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Fatty Acid Composition of Four *Lathyrus aphaca* L. Varieties, A Chemotaxonomic Approach

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Abstract: Some natural *Lathyrus aphaca* L. varieties have been investigated from chemotaxonomic point of view. Seed fatty acid composition of four *L. aphaca* varieties (*affinis, biflorus, pseudoaphaca* and *modestus*) of genus *Lathyrus* was determined and their taxonomic relationships were also evaluated using chemical and numerical taxonomic techniques. The *Lathyrus aphaca* varieties seed contained profitable composition of poly-unsaturated fatty acids, linoleic and palmitic acid. The lowest fatty acid component was margaric, pentadecanoic and myristic acid. It is determined that the seed lipids contained a relatively large proportion of un-saturated fatty acids with linoleic acid as predominant. The results showed that intraspecific variation of fatty acid patterns was low in *L. aphaca* varieties with the exception of *modestus*, which was found partly different by fatty acid composition from the others.

Key words: *Lathyrus*, chemotaxonomy, fatty acid, cluster analysis.

Introduction

The fact that, species producing unusual fatty acids may become extinct before their genetic potential to produce technically interesting raw material has been fully recognized and investigated. The rate at which progress in new techniques, e.g. gene transfer from wild plant species into agronomically developed crops as rapeseed, can be achieved is in direct competition with the rate at which plant species, and their genetical potential, are disappearing from the globe (Aitzetmüller & Tsevegsüren, 1994). Thus, the study of plant chemicals in terms of technical and pharmaceutical importance is increased all over the world. Kupicha (1983) lists 152 species in the *Lathyrus* genus (Fabaceae) which are distributed from sub-arctic to sub-tropics. It includes a range of grain, forage, pasture and ornamental crops (Enneking, 1998). The genus comprises of 67-70 taxa in the level of species, subspecies and variety (Davis, 1970; Maxted *et al.*, 1988; Ertekin & Saya, 1996) and it is represented by 10 sections in Flora of Turkey. The *Aphaca* (Adans) Reichb. (Guss.) Arc. section is represented by 2 species, *L. aphaca* (L.) and *L. stenolobus* (Boiss), according to Davis (1970). *L. aphaca* is comprised of 6 varieties in Flora of Turkey and its complex reaches its maximum variability in Anatolia. Several of the varieties have ecogeographical preferences and intermediates are relatively uncommon in Turkey. The classification accommodates most of the variation found in turkey, but varietal misidentification is easy in the absence of notes on flower colour. Seed - coat pattern is probably not a reliable diagnostic character.

L. aphaca var. *biflorus* Post. is widespread except in N. east Anatolia. *L. aphaca* var. *pseudoaphaca* (Boiss.) Davis usually has a narrow standard than others. Some morphological differences were reported among the varieties, especially flowers colour, peduncle and the stipule characters. However, *Lathyrus* genus is very important plant as a cultivar and crop in Turkey and other parts of the world. Doğan *et al.* (1992) divided *Lathyrus* genus into *Orobus* and *Lathyrus* sub-genera. Then they reducted the section number of *Lathyrus* from 10 to 9.

Many wild species of plants are capable of synthesizing unusual and technically interesting fatty acid structures in their seed lipids (Smith, 1970; Badami & Patil, 1981; Aitzetmüller & Tsevegsüren, 1994). Unusual FAs accumulated in the seeds of higher plants can be of chemotaxonomical interest and their presence or absence may indicate closer or more distant relationships between the considered species (Aitzetmuller, 1993) as it has already been demonstrated (Hegnauer, 1989; Harborne & Turner, 1984; Jmbs and Pham, 1996; Valesco and Goffman, 1999). There is some evidence that the rare fatty acids, may be harmful to animals eating the seeds. Erucic acid, a characteristic acid of the Cruciferae, has been described as having toxic effects in mammals, if ingested in sufficient amounts(Harborne & Turner, 1984).

It is aimed in this research to indicate the fatty acid composition of *L. aphaca* varieties and chemical relationships among these taxa and also compare them chemotaxonomically. In this way it was studied whether the fatty acid made a positive contribution to the taxonomy of these plants or not. The study is very important in view of infraspesific chemical variation in the *Lathyrus* taxa. On the other hand, it is very useful to determine these chemicals in plant, in field of chemosystematics, pharmacology and agronomy.

Materials and Methods

Plant material: Seeds of four *L. aphaca* varieties were collected from natural habitats, shown in Table 1.

Lipid Extraction: Seeds (2g) of each sample were homogenized with isopropanol. The latter procedure - extraction and purification - was made according to Kates (1986) method. Methylation was carried out according to Christie (1984).

Gas Chromatography: Analysis of fatty acid methyl esters was carried out and the resultant mixture of fatty acid methyl ester was injected onto a UNICAM – 610 GC. FID detector, carrier gas (N, ml /min) 2.5 capillary column (15 m X 0.32 mm) packed with 70% BPX –70 were used for analysis. The temperature of column was 185 ^{0}C. One μL was introduced onto the column.The quantity of fatty acid was calculated as μg/ g. Identification of fatty acid components was carried out with the help of authentic standard and retention time of components used in fatty acid analysis. For all samples the procedure was performed in triplicate and the mean values were stated.

Cluster analysis technique – Euclidean distance – in SPSS 9.05 computer program was applied on the chemical data.

Results and Discussion

The data obtained from the analysis is presented in Tables 2a,b. Despite the mentioned morphological differences (Davis, 1970: Şahin *et al.*, 2000) that exist among the *L. aphaca*

Table 1: Locations of studied *Lathyrus aphaca* varieties.

Species	Region – Locality	Altitude	Latitude.	Longitude.
L. aphaca var. *psudoaphaca*	C_3 Isparta Aksu	1600 m.	37°47′	31°4′
L. aphaca var. *affinis*	C_2 Muğla -Startonikia Antic city	550 m.	37° 19′	28° 8′
L. aphaca var *biflorus*	C_2 Antalya - Korkuteli Avdan Village	1000 m.	37° 4′	30° 13′
L. aphaca var.*modestus*	C_3 Isparta - Eğirdir, Balkırı Village	910 m.	37° 52′	30° 51′

Table 2a: Fatty acid composition of seed lipids of *Lathyrus aphaca* varieties (µg/g, %)

Taxa	12:0	13:0	14:0	15:0	16:0	17:0	18:0	18:1	18:2	18:3	19:0	20:0
Lathyrus aphaca var.	--	7.94	2.16	2.36	7.57	0.22	24.4	33.83	312.2	49.9	--	--
psudoaphaca	--	1.80	0.49	0.54	1.72	0.08	5.54	7.68	70.84	11.32	--	--
Lathyrus aphaca var. *affinis*	--	57.0	54.0	14.3	96.4	0.35	36.3	31.8	370.2	39.3	--	--
	--	8.15	7.72	2.04	13.8	0.06	5.19	4.54	52.91	5.62	--	--
Lathyrus aphaca var *biflorus*	--	27.6	11.46	1.8	118.9	2.27	46.46	46.1	451.7	62.96	--	--
	--	3.59	1.49	0.23	15.46	0.31	6.04	5.99	58.7	8.18	--	--
Lathyrus aphaca var.*modestus*	--	9.95	8.51	16.7	60.56	5.30	14.2	25.1	242.9	10.9	--	--
	--	2.52	2.16	4.24	15.37	1.35	4.95	6.37	68.0	2.77	--	--

Fatty acids: 13:0 (Tridecanoic acid), 14:0 (Myristic acid), 15:0 (Pentadecanoic acid), 16:0 (Palmitic acid), 17:0 (Margaric acid), 18:0 (Stearic acid), 18:1 (Oleic acid), 18:2 (Linoleic acid), 18:3 (Linolenic acid).

Table 2b: SFA (Saturated fatty acids) and USFA (Unsaturated fatty acid) amount of *Lathyrus aphaca* varieties studied (µg/g).

Taxa	SFA	USFA
L. aphaca var. *psudoaphaca*	44.78	395.93
L. aphaca var. *affinis*	258.42	441.34
L. aphaca var. *biflorus*	208.54	560.76
L. aphaca var. *modestus*	115.22	278.9

varieties they had similar fatty acid patterns. The major FAs encountered were 18:2, 16:0, 18:3, 18:1 and 18:0 respectively.

Comparison of fatty acid composition of *L. aphaca* varieties revealed specific features in FA composition, characteristic of these varieties that would have a taxonomic value. Four *L. aphaca* varieties were found different in fatty acid composition quantitatively.

In investigation, 9 fatty acid components were determined in each taxa, 12:0, 19:0 and 20:0 fatty acid components were not found throughout. The amount of saturated fatty acids (SFA- 13:0, 14:0, 15:0, 16:0:17:0,18:0) and also unsaturated fatty acid (USFA - 18:1 + 18:2 + 18:3) are shown in Table 2b. It can be said that *Lathyrus* taxa studied were not rich in point of saturated fatty acid component and amount (Table 2a and b).

The lowest fatty acid component was Margaric acid (17:0) in all of the taxa, this is very rare fatty acid component in the plant seed lipids. Pentadecanoic and myristic acid also were the second lowest components. The highest fatty acid component was the Linoleic acid in all of the *Lathyrus* taxa. While Palmitic acid was the second abundant fatty acid component in all varieties except *pseudoaphaca*. It is reported that γ- linolenic acid had great interest for the pharmaceutical industry and is now produced from oilseeds, where it is present at 5-10% levels. Some of unusual fatty acids were present in small amounts only in the seed oils and they were chemotaxonomically significant becuse of their constant presence in all the species of one genus or a few genera, combined with their constant absence throughout the species of other genera (Aitzetmüller & Tsevegsüren, 1994).

The ratio of 18:3 and 18:2 (18:3/18:2) fatty acid components were found 0.16 in var. *pseudoaphaca*, 0.11 in var. *affinis*, 0.14 in var. *biflorus* and 0.04 in var. *modestus*. It is reported that the fatty acid composition and the ratio of (18:3/18:2)can serve as taxonomic markers in some family member (Marin *et al.*, 1991). The differences among the taxa by 18:3/18:2 ratio, showed that first three varieties were very similar and var. *modestus* was found highly different from others. It is reported that some morphological confusions and

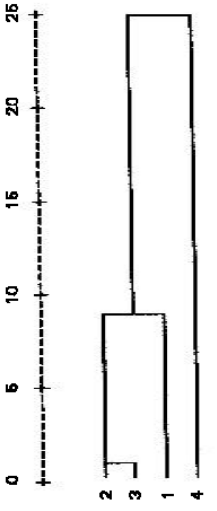

Fig. 1: Affinities relationships between samples of *Lathyrus aphaca* varieties, based on the distribution of seed fatty (1. *L. aphaca* var. *psudoaphaca*, 2. *L. aphaca* var. *affinis*, 3. *L. aphaca* var. *biflorus*, 4. *L. aphaca* var. *modestus*).

controversies were determined among the *Lathyrus* species and varieties. *L. aphaca* var. *affinis* (Guss.) Arc.- was reported as depauperate plant and easily mistaken for var. *pseudoaphaca* (Davis, 1970).

It can be said that fatty acid amount of *Lathyrus* sp. studied in general were similar in point of both SFA and USFA. In the same way, oleic, linoleic and palmitic acid were the main components in seed oil. This phenomenon was reported by other researchers (Daulatabad, 1987; Kwieicinska & Matyka, 1986; Bağcı & Şahin, unpublished).

The results of the Euclidean distance method of cluster analysis for the specimen data set are summarized in Fig. 1. In the dendrogram, *L. aphaca* varieties studied connected with each other at top level. These varieties were found very similar in view of fatty acid composition as shown in dendrogram. But *L. aphaca* var. *modestus* was determined as a single taxon. They formed two groups, first three varieties – var. *pseudoaphaca*, var. *affinis*, var. *biflorus*- and the last, that stayed as single specie in dendrogram. *L. aphaca* varieties *affinis* and *biflorus* were found the closest two taxon in the dendrogram and var. *pseudoaphaca* was the third closest

taxon with the both.

Chowdury & Banerjii (1995) analyzed the seeds from 3 plant species belonging to *Crotalaria* for fat and fatty acids and found that all the seed fats resembled the simple linoleic-oleic-palmitic type. The results obtained in present study confirmed that the linoleic – palmitic and linolenic type fatty acid composition was dominant in the *Lathyrus* taxa, particularly comparing with the other plant species. Nine samples of the legume seeds including *L. sativus* were analyzed for fatty acids and tocopherol contents. In the result, they reported that the legume seeds had profitable polyunsaturated fatty acids composition especially, from linoleic and linolenic acid point of view (Grela & Günter, 1999).

In morphologic and karyologic analysis of some *Lathyrus* sp., *L. aphaca*, var. *modestus* (Şahin, 1993) was found to be different from others (*Lathyrus rotindifolius* Willd., subsp. *miniatus* (Bieb.ex.stev) Davis, *L. cassius* Boiss., *L. cicera* L.) in terms of chromosomal karyology. Şahin *et al.* (2000) studied, chromosome number of *Lathyrus* taxa and *L. aphaca* varieties - *modestus*, *affinis* and *pseudoaphaca*, these were found same (2n = 14). In the dendrogram of karyologic analysis *L. aphaca* var. *affinis* and var. *pseudoaphaca* formed a small duplet group and var. *modestus* was found similar with the other big *Lathyrus* group containing var. *modestus*, *L. sativus*, *L. stenophyllus* and the others (Şahin *et al.*, 2000). The dendrogram result in this study was supported with this finding.

In the Lamarque and Guzma' n (1997) study on the *Prosopis chilensis* stated that fatty acids had no significant correlation with geographic parameters analyzed, some unsaturated fatty acids correlated significantly with altitude and the variability observed might indicate that there is sufficient intraspecific difference to permit improvement by selection and breeding. The results obtained also were very important by showing infraspecific variation determined in the *Lathyrus* taxa. It is required to enlarge the study with different parameters especially inter and intraspecific variation.Velasco & Goffman, (2000) reported that the fatty acid composition of the seed oil has been used as a criterion in searching for a ratio of infrageneric classification of *Linum* L.

Previous studies have revealed the chemotaxonomic value of fatty acids in several plant families and species (Senatore, & Basso, 1994: Agnase *et al.*, 1998) which is confirmed for the *Lathyrus aphaca* varieties in the present study. In conclusion fatty acid composition of *Lathyrus* sp. studied can be used as taxonomic markers, but it will require to make population studies by means of biosystematics in detail. A more detailed investigation could afford a proper evaluation of polymorphisms based on fatty acid distributions in *Lathyrus* genus. It is reported that, especially in the interspecific hybridization studies in genus *Lathyrus* are essential for clarifying the genetic relationships among species of this important genus (Yamamoto, 1984). Unusual and technically interesting fatty acids and their sporadic occurence in seed fats is genetically determined and they are highly significant indicators of phylogenetic relationships (Aitzetmuller, 1995).

References

Agnase, M.A., R.H. Juliani and J.L. Cabrera, 1998. Chemotaxonomic features of Lipophilic components in *Adesmia* species(Leguminosae). Pl. Syst. Evol., 210:1-2.

Aitzetmuller, K., 1993. J. High Resol.Chromatograph, 16: 488.

Aitzetmüller, K. and N. Tsevegsüren, 1994. Seed Fatty Acids, "Front–End" Desaturases and Chemotaxonomy- a case study in the Ranunculaceae, J. Pl. Physiol., 143: 538-543.

Aitzetmüller, K., 1995. Fatty acid patterns of Ranunculaceae seed oils: Phylogenetic relationships. Pl. Syst. Evol., 9: 229-240.

Bağcı, E. and A. Şahin : A Chemotaxonomic Study On Some *Lathyrus* L. (Fabaceae) Species, In View of Fatty Acid Composition. Annales Botanici Fennici, (submitted)

Badami, R.C. and K.B. Patil, 1981: Structure and Occurrence of unusual fatty acids in minor seed oils. Prog. Lipid Res., 19:119-153.

Chowdury, A.R. and R. Banerjii, 1995. Studies on Leguminosae seeds. Fett Wissenschaft Technologie – Fat Sci. Technol., 97: 457--458.

Christie, W.W., 1989. Gas Chromatography and Lipids. The Oily Press.

Daulatabad, C.D., K.M. Hosamani, V.A. Desai and K.R. Alagawadi, 1987. Cyclopropenoid Fatty acids in Leguminosae Oils. Jaocs, 64. 10: 1423

Davis, P.H., 1970. Flora of Turkey and East Aegean Island, Edinburgh University Press, 3: 328-369.

Doğan, A., A. Kence and C. Tigin, 1992. Numerical Taxonomic Study on Turkish *Lathyrus* (Leguminosae). Edinburgh, J. Bot., 49: 333-341.

Enneking, D., 1998: A bibliographic database for the genus *Lathyrus*. Co-operative Research Centre for Legumes in Mediterranean Agriculture. Occasional Publication, No. 18.

Ertekin, S.A. Ö. Saya, 1996. Türkiye Florası için yeni bir kayıt, Doğa Tr. J. of Botany, 15: 75-77.

Harborne, J.B. and B.L. Turner, 1984. Pl. Chemosyst. pp: 180-191. Academic press, London.

Hegnauer, R., 1989. Chemotaxonomie der pflanzen, 8: 611-612.

Jmbs, B.A. and L.Q. Pham, 1996. Fatty acids and Triacylglycerols in Seeds of Pinaceae Species. Phytochem., 42: 1051-1053.

Kates, M., 1986. Techniques of Lipidology , Isolating analysis and identification of Lipids. P 105. Elsevier New york.

Kupicha, F.K., 1983. The Infrageneric Structure of *Lathyrus*, Notes R. B.,G. Edinburgh, 41: 209-244.

Kwiecinska, B. and S. Matyka, 1986. Fatty acids in seeds of pea, Field pea, and Horse bean. Biuletyn Informacyjny Przemyslu Paszowego, 25. 3: 11—16.

Marin, P. D., V. Sajdl, S. Kapor, T. Budislav and B. Petkovic, 1991. Fatty acids of the Saturojoideae, Ajogoideae and Scutellarioideae (Lamiaceae). Phytochem., 30: 2982.

Markham, K.R. and K.R.W. Hammett, 1994. The Basis of yellow Colouration in *Lathyrus aphaca* flowers. Phytochem., 37:163.

Maxted, N. and D. J. Goyder, 1988. A New Species of *Lathyrus* Sect. *Lathyrus* (Leguminosae, Papilionoidae) form Turkey, Kew Bulletin, 4: 711-714.

Senatore, F. and F. Basso, 1994. Fatty acid and sterol composition from seeds of some *Lathyrus* ecotypes. Rivista Itallana Delle Sostanze Grasse, 71:11, 313.

Smith, C.R., 1970. Occurrence of Unusual Fatty Acids in Plants. In Holman, R.T. (ed.): Prog. Chem. Fats other Lipids, 11: 139-177.

Şahin, A., 1993: Türkiye'nin Bazı *Lathyrus* L. türleri [L. *rotundifolius* Willd., subsp. *miniatus* (Bieb. ex Stev.) Davis, *L. cassius* Boiss. *L. cicera* L., *aphaca* L. var. *modestus* P.H. Davis] 'in Karyotip Analizleri I. Doğa Tr. J. Bot., 17: 65-69.

Şahin, A., H. Genç and E. Bağcı, 2000. Cytotaxonomic Investigations on Some *Lathyrus* L. Species Growing in West Mediterranean and Southern Aegean Regions II. Acta Botanica Gallica, 147: 243-256.

Velasco, L. and F.D. Goffman, 2000: Tocopherol, Plastochromanol and fatty acid patterns in the genus *Linum*. Plant, Systematics and Evolution, 1-2.

Yamamoto, K., Y.Fujiwara and T. Blumenreich, 1984. Karyotypes and Morphological Characteristics of Some Species in the *Lathyrus* L., Japan J. Breed, 34: 273-284.