# Amino acid contents and biological value of protein in various amaranth species

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**ABSTRACT**: Amino acid content before and after heat treatment was assessed in grain of six selected amaranth varieties and four species: *Amaranthus cruentus, A. hypochondriacus, A. caudatus* and *A. hybridus*, cultivated in the Czech Republic. High content of Lys and Arg was detected in both heat treated and untreated grains, as well as satisfactory content of Cys and lower levels of Met, Val, Ile and Leu. The latter three amino acids appear as limiting. Chemical scores of essential amino acids and essential amino acid index (EAAI) were determined. EAAI value of 90.4% shows the favourable nutritional quality of amaranth protein, which is almost comparable with egg protein. Heat treatment by popping at 170 to 190°C for 30 s resulted in decreased EAAI to 85.4%. Of the essential amino acids under study, Val and Leu contents decreased significantly ($P < 0.05$). The relatively high content of essential amino acids in amaranth grain predetermines its use as a substitution of meat-and-bone meals.

**Keywords**: amaranth grain; popping; essential amino acids; chemical score; EAAI

Alternative sources of feeds offering high-quality nutrients, especially proteins, limiting amino acids but also energy, could be found in unconventional plants. One of the possible ways of replacing feeds of animal origin in the rations for monogastric animals is the use of amaranth and its processed products, which meet the demands for substitution of meat-and-bone meals (Herzig, 2001).

Grain of the current amaranth species is of high nutritional value. Dry matter content ranges from 90 to 94%, N-substances from 15 to 18%, ether extract (fat) from 6 to 8%, crude fibre from 3 to 5%, ash from 2 to 3%, and nitrogen-free extracts range between 60 and 65%. The advantage of amaranth grains compared to conventional cereals is a relatively high content of proteins and more balanced composition of essential amino acids (Carlsson, 1979; Bressani *et al.*, 1987b; Szelényi-Galántai and Zsolnai-Harszi, 1992; Andrasofszky *et al.*, 1998).

All species under study show favourable amino acid composition. Amaranth grain is rich in lysine and tryptophan, which are comparable to proteins of animal origin; the levels of sulphur amino acids are also higher, the content of leucine is low (Bressani *et al.*, 1992), and the limiting amino acid is threonine (Jarošová *et al.*, 1997). Lysine content of *A. hypochondriacus* 5.95 g/16 g N is higher compared to 2.90 g/16 g N in wheat flour (Dodok *et al.*, 1997). A suitable content of lysine and tryptophan together with low content of leucine makes it a high-quality supplement e.g. for maize, which is rich in leucine but poor in lysine and tryptophan (Vetter, 1994). Amaranth grain contained (g/16 g N) 7.00 lysine, 4.26 methionine + cystine, 5.56 valine, 7.48 leucine, 4.85 isoleucine, and 13.05 arginine (Tovar *et al.*, 1989). According to Bressani *et al.* (1987a), lysine content (g/16 g N) varied in *Amaranthus caudatus* from 5.55 to 6.44, in *A. cruentus* from

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4.94 to 5.50, and in *A. hypochondriacus* from 4.68 to 5.62. Tryptophan levels varied from 1.36 to 1.70 in *A. caudatus*, 1.10 to 1.87 in *A. cruentus*, 1.19 to 1.79 in *A. hypochondriacus*, and 1.45 in *A. hybridus*. High levels of lysine, methionine and arginine in amaranth grains were demonstrated by Gorinstein *et al.* (2002).

The relatively high content of essential amino acids in amaranth grain predetermines its use as a substitution of conventional cereals (Gorinstein *et al.,* 2002). According to Aritsuka *et al.* (1994), supplementation of 5% amaranth into the basic diet of rats induced growth inhibition, but addition of 5 or 10% sugar beet fibre and soybean protein reduced growth inhibition. To increase the nutritional value of amaranth, grain is treated by popping and roasting at 170 to 190°C, either at normal or increased pressure (Jarošová *et al.,* 1997). At popping, trypsin inhibitors and other antinutritional substances are denatured (Correa *et al.,* 1986; Imeri, 1987; Stratil, 1993). The conventional way of popping is done on hot plates at a temperature higher than 190°C (Tovar *et al.,* 1989).

Heat processing can cause damage of essential amino acids resulting in decreased contents or transfer into a racemic mixture (Bressani *et al.,* 1987b; Tovar *et al.,* 1989). Temperatures above 100°C induce reactions in both bind and free amino acids, and especially in essential amino acids valine, leucine, isoleucine, lysine, threonine, methionine, phenylalanine and tryptophan, which then become non-utilizable for humans, thus decreasing the biological value of foodstuffs (Velíšek, 1999).

The aim of our study was to determine amino acid levels in grain of selected varieties in *Amaranthus cruentus*, *A. hypochondriacus*, *A. caudatus* and *A. hybridus* before and after heat treatment, and to determine the biological value of proteins using chemical scores and the index of essential amino acids (EAAI).

## MATERIAL AND METHODS

Grain samples ($n$ = 14) of selected varieties of four amaranth species – *A. cruentus* (variety Olpir, K 283), *A. hypochondriacus* (variety Koniz), *A. caudatus* (variety Elbrus), *A. hybridus* (variety K 432, D 701), and one variety mixture before ($n$ = 7) and after ($n$ = 7) heat treatment were analysed. All the varieties were grown in the Czech Republic.

Heat treatment was performed by a commercial system of current popping when the grain passes through hot air of 170 to 190°C for 30 s. Treatment of samples was carried out in accordance with the Czech Standards (Anonym, 1986).

The content of N-substances (N × 6.25) was determined using the equipment 2 300 Kjeltec Analyzer Unit (Foss Tecator, Sweden), fat content after extraction with ethyl ether was determined according to Soxhlet-Henkel. The methods for laboratory testing of feeds were carried out according to the Regulation of the Ministry of Agriculture of the Czech Republic (Anonym, 2001).

Samples for amino acid determination were adjusted using acidic and oxidative acidic hydrolysis HCl ($c$ = 6 mol/dm$^3$). The chromatographic analysis of sample hydrolysates was performed in the analyser AAA 400 (manufacturer INGOS Prague, CR) and using Na-citrate buffers and ninhydrin detection (Official Journal, 1978; Kráčmar *et al*., 1998).

Biological value of protein was assessed using chemical scoring and the basic index of essential amino acids (Davídek *et al.,* 1983). Chemical score (CS) expresses the percentage of the given amino acid in protein of the sample under study from the volume of the same amino acid in a standard protein. Egg albumin has been used as a standard.

The results of the determined amino acid values characters were converted into 100% dry matter. Basic statistical characteristics were obtained using the programme Stat Plus (Matoušková *et al.,* 1992).

## RESULTS AND DISCUSSION

Table 1 shows the content of dry matter, N-substances and fat in grain of selected varieties of four amaranth species – *Amaranthus cruentus* (Olpir and K 283), *A. hypochondriacus* (Koniz), *A. caudatus* (Elbrus), *A. hybridus* (D 701 and K 432) and one variety mixture, prior to and after heat treatment.

Heat treatment resulted in an increase of dry matter content by 4.2%, N-substances by 8.4%, and fat content decreased by 4.7%. The obtained data are in accordance with those reported by Carlsson (1979), Bressani *et al.* (1987b), Szelényi-Galántai and Zsolnai-Harszi (1992), Andrasofszky *et al.* (1998).

Amino acid values obtained in grain of selected amaranth species prior to and after heat treatment are shown in Tables 2 and 3, the average values and chemical scores are presented in Table 4.

Table 1. Content of dry matter, N-substances (N × 6.25) and fat (g/kg) in grain of selected varieties of *Amaranthus cruentus, A. hypochondriacus, A. caudatus* and *A. hybridus* before and after heat treatment (popping)

| Varieties | Dry matter | | N-substances | | Fat | |
|---|---|---|---|---|---|---|
| | raw | popping | raw | popping | raw | popping |
| Olpir | 924.0 | 949.2 | 144.6 | 148.6 | 75.9 | 77.1 |
| K 283 | 917.5 | 925.4 | 128.4 | 158.0 | 81.1 | 74.0 |
| Koniz | 881.2 | 932.8 | 125.0 | 141.3 | 71.4 | 69.2 |
| Elbrus | 917.0 | 947.8 | 125.0 | 144.8 | 75.2 | 77.3 |
| D 701 | 906.3 | 953.8 | 142.9 | 142.7 | 76.8 | 68.5 |
| K 432 | 898.2 | 965.2 | 136.3 | 147.4 | 73.0 | 67.1 |
| Mixture of varieties | 907.6 | 945.8 | 142.8 | 143.8 | 80.6 | 75.6 |
| Mean | 907.4 | 945.7 | 135.3 | 146.7 | 76.3 | 72.7 |
| ± S.D. | 14.4 | 13.2 | 9.08 | 5.61 | 3.61 | 4.32 |

Table 2. Amino acid contents in untreated grain of selected varieties of *Amaranthus cruentus, A. hypochondriacus, A. caudatus* and *A. hybridus* (g/16 g N)

| Amino acids | Olpir | K 283 | Koniz | Elbrus | K 432 | D 701 | Mixture of varieties |
|---|---|---|---|---|---|---|---|
| Cys | 3.1 | 3.4 | 2.9 | 3.2 | 3.1 | 3.6 | 3.5 |
| Asp | 10.0 | 10.4 | 10.7 | 9.6 | 10.2 | 10.5 | 10.4 |
| Met | 2.0 | 2.0 | 2.3 | 2.0 | 1.7 | 2.3 | 2.2 |
| Thr | 4.9 | 4.9 | 4.5 | 4.7 | 4.2 | 5.0 | 5.1 |
| Ser | 8.8 | 8.9 | 9.3 | 7.7 | 7.6 | 8.8 | 9.2 |
| Glu | 15.5 | 16.6 | 17.7 | 15.8 | 16.1 | 16.8 | 15.6 |
| Pro | 4.6 | 3.7 | 3.7 | 4.0 | 3.9 | 3.9 | 4.0 |
| Gly | 14.3 | 14.7 | 15.2 | 13.2 | 13.2 | 15.0 | 14.4 |
| Ala | 6.2 | 6.2 | 6.2 | 5.9 | 5.0 | 5.8 | 6.5 |
| Val | 4.8 | 4.9 | 5.3 | 4.7 | 4.6 | 4.9 | 5.1 |
| Ile | 3.6 | 3.7 | 3.8 | 3.4 | 3.7 | 3.2 | 3.9 |
| Leu | 6.2 | 6.5 | 6.9 | 5.9 | 6.8 | 5.8 | 6.5 |
| His | 2.0 | 1.9 | 1.7 | 1.7 | 2.3 | 1.6 | 2.0 |
| Lys | 8.0 | 8.0 | 8.0 | 7. 6 | 6.1 | 7.8 | 6.9 |
| Arg | 12.7 | 13.2 | 14.5 | 13.5 | 15.6 | 13.9 | 14.1 |

Heat treatment resulted in decreased levels of all the amino acids under study (Table 4, Figure 1) by 4.6% (from 0.61 to 9.6%) on average. Of the essential amino acids, His drop by 9.6% was the most marked, followed by Arg 7.9% ($P < 0.05$), Lys 7.7%, Leu 7.2% ($P < 0.05$), Val 5.5% ($P < 0.05$), Cys 5.2% and Thr 2.3%. The least decrease (by 0.96%) was recorded in Met. Pant (1985) stated the decrease of lysine level by 36% (from 4.83 to 3.08 g/16 g N) in traditionally popped grain while in commercially popped grain the lysine level decreased by 25%. Tovar *et al.* (1989) found the decrease of lysine, arginine and cystine levels while using both the methods of popping.

High chemical scores were demonstrated in Cys, Asp, Thr, Glu, Pro, Gly, His, Lys and Arg; lower scores

Table 3. Amino acid contents in heat-treated grain of selected varieties of *Amaranthus cruentus, A. hypochondriacus, A. caudatus* and *A. hybridus* (g/16 g N)

| Amino acids | Olpir | K 283 | Koniz | Elbrus | K 432 | D 701 | Mixture of varieties |
|---|---|---|---|---|---|---|---|
| Cys | 2.9 | 3.0 | 3.4 | 3.1 | 2.9 | 3.2 | 3.0 |
| Asp | 9.4 | 9.4 | 10.2 | 10.3 | 9.6 | 10.1 | 9.8 |
| Met | 1.8 | 2.0 | 2.1 | 2.2 | 2.0 | 2.2 | 2.1 |
| Thr | 4.4 | 4.6 | 4.9 | 4.4 | 4.7 | 4.8 | 4.6 |
| Ser | 8.3 | 8.2 | 8.5 | 8.5 | 8.2 | 8.3 | 8.3 |
| Glu | 16.8 | 16.8 | 16.3 | 16.2 | 16.5 | 15.9 | 15.3 |
| Pro | 4.1 | 3.3 | 3.2 | 3.7 | 4.4 | 3.4 | 3.9 |
| Gly | 13.4 | 13.8 | 14.4 | 13.9 | 13.9 | 14.0 | 13.7 |
| Ala | 5.7 | 5.9 | 5.8 | 6.0 | 5.9 | 5.6 | 6.2 |
| Val | 4.5 | 4.5 | 4.8 | 4.6 | 4.6 | 4.6 | 4.8 |
| Ile | 3.4 | 3.6 | 3.7 | 3.6 | 3.4 | 3.2 | 3.6 |
| Leu | 5.9 | 6.0 | 5.8 | 6.1 | 6.0 | 5.6 | 6.1 |
| Tyr | 0.3 | 0.2 | 0.5 | 0.3 | 0.3 | 0.2 | 0.3 |
| His | 2.1 | 1.5 | 1.6 | 1.7 | 1.8 | 1.5 | 1.8 |
| Lys | 7.0 | 6.6 | 7.4 | 7.4 | 6.3 | 7.0 | 6.7 |
| Arg | 12.2 | 12.6 | 13.2 | 13.0 | 12.2 | 13.1 | 13.5 |

Table 4. Content of amino acids (g/16 g N) and chemical scores (CS %) in grain of selected varieties of *Amaranthus cruentus, A. hypochondriacus, A. caudatus* and *A. hybridus* before and after heat treatment

| Amino acids | Egg protein | Raw ($n$ = 7) | | | Popping ($n$ = 7) | | |
|---|---|---|---|---|---|---|---|
| | | Mean | S.D. | CS | Mean | S.D. | CS |
| Cys | 1.8 | 3.3 | 0.24 | 181 | 3.1 | 0.17 | 172 |
| Asp | 8.2 | 10.2 | 0.38 | 124 | 9.8[a] | 0.37 | 120 |
| Met | 4.1 | 2.1 | 0.20 | 51 | 2.1 | 0.14 | 50 |
| Thr | 4.0 | 4.7 | 0.33 | 118 | 4.6 | 0.21 | 116 |
| Ser | 9.1 | 8.6 | 0.68 | 95 | 8.3 | 0.13 | 91 |
| Glu | 13.2 | 16.3 | 0.78 | 124 | 16.2 | 0.53 | 123 |
| Pro | 3.7 | 4.0 | 0.30 | 107 | 3.7 | 0.43 | 100 |
| Gly | 4.8 | 14.3 | 0.80 | 298 | 13.8 | 0.30 | 288 |
| Ala | 8.9 | 6.0 | 0.46 | 67 | 6.0 | 0.19 | 66 |
| Val | 7.1 | 4.9 | 0.24 | 69 | 4.6[a] | 0.12 | 65 |
| Ile | 6.3 | 3.6 | 0.21 | 57 | 3.5 | 0.16 | 56 |
| Leu | 8.3 | 6.4 | 0.42 | 77 | 5.9[a] | 0.17 | 71 |
| Tyr | 2.4 | – | – | – | 0.3 | 0.10 | – |
| His | 1.8 | 1.9 | 0.22 | 104 | 1.7 | 0.19 | 94 |
| Lys | 5.1 | 7.5 | 0.75 | 147 | 6.9 | 0.40 | 135 |
| Arg | 3.9 | 13.9 | 0.95 | 356 | 12.8[a] | 0.51 | 328 |

[a]mean values marked with superscript letters are significantly ($P < 0.05$) different

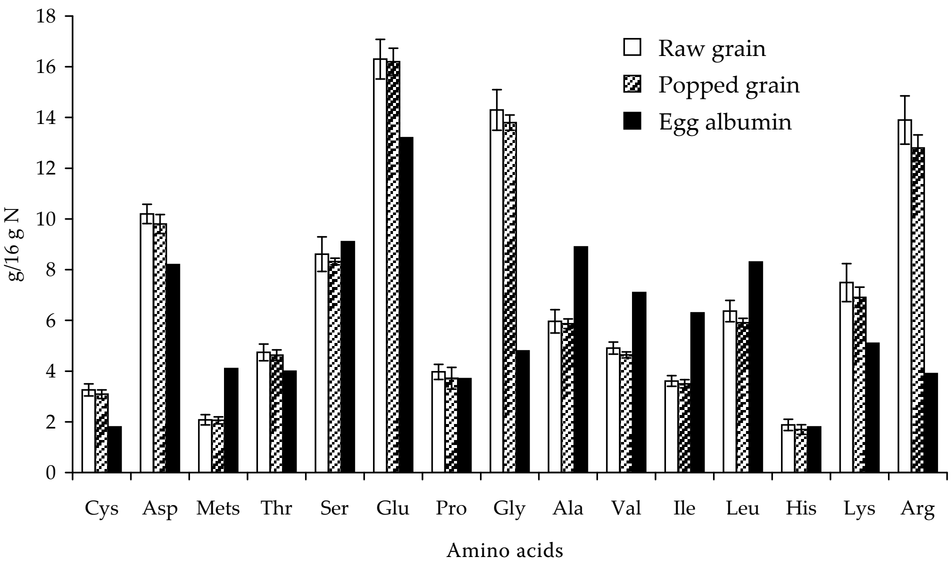


Figure 1. Amino acids contents in untreated and heat treated grain of selected variants of *Amaranthus cruentus, A. hypochondriacus, A. caudatus* and *A. hybridus* (g/g N) and in a standard

were recorded in Met, Ser, Ala, Val, Ile and Leu. The obtained amino acid levels in untreated and heat-treated amaranth grains are consistent with the data from the literature (Tovar *et al.,* 1989; Bressani *et al.,* 1992; Dodok *et al.,* 1997; Jarošová *et al.,* 1997; Gorinstein *et al.,* 2002).

Chemical scores in Val from 78.4 to 86.8%, Ile from 86.4 to 88.5% and Leu from 79.1 to 86.8% are mentioned by Paredes-Lopez (1994) in *Amaranthus caudatus, A. hypochondriacus, A. cruentus* and *A. edulis.*

Table 5. Chemical scores (CS %) and essential amino acid index (EAAI %) in grain of selected varieties of *Amaranthus cruentus, A. hypochondriacus, A. caudatus* and *A. hybridus* before and after heat treatment

| Amino acids[x)] | Chemical score (CS) | |
|---|---|---|
| | raw | popping |
| His | 104 | 94 |
| Ile | 57 | 56 |
| Leu | 77 | 71 |
| Lys | 147 | 135 |
| Met + Cys | 90 | 87 |
| Thr | 118 | 116 |
| Val | 69 | 65 |
| EAAI | 90.4 | 85.4 |

[x)]Food and Agricultural Organization, WHO (1983); Crim and Munro (1984)

Biological values of protein in untreated and heat-treated amaranth grain are summarized in Table 5. High values of chemical scores were found in Lys, Thr, His and Arg. Lower values were found in Leu, Val and Ile (Table 4). Val, Leu and Ile seem to be the limiting amino acids. Tovar *et al.* (1989) stated the chemical scores of essential amino acids of amaranth grain prior to and after popping as follows: Lys 127 : 56%, Thr 115 : 110%, Val 111 : 112%, Ile 122 : 126%, and Leu 107 : 110%. He indicated Lys and sulphur amino acids to be limiting. Leu was also indicated as a limiting amino acid by Becker *et al.* (1981), Pedersen *et al.* (1987) and Bressani *et al.* (1992).

The biological value of untreated amaranth grain based on the chemical scores of the essential amino acids and expressed by the EAAI character was 90.4%. After heat processing, the value dropped to 85.4%. The relatively high content of essential amino acids in amaranth grain predetermines its use as a substitution of meat-and-bone meals.

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