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GEOGRAPHICAL VARIATION IN TERPENES FROM *LINDERA UMBELLATA* AND *LINDERA SERICEA*

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Key Word Index—*Lindera umbellata*; *L. sericea*; Lauraceae; leaf terpenes; geographical races.

Abstract—An investigation of the leaf terpenes in *Lindera umbellata* and *L. sericea* revealed the presence of four geographical races among these taxa.

FIVE varieties of two species of *Lindera umbellata* and *Lindera sericea* are all called Kuromoji in Japan. These plants are small deciduous trees and grow on the mountains from Hokkaido to Kyushu region in Japan. Morphologically, they are all very similar plants, but systematically they are treated as five taxa: *L. umbellata* Thunb. var. *umbellata*; *L. umbellata* var. *membranacea* (Maxim.) Momiyama; *L. umbellata* var. *lancea* Momiyama; *L. sericea* (Sieb. et Zucc.) Blume; *L. sericea* var. *glabrata* Blume.

Many workers have reported the analysis of the essential oils of Kuromoji,[1–4] Recently, von Rudloff[5] has reviewed the use of terpenes in chemosystematic studies and, from this, we decided to investigate chemotaxonomy of Kuromoji.[6,7]

RESULTS AND DISCUSSION

The smell of four varieties of *L. umbellata* and *L. sericea* resembled each other, while that of *L. sericea* var. *glabrata*, which has a leaf-like odour, is different from the others. The leaf oils were isolated by the steam distillation from the fresh leaves of Kuromoji collected from 26 geographical locations (Table 1). The seasonal changes in terpene composition of *L. umbellata* var. *umbellata* is shown in Table 2. From this, it can be seen that the seasonal variation is minimal from July to October, so that chemotaxonomic analyses can be carried out during this period.

The individual terpenes were isolated by column chromatography followed by preparative GLC and identified by IR spectral and GLC comparison with authentic compounds. The results are shown in Table 3. The percentage of the composition of the oil was calculated from the areas of the peaks of the gas chromatogram.

[1] KATO, M. (1951) *Kogyo Kagaku Zasshi* **54**, 519.
[2] HAGINIWA, T., HARADA, M., NAKAJIMA, M. and SAKAI, K. (1962) *Yakugaku Zasshi* **82**, 1441.
[3] FURUHATA, M., HORIGUCHI, T. and KATO, H. (1966) *Yakugaku Zasshi* **86**, 683.
[4] HAYASHI, N., TAKESHITA, K., NISHIO, N. and HAYASHI, S. (1970) *Flavour Ind.* **1**, 405.
[5] VON RUDLOFF, E. (1969) *Recent advances in Phytochemistry*, Vol. 2, p. 127.
[6] KOMAE, H., HAYASHI, N., KOSELA, S. and ARATANI, T. (1972) *Flavour Ind.* **3**, 208.
[7] HAYASHI, N. and KOMAE, H. (1973) *Z. Naturforsch* **28C**, 227.

Kuromoji plants from 26 geographic locations (Fig. 1) can be divided into four groups from the composition of the leaf oils. These groups are named Linalool-Kuromoji, Carvone-Kuromoji, Caryophyllene-Kuromoji, and Cineole-Kuromoji on the basis of the characteristic oils. Caryophyllene-Kuromoji is found in both Tohoku and Hokkaido. Linalool-Kuromoji occurs at Kanto and Chubu, both Linalool-Kurumoji and Carvone-Kuromoji grow at Chugoku, while Cineole-Kuromoji is found in the south part of Japan, in Shikoku and Kyushu.

Thus, all forms of Kuromoji, except for *L. sericea* var. *glabrata*, appear to be geographical races of a single taxon.

TABLE 1. KUROMOJI POPULATIONS SAMPLED

Population No.	Empirical designation	Elevation (m)	Identification	Chemotaxonomy
1	Mt. Hakodate, Hokkaido	350	D	d
2	Mt. Kutogi, Aomori	300	D	d
3	Rikuchukawaziri, Iwate	250	D	d
4	Yonezawa, Yamagata	300	D	d
5	Okuni, Yamagata	200	D	d
6	Shiobara, Tochigi	500	D	d
7	Mt. Tsukuba, Ibaragi	500	A	a
8	Tomiura, Chiba[2]	—	—	a
9	Izunagaoka, Shizuoka	450	A	a
10	Okatani, Nagano[3]	—	—	a
11	Mt. Hongu, Aichi	700	B	b
12	Takayama, Gifu	700	A	a
13	Yogo, Shiga	500	A	b
14	Mt. Yoshino, Nara	400	A	b
15	Haga, Hyogo	800	A	a
16	Mt. Daisen, Tottori	600	A	a
17	Mt. Sanbe, Shimane	400	A	b
18	Saijo, Hiroshima	350	A	b
19	Yoshiwa, Hiroshima	700	A	b
20	Mt. Aono, Shimane	800	A	a
21	Kido Pass, Yamaguchi	350	A	a
22	Mt. Nishihohen, Yamaguchi	400	A	a
23	Shirogano Pass, Kochi	400	A	a
24	Omogo, Ehime	900	C	c
25	Kuromori Pass, Ehime	1000	C	c
26	Mt. Hiko, Fukuoka	800	C	c

A: *L. umbellata* Thunb. var. *umbellata*. B: *L. umbellata* var. *lancea* Momiyama. C: *L. sericea* (Sieb. et Zucc.) Blume. D: *L. umbellata* var. *membranacea* (Maxim.) Momiyama.

a: Linalool-Kuromoji, b: Carvone-Kuromoji, c: Cineole-Kuromoji, d: Caryophyllene-Kuromoji.

EXPERIMENTAL

Isolation of the leaf oil. Samples were collected in 26 localities listed in Table 1. The fresh leaves (1–3 Kg) were subjected to steam distillation. The leaf oils were extracted with ether and dried. After the evaporation of the ether, leaf oils were obtained. The physical constants of four typical kuromojis (linalool-kurumoji, carvone-kuromoji, caryophyllene-kuromoji, cineole-kuromoji) are as follows: linalool-kuromoji (Izunagaoka), sampled at Izunagaoka (population No. 9) in Shizuoka Prefecture on September in 1970, 0·2% yield of fresh leaves, d_4^{25} 0·8925, n_D^{25} 1·4658, α_D^{25} −17·0°. Carvone-kuromoji, sampled at Mt. Hongû (population No. 11) in Aichi Prefecture in October 1971, 0·4% yield, d_4^{25} 0·9055, n_D^{25} 1·4789, α_D^{25} +10·7°. Caryophyllene-kuromoji, sampled at Yonezawa (population No. 4) in Yamagata Prefecture in September 1971, 0·3% yield, d_4^{25} 0·8470, n_D^{25} 1·4717, α_D^{25} −20·5°. Cineole-kuromoji, samples at Kuromori Pass (population No. 26) in Ehime Prefecture in July 1970, 0·2% yield, d_4^{25} 0·8979, n_D^{25} 1·4646, α_D^{25} −16·0°.

TABLE 2. SEASONAL VARIATION OF THE ESSENTIAL OIL OF *Lindera umbellata* VAR. *umbellata*

Compound	t_R (min)	Seasons May	July	October
		Composition (%)		
α-Pinene	6·9	1·6	3·9	1·5
	7·9	0·7	2·3	0·8
	9·0	0·4	0·8	0·3
	10·1	0·4	0·7	0·3
	10·6	0·4	0·8	0·3
	11·1	0·4	0·8	0·5
	11·4	0·4	—	—
	11·8	4·3	5·9	4·7
1,8-Cineole	12·4	25·0	16·6	16·3
	13·4	1·4	1·4	1·8
	14·1	0·8	1·6	1·2
	14·9	0·2	0·4	0·3
	20·6	0·1	0·2	—
	24·8	—	0·2	—
Linalool	25·3	27·3	27·4	29·4
	26·4	0·5	—	0·9
	27·9	0·2	0·6	2·6
	28·2	2·8	2·3	0·1
	28·5	—	0·2	—
	29·2	0·1	0·7	1·9
	30·0	2·6	0·6	4·7
	32·1	2·0	1·8	2·0
	32·8	1·0	0·7	0·9
Carvone	33·1	24·7	28·4	26·1
	33·9	0·6	0·2	0·4
	34·3	—	—	0·3
	34·6	—	—	0·4
	34·7	—	—	0·5
	35·1	1·6	0·4	0·9
	39·4	0·5	1·1	0·9

TABLE 3. COMPOSITIONS OF THE ESSENTIAL OILS OF KUROMOJI

Compound	Samples a	b	c	d
	Composition (%)			
α-Pinene	7·1	5·1	11·2	6·0
Camphene	4·4	3·3	5·1	3·0
β-Pinene	1·3	0·8	3·6	1·0
Δ^3-Carene	0·7	1·0	1·6	1·0
Myrcene	1·6	1·4	2·0	1·1
Limonene	6·7	15·5	8·0	5·8
1,8-Cineole	30·6	13·7	42·3	30·1
γ-Terpinene	4·9	2·2	2·6	4·0
Linalool	28·6	18·2	7·0	22·9
Borneol	0·5	0·4	0·6	0·8
Carvone	—	23·7	0·6	0·5
Geraniol	1·3	3·2	0·6	0·8
Bornyl acetate	0·6	0·4	1·0	0·8
Geranyl acetate	1·3	1·1	3·2	0·5
Caryophyllene	0·2	1·1	1·2	7·8
γ-Cadinene	0·5	0·6	0·4	0·5
Unidentified	9·7	8·3	9·0	13·4

SE-30 (5%), 70–250 °/min, column 1 m.

a: Linalool-Kuromoji. b: Carvone-Kuromoji. c: Cineole-Kuromoji. d: Caryophyllene-Kuromoji.

Gas chromatography. For identification, a Hitachi Model K53 gas chromatograph equipped with an ionization detector was used. The stainless columns were packed with 10% SE-30 or 10% Carbowax 20M on Chromosorb W. Temperature programmed from 70 to 250° at 3°/min. For preparative work a JEOL-JGC-20KT gas chromatograph was used with 20% Carbowax 20M on Chromosorb WAW at 80 or 170°.

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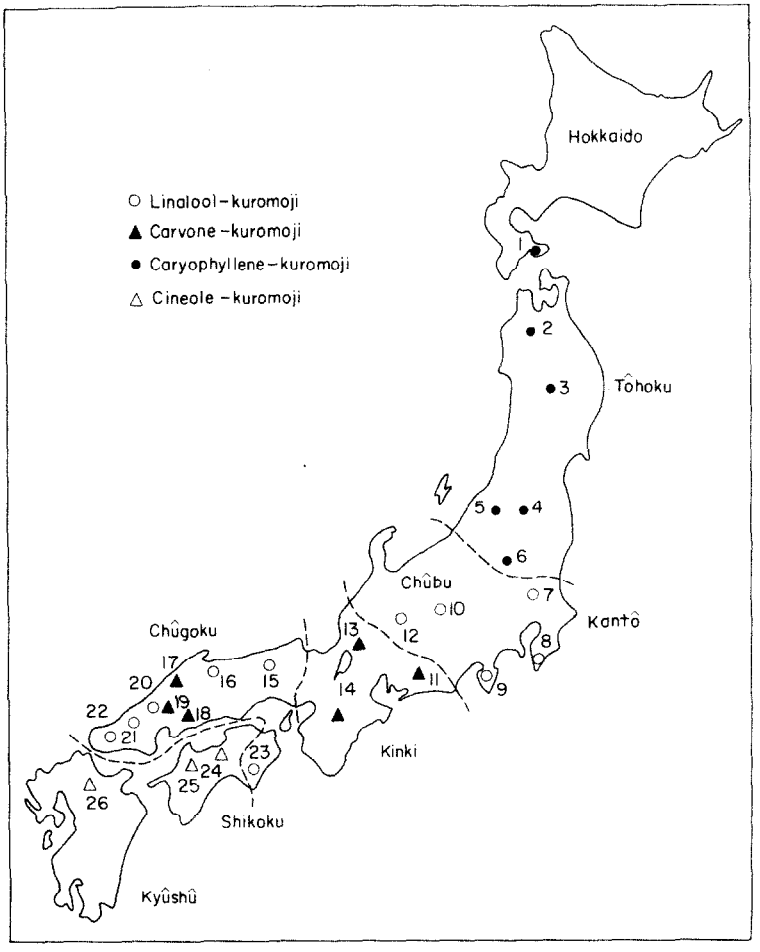

FIG. 1. DISTRIBUTION OF DIFFERENT LEAF OIL TYPES OF KUROMOJI IN JAPAN.