ACTA SOCIETATIS
BOTANICORUM POLONIAE
vol. XL — nr 3
1971

Free amino acids in different *Lotus* species

J. PRZYBYLSKA and B. CHWAŁEK

Institute of Plant Genetics, Polish Academy of Sciences, Poznań, Strzeszyńska 2/4, Poland

(Received: November 15, 1970)

Abstract:

Studies on free amino acid pools in various organs of different *Lotus* species showed mature seeds to be distinguished by a high concentration of canavanine and by the presence of γ-glutamyltyrosine and γ-glutamylphenylalanine. In seeds of *L. helleri* and *L. purshianus* homoarginine has been found which is interesting as the occurrence of this amino acid seemed to be restricted to the genus *Lathyrus*.

INTRODUCTION

The composition of free amino acids in different plant organs provides information on nitrogen metabolism in plants. High concentration of a given amino acid in conductive tissues suggests the compound to play some role in nitrogen transport. Accumulation of a particular amino acid in storage organs indicates that this very compound is of significance in nitrogen storage.

Data on free amino acid composition in plants may also be of value in taxonomic studies. Besides the twenty amino acids representing well-known protein constituents, plants are capable of synthesizing numerous related compounds usually occurring in the free state. So far about two hundred so-called "non-protein" amino acids have been recorded to occur in plants (Bell 1967). Only a few non-protein amino acids, e.g. γ-aminobutyric acid, are ubiquitous in the plant kingdom. Most of the non-protein amino acids have a restricted distribution, they therefore attract attention in chemotaxonomic studies with plants (see reviews: Przybylska 1963; Bell 1966; Kjaer 1966).

As regards food and fodder plants, the practical aspect of free amino acid studies is obvious; some non-protein amino acids are toxic and their presence in food or forage may be harmful to man and animal (see reviews: Przybylska 1965; Tschiersch 1966; Bell 1967).

Numerous studies on free amino acid composition in *Papilionaceae*, carried out in a number of laboratories, showed this family to synthesize many non-protein amino acids, some of them characteristic of tribes, genera or even groups of species

within a single genus (Tschiersch 1961; Bell 1962a; Bell and Tirimanna 1965; Przybylska and Rymowicz 1965; Dunnill and Fowden 1967; Tschiersch and Hanelt 1967; Turner and Harborne 1967). These observations gave rise to various problems at the same time inducing researchers to determine free amino acid composition in those members of *Papilionaceae* which have so far not been thoroughly examined from this point of view.

The present paper presents studies on free amino acid composition in various *Lotus* species. The genus *Lotus* comprises about 150 species, both annual and perennial, the taxons being extremely diverse in form and adapted to a wide range of ecological conditions (Hitchcock et al. 1959). The European species, *Lotus corniculatus* L., represents a successful forage crop.

MATERIAL AND METHODS

Plant material

Test material consisted of different organs of 20 *Lotus* species. Stems and leaves collected at time of bloom, flowers, young pods collected about two weeks after perianth shedding, and mature seeds were analysed. Collection of the species for analysis was started by the authors in 1965, seed material being supplied by the Forage and Range Research Branch, Beltsville, Md., U.S.A. *Lotus corniculatus* L. was represented by Polish and American cultivars, Bursztyn and Granger, respectively. Samples for analyses were harvested in 1967. A list of the examined material is presented in table 1.

Table 1

List of analysed samples

Lotus species / Organs	*L. angustissimus* L.	*L. arabicus*	*L. arenarius* Brot.	*L. chihuahuanus* Greene	*L. conimbricensis* Brot.	*L. corniculatus* L.	*L. decumbens*	*L. divaricatus*	*L. filicaulis*	*L. helleri* Britt.	*L. maroccanus* Ball	*L. mearnsii* Greene	*L. ornithopodioides* L.	*L. palustris* Willd.	*L. parviflorus* Desf.	*L. pedunculatus* Cav.	*L. purshianus*	*L. schoelleri* Schweinf.	*L. tenuis* W. et K.	*L. weilleri*
Stems	+	+	+	+	+	+	+	+	+	—	+	+	+	+	+	+	+	+	+	+
Leaves	+	+	+	+	+	+	+	+	+	—	+	+	+	+	+	+	+	+	+	+
Flowers	+	+	+	+	—	+	+	+	+	—	+	+	+	+	+	+	+	+	+	+
Young pods	—	—	—	—	+	—	+	+	+	+	—	+	—	+	+	+	+	+	+	—
Seeds	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

+ analysed, — not analysed.

In addition to the samples listed in table 1, mature seeds were analysed of 19 *Lotus* species not included in the collection. The species were as follows: *L. cytisoides*, Bizerte *L. caucasicus* Kupr., *L. commutatus* Guss., *L. conjugatus* L., *L. creticus* L., *L. cruentus*, *L. edulis* L., *L. gebelia* Vent., *L. glaucus*, *L. hispidus* Desf., *L. jacobaeus* L., *L. krylovii* Schischk., *L. major* Ldb., *L. maritima* L., *L. scoparius* (Nutt) Ottley, *L. siliquosus* L., *L. strictus* Fisch., *L. tenuifolius* L., *L. tetragonolobus* L.

For some *Lotus* species under study seed samples from different sources were analysed (see Table 2).

Seed sources:

A. North Central Regional Plant Introduction Station, Ames, U.S.A.,
B. Northeast Regional Plant Introduction Station, Geneva, N.Y., U.S.A.,
C. Department of Genetics of McGill University, Montreal, Canada,
D. Forage and Range Research Branch, Beltsville, Md., U.S.A.,
E. Botanical Garden, Hungarian Academy of Sciences, Vacratöt, Hungary,
F. Botanical Garden, Stockholm, Sweden,
G. All-Union Institute of Plant Industry, Leningrad, U.S.S.R.,
H. Royal Botanic Gardens, Kew, England,
I. Botanic Garden, Cambridge, England,
J. Jardin Botanique, Bruxelles, Belgium,
K. Institut National de la Recherche Agronomique, Ariana, Tunisie,
L. Jardin Botanique de l'Université, Copenhagen, Denmark,
M. Jardin des Plantes de la Ville, Caen, France,
N. Institut für Kulturpflanzenforschung, Deutsche Academie der Wissenschaften zu Berlin, D.D.R.,
O. Royal Botanic Gardens, Sydney, Australia.

No attempt was made by the authors to check the identification of seed samples.

Analytical procedures

The extraction of plant material was performed as reported previously (Przybylska and Rymowicz 1965). Analysis of free amino acids was carried out by two-dimensional paper chromatography and paper electrophoresis, as described in the former papers (Przybylska 1964; Przybylska and Rymowicz 1965). All the extracts were analysed before and after mild acid hydrolysis (1 N HCl for 3.5 h at 100°C, under reflux).

Sakaguchi's reagent (Smith 1960) was used to detect guanidino amino acids. Canavanine was identified with the aid of the pentacyanoammonioferrate reagent (Fearon and Bell 1955).

Identification of amino acids and of γ-glutamylpeptides was based on the comparison of their chromatographic and electrophoretic behaviour as well as of their colour reactions with corresponding features of authentic compounds. Except for L-homoarginine, γ-glutamyltyrosine and γ-glutamylphenylalanine which were supplied as gifts (see: Acknowledgements), authentic compounds used for comparison were derived from commercial sources.

RESULTS AND DISCUSSION

1. Protein amino acids

In all the analysed organs of the examined *Lotus* species high concentrations of aspartic acid with asparagine, glutamic acid with glutamine, alanine and serine have been revealed. Proline has proved to be the main free amino acid in flowers, while high concentrations of arginine were found in some seed samples only. The remaining protein amino acids — except tryptophan undetected on paper chromatograms — represented only minor constituents of ethanolic extracts.

Among the analysed organs, stems and young pods showed a particularly pronounced predominance of aspartic acid with asparagine over other free amino acids — this being due to large amounts of asparagine. The high concentration of asparagine in the above organs confirms the well known role of this amide in nitrogen transport in plants (McKee 1962).

A considerable proportion of proline in the free amino acid pool was a characteristic feature of flowers. High level of free proline in flowers of leguminous plants and in inflorescences of grasses has been already reported in the previous papers (Przybylska and Rymowicz-Dąbrowska 1970b; Rymowicz-Dąbrowska and Przybylska 1970; Dąbrowska and Przybylska 1970) and was interpreted as accumulation of free proline in pollen grains — a phenomenon observed by other authors (Bathurst 1954; Tupy 1963, 1964; Britikov et al. 1964).

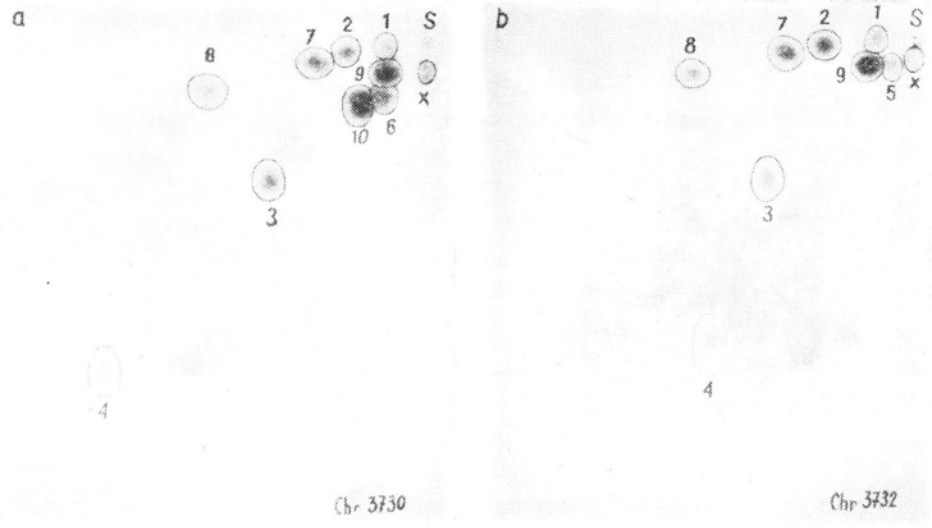
2. Detection of γ-glutamyltyrosine and γ-glutamylphenylalanine

In seeds of all the *Lotus* species examined two γ-glutamylpeptides identified tentatively as γ-glutamyltyrosine and γ-glutamylphenylalanine were observed. According to a rough estimation, on the basis of colour intensity of the respective spots, in most of the examined species the level of γ-glutamyltyrosine was several times higher than that of γ-glutamylphenylalanine and ranged from 0.05 to 0.2 per cent of dry matter. In a separate paper (Przybylska and Chwałek 1970) isolation of these compounds has been reported, in crystalline form, from seeds of *Lotus corniculatus* L.; tentative identification has been confirmed and the isolated peptides have been shown to have an L-configuration at both centres.

γ-Glutamylpeptides of tyrosine and phenylalanine seem to be widely distributed in plants. The peptides were reported to occur in different members of *Papilionaceae*: *Glycine max* (Morris and Thompson 1962), *Lupinus angustifolius* and *L. albus* (Wiewiórowski and Augustyniakowa 1962), and *Astragalus* spp. (Dunnill and Fowden 1967). Moreover, both peptides were found in *Aubrietia deltoidea* of the *Crucifereae* family (Larsen and Sorensen 1967), while γ-glutamylphenylalanine also occurred in *Allium cepa* of the *Liliaceae* family (Virtanen and Matikkala 1960). It is worth noting that the concentration of γ-glutamyltyrosine usually prevails over that of γ-glutamylphenylalanine.

3. Non-protein amino acids

From among non-protein amino acids, γ-aminobutyric acid and β-alanin were observed, in low concentrations, in a vast majority of the samples analysed. Pipecolic acid, the higher homologue of proline, was found — in rather large amounts — in young pods of 11 *Lotus* species, namely: *L. corniculatus*, *L. conimbricensis*, *L. decumbens*, *L. divaricatus*, *L. filicaulis*, *L. helleri*, *L. mearnsii*, *L. palustris*, *L. parviflorus*, *L. schoelleri* and *L. tenuis*. A high proportion of canavanine (α-amino-γ-guanidinoxybutyric acid) in the free amino acid pool (Phot. 1a, b) was a characteristic feature of mature seeds of all the species analysed. In seeds of two species, *L. helleri* and *L. purshianus*, considerable amounts of homoarginine were found (Phot. 1a).



Phot. 1. Chromatograms (fragments) of free amino acids from seeds of *Lotus purshianus* (a) and *L. conjugatus* (b).

Key to spots: *1* — aspartic acid; *2* — glutamic acid; *3* — alanine; 4 — methionine and/or valine; *5* — lysine; *6* — arginine; *7* — γ-glutamyltyrosine; *8* — γ-glutamylphenylalanine; *9* — canavanine; *10* — homoarginine; *x* — unidentified compound. *S* — starting point

Both γ-aminobutyric acid and β-alanine are very widely distributed in plants and their detection is to be expected in any plant sample thoroughly examined. Pipecolic acid is also a rather common plant constituent and is known to occur in various *Papilionaceae* species (Morrison 1953). Its relatively high concentration in young fruits or young seeds of some of these plants should be emphasized (Simola 1968; Przybylska and Rymowicz-Dąbrowska 1970a, 1970b). Large amounts of pipecolic acid in young seeds, and lack of detectable amounts of this amino acid in mature seeds of *Vicia villosa euvillosa*, suggested that pipecolic acid may be used for metabolic processes in the course of seed ripening (Przybylska and Rymowicz-Dąbrowska 1970a).

As regards canavanine, this amino acid is known as a nitrogen-storing compound in *Papilionaceae* (Tschiersch 1959; Bell 1960). Within *Papilionaceae*, canavanine is a very widespread compound (Turner and Harborne 1967). It must be, however, mentioned that within particular tribes the occurrence of canavanine is restricted to some genera only, and within individual genera there are canavanine-containing species and species in which this amino acid has not been detected. Turner and Harborne (1967) express the opinion that "canavanine arose very early in the evolution of the subfamily* and that it has been maintained in several divergent stem lines arising out of the ancestral complex".

As canavanine was found to be toxic for animals (see review: Tschiersch 1966), it should be noted that except for seeds no detectable amounts of this amino acid have been found in the analysed material.

3a. Detection of homoarginine

Detection of homoarginine in seeds of two *Lotus* species should be emphasized as its occurrence in plants seemed to be restricted to a number of *Lathyrus* species. To the authors' knowledge, despite of extensive surveys of free amino acids in *Papilionaceae*, homoarginine has not been found in any other genus of this family. Moreover, to the authors' knowledge, the occurrence of homoarginine in other plant families has not been reported.

Homoarginine belongs to recently-discovered non-protein amino acids. The first report on the natural occurrence of homoarginine comes from Bell, who identified it in many *Lathyrus* species (1962 a) and who isolated L-homoarginine from *Lathyrus cicera* (1962 b). Later, Rao and coworkers (1963) isolated L-homoarginine from *Lathyrus sativus*. A high level of homoarginine in seeds of many *Lathyrus* species (Bell 1962 a; Przybylska and Rymowicz 1965) suggests the compound to play some role in nitrogen storage. Experiments with chicks (Rao et al. 1964) and rats (see review: Fowden et al. 1967) did not reveal any toxic effect of homoarginine. The studies of Ryan and Wells (1964) indicate that in rat and man homoarginine is the product of lysine metabolism; ingestion of lysine by human adults resulted in an increased urinary excretion of homocitrulline and homoarginine.

To confirm the tentative identification of homoarginine in *Lotus*, the compound was isolated, in crystalline form, from seeds of *Lotus helleri* (Chwałek and Przybylska 1970). Characterization of the isolated compound and its comparison with an authentic amino acid showed the compound to be L-homoarginine.

The occurrence of homoarginine in various species of *Lathyrus* and in two species of *Lotus* — genera belonging to different tribes, *Vicieae* and *Loteae*, respectively — indicates that homoarginine may be more widely distributed within *Papilionaceae*.

---

* *Papilionoideae*

Simultaneous occurrence of large amounts of homoarginine and canavanine in both *Lotus helleri* and *L. purshianus* should be noted. Though canavanine is widely distributed within *Papilionaceae*, it could not be detected in *Lathyrus* (Bell 1966) — the genus being distinguished by the occurrence of homoarginine in many species.

In the present studies, covering seeds of 39 *Lotus* species, only two of the examined species, *L. helleri* and *L. purshianus*, were shown to contain homoarginine. The homoarginine-containing *Lotus* species are North-American species which, according to some systematists (Torrey and Gray 1838; Gray 1863; Watson 1876), should be referred to a separate genus *Hosackia*. It must be, however, noted that three other North-American *Lotus* species under study, *L. scoparius*, *L. chihuahuanus* and *L. mearnsii*, did not exhibit any detectable amounts of homoarginine in seeds. As North-American *Lotus* species represent a large group, comprising about 60 taxons (Zandstra and Grant 1968), it would be interesting to check whether the occurrence of homoarginine is a rather common feature in this group or if it is perhaps restricted to only a few species.

4. Variations in free amino acid levels in seeds of some *Lotus* species

As it is known, the chemical composition of plants depends considerably on growth conditions. The composition of seeds, as compared to that of other organs, is relatively little affected by environmental factors prevailing during plant growth (Dunnill and Fowden 1965). Nevertheless, significant variations in free amino acid concentrations in seeds of a single species from different sources have been reported (Bell and Tirimanna 1965).

To gain information on variations in the free amino acid levels in seeds of individual *Lotus* species, for some of the analysed species seed samples from different sources were examined. Rough estimations of several ethanol-soluble ninhydrin-positive compounds in this material are presented in Table 2. The remarkable differences between the samples of a single species should be emphasized. Variations in free arginine level are most marked in *Lotus hispidus*, *L. maritima* and *L. purshianus*. Within *L. edulis* and *L. purshianus* likewise significant differences were observed in the relative concentration of γ-glutamyltyrosine. The level of γ-glutamylphenylalanine also varied depending on the seed source, however, as the peptide occurred in low concentration, the variations were not so conspicuous. From among seven samples of *Lotus purshianus* seeds, five samples contained large amounts of homoarginine, in one sample homoarginine occurred as a minor free amino acid, and in one sample homoarginine was not detected by the analytical procedure employed. As regards canavanine, because of a very high concentration of the compound in most of the analysed samples, variations in its level could not be observed under the conditions applied.

The above presented variations in the concentrations of several ninhydrin-positive compounds in seeds of individual *Lotus* species visualize the hazard of using an arbitrary concentration of a single compound as a criterion of relationship

Table 2

Variations in the relative concentrations of some ninhydrin-positive compounds in seeds of individual *Lotus* species from different sources as judged by ninhydrin reaction

(Capital letters indicate seed source — for key see ,,Materials and methods", letter "a" beside capital letters indicates that seeds were reproduced in authors' own collection)

Species	Seed source	Spot intensities				
		Can	Arg	H-arg	Glu-tyr	Glu-phe
L. angustissimus	A	S	M	O	W	T
	M	M	M	O	W	T
L. creticus	K	W	T	O	W	T
	L	M	W	O	W	W
L. edulis	D	M	T	O	W	T
	Da	M	M	O	M	T
	H	M	T	O	W	T
L. helleri	D	M	T	S	W	T
	Da	S	T	M	W	T
L. hispidus	A	M	W	O	M	W
	M	S	S	O	W	T
	H	M	M	O	W	W
L. major	G	M	O	O	T	T
	G_1	M	O	O	T	T
L. maritima	K	M	W	O	W	T
	I	W	S	O	M	W
	Ia	M	W	O	W	W
L. ornithopodioides	A	S	W	O	W	T
	L	M	T	O	W	T
	H	S	T	O	W	T
L. parviflorus	C	S	M	O	W	T
	F	S	M	O	W	T
	A	S	W	O	W	W
L. purshianus	D	S	S	S	T	T
	Da	S	W	S	T	T
	F	S	T	S	W	T
	Aa	S	T	S	M	W
	C	S	M	W	S	W
	B	S	S	S	W	T
	B_1	S	W	O	W	W
L. tenuis	A	S	T	O	W	T
	G	M	T	O	W	T
L. tetragonolobus	H	M	O	O	W	W
	I	S	O	O	W	T

[1]) another origin.

Abbreviations: Can — canavanine, Arg — arginine, H-arg — homoarginine, Glu-tyr — γ-glutamyltyrosine, Glu-phe — γ-glutamylphenylalanine.

Symbols denote relative strengths of spots on two-dimensional chromatograms: O—absent; T—trace; W—weak; M—medium; S—strong;

Remark: volumes of extracts applied on chromatograms were equivalent to 20 mg of original seed meal.

between species. The presence of an unusual amino acid, irrespective of its concentration, is more valuable for taxonomic purposes as it indicates the existence of a genetically-controlled metabolic pathway, characteristic of the plant group containing the unusual compound. It should, however, be borne in mind that the concentration of the unusual compound may remain beyond the level detectable by the analytical procedures employed. Consequently, data on distribution of unusual compounds in plants may be of value for taxonomy when linked with other characteristics of the taxons investigated.

SUMMARY

Free amino acid composition in different organs of 20 *Lotus* species has been determined by means of paper chromatography and electrophoresis. Stems and leaves sampled at time of bloom, flowers, young pods and mature seeds were analysed. Moreover, for 19 other *Lotus* species, in the same way, exclusively mature seeds were analysed. In respect to 12 *Lotus* species seed samples from different sources were examined.

As regards characteristic features of the analysed organs, high levels of free aspartic acid and asparagine in stems and young pods, and considerable participation of proline in the free amino acid pool of flowers should be pointed out. Mature seeds were distinguished by a high concentration of canavanine — amino acid unobserved in other organs under study.

In mature seeds of all the analysed *Lotus* species, γ-glutamyltyrosine and γ-glutamylphenylalanine were found; the latter peptide occurred in smaller amounts.

From among non-protein amino acids, apart from the above mentioned canavanine, in most of the analysed samples γ-aminobutyric acid and β-alanine were observed, while in young pods of 11 *Lotus* species pipecolic acid was detected. In seeds of *Lotus helleri* and *L. purshianus* homoarginine was found. As the occurrence of homoarginine in the plant kingdom seemed to be restricted to the genus *Lathyrus* only, the discovery of this amino acid in *Lotus* species is interesting from the taxonomical point of view.

Seed samples of individual *Lotus* species from different sources varied in respect to free amino acid composition.

Acknowledgements

The authors are grateful to Professor C. J. Morris, U.S. Department of Agriculture, Ithaca New York, for the samples of authentic γ-glutamyltyrosine and γ-glutamylphenylalanine. More over, the authors are greatly indebted to Professor E. A. Bell, University of Texas, and to Professo C. E. Dent, University of London, for samples of authentic L-homoarginine. The skilful technical assistance of B. Czerwińska, Sc. B. is acknowledged.

This work was supported in part by a research grant from the U.S. Department of Agriculture

REFERENCES

Bathurst N. O., 1954, The amino acids in grass pollen, J. Exp. Bot. 5: 253–256.

Bell E. A., 1960, Canavanine in the *Leguminosae*, Biochem. J. 70: 618–620.

Bell E. A., 1962a, Associations of ninhydrin-reacting compounds in the seeds of 49 species of *Lathyrus*, Biochem. J. 83: 225–229.

Bell E. A., 1962b, The isolation of L-homoarginine from seeds of *Lathyrus cicera*, Biochem. J. 85: 91–93.

Bell E. A., Tirimanna A. S. L., 1965, Associations of amino acids and related compounds in

the seeds of forty-seven species of *Vicia*: their taxonomic and nutritional significance, Biochem. J. 97: 104—111.

Bell E. A., 1966, Amino acids and related compounds. In: Swain T. (ed.). Comparative Phytochemistry, Academic Press, London and New York.

Bell E. A., 1967. Some unusual plant amino acids and their effects in animals, Seventh International Congress of Biochemistry, Tokyo, August 19–25, Abstracts Col. IV-5.

Britikov E. A., Musatova N. A., Vladimirtseva S. V. and Protsenko M. A., 1964, Proline in the reproductive system of plants. [In: Linskens H. F. (ed.). Pollen physiology and fertilization. North Holland Publishing Co., Amsterdam, pp. 77–85].

Chwałek B., Przybylska J., 1970, L-Homoarginine in *Lotus helleri*, Bull. Acad. Polon. Sci., Ser. sci. biol., 18:603-605.

Dąbrowska T., Przybylska J., 1970, Free amino acids in leaves and inflorescences of 34 grass species, Acta Soc. Bot. Polon., 39:445-452.

Dunnill P. M., Fowden L., 1965, The amino acids of seeds of the *Cucurbitaceae*, Phytochemistry 4: 933–944.

Dunnill P. M., Fowden L., 1967, The amino acids of the genus *Astragalus*, Phytochemistry 6: 1659–1663.

Fearon W. R., Bell E. A., 1955, Canavanine: detection and occurrence in Colutea arborescens, Biochem. J. 59: 221–223.

Fowden L., Lewis D. and Tristram H., 1967. Toxic amino acids: their action as antimetabolites, Adv. in Enzymol. 29: 90–163.

Gray A., 1863, Synopsis of the species of Hosackia. Proc. Acad. Nat. Sci. Phila 15: 346–352*.

Hitchcock C. L., Cronquist A. and Thompson J. W., 1959, Vascular plants of the Pacific Northwest, Part 3, University of Washington Press, Seattle.

Kjaer A., 1966, The distribution of sulphur compounds. [In: Swain T. (ed). Comparative Phytochemistry, Academic Press, London and New York].

Larsen P. O., Sorensen H., 1967, γ-Glutamyl-phenylalanine and γ-L-glutamyl-L-tyrosine from seeds of Aubrietia deltoidea D. C., Acta Chem. Scand. 21: 2908–2909.

McKee H. S., 1962, Nitrogen metabolism in plants, Clarendon Press, Oxford.

Morris C. J., Thompson J. F., 1962, The isolation and characterization of γ-L-glutamyl-L-tyrosine and γ-L-glutamyl-L-phenylalanine from soybeans, Biochemistry 1: 706–709.

Morrison R. J., 1953, The isolation of L-pipecolic acid from Trifolium repens, Biochem. J. 53: 474–478.

Przybylska J., 1963, Wolne aminokwasy, ich rola w metabolizmie i znaczenie w taksonomii roślin, Post. Biochem. 9: 105–127.

Przybylska J., 1964, The role of tingitanine in nitrogen metabolism of Lathyrus tingitanus, Acta Soc. Bot. Polon., 33: 211–241.

Przybylska J., 1965, Niebiałkowe aminokwasy w pokrewnych rodzajach Lathyrus i Vicia oraz ich działanie na organizm zwierzęcy, Post. Nauk Roln. 3 (93): 51–66.

Przybylska J., Chwałek B., 1970, γ-L-glutamyl-L-tyrosine and γ-L-glutamyl-L-phenylalanine in *Lotus corniculatus* L., Bull. Acad. Polon. Sci., Ser. sci. biol. 18: 249–252.

Przybylska J., Rymowicz T., 1965, Free amino acids in different organs of 16 Lathyrus species, Genetica Polonica 6: 91–124.

Przybylska J., Rymowicz-Dąbrowska T., 1970a, Wolne aminokwasy w roślinach pastewnych. I. Wolne aminokwasy w różnych organach niektórych gatunków jednorocznych roślin motylkowych, Roczn. Nauk Roln. 96-A-3: 189–201.

Przybylska J., Rymowicz-Dąbrowska T., 1970b, Wolne aminokwasy w roślinach pastewnych. II. Wolne aminokwasy w różnych organach niektórych gatunków wieloletnich roślin motylkowych, Roczn. Nauk Roln., 96-A-4:25-33.

Rao S. L .N., Ramachandran L. K., Adiga P. R., 1963, The isolation and characterization of L-homoarginine from seeds of *Lathyrus sativus*, Biochemistry 2: 298–300.

Rao S. L. N., Adiga P. R. and Sarma P. S., 1964, The isolation and characterization of

β-N-oxalyl-L-α, β-diaminopropionic acid: a neurotoxin from the seeds of Lathyrus sativus Biochemistry 3: 432–435.

Ryan W. L., Wells I. C., 1964, Homocitrulline and homoarginine synthesis from lysine, Science, 144: 1122–1127.

Rymowicz-Dąbrowska T., Przybylska J., 1970, Wolne aminokwasy w roślinach pastewnych. III. Wolne aminokwasy w różnych organach niektórych gatunków traw, Roczn. Nauk Roln., 97-A-1: 15-19.

Simola L. K., 1968, Comparative studies of the amino acid pools of three *Lathyrus* species, Acta bot. fenn. 81: 1–62.

Smith I., 1960, Chromatographic and electrophoretic techniques, Heineman W. (éd.). Medical Books, Ltd., London.

Torrey G. S., Gray A., 1838, Flora of North America.*

Tschiersch B., 1959, Über Canavanin, Flora 147: 405–416.

Tschiersch B., 1961, Über das Vorkommen von Canavanin, Flora 150: 76–94.

Tschiersch B., 1966, Toxische Aminosäuren, Die Pharmatzie 21: 445–457.

Tschiersch B., Hanelt P., 1967, Die freien Aminosäuren der Samen von Vicia L. und die systematische Gliederung der Gattung, Flora 157: 389–406.

Tupy J., 1963, Free amino acids in apple pollen from the point of view of its fertility, Biol. Plant. 5: 154–160.

Tupy J., 1964, Metabolism of proline in styles and pollen tubes of Nicotiana alata. [In: Linskens H. F. (ed.). Pollen physiology and fertilization. North Holland Publishing Co., Amsterdam].

Turner B. L., Harborne J. B., 1967, Distribution of canavanine in the plant kingdom, Phytochemistry 6: 863–866.

Virtanen A. I., Matikkala E. J., 1960, Neue γ-Glutamylpeptide in der Zwiebel (*Allium cepa*), Hoppe-Seyler's Z. Physiol. Chem. 322: 8–20.

Watson S., 1876, [In: Botany of California, vol. 1, Brewer W. W., Watson S. and Gray A. (ed.)]*

Wiewiórowski M., Augustyniakowa H., 1962, Occurrence of γ-L-glutamyl-L-tyrosine and γ-L-glutamyl-L-phenylalanine in seeds of *Lupinus angustifolius* and *Lupinus albus*, Acta Biochim. Polon. 9: 399–409.

Zandstra I. I., Grant W. F., 1968, The biosystematics of the genus *Lotus* in Canada. I. Cytotaxonomy, Can J. Bot. 46: 557–583.

Wolne aminokwasy u różnych gatunków Lotus

Streszczenie

Przy pomocy chromatografii i elektroforezy bibułowej oznaczono skład wolnych aminokwasów w różnych organach 20 gatunków *Lotus*. Analizowano łodygi i liście pobrane w fazie kwitnienia roślin, kwiaty, młode strąki oraz dojrzałe nasiona. Ponadto, dla 19 innych gatunków *Lotus* analizowano wyłącznie dojrzałe nasiona. W przypadku 12 gatunków *Lotus* analizowano nasiona pochodzące z różnych źródeł.

Odnośnie charakterystycznych cech poszczególnych typów organów, podkreślić należy szczególnie wysoki poziom kwasu asparaginowego i asparaginy w łodygach i w młodych strąkach oraz znaczny udział proliny w puli wolnych aminokwasów kwiatów. W dojrzałych nasionach dominującym wolnym aminokwasem była, nie wykryta w innych analizowanych organach, kanawanina.

W dojrzałych nasionach wszystkich analizowanych gatunków zaobserwowano γ-glutamylotyrozynę oraz, występującą w mniejszej ilości , γ-glutamylofenyloalaninę.

---

* Cited after Callen E. O., 1959, Studies in the genus *Lotus* (*Leguminosae*). I. Limits and subdivisions of the genus, Can. J. Bot. 37: 157–167.

Spośród niebiałkowych aminokwasów, oprócz wspomnianej wyżej kanawaniny, w większości analizowanych prób zaobserwowano kwas γ-aminomasłowy i β-alaninę, a w młodych strąkach 11 gatunków kwas pipekolinowy. W dojrzałych nasionach *Lotus helleri* oraz L. *purshianus* stwierdzono obecność homoargininy. Ponieważ występowanie homoargininy w świecie roślin zdawało się ograniczać do rodzaju *Lathyrus*, stwierdzenie tego aminokwasu w rodzaju *Lotus* ciekawe jest z taksonomicznego punktu widzenia.

Próby nasion poszczególnych gatunków *Lotus*, pochodzące z różnych źródeł, wykazywały różnice w składzie wolnych aminokwasów.