

THE EFFECT OF THERMIC TREATMENT CONDITIONS ON THE AMINO ACID COMPOSITION OF SOYBEAN AND MAIZE

DJELOVANJE TERMIČKOG TRETIRANJA NA SASTAV AMINO KISELINA SOJE I KUKURUZA

Éva Varga-Visi, Gabriella Pohn, Csilla Albert, Z. Mándoki, J. Csapó

Original scientific paper - Izvorni znanstveni članak
Received - Primljeno: 24. march - ožujak 2009.

SUMMARY

Protein is one of the most valuable components of fodder, therefore there is a concern preserving over these components in an available form during different treatments wherein heating is applied. The aim of the research was to investigate the influence of toasting of fullfat soybean and air-drying of maize on the amino acid profile of the products.

Dried maize was sampled at two Hungarian drying plants. The maize was dried with an industrial Bábolna B1-15 type gravitational drying tower.

The influence of toasting on two sorts of fullfat soybean products (hydrothermic soy coarse and 'natural' hydrothermic soy) was investigated. Soybeans were heat processed with an industrial KAHL HR-1600 type hydrothermic reactor after cracking.

The amino acid analysis of samples was carried out with amino acid analyzer and amino acid enantiomers were quantified with a LaChrom type MERCK-Hitachi high performance liquid chromatograph.

The D-amino acid content of the samples before and after drying did not differ significantly, not even at the applied highest temperatures (100 °C drying air temperature and 40-45 °C kernel temperature).

Pressurized steam cooking of fullfat soy did not result in a significant increase of the amount of D-enantiomers or a decrease of the concentration of L-amino acids ($P>0.05$), while the trypsin inhibitor activity (TIA) was reduced to the required level ($TIA<1.5$ mg/g) and the results of the urease test ($\Delta pH<0.2$) also verified the adequate intensity of the heat treatment.

Key words: thermic treatment, soybean, maize, toasting, air-drying, amino acids

Éva Varga-Visi, Gabriella Pohn, Zsolt Mándoki, János Csapó, University of Kaposvár, Faculty of Animal Sciences, Department of Chemistry-Biochemistry; Csilla Albert, János Csapó, Sapientia Hungarian University of Transylvania, Csíkszereda Campus, Department of Food Sciences, Hungary.

INTRODUCTION

Processing steps can increase considerably the amount of D-amino acids, when they include heat treatment, alkaline treatment or fermentation (Imai et al., 1996; Man and Bada, 1987; Friedman, 1991). Heat treatment processes accelerate racemization, since higher temperatures result in increasing reaction rate of the racemization according to the Arrhenius equation. In case of a given production process a concrete experiment is required in order to decide whether the degree of the conversion into D-amino acids is considerable.

When feeding crude soybean without heat treatment fodder utilization can be reduced even by 20 to 30% in the case of monogastric species and young ruminants, as soya contains considerable amount of antinutritive substances (eg. protease inhibitors, lectins). Some of these losses their activity when high temperature is applied, so there is a chance to lessen the unfavourable effect. There are several thermal procedures to heat treat soybean: extrusion, puffing-up, as well as toasting, when soybean is cooked in the presence of water steam at 115-120 °C for 15-40 min (Bódis and Manninger, 2002). Any of these treatments are used, the working parameters (temperature, moisture content, particle size, other physical effects) should be chosen so that the level of antinutritive substances is appropriately reduced without deterioration of the nutritive value of the protein (Monary, 1996).

In Hungary, among cereals maize is used in the highest amount for foddering purpose. Maize needs to be dried in order to be stored safely. Drying maize fodders is carried out by blowing-through high temperature air, and in most of the agricultural plants so-called drying towers are used (Francsics and Parti, 1987). Inappropriate drying (overdrying) decreases the digestibility of the feeding stuff proteins. Deterioration of proteins of the maize fodder during the drying can be avoided if the temperature of the drying air does not exceed 80-130 °C, and so the temperature of the kernels does not rise above 60-80 °C. If still overdrying occurs, digestibility of the proteins decreases, utilizability of amino acids reduces. Utilizability of lysine reduces to the highest degree which is especially harmful as to the nutritive value of maize, since in zein lysine is the primary limiting amino acid.

The effect of the above thermic treatments on the amino acid content has already been reported in

a number of publications, however, according to our knowledge it has not been measured yet in what ratio the amino acids bound in proteins convert into D-amino acids during toasting of soybean and drying of maize.

MATERIAL AND METHODS

Conditions of the drying of maize and toasting of soybeans

The dried maize samples were taken partly at the drying plant of Agria Mezőgazdasági és Szolgáltató Szövetkezet (Agria Agricultural and Servicing Co-operative) in Szentgáloskér, partly at the drying plant of Kapostáj Mezőgazdasági Termelőszövetkezet (Kapostáj Agricultural Co-operative) in Zimány. Drying of the crops was performed at both sites with a *Bábolna B1-15* dryer that is commonly used in Hungary. This equipment belongs to the group of the gravitational tower dryers, where the drying (and cooling, respectively) air touches indirectly the particles of the crop. On the border of the drying and the cooling zone sensors are installed measuring the moisture content of the seeds, the maize temperature and the temperature of the drying air on both sides of the hot air canal (T_1 , T_2) at a certain height. Parameters read from the displays during the drying and data taken in the collected samples at the Analytical Laboratory are shown in Table 1.

A sample was taken from a given lot before drying, then from the dried maize continuously until the given lot exited the drier.

The hydrothermal treatment was carried out at the feeding stuff plant of Bóly Rt. (Bóly-Állomáspuszta) in Törökdomb. The whole soybeans stored in silos were carried through two rolling mills after cleaning, where the seeds were cracked into 9 to 12 pieces, then the material entered the toaster (*KAHL HR-1600* hydrotermic reactor). This autoclave with a mixing part cooked the soya in pressurized steam with an average residence time of 30 min at 120 °C. Due to the wet heat treatment most of the anti-digestive trypsin inhibitors present in the soybean became inactivated. During production in the first case the heat treatment was followed by milling in a hammer mill, and the product obtained sold as „hydrothermal soya grits”. In the case of the second product that milling was omitted, and the soybean remained in cracked form, called „natural hydrothermic soya”.

Table 1. Data of plant drying of maize

Tablica 1. Podaci o sušenju kukuruza

Conditions of drying - Uvjeti sušenja		Place of sampling - Mjesto uzorkovanja		
		Szentgáloskér	Zimány (1)	Zimány (2)
Values measured at drying - Vrijednosti mjerene kod sušenja				
Average temperature of the drying air (°C) - Prosječna temperatura zraka za sušenje				
	T1	100	78	45
	T2	100	83	39
Moisture content above the cooling zone (%) Sadržaj vlage iznad zone hlađenja		20-21	13-14	12-13
Maize temperature (°C) - Temperatura kukuruza		40-45	39-40	32-33
Values measured in the laboratory - Vrijednosti mjerene u laboratoriju				
Moisture content before drying (%) - Sadržaj vlage prije sušenja		16,6	17,1	16,6
Moisture content after drying (%) - Sadržaj vlage poslije sušenja		13,1	12,5	13,1

Chemical examinations

The chemical analysis of the samples were carried out at the Department of Chemistry and Biochemistry, University of Kaposvár, Faculty of Animal Science. Moisture content of the samples was determined according to the standard MSZ ISO 1442, and trypsin inhibitor activity of the soya products was determined according to the standard EN ISO 14902. In order to determine the amino acid enantiomers, after hydrolysis of the protein (6 M hydrochloric acid, 24h, 105 ±1°C) diastereomer pairs were formed from the D- and L-amino acids with o-phthalaldehyde (OPA) and 1-thio-β-D-glucose tetra acetate (TATG) according to the method of Einarsson et al. (1987), as well as Csapó et al (1995). The analyses were performed using a MERCK-Hitachi LaChrom high-performance liquid chromatograph. The derivatives were separated on a Superspher 60 RP-8e column (125 mm x 4 mm i.d.). Fluorescent signals of the diastereomers were measured (λ_{ex} : 325nm, λ_{em} : 420nm).

The statistical evaluation was performed with SPSS for Windows 10.0 (1999) software.

RESULTS AND DISCUSSION

The effect of toasting on the D-amino acid content of the fullfat soya

L- and D-amino acid content of the untreated soybean as well as hydrothermally treated fullfat soybean products is shown in Table 2. Because of the difference in the water content of the samples the analysis results are given on dry matter basis. D-amino acids found in the untreated control sample were formed probably during chemical preparation of the heat treated samples due to acidic hydrolysis of the proteins (Masters and Friedman, 1980). For the time being, according to our knowledge there is no chemical method available, using which this phenomenon could be excluded. If the mean values of D-amino acid content of samples taken before and after the heat treatment differ considerably, this difference can be regarded as the increase of D-amino acids due to the heat treatment (de Vrese et al., 2000).

Practically no detectable change occurred in the amount of either D- or L-amino acids due to cooking in the pressurized steam, whereas the activity of trypsin inhibitors reduced appropriately in both of the soya products due to the heat treatment (Table 2).

Table 2. Amino acid content (g/100g dry matter) (n=5) and trypsin inhibitor activity (TIA) (n=3) of non-heat treated soya and toasted fullfat soya products

Tablica 2. Sadržaj amino kiselina (g/100g suhe tvari)(n=5) i aktivnost inhibitora tripsina (TIA)(n=3) netoplinski tretirane soje i tostiranih proizvoda punomasne soje

Amino acid (g/100g dry matter) Amino kiselina(g/100g suhe tvari)	Untreated soybean Netretirano zrno soje	Hydrothermal soya grits Hidrotermalna sojina prekrupa	Natural hydrothermic soya Prirodna hidrotermička soja
L-Asp	3,90 ± 0,10	4,20 ± 0,70	4,10 ± 0,20
D-Asp	0,55 ± 0,04	0,62 ± 0,11	0,65 ± 0,04
L-Glu	6,60 ± 0,20	7,00 ± 1,30	7,10 ± 0,50
D-Glu	0,11 ± 0,03	0,12 ± 0,02	0,12 ± 0,01
L-Ser	1,68 ± 0,05	1,76 ± 0,36	1,78 ± 0,13
D-Ser	0,13 ± 0,01	0,13 ± 0,03	0,13 ± 0,01
L-Val	1,70 ± 0,05	1,78 ± 0,31	1,78 ± 0,14
L-Met	0,25 ± 0,04	0,26 ± 0,04	0,28 ± 0,03
L-Phe	1,81 ± 0,07	1,89 ± 0,37	1,92 ± 0,13
D-Phe	0,08 ± 0,02	0,08 ± 0,01	0,09 ± 0,01
L-Leu	2,70 ± 0,06	2,79 ± 0,59	2,79 ± 0,26
D-Leu	0,27 ± 0,01	0,26 ± 0,06	0,26 ± 0,03
L-Lys	1,56 ± 0,14	1,62 ± 0,43	1,60 ± 0,12
D-Lys	0,03 ± 0,01	0,03 ± 0,01	0,04 ± 0,01
TIA (mg/g)	17,2 ^b ± 0,5	1,1 ^a ± 0,2	1,2 ^a ± 0,3

Mean values without marks being in the same row do not differ (P<0.05).

^{ab} There is no difference between mean values marked with the same letters and being in the same row.

The effect of drying of the maize on the D-amino acid content

The amount of the examined D- and L-amino acids contained in the maize before and after the heat treatment can be found in Table 3. In the samples taken before and after drying the amount of the examined D-amino acids did not differ significantly even in the case of drying at the highest temperature (100 °C air temperature, 40-45 °C kernel temperature, sampling in Szentgáloskér), however, it should be kept in mind that what we measured was an average value which is

important in practice. Layers lying closer to the surface of the seeds were exposed to a heat impact of higher degree than the inner parts, and also inside the dryer temperature distribution could be inhomogeneous, with centers of higher and lower heat burden.

In the amount of the measured L-amino acids there was no difference between the control and the heat-treated groups with one exception, in the second sampling (Zimány (1)) L-methionine content of the dried maize was a little lower than that of the untreated sample.

Table 3. Amount of the examined L- and D-amino acids in the maize before and after drying (g/100g dry matter) (n=5)

Tablica 3. Količina ispitanih L- i D-amino kiselina u kukuruzu prije i poslije sušenja (g/100 suhe tvari (n=5))

Amino acid Amino kiselina	Place of sampling - Mjesto uzorkovanja					
	Szentgáloskér		Zimány (1)		Zimány (2)	
	Drying - Sušenje		Drying - Sušenje		Drying - Sušenje	
	before - prije	after - poslije	before - prije	after - poslije	before - prije	after - poslije
L-Asp	0,66±0,05	0,68±0,03	0,64±0,04	0,65±0,01	0,70±0,04	0,70±0,03
D-Asp	0,079±0,006	0,081±0,004	0,077±0,005	0,079±0,002	0,095±0,005	0,093±0,006
L-Glu	1,70±0,10	1,74±0,08	1,86±0,11	1,85±0,07	1,81±0,13	1,89±0,10
D-Glu	0,026±0,002	0,027±0,001	0,030±0,003	0,030±0,001	0,031±0,002	0,032±0,002
L-Ser	0,48±0,03	0,49±0,02	0,50±0,03	0,49±0,03	0,49±0,03	0,49±0,02
D-Ser	0,040±0,008	0,043±0,009	0,034±0,01	0,039±0,01	0,068±0,008	0,065±0,007
L-Val	0,49±0,03	0,49±0,02	0,47±0,03	0,046±0,03	0,50±0,03	0,50±0,02
L-Met	0,04±0,005	0,04±0,003	0,04*±0,006	0,03*±0,002	0,04±0,005	0,05±0,01
L-Phe	0,44±0,03	0,45±0,02	0,49±0,03	0,48±0,02	0,47±0,03	0,49±0,02
D-Phe	0,26±0,01	0,23±0,02	0,17±0,02	0,16±0,04	0,21±0,02	0,21±0,02
L-Leu	1,11±0,05	1,11±0,04	1,28±0,08	1,23±0,09	1,14±0,08	1,22±0,06
D-Leu	0,10±0,03	0,11±0,008	0,15±0,01	0,14±0,02	0,16±0,01	0,16±0,008
L-Lys	0,30±0,05	0,32±0,02	0,30±0,03	0,27±0,06	0,29±0,04	0,29±0,02

Remark: Mean values marked with asterisk differ significantly (P<0.05).

CONCLUSION

It can be said that the aim of the examined thermal procedures was accomplished without significant increase in the amount of D-amino acids. During toasting of the soybean the trypsin inhibitors were appropriately inactivated, and at the same time the L-amino acid content did not change. The moisture content decreased to the required level during the drying of maize without a considerable loss of amino acids.

REFERENCES

- Bódis, L., Manninger, S. (2002): Fehérjéggazdálkodás Magyarországon. In Magyarország fehérjéggazdálkodásának helyzete és fejlesztési stratégiája. Ed. Babinszky L.; Agroinform Kiadó, Budapest 45-91.
- Csapó, J., Csapó-Kiss, Zs., Einarsson, S., Folestad, S., Tivesten, A. (1995): Methods for determination of D-amino acid content of foods and feeds. *Acta Alimentaria*, 24: 125-126.
- Einarsson, S., Folestad S., Josefsson, B. (1987): Separation of amino acid enantiomers using precolumn derivatization with o-phthalaldehyde and 2,3,4,6,-tetra-O-acetyl-1-thio-β-glucopyranoside. *J. Liquid Chrom.* 10. 1589.
- Francsics, P., Parti, M. (1987): Energiatakarékos szemettermény-szárítás. Mezőgazdasági Kiadó, Budapest.
- Friedman, M. (1991): Formation, nutritional value, and safety of D-amino acids. In Nutritional and toxicological consequences of food processing. Ed. M. Friedman, Plenum Press, New York. 447-481.
- Imai, K., Fukushima, T., Santa, T., Homma, H., Hamase, K., Sakai, K., Kato, M. (1996): *Analytical chemistry and biochemistry of D-amino acids. Biomedical Chromatography*, 10. 303-312.
- Man, H., Bada, J. L. (1987): Dietary D-amino acids. *Ann. Rev. Nutr.* 7. 209-225.
- Masters, P.M., Friedman, M. (1980): Amino acid racemization in alkali-treated food proteins – chemistry, toxicology, and nutritional consequences. In Chemical Deterioration of Proteins. Ed. J. R.

- Whittaker, M. Fujimaki, Washington DC, Am. Chem. Soc. ACS Symp. 123. 165-194.
9. Monary, S. (1996): Fullfat soya handbook. American Soybean Association, Brüssel, Belgium.
10. de Vrese, M., Frik, R., Roos, N., Hagemester, H. (2000): Protein-bound D-amino acids, and to a lesser extent lysinoalanine, decrease true ileal protein digestibility in minipigs as determined with ¹⁵N-labeling. *J. of Nutr.* 8. 2026-2031.

SAŽETAK

Bjelančevina je jedan od najvrjednijih sastojaka krmiva, pa je zato važno očuvanje tih sastojaka u dostupnom obliku za vrijeme raznih postupaka u kojima je primijenjeno grijanje. Cilj je rada bio istražiti djelovanje tostiranja punomasne soje i zrakom sušenog kukuruza na profil amino kiselina proizvoda.

Sušeni kukuruz uzorkovan je u dva pogona za sušenje u Mađarskoj. Kukuruz je sušen u industrijskom gravitacijskom tornju za sušenje tipa Babolna B1-15.

Istraživan je utjecaj tostiranja na dvije sorte punomasne soje (hidrotermička gruba soja i "prirodna" hidrotermička soja). Soja je toplinski obrađena industrijskim tipom KAHL HR-1600 hidrotermičkog reaktora nakon pucanja.

Analiza amino kiselina uzoraka obavljena je pomoću analizatora amino kiselina, a emancioneri amino kiselina kvantificirani su pomoću LaChrom tipa MERCH-Hitachi tekućim kromatografom visoke performanse.

Sadržaj D-aminokiseline u uzorcima nije se značajno razlikovao prije i poslije sušenja, čak niti kod primjene najviših temperatura (100 °C temperatura zraka sušenja, 40-45 °C temperatura zrna).

Kuhanje punomasne soje na pari pod pritiskom nije znatno povećalo količinu D-emancionera niti smanjilo koncentraciju L-amino kiselina ($p > 0.05$), dok je aktivnost inhibitora tripsina (TIA) smanjena na potrebnu razinu.

Ključne riječi: termički postupak, soja, kukuruz, pečenje, sušenje, amino kiseline