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PROTEIN, AMINO ACID AND MINERAL COMPOSITION OF SOME CULTIVARS OF BREAD WHEAT

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Total protein, amino acids and Ca, Zn, Cu, Fe, Na and Mg contents were determined in eight selected cultivars of bread wheat, *Triticum aestivum* (AABBDD, 2n = 6x = 42) and compared with their respective mutants. The material exhibited pronounced genetic variability in protein content and quality. Some mutants were found to differ significantly from the parent cultivars in their amino acids and mineral compositions. Enhanced production of essential amino acids such as lysine, threonine, valine, metheonine, leucine and isoleucine, were found in some wheat mutants. A mutant, M-11 has been identified with significantly higher ($P < 0.05$) proline value than its mother cultivar C- 591. The present study shows that induced mutations can be utilized for alteration / enhancement of amino acids and mineral composition of cereals.

Key words: Protein, Amino acid, Wheat.

Introduction

Wheat, *Triticum aestivum* (AABBDD, 2n = 6x = 42) is the world's most important food crop which provides energy, protein, minerals and vitamins to most people [1]. In Pakistan self - sufficiency in wheat has been of prime consideration by the government [2]. Modern plant breeding has played a vital role in enhancing the quantity and quality of wheat in Pakistan. The determination of protein, amino acids and mineral quality is therefore, essential when working with new cultivars of wheat . Various methods have been developed to study the wheat quality, based on simple chemical and physical tests [3,4].

The present study reports total protein, amino acids and mineral composition (Ca, Zn, Cu, Fe, Na and Mg) of eight cultivars of bread wheat and their respective mutants, grown under similar conditions at the experimental farm of the AEARC, Tandojam.

Experimental

Sample description. The details of the wheat cultivars and their respective mutants are as under:

Parent	Mutant	Parent	Mutant
C-591	M - 11	Sarsabz	M - 147
Indus-66	M - 37	Pavon	M - 143
6134 x C-271	TDS	Sonalika	M - 233
Sind - 81	M - 179	Nayab	Jauhar-78

Procedure. All wheat samples were ground in a hammar mill to pass a 0.024 mm screen. Nitrogen and total protein was determined by Kjeldahl method [5].

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The 18 commonly occuring amino acids were determined in all samples by 6N HCl hydrolysis of wheat flour and subsequent ion-exchange chromatography on a Hitachi Model 835A Amino Acid Analyser. Determination of cystine and methionine was done as per procedure of Gehrke *et al.* [6]. Tryptophan was determined using spectrophotometric method [7].

Mineral analysis was carried out by atomic absorption spectrophotometer, 1 g samples were wet - ashed with nitric acid as per procedure of Lorenz and Loewe [8]. Absorption measurement were made in clear digests by Hitachi Model 180-50 atomic absorption spectrophotometer. Optimum instrument parameters were adjusted according to manufacturer's instructions. An air-acetylene flame was used for all elements. All the samples were analysed in triplicate.

Results and Discussion

The total protein content (N x 5.7) of the wheat cultivars analysed ranged from 9.75 to 13.62% (Table 1). Mutants M-143, and M-233 which are derived from Pavon and Sonalika respectively, showed higher protein than their parent cultivars. Total protein contents were found to be higher in the parent cultivars C-591, 6134 x C-271, and Sind - 81. The protein values are within the acceptable limits and compare favourably with the values reported by others [9,10].

Table 1 also gives a detailed survey of eighteen amino acid contents in the present bread wheat cultivars and their mutants. The data is obtained after optimization of the conditions for 6N HCl hydrolysis prior to chromatographic analysis as recommended by Zumwalt *et al.* [11].

Statistical analysis: The t - test analysis of the amino acids contents for all samples was done to establish any statis-

TABLE 1. PROTEINS, AMINO ACIDS CONTENTS OF WHEAT CULTIVARS.

Wheat cultivars	C-591	M-11	Indus -66	N-37	6134 x C-271	TDS	Sindh -81	M-179	Sarsabz	N-147	Pavon	N-143	Sonalika	N-233	Nayab	Jauhar -78
Total Protein Nx5.7	12.77	12.14	11.26	9.75	12.80	10.94	13.62	11.91	10.93	11.23	11.12	12.03	11.40	12.08	10.77	10.1
Asp	4.72	4.76	4.66	4.70	5.10	4.70	4.19	4.56	5.69	4.73*	4.36	4.85*	5.49	4.19*	3.54	2.75
Thr	2.36	1.66*	2.51	1.59*	1.71	2.54*	2.31	2.47	2.79	2.46*	2.50	2.54	3.08	2.49*	2.08	1.66*
Ser	3.17	1.33*	3.62	1.20*	1.29	3.60*	3.01	3.76	4.06	3.60	3.30	3.26	4.63	3.54*	2.92	2.26
Glu	31.16	28.09*	29.90	29.11*	32.31	29.86*	23.50	28.43*	33.43	31.44	30.64	30.78	29.68	25.21*	25.79	20.71*
Pro	9.39	11.14*	8.61	8.36*	9.46	8.96	7.24	7.98	9.35	8.57	8.44	9.08	7.06	5.97	6.99	6.70
Gly	3.63	3.64	3.34	3.65*	3.61	3.56	3.31	3.75	4.42	4.23	3.83	3.57	4.13	3.20*	3.36	1.88
Ala	2.92	2.91	2.78	2.91	2.76	2.82	2.76	3.01	3.66	3.61	3.06	2.83	3.32	2.49	2.69	2.41
Cys	1.34	1.15*	1.87	1.79	2.00	2.05	1.17	1.55	1.36	1.24	1.43	1.26*	1.48	1.74	2.17	2.16
Yal	4.07	3.79*	3.79	3.51*	3.95	3.66	3.58	4.25*	4.52	4.51	4.76	4.14*	3.79	3.25	3.73	2.84
Met	1.82	1.65*	1.55	1.63	1.86	1.73	1.43	1.39	1.91	2.01*	1.87	1.90	2.12	1.94*	2.01	1.84*
Ileu	3.07	2.63*	2.95	2.73	2.88	2.82	2.57	3.21	3.56	3.51	1.27	2.97	2.99	2.65*	2.74	2.27*
Leu	5.74	5.22*	5.43	5.29	5.68	5.27	4.96	5.64	6.47	6.18	6.24	5.68	5.48	4.93	5.29	4.12*
Tyr	2.67	3.10*	2.95	2.74*	3.32	3.27	2.26	3.43	3.93	3.38*	3.66	3.85	3.90	3.93	3.83	2.27*
Phe	4.18	3.49*	4.02	3.41	3.97	3.70	3.62	3.87	4.25	3.77	4.32	4.01	3.72	3.76	3.43	2.48*
Lys	1.98	1.94	1.87	1.95	2.01	1.83	1.75	2.16*	2.25	2.52*	1.93	1.95	2.23	1.82*	1.89	1.48*
NH_3	3.59	4.55*	3.74	4.54*	4.16	4.17	3.79	5.88*	6.84	4.80	4.63	4.28	5.05	4.65*	7.54	8.04
His	2.14	1.87*	1.66	1.79	2.05	1.73	1.42	1.84	2.25	1.84*	1.75	1.71	1.78	1.59	1.74	1.67
Arq	3.65	3.58	3.95	3.53	3.85	3.86	3.77	3.68	4.39	4.52*	3.94	1.53*	1.46	3.89	3.60	2.66
Trp	1.12	1.63*	1.30	1.03*	1.82	1.36	1.50	1.99*	1.16	2.04*	1.21	1.84*	1.30	1.74*	1.18	1.13

* Denotes that the mutant value is significantly different from its mother variety.

tically significant difference between the parent and the mutant wheat cultivars. Table 1 also reveals such differences. Mutant M-11 derived from 350 Gy treatment of wheat cultivar C-591 have significantly more tyrosine. M-11 and M-143 also contain more ($p< 0.05$) proline than their mother cultivars. Its production was also found to be significantly higher ($P<0.05$) in Indus-66 and Sonalika. These cultivars have exhibited wide adaptability at international level.

Mutant-37 derived from Indus-66 (200 Gy) has radiation-induced necrotic spots on the leaf and the grain yield of M-37 is significantly lower than Indus-66 [12] apparently due to inherent damage to the photosynthetically active tissues. M-37 produced significantly more ($P < 0.05$) glycine, which is perhaps a reflection of the implication of glycine in physiological mechanism connected with photosynthesis.

Since 1979, Pavon has remained the dominant wheat cultivar in Sindh (Pakistan). The area occupied by Pavon has been partially replaced by wheat cultivars namely Jauhar - 78 in Sind Province. Th release of Mutant-143 as Saughat-90 is a new scientific development [2]. The present results suggest that M-143 produced significantly higher ($P < 0.05$) quantity of aspartic acid in comparison with Pavon, while no significant increase in the amino acids compsition of the mutants (M-233 and Jauhar-78) were observed. Valine was significantly higher in C-591, Indus-66 and Pavon of all the wheat cultivars. Glutamic acid production was higher in Sonalika followed by C-591, Nayab, Indus-66 and 6134 x C-271. In serine production Indus-66 was higher followed by C-591 and Sonalika. Similarly in glycine production Nayab surpassed all the cultivars. Pavon and C-591 produced significantly higher ($P < 0.05$) cystine in comparison with other cultivars. Pavon was also most outstanding with respect to arginine production. TDS which is derived from 6134 x C-271, produced significantly higher ($P < 0.05$) quantities of serine and arginine. This mutant is short which provides effective protection against lodging and may tolerate high nitrogenous fertilizers. Mutant M-179 which is derived from Sindh-81 produced significantly more ($P < 0.05$) valine in addition to serine, glutamic acid, glycine and cystine. This is perhaps the most outstanding example of a wheat mutant representing induced genetic changes for the production of different amino acids.

Essential amino acids. The concentration of essential amino acids such as threonine, lysine and methionine is often expressed as a function of nitrogen content of grain. Considerable efforts have been devoted at international level for increasing the lysine content of wheat [13, 15]. In the present work a new wheat mutant M-179 has been identified which produced significantly higher ($P < 0.05$) lysine content than the mother cultivar, Sind-81. Mutant-147 derived from an outstanding, genetically verstile variety produced significantly higher ($P < 0.05$) quantities of two essential amino acids namely lysine and methionine than Sarsabz. This is a significant finding. Mutant-147 can be utilized for improving the lysine content of Sarsabz through a series of backcrossing. TDS which is mutant of 6134 x C-271 produced significantly higher quantities of threonine. Trypophan, a nutritionally essential amino acid, has been found to be significantly higher ($P < 0.05$) in M-11, M-179, M-147, M-143 and M-233.

Among the parent wheat cultivars lysine contents were significantly higher ($P < 0.05$) in Sonalika followed by Nayab. Likewise, leucine contents were significantly higher ($P < 0.05$) in C-591, Nayab and Pavon in comparison with all

TABLE 2. MINERAL COMPOSITION OF WHEAT CULTIVARS.

Wheat cultivars	Calcium (μg/g)	Zinc (μg/g)	Copper (μg/g)	Iron (μg/g)	Sodium (%)	Magnesium (%)
C-591	221.43 ±10.10	49.94 ± 0.85	4.73 ± 0.61	58.59 ± 1.52	0.107 ± 0.001	0.171 ± 0.00
M-11	196.43 ± 5.01	53.51 ± 3.56	5.98 ± 0.53	42.88 ± 1.52	0.140 ± 0.001	0.187 ± 0.00
Indus-66	178.57 ± 1.20	44.37 ± 1.10	6.42 ± 0.00	35.79 ± 0.00	0.891 ± 0.002	0.142 ± 0.001
M-37	210.72 ± 5.06	50.33* ± 1.31	4.53 ± 0.00	29.65* ± 1.52	0.086 ± 0.001	0.139 ± 0.001
6134 x C271	20.00 ±10.10	38.85 ± 0.77	2.64 ± 0.00	N.A.	0.114 ± 0.003	0.170 ± 0.001
TDS	19.10 ± 9.21	52.10* ± 2.20	4.84* ± 0.40	N.A.	0.130 ± 0.002	0.173 ± 0.002
Sind-81	185.72 ±10.01	39.29 ± 1.01	4.53 ± 0.00	20.88 ± 1.52	0.134 ± 0.00	0.129 ± 0.001
M-179	235.72* ±10.10	33.11 ± 4.59	4.53 ± 0.00	1.58* ± 0.00	0.005 ± 0.001	0.121 ± 0.001
Sarsabz	167.86 ± 5.05	44.02 ±13	5.10 ± 0.49	32.28 ± 3.04	0.168 ± 0.005	0.153 ± 0.001
M-147	150.01* ± 5.05	43.74 ± 0.73	6.07 ± 0.60	44.56* ± 1.56	0.095 ± 0.001	0.163 ± 0.001
Pavon	182.14 ± 5.10	48.76 ± 0.73	5.10 ± 0.49	46.48 ± 1.16	0.076 ± 0.003	0.117 ± 0.001
M-143	185.72 ± 5.05	53.13 ± 2.17	5.73 ± 0.60	56.39* ± 0.58	0.085 ± 0.003	0.155 ± 0.002
Sonalika	189.29 ± 5.11	48.13 ± 1.53	4.07 ± 0.40	53.33 ± 1.52	0.235 ± 0.004	0.177 ± 0.002
M-233	171.00* ± 1.01	44.37 ± 1.23	2.64* ± 0.00	12.99* ± 1.52	0.143 ± 0.003	0.144 ± 0.003
Nayab	228.57 ± 0.00	44.01 ± 0.72	4.81 ± 0.49	16.49 ± 1.52	0.069 ± 0.004	0.155 ± 0.001
Jauhar	210.72* ± 5.06	39.96* ± 0.77	6.07 ± 0.60	12.99 ± 1.52	0.109* ±0.001	0.122* ± 0.002

* Denotes that the mutant value is significantly different from its mother variety. Mean of triplicate determinations.

other cultivars. C-591, and Nayab possessed significantly higher ($P<0.05$) quantities of phenylalanine. Methionine contents were significantly higher ($P < 0.05$) in Sonalika, C-591 and Nayab. Isoleucine contents were significantly higher ($P < 0.05$) in C-591, Nayab and Sonalika. Threonine contents were significantly higher ($P < 0.05$) in Indus-65, Nayab, Sarsabz, and Sonalika.

The nutritional quality of all the cultivars of wheat studied in the present work , was evaluated by comparing the essential amino acid composition with that of FAO provisional pattern (Table 3). Except for lysine adequate amount of the essential amino acids were present in the wheat cultivars.

Mineral composition. The average values for the following minerals; Ca, Zn, Cu, Fe, Na and Mg are presented in Table 2. These values are within the range of mineral contents of wheat reported by others [8, 9, 16]. Statistical evaluation of the results of mineral compositions shows that the dwarf mutant namely TDS contained almost doubled the amount of copper as compared to 6134 x C-271, while M-179 has the highest amount of calcium. Significantly higher ($P < 0.05$) quantities of iron were detected in M-147 and M-143. In magnesium uptake M-147 was most efficient, followed by M-143. The zinc uptake was highest in M-11 followed by M-143. The zinc uptake was highest in M-37 followed by TDS. These results have theoretical as well as practical implication.

TABLE 3. ESSENTIAL AMINO ACIDS COMPOSITION OF THE PRESENT WHEAT CULTIVARS IN COMPARISON TO THE LEVELS SET BY FAO.

Amino acid	Mean value in 16 cultivars of wheat (g/16g nitrogen)	FAO level*
Lvs	1.97 (1.48 - 2.52)	4.3
Thr	2.30 (1.59 - 3.08)	3.3
Val	3.89 (2.84 - 4.76)	2.8
Ile	2.94 (2.27 - 3.56)	4.3
Met	1.79 (1.39 - 2.12)	N.A.
Leu	5.48 (4.12 - 6.47)	4.9
Tvr	3.22 (2.26 - 3.93)	2.5
Phe	3.75 (2.48 - 4.32)	2.9

* Source [17].

The genetic variation for mineral uptake was also observed in the contemporary wheat cultivars. Of all the cultivars sonalika was most efficient in uptake of copper, calcium, iron, sodium, and magnesium. Similarly Nayab exhibited efficient utilization of calcium, magnesium and zinc. Sarsabz was characterized by efficient uptake of calcium and sodium. Sind-81 was efficient in the uptake of iron, sodium and magnesium. Likewise, C-591, Indus-66 and 6134° C-271 were efficient in the uptake of iron. Indus-66 possessed significantly more ($P < 0.05$) magnesium than the respective mutant namely M-37.

Conclusion

The present investigations have elucidated the genetic variation for amino acids and mineral composition in the contemporary bread wheat cultivars. It was also found possible to induce mutants with higher protein in wheat cultivars Pavon,

Sonalika and Sarsabz. Mutant 11 originated from indigenous tall variety C-591 possessed significantly higher proline, seems to have the capabilities of adaptation under stress conditions. Lysine and methionine - two important essential amino acids were higher in the mutant 147. Improvement in the level of trypotophan in some mutants is also significant. This study shows that induced mutations can be utilized for alteration or enhancement of the mineral composition of cereals.

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References

1. S. Bajaj, Biotechnology in Nutritional Improvemnt of Wheat, in "*Biotechnology in Agriculture and Forestry*", Wheat (ed.) Bajaj, (Springer-Verlag, Berlin YPS. 13, 615 (1990).
2. K.A. Siddiqui, G. Mustafa, M.A. Arain, and K.A. Jaffri, Realities and Possibilities of Improving cereal Crops Through Mutation Breedings, "Proc. Contribution of Plant Mutation Breeding to Crop Improvement". FAO/ IAEA Vienna, 173, (1991).
3. G. Sarwar, D.A. Christensen, A.J. Finlayson, M. Friendman, L.R. Hackler, S.L. Macknenzie, P.L. Pellet and R. Tkachuk, J.Fd. Sci., **48**, 526, (1983).
4. P.C. Williams, K.R. Prestion, K.H. Norris, and P.M. Starkey, J.Fd. Sci., **49**, 17 (1984).
5. American Association of Cereal Chemistry, Approved Methods of the AACC Methods 46-12 (1969). Minneeapolis, MN.
6. C.W. Gehrke, L.L. Wall, J.S. Absheer, F.E. Kaiser, and R.W. Zumwalt, J. Assoc. Off. Anal. chem., **68**, 811 (1985).
7. K.K. Verma A. Jain and G. Gasparic, Talanta, **35**, 35 (1988).
8. K. Lorenz and R. Loewe, J. Agric. Fd. Chem., **25**, 806 (1977).
9. E. Dikeman, Y. Poeranz, and F.S. Lai, Cereal Chem., **59** 139 (1982).
10. H.A. Khatchadourian, W.N. Sawaya and M.I. Bayoumi, Cereal Chemistry, **62**, 5, 417 (1985).
11. R.W. Zumwalt, J.S. Absheer, F.E. Kaiser, and C.W. Gehrke, J. Assoc. Off. Anal. Chem., **70**, 1, 147, (1987).
12. K.A. Siddiqui and G. Arain, Euphytica, **23**, 585, (1974).
13. K.A. Siddiqi, Hereditas, **71**, 157 (1972).
14. V.A. Johnson, *Genetic Diversity in Plants* (Plenum Press New York and London, 1977), vol. 8, pp. 349.
15. B.O. Eggum, *Genetic Diversity in Plants"* (Plenum Press, New York and London 1977), Vol. 8, pp.391.
16. C.P. Czerniejewski, C.W. Shank, W.G. Bechtel and W.B.Bradley, Cereal Chem. **41**,2,65 (1964).
17. P. N. Okah, L. B. Olugbemi and S. M. Abed, J. Agric. Fd. Chem., **33**, 688 (1985).