	**25.9**
969	sabinene	t	t	t	t	t	t	0.1
974	**β-pinene***	**42.0**	**47.1**	**45.2**	**52.2**	**43.9**	**41.5**	**3.1**
988	myrcene*	2.0	2.2	1.4	2.2	2.1	1.7	2.0
1002	α-phellandrene	0.4	0.4	0.3	0.3	0.2	0.4	0.1
1008	δ-3-carene*	0.1	0.1	0.2	0.5	1.6	1.1	0.8
1014	α-terpinene	0.2	0.2	0.2	0.1	0.1	0.2	t
1020	p-cymene	t	t	t	t	t	t	t
1024	**limonene***	**23.0**	**23.0**	**17.6**	**9.0**	**9.3**	**21.2**	**9.1**
1025	β-phellandrene	2.5	2.5	2.0	1.1	1.1	2.5	1.2
1032	(Z)-β-ocimene	t	-	t	0.3	0.2	0.1	t
1038	2-heptyl acetate	-	-	-	-	t	-	-

RI	compound	Lee S	Plac	Klam	AZ	UT	Onyx	NM
1044	**(E)-β-ocimene***	**-**	**-**	**-**	**0.3**	**0.3**	**0.1**	**0.5**
1054	γ-terpinene	0.2	0.2	0.1	0.2	0.2	0.2	0.2
1077	(6-methyl-5-octen-2-one)	-	-	-	0.1	0.1	0.1	-
1086	terpinolene*	1.4	1.7	1.0	0.7	1.7	1.4	1.2
1087	2-nonanone*	0.3	0.4	0.1	0.5	0.6	0.4	0.1
1095	**linalool***	**0.4**	**0.2**	**0.1**	**1.4**	**1.8**	**1.4**	**3.1**
1118	endo-fenchol*	0.4	0.4	0.3	0.2	1.8	1.6	t
1118	cis-p-menth-2-en-1-ol	0.4	0.4	0.3	0.1	0.2	0.4	t
1122	α-campholenal	0.1	0.1	t	0.1	0.2	0.2	t
1136	trans-p-menth-2-en-1-ol	0.3	0.3	0.1	t	0.1	0.3	t
1141	camphor	0.1	t	t		0.1	t	0.2
1145	**camphene hydrate***	**0.3**	**0.2**	**0.1**	**1.8**	**4.1**	**0.5**	**1.2**
1148	citronellal*	0.1	0.1	0.3	0.6	0.3	0.2	0.2
1155	iso-borneol	t	t	t	t	0.1	t	t
1165	borneol*	0.5	0.3	0.2	0.2	0.6	0.8	1.5
1172	cis-pinocamphone	t	t	t	t	-	t	t
1174	terpinen-4-ol*	0.4	0.4	0.3	0.3	0.6	0.7	0.8
1183	cryptone	t	t	t	t	-	t	-
1186	**α-terpineol***	**6.5**	**6.9**	**4.8**	**2.2**	**3.5**	**4.5**	**0.8**
1195	cis-piperitol	0.2	0.2	0.1	t	t	0.1	t
1201	n-decanal	0.1	0.3	-	0.2	0.1	0.2	t

RI	compound	Lee S	Plac	Klam	AZ	UT	Onyx	NM
1207	trans-piperitol	0.2	0.1	t	t	t	0.1	-
1218	endo-fenchyl acetate	-	t	t	0.1	t	t	t
1223	citronellol*	0.5	0.2	0.3	0.4	0.6	1.0	0.3
1235	neral	0.3	0.1	-	-	t	t	-
1249	**piperitone***	**0.4**	**0.1**	**t**	**t**	**1.1**	**1.7**	**0.2**
1264	geranial	0.6	0.4	-	t	0.2	0.2	t
1287	**bornyl acetate***	**2.8**	**1.2**	**6.6**	**8.8**	**6.4**	**0.6**	**28.7**
1289	**thymol***	**t**	**t**	**-**	**t**	**1.3**	**0.6**	**-**
1293	2-undecanone	0.1	0.3	0.2	0.4	0.1	0.4	-
1300	tridecane*	0.2	0.1	t	0.3	0.2	0.8	-
1350	citronellyl acetate*	0.2	0.2	0.2	0.5	t	0.1	0.4
1374	α-copaene	-	-	-	-	-	-	0.1
1379	geranyl acetate*	0.5	0.8	0.4	0.2	t	t	0.3
1395	sesquiterpene,43,55,86,206	0.1	0.1	-	0.3	t	-	t
1408	dodecanal	t	t	-	t	t	0.1	-
1417	(E)-caryophyllene	t	t	t	t	0.2	0.3	t
1478	γ-muurolene	0.1	t	0.2	-	-	t	-
1480	**germacrene D***	**-**	**-**	**-**	**-**	**-**	**-**	**0.2**
1496	valencene	0.2	0.2	0.3	t	-	0.2	-
1500	**α-muurolene**	**-**	**-**	**-**	**-**	**-**	**-**	**0.1**
1514	cubebol	-	-	0.2	0.1	-	0.3	-

RI	compound	Lee S	Plac	Klam	AZ	UT	Onyx	NM
1522	δ-cadinene	-	-	t	0.1	-	0.4	0.1
1559	germacrene B	0.1	t	-	t	-	t	-
1561	(E)-nerolidol	0.1	t	-	t	0.1	t	-
1617	**sesquiterpene,81,161, 189,222***	**0.5**	**0.3**	**0.4**	**t**	**t**	**-**	**-**
1627	1-epi-cubenol	0.1	t	-	t	-	0.3	
1630	α-acorenol	-	-	-	-	-	-	0.1
1649	β-eudesmol	0.2	0.1	-	-	t	-	-
1652	α-eudesmol	0.2	0.1	-	-	t	-	-
1652	α-cadinol	0.2	0.1	-	-	t	-	0.4
1665	**intermedeol***	**-**	**-**	**6.4**	**-**	**-**	**-**	**-**
1685	**germacra-4(15),5,10(14)-trien-1-al**	**-**	**-**	**-**	**-**	**-**	**-**	**0.2**
1700	eudesm-7(11)-en-4-ol	0.1	0.1	0.1	-	t	-	-
1756	**benzyl benzoate**	**-**	**-**	**-**	**-**	**-**	**-**	**0.1**
1937	**cembrene**	**-**	**-**	**-**	**-**	**-**	**-**	**0.1**
1987	manoyl oxide	t	-	t	0.1	t	t	t
2014	palustradiene(=abieta-8, 13-diene) *	0.1	t	t	t	t	0.3	t
2056	manool	0.1	0.1	t	t	t	t	t
2149	abienol	0.1	0.1	t	t	t	0.2	t