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Characteristic of the nutritive value of the protein from rye caryopses[1]
I. Amino acid composition of protein and the nitrogen forms in the caryopses of ten rye varieties from the breeding collection

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Abstract

Total and soluble nitrogen, protein and non-protein nitrogen was determined as well as the amino acid composition of the caryopses of ten rye varieties including three bred in Poland and cultivated on a commercial scale: 'Dańkowskie Złote', 'Dańkowskie Selekcyjne' and 'Borkowskie Tetra', and seven foreign varieties characterized by a high total protein content (11.9-16.4% in dry weight). In the varieties examined the amount of protein nitrogen increased in the same degree as did the content of total nitrogen. The amino acids limiting the nutritive value of the protein in rye caryopses were mostly lysine and methionine, and in the varieties with high protein content tryptophan. The low-protein varieties had a relatively higher content of lysine, sulphur amino acids, tryptophan and other amino acids (as % of protein) than the high protein ones, but their absolute amino acid content (as % of dry weight) was lower.

INTRODUCTION

In some investigations of the protein and exogenous amino acids content in rye caryopses the intervarietal differences were found to be small. Wider differences were due to soil and climatic conditions (Trzebska-Jeske and Morkowska-Gluzińska 1963; Janicki et al. 1967). The results of Dimenstein and Ermakov (1958), Focke (1956) and Balint and Kotvics (1970) indicate, ho-

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wever, that the greatest differences, as regards the content of protein and some amino acids, occure between rye varieties and species. Some varieties of *Secale montanum* contained about 24 per cent protein, and after crossing with *Secale cereale* gave forms with a high content of protein, rich in lysine and methionine (Balint and Kotvics 1970). Introduction for cultivation of ryes with increased protein and exogenous amino acids content, preserving at the same time other valuable traits such as high yield, resistance to drought, diseases, lodging and frost would be of great importance for improving in Poland the balance of fodder protein, the deficit of which is evaluated at 600 000 tons (Janicki 1972).

The collaboration established with Plant Breeding Stations allows an analysis of the biological value of the material of current interest to rye breeders.

MATERIAL AND METHODS

Samples of the three Polish rye varieties and of the seven foreign ones ('Svalöf Urm', 'Dominant', 'Lvovskaya', 'Toivo', 'Weser', 'Bonel', 'Elbon') originated from 1970 harvest at the Plant Breeding Station in Laski. The caryopses were ground in a Wiley mill to a 40 mesh meal. Total, soluble (in 0.1 M phosphate buffer pH 7.0) and non-protein nitrogen content (after precipitation of the protein fraction from the soluble one with trichloroacetic acid of 12% final concentration) was determined by Kjeldahl's method. For converting total nitrogen to protein the coefficient 5.67 (Janicki et al. 1972) was used. From the difference between total and soluble non-protein nitrogen, the value of protein nitrogen was calculated. This method doesn't distinguish between many nitrogen fractions, but it is sufficiently accurate to find application in comparative studies of protein nitrogen forms. The results are given in grams of nitrogen per 100 grams of caryopses (on a dry weight basis), and, for the particular fractions in per cent in relation to the total nitrogen content (Table 1).

The full amino acid composition of the caryopses of the varieties studied (ground to 40 mesh) was determined by the method of column chromatography with the use of ion exchangers, of Spackman et al. (1958) in the modyfication of Korolczuk and Rakowska (1972) on an amino acid analyser (Technicon). Protein was hydrolysed for 20 h with 6 N hydrochloric acid in high excess with the addition of 1% phenol (Blackburn 1968) at 105° ± 2°C, and for complete release of isoleucine, leucine and valine for 70 h (Smith and Stockell 1954). For cysteine, cystine and methionine determination hydrolysis was run after previous oxidation of these amino acids by means of performic acid

Table 1

Content of different nitrogen fractions in rye caryopses

No.	Variety	N_t*	N_s**		N_n***	$N_t - N_n$	
		g/100 g of grains d.wt. basis	g/100 g of grains d.wt. basis	N_t %	g/100 g of grains d.wt. basis	g/100 g of grains d.wt. basis	N_t %
1	Dańkowskie Złote	1.44	0.44	30.6	0.24	1.20	83.3
2	Dańkowskie Selekcyjne	1.57	0.52	33.1	0.22	1.35	86.0
3	Borkowskie Tetra	1.93	0.55	28.5	0.29	1.64	85.0
4	Dominant	2.10	0.58	27.6	0.28	1.82	86.7
5	Svalöf Urm	2.14	0.66	30.8	0.33	1.81	84.6
6	Lvovskaya	2.34	0.67	28.6	0.30	2.04	87.2
7	Toivo	2.60	0.74	28.5	0.37	2.23	85.8
8	Bonel	2.79	0.79	28.3	0.27	2.52	90.3
9	Weser	2.88	0.80	27.8	0.24	2.64	91.7
10	Elbon	2.89	0.85	29.4	0.34	2.55	88.2

* N_t — total nitrogen;
** N_s — soluble nitrogen;
*** N_n — non-protein nitrogen.

(9 parts of formic acid and 1 part of 30% hydrogen peroxide) to cysteic acid and methionine sulphone. The oxidated forms of these amino acids are not decomposed during acid hydrolysis of protein (Schram et al. 1954).

Tryptophan was determined colorimetrically after reaction with p-dimethylaminobenzaldehyde by the method of Horn and Jones (1945), in the modyfication of Lombard and de Lange (1965) and Skibińska and Kakowska-Lipińska (1970) after enzymatic hydrolysis with papain activated with sodium cyanide. The content of the particular amino acids is given in mg/g of nitrogen and in mg/100 g dry weight of caryopses. The correlation between protein and lysine, methionine and tryptophan contents was calculated. The coefficients of the nutritive value of the protein of caryopses of the varieties studied are calculated on the basis of their amino acid composition, Chemical Score (CS) (Mitchell and Block 1946) and Essential Amino Acid Index (EAAI) (Oser 1951). The coefficients of correlation between total protein content and that of the particular exogenous amino acids, and between protein content and the coefficients of nutritive value CS and EAAI, as well as between total protein and the amount of "useful" (%N×5.67×CS%) protein were calculated.

RESULTS AND DISCUSSION

The result of determination of the content of different nitrogen forms in the ten rye varieties examined are compiled in Table 1. As total nitrogen content increases in the caryopses, the contribution of the non-protein form to the pool of total nitrogen decreases, and that of the protein form rises (Fig. 1), the coefficient of correlation being r = 0.996.

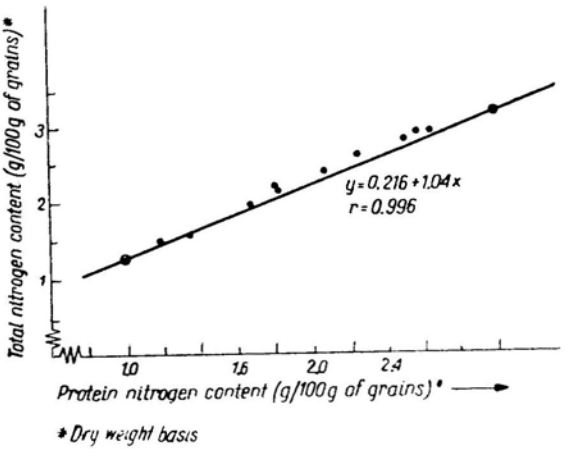

Fig. 1. Correlation between total and protein nitrogen content in caryopses of various rye varieties

Tab

Amino acid composition of 10 rye varieties in mg/gN (*a*)

Variety / Amino acid	Dańkowskie Złote		Dańkowskie Selekcyjne		Borkowskie Tetra		Dominant	
	a	*b*	*a*	*b*	*a*	*b*	*a*	*b*
Tryptophan	65.2	93.9	69.0	108	68.5	132	51.9	109
Methionine	138	199	132	207	123	237	111	233
Cystine+Cysteine	169	243	165	259	159	307	170	357
Aspartic acid	538	774	588	923	473	913	529	1110
Threonine	245	353	226	355	246	475	239	502
Serine	298	429	283	444	324	625	306	645
Glutamic acid	1510	2170	1400	2200	1650	3180	1670	3510
Proline	664	956	649	1020	793	1530	766	1610
Glycine	308	444	267	419	247	477	297	624
Alanine	305	439	299	469	290	560	279	586
Valine	324	467	305	479	293	565	270	567
Isoleucine	208	300	237	372	217	419	269	565
Leucine	439	632	412	647	393	758	423	888
Tyrosine	180	259	154	242	159	307	156	328
Phenylalanine	294	423	268	421	293	565	333	699
Lysine	260	374	238	374	210	405	230	483
Histidine	133	192	144	226	125	241	135	284
Arginine	333	480	341	535	321	620	354	743

When we compare amino acids determinations in rye by various authors (Trzebska-Jeske and Morkowska-Gluzińska 1963, and others) a wide dispersion of results is observed, particulary as regards lysine, cystine + cysteine, tyrosine and histidine. In the case of lysine and histidine this is mainly due to intervarietal differences, whereas for cystine + cysteine and tyrosine, which readily undergo oxidation, it may result from differences in the preparation of hydrolysates. Table 2 lists the contents of all protein amino acids in mg/g nitrogen and in mg/100 g dry weight of caryopses from 10 rye varieties containing 8.16-16.4 per cent protein (converted to dry weight). The content of cysteic acid corresponding to the sum of cystine and cysteine exceeds somewhat the literature data referring to the content of these amino acids in the protein of rye caryopses, with the exception of the results of Tkachuk and Irvine (1969) and those of Schram et al. (1954), obtained after cystine and cysteine oxidation. In varieties with high protein content a relatively lower level of the following amino acids is observed (mg/g N): tryptophan, lysine, methionine, cystine, cysteine, aspartic acid, threonine, glycine, alanine, valine and leucine than in the low-protein varieties (Table 2, Fig. 2*A*). At the same time, the tendency to a rise of all amino acids level per weight unit of the caryopses observed earlier in wheat (Janicki et al. 1967; Lawrance

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and mg/100 g of caryopses (d.wt.basis) (*b*)

Svalöf Urm		Lvovskaya		Toivo		Bonel		Weser		Elbon	
a	*b*	*a*	*b*	*a*	*b*	*a*	*b*	*a*	*b*	*a*	*b*
57.1	122	58.4	137	49.6	129	42.8	119	45.1	130	48.1	139
115	246	112	262	109	283	88.0	246	97.9	282	92.1	266
181	387	166	388	137	356	134	374	129	372	127	376
475	1020	438	1020	413	1070	357	1000	426	1230	408	1180
237	507	217	508	218	567	194	541	195	562	186	538
306	655	294	688	271	705	246	686	279	804	261	754
1810	3870	1690	3950	1510	3930	1440	4020	1760	5070	1700	4910
668	1430	711	1660	683	1780	751	2100	802	2310	800	2310
262	561	271	634	265	689	209	583	233	671	225	650
293	627	282	660	255	663	241	672	253	729	242	699
317	678	260	608	297	772	206	854	274	789	227	656
236	505	240	562	235	536	224	625	206	662	230	665
400	856	399	931	398	959	399	1110	369	962	334	965
159	340	154	360	154	400	158	441	150	432	154	445
313	670	307	718	293	761	307	857	291	838	304	879
225	482	209	489	218	567	210	586	183	527	205	592
137	293	132	309	139	361	137	382	131	377	126	364
386	826	363	849	391	1020	330	921	304	876	315	910

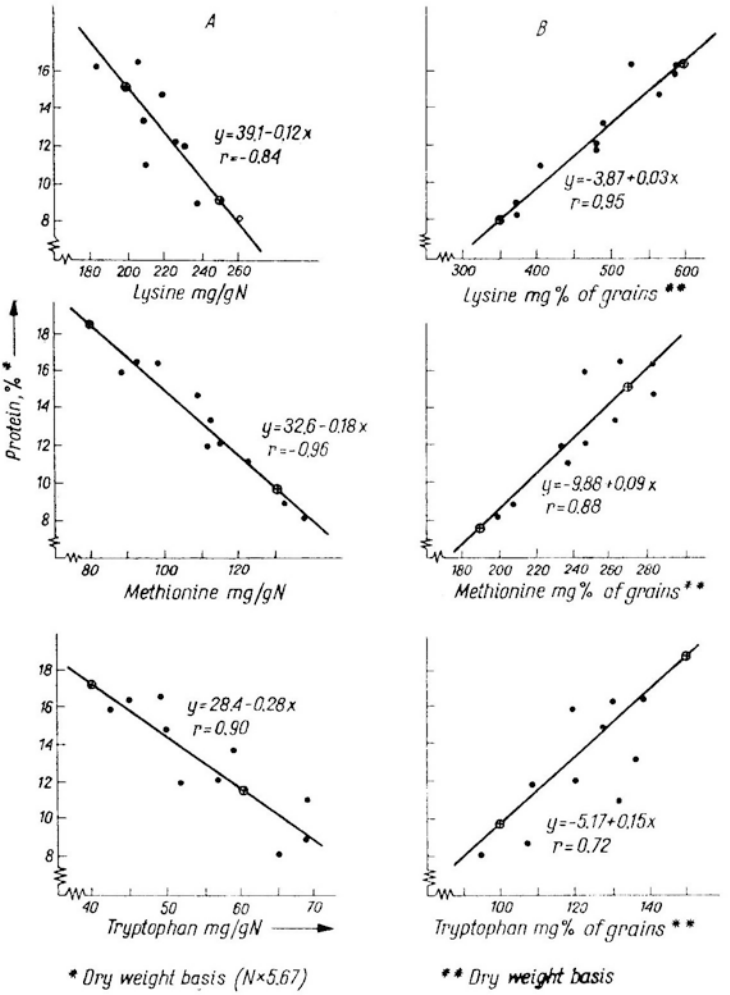

Fig. 2. *A*. Correlation between protein content and the content of lysine, methionine and tryptophan in the protein of caryopses of various rye varieties

B. Correlation between the protein content and of lysine, methionine and tryptophan content expressed in mg % of caryopses of various rye varieties (expressed as samples)

et al. 1958; Mc Dermott and Pace 1960) and in maize (Bressani et al. 1962; Dumanovic and Denic 1969) was confirmed in rye as the total protein content increased in various varieties (Table 2, Fig. 2*B*).

The varieties with high protein content exhibited a lower nutritive value coefficients than the low-protein ones, as calculated on the basis of amino acids composition, thus CS and EAAI (Table 3). The amount of "useful" protein, however, obtained per weight unit of caryopses of

Table 3

Nutritional value and "useful" protein content in 10 rye varieties

No.	Variety	Total protein % d.w.t. basis	Chemical Score (CS) %	Limiting amino acids	Essential Amino Acid Index (EAAI)	"Useful" protein (total protein × × CS) % d.wt. basis
1	Dańkowskie Złote	8.16	65	Try, Lys, Met	71.7	5.39
2	Dańkowskie Selekcyjne	8.88	59	Lys, Met, Try	68.5	5.24
3	Borkowskie Tetra	11.0	53	Lys, Met, Try	66.8	5.83
4	Dominant	11.9	52	Try, Lys, Met	68.0	6.90
5	Svalöf Urm	12.1	56	Lys, Try, Met	67.1	6.78
6	Lvovskaya	13.3	52	Lys, Met, Try	65.3	6.92
7	Toivo	14.7	50	Try, Lys, Met	63.2	8.09
8	Bonel	15.9	43	Try, Met, Lys	64.1	7.16
9	Weser	16.3	45	Try, Lys, Met	59.8	7.34
10	Elbon	16.4	48	Try, Met, Lys	58.6	7.87

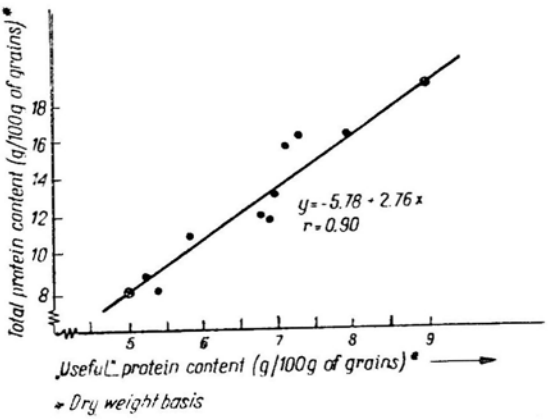

Fig. 3. Increase in „useful" protein content (total protein × CS) with rise in total protein content in rye caryopses

the high protein rye varieties is higher than that found in low-protein varieties (Fig. 3). It therefor seems reasonable to try to select varieties with a high protein content which would at the same time be highly fertile, without losing sight of the biological value of the breeding material protein (Kubiczek and Molski 1973).

REFERENCES

Balint A., Kotvics G., 1970, Investigations on increasing the protein content of *Secale cereale* L., in Protein Growth by Plant Breeding, Akademiai Kiado, Budapest, 89-98.

Blackburn S., 1968, Amino acid determination, Methods and Techniques, Marcel Dekker, New York.

Bressani R., Elias L. G., Scrimshaw N. S., Guzman M. A., 1962, Nutritive value of Central American Crops; VI Varietal and environmental influence on the nitrogen, essential amino acid and fat content of ten varieties, Cereal Chem. 39 (1): 59-67.

Dimenstein F. J., Ermakov A. I., 1958, Biokhimiya rzhi, Sel'skokhoz. Izdat. Moskva, I: 165-234.

Dumanovic J., Denic M., 1969, Variation and heritability of lysine content in maize, New Approaches to Breeding for Improving Plant Protein, IAEA/FAO, Vienna, 109-116.

Focke R., 1956, Über den Rochproteingehalt von *S. cereale* und *S. montanum* und den prozentualen Anteil einiger Aminosäuren, Züchter, 26, 1/2, 40-41.

Horne M. J., Jones D. B., 1945, J. biol. Chem., 157, 153 [after Lombard and de Lange 1965].

Janicki J., Kowalczyk I., Riewe I., 1967, Skład animokwasowy niektórych polskich odmian pszenicy i żyta, Roczniki Technologii i Chemii Żywności 13 (5): 5-16.

Janicki J., 1972, Wybrane zagadnienia z biochemii ziarna zbóż. Zeszyty Problemowe Postępów Nauk Rolniczych 125: 37-53.

Janicki J., Warchalewski J., Stasińska B., Kowalczyk I., 1972, Badania składu aminokwasowego niektórych mąk, Roczniki Technologii i Chemii Żywności 22 (1): 71-72.

Korolczuk J., Rakowska M., 1972, unpublished data.

Kubiczek R., Molski B., 1973, Wartość odżywcza białka ziarniaków żyta oraz metody jej oceny, (in press).

Lawrance J. M., Day K. M., Huey R., Lee B., 1958, Lysine content of wheat varieties, species and related genera, Cereal Chem. 35 (3): 169-178.

Lombard J. H., de Lange D. J., 1965, The chemical determination of tryptophan in foods and mixed diets, Anal. Biochem. 10: 260-265.

Mc Dermott E. E., Pace I., 1960, Comparison of the amino-acid composition of the protein in flour and endosperm from different types of wheat, with particular reference to variation in lysine content, J. Sc. Food Agricult. 11: 109-115.

Mitchell H. H., Block R. J., Some relationships between the amino acid content of proteins and their nutritive values for the rat, J. biol. Chem. 163, 599.

Oser B. L., 1951, Method for integrating essential amino acid content in the nutritional evaluation of protein, J. A. Dietet. Ass., 27: 396-402.

Schram E., Moore S., Bigwood E. J., 1954, Chromatographic determination of cystine as cysteic acid, Biochem. J., 57: 33.

Skibińska T., Kakowska-Lipińska I., 1970, Oznaczanie tryptofanu w produktach spożywczych, Roczniki Państwowego Zakładu Higieny 21 (3): 303-310.

Smith E. L., Stockell A., 1954, Amino acid composition of crystalline carboxypeptidase, J. biol. Chem. 207 (2): 501-514.

Spackaman D. H., Stein W. H., Moore S., 1958, Automatic recording apparatus for use in the chromatography of amino acids, Anal. Chem. 30: 1190-1206.

Tkachuk R., Irvine G. N., 1969, Amino acid composition of cereals and oilseeds meales, Cereal Chem. 46 (2): 206-218.

Trzebska-Jeske I., Morkowska-Gluzińska W., 1963, Zawartość azotu i niektórych aminokwasów niezbędnych w ziarnie żyta polskiego ze zbiorów 1959, Roczniki Państwowego Zakładu Higieny 14 (2): 153-164.

Charakterystyka wartości odżywczej białka ziarniaków żyta
I. Skład aminokwasowy białka oraz formy azotu w ziarniakach 10 odmian żyta z kolekcji hodowlanej

Streszczenie

Oznaczono zawartość azotu ogólnego, azotu rozpuszczalnego, niebiałkowego i białkowego oraz skład aminokwasowy ziarniaków dziesięciu odmian żyta, w tym trzech odmian polskich — będących w rejonizacji 'Dańkowskiego Złotego', 'Borkowskiego Tetra' i wychodzącego z rejonizacji 'Dańkowskiego Selekcyjnego' oraz siedmiu odmian obcych charakteryzujących się wysoką zawartością białka ogólnego (11,9-16,4% w suchej masie).

W przebadanych odmianach wraz ze wzrostem zawartości azotu ogólnego zwiększyła się w tym samym stopniu ilość azotu białkowego.

Aminokwasami ograniczającymi wartość odżywczą białka ziarniaków żyta były najczęściej: lizyna, metionina oraz w odmianach wysokobiałkowych tryptofan. Odmiany niskobiałkowe przewyższały wysokobiałkowe względną zawartością lizyny, aminokwasów siarkowych, tryptofanu i innych (w % białka), ale ustępowały im bezwzględną zawartością aminokwasów (w % suchej masy).

Cena zł 26.—