

## AMINO ACID COMPOSITON OF EMMER LANDRACES GRAIN

P. KONVALINA<sup>1</sup>, J. MOUDRÝ Jr.<sup>1</sup>,  
Z. STEHNO<sup>2</sup>, J. MOUDRÝ<sup>2</sup>

<sup>1</sup>University of South Bohemia, Faculty of Agriculture  
Studentská 13 České Budějovice  
370 05 - Czech Republic

phone number: +420387772547

e-mail: konvalina@zf.jcu.cz

<sup>2</sup>Crop research institut, Drnovská 507,  
Praha 6 – Ruzyně 161 06 – Czech Republic

*This article deals with a study of amino acid composition of 6 varieties of emmer coming from the genetic resources. In the year 2007, small-parcel double trials were set up at two locations, at the Research Institute of the Crop Production in Prague (fertile location) and at the Faculty of Agriculture in České Budějovice (marginal location). Amino acid content was analysed by method of acidolysis at AAA 400 apparatus based on liquid chromatography. According to the findings, lysine is the limiting amino acid in emmer wheat and bread wheat too. There are no considerable differences in lysine content between the tested varieties (chemical score varies from 0,37 to 0,44). The proportion of amino acids was obviously ( $p<0,05$ ) influenced by crude protein content (valine  $r=-0,72$ ; tyrosine  $r=-0,56$ ; phenylalanine  $r=-0,73$ ) in negative way. The proportions of valine, leucine, tyrosine, phenylalanine were obviously ( $p<0,05$ ) influenced by the relation to species and variety; the controlling varieties of wheat achieved higher values. The correlation analysis of essential amino acids also provides very interesting figures; threonine content is in positive correlation to isoleucine content ( $r=0,96$ ), leucine ( $r=0,91$ ) and lysine ( $r=0,95$ ). The proportion of valine is in positive correlation to phenylalanine content ( $r=0,99$ ). Isoleucine is in positive correlation to leucine content ( $r=0,98$ ) and to lysine content ( $r=0,95$ ). Emmer wheat contains the same amino acids as modern varieties of wheat. When higher crude protein content in flour and the conversion to 1 000 g of flour taken into account, it is characterised by higher protein content in grain and higher content of amino acids in g/1 000 g of flour. Therefore, the grains of emmer wheat can be used for the production of nutritional valuable diet (organic foodstuffs).*

**Key words:** emmer wheat, amino acid composition, landrace

Emmer wheat [*Triticum dicoccum* (SCHRANK) SCHUEBL] belongs, among others, to historical tetraploid wheat species (AABB genome,  $2n=28$ ). It is considered to be a predecessor of most of wheat species [KÖRNICKE, 1889]. The first forms of wheat were formed by a spontaneous hybridization of emmer wheat

and *Aegilops squarosa* [DIAMOND, 1997]. It is an annual self-pollinating type of cereals which can be found in the transitional area between the Mediterranean and steppe region. Most of genotypes of emmer wheat belong to spring with awned spike [FOLTÝN *et al.*, 1970]. Emmer wheat is divided into 99 varieties.

The tradition of growing and use of emmer wheat for human diet is very long and very old. The human interest is even more strengthened by the fact it was probably domesticated more than once [ALLABY, 1999]. Near East is supposed to be an area of origin of wheat. Emmer wheat was domesticated more than 10 000 years ago [ZOUHARY, HOPF, 1988]. Emmer wheat was considered as the most important crop for 7 000 years [FELDMAN, 2001]. Several wild and domesticated forms of emmer wheat and einkorn were founded in Levant area (South-Eastern Turkey, Northern Syria) in 7 700-7 500 BC. It was and still is rarely grown in submontane and montane areas in Caucasus, Baskyria, on the Balkans, in Spain, Turkey, Iran, Yemen, India, Morocco and Etiopia [DOROFEEV *et al.*, 1987].

In spite of its considerable importance, it has seldom been bred. Nowadays, just wild forms, land races or obsolete traditional cultivars are available. A lot of land races are grown in Italy [BOZZINI *et al.* 1994; GALTERA *et al.*, 1994; PERRINO *et al.*, 1996; PISANTE *et al.*, 1996; ANTUONO *et al.*, 1998]. Some original land races of emmer wheat are also stored and evaluated by the Czech gene bank. The interest in emmer wheat has still been increasing; nevertheless, it is conditioned by better possibilities of market sales [D' ANTUONO *et al.*, 1996]. Recently, the interest in this crop has also increased in the Czech Republic – the demands and requirements on colourfulness and quality of foodstuffs have been becoming more and more high (KONVALINA, MOUDRÝ, 2007a; KONVALINA, MOUDRÝ, 2007b).

Nutritional grain value is one of the reasons of the increasing interest in products made of emmer wheat. The target of the work is to evaluate the nutritional protein value in emmer wheat due to the proportion and chemical score of essential amino acids. The essential amino acid showing the lowest percentage of the content of the same amino acid in the same quantity of protein (real or hypothetical) selected as a reference protein is called the (first) limiting amino acid, and this percentage is the chemical score [FAO/WHO, 1991].

## MATERIAL AND METHOD

On the basis of the previous screening, 6 genotypes of emmer (Table 1) and 4 landraces (Kundan, Jara, Praga, Rosamova česká přesívka) and 2 modern (M6 - Vánek, M10 – SW Kadrij) controlling cultivars of bread wheat were choosen. In the year 2007, small-parcel double trials were set up at two stations, at the Research Institute of the Crop Production in Prague (CRI) - fertile location and at the Faculty of Agriculture in České Budějovice (CB) - marginal location.

Amino acid content was analysed by method of acidolysis. The bounded amino acids are losed from proteinic string at first. The salt acid as hydrogenation agent is used. The analysis continue at AAA 400 apparatus based on liquid chromatography (in group essential amino acod content was analysed content of threonine, valine, leucine, isoleucine, tyrosine, phenylalanine a lysine). The amino acid content in flour have been

converted into amino acid content per 100 g protein. In subsequence have been counted chemical score, based on the FAO 1985 pattern (threonine = 3,40; valine = 3,5; leucine = 6,6; isoleucine = 2,8; tyrosine + phenylalanine = 6,3; lysine = 5,8) (FAO/WHO, 1991). The data were analysed in STATISTICA programme.

Table 1

**List of varieties**

Code of variety	ECN <sup>1</sup>	BCHAR <sup>2</sup>	name	SP <sup>3</sup>	<i>Triticum dicoccum</i> (SCHRANK) SCHUEBL:
D1	01C0200117	412064	Horny Tisovnik	CZ	var. <i>rufum</i> SCHUEBL
D2	01C0200947	412048	Ruzyne	-	var. <i>rufum</i> SCHUEBL
D3	01C0201262	412051	Tapioszele 1	-	var. <i>serbicum</i> A. SCHULZ
D4	01C0201282	412017	Tapioszele 2	-	var. <i>rufum</i> SCHUEBL.
D7	01C0203989	412013	Kahler Emmer	D	var. <i>dicoccum</i>
D10	01C0204501	412013	No. 8909	-	var. <i>dicoccum</i>
Note: <sup>1</sup> ECN = identifier ; <sup>2</sup> BCHAR = taxonomical code ; <sup>3</sup> SP = origin					

**RESULTS AND DISCUSSIONS**

Lysine is the limiting amino acid of the controlling varieties of wheat (the statement is in accordance with the literature). Concerning the tested varieties, the proportion of lysine varies from 1,72 g/100 g of proteins (location CB, variety D4 Tapioszele 2) to 2,89 g/100 g of proteins (location CRI, variety D1 Horní Tisovnik) (table 2). It may be stated, according to the variance analysis, that the proportion was not influenced neither by fertility of station, nor by species (emmer wheat), variety or by protein content in grain (Table 3). Chemical score of 6 varieties of emmer wheat achieves quite low values of 0,38 (CB), 0,43 (CRI), the same as the controlling varieties (0,42 - CB and 0,43 - CRI). The values mentioned above are in accordance with the data stated by GALTERIO *et al.* (1994) and CUBADDA, MARCONI (1996).

The registered proportion of threonine was also lower than the figures stated by FAO/WHO (1991). It varied from 1,77 g/100 g of proteins (location CB, variety D4 Tapioszele 2) to 2,77 g/100 g of proteins (location CRI, variety D1 Horní Tisovnik) (table 2). The proportion of threonine was not considerably influenced neither by fertility of station, nor by species (emmer wheat), variety or protein content in grain (table 3) (the same as lysine). Chemical score achieved 0,66 (CB) and 0,73 (CRI); it is supposed to be the second limiting amino acid.

Leucine is the third limiting amino acid (chemical score 0,80 – CB and 0,84 – CRI). Concerning modern varieties, it achieved 0,90 (CB) and 0,91 (CRI). The proportion of leucine was not considerably influenced neither by fertility of station, nor by species (emmer wheat), variety or crude protein content in grain (Table 3). The proportion of leucine varied from 4,23 g/100 g of proteins (location CB, variety D4 Tapioszele 2) to 6,02 g/100 g of proteins (location CRI, variety D1 Horní Tisovnik) (table 2).

Table 2

**Essential amino acid patterns (g/100 g protein) of emmer**

Variety	Threonine			Valine			Isoleucine			Leucine			Tyrosine + Phenylalanine						Lysine			chemical score		
	Location		CB	Location		CB	Location		CB	Location		CB	Location		CB	Location		CB	Location		CB		CRI	mean
Emmer																								
D1	2,28	2,77	3,82	3,87	5,45	6,02	2,83	3,23	2,50	2,69	3,82	3,92	2,24	2,89	2,56									<b>0,44</b>
D2	2,25	2,52	3,65	3,52	5,53	5,68	2,85	2,94	2,26	2,38	3,65	3,62	2,41	2,50	2,45									<b>0,42</b>
D3	2,38	2,20	3,69	3,64	5,41	5,30	2,85	2,70	2,26	2,40	3,69	3,70	2,32	2,14	2,29									<b>0,40</b>
D4	1,77	2,47	2,88	3,45	4,23	5,31	2,16	2,87	1,93	2,35	2,96	3,48	1,72	2,54	2,13									<b>0,37</b>
D7	2,39	2,33	3,58	3,49	5,42	5,30	2,81	2,72	2,18	2,17	3,59	3,50	2,39	2,31	2,35									<b>0,41</b>
D10	2,45	2,61	3,62	3,66	5,51	5,59	2,82	2,98	2,22	2,43	3,63	3,68	2,26	2,66	2,46									<b>0,42</b>
mean	2,25	2,48	3,54	3,60	5,26	5,53	2,72	2,91	2,23	2,40	3,56	3,65	2,22	2,51	-									-
chemical score <sup>1</sup>	0,66	0,73	1,14	1,03	0,80	0,84	0,97	1,04	-	-	0,92	0,96	<u>0,38</u>	<u>0,43</u>	-									-
Controll variety																								
M6	2,43	2,65	3,87	4,10	5,68	6,12	2,98	3,14	2,61	2,53	4,02	4,10	2,33	2,53	2,43									0,42
M10	2,63	2,50	4,24	4,07	6,18	5,82	3,15	3,04	2,54	2,60	4,14	4,12	2,56	2,45	2,50									0,43
mean	2,53	2,57	4,05	4,08	5,93	5,97	3,07	3,09	2,58	2,56	4,08	4,11	2,44	2,49	-									-
chemical score <sup>1</sup>	0,74	0,76	1,16	1,17	0,90	0,91	1,10	1,10	-	-	1,06	1,06	<u>0,42</u>	<u>0,43</u>	-									-
<sup>1</sup> Based on the FAO 1985 pattern (threonine = 3,4; valine = 3,5; leucine = 6,6; isoleucine = 2,8; tyrosine + phenylalanine = 6,3; lysine = 5,8) (FAO/WHO, 1991); the first limiting amino acid is underlined																								

<sup>1</sup>Based on the FAO 1985 pattern (threonine = 3,4; valine = 3,5; leucine = 6,6; isoleucine = 2,8; tyrosine + phenylalanine = 6,3; lysine = 5,8) (FAO/WHO, 1991); the first limiting amino acid is underlined

Table 3

**Correlation analysis of factor vs. amino acid**

Factor	Threonine	Valine	Methionine	Isoleucine	Leucine	Tyrosine	Phenylalanine	Lysine
Crude protein content	-0,20	-0,72*	0,04	-0,39	-0,44	-0,56*	-0,73*	-0,03
Location	0,41	0,09	0,52*	0,31	0,25	0,34	0,13	0,45
Species	0,36	0,70*	0,15	0,48	0,55*	0,57*	0,73*	0,18
Variety	-0,21	-0,53*	-0,32	-0,43	-0,45	-0,65*	-0,58*	-0,21

\*Correlations are significant at  $p < 0,05$

Tyrosine and phenylalanine were evaluated together (FAO/WHO, 1991). The proportion of these two elements varied from 4,88 g/100 g of proteins (location CB, variety D4 Tapoiszele 2) to 6,61 g/100 g of proteins (location CRI, variety D1 Horní Tisovnik); chemical score achieved 0,92 (CB) and 0,96 (CRI) (*table 2*). Concerning the controlling varieties, the proportion and chemical score were higher (1,06) at both locations; in such case, the amino acids are not supposed to be limiting ones any more. According to the figures stated by the analysis of variance, the proportion of both amino acids was considerably influenced ( $p < 0,05$ ) by species factor (positive dependance,  $r = 0,57$  - tyrosine,  $r = 0,73$  phenylalanine) and variety factor (negative dependance,  $r = -0,65$  tyrosine,  $r = -0,58$  phenylalanine). The negative relation between tyrosine content ( $r = -0,56$ ) and phenylalanine content ( $r = -0,73$ ) and crude protein content (*table 3*) was also registered.

Chemical score of isoleucine achieved 0,97 (CB) and 1,04 (CRI). Concerning the controlling varieties, it is not supposed to be limiting amino acid (chemical score = 1,1 at both locations). The proportion of isoleucine varied from 2,16 g/100 g of proteins (location CB, variety D4 Tapoiszele 2) to 3,23 g/100 g of proteins (location CRI, variety D1 Horní Tisovnik) (*table 2*). It was not considerably influenced neither by fertility of station, nor by species (emmer wheat), variety or crude protein content (*table 4*).

Table 4

**ANOVA, values of the test criterion of amino acid content**

Amino acid	Crude protein content	Location	Species	Variety
Threonine	0,42 <sup>n</sup>	2,82 <sup>n</sup>	2,06 <sup>n</sup>	1,24 <sup>n</sup>
Valine	4,94*	0,11 <sup>n</sup>	13,57**	3,31 <sup>n</sup>
Methionine	0,13 <sup>n</sup>	5,20*	0,31 <sup>n</sup>	0,95 <sup>n</sup>
Isoleucine	0,91 <sup>n</sup>	1,51 <sup>n</sup>	4,18 <sup>n</sup>	2,26 <sup>n</sup>
Leucine	1,40 <sup>n</sup>	0,91 <sup>n</sup>	6,22*	3,41*
Tyrosine	1,52 <sup>n</sup>	1,78 <sup>n</sup>	6,62*	3,66*
Phenylalanine	4,32*	0,25 <sup>n</sup>	16,16**	4,30*
Lysine	0,42 <sup>n</sup>	3,64 <sup>n</sup>	0,46 <sup>n</sup>	0,79 <sup>n</sup>
* Marked effects are significant at $p < 0,05$ ; ** Marked effects are significant at $p < 0,05$ ; <sup>n</sup> statistically insignificant				

Valine is the most frequent essential amino acid. Chemical score achieved 1,14 (CB) and 1,03 (CRI). It is not supposed to be limiting amino acid, nor in case of the controlling varieties (chemical score = 1,16 and 1,17). According to the findings of the analysis, the negative dependance ( $p < 0,05$ ) between crude protein content in grain and valine ( $r = -0,72$ ) is evident. According to the findings of analysis of variance, the proportion of valine was considerably ( $p < 0,05$ ) influenced by species factor (positive dependance,  $r = 0,70$ ) and variety factor (negative

dependance,  $r=-0,53$  tyrosine). GALTERIO *et al.* (1994) and CUBADDA, MARCONI (1996) also confirmed the high proportion of valine.

Correlation analysis of each essential amino acid also provides very interesting findings (*table 5*). They are evident and obvious from the statistics ( $p<0,05$ ). Threonine content is in positive correlation to isoleucine content ( $r=0,96$ ), leucine content ( $r=0,91$ ) and lysine content ( $r=0,95$ ). The proportion of valine is in positive correlation to phenylalanine content ( $r=0,99$ ). Isoleucine is in positive correlation to leucine content ( $r=0,98$ ) and lysine content ( $r=0,95$ ).

Table 5

**Correlations between essential amino acid in emmer flour**

Amino acid	Thr							
Threonine (Thr)	1,00	Val						
Valine (Val)	0,73*	1,00	Met					
Methionine (Met)	0,89*	0,59*	1,00	Iso				
Isoleucine (Iso)	0,96*	0,86*	0,89*	1,00	Leu			
Leucine (Leu)	0,91*	0,90*	0,84*	0,98*	1,00	Tyr		
Tyrosine (Tyr)	0,76*	0,81*	0,80*	0,85*	0,81*	1,00	Phe	
Phenylalanine (Phe)	0,76*	0,99*	0,65*	0,88*	0,92*	0,86*	1,00	Lys
Lysine (Lys)	0,95*	0,69*	0,92*	0,95*	0,88*	0,77*	0,71*	1,00
*Correlations are significant at $p < 0,05$								

According to the findings mentioned above, the average proportion of amino acid in 6 tested forms of emmer wheat is lower than the same proportion tested in 2 controlling varieties of wheat. It is necessary to take specific proportion of amino acids in each variety and the total protein content into account. E.g. variety D2 Ruzyně contains 2,45 g of lysine/100 g of proteins (chemical score of 0,42). The controlling variety called M6 Vánek achieved the same figures (2,43 g of lysine/100 g of proteins, chemical score of 0,42). However, if the figures converted to 1 000 g of flour, then the protein content in emmer wheat grains is higher, so the total lysine content becomes higher too (D2 Ruzyně 4,48g/1000 g of flour - CB and 5,09g/1000 g of flour - CRI) contrary to the controlling variety called M6 Vánek (3,45 g/1000 g of flour- CB and 3,99g/1000 g of flour - CRI). There is the difference; D2 Ruzyně variety has higher proportion of lysine (1,03 or 1,1 g of lysine/1000 g of flour or more in dependence of location).

Table 6

## Conversion of amino acid content from g/100 g protein to g/1000 g flour

	Crude protein content (%)			Lysine (g/100 g protein)				chemical score	Lysine content in flour (g/1000 g flour)		
	Location			Location					Location		
	CB	CRI		CB	CRI		mean		CB	CRI	
Emmer											
D1	15,6	18,0		2,237	2,889		2,56	0,44	3,49		5,2
D2	18,6	20,4		2,409	2,495		2,45	0,42	4,48		5,09
D3	14,4	15,9		2,319	2,138		2,29	0,40	3,34		3,4
D4	20,0	20,2		1,715	2,540		2,13	0,37	3,43		5,13
D7	19,2	19,6		2,385	2,311		2,35	0,41	4,56		4,53
D10	18,3	19,0		2,257	2,663		2,46	0,42	4,13		5,06
mean	17,7	18,9		2,22	2,51		2,37	0,41	3,91		4,74
chemical score	-	-		0,38	0,43		-	-	-		-
Modern varieties											
M6	14,8	15,8		2,331	2,525		2,43	0,42	3,45		3,99
M10	15,3	15,0		2,556	2,447		2,50	0,43	3,9		3,67
mean	15,1	15,4		2,44	2,49		2,47	0,43	3,68		3,83
chemical score	-	-		0,42	0,43		-	-	-		-

## CONCLUSIONS

Crude protein content is not the crucial factor of grain quality. It is also necessary to know the structure of protein fractions, baking qualities and the composition of amino acids. According to the findings, lysine is the limiting amino acid in emmer wheat and bread wheat too. There are no considerable differences in lysine content between the tested varieties (chemical score varies from 0,37 to 0,44). Threonine is the second limiting amino acid (chemical score varies from 0,66 to 0,73). It is followed by leucine (chemical score varies from 0,80 to 0,84), tyrosine and phenylalanine (chemical score varies from 0,92 to 0,96). On the other hand, the proportions of isoleucine and valine are not sufficient.

The proportion of amino acids was obviously ( $p < 0,05$ ) influenced by crude protein content (valine  $r = -0,72$ ; tyrosine  $r = -0,56$ ; phenylalanine  $r = -0,73$ ) in negative way. The proportions of valine, leucine, tyrosine, phenylalanine were obviously ( $p < 0,05$ ) influenced by the relation to species and variety; the controlling varieties of wheat achieved higher values. The correlation analysis of essential amino acids also provides very interesting figures; threonine content is in positive correlation to isoleucine content ( $r = 0,96$ ), leucine ( $r = 0,91$ ) and lysine ( $r = 0,95$ ). The proportion of valine is in positive correlation to phenylalanine content ( $r = 0,99$ ). Isoleucine is in positive correlation to leucine content ( $r = 0,98$ ) and to lysine content ( $r = 0,95$ ).

According to the findings, the proportion of amino acids (converted to 100 g of proteins) in emmer wheat grains is higher than the proportion of amino acids in grains of the controlling varieties. However, it is very important to take the specific proportion of amino acids in relation to the total protein content in each variety into account. E.g. D2 Ruzyně variety contains the same amount of lysine (g/100 g of proteins) as the controlling variety M6 Vánek. When higher crude protein content in flour and the conversion to 1 000 g of flour taken into account, the proportion of lysine is by 1,1 g/1 000 g of flour higher (contrary to the controlling variety).

According to the findings mentioned above, emmer wheat contains the same amino acids as modern varieties of wheat. However, it is characterised by higher protein content in grain and higher content of amino acids. Therefore, the grains of emmer wheat can be used for the production of nutritional valuable diet (organic foodstuffs).

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