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The amino acid composition of cotyledons, testa, embryo and protein fractions of bean seeds

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Abstract

The contribution of cotyledons, embryo and testa to the whole seed, was analyzed in the bean cultivar, Wiejska. The total nitrogen content and amino acid composition of morphological parts of the seed were determined. The average amino acid composition of globulins and albumins and the content of free amino acids in seeds of six Polish cultivars were estimated as well. It was found that the embryo contained the highest quantity of total nitrogen and the lowest of protein nitrogen. The exogenous amino acid content in the embryo was higher than in cotyledons and testa. Both albumins and globulins were shown to contain 42% exogenous amino acids. The content of methionine – the first limiting amino acid of bean proteins – did not exceed 0.30% of the total amino acid content in albumins and globulins. Free glutamic and aspartic acids made up more than 60% of the total free amino acids.

INTRODUCTION

Cotyledons are the dominating morphological components of bean seeds. The proportions, by weight, of different parts of the seed are approximately cotyledons – 90%, testa – 9%, embryo – 1% (Otoul, 1969; Singh et al., 1968). Woolfe and Hamblin (1974) tested two cultivars of bean and although there was a considerable difference in the seed weights, the percentage contributions of the main fractions were almost identical and similar to those reported elsewhere (Otoul, 1969; Singh et al., 1968). The crude protein content of the testa in those varieties was low.

From a nutritional point of view, the quantitative ratio between albumins and globulins in seeds is very important. It is suggested that albumins contain at least twice as much methionine as globulins (Marquez and Lajolo, 1981). Other investigators (Pant and Tulsiani, 1969; Palmer et al., 1973) have shown that the methionine content in albumins approximates that of globulins.

There is no information available in literature concerning the amino acid composition of bean embryo, cotyledons or testa. For this reason such studies were undertaken. Analysis of total nitrogen content and amino acid composition of seed parts was carried out on the bean cultivar Wiejska. The amino acid composition of albumins, globulins and the free amino acid content of whole seeds were estimated on 6 Polish bean cultivars grown for dry seeds.

METERIALS AND METHODS

The percentage contribution of cotyledons, embryo and testa was analyzed in seeds of the Wiejska cultivar. The total nitrogen content was analyzed by the Kjeldahl method.

Hydrolysis of proteins was carried out in 6 N hydrochloric acid at a temperature of 110°C for a period of 24 hours (Bkackburn, 1968; Kosson and Horbowicz, 1979). Amino acids were separated and determined quantitatively using an AAA 881 automatic amino acid analyzer by ion-exchange chromatography according to Spackman et al. (1958). The free amino acids were extracted from homogenized bean seeds with an 80% solution of ethanol and then analyzed as the protein amino acids. The amino acid compositions of albumins, globulins and free amino acids of seeds were determined in cultivars: Bomba, Słowianka, Igołomska, Piękny Jaś, Biała Wyborowa and Jubilatka.

RESULTS AND DISCUSSION

1. Total nitrogen content in cotyledons, embryo and testa

It was shown that the total nitrogen content and total protein content, as N total $\times 6.25$, was the highest in the embryo and lowest in the testa.

The total protein content in cotyledons determines the average protein content of whole seeds because of its contribution, by weight, in the seed. In the case of the Wiejska cultivar, cotyledons make up 92.8% of seed weight, testa and embryo 6.5% and 0.7%, respectively (Table 1).

According to Singh et al. (1968) and Otoul (1962) the cotyledons: testa : embryo ratio, by weight, is 90:9:1. The high protein contetn in the embryo and low protein content in the testa, given in Table 1, confirm the earlier observations of the above mentioned authors. Woolfe and Hamblin (1974) reported that cotyledon + embryo : testa ratio can be as 9.1:8.7 or 90.5:9.5. Our results indicate a lower percentage contribution of the testa in whole seeds. The protein content in the cotyledons and embryo of the investigated bean seeds was 3 times higher as compared to the testa.

Table 1

The content of total nitrogen and total protein in the different parts of the bean seed of cultivar Wiejska

	Whole seed	Cotyledon	Testa	Embryo	The sum of cotyledon + embryo
Total nitrogen (%of dry matter)	3.93±0.11*	4.09±0.08	1.29±0.08	7.09±0.10	4.11
Total protein (% of dry matter)	24.58±0.68	25.56±0.50	8.06±0.50	44.31±0.63	25.71
The proportions, by weight, of different parts of the seed	100	92.8	6.5	0.7	93.5

*Mean value ± confidence limits at p = 0.95 evaluated according to the Dean and Dixon test.

2. The amino acid content in cotyledons, testa and embryo

The exogenous and endogenous amino acid content in cotyledons, testa and embryo of the bean Wiejska is given in Table 2. The sum of the amino acid content is the highest in the testa, 95.84 g/16 g of nitrogen and the lowest in the embryo, 71.81 g/16 g of nitrogen. The sum of the amino acid content indicates indirectly the protein nitrogen content. It appears that morphological fractions with a higher content of total nitrogen are characterized by a lower protein nitrogen content and vice versa. Each exogenous amino acid content, expressed in g/16 g of nitrogen, was lower in the embryo as compared to cotyledons and testa. This results from the lower percentage of protein nitrogen in the embryo than in other fractions. In the case of some amino acids in cotyledons, testa and embryo, these differences are statistically significant. Differences in the endogenous amino acid content between the analyzed fractions were found to be the highest for arginine, aspartic acid, serine and glycine. It was observed that the embryo contained 2 times as much arginine and 2 times less serine, as compared to cotyledons. In testa, 2 times as much glycine was found than in cotyledons and embryos.

3. Amino acid composition of protein fractions

The contents of all amino acids (in g/16 g of N) of one protein fraction in seeds of one cultivar were added up and then the percentage of each amino acid was calculated. Results given in Table 3 for albumins and globulins are the average value for six cultivars of bean.

It was found that globulins contain 42.6% exogenous amino acids, and albumins contain 42.3%. Among the exogenous amino acids, the leucine

Table 2

Amino acid composition of different parts of bean seed; cultivar – Wiejska (g/16 g of N)

Exogenous amino acids	Part of seed			Endogenous amino acids	Part of seed		
	testa	cotyledon	embryo		testa	cotyledon	embryo
Lysine	6.00±0.38*	5.69±0.17	4.81±0.26	Arginine	7.65±0.38	5.71±0.26	12.32±0.32
Histidine	2.97±0.00	2.54±0.03	2.19±0.00	Aspartic acid	11.35±0.73	11.47±0.17	7.55±0.13
Threonine	4.31±0.39	3.57±0.00	3.40±0.26	Serine	6.75±0.46	6.08±0.00	3.75±0.10
Valine	6.16±0.17	2.97±0.14	2.30±0.23	Glutamic acid	14.01±0.05	14.26±0.47	11.43±0.21
Methionine	1.06±0.18	0.85±0.06	1.06±0.19	Proline	5.32±0.08	4.00±0.18	4.95±0.39
Isoleucine	2.15±0.29	6.11±0.26	4.30±0.21	Glycine	7.75±0.55	3.42±0.26	3.36±0.22
Leucine	6.90±0.29	6.11±0.26	4.30±0.21	Alanine	5.21±0.41	3.54±0.65	4.03±0.23
Tyrozine	3.95±0.29	3.14±0.26	2.30±0.09				
Phenylalanine	4.40±0.20	4.64±0.16	2.45±0.17				
Sum of amino acids	37.90	35.62	27.11	Sum of amino acids	58.04	48.48	47.39

Table 3

Amino acid content in albumins and globulins of bean proteins (as percent of total amino acid content)

Exogenous amino acids	Albumins	Globulins	Endogenous amino acids	Albumins	Globulins
Lysine	6.95±0.68*	6.38±1.06	Arginine	3.95±1.00	5.65±0.62
Histidine	2.09±0.35	3.29±0.76	Aspartic acid	12.01±1.12	12.55±1.17
Threonine	7.16±0.72	4.41±0.22	Serine	7.77±1.21	6.13±0.18
Valine	6.93±0.87	5.05±0.27	Glutamic acid	13.40±0.88	20.56±0.67
Methionine	0.18±0.10	0.29±0.10	Proline	7.30±2.23	3.58±0.40
Isoleucine	4.03±0.68	4.28±0.23	Glycine	4.24±0.37	4.53±0.24
Leucine	6.73±0.78	8.30±0.26	Alanine	5.68±0.34	4.49±0.18
Tyrozine	4.56±0.58	3.98±0.16			
Phenylalanine	5.49±0.35	6.59±0.21			
Sum of amino acids	44.12	42.57	Sum of amino acids	54.35	57.49

*Average value for six cultivars.

content appeared to be the highest in albumins and globulins. A significant differences in the threonine content between albumins and globulins was also observed. Methionine is the limiting amino acid for albumins and globulins, and its content in both fractions did not exceed 0.30% of total amino acids. A significantly higher quantity of serine, alanine and valine was found in albumins, while of histidine, aspartic acid, arginine, leucine and phenylalanine in globulins. In the case of the proline content albumins and globulins differed by a factor of two.

Aspartic acid and glutamic acid were the dominating endogenous amino acids of both fractions. At the same time, albumins contained equal quantities of both amino acids but globulins contained one and a half times more glutamic acid as compared to aspartic acid.

The results obtained for the amino acid composition of albumins and globulins confirm the earlier investigations of Palmer et al. (1973), and Pant and Tulsiani (1969), who stated that albumins do not differ from globulins in their methionine content.

4. Occurrence of free amino acids in bean seeds

Part of amino acids in bean seeds occur as non-protein amino acids. Investigations have shown that the total free amino acid content does not exceed 1.5% of the protein content. In the total amino acid balance of beans, the free amino acids make only a slight contribution and they do not affect the nutritive value of bean seeds. The high contribution of aspartic and

glutamic acids to free amino acids is the peculiarity of the bean. These amino acids make up more than 60% of the total free amino acids in each of the investigated cultivars (Table 4). It is interesting, that exogenous amino acids, lysine, isoleucine, leucine, tyrosine and phenyloalanine make a low contribu-

Table 4

The free amino acid content in bean seeds (as a percent of total amino acid contents)

Amino acid	Cultivar					
	Bomba	Słowianka	Igołomska	Piękny Jaś	Biała Wyborową	Jubilatka
Lysine	0.66±0.33	0.52±0.16	0.66±0.17	0.49±0.17	0.46±0.13	0.46±0.16
Histidine	2.59±0.53	1.67±0.38	2.76±0.41	1.05±0.03	1.73±0.35	1.73±0.40
Arginine	12.58±0.43	7.67±1.13	7.80±2.58	3.33±0.30	10.57±1.72	11.34±0.00
Aspartic acid	42.73±6.58	45.02±2.42	44.35±3.25	42.87±0.04	43.74±5.48	49.82±6.59
Threonine	6.36±1.01	9.89±1.44	10.5±0.18	5.23±1.39	9.18±0.29	4.87±0.12
Glutamic acid	14.23±1.11	19.59±1.92	16.62±0.43	33.62±2.89	22.77±1.54	21.81±0.54
Glycine	1.96±0.36	1.80±0.35	1.83±0.44	3.42±0.29	1.08±0.21	0.95±0.30
Alanine	4.07±0.56	4.53±0.83	2.65±0.73	10.76±1.22	3.86±0.43	2.44±0.56
Valine	2.42±0.27	3.83±0.44	3.51±0.30	3.44±0.73	2.55±0.39	3.36±0.51
Isoleucine	0.69±0.36	2.15±0.27	0.73±0.19	0.55±0.00	0.45±0.13	0.33±0.07
Leucine	0.91±0.04	1.84±0.18	0.99±0.16	1.22±0.05	1.16±0.08	0.75±0.13
Tyrozine	0.93±0.16	1.11±0.30	0.79±0.18	1.00±0.33	1.12±0.29	0.68±0.16
Phenylalanine	1.03±0.26	1.37±0.10	1.15±0.20	0.87±0.17	1.34±0.20	1.14±0.26

tion to the free amino acids and that their content does not exceed 3% of the total free amino acids. Methionine occurs only in trace quantities similarly to proline and serine (not shown in Table 4). The above results confirm earlier investigations by Posypanov and Bukhanova (1980) who have stated among others that aspartic and glutamic acid make up 56% of the total free amino acid content.

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Skład aminokwasowy bielma, pokrywy nasiennej, zarodka i frakcji białkowych nasion fasoli

Streszczenie

Na przykładzie odmiany Wiejska analizowano wagowy udział poszczególnych części morfologicznych: okrywy, zarodka i bielma w nasionach fasoli. Określono również zawartość azotu ogólnego w wymienionych częściach oraz ich skład aminokwasowy. Na przykładzie sześciu odmian fasoli: Bomba, Słowianka, Igołomska, Piękny Jaś, Biała Wyborowa i Jubilatka oznaczono średni skład aminokwasowy albumin i globulin oraz zawartość wolnych aminokwasów w nasionach. Stwierdzono m.in., że zarodek charakteryzuje się najwyższą zawartością azotu ogólnego, a najniższą azotu białkowego. Zawartość aminokwasów egzogennych w zarodku jest wyższa w porównaniu z bielmem i okrywą, i wynika to z wyższej zawartości azotu białkowego w zaroku. Ponad 60% ogólnej zawartości wolnych aminokwasów stanowi kwas glutaminowy i asparaginowy. Albuminy nie różnią się od globulin sumaryczną zawartością aminokwasów egzogennych. Zawartość metioniny w albuminach i globulinach nie przekracza poziomu 0,30% ogólnej puli aminokwasów.