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PROTEIN IMPROVEMENT IN WHEAT

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Abstract. Mutation breeding has been successfully employed to generate variability for protein characteristics and a few high protein wheat strains have been selected. Different levels of N fertilization have increased the protein and grain yields, and this increase is proportionately high in the radiation-induced protein-rich wheat mutant Rageni-15. It is suggested that we should consider adopting concept of 'crop protein yield' expressed for example, as kg protein/ha.

Seventy per cent of the total world supply of edible protein comes from plants.[14] In plants, wheat being the basic food for more than a billion people in the world, is a very important source of protein. Any improvement in the quality and quantity of its protein would have a significant and far-reaching consequence.

The environment under which the crop is grown has great influence on its grain protein. The protein values range as high as 18% and as low as 7% in different regions of the world. Increase in protein quantity can be accomplished with applications of nitrogen fertilizer in some areas but has serious limitations in other areas. Improved levels of grain protein probably will require a combination of fertilizer application and the use of varieties with capacity to synthesize more protein in their grain than currently grown varieties.

Mutation breeding may be of particular value in the development of high protein wheat, since there is evidence that variability can be generated for protein characteristics without disturbing a superior and well-adapted genotype.[5,9]

Materials and Methods

Seeds of a standard variety Mexi–Pak-65 were irradiated with 400 rad neutron (fast), and were space-planted. Single-spike progenies were grown in the M_2 generation for the selection of desirable mutant plants. Through this procedure, seven dwarf and early maturing mutants were selected and their breeding behaviour was confirmed in M_3 generation.

Effect of fertilizer (90, 110 and 130 kg N/ha) on grain yield and protein was tested with three varieties —Mexi–Pak-65, Chenab-70 and radiation induced mutant Rageni-15. The treatments were laid in a split plot design with four replicates. The individual plot was 5.5×1.20 m with 6 rows of wheat plants 20 cm apart.

One gram seed sample from each replication of all the varieties was taken for protein estimation by Kjeldahl method. The amino acid analysis was conducted on the EEL-193 high speed amino acid analyser.

Results and Discussion

Radiation-Induced High-Protein Wheat Mutants. M_2 generation of neutron-irradiated (400 rad) Mexi-Pak-65 wheat material was screened for preliminary selection of wheat mutants. Five hundred dwarf, semi-dwarf, early maturing and high tillering mutants were isolated.[7] These mutants were sown in M_3 for further confirmation and 336 true breeding promising mutants were isolated. The protein values of these mutants ranged from 9 to 18% and most of them had more protein than the parent variety Mexi–Pak-65 which ranges from 11 to 12%. The protein content of wheat grain is heritable trait and its inheritance is relatively simple.[5] Many workers reported the radiation-induced high protein mutants in various crops; Swaminathan *et al.* in wheat;[11] Hagberg *et al.*[2] in barley and Tanaka and Tamura in rice.[12]

Wheat mutants were further subjected to laboratory tests to study their yield components such as spike length, fertility, grain and weight. Seven most promising mutants were isolated on the basis of these tests and put in the microplot trials for yield and quality tests. The grain yield in all the 7 mutants was less than the parent variety but the total protein values on acre basis were much higher in mutants than control (Table 1).

One high protein (17.04%) bold grain wheat mutant (M-15: Rageni) was dwarf, 3 weeks early and its 'chapati' making quality was superior than that of Mexi–Pak-65, Chenab-70 and C-273. The amino acid analysis of mutant Rageni-15 and the parent variety Mexi–Pak-65 showed an increase in case of mutant in all the 16 amino acids analysed due to its high protein content. Knowing that the amino acid composition does not give an estimate of milling

TABLE 1. AVERAGE GRAIN YIELD AND PROTEIN CONTENT OF 7 WHEAT MUTANTS FROM 4 TRIALS CONDUCTED AT NIAB IN 1971.

Mutant	Protein (%)	Yield (kg/ha)	
		Grain	Protein
M-3	15.60	5082	793
M-9	14.67	4025	620
M-11	16.26	3506	570
M-15	17.04	5199	886
M-17	14.87	5230	778
M-27	16.88	4396	743
M-244	13.68	4930	674
Mexi–Pak-65 (control)	11.60	5452	633

qualities, the increase in total production of amino acids of this magnitude is important.[4]

Four amino acids in wheat protein are in short supply according to FAO determinations for human requirements (Fig. 1). The lysine in wheat protein provides less than one half of man's daily requirement and is the most critical amongst essential amino acids. Isoleucine, methionine and threonine are also deficient. Phenylalanine and leucine are strongly in excess of requirements. A significant increase of 45.16% was found in the lysine analysis of Rageni-15 mutant (0.45%) over the parent variety Mexi–Pak-65 (0.31%). Similarly, a significant increase in Rageni-15 over the control was found in the other two deficient amino acids, namely methionine and tryptophan. It was clear from our results that high lysine wheat varieties can be evolved without altering the other quality attributes. Johnson *et al.*[6] have also successfully evolved high lysine strains of wheat by hybridization. They have carried out analysis about the interrelationship of amino acids in wheat and suggested that the selection for high lysine may not be associated with adverse downward shifts in leavels of other essential amino acids.

Effect of N Fertilization on Grain and Protein Yield. The application of nitrogen at the three levels increased grain yield significantly in all the three varieties of wheat (Table 2). With no nitrogen, the yield is lower in Rageni-15 than Mexi–Pak-65 and is highest in Chenab-70. But in the 130 kg N/ha treatment, Rageni-15 has shown maximum increase that is 20% over control. At 130 kg N/ha level the difference of yield between Mexi–Pak-65 and Rageni-15 is insignificant.

There is an increase in the protein content of the three wheat varieties at all the levels of N fertilizations (Table 2). This is in confirmation with other research workers who have shown that N fertilization increases the protein content of wheat grain.[1,8,10] Maximum protein yield is produced in Rageni-15 (12.89 kg/ha) at 130 kg N/ha fertilization. The protein content in Rageni-15 was increased significantly with 130 kg N/ha over that with 90 or 110 kg N/ha. In case of Chenab-70 and Mexi–Pak-65 protein content increased significantly over that in control, the levels of N having no significant effect on protein control. Grain protein content cannot be fixed at a specified high level by breeding. The plant environment, particularly soil fertility, has strong influence on protein content as well as on grain yield. However, our data do show that high protein variety like Rageni-15 can be expected to be superior in protein level to ordinary varieties in an array of soil fertility situations.

The overall increase in the protein and grain yield of wheat after N fertilization contradicts the concept that the grain yields are negatively correlated with percentage grain protein.[13] Proper management of N fertilization of wheat, i.e. time and rate of application can further improve the protein content of grain.[3]

These data indicate that higher yields of high protein food can be produced economically by the use of inorganic fertilizers. Genotype of the crop is another important factor because it controls the complete translocation of nitrogen from the soil to wheat plant which results in the production of high grain protein.

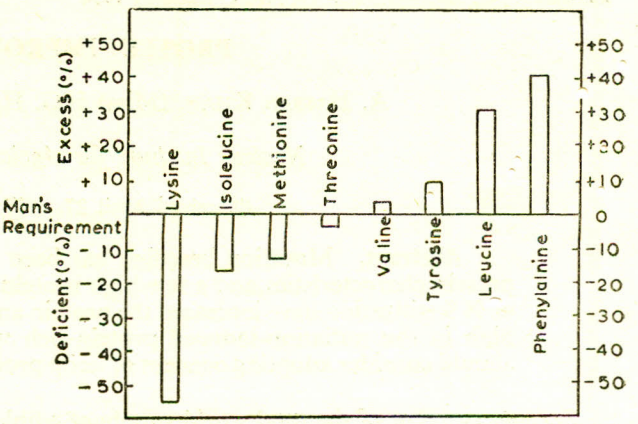

Fig. 1. Deviation of essential amino acids in wheat protein from the requirements of man, FAO 1957 (value for tryptophan not included in the FAO report).

TABLE 2. EFFECT OF NITROGEN APPLICATION ON YIELD OF GRAIN AND GRAIN PROTEIN OF WHEAT.

Rates of varieties (lb/acre)	0	80	100	120
Grain Yield (kg/ha)				
Chenab-70	6229^{c}	7021^{b}	7025^{b}	7322^{a}
Mexi–Pak-65	5823^{b}	6609^{a}	6762^{a}	6864^{a}
Rageni-15	5593^{c}	6352^{b}	6363^{b}	6661^{a}
*Grain Protein** (%)/kg protein/ha				
Chenab-70	11.71^{b}/730	13.88^{a}/975	14.47^{a}/1016	14.56^{a}/1066
Mexi–Pak-65	11.01^{b}/641	13.72^{a}/907	13.92^{a}/941	14.54^{a}/998
Rageni-15	15.43^{c}/863	17.71^{b}/1125	17.49^{b}/1112	19.34^{a}/1289

*Protein measured as Kjeldahl-N × 5.7, oven-dry basis. Figures followed by similar letters a,b,c are insignificant at 95% level of confidence.

With the introduction of high protein dwarf mutant Rageni-15 and the precise application of fertilizer, it is possible to get much higher yield potentials.

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References

1. E. Evald and G. Wenzel, Field Crop Abstr., **20,** 15 (1967).
2. A. Hagberg and K. E. Karlsson, New Approaches to Breeding for Improvement Plant Protein, FAO/IAEA Proc. Sweden, 1968, 17 (1969).
3. A. Hamid and G. Sarwar, Technical Report submitted to IAEA (unpublished), (1971).
4. D.P. Hucklesby, C.M. Brown, S.E. Howell and R.H. Hageman, Agron., J., **63,** 274 (1971).
5. V.A. Johnson, P.J. Mattern, D.A. Whited and J.W. Schmidt, New Approaches to Breeding for Improvement Plant Protein, FAO/IAEA Proc. Sweden, 1968, 29 (1969).
6. V.A. Johnson, P.J. Mattern, J.W. Schmidt, Rice Breeding—Irri. Symp., 409 (1972).

7. A. H. Khan and G. Bari, Pakistan J. Sci., **23,** 325 (1971).
8. J.M. Lawrence, K.M. Day, E. Huey and B. Lee, Cereal Chem., **35,** 169 (1958).
9. E.T. Mertz, L.S. Bates and O.E. Nelson, Science, **145,** 279 (1964).
10. F.C. Stickler, A.W. Pauli and J.A. Johnson, Agron. J., **56,** 592 (1964).
11. M.S. Swaminathan, A. Austin, A.K. Kaul and M.S. Naik, New Approaches to Breeding for Improvement Plant Protein, FAO/IAEA Proc. Sweden, 1968, 71 (1969).
12. S. Tanaka and S. Tamusa, Japan Agron. Res. (Ouat), **3,** 32 (1968).
13. G.L. Terman, R.E. Raming, A.F. Dreier and H.D. Fryer, Agron. J., **61,** 755 (1969).
14. M. Thielebein, New Approaches to Breeding for Improvement Plant Protein, FAO/IAEA Proc. Sweden, 1968, 3 (1969).
15. Report of a Joint FAO/WHO Expert Group, WHO Tech. Rept. Ser. (Rome), 71 (1957).