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Chlorophyll concentration during grain filling period was correlated with grain yield and total plant protein. High grain protein cultivars had longer chlorophyll duration after anthesis than low protein cultivars.

Nitrogen harvest index, total plant protein, and chlorophyll duration after anthesis accounted for 88% and 94% in 1984, and 94% and 90% in 1985, of the total variation among cultivars in grain yield and percent grain protein, respectively. Simultaneous increases in grain yield and percent grain protein could be achieved by selecting for high total plant protein, high N-translocation efficiency, and longer chlorophyll duration after anthesis in wheat.

GRAIN PROTEIN AND GRAIN YIELD AS FUNCTIONS OF DRY MATTER, PLANT PROTEIN, AND CHLOROPHYLL CHARACTERISTICS IN ELITE INTERNATIONAL WINTER WHEATS

by

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A thesis submitted in partial fulfillment
of the requirements for the degree

of

Master of Science

in
Agronomy

MONTANA STATE UNIVERSITY
Bozeman, Montana

May 1986

APPROVAL

of a thesis submitted by

Mohamed Ali Al-Khawlani

This thesis has been read by each member of the author's graduate committee and has been found to be satisfactory regarding content, English usage, format, citations, bibliographic style, and consistency, and is ready for submission to the College of Graduate Studies.

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ACKNOWLEDGMENTS

I wish to express my sincere appreciation to the following:

Dr. G. Allan Taylor, my major advisor, for his advice, friendship, and encouragement through the course of this study.

Dr. Jarvis H. Brown, Dr. C. F. McGuire, and Dr. John M. Martin for sharing their time, their advice, and their enthusiasm while serving on my committee.

The World Bank and Food and Agriculture Organization for financial support that made this program successful.

My wife, Fathia, and son, Nabel, for their patience and love.

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ABSTRACT

Wheat grain protein content is important for product utilization and nutritional qualities. Improving protein content is difficult because of the inverse relationship with grain yield. The relationship of dry matter, plant protein and chlorophyll characteristics to grain protein and grain yield was studied in 66 international winter wheats (Triticum aestivum L.). Field experiments include low, medium, and high N fertility regimes in 1984 and 1985.

The cultivars differed for all measured traits. A significant but low negative correlation between grain yield and percent grain protein was found only in 1984. This suggested simultaneous increases in grain yield and grain protein could be achieved by selection. Total plant protein was positively correlated with biological yield and grain yield, but not correlated with percent grain protein.

Nitrogen harvest index (NHI) decreased with increasing soil N levels. High grain protein cultivars were more efficient than medium and low grain protein cultivars at any soil N level above 110 kg/ha. A positive correlation between nitrogen harvest index (NHI) and percent grain protein was found in 1985.

Chlorophyll concentration during grain filling period was correlated with grain yield and total plant protein. High grain protein cultivars had longer chlorophyll duration after anthesis than low protein cultivars.

Nitrogen harvest index, total plant protein, and chlorophyll duration after anthesis accounted for 88% and 94% in 1984, and 94% and 90% in 1985, of the total variation among cultivars in grain yield and percent grain protein, respectively. Simultaneous increases in grain yield and percent grain protein could be achieved by selecting for high total plant protein, high N-translocation efficiency, and longer chlorophyll duration after anthesis in wheat.

INTRODUCTION

Wheat grains constitute the stable food of a large proportion of the world population. Therefore, wheat protein represents a major source of protein for both humans and animals. Grain protein is an important factor for both baking and nutritional properties of bread wheat. Increasing grain yield and grain protein simultaneously is the ultimate goal for many wheat breeding programs around the world. The simultaneous improvement of grain yield and grain protein is difficult because of their inverse relationship. However, cultivars with high grain yield and high percent grain protein have been obtained (Johnson et al., 1967). This suggests improving both grain yield and protein content by selection is possible. Increased grain yield and percent grain protein may be associated with increased use of nitrogenous fertilizers or increased efficiency in translocating nitrogenous compounds from vegetative parts of the plant to the grain. Higher costs of nitrogenous fertilizers in recent years have drawn attention to the creation of genotypes with an improved efficiency of nitrogen utilization (Austin et al., 1977).

This study was conducted to examine the genetic variation and the relationships of characters related to grain yield and percent grain protein in elite international winter wheats.

LITERATURE REVIEW

Nitrogen and Dry Matter

Numerous investigators have reported significant inverse relationships between grain yield and percent grain protein in spring and winter wheats (Terman et al., 1969; Halloran, 1981; Bhatia, 1975; Loffler and Busch, 1982). The range of correlation coefficients from -0.48 to -0.58 suggests no genetic limitations for improving both grain yield and grain protein percentage in wheat. Stuber et al. (1962) reported high-yielding lines with high grain protein content were found in an F2 population of a high x low protein wheat cross. This suggests simultaneous improvement of grain yield and grain protein is possible.

Wheat plants are known to accumulate most of their nitrogen in the vegetative parts prior to anthesis. The nitrogen is translocated to the developing grain after anthesis. McNeal et al. (1968) found the nitrogen content of vegetative parts of seven spring wheat genotypes (leaves, stems, and head chaff) decreased after anthesis, while grain nitrogen content increased. This is indirect evidence for translocation of nitrogenous compounds from vegetative organs to the developing grains. Translocation of labeled amino acids from culms to grains is direct evidence of mass translocation of nitrogen to the grain (Mikesell et al., 1971). Nitrogen translocation efficiency represents the ability of a genotype to translocate nitrogenous compounds from the vegetative parts to the grains. The efficiency of partitioning of nitrogen between straw and grains is expressed as nitrogen partitioning efficiency (Loffler and Busch, 1982), nitrogen translocation efficiency (Halloran and Lee, 1979), or nitrogen harvest index (Desai and Bhatia, 1978). It is calculated as the ratio of grain nitrogen to total plant nitrogen. Significant differences among wheat cultivars were found for nitrogen translocation efficiency (Halloran and Lee, 1979; Dubois and Fossati, 1981; Loffler and Busch, 1982; Loffler et al., 1985). Halloran (1981) found nitrogen translocation efficiency decreased with increasing availability of soil nitrogen, but the high grain protein cultivars remained highly efficient at high soil nitrogen levels. Loffler and Busch (1982) reported selection for high nitrogen harvest index significantly improved grain yield in the progeny of three crosses of hard spring wheat genotypes. Grain protein content significantly increased in one population, with no reductions in the others. Nitrogen translocation efficiency can be an important criterion for improving grain yield (Dubois and Fossati, 1981), or grain protein content (Halloran and Lee, 1979; Loffler et al., 1985).

Total plant nitrogen can be considered as an indicator of the efficiency of nitrogen uptake (Desai and Bhatia, 1978). The physiological basis for high grain yield and high percent grain protein in wheat appeared to be associated with greater nitrogen uptake. Increased total plant nitrogen at maturity or more efficient and complete translocation of nitrogenous compounds from the vegetative plant parts to the grains are likely. Total plant nitrogen at maturity was positively correlated with grain yield, but not significantly correlated with grain protein content (Desai and Bhatia, 1978; Cox et al., 1985b; Loffler et al., 1985). This suggests selection for high total plant nitrogen could improve grain yield without reducing percent grain protein. Johnson et al. (1967) found nitrogen uptake and nitrogen translocation efficiency functions were separate and independent physiological systems in wheat plants. Both total plant nitrogen and nitrogen translocation efficiency could be used in selecting wheat genotypes for efficient nitrogen utilization (Rao et al., 1977).

Studies of characteristics related to nitrogen utilization provide useful information for parent selections and planned crosses. Edwards et al. (1978) crossed two spring wheat genotypes with complementary values of total reduced plant nitrogen and nitrogen translocation efficiency. The high grain protein progeny had a combination of both high total reduced plant nitrogen and high nitrogen translocation efficiency. Bhatia (1975) concluded that grain protein yield per unit ground area provides a good selection criterion for improving protein productivity in spring wheat cultivars. McNeal et al. (1982) also found selection for high grain protein yield increased grain yield and protein productivity in the progeny of spring wheat crosses.

Chlorophyll Content

The negative association between grain yield and percent grain protein is due in part to the competition between carbohydrate and protein accumulation for assimilates and energy in plants (Bhatia and Robson, 1976). Penning de Vries et al. (1974) concluded 1 g of glucose produced by photosynthesis in plants can be used to produce 0.83 g of carbohydrate or 0.40 g of protein (assuming nitrate to be the N source).

Increasing both grain yield and grain protein content could be achieved by increasing photosynthetic output, by increasing the rate of photosynthesis, and by extending the period of photosynthetic activity (Bhatia and Robson, 1976). Cox et al. (1985b) found 10 to 22% of the total plant nitrogen accumulated after anthesis. They concluded cultivars with longer green tissue duration assimilate more nitrogen than cultivars with short green tissue duration. Mikesell et al. (1971) found similar N content in flag leaves of high and low protein wheat lines at anthesis. They reported removing the flag leaves at anthesis had little effect on grain N content of low protein wheat lines, but greatly decreased grain N content of high protein wheat lines. They concluded the viability and longevity of flag leaves are important for high protein lines. High protein lines continued assimilation of N after anthesis. Neales et al. (1963) examined the effect of leaf removal at anthesis on grain N content. They found leaf removal at anthesis reduced the N uptake and the grain N content at maturity. Spiertz et al. (1971) concluded from 61 to 81% of the variation of grain yield could be statistically predicted by green area duration of flag leaf and peduncle. Rahman (1983) found a positive correlation between net photosynthetic rate and chlorophyll content per unit leaf area in couchgrass.

MATERIALS AND METHODS

Cultivars

Sixty-six cultivars selected from four International Winter Wheat Nurseries (1980, 1981, 1983, 1984) were used in this study (Table 1).

Table 1. Names and origins of cultivars used.

Name	Origin	Name	Origin	Name	Origin
Brule	USA,NE	Sutjeska	Yugoslavia	Vratza	Bulgaria
Grana	Poland	Lavrin-32	Romania	Daws	USA,WA
Bezos.1	USSR	Orov.	Yugoslavia	MV-22-27	Hungary
AW12399	E.Germany	F29-75	Romania	Vala	Czechsl.
Arina	Switzer.	MV-7	Hungary	Stephens	USA,OR
NE7060	USA,NE	Horosh.	Japan	Ogosta	Bulgaria
Odissa-4	USSR	CA8055	China	WWP.4258	Austria
Atlas-66	USA,IN	NS-15-89A	Yugoslavia	Vega	Bulgaria
Lavrin-24	Romania	Katya a-1	Bulgaria	NS2630-1	Yugoslavia
Lethb.32	Canada	Feng-Kang	China	Sudova S.	Bulgaria
Martonv.5	Hungary	Saiente	Italy	Loudog.	Bulgaria
Purdue	USA,IN	F29-76	Romania	Bounty	England
Blueboy	USA,NC	GK-Prot.	Hungary	Pai Yu P.	China
Houser	USA,NY	Clement	Netherlands	NS2699	Yugoslavia
Jana	Poland	Doina	Romania	NE79Y90576	USA,NE
Alcedo	E.Germany	WWP.4394	Austria	NSR-1	Yugoslavia
Aura	Finland	Bastion	Netherlands	Adams	Austria
MV-6	Hungary	TX71A562-6	USA,TX	Inernio	Italy
Martonv.	Hungary	WW330	Australia	Trakia	Bulgaria
NS2699	Yugoslavia	Chokwang	Korea	Marisml.	England
Redwin	USA,MT	Centurk	USA,NE	PL V	USA,KA
Lancota	USA,NE	MT 7811	USA,MT	Norwin	USA,MT

The cultivars were selected to provide a wide range of grain protein content, grain yield, and genetic backgrounds. The cultivars originated from 20 countries. They were grouped by height into three groups to reduce interplot competition with 22 cultivars in each group:

75 - 85 cm -- Short

85 - 95 cm -- Medium

>95 cm -- Tall

Experimental Design

The three height groups were planted separately in a completely randomized block field experiment with six replicates per harvest in 1984 and 1985 near Bozeman. The groups were randomized within each nitrogen level. The cultivars were planted in hill plots with 0.3 m spacing. Thirty seeds of each cultivar were planted in each hill plot. Seeds were dropped in 3-4 cm deep holes dug with hoes. Each replicate was surrounded by a short winter wheat cultivar to avoid shading.

Soil Nitrogen

The soil contained 110 and 22 kg/ha NO3-N in one meter depth in 1984 and 1985 harvest years, respectively. The experiments included three soil nitrogen levels in 1984 and 1985. The first experiment had no nitrogen added in both years (N0). The second experiment had 85 and 110 kg/ha nitrogen added as ammonium nitrate (NH_4NO_3) (34-0-0) in 1984 and 1985, respectively (N1). The third experiment had 170 and 220 kg/ha NH_4NO_3 added in 1984 and 1985, respectively (N2). Fertilizer was applied in mid-May. These soil nitrogen levels in two years totaled six soil nitrogen environments.

Chlorophyll Estimations

The need to estimate chlorophyll concentrations in flag leaves of field grown wheat cultivars required a nondestructive method of analysis. Rahman (1983) suggested visual color ratings provide an estimate of chlorophyll content per unit area. He found a positive correlation between visual color ratings and chlorophyll content per unit area.

A chlorophyll meter similar to one used by Wallihan (1973) to estimate chlorophyll concentrations in leaves of citrus trees was used in this study. Calibration of the chlorophyll meter included samples of leaves taken from 17 field grown wheat cultivars. The leaves were selected to provide a wide range of chlorophyll content on the basis of their color (dark green to yellow). Light absorbance readings were taken by chlorophyll meter and chlorophyll concentrations were estimated by extraction (Arnon, 1949).

Plotting chlorophyll concentration (mg/g fresh weight) against absorbance readings showed 91% of the total variation in chlorophyll concentration could be statistically predicted by absorbance readings taken by chlorophyll meter. The chlorophyll meter provided a good estimate of chlorophyll concentration in wheats.

Measurements

Heading dates and plant height were recorded for each hill plot. The heading date was the number of days from January 1 until 50% of the heads in a plot were fully out of the boot. Plant height was measured in centimeters from the soil surface to tip of the majority of spikes within a plot, excluding awns.

Flag leaf chlorophyll concentration of the 66 cultivars was estimated using the chlorophyll meter during grain filling period; four times in 1984 and five times in 1985. Chlorophyll duration was the number of days from anthesis until 75% of flag leaves in a plot turned yellow.

The following data were recorded for each experimental unit:

1. Grain Protein Percentage (GPP) -- amount of protein expressed as percent of the dry weight of the grain.
2. Grain Yield (GY) -- grain weight (g/plot).
3. Biological Yield (BY) -- total dry weight (g/plot) of aerial biomass including grain.
4. Harvest Index (HI) -- GY/BY.
5. Total Plant Protein (TPP) -- amount of protein (g/plot) in the aerial biomass at maturity including grain.
6. Grain Protein Yield (GPY) -- amount of protein (g/plot) in the grain (GY x GPP).
7. Nitrogen Harvest Index (NHI) -- GPY/TPP.

Percent protein in the grain (Williams, 1979) and the straw were estimated using the Near Infrared Reflectance analyzer (NIR) (Noaman et al., 1984).

Statistical Analyses

Analyses of variance were computed for data from each year and combined over years and soil nitrogen levels. The pooled mean square error was used to test the cultivar mean squares and the cultivar x environment interaction (McIntosh, 1983). All factors were considered fixed. Phenotypic correlations among traits were computed for each year using entry means over soil nitrogen levels. The relationships among traits were analyzed using a multiple regression procedure (Neter and Wasserman, 1974).

The relationships among traits were examined using only cultivars consistent for high and low percent grain protein. A cultivar had high grain protein if it ranked in the top 10 (of 66) in at least four of six soil N environments. A cultivar was considered low grain protein if it ranked in the lowest 10 (of 66) in at least four of six soil N environments. The cultivars were divided by percent grain protein content into three groups:

High grain protein cultivars	--	>15%	grain protein
Medium grain protein cultivars	--	13-15%	grain protein
Low grain protein cultivars	--	<13%	grain protein

RESULTS AND DISCUSSION

Growing conditions differed between the two years of this study. Environmental conditions were ideal for wheat growth in 1984. The soil nitrogen was high (110 kg/ha) and water stress occurred only late in the grain filling period. In the 1985 experiment the soil nitrogen was low (22 kg/ha) with the drought beginning at heading. Although irrigation was applied at heading in 1985, grain yield was about 50% higher in 1984 (Tables 11-16, Appendix).

Cultivar Variation in Grain Protein and Other Traits

The analyses of variance showed highly significant differences for all traits among cultivars within each year and combined across years. Significant cultivar x environment interactions were attributed largely to years rather than soil N levels. Cultivar mean squares were much larger than cultivar x environment interactions (Tables 7-10, Appendix). In 1984, the check treatment (N0) soil nitrogen content was high (110 kg/ha). Nitrogen added to the soil (N1 and N2) caused no significant changes in grain yield, biological yield, and total plant protein. Grain protein percentage significantly increased with increasing soil nitrogen. In 1985, the soil nitrogen content was low (22 kg/ha) in the check (N0) treatments. Significant differences for all measured traits were obtained among nitrogen levels.

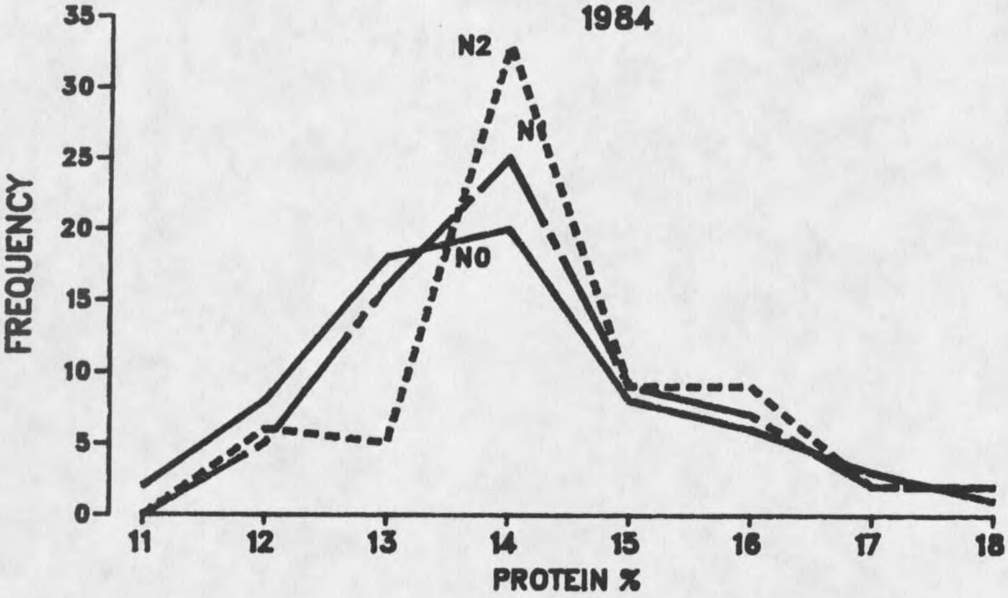


Figure 1. Frequency distribution of percent grain protein for 66 cultivars at 3 soil N levels (1984).

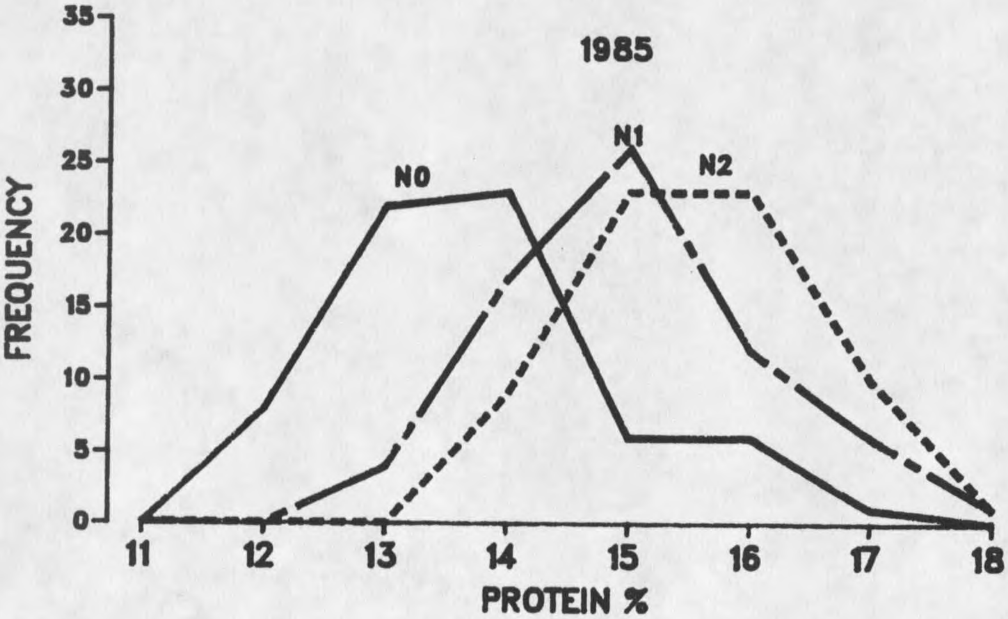


Figure 2. Frequency distribution of percent grain protein for 66 cultivars at 3 soil N levels (1985).

This suggested genetic variability in the elite international winter wheats for grain yield, percent grain protein, and other traits evaluated. Percent grain protein of the 66 cultivars was symetrically distributed around the means in 1984 for all three soil nitrogen regimes (Figure 1). The low protein cultivars increased their protein content with increased soil N, while few changes in grain protein content occurred in the high grain protein cultivars. Although the analysis of variance revealed significant differences among soil N levels, the range of grain protein content was the same at three soil N levels (from 11 to 18%).

Percent grain protein of the 66 cultivars in 1985 was symetrically distributed around individual means for each of the three soil N levels (Figure 2). Increased soil N significantly increased mean percent grain protein. The range and the variation for percent grain protein were similar at the three soil N regimes.

The significant differences for grain protein content in 1984 and 1985 among cultivars and the wide range of grain protein at all soil N levels (Figures 1 and 2) suggested differing genetic capacities for accumulation of protein in the grain. This is supported by the high heritability of percent grain protein in wheat (Stuber et al., 1962; Loffler and Busch, 1982; Cox et al., 1985a). Frequency distributions of grain protein content in the 66 cultivars approaching normality at all soil N levels could be an evidence for quantitative gene control of percent grain protein in these cultivars.

Relationships Among Traits

Dry Matter

Positive correlations were found between biological yield and grain yield in both years (Table 2). Biological yield and percent grain protein were not correlated. Thus selection for high biological yield could improve grain yield without reducing grain protein. Significant increases in grain yield without reduction in percent grain protein were achieved by selection for biological yield (Loffler et al., 1982).

Table 2. Correlation coefficients (r) among trait means of 66 cultivars over 3 soil N levels in two years.

	Biological Yield	Grain Yield	Harvest Index	% Grain Protein	Total Plant Protein
Grain Yield	0.96 ** + 0.97 ** @				
Harvest Index	0.14 ns 0.17 ns	0.42 ** -0.06 ns			
% Grain Protein	-0.04 ns -0.03 ns	-0.27 * -0.09 ns	-0.76 ** -0.20 ns		
Total Plant Protein	0.97 ** 0.97 **	0.91 ** 0.96 **	0.07 ns -0.07 ns	0.13 ns 0.13 ns	
Grain Protein Yield	0.97 ** 0.95 **	0.93 ** 0.97 **	0.15 ns 0.01 ns	0.21 ns 0.16 ns	0.99 ** 0.99 **
N Harvest Index	0.03 ns -0.24 ns	0.19 ns -0.11 ns	0.60 ** 0.52 **	0.22 ns 0.25 *	-0.10 ns -0.17 ns

*, ** = Significant at P<0.05 and <0.01, respectively
ns = Not significant
\+ = 1984
@ = 1985

The correlation between grain yield and grain protein percentage was negative and significant in 1984, and not significant in 1985 (Table 2).

Consistent high or low grain protein across environments is important in wheat breeding. Therefore, the relationships among traits were examined using only cultivars consistent for high and low grain protein, deleting the intermediate groups causing an increase in cultivar x environment interactions. The grain yield and percent grain protein of high and low grain protein cultivars are plotted in Figure 3.

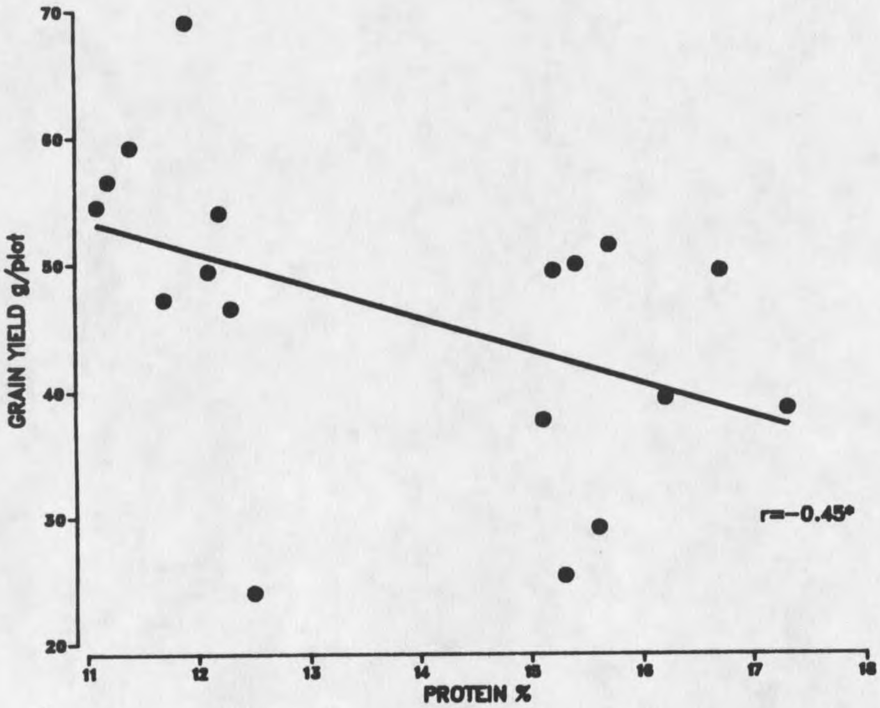


Figure 3. The relationship between grain yield and grain protein (%) for high and low grain protein cultivars over 3 soil N levels (1984).

Despite the negative correlation between grain yield and grain protein, cultivars with high percent grain protein and reasonably high grain yield were identified (Figure 3). The simultaneous improvement of both grain yield and grain protein percentage could be achieved by selection. Similar conclusions were reported by Loffler et al. (1985) and Halloran (1981).

Negative correlations between percent grain protein and harvest index of -0.76** and -0.20 were noted in 1984 and 1985, respectively (Table 2). High grain protein cultivars had low harvest index and vice versa (Figure 4). Selection for high harvest index to improve grain yield could result in reduced grain protein percentage. Similar high negative correlations between harvest index and grain protein were

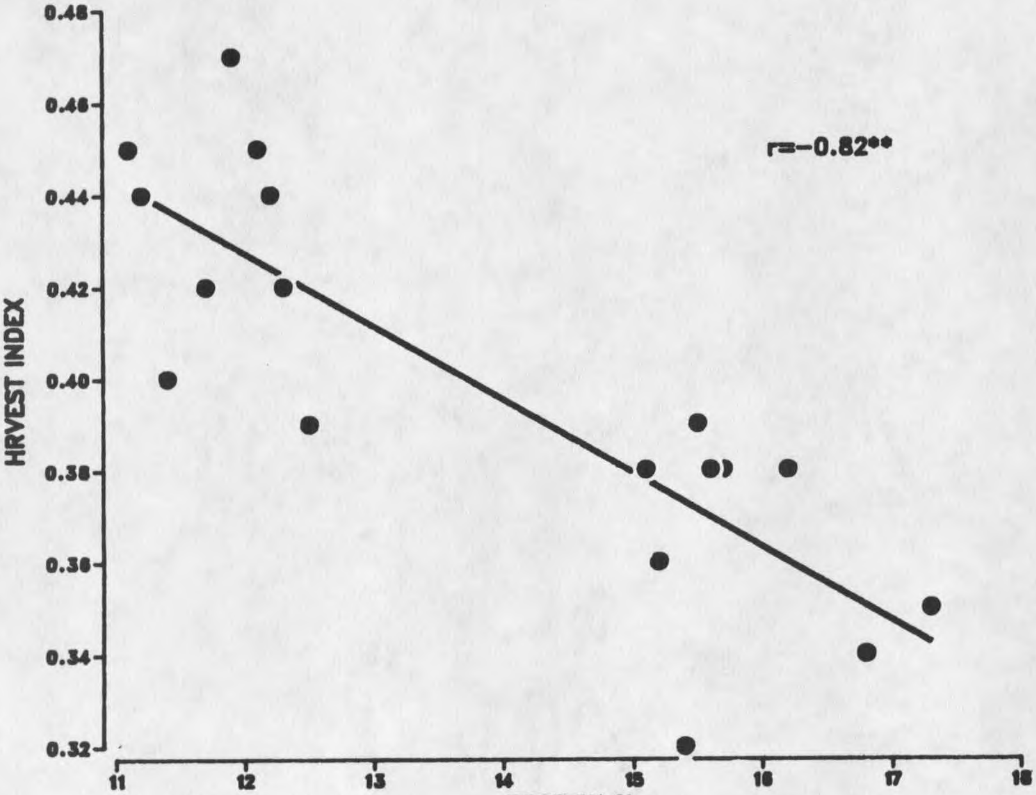


Figure 4. Grain protein (%) and harvest index for high and low grain protein cultivars over 3 soil N levels (1984).

reported by McNeal et al. (1968), Bhatia (1975), and Loffler et al. (1982). They suggested as HI increases, the biomass of vegetative plant parts which serve as an N reservoir decreases. Therefore, less nitrogen is available for translocation to the developing grains.

Plant Protein

Total plant protein accumulated in above-ground parts of the wheat plants was positively correlated with biological yield and grain yield, but not correlated with percent grain protein in 1984 and 1985 (Table 2). Selection for high total plant protein could increase grain yield without decreasing grain protein. Total plant protein was strongly associated with both grain yield and grain protein yield accounting for 83 to 98% of their variation (r = 0.91 and 0.99, respectively; Table 2). This is in agreement with Neales et al. (1963) and Cox et al. (1985b).

Nitrogen harvest index is the genotype's ability to partition nitrogenous compounds between the grain and the vegetative plant parts. The NHI values from 0.60 to 0.83 are within the range reported by Dubois and Fossati (1981) for winter wheat cultivars. This range was higher than that reported by Halloran and Lee (1981), and Loffler et al. (1985), for spring wheat cultivars. Significant positive correlations between NHI and harvest index of 0.60 and 0.53 were found in 1984 and 1985, respectively (Table 2).

This suggests translocation of carbohydrate and nitrogenous compounds are associated. The correlation between NHI and percent grain protein ($r = 0.25$ $P<0.05$) in 1985 was significant but low, and not significant in 1984 (Table 2). Factors other than NHI are important for grain protein percentage. However, the high grain protein cultivars had higher NHI than the low grain protein cultivars (Figure 5). Grain yield

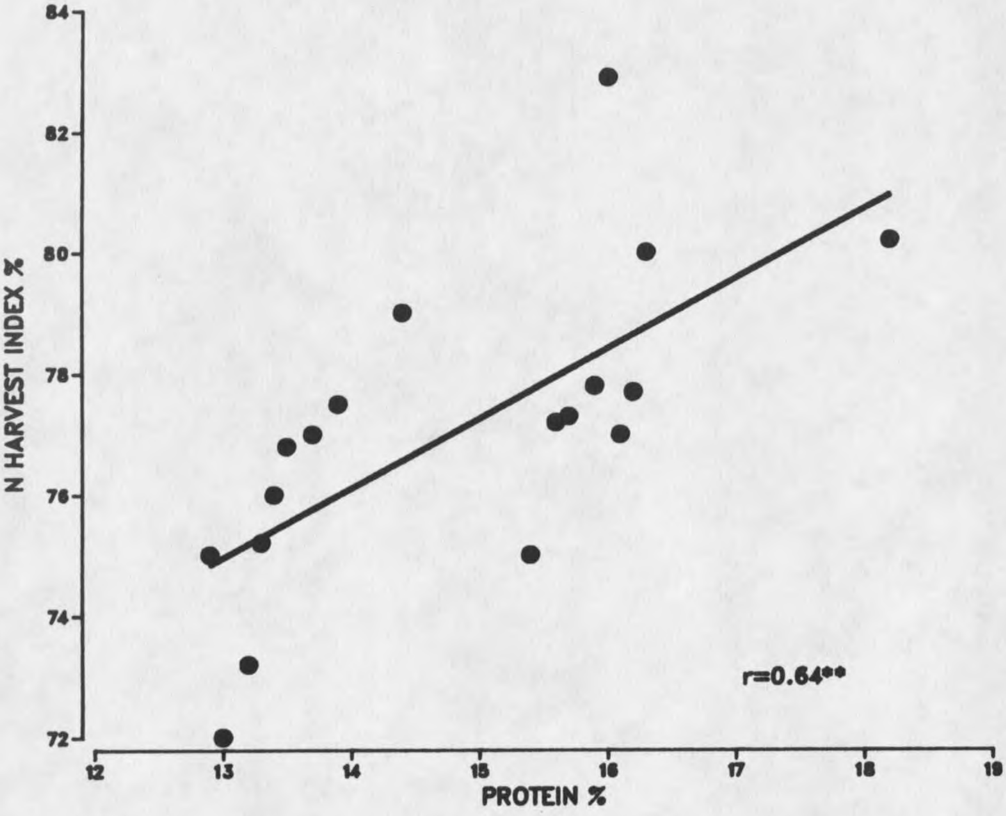
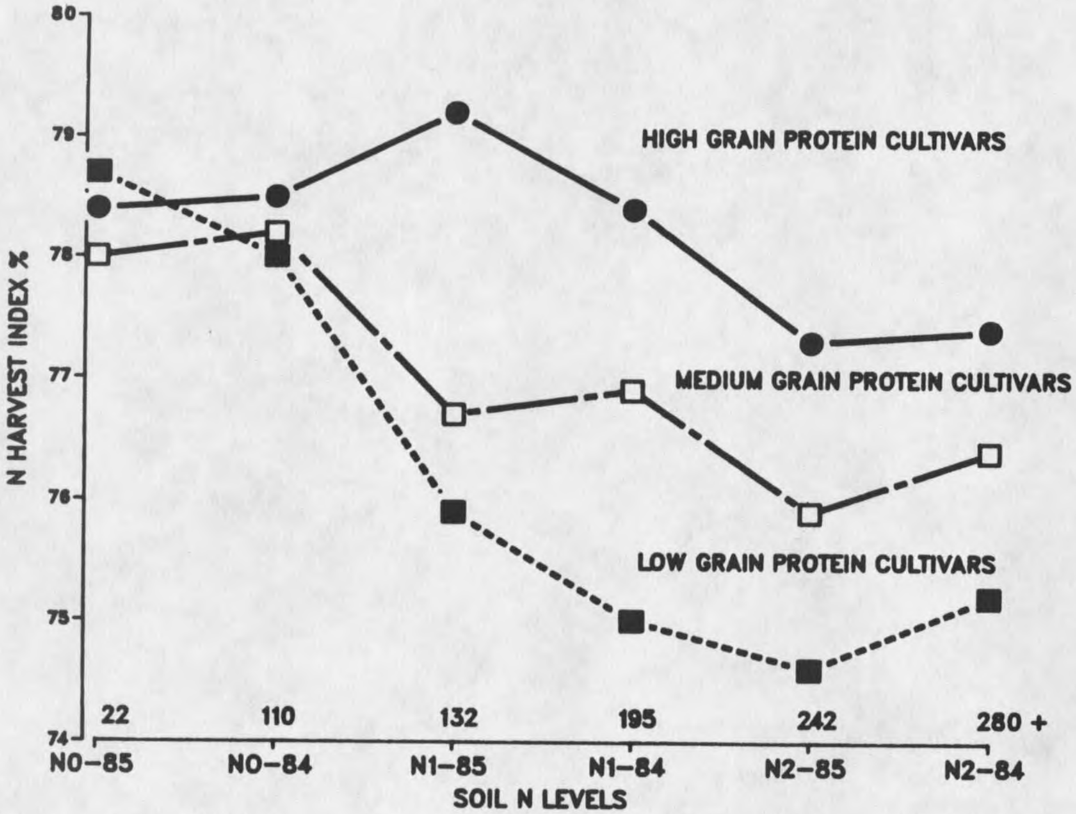


Figure 5. N harvest index and grain protein (%) for high and low grain protein cultivars over 3 soil N levels (1985).

was not correlated with NHI in either year. A greater efficiency for translocation of nitrogen to the grain should result in increased grain protein content at existing grain yield levels. These results are in contrast with those reported in other wheat studies (Cox et al., 1985; Loffler et al., 1982). They found NHI was positively correlated with grain yield but not correlated with grain protein. They concluded selection for high NHI could increase grain yield without reduction in grain protein. However, Halloran and Lee (1979) suggested selection for high NHI could increase grain protein content without changing grain yield.

Nitrogen harvest index (%) for high, medium, and low grain protein cultivars at six soil nitrogen levels is shown in Figure 6.



+ = kg/ha nitrogen (available + added)

Figure 6. N harvest index for 3 grain protein wheat groups at 6 levels of soil N.

Similar nitrogen harvest index occurred in high, medium, and low grain protein cultivars at low soil nitrogen levels. Nitrogen harvest index decreased with increased soil nitrogen. The high grain protein cultivars were more efficient in translocating N to the grain than low grain protein cultivars above 110 kg/ha of soil nitrogen. Separation of high, medium, and low grain protein cultivars on the basis of nitrogen harvest index could be achieved at any soil N levels above 110 kg/ha. High soil nitrogen is required for the phenotypic expression of the genes controlling the nitrogen harvest index. A similar conclusion was reported by Halloran (1981) in a study of nitrogen harvest index of six spring wheat cultivars at different soil N levels. He reported the high grain protein cultivars maintained their high efficiency in translocating N to the grain at high soil N levels. He suggested selection for high nitrogen harvest index should be conducted at high soil nitrogen levels.

Chlorophyll Content

Significant differences among cultivars for flag leaf chlorophyll concentration (mg/gFW), estimated by a chlorophyll meter, occurred at anthesis, early dough, mid-dough, and hard dough in both years and combined over years (Tables 9 and 10, Appendix). Chlorophyll content declined progressively from anthesis to maturity. Although the range of chlorophyll content was very small at anthesis (2.11 to 2.79 mg/g FW), the range increased at the subsequent stages. The chlorophyll concentrations of 2.11 to 2.79 mg/g FW were within the ranges reported by Purves and Barmore (1981), Thimann (1985), and Wallihan (1973).

Significant differences among cultivars for chlorophyll duration were found in both years and combined over years (Table 10, Appendix). The range was 36 to 49 days in 1984 (Tables 11-13, Appendix) and 30 to 46 days in 1985 (Tables 14-15, Appendix). Significant differences among soil nitrogen levels for chlorophyll content occurred in both years.

Chlorophyll content and chlorophyll duration decreased with added nitrogen (Tables 17-19, Appendix). Wide ranges of chlorophyll content in the flag leaves of the 66 cultivars were obtained at early dough, mid-dough, and hard dough (Tables 11-16, Appendix). Significant positive correlations were found between flag leaf chlorophyll concentration at early dough, mid-dough, and hard dough and biological yield, grain yield, total plant protein, and grain protein yield in 1984 and 1985 (Table 3). Significant negative correlations occurred between chlorophyll content and percent grain protein in 1984, but not in 1985 (Table 3). Harvest index was positively correlated with chlorophyll content in 1984, but not in 1985.

Table 3. Correlation coefficients among traits of 66 genotype means over 3 soil N levels in two years.

	BY	GY	HI	GPP	TPP	GPY	NHI
CHLOROPHYLL CONCENTRATION:							
E.dough	0.42**+ 0.48**@	0.50** 0.51**	0.40** 0.15ns	-0.50** -0.16ns	0.35** 0.49**	0.34** 0.48**	-0.06ns -0.27*
M.dough	0.45** 0.47**	0.51** 0.49**	0.32* 0.12ns	-0.49** -0.15ns	0.37** 0.49**	0.35** 0.46**	-0.13ns -0.39**
H.dough	0.40** 0.39**	0.46** 0.40**	0.30* 0.05ns	-0.41** -0.09ns	0.33* 0.42**	0.32* 0.38**	-0.06ns -0.39**
DURATION:	-0.18ns 0.29*	-0.22ns 0.28*	-0.23ns 0.05ns	0.39** 0.07ns	-0.07ns 0.29*	-0.09ns 0.31*	-0.18ns 0.13ns

BY = Biological Yield; GY = Grain Yield; HI = Harvest Index; GPP = Grain Protein Percentage; TPP = Total Plant Protein, GPY = Grain Protein Yield, NHI = Nitrogen Harvest Index

*,** = Significant at P<0.05 and P<0.01, respectively

ns = Not significant

+ = 1984; @ = 1985

Grain yield was more positively associated with chlorophyll concentration in the flag leaf during grain filling than with grain protein content. Most of the carbohydrate available for grain filling is dependent on size and duration of green tissue during the grain filling period. Only 5 to 10% of the carbohydrate formed before anthesis is available for redistribution to the grain (Lupton, 1968). However, nitrogen is mostly accumulated in the vegetative plant parts prior to anthesis and then translocated to the grain. Only 10 to 22% of the total nitrogen is assimilated during the grain filling period (Cox et al., 1985b). The photosynthesis of the flag leaf in wheat makes a major contribution of the carbohydrate to the developing grain (Spiertz et al, 1971). The negative association between chlorophyll content and grain protein is indirect evidence of the positive association of breakdown of chlorophyll and leaf protein. Peoples et al. (1980) reported ribulose bisphosphate carboxylase (RuBPCase) was the major leaf storage protein as well as a major catalyst of CO_2 fixation. RuBPCase accounted for 60% of the total soluble leaf protein in wheat. RuBPCase degradation is associated with degradation of chlorophyll (Hall et al, 1978). Percent grain protein was not correlated with chlorophyll concentration at three grain filling stages in 1985 (Table 3). Drought stress early in the growing season which led to early heading and early maturity may have been a factor. Nitrogen harvest index was negatively correlated with chlorophyll concentration at mid-dough and hard dough in 1985, but not in 1984 (Table 3). This suggests high percent grain protein was not associated with higher concentration of chlorophyll in the flag leaves at any growth stage. Similar conclusions were reported by Cox et al.

(1985a). They found that cultivars with lower percent green tissue (estimated 47 days after anthesis) translocated more N to the grain than cultivars with higher percent green tissue.

Chlorophyll duration estimated from anthesis to the complete yellowing of the flag leaf was significantly correlated with biological yield and grain yield in 1985 (r = 0.29 and 0.28 $P<0.05$, respectively), but not in 1984 (Table 3). Grain protein percentage was positively correlated with chlorophyll duration in 1984 (Table 3), but not in 1985. Although the correlation coefficient between chlorophyll duration and grain protein was low, the high grain protein cultivars had longer chlorophyll duration than the low grain protein cultivars (Figure 7). The chlorophyll duration for high and low grain protein cultivars at three soil N levels for two years is shown in Figure 8. The high grain protein cultivars had longer chlorophyll duration than low grain protein cultivars at any level of soil nitrogen in both 1984 and 1985. This indicates long chlorophyll duration is associated with high grain protein cultivars.

The importance of the longevity of flag leaves and high protein wheat was reported by Neales et al. (1963) and Johnson et al. (1968). They concluded the retention of leaves is important for phenotypic expression of high protein in wheat. Diseases that damage the chlorophyll in the leaves would adversely affect the level of protein. Close association of leaf rust resistance and high protein in wheat was reported by Haunold et al. (1962).

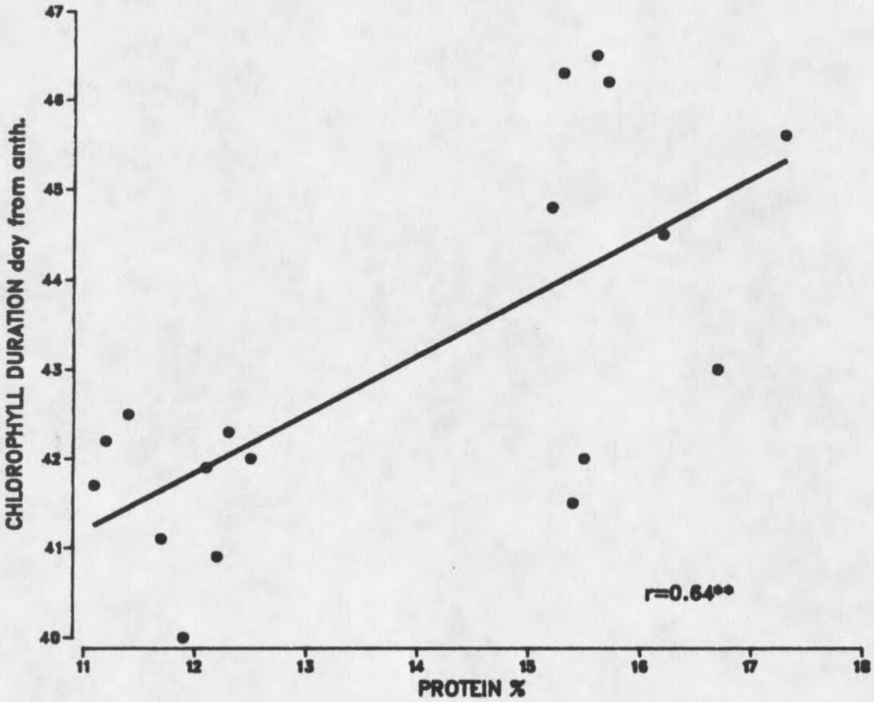


Figure 7. Chlorophyll duration and grain protein (%) for high and low protein cultivars over 3 soil N levels (1984).

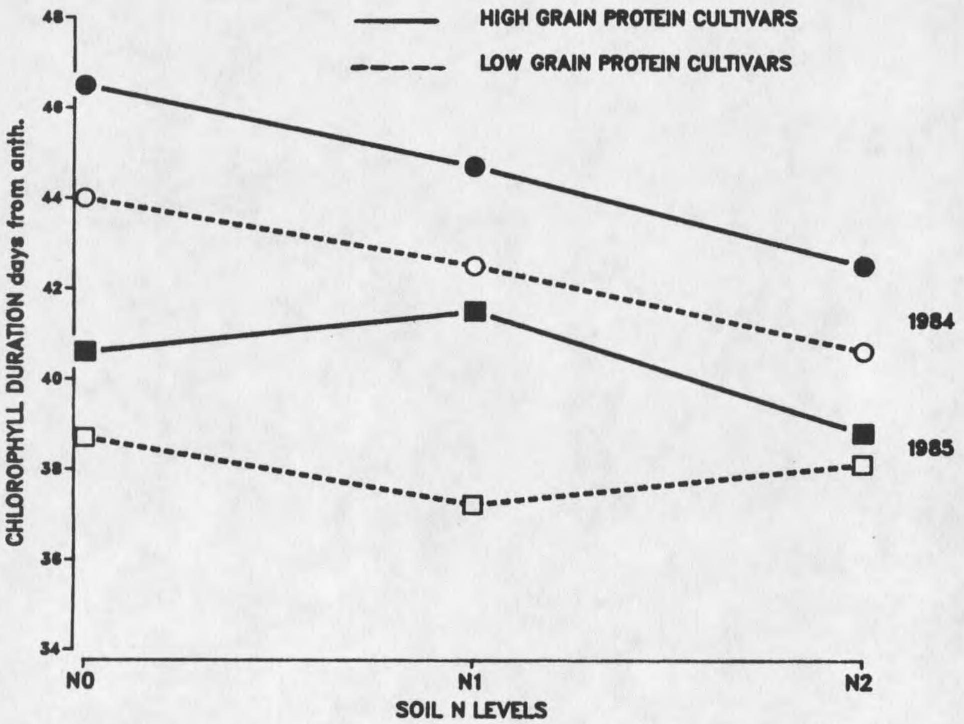


Figure 8. Chlorophyll duration for high and low grain protein groups at 3 soil N levels in two years.

Grain Yield and Grain Protein as Functions of Several Traits

Percent grain protein and grain yield as an expression of several traits was examined using multiple regression. The prediction of GPP including GY, NHI, and TPP as independent variables accounted for 93% and 90% of the total variation of GPP among cultivars in 1984 and 1985, respectively (Tables 4 and 5). Both NHI and TPP had positive regression coefficients, while GY produced a negative regression coefficient. At a given grain yield level, increasing both NHI and TPP could increase GPP.

Table 4. Multiple regression analyses expressing percent grain protein and NHI as functions of several traits (1984).

Dependent Variable	Independent Variables	Regression Coefficients B1	B2	B3+	R^2
Grain Protein (%)	GY,TPP,NHI	-0.28 **	1.56 **	15.40 **	0.83
NHI	GY,TPP,GPP	0.01 **	0.06 **	0.03 **	0.60
NHI	GPY,SPY	0.03 **	-0.09 **	--	0.93

GY = Grain Yield; TPP = Total Plant Protein; NHI = Nitrogen Harvest Index; GPY = Grain Protein Yield; SPY = Straw Protein Yield

+ = B1, B2, B3 = Multiple regression coefficients

** = Significant at P<0.01

The prediction of NHI including GY, TPP, and GPP (Tables 4 and 5) as independent variables accounted for 60% and 73% of the total variation among cultivars in 1984 and 1985, respectively.

Table 5. Multiple regression analyses expressing percent grain protein and NHI as functions of several traits (1985).

Dependent Variable	Independent Variables	Regression Coefficients B1	B2	B3+	R^2
Grain Protein (%)	GY,TPP,NHI	-0.81 **	4.40 **	18.20 **	0.90
NHI	GY,TPP,GPP	0.03 **	-0.18 **	0.04 **	0.73
NHI	GPY,SPY	0.06 **	-0.21 **	--	0.91

GY = Grain Yield; TPP = Total Plant Protein; NHI = Nitrogen Harvest Index; GPY = Grain Protein Yield; SPY = Straw Protein Yield

+ = B1, B2, B3 = Multiple regression coefficients

** = Significant at P<0.01

Grain yield and percent grain protein had positive regression coefficients, while total plant protein had a negative regression coefficient.

The prediction of NHI using only grain protein yield (GPY) and straw protein yield (SPY) accounted for 93% and 91% of the total variation in 1984 and 1985, respectively (Tables 4 and 5). Grain protein yield had a positive regression coefficient, while amount of protein remaining in the straw had a negative regression coefficient. This suggests that high efficiency of translocating N from the straw to the grain is associated with high grain yield and high percent grain protein.

Multiple regression equations expressing percent grain protein and grain yield as functions of several traits are shown in Table 6. The prediction of percent grain protein included total plant protein, NHI, chlorophyll duration, and grain yield. This combination of independent variables accounted for 94% and 90% of the total variation in percent grain protein in 1984 and 1985, respectively. The grain yield prediction equation including total plant protein, NHI, and chlorophyll duration accounted for 88% and 94% of the total variation in grain yield in 1984 and 1985, respectively. Total plant protein, NHI, and chlorophyll duration were the most important variables in all prediction equations. None of the other traits measured met the required significance levels for entry into the equations.

Table 6. Multiple regression analyses expressing percent grain protein and grain yield as functions of several traits for 66 cultivars.

	Dependent Variable	Independent Variables+	R^2
1984	Grain Protein (%)	TPP, NHI, ChL.D., GY	0.94
	Grain Yield	TPP, NHI, ChL.D.	0.88
1985	Grain Protein (%)	TPP, NHI, ChL.D., GY	0.90
	Grain Yield	TPP, NHI, ChL.D.	0.94

+TPP = Total Plant Protein; NHI = Nitrogen Harvest Index; ChL.D. = Chlorophyll Duration; GY = Grain Yield

Selection for high NHI, high total plant protein, and long chlorophyll duration could increase both grain yield and grain protein percentage. Johnson et al. (1967) reported nitrogen uptake and nitrogen partitioning function as separate and independent physiological systems in the wheat plant. Both NHI and total plant protein independently contributed to percent grain protein in the current study. Therefore, inclusion of both characters in wheat genotypes through breeding should result in higher grain yield and higher grain protein content. Rao et al. (1977) suggested that selection for efficient nitrogen utilization must include two or more factors simultaneously, such as high N uptake and high efficiency of translocating vegetative nitrogen to the grain.

SUMMARY

The low negative correlation between grain yield and percent grain protein in this study indicates simultaneous increases in grain yield and protein content could be achieved by selection. Simple correlation analyses suggested that increases in total plant protein may increase grain yield without reducing percent grain protein. High grain protein content was associated with high NHI. Selection for high NHI at high soil nitrogen could increase percent grain protein without reducing grain yield.

Although significant correlations were found between grain yield, biological yield, total plant protein and chlorophyll concentration in the flag leaves at three grain filling stages, the values were too low for predictive purposes. Chlorophyll duration was associated with high grain protein content. Separation of high and low grain protein cultivars was possible by chlorophyll duration.

The multiple regression analyses suggested total plant protein, NHI, and chlorophyll duration contributed to both high grain yield and high percent grain protein. Combining high NHI, high total plant protein, and long chlorophyll duration in hybridization could provide wheat cultivars with high yield and high protein content.

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LITERATURE CITED

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APPENDIX

Table 7. Mean squares of grain yield and percent grain protein of three height groups of cultivars combined over years and soil nitrogen levels.

		Grain Yield			Percent Grain Protein		
Source	d.f.	Tall	Medium	Short	Tall	Medium	Short
Year(Y)	1	193300 **	187300 **	178200 **	131.6 **	48.61 **	140.5 **
Nitrogen(N)	2	284.9 ns	294.3 ns	56.2 ns	63.4 **	68.8 **	115.8 **
Y x N	2	134.3 ns	41.0 ns	130.9 ns	37.4 **	21.6 **	41.7 **
BLK/N/Y	30	118.7	142.3	111.0	0.564	0.53	0.52
Cultivar(C)	21	1124 **	2694 **	2202 **	54.0 **	36.8 **	29.6 **
Y x C	21	390.6 **	1294 **	780.6 **	6.8 **	7.4 **	6.4 **
N x C	42	72.4 ns	99.9 ns	129.8 ns	0.89 **	1.76 **	1.9 **
Y x N x C	42	106.3 ns	89.1 ns	133.0 ns	0.99 **	1.3 **	1.21 **
Pooled Error	630	96.06	88.21	88.26	0.369	0.273	0.238

** = Significant at P<0.01
ns = Not significant

Table 8. Mean squares of biological yield and total plant protein of three height groups of cultivars combined over years and soil nitrogen levels.

		Biological Yield			Percent Grain Protein		
Source	d.f.	Tall	Medium	Short	Tall	Medium	Short
Year(Y)	1	1373000 **	114000 **	998800 **	5936 **	5333 **	4505 **
Nitrogen(N)	2	233.4 ns	4343 **	488.1 ns	2.46 ns	30.3 **	11.9 *
Y x N	2	4203 **	1335 ns	846.9 ns	17.9 *	2.7 ns	6.7 ns
BLK/N/Y	30	696.4	657.5	859.8	5.0	3.9	3.5
Cultivar(C)	21	4809 **	12910 **	110600 **	36.8 **	68.1 **	66.4 **
Y x C	21	1879 **	5542 **	3851 **	17.4 **	26.8 **	25.2 **
N x C	42	392.1 ns	568.4 ns	544.6 ns	2.4 ns	3.1 ns	3.9 **
Y x N x C	42	441.8 ns	505.3 ns	552.1 ns	3.2 ns	3.0 ns	4.3 **
Pooled Error	630	476.7	428.0	416.8	2.65	2.47	2.38

*, ** = Significant at $P<0.05$ and $P<0.01$, respectively.
ns = Not significant

Table 9. Mean squares of chlorophyll concentration (mg/g FW) at anthesis and early dough of three height groups of cultivars combined over years and soil nitrogen levels.

		Chlorophyll Concentration					
		Anthesis			E.Dough		
Source	d.f.	Tall	Medium	Short	Tall	Medium	Short
Year (Y)	1	11.93 **	17.79 **	18.42 **	1.42 ns	0.052 ns	0.103 ns
Nitrogen(N)	2	0.582 **	0.18 **	0.475 **	17.9 **	25.4 **	37.7 **
Y x N	2	0.635 **	0.439 **	0.301 **	6.84 **	9.29 **	6.16 **
BLK/N/Y	30	0.0534	0.029	0.031	0.27	0.497	0.337
Cultivars(C)	21	0.2273 **	0.274 **	0.347 **	2.25 **	2.85 **	3.75 **
Y x C	21	0.019 ns	0.068 **	0.90 **	0.792 **	0.727 **	1.13 **
N x C	42	0.0196 ns	0.0185 ns	0.0265 ns	0.318 **	0.326 **	0.364 **
Y x N x C	42	0.0221 ns	0.0313 ns	0.0236 ns	0.137 **	0.238 **	0.274 **
Pooled Error	630	0.0224	0.0239	0.0233	0.0626	0.10	0.07

** = Significant at P<0.01
ns = Not significant

Table 10. Mean squares of chlorophyll concentration at hard dough and chlorophyll duration of three height groups of cultivars combined over years and soil nitrogen levels.

Source	d.f.	Chlorophyll (mg/g FW)			Chlorophyll Duration		
		Tall	Medium	Short	Tall	Medium	Short
Year (Y)	1	4.21 **	3.57 **	1.68 *	2190 **	4558 **	5208 **
Nitrogen(N)	2	2.78 **	4.60 **	1.74 **	554.7 **	243.5 **	548.5 **
Y x N	2	8.88 **	8.81 **	9.34 **	112.3 **	241.9 **	113.6 **
BLK/N/Y	30	0.258	0.229	0.225	4.58	6.45	4.66
Cultivars(C)	21	4.01 **	4.86 **	6.77 **	145.2 **	64.2 **	97.6 **
Y x C	21	0.877 **	1.13 **	0.814 **	40.1 **	81.2 **	75.2 **
N x C	42	0.183 **	0.273 **	0.218 **	22.6 **	26.5 **	37.2 **
Y x N x C	42	0.311 **	0.265 **	0.384 **	24.7 **	29.7 **	32.5 **
Pooled Error	630	0.0843	0.0739	0.0714	4.61	4.69	4.7

*, ** = Significant at P<0.05 and P<0.01, respectively.

Table 11. Eight trait means of 22 winter wheat cultivars (tall group) grown at 3 soil N levels in 1984.

					Chlorophyll Concent.			
Cultivars	GY	GPP	TPP	NHI	E. Dough	M. Dough	H. Dough	ChL.D.
Brule (USA,NE)	50.2	12.7	8.1	0.74	1.92	1.38	0.145	44.5
Grana (Poland)	59.2	12.5	9.7	0.76	2.30	1.30	0.561	41.8
Bezost.1 (USSR)	55.3	14.0	9.9	0.78	1.91	0.77	0.178	41.8
AW12399 (E.Germ.)	62.4	14.4	11.6	0.77	2.24	1.18	0.324	42.2
Arena (Switzerl.)	47.5	14.9	9.0	0.78	2.22	1.10	0.299	40.7
NE7060 (USA,NE)	49.4	16.7	10.4	0.79	1.48	0.32	0.035	42.3
Odissa-4 (USSR)	49.9	15.4	10.0	0.77	1.94	0.81	0.068	41.6
Atlas-66 (USA,IN)	25.4	15.3	5.1	0.77	1.72	0.69	0.067	40.2
Lavrin24 (Romania)	60.0	14.1	10.4	0.81	2.18	1.10	0.218	40.6
Lethb.32 (Canada)	38.2	14.9	7.3	0.78	0.74	0.16	0.051	44.4
Martonv.5 (Hungary)	50.7	14.5	9.8	0.75	2.00	0.85	0.110	42.3
Purdue (USA,IN)	37.8	13.5	6.9	0.74	1.82	0.65	0.011	43.2
Blueboy (USA,NC)	56.6	12.8	9.1	0.80	1.92	0.83	0.262	42.3
Houser (USA,NY)	54.5	11.1	7.8	0.78	2.01	0.89	0.257	41.7
Jana (Poland)	47.2	11.7	7.3	0.76	2.30	1.34	0.710	41.1
Alcedo (E.Germ.)	48.2	12.1	7.9	0.77	2.15	1.12	0.388	40.0
Aura (Finland)	41.5	13.0	7.1	0.76	1.54	0.50	0.305	37.4
MV-6 (Hungary)	49.0	13.7	9.0	0.74	2.10	1.00	0.280	41.3
Martonv. (Hungary)	59.0	13.8	10.8	0.75	1.95	1.04	0.196	42.8
NS2699 (Yugoslav.)	49.2	13.5	8.6	0.77	1.41	0.46	0.255	42.8
Redwin (USA,MT)	49.4	15.2	10.3	0.73	2.17	1.26	0.419	44.8
Lancota (USA,NE)	51.4	15.7	10.7	0.75	1.95	0.96	0.253	46.2
Means	49.6	13.9	9.0	0.77	1.90	0.89	0.245	42.1
C.V.	19.0	3.2	17.6	5.0	11.1	7.5	18.7	4.1
LSD.05	5.2	0.2	0.9	0.02	0.12	0.12	0.076	1.0
LSD.01	7.3	0.3	1.2	0.30	0.16	0.17	0.107	1.3

GY = Grain Yield; GPP = Grain Protein Percentage; TPP = Total Plant Protein; NHI = Nitrogen Harvest Index; ChL.D. = Chlorophyll Duration

Table 12. Eight trait means of 22 winter wheat cultivars (medium height group) grown at 3 soil N levels in 1984.

Cultivars	GY	GPP	TPP	NHI	Chlorophyll Concent. E. Dough	Chlorophyll Concent. M. Dough	Chlorophyll Concent. H. Dough	ChL.D.
Sutjeska(Yugos.)	51.7	13.8	9.4	0.76	2.07	1.02	0.292	41.5
Lavrin-32(Romania)	45.3	13.3	7.5	0.80	1.90	0.69	0.103	43.5
Orov.(Yugoslavia)	21.7	14.2	4.1	0.76	1.55	0.34	0.058	43.6
F29-75(Romania)	64.2	13.1	10.9	0.76	2.15	1.12	0.236	42.2
MV-7(Romania)	49.5	13.1	8.3	0.78	1.96	0.75	0.045	42.9
Horosh(Japan)	62.0	13.4	10.6	0.79	2.15	1.04	0.275	43.3
CA8055(China)	47.3	13.8	8.4	0.78	1.82	0.61	0.045	44.8
NS-15-89A(Yugosl.)	39.1	14.0	7.2	0.75	1.95	0.83	0.099	43.0
Katia-1(Bulgaria)	41.1	13.6	7.3	0.76	1.88	0.79	0.134	45.3
Feng-Kang(China)	33.1	17.2	7.6	0.74	1.15	0.26	0.058	46.8
Saiente(Italy)	38.7	13.1	6.5	0.78	1.63	0.49	0.045	42.2
F29-76(Romania)	51.8	12.9	8.5	0.78	2.20	1.00	0.281	43.1
GK-Prot.(Hungary)	39.4	16.2	8.0	0.80	1.53	0.32	0.080	44.5
Clement(Netherld.)	55.5	11.2	8.1	0.77	2.35	1.43	1.010	42.4
Doina(Romania)	45.7	13.0	7.5	0.79	1.82	0.75	0.171	45.7
WWP.4394(Austria)	41.3	14.7	7.7	0.79	1.89	0.79	0.265	45.7
Bastion(Netherld.)	23.4	12.5	3.9	0.75	1.99	1.00	0.316	44.0
TX71A562-6(USA,TX)	69.1	11.9	10.2	0.80	1.74	0.55	0.149	43.9
WW330(Australia)	27.0	13.1	4.9	0.73	1.93	0.77	0.149	44.6
Chokwang(Korea)	29.0	15.6	6.1	0.74	0.88	0.20	0.087	46.3
Centurk(USA,NE)	66.9	13.8	12.0	0.79	1.97	0.90	0.380	42.2
MT 7811(USA,MT)	66.0	13.3	11.1	0.79	2.28	1.26	0.477	44.4
Means	45.9	13.7	8.0	0.77	1.85	0.77	0.216	43.9
C.V.	20.0	2.9	19.1	5.6	11.6	8.8	16.7	4.0
LSD.05	5.0	0.2	0.8	0.02	0.12	0.11	0.062	1.0
LSD.01	7.1	0.3	1.2	0.03	0.17	0.15	0.088	1.3

GY = Grain Yield; GPP = Grain Protein Percentage; TPP = Total Plant Protein; NHI = Nitrogen Harvest Index; ChL.D. = Chlorophyll Duration

Table 13. Eight trait means of 22 winter wheat cultivars (short group) grown at 3 soil N levels in 1984.

Cultivars	GY	GPP	TPP	NHI	Chlorophyll Concent. E. Dough	M. Dough	H. Dough	ChL.D.
Vratza (Bulgaria)	33.6	13.4	5.2	0.82	1.34	0.22	0.011	42.7
Daws (USA,WA)	59.2	11.4	8.4	0.80	2.25	1.16	0.329	42.5
MV-22-27 (Hungary)	50.6	13.2	8.8	0.76	1.78	0.65	0.156	42.1
Vala (Czechslov.)	53.1	12.5	8.4	0.79	2.08	0.94	0.251	42.4
Stephens (USA,OR)	48.9	12.1	7.2	0.82	2.14	1.02	0.238	40.9
Ogosta (Bulgaria)	55.1	13.2	9.2	0.79	2.14	1.04	0.293	44.6
WWP.4258 (Austria)	49.4	12.0	7.3	0.81	1.97	0.87	0.144	41.9
Vega (Bulgaria)	34.3	14.1	6.1	0.80	1.31	0.20	0.050	43.9
NS2630-1 (Yugosl.)	58.6	13.7	10.2	0.79	1.95	0.73	0.120	42.8
Sudova S. (Bulgar.)	50.2	12.7	8.2	0.79	2.09	1.00	0.192	43.6
Loudog. (Bulgaria)	71.9	14.0	13.6	0.75	2.24	1.10	0.466	44.3
Bounty (England)	43.7	12.5	6.8	0.80	2.24	1.20	0.363	40.3
Pai Yu P. (China)	37.7	15.2	7.3	0.79	1.05	0.20	0.033	46.3
NS2699 (Yugoslav.)	51.7	13.4	8.9	0.78	1.57	0.56	0.186	42.6
NE79Y90576 (USA,NE)	40.0	13.2	6.4	0.83	0.68	0.13	0.030	43.7
NSR-1 (Yugoslavia)	31.0	14.2	5.7	0.77	1.71	0.55	0.011	44.8
Adams (Austria)	48.2	13.0	7.6	0.82	2.06	1.20	0.278	42.9
Inernio (Italy)	18.3	12.9	3.1	0.76	1.81	0.71	0.054	44.0
Trakia (Bulgaria)	44.9	13.3	7.4	0.80	1.92	0.77	0.084	44.0
Marisml. (England)	35.5	12.8	5.7	0.79	2.46	1.36	0.369	41.5
PL V (USA,KA)	38.6	17.3	8.5	0.79	1.20	0.36	0.226	45.6
MT 7877 (USA,MT)	61.1	13.1	9.9	0.81	2.19	1.16	0.443	42.2
Means	41.2	13.3	7.7	0.79	1.83	0.77	0.196	43.2
C.V.	22.2	3.0	19.5	4.0	12.4	9.3	16.4	4.1
LSD.05	5.0	0.2	0.8	0.02	0.12	0.11	0.056	1.0
LSD.01	7.1	0.3	1.2	0.03	0.18	0.15	0.079	1.4

GY = Grain Yield; GPP = Grain Protein Percentage; TPP = Total Plant Protein; NHI = Nitrogen Harvest Index; ChL.D. = Chlorophyll Duration

Table 14. Eight trait means of 22 winter wheat cultivars (tall group) grown at 3 soil N levels in 1985.

Cultivars	GY	GPP	TPP	NHI	Chlorophyll Concent. E. Dough	M. Dough	H. Dough	ChL.D.
Brule (USA,NE)	17.5	13.3	3.0	0.77	1.99	1.32	0.677	40.1
Grana (Poland)	25.8	13.7	4.5	0.78	2.09	1.61	1.050	39.3
Bezost.1 (USSR)	20.2	14.1	3.7	0.78	1.90	0.96	0.463	37.6
AW12399 (E.Germ.)	22.7	15.6	4.5	0.77	2.04	1.58	1.043	37.6
Arena (Switzerl.)	16.3	15.2	3.3	0.75	2.17	1.73	0.910	37.1
NE7060 (USA,NE)	18.4	16.1	3.7	0.80	1.89	1.15	0.663	43.2
Odissa-4 (USSR)	17.6	15.6	3.6	0.77	2.04	1.47	0.753	39.3
Atlas-66 (USA,IN)	13.8	18.3	3.2	0.80	1.95	1.48	1.050	38.9
Lavrin24 (Romania)	19.3	14.1	3.4	0.81	2.03	1.48	0.803	40.7
Lethb.32 (Canada)	11.8	14.7	2.2	0.78	1.58	0.77	0.190	38.0
Martonv.5 (Hungary)	19.2	14.8	3.6	0.79	2.02	1.41	0.583	37.6
Purdue (USA,IN)	13.2	14.2	2.5	0.76	1.76	0.92	0.523	42.5
Blueboy (USA,NC)	15.7	14.4	2.9	0.78	1.98	1.23	0.817	40.7
Houser (USA,NY)	22.6	13.5	4.0	0.78	1.98	1.43	0.767	40.9
Jana (Poland)	20.8	13.0	3.7	0.72	2.11	1.84	1.383	34.0
Alcedo (E.Germ.)	19.0	13.7	3.6	0.74	2.09	1.70	1.223	35.4
Aura (Finland)	16.0	14.9	3.2	0.75	2.10	1.74	1.233	32.6
MV-6 (Hungary)	23.2	14.5	4.2	0.81	1.97	1.36	0.683	36.8
Martonv. (Hungary)	20.6	13.8	3.8	0.76	1.99	1.37	0.713	40.7
NS2699 (Yugoslav.)	13.6	14.1	2.4	0.78	1.93	1.24	0.650	38.7
Redwin (USA,MT)	21.0	16.1	4.4	0.77	2.13	1.69	1.280	40.4
Lancota (USA,NE)	16.3	15.6	3.4	0.75	2.00	1.30	0.743	40.8
Means	18.4	14.7	3.5	0.77	1.99	1.40	0.827	38.8
C.V.	14.4	2.8	10.6	5.4	6.7	12.8	21.0	3.3
LSD.05	1.5	0.2	0.2	0.02	0.07	0.10	0.096	0.7
LDS.01	2.1	0.3	0.3	0.03	0.10	0.14	0.135	1.0

GY = Grain Yield; GPP = Grain Protein Percentage; TPP = Total Plant Protein; NHI = Nitrogen Harvest Index; ChL.D. = Chlorophyll Duration

Table 15. Eight trait means of 22 winter wheat cultivars (medium height group) grown at 3 soil N levels in 1985.

Cultivars	GY	GPP	TPP	NHI	Chlorophyll Concent. E. Dough	M. Dough	H. Dough	ChL.D.
Sutjeska (Yugos.)	13.2	13.3	2.6	0.75	2.13	1.46	0.847	39.9
Lavrin-32 (Romania)	23.0	13.5	4.0	0.78	1.89	1.42	0.860	42.3
Orov. (Yugoslavia)	13.2	14.0	2.3	0.81	2.01	1.29	0.897	38.7
F29-75 (Romania)	18.5	14.5	3.5	0.76	2.08	1.29	0.697	41.5
MV-7 (Romania)	14.2	13.7	2.5	0.77	1.95	1.02	0.963	34.8
Horosh (Japan)	21.4	13.5	3.9	0.75	1.89	1.25	0.650	39.6
CA8055 (China)	11.4	14.0	2.0	0.78	1.31	0.67	0.307	39.8
NS-15-89A (Yugosl.)	18.5	13.9	3.3	0.78	2.02	1.53	0.990	41.6
Katia-1 (Bulgaria)	10.9	13.4	1.9	0.76	2.04	1.48	0.747	41.9
Feng-Kang (China)	9.9	15.0	1.9	0.78	1.19	0.38	0.050	33.2
Saiente (Italy)	13.1	13.7	2.4	0.76	1.81	1.19	0.473	39.0
F29-76 (Romania)	15.5	13.8	2.9	0.74	2.05	1.44	0.877	38.7
GK-Prot. (Hungary)	16.0	15.8	3.1	0.83	1.98	1.22	0.533	38.3
Clement (Netherld.)	16.8	12.9	2.9	0.76	2.08	1.69	1.320	37.1
Doina (Romania)	13.6	14.1	2.5	0.77	1.95	1.33	0.613	41.5
WWP.4394 (Austria)	14.8	15.7	2.9	0.82	1.95	1.48	0.700	41.3
Bastion (Netherld.)	9.5	13.2	1.7	0.75	1.82	1.64	1.260	34.5
TX71A562-6 (USA,TX)	17.8	13.8	3.2	0.77	1.75	1.03	0.373	38.3
WW330 (Australia)	10.5	13.6	1.9	0.78	1.80	0.78	0.357	40.2
Chokwang (Korea)	12.1	15.7	2.4	0.78	1.51	0.51	0.187	38.4
Centurk (USA,NE)	18.3	13.7	3.4	0.74	1.84	1.03	0.580	40.0
MT 7811 (USA,MT)	22.0	15.1	4.6	0.72	2.03	1.50	1.070	40.0
Means	15.2	14.2	2.8	0.77	1.87	1.21	0.693	39.1
C.V.	12.8	2.4	13.3	4.4	12.4	14.0	22.4	3.3
LSD.05	1.1	0.2	0.2	0.02	0.13	0.09	0.085	0.7
LSD.01	1.5	0.3	0.3	0.03	0.18	0.13	0.120	1.0

GY = Grain Yield; GPP = Grain Protein Percentage; TPP = Total Plant Protein; NHI = Nitrogen Harvest Index; ChL.D. = Chlorophyll Duration

Table 16. Eight trait means of 22 winter wheat cultivars (short group) grown at 3 soil N levels in 1985.

Cultivars	GY	GPP	TPP	NHI	Chlorophyll Concent. E. Dough	M. Dough	H. Dough	ChL.D.
Vratza (Bulgaria)	15.5	14.0	2.8	0.79	1.91	1.07	0.327	39.1
Daws (USA,WA)	21.0	13.3	3.6	0.77	1.99	1.42	0.820	39.3
MV-22-27 (Hungary)	19.4	13.4	3.5	0.74	1.93	1.05	0.547	37.6
Vala (Czechslov.)	20.0	13.2	3.6	0.73	1.82	1.52	0.787	38.7
Stephens (USA,OR)	20.5	13.7	3.5	0.79	2.05	1.42	0.640	38.2
Ogosta (Bulgaria)	13.9	14.4	2.6	0.80	1.96	1.35	0.883	37.9
WWP.4258 (Austria)	12.0	14.2	2.1	0.80	1.61	0.88	0.193	36.2
Vega (Bulgaria)	16.6	14.5	3.1	0.78	1.88	0.97	0.357	37.7
NS2630-1 (Yugosl.)	13.5	13.7	2.4	0.80	1.90	1.23	0.527	41.3
Sudova S. (Bulgar.)	21.9	14.3	4.2	0.75	1.79	1.39	0.890	41.7
Loudog. (Bulgaria)	13.9	13.9	2.5	0.77	1.95	1.63	1.377	33.6
Bounty (England)	11.1	15.8	2.3	0.78	1.22	0.37	0.083	35.2
Pai Yu P. (China)	16.6	13.6	2.9	0.79	1.99	1.32	0.680	38.6
NS2699 (Yugoslav.)	11.0	14.6	2.1	0.78	1.53	0.78	0.170	37.9
NE79Y90576 (USA,NE)	12.6	14.0	2.3	0.77	1.81	1.22	0.400	38.8
Adams (Austria)	25.3	14.7	4.6	0.81	2.00	1.62	1.117	39.8
Inernio (Italy)	12.1	13.6	2.1	0.81	1.96	1.06	0.297	31.5
Trakia (Bulgaria)	12.7	13.6	2.2	0.79	1.83	1.17	0.500	40.7
Marisml. (England)	9.5	14.2	1.9	0.69	1.99	1.71	1.363	32.6
PL V (USA,KA)	11.2	16.1	2.3	0.77	1.54	0.71	0.370	39.7
MT 7877 (USA,MT)	21.8	14.3	4.4	0.71	1.97	1.43	0.927	40.8
Means	16.2	14.2	3.0	0.77	1.85	1.20	0.637	38.0
C.V.	12.6	1.9	11.5	3.4	7.4	13.2	22.5	3.3
LSD.05	1.1	0.2	0.2	0.01	0.08	0.07	0.078	0.7
LSD.01	1.6	0.2	0.3	0.02	0.11	0.12	0.111	1.0

GY = Grain Yield; GPP = Grain Protein Percentage; TPP = Total Plant Protein; NHI = Nitrogen Harvest Index; ChL.D. = Chlorophyll Duration

Table 17. Trait means of 22 tall cultivars grown in 1984 and 1985 at 6 soil N levels.

Soil N Levels	GY	HI	GPP	TPP	NHI	Chlorophyll Concent. Anth.	E.D.	M.D.	H.D.	ChL.D.
1984										
NO-(110)+	50.7	0.39	13.8	9.2	0.756	2.45	2.36	1.15	0.27	44.2
N1-(195)	49.8	0.41	13.9	8.9	0.779	2.50	1.78	1.04	0.22	42.1
N2-(280)	48.4	0.38	14.0	8.8	0.763	2.52	1.57	1.57	0.20	40.0
LSD.05	4.7	0.02	0.2	0.9	0.028	0.05	0.15	0.14	0.08	0.8
LSD.01	9.7	0.05	0.5	2.0	0.058	0.11	0.30	0.28	0.16	1.7
1985										
NO-(22)	17.9	0.45	13.8	3.1	0.784	2.23	2.08	1.42	0.80	39.4
N1-(132)	19.9	0.43	14.9	3.8	0.774	2.35	2.02	1.46	0.84	39.2
N2-(242)	17.3	0.43	15.5	3.5	0.756	2.17	1.86	1.31	0.77	37.8
LSD.05	0.7	0.05	0.2	0.1	0.014	0.06	0.06	0.05	0.06	0.3
LSD.01	1.5	0.10	0.3	0.2	0.029	0.12	0.12	0.10	0.13	0.7

GY = Grain Yield; HI = Harvest Index; GPP = Grain Protein Percentage; TPP = Total Plant Protein; NHI = Nitrogen Harvest Index; E.D. = Early Dough; M.D. = Medium Dough; H.D. = Hard Dough; ChL.D. = Chlorophyll Duration

+ = Kg/ha Soil Nitrogen (available + added)

Table 18. Trait means of 22 medium height cultivars grown in 1984 and 1985 at 6 soil N levels.

Soil N Levels	GY	HI	GPP	TPP	NHI	Chlorophyll Concent. Anth.	E.D.	M.D.	H.D.	ChL.D.
1984										
NO-(110)+	45.1	0.42	13.4	7.7	0.776	2.49	2.38	0.99	0.25	45.5
N1-(195)	46.7	0.43	13.7	8.0	0.787	2.51	1.74	0.86	0.18	44.3
N2-(280)	46.0	0.39	13.9	8.3	0.753	2.54	1.43	1.44	0.14	41.9
LSD.05	3.9	0.02	0.2	0.6	0.022	0.05	0.17	0.16	0.06	0.8
LSD.01	8.0	0.05	0.5	1.3	0.045	0.10	0.36	0.33	0.13	1.6
1985										
NO-(22)	13.6	0.44	13.3	2.3	0.774	2.22	2.00	1.33	0.58	39.4
N1-(132)	15.8	0.44	14.4	3.0	0.764	2.29	1.83	1.21	0.57	38.6
N2-(242)	16.1	0.44	14.8	3.1	0.766	2.16	1.77	1.15	0.61	39.3
LSD.05	0.3	0.01	0.1	0.1	0.011	0.04	0.13	0.05	0.07	0.3
LSD.01	0.5	0.02	0.2	0.2	0.023	0.08	0.27	0.11	0.14	0.7

GY = Grain Yield; HI = Harvest Index; GPP = Grain Protein Percentage; TPP = Total Plant Protein; NHI = Nitrogen Harvest Index; E.D. = Early Dough; M.D. = Medium Dough; H.D. = Hard Dough; ChL.D. = Chlorophyll Duration

+ = Kg/ha Soil Nitrogen (available + added)

Table 19. Trait means of 22 short cultivars grown in 1984 and 1985 at 6 soil N levels.

Soil N Levels	GY	HI	GPP	TPP	NHI	Chlorophyll Concent. Anth.	E.D.	M.D.	H.D.	ChL.D.
1984										
NO-(110)+	47.1	0.44	13.1	7.7	0.800	2.50	2.38	1.01	0.22	45.2
N1-(195)	46.2	0.44	13.3	7.7	0.800	2.58	1.71	0.93	0.17	42.9
N2-(280)	45.7	0.41	13.6	7.8	0.779	2.55	1.39	1.38	0.14	41.4
LSD.05	4.6	0.03	0.2	0.8	0.025	0.04	0.08	0.09	0.04	0.7
LSD.01	9.6	0.05	0.4	1.7	0.051	0.08	0.16	0.19	0.08	1.4
1985										
NO-(22)	15.5	0.45	13.3	2.5	0.788	2.25	2.06	1.36	0.63	39.3
N1-(132)	16.8	0.43	14.4	3.1	0.773	2.31	1.91	1.19	0.53	37.0
N2-(242)	16.2	0.44	15.1	3.2	0.756	2.18	1.57	1.04	0.38	37.8
LSD.05	0.6	0.01	0.1	0.1	0.013	0.04	0.06	0.05	0.03	0.3
LSD.01	1.2	0.02	0.3	0.2	0.026	0.08	0.13	0.10	0.07	0.7

GY = Grain Yield; HI = Harvest Index; GPP = Grain Protein Percentage; TPP = Total Plant Protein; NHI = Nitrogen Harvest Index; E.D. = Early Dough; M.D. = Medium Dough; H.D. = Hard Dough; ChL.D. = Chlorophyll Duration

+ = Kg/ha Soil Nitrogen (available + added)