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Variation of the Essential oil and Main Component from Different Origin of *Levisticulum officinale* Koch.

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ABSTRACT : The essential oils were isolated by hydrodistillation and their composition determined capillary GC method with standards. The essential oil content showed significant differences between the two populations on the vegetative organs. The essential oil level of the leaves and roots was considerably higher in the Korean population at full flowering and waxy ripening stage but essential oil content of the roots was significantly higher in the Hungarian taxon at leaf rosette stage. We observed the essential oil accumulation tendency was mianly dependent on plant organs and intra-specific taxon during the vegeation period. Butylidene-phthalide was proved to be the main component of the oil in both population roots (50.9-73.3%), while dimethyl-acetate was showed as a major compound on the over-ground parts (56.7-62.0%). The qualitative composition of the essential oil in the reproductive organs concerning the identified compounds was the same as the vegetative parts with the main component α-phellandrene (4.8-28.1%) and butylidene-phtalide (9.7-16.1%). The quantitative composition showed some changes during the ontogenesis phases. Most characteristic ones are the decreasing proportion of dimethyl-acetate (from 7.3% to 1.1%) and the appearance of α-pinene (from 0.5% to 1.5%) only after fruit setting in both population.

Key words : Lovage, essential oil, plant organ, main components, α-pinene, α- phellandrene, Phthalide, dimethyl-acetate

INTRODUCTION

Levisticum officinale Koch. is a perennial herbaceous plant belonging to the Apiaceae family. It has been cultivated as a medicinal and spice plant since ancient times. Lovage leaves are often used to be seasoning, especially for soup, while the underground parts (rhizome and roots) are used for medicinal purposes (Tyler. 1990 and Keville, 1991). The essential oil of lovage (*Aetheroleum levistici radicis*), root (*Levistici radix*), fruit (*Levistici fructus*), leaf (*Levistici folium*) and sometimes its herb (*Levistici herba*) are used as drugs (Hornok, 1992; Tyler, 1990). The essential oil content of leaf is usually 0.08-0.24%, in ripe seed investigated 0.8-1.5% and root contains 0.6-1.0% (Szebeni and Galambosi, 1992; Lenches, 1993). The main components of the lovage in essential oil were mainly phthalides and terpenoids, including n-butylidine-phthalide, sedanonic anhydride, d-α-terpineol, carvacrol and eugenol (Tyler, 1990; Szebeni and Galambosi; Stahl-Biskup and Wichtmann). This study was carried out to know the variation of essential oil content and the main components in essential oil by different ecological conditions between the Korean and the Hungarian lovage populations during the vegetation periods and ontogenesis.

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MATERIALS AND METHODS

Taxa

Intraspecific taxa of the two *Levisticulum officinale* Koch. a variety of Hungarian origin "Budakalaszi" the Gene bank of the Department of Medicinal and Aromatic Plants, SZIE, and a Far eastern origin commonly grown in Korea (from Gene bank of the Rural Development Administration in Suwon, Korea) Were investigated and compared from 1997 to 1999.

Samplings

The essential oil content and composition was measured from several plant organs during vegetation and ontogenesis in both populations. The vegetative plant organs were sampled at the flowering phenophases in the three-year-old stands :

- o Leaf-rosette-stage : over-ground parts and roots
- o Full flowering stage : leaves, stems and roots
- o At waxy ripening stage : leaves, stems and roots.

The generative organs(without stalks) were sampled at the following phenophases in the 2nd and 3rd vegetation cycles:

- o yellow bud stage
- o full flowering stage green fruit stage (2-3weeks after full flowering)
- o Milky fruit stage (fruits of full size, not solid, yellowish)
- o waxy fruit stage (fruits are of full size, start of hardening, yellowish-brown)
- o Ripening fruit stage (usual harvest time : brown and solid)
- o Over-ripening stage (2-3weeks after usual harvest time, brown-grayish). At every stage and every organ, representative, average population samples were taken in 3replications and drying was carried out under natural circumstances, in closed place.

Laboratory investigations

The essential oil was obtained by hydro-distillation method from 20g of each sample with Clevenger apparatus according to the descriptions of the phHgVII. The oil composition was analyzed by capillary GC (Shimadzu GC-B14 with Shimadzu Class-VP Chromatography Data System 4.2 computer system). An SE - 30 25 M × 0.25 MM (film thickness 0.25 μm) column was used. The injector and detector temperatures were 220℃ and 250℃ and the column temperature program was 110℃ (3min), 110℃ - 250℃ (8℃/min), 250℃ (5 min). The carrier gas was nitrogen, 1 ml/min. the identification of compounds was performed by comparison of their retention times with those of pure substances by peak enrichment with standards of GC-MSD.

RESULTS AND DISCUSSION

The essential oil content of the leaves and wooden part of stem proved to be higher in the Korean population at leaf rosette stage. However, the difference is not significant and on the contrary, the essential oil content of the roots was significantly high in the Hungarian taxon at the same phase (Table1.).

Table 1. Essential oil content of the three-year-old vegetative organs at different phenophases of different origins of *Levisticum officinale* Koch.

(ml/100g d. w.)

Phase	Shoots		Roots	
	Hungarian	Korean	Hungarian	Korean
Leaf-rosette	0.60	0.66	0.63	0.39

	Leaf		Stem		Root	
	Hungarian	Korean	Hungarian	Korean	Hungarian	Korean
Flowering	0.67	0.83	0.09	0.11	0.11	0.56
Waxy ripening	0.88	1.10	0.10	0.11	0.30	0.44

The tendency of the accumulation oil in the aerial parts, especially in the leaves during the vegetation period seems to be increasing in both population, maximum values reached was at the waxy ripening stadium. The contents of the stem were quite constant. The roots showed different tendencies in both taxa, decreasing-increasing tendency in the Hungarian taxon but increasing-decreasing in the Korean population. The maximum values of the leaves and roots can be considered as high compared with the essential oil values -0.12-1.36% in the roots and 0.16-0.31% in the leaves- mentioned by Szebeni and Galambosi (1992).

The Korean population proved to be superior to the Hungarian one concerning the essential oil content of their generative organs. Exceptions were only found in green-milky fruit stage at two-year-old and milky fruit stage at three-year-old stands. The essential oil content of the generative organs showed a characteristic tendency during the ontogenesis, which was similar in both studied taxa (Table 2.). A considerable increasing step can be observed after the start of fruit development, in the "green fruit" stage. In the two - year - old Hungarian population the accumulation level even reaches its maximum (1.86% d.w.) at this phase. Interestingly, at the following milky fruit stage the levels went down to about their one third, while increased thereafter again. In the three-year-old populations this drawback was measured at "waxy fruit" stage. The decreasing period is followed in every case by a second accumulation step and the fruits had high essential oil content at the end of the vegetation period.

The main component in the essential oil of the over ground parts was dimethyl-acetate at leaf-rosette-stage (Table 3). In the oil of the shoots, the monoterpene 1, 8 cineole as well as the butylidene-phtalide accumulated almost at the same level (12-17% of the oil). As minor components, α- and β-pinenes were identified. This composition both quantitatively and qualitatively was practically equal in the studied taxa. In the roots, phthalide proved to be the main component in both populations, however, in its proportion, considerable differences were measured : the proportion in the Korean plants reached 73%, while it appeared to be 51% in the Hungarian one. In the contrary, dimethyl-acetate concentrated in the Hungarian populations as twice high level as in the Korean taxon. The other identified components were qualitatively the same as in the shoots, except β-phellandrene in the Korean taxon. With this result, our data partly contradict to Galambosi and Szebeni (1992), who mentioned phellandrene, ligustilide, pentyl-cyclohexa-dine and germacrene-D as the main components of the roots, however it was coincide with the statement of Tyler (1994) pointed out that the phthalides as the main compound of the lovage root. The quantitative composition of the essential oil in the reproductive organs concerning the identified compounds was the same as the vegetative parts. However, some interesting quantitative changes could be measured during the development of the fruits (Table 4). β-Phellandrene and phthalide are the main components of the reproductive organs of both the Hungarian and the Korean population as well. In this way our findings was in agreement with the results of Toulemound and Nolean (1988) as well as of Tyler (1994). β-Phellandrene's proportion in the essential oil lies between 27-54%. The variation however is not

Table 2. Essential oil content of the reproductive organs during the fruit developing phases of different origin of lovage

(ml/ 100g)

Plant Age	Population	Bud	Flower-ring	Green fruit	Milky fruit	Waxy fruit	Ripening fruit	Over-ripening	LSD
Two	Hungarian	0.17	0.35	1.86	0.53	1.60	1.81	-	0.22
	Korean	0.35	0.53	1.33	0.43	1.82	1.81	-	0.14
Three	Hungarian	-	-	1.07	1.61	0.86	1.40	1.40	2.14
	Korean	-	-	1.61	1.30	0.86	2.58	3.02	1.90

Table 3. Main components in the essential oil (in % of the oil) of the three-year-old different populations on the different plant organs at the leaf rosette stadium

(%)

Compounds	Shoots		Roots	
	Hungarian	Korean	Hungarian	Korean
α-Pinene	0.4	0.7	1.1	0.4
β-Pinene	1.9	2.4	3.4	1.4
1.8-Cineole	12.7	17.7	5.9	4.8
Dimethyl acetate	62.0	56.7	16.6	7.4
Butylidene-phthalide	16.9	14.4	50.9	73.3
β-Phellandrene	-	-	-	2.0

Table 4. Main components in the essential oil (in % of the oil) of the two -year- old different populations during the ontogeny phases

(%)

Compound	Bud stage		Flowering stage		Green fruit		Milky fruit		Waxy fruit		Ripening fruit	
	Hun	Kor	Hun	Kor	Hun	Kor	Hun	Kor	Hun	Kor	Hun	Kor
α-Pinene	-	-	-	-	1.5	1.1	0.6	0.8	0.5	0.8	-	0.5
β-Pinene	0.7	0.	1.0	1.5	1.6	1.2	1.4	0.9	1.9	1.3	0.9	1.5
β-Phellandrene	27.3	32.2	35.5	45.7	48.3	41.7	39.4	38.2	49.4	43.4	34.8	43.8
1.8 cineol	1.0	-	0.6	0.9	0.6	1.6	0.1	0.1	-	-	0.9	-
Dimethyl acetate	5.7	7.3	6.3	6.2	1.5	6.8	0.7	1.1	1.1	0.9	13.5	1.8
Butylidenephthalide	59.3	51.0	47.0	38.2	38.1	43.0	47.6	60.5	40.5	45.7	41.7	47.6

depending on the genotype and it hardly depends also on the phenological phases. The situation was similar in case of phthalide, the level was between 38-59% in the Hungarian and 38-60% in the Korean population, which means no difference between the two taxa. Among the minor components, the monoterpene α-pinene appears only after fruit setting and was measured from the green fruit stadium in both taxa. β-Pinene was detected during the whole investigation period in both population and no significant changes were measured in the proportion. The content of 1,8-cineole could be considered as low in both taxa. Propotion of dimethyl-acetate showed to be decreasing during the fruit development in both populations.

SUMMARY

We investigated the essential oil and main components of different plant organs during the vegetative and reproductive periods and found out the essential oil content and chemical composition were mainly depended on plant organs though it was cultivated different ecological conditions in both populations, respectively. The tendency of the essential oil accumulation in aerial parts, especially in the leaves during the vegetation period seems to be increasing in both population and maximum values (1.60-1.82%) reached at the waxy ripening stadium. Korean population proved to be superior to the Hungarian taxon concerning the essential oil content of their generative organs (Kor 2.58, Hun 1.81%). The main component of vegetative organs of leaves and stems was dimethyl acetate (Kor 76.6, Hun 72.2%); meanwhile in root was the butylidene phthalide (Kor 72.8, Hun 71.0%). However, β-phellandrene (from 27.3 to 53.9%) and butylidene phthalide (from 38.1 to 60.5%) were the main component of

generative organs in both populations. The quantitative composition showed some changes during the development of the fruit and the most characteristic ones are the decreasing proposition of dimethyl acetate and the appearance of α-pinene only after the fruit setting in both taxa, respectively.

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