

AN ABSTRACT OF THE THESIS OF

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Title: EFFECT OF LATE FOLIAR APPLICATIONS OF UREA ON PROTEIN,  
HARDNESS, AND YIELD OF WINTER WHEAT CULTIVARS (TRITICUM  
AESTIVUM L.)

Abstract approved: \_\_\_\_\_

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Willis L. McCuiston

The response of cultivars of winter wheat to foliar applications of urea was examined for grain protein content, yield, and kernel hardness. The objectives of this study were to measure the effect of urea on these three traits in diverse environments and to explain how potential responses could occur by (1) determining potential uptake of N15 labelled urea from foliar application under greenhouse conditions using different cultivars, light duration, and soil fertility levels; (2) analyzing immature spike samples collected following foliar applications in the field during the flowering period; and (3) examining grain from primary and secondary tillers and from central and lateral florets within spikelets.

Foliar applications significantly increased grain protein content over the control and standard topdressing treatment in most cultivars. For example, for a single cultivar at the Hyslop location in 1979-1980, protein percents of 11.18, 12.22, and 13.60 were obtained for the

control, standard topdressing, and foliar urea treatments respectively.

Generally, foliar applications of urea failed to increase grain yield over the control, and yield was significantly below the standard topdressing treatment. However, the combination of topdressed nitrogen with foliar sprays produced yields equal to topdressing alone while significantly increasing protein. Mean yields at the Wasco location of the topdressing treatment and the combination of topdressing and one foliar application were 4.05 and 4.04 T/ha, whereas the grain protein content averaged 9.34 and 10.45 percent respectively.

Foliar applications of urea did not significantly alter kernel hardness. Significant cultivar x nitrogen treatment interactions were found for protein, yield, and hardness, and split applications of foliar nitrogen did not significantly change these results.

A potential uptake of 29.2 to 61.4 percent of actual foliar applications was found with N15 labelled urea in a greenhouse experiment. A significant effect for light duration was observed with recovery values of 41.4 and 46.6 percent for 12 and 18 hour photoperiods respectively. The varieties Stephens and Centurk had a significant difference in uptake of 48.1 and 39.9 percent recovery respectively.

Following urea sprays which were initially applied at the heading stage, nitrogen treatment differences were found within three weeks after anthesis. The ranking of treatments from immature spike analysis corresponded to grain protein analysis at harvest. Nitrogen treatments did not change the protein or hardness levels disproportionately for mature spikes in comparing grain from primary and secondary tillers or from central and lateral florets within the spikelets.

Effect of Late Foliar Applications of Urea on Protein,  
Hardness, and Yield of Winter Wheat  
Cultivars (Triticum aestivum L.)

by

David Wayne Altman

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## TABLE OF CONTENTS

	<u>page</u>
INTRODUCTION .....	1
LITERATURE REVIEW .....	3
I. Effect of Nitrogen on Protein and Hardness of Wheat ...	3
II. Effect of Nitrogen on Yield of Wheat .....	7
III. Foliar Application of Urea on Wheat .....	9
MATERIALS AND METHODS .....	15
I. Field Establishment and Treatments .....	15
II. Greenhouse Experiment .....	19
III. Application and Measurement Techniques .....	20
IV. Statistical Analysis .....	22
RESULTS .....	25
I. All Trials, 1978-1979 .....	25
II. Immature Spikes .....	29
III. Main Trial, Hyslop 1979-1980 .....	34
IV. Main Trial, Wasco 1979-1980 .....	43
V. Primary and Secondary Tillers, 1979-1980 .....	51
VI. Greenhouse Experiment .....	54
DISCUSSION .....	57
CONCLUSIONS .....	64
BIBLIOGRAPHY .....	66
APPENDICES .....	72
Appendix 1: Variety Descriptions .....	72
Appendix 2: Techniques for Protein Determination .....	75

## LIST OF FIGURES

<u>Figure</u>		<u>Page</u>
1	Nitrogen treatment means of immature spikes, Hyslop 1979-1980	32
2	Nitrogen treatment means of immature spikes, Wasco 1979-1980	33



## LIST OF TABLES

<u>Table</u>		<u>Page</u>
1	Meteorological data for four location-years of yield trials	16
2	Treatments applied to subplots at Hyslop and Wasco, 1979-1980	18
3	Soil analysis for 1979-1980 locations	18
4	Coefficients for partitioning the sums of squares for the data from 1979-1980 yield trials	24
5	Analysis of variance for main trial, Hyslop 1978-1979	26
6	Analysis of variance for main trial, Pendleton 1978-1979	27
7	Analysis of variance for protein of immature spikes, Hyslop 1979-1980.	30
8	Analysis of variance for protein of immature spikes, Wasco 1979-1980	31
9	Analysis of variance for main trial, Hyslop 1979-1980	35
10	Hyslop varietal means 1979-1980	37
11	Hyslop nitrogen treatment means 1979-1980	38
12	Yield means for nitrogen treatments within varieties, Hyslop 1979-1980	39
13	Hardness means for nitrogen treatments within varieties, Hyslop 1979-1980	40
14	Protein means for nitrogen treatments within varieties, Hyslop 1979-1980	41
15	Height means for nitrogen treatments within varieties, Hyslop 1979-1980	42
16	Analysis of variance for main trial, Wasco 1979-1980	44
17	Wasco varietal means 1979-1980	45
18	Wasco nitrogen treatment means 1979-1980	46

<u>Table</u>		<u>Page</u>
19	Yield means for nitrogen treatments within varieties, Wasco 1979-1980	47
20	Hardness means for nitrogen treatments within varieties, Wasco 1979-1980	48
21	Protein means for nitrogen treatments within varieties, Wasco 1979-1980	49
22	Height means for nitrogen treatments within varieties, Wasco 1979-1980	50
23	Partial analysis of variance for spike type, Wasco 1979-1980	52
24	Partial analysis of variance for spike type, Hyslop 1979-1980	52
25	Protein means for spike type within varieties, Hyslop 1979-1980	53
26	Hardness means for spike type within varieties, Hyslop 1979-1980	53
27	Analysis of variance for recovery of N15	55

EFFECT OF LATE FOLIAR APPLICATIONS OF UREA ON PROTEIN,  
HARDNESS, AND YIELD OF WINTER WHEAT CULTIVARS  
(TRITICUM AESTIVUM L.)

INTRODUCTION

The most important single crop grown in the Pacific Northwest is wheat. Of the total crop produced, 85 percent is soft white wheat, and approximately 80 percent of all wheat produced is exported (Dr. N. Goetze, personal communication). In recent years, there has been an interest in expanding the production of hard red winter wheat in Oregon and adjacent states due to fluctuations in demand for white wheat. Diversification of wheat production could offer the Oregon farmer more options for marketing along with possibly greater financial returns.

Unfortunately, most hard red winter varieties commonly grown throughout the United States are poorly adapted to Pacific Northwest conditions. Wanser is the major hard red winter wheat variety grown in this region, but it has low yield potential and is not preferred in the better wheat growing regions. Also, Wanser has variable levels of quality from year to year causing uncertainty as to whether or not it will meet grading requirements for export as hard red wheat.

Therefore, it is of interest to examine soil fertility and plant nutrition with the objective of properly managing the wheat crop for optimal yield and quality. Grain protein content is foremost among the

quality factors essential for an acceptable hard red wheat. Foliar applications of nitrogen, particularly urea, have been used in a number of crops to increase protein levels and yields at critical times, and this could be especially appropriate under the variable conditions found in the Pacific Northwest.

In light of these considerations, this study was conducted to answer several questions about foliar fertilization. First, is foliar application of nitrogen in the form of urea effective in increasing protein, and what is the corresponding influence on yield and another quality factor, hardness? Second, what is the potential uptake of nitrogen from foliar applications and how soon do differences between treatments show up following application? Finally, is foliar applied nitrogen translocated preferentially to those types of seed that are normally higher or lower in protein content?

In answering the first question, an experiment was designed including 11 cultivars representing diverse winter wheat germplasm. Several experiments were planned for addressing the last two questions. The objectives were to decide: (1) if an increase in nitrogen uptake occurred mainly in primary or secondary tillers or in central or lateral seeds within the spikelet; (2) if differences in nitrogen content could be found in the ripening spikes over a period of four weeks after foliar applications; and (3) how much of the additional nitrogen could be shown to actually reach the grain from a foliar application using N15 labelled urea.

## LITERATURE REVIEW

I. Effect of Nitrogen on Protein and Hardness of Wheat

Increases in protein as a result of nitrogen applications have been frequently reported (Davidson and LeClerc, 1917; Lamb, 1967; Hucklesby et al, 1971; Daigger et al, 1976; Rousset, 1978). However, some researchers have shown no response from supplemental nitrogen (McKercher, 1964; Schlehuber and Tucker, 1967). McKercher postulated that differences within a variety can be attributed more to specific soil profiles and associated micro-climates in the field.

Variation in grain protein content does seem to relate mainly to environmental factors, nitrogen supply being generally one of the most important variants, but the response curve does not parallel that for yield (Kramer, 1979). In a greenhouse experiment, Alkier et al (1972) showed an equal response to nitrogen for both yield and protein, but protein continued to increase with additional fertilizer after yield had plateaued. However, Schlehuber and Tucker (1967) have concluded that often even moderate rates of fertilization will improve yield with no subsequent increase in protein content. Kramer (1979, 1980) has hypothesized a more complicated response curve directly related to the yield response. The correlation with grain yield will be positive, negligible, or negative within a cultivar, and the overall range of grain protein content will be cultivar dependent.

Rousset (1978) emphasizes that fluctuating results of nitrogen

studies can be attributed to date and amount of application as well as variable response from different genotypes. Other workers have reported on the importance of date of application to nitrogen response for protein content. Whereas the amount of available nitrogen after heading may have little effect on grain yield (Thorne, 1962; Langer and Liew, 1973), a protein response can result from either soil or foliar applications (Lamb, 1967; Croy and Hageman, 1970; Hamid and Sarwar, 1976). Gericke (1922) reported that the latest applications consistently produced the highest protein content in the seed for both winter and spring wheats. His data for the spring variety White Australian showed a progressive increase in protein that corresponded to each increase in the length of time after planting when nitrogen was applied. Seth et al (1960) found soil applications were much more effective after heading in raising protein content in a greenhouse experiment. Nitrogen fertilization before heading increased grain protein content by 3.6 percent, but applications after heading raised the level by 8.4 percent. Mesdag (1964) used 30 kg/ha at three growth stages and found no increase in average protein content when applied at sowing, an 11 percent increase at heading, and an increase of 28 percent during flowering. The effect of late foliar nitrogen treatments on protein content is discussed in detail in section III.

In addition to date of application, protein content can be affected by the type of fertilizer treatments. Rankin (1946) reported that several small applications spread throughout the season were more efficient than one application in increasing plant uptake of nitrogen. Hamid and Sarwar (1976) using N15 showed that six split applications

significantly increased protein content over either one or two treatments. They also reported that ammonium nitrate was more effective than urea. Spratt (1974) concurred that nitrate nitrogen is more effective at later growth stages in increasing the percent protein in the grain. However, Schlehner and Tucker (1967) found that, although under specific conditions one form may have an advantage over another, the field experimentation in the Wheat Belt of the United States has shown all sources to be equally effective when properly applied. Allison (1966) notes that some of the conditions allowing for inefficiency of urea can be heavy rates, lack of soil incorporation, and certain climatic variables. Ayoub (1974) reported no significant differences in the percent grain nitrogen from application of urea and two sources of nitrate.

Climatic factors also appear to influence the effect of nitrogen response. Some researchers supported the viewpoint that climate was the most limiting factor (Thatcher, 1913; Shaw, 1913). Interactions of fertilizer and available soil moisture have been reported (Pushman and Bingham, 1976). At high moisture levels, it has been shown that nitrogen applications will increase both yield and protein, but for the same conditions in the absence of fertilizer, yield will increase and protein decrease (Hutcheon and Paul, 1966). At low moisture levels the nitrogen recovery decreases with increasing rates of nitrogen (Humbert and McVickar, 1963), and apparently there is a critical soil moisture level for the protein response reported by Hutcheon and Paul.

Temperature also affects the uptake of nitrogen during the growing season (Lamb, 1967; Partridge and Shaykewich, 1972; Smika and Greb,

1973). Higher soil temperatures prior to the period of maturation have produced increased uptake and higher protein levels (Smika and Greb, 1973; Mifflin, 1980). High ambient temperatures during the latter stages of plant development can lower the grain protein (Rousset, 1978) as well as loaf volume (Lamb, 1967).

Cultivar differences for fertilizer response have also been examined but with mixed results. Seth et al (1960) reported differences in nitrogen uptake for several high and low protein cultivars, including Atlas 66, and attributed this to continued uptake later in the season. Brunori et al (1977) substantiated this finding by demonstrating that high protein cultivars like Atlas 66 have an extended period of protein synthesis associated with a delayed decrease in RNA activity. Atlas 66 was able to continue protein synthesis until seed water content dropped to 28 percent, whereas Irnerio stopped synthesizing at the 50 percent level. Other researchers have not reached the same conclusion (Syme et al, 1975; Miezán, 1977), and many fertilizer experiments have shown no cultivar x fertilizer interaction (Reeves, 1954; McNeal et al, 1971; Pushman and Bingham, 1976). Possibly, many of the cultivars that are the result of selection for adaptation to modern farming practices do not have large differences in response, however exceptions, such as Atlas 66, do exist that are more efficient in nitrogen utilization.

Very little research has been reported specifically on the effect of supplemental nitrogen on kernel hardness. Although the importance of hardness in determining the milling characteristics of wheat is accepted (Finney and Yamazaki, 1967), less is known about the factors that influence hardness. Carrillo et al (1976) reported that nitrogen



fertilization didn't affect kernel hardness of the cultivar Mexifen over the three years of their study. However, Mesdag (1964) noted harder and more vitreous grains were found in the fertilizer-induced high protein samples for the 19 cultivars tested. The increase in hardness was associated with lighter crumb color, higher loaf volume, and higher scores for crumb texture and appearance and brake shred.

Schlehuber and Tucker (1967) stated that percent hard kernels may be increased by additional nitrogen fertilization, and they also equated yellowberry and hardness. Yet, Smika and Greb (1973) emphasize that yellowberry is not a soil management problem, but a disease that decreases nitrogen translocation to the grain. A decrease in protein of 0.4 percent was found for each 10 percent increase in incidence of yellowberry. This second finding has been corroborated by Waines et al (1978). They decided to classify yellowberry as a physiological disorder. Therefore, the literature on hardness appears to be confusing, and any conclusions are to be examined in light of what definitions and assumptions are used about the nature of hardness and its relation to other traits.

## II. Effect of Nitrogen on Yield of Wheat

The increased yields of wheat with supplemental nitrogen fertilization has been well documented (Davidson and LeClerc, 1917; Burke, 1925; Schlehuber and Tucker, 1967; Kramer, 1979). Although this is generally the case, yield increases have not always been reported from applications of nitrogen (Pittman and Tipples, 1978).

Response to nitrogen is a function of many environmental, cultural, and genetic factors. Timing of application has been shown to influence yield response (Davidson and LeClerc, 1917; Rankin, 1946; Ayoub, 1974). Also, splitting the application can result in substantially better response although this may not always be feasible. Hamid and Sarwar (1976) found that splitting nitrogen applications into two treatments resulted in superior response from both urea and ammonium nitrate. Jain et al (1971) reported that two applications increased the efficiency of applied nitrogen while three doses did not augment efficiency.

Cultural practices and the particular environment will influence the response from given types of nitrogen fertilizers. Usually, ammonium forms of nitrogen are more prone to loss (Allison, 1966; Alessi and Power, 1973). However, delaying application of urea has resulted in increased uptake (Mason et al, 1972), and Allison (1966) reports that if moderate rates are used, differences between forms of nitrogen are not significant. Workers have corroborated that ammonium and nitrate sources of nitrogen do not have a differential effect on yield of wheat when properly applied (Spratt and Gasser, 1970; Spratt, 1974).

Moisture supply is a major factor governing yield responses to nitrogen. Investigators have noted increased uptake of nitrogen with more available water (Power et al, 1961; Humbert and McVickar, 1963). However, the influence of moisture stress on response to nitrogen can depend on the growth stage of the wheat plant. Hutcheon and Paul (1966) showed that changes in moisture were particularly detrimental at the soft dough stage and resulted in a reduced response to nitrogen fertilization.

Cultivar differences are another consideration. McNeal et al (1971) found no cultivar x fertilizer interaction when comparing short, medium, and tall cultivars for yield. They concluded that similar fertilization could be practiced across these genotypes. However, Johnson et al (1973) found a difference in response of two cultivars of winter wheat. These differences were evident at high rates of application but not obvious at the lower levels.

### III. Foliar Application of Urea to Wheat

Due to the successful utilization of urea and other nitrogen sprays in several horticultural crops such as apples, researchers began investigations of the potential of foliar applications in small grains. Finney et al (1957) were the first workers to do large scale testing of liquid urea applications of wheat. They reported results for yield, test weight, protein content, loaf volume, and several other quality factors with Pawnee winter wheat for two growing seasons. During the first year, significant yield increases were obtained with a number of treatments, especially higher concentrations before flowering.

Flowering stage was the best time to spray for increased protein content, and split applications further increased the effect, however this was not additive. The increases in protein did not result in corresponding improvement of other quality factors such as loaf volume which was attributed to incomplete gluten protein synthesis. There was less increase in protein content for the second year's trial, and no

significant increases in yield were reported. Finney explained the reduced response by the higher soil fertility and a 50 percent longer fruiting period. After testing rates of 10, 30, and 50 lb/A with the first year's trial, the latter was chosen for all applications in the second season. Large scale aerial applications have also been tried which have supported Finney's conclusions (Murphy et al, 1977; Gallagher et al, 1977; Lamond et al, 1978; Scott et al, 1978). Kansas has also decided to use late boot applications of liquid urea to increase the protein content of their breeding material analyzed for quality as a result of this research (Heyne, 1979).

Reeves (1954) initially tested rates of 10, 20, 40, and 80 lb/A applied as a urea spray with one cultivar of winter wheat in Australia. Three timings of the applications were attempted: (1) five weeks before heading, (2) heading, and (3) three split applications five weeks before heading, at heading, and flowering. Reeves concluded that 40 lb/A was the best rate, that significant increases for yield were obtained if the application was prior to heading, and that significantly higher protein was observed with applications at any time near heading stage. In addition, the response of four cultivars to one application of 40 lb/A at the heading period was tested, and a comparison of equal nitrogen amounts applied to the soil versus a foliar application was made for one cultivar. No significant difference was found for cultivar response, but the foliar application of urea produced significantly higher protein levels. However, the urea treatment did not increase yield or 1000 kernel weight when compared to the soil application.

Sadaphal and Das (1956) found significant increases in protein,

yield, and 1000 kernel weight for rates of urea spray as low as 4.25 lb/A. All progressive rates up to 76 lb/A resulted in increases above the control for two growing seasons. Sadaphal and Das (1966) reported more extensively on the same study covering three years' data and concluded that sprays of up to 51 lb/A were effective when applied at the stages of heading for yield increases and post-flowering for increasing protein. They also decided that this increase in protein was due to higher accumulation of nitrogen in the terminal spikelets although separate analysis was not reported for different segments of spikes. In addition, they claimed that foliar urea treatment resulted in a decrease of mottling in the grain at harvest. This must mean that there was less incidence of yellowberry following foliar application of urea.

Not all of the initial investigations of the effect of foliar urea applications showed significant increases for yield and/or grain quality. Juarez and Swanson (1956) used low, moderate, and high rates of liquid urea sprayed 15 days before, 15 days after, and just at flowering stage in Peru. They found higher yields from pre-flowering application at low and moderate rates, however these were not significant. Although protein was higher at all concentrations applied at post-flowering stage, these differences also were not significant. Jain et al (1971) reported no response to foliar sprays at heading time with rates of 60, 100, and 140 kg/ha. Thorne and Watson (1955) attempted to retard leaf area decline and compared 50 lb/A of nitrogen applied as a liquid to either the soil or the foliage. The foliar treatments were either one spray of urea or eight sprays of ammonium

nitrate. Although the urea spray gave significantly higher nitrogen content of the grain than the ammonium nitrate treatments, there were no differences in yield or other characters for comparisons of soil and foliar applications.

Subsequent studies expanded upon the early research. Seth et al (1960) tried to determine if high or low protein cultivars could have differing genetic potential for ability to use nitrogen as nitrate or urea. Both urea and nitrate sprays were applied in a greenhouse experiment, and they substantially increased protein when applied after heading stage. The two high protein cultivars gave a much greater response than the two low protein cultivars, but no differences in nitrogen content of vegetative parts were noted prior to heading. This response was hypothesized as due to a higher rate of protein synthesis occurring in the kernels and possibly better translocation.

Ries et al (1976) studied the effectiveness of urea sprays in increasing the nitrogen content of certain portions of the spike more than others. The seeds of lateral florets had higher protein content than those of the central florets within the spikelets, and the lower ten spikelets were higher than those in the upper portion of the spike. Foliar urea application resulted in higher levels of protein for all seeds in the spike, but more of the additional nitrogen went to those that were already high in protein content. These results are contrary to the hypothesis advanced by Sadaphal and Das (1966). Forty kg/ha of nitrogen applied as a split application (anthesis and 20 days post-anthesis) gave the most response, and no significant cultivar x treatment interaction was reported. The results of Ries' work would

seem to be in agreement with other research on assimilate limitations to distal kernels which pointed out the difficulty in overcoming nitrogen deficiencies even after excision of the laterals (Simmons and Moss, 1978).

Pushman and Bingham (1976) have done one of the most complete studies of the effect of one additional spray of urea after anthesis and its influence on quality factors. Comparing three rates of nitrogen, one of which had an additional spray of 45 kg/ha of urea, under both natural precipitation and supplemental irrigation, they measured yield, number of heads/square meter, grain protein, protein production, 1000 kernel weight, test weight, kernel alpha amylase, flour alpha amylase, milling extraction, flour protein content, water absorption, loaf volume, and loaf score. Protein content had no interactions of significance for fertilizer x cultivar but showed an irrigation x fertilizer effect. A significant increase in protein for the urea application was noted (12.4 percent increase under irrigation and 8.8 percent increase with natural precipitation). No advantage was noted for yield, and the type of fertilizer did not affect the negative correlation between yield and grain protein for the ten cultivars tested. Although the urea application increased water absorption and loaf score as well as protein content, it did not improve loaf volume, and flour extraction dropped.

Investigators have attempted to verify the effect of urea sprays with labelled nitrogen (Alkier et al, 1972). Sprays and granular applications were equally effective in the field, but only one percent of the N15 applied through the foliage was found in the grain in a

greenhouse test. This compared with 30 percent recovered when applied via the soil.

Singh and Seth (1978) looked at differences in percent nitrogen of vegetative parts when fertilizer urea was applied to the soil or the foliage. They found that all vegetative parts were significantly lower in nitrogen at the end of the growing season when a foliar application was used.

Foliar burning has frequently been reported for liquid urea applications (Reeves, 1954; Finney et al, 1957; Sadaphal and Das, 1966). A possible explanation is that urea assimilation and breakdown results in high ammonium concentrations in the foliage which are toxic to plant cells (Mifflin, 1980). Sufficient carbohydrate production through photosynthesis will result in ammonium assimilation (Allison, 1966). Van Vuurde and Tonneyck (1978) attempted to test such an explanation by conducting a greenhouse experiment with high and low nutrient levels of NPK and high and low light intensity. Solutions of 1.5 percent urea were applied four times at 24 hour intervals to one week and one month old seedlings of Kaspar spring wheat. A significant difference in dry weight production was found for the high light intensity treatment, but the level of fertility was not significant for increasing dry weight.



## MATERIALS AND METHODS

I. Field Establishment and Treatments

This study was planted at Hyslop Agronomy Farm near Corvallis, Oregon and at an experimental site on Mr. Quinton Rugg's farm near Pendleton, Oregon for the 1978-1979 crop year. The 1979-1980 experiments were grown at Hyslop and on Mr. Larry Kaseberg's farm near Wasco, Oregon. Soil types were a Woodburn silt loam at Hyslop and a Walla Walla silt loam at Pendleton and Wasco. Precipitation and temperature data for the four location-years are presented in Table 1. Seeding rates of 100 kg/ha at Hyslop and Pendleton and 67 kg/ha at Wasco were used with all cultivars adjusted according to 1000 kernel weight in order to insure equal seed number per plot. All soil amendment rates were determined after consultation with Dr. T. Jackson of the Soils Dept. and/or researchers with the Dryland Cereal Production Project of the Crop Science Department at Oregon State University.

In the first year, the study was designed as a split block. Each plot was three meters long and four rows wide with 20 cm between rows at Hyslop and 30 cm at Pendleton. Five cultivars having red seed, Centurk, Kavkaz, Vorochilovskaja, Pumafen/Lilifen, and Centurk/Ciano, were sown in four replications. Varietal descriptions are found in Appendix 1. Fall and early spring applications of nitrogen corresponded to conventional farming practices for that year and totalled 112 kg N/ha at Pendleton and 168 kg N/ha at Hyslop. In addition, one half of each plot

Table 1. Meteorological data for four location-years of yield trials

Month	Pendleton 1978-79			Hyslop 1978-79		
	Av Max (oC)	Av Min (oC)	Precip (mm)	Av Max (oC)	Av Min (oC)	Precip (mm)
Aug	28.6	11.0	35	26.7	11.8	59
Sept	22.6	7.1	41	21.1	9.8	86
Oct	19.9	-1.0	T	19.5	4.7	25
Nov	6.4	-5.7	43	9.4	-0.6	80
Dec	3.8	-9.7	58	6.1	-1.3	107
Jan	-4.7	-13.9	33	3.1	-3.5	65
Feb	6.7	-2.7	39	8.4	2.2	212
Mar	13.0	0.8	44	14.6	3.7	73
Apr	15.4	3.4	46	15.1	5.2	74
May	21.8	6.3	29	20.0	6.4	54
Jun	27.5	7.7	5	24.3	8.0	10
July	31.9	11.1	3	26.2	10.5	11

Month	Hyslop 1979-80			Wasco 1979-80		
	Av Max (oC)	Av Min (oC)	Precip (mm)	Av Max (oC)	Av Min (oC)	Precip (mm)
Aug	26.0	10.4	68	26.1	12.2	27
Sept	24.7	10.0	55	24.8	8.6	13
Oct	19.7	7.7	183	17.2	5.6	66
Nov	10.7	2.3	104	4.4	-2.2	57
Dec	9.9	2.8	159	5.8	-1.4	17
Jan	7.1	-1.2	170	-0.3	-5.9	87
Feb	10.4	1.9	99	3.1	-1.9	47
Mar	12.1	3.1	102	8.3	0.6	24
Apr	16.7	4.5	92	15.0	3.9	23
May	18.3	6.2	37	17.8	6.7	32
Jun	19.9	9.0	44	20.0	7.8	35
July	27.0	11.4	6	27.2	12.2	4

was treated with a foliar application of six percent urea ten days post-anthesis at a rate of 34 kg/ha. Two percent of the solution consisted of the surfactant Triton X-100. After trimming 30 cm from the ends of each half plot, the center two rows were harvested and analyzed for yield, protein, and hardness. Spike samples were collected from each plot prior to threshing. Primary and secondary tillers were designated according to the procedure used by McNeal and Davis (1966). Another set of spike samples were also used to separate central and lateral seeds within the spikes.

In the second year, the study was designed as a split plot with six treatments per main plot. These treatments at the two locations are shown in Table 2. Whole plots were six meters long and 12 rows wide with 20 cm between rows at Hyslop and 36 cm between rows at Wasco. Four replications of ten entries of winter wheat were grown in 1979-1980. All cultivars from 1978-1979 were included, except Centurk/Ciano, and additional cultivars were Stephens, Wanser, Hatton, GK-Protein, NE 7060, and NE 95021 (See Appendix 1). Soil samples were taken twice during the growing season at each location, and the results are listed in Table 3. Foliar applications were made at the heading stage for the first series and at one week post-anthesis stage for the second. Concentrations and surfactant were identical to those for 1978-1979 although rates were different (See Table 2). Spike samples were collected for protein analysis at the stages of heading, two weeks after heading, and four weeks after heading. Harvested plot areas were 1.5 meters from the center four rows of each subplot at Hyslop and 1.2 meters at Wasco, and mature spikes for comparing primary and secondary tillers were again

Table 2. Treatments applied to subplots at Hyslop and Wasco,  
1979-1980

Location	Nitrogen Fertilizer Treatments (kg/ha)					
	C	T	S <sup>2</sup>	T+S <sup>2</sup>	S <sup>1</sup> +S <sup>2</sup>	T+S <sup>1</sup> +S <sup>2</sup>
Hyslop	0	100	50	50+50	25+25	50+25+25
Wasco	0	28	28	28+28	14+14	28+14+14

C=control, T=topdress, S<sup>1</sup>=first series' spray, S<sup>2</sup>=second series' spray

Table 3. Soil Analysis for 1979-1980 locations

Location	Date of sample	Depth (cm)	pH	P (ppm)	K (ppm)	NO <sub>3</sub> (ppm)	NH <sub>4</sub> (ppm)	S (ppm)
Hyslop								
sample 1	9/10/79	0-15	5.5	-	-	16.2	3.0	-
sample 2	9/10/79	16-30	5.5	-	-	9.3	2.8	-
sample 3	3/4/80	0-30	6.1	-	-	3.03	3.21	-
sample 4	3/4/80	31-90	6.1	-	-	2.33	3.20	-
Wasco								
sample 1	9/21/79	0-15	6.0	-	-	19.0	1.3	3.82
sample 2	9/21/79	16-60	6.7	-	-	3.3	2.2	3.62
sample 3	2/28/80	0-30	6.8	24	335	1.1	1.81	-
sample 4	2/28/80	31-60	6.9	27	281	6.0	3.84	-
sample 5	2/28/80	61-90	7.3	27	250	8.2	2.35	-

sampled as in 1978-1979. All spike samples in the second season were only collected from Stephens, Wanser, Vorochilovskaja, GK-Protein, and NE 95021.

## II. Greenhouse Experiment

Two of the cultivars from the field studies, Centurk and Stephens, were planted in the greenhouse at Corvallis on July 6, 1980 after five weeks of vernalization at 4.4 degrees Centigrade. Two seedlings were planted in each pot, and only the main tiller was allowed to grow. A completely randomized factorial design was used with four plants treated for each factor level. The factors were cultivars (the two mentioned above), daylength (12 and 18 hour days), and fertilizer (no initial fertilization and two tablespoons of slow-release Osmocote 18-6-12).

Approximately three times the number of plants needed were grown in order to insure that uniform plants could be selected for treatment. At anthesis, the selected plants were treated with 0.5 mg of N15 in the form of urea. A 1000 ppm solution of 99.5 percent excess N15 was used. Two percent of the solution was the surfactant Triton X-100 that was used in the field experiments. Extreme care was taken to insure that all of the material was actually applied to the foliage. The precise amount to be applied to each plant was measured with a pipette in the laboratory and then placed into four dram plastic pharmaceutical vials. Plastic vials were utilized so that the solution would form droplets and thus prevent any liquid being left in the vial. The solution was then

painted on the spike and upper leaves of each uni-culm plant with a number five camel hair brush. A rinse solution of distilled water was also painted on untreated leaves to insure that little N15 remained in the vial or on the brush. Single plant applications were made in a sink isolated from the other plants. Handwashing and rinsing of the brushes were done in another sink between the treatment of each plant.

After treatment, when the solution had dried on the foliage, the plant was then placed in one of two identical growth chambers with either a 12 or 18 hour daylength. Temperature was kept constant between 21.1 and 26.7 degrees Centigrade and monitored several times a day. No fluctuations beyond the control limits in either chamber were recorded. Several control plants of each variety were placed in each chamber to calibrate the data.

### III. Application and Measurement Techniques

Foliar applications of urea were made with an AZ Field Test Service sprayer which was powered by a portable carbon dioxide cartridge carried on a belt. Constant pressure of 30 psi was maintained for all applications, and the same quantity of liquid as well as amount of urea was applied for equivalent sprays within a given series. Some foliage burning was noted after each spray, but the plants generally recovered within several weeks. No measurable precipitation was recorded within 24 hours for all treatments except for one series for one variety at Hyslop in 1979-1980 when a light rain occurred seven hours after

application. Topdressing treatments were done by hand, and all soil applied nitrogen was 46-0-0 granular urea.

Yield was measured in grams with a Mettler electronic balance. Notes on height, lodging, and shattering were taken on all plots in 1979-1980 with the intent of modifying yield, protein, or hardness values if significant correlations between traits existed.

Protein measurements were taken by two methods depending on the nature of the samples. The nitrogen content of the three dates of immature spike samples were determined by the micro-Kjeldahl method (Nelson and Sommers, 1973). This analysis was also used to verify results of grain protein determination. Grain protein was analyzed with a Technicon InfraAnalyzer 400. Other researchers have reported on the reliability of this measurement (Williams, 1975; Rubenthaler and Bruinsma, 1978; Schumaker, 1980), and a correlation coefficient of 0.972 was obtained between Technicon and micro-Kjeldahl values. A more detailed description of the micro-Kjeldahl procedure and the calibration of the InfraAnalyzer are presented in Appendix 2.

Hardness was also determined by the InfraAnalyzer. F values were initially obtained from the Western Wheat Quality Laboratory in Pullman, Washington (Dr. G. Rubenthaler, personal communication). Adjustments were then made by comparing Technicon values with hardness values measuring the joules required to mill a 50 g sample as determined by the Hard Red Winter Wheat Regional Laboratory in Manhattan, Kansas.

N15 analysis was done by the Los Alamos Scientific Lab. An automated mass spectrometer was used with an accuracy of 0.001% (Dr. B. McInteer, personal communication). Samples were prepared in

Corvallis using a modified micro-Kjeldahl procedure (W. Silvester. Use of  $^{15}\text{N}$  in plant nutrition studies. Presented at the Stable Isotope Workshop, Corvallis on February 29, 1980). This procedure uses the same method described in Appendix 2 except that the indicator solution used in the distillation did not contain boric acid. Also, after the titration, one drop of dilute sulfuric acid was added to the solution, and then the solution was oven dried and placed in one dram glass vials for shipment.

#### IV. Statistical Analysis

Most of the data were punched onto IBM computer cards and analyzed by the Cyber-Nos system of the Milne Computer Center at OSU. All calibration analysis of variance and regression was computed by desk top calculator. The procedures for all design analysis were obtained from statistical reference texts (Cochran and Cox, 1957; Little and Hills, 1978). Split plot analysis for the immature spikes and the main experiment for protein, yield, and hardness in 1979-1980 required a pooled subplot error of the terms replication x nitrogen level and replication x cultivar x nitrogen level. The split block analysis for the main experiment in 1978-1979 required the separation of the two error terms to test subplots and the interaction between main plots and subplots respectively. The additional experiments examining either central and lateral seeds or main and secondary tillers were analyzed by a split-split plot design (1979-1980) or a split-split block design



(1978-1979) and required an extension of the same methods mentioned above. Finally, the N15 greenhouse study utilized a factorial design with the corresponding analysis.

Two comparisons were planned for the 1979-1980 trial, but these were different at the two locations because of the differences in environment and cultural practices. At Wasco, a comparison between soil vs. foliar applications was planned, whereas at Hyslop, the comparison was between all of the additional nitrogen applied to the soil vs. half applied to the soil and half to the foliage. The other two comparisons which were identical at both locations were a single foliar application vs. a split application and the control vs. all other nitrogen treatments. Coefficients used in these orthogonal contrasts are presented in Table 4.

Table 4. Coefficients for partitioning the sums of squares  
for the data from 1979-1980 yield trials

Location Comparison	Treatments					
	C	T	$S^2$	$T+S^2$	$S^1+S^2$	$T+S^1+S^2$
Wasco						
1: soil vs. foliar	0	+2	-1	0	-1	0
2: split vs. single	0	0	+1	+1	-1	-1
3: control vs. nitrogen	+5	-1	-1	-1	-1	-1
Hyslop						
1: all top vs. half and half	0	+2	0	-1	0	-1
2: split vs. single	0	0	+1	+1	-1	-1
3: control vs. nitrogen	+5	-1	-1	-1	-1	-1

## RESULTS

Because of the diverse nature of the experiments conducted in this study, the results will be presented in six groupings. The environments selected for this study varied greatly since Wasco exemplified summer fallow dryland farming, Hyslop high rainfall continuous cropping, and Pendleton intermediate rainfall continuous cropping. The meteorological data presented in Table 1 points out some of these differences. Therefore, all analyses for different locations are reported on a separate basis.

### I. All Trials, 1978-1979

Analyses of variance for the main trials at Hyslop and Pendleton are presented in Tables 5 and 6 respectively. At both locations there were no significant nitrogen treatment effects in comparing the sprayed plots and control for all traits measured. This was due in part to the small degrees of freedom for subplot error 1. For example, the observed mean square value for protein at Pendleton was relatively large, but the F statistic of 9.85 was just below the .05 significance level for 1,3 df. The observed mean square of nitrogen treatment for protein was significant in the analysis of spike samples selected for central and lateral seeds with an F statistic of 13.92 for the same df. This suggested that for protein, there was a possibility of an effect of a single foliar spray that should be further investigated with more

Table 5. Analysis of variance for main trial, Hyslop 1978-1979.

Source of variation	df	Observed M.S.		
		Yield	Hardness	Protein
Variety	4	9261.65	7895.19**	8.567**
Replication	3	7359.29	16.79	1.530**
Main plot error	12	5953.08	74.47	.248
Nitrogen treatment	1	2640.63	331.20	1.914
Subplot error 1	3	1857.09	88.29	1.749
Nitrogen treatment x variety	4	1025.75	249.14	.234
Subplot error 2	12	2025.72	264.84	.238
Coefficient of variation		16.9%	17.4%	3.6%

\*,\*\* indicate probability of Type I error at the .05 and .01 level respectively

Table 6. Analysis of variance for main trial, Pendleton 1978-1979

Source of variation	df	Observed M.S.		
		Yield	Hardness	Protein
Variety	4	13577.4	10087.60**	18.251**
Replication	3	5508.8	135.29	10.849*
Main plot error	12	4379.8	215.73	2.814
Nitrogen treatment	1	409.6	198.03	2.401
Subplot error 1	3	883.0	93.23	.243
Nitrogen treatment x variety	4	646.9	16.96	.027
Subplot error 2	12	798.3	28.00	.574
Coefficient of variation		10.1%	7.7%	4.9%

\*,\*\* indicate probability of Type I error at the .05 and .01 level respectively

precise measurement.

All of the spike samples were not analyzed. This was because some sample groups had missing samples which would have further reduced the degrees of freedom, the results of the main trial suggested an extensive investigation would not be necessary, and the amount of time required for analyzing the different types of seeds and tillers was prohibitive. Therefore, the analysis of central and lateral seeds was completed for Pendleton, and the analysis of primary and secondary tillers was accomplished for Hyslop.

The major result of these determinations was that there were no significant interaction terms. This part of the experiment was set up to test whether a foliar spray could provide extra nitrogen to those seeds that were deficient or whether any additional nitrogen that was metabolized would be partitioned equally or preferentially to the seeds normally higher in nitrogen. Since there were no interactions, there was an indication that foliar sprays provided nitrogen equally to primary and secondary tillers and central and lateral seeds.

Other results of the spike analysis were of interest as well. For protein, there were significant differences for both type of tiller and seed location in the spike. Therefore, this confirmed earlier reports of differences for these traits (McNeal and Davis, 1966; Ries et al, 1976). As mentioned above, a significant nitrogen treatment term for protein was noted at Pendleton. Also, a significant difference for nitrogen treatment was found for hardness at Hyslop. Both of these results indicated the necessity to further study the possibility of foliar urea applications. Therefore, a more extensive investigation was

planned for 1979-1980.

## II. Immature Spikes

Tables 7 and 8 show the analyses of variance for protein of the three successive sampling dates for Hyslop and Wasco respectively. Of interest was the general agreement of the results from the two locations. One important point about the sampling technique should be noted in order to fully understand the findings. Sampling was based on heading as mentioned in the section on methods, but only at Hyslop was it possible to sample every cultivar at exactly the same growth stage. Because the Wasco location was not conveniently accessible, average heading date across cultivars was used. This is reflected by the consistently higher coefficients of variation at Wasco, the larger observed mean squares for nitrogen treatments at Hyslop, and possibly the only observed interaction for date two at Wasco. Also, the graphs of nitrogen treatment means presented in Figures 1 and 2 demonstrate that more distinct treatment differences are evident from the data of Hyslop and that the results from Wasco tend to merge into a similar response pattern.

However, the close agreement of the data from the two locations is striking. Both locations show no nitrogen effect for the first date which corresponded to heading, but a significant variety response. The mean square for nitrogen then steadily increases across the next two dates and is significant at both locations for date three. In addition,

Table 7. Analysis of variance for protein of immature spikes , Hyslop 1979-1980.

Source of variation	df	Observed M.S.		
		Date 1	Date 2	Date 3
Variety	4	18.4392*	7.3581	5.4833
Replication	1	2.4160	10.9483	1.7682
Main plot error	4	1.6874	3.5595	6.4823
Nitrogen treatment	5	2.8047	1.7886*	4.5948**
Variety x nitrogen treatment	20	1.1443	.6534	.9689
Subplot error	20	1.1884	.5636	.5259
Coefficient of variation		11.0%	8.0%	6.9%

\*,\*\* indicate probability of Type I error at the .05 and .01 level respectively



Table 8. Analysis of variance for protein of immature spikes , Wasco 1979-1980.

Source of variation	df	Observed M.S.		
		Date 1	Date 2	Date 3
Variety	4	47.1354**	4.0785*	4.1062
Replication	1	10.6513	1.9189	.5802
Main plot error	4	1.6018	.4664	.9501
Nitrogen treatment	5	1.7851	1.4330	1.9795*
Variety x nitrogen treatment	20	2.3459	1.4751*	1.1720
Subplot error	20	2.1427	.6933	.6527
Coefficient of variation		14.0%	10.3%	7.9%

\*,\*\* indicate probability of Type I error at the .05 and .01 level respectively

Figure 1. Nitrogen treatment means of immature spikes ,  
Hyslop 1979-1980

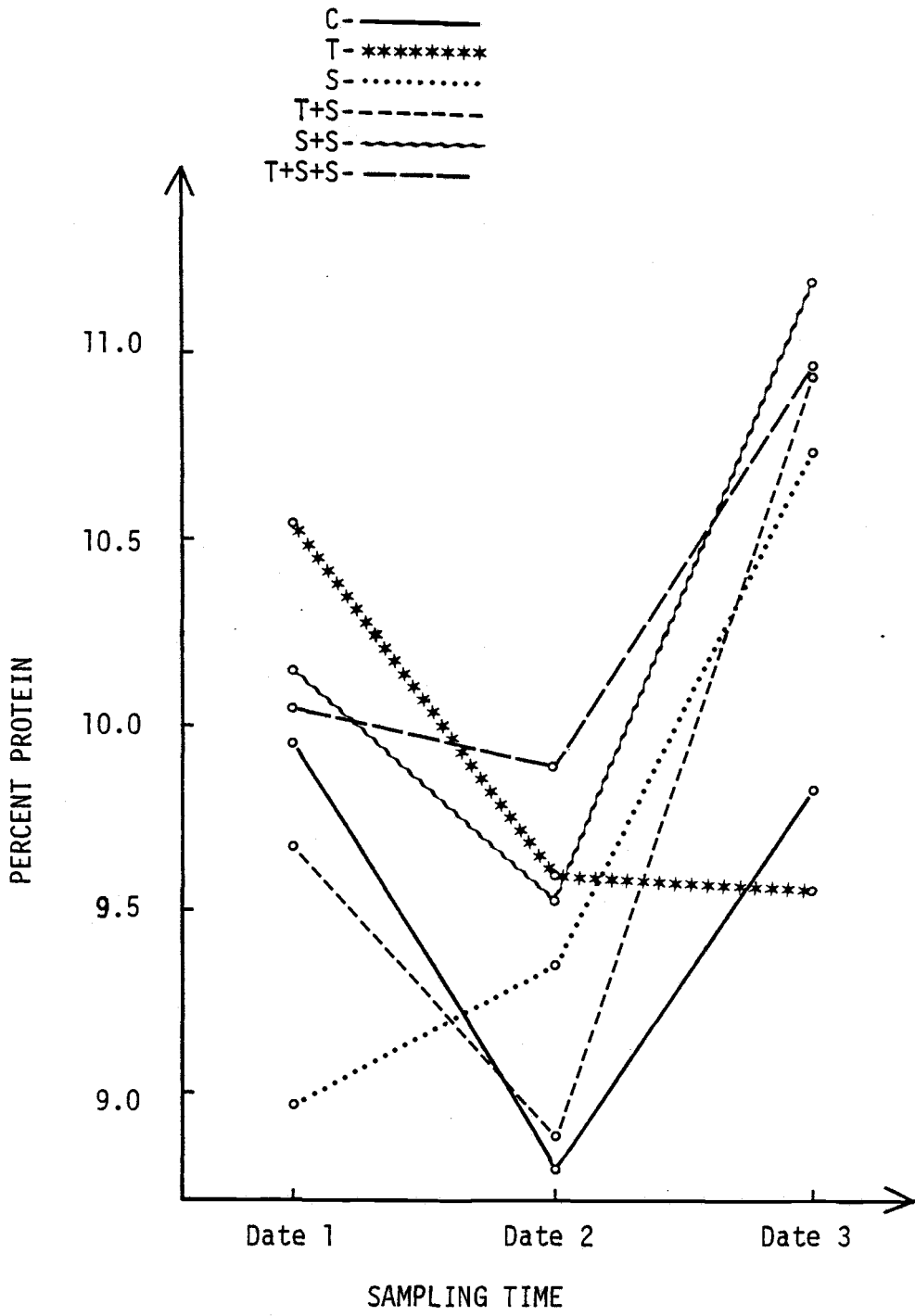
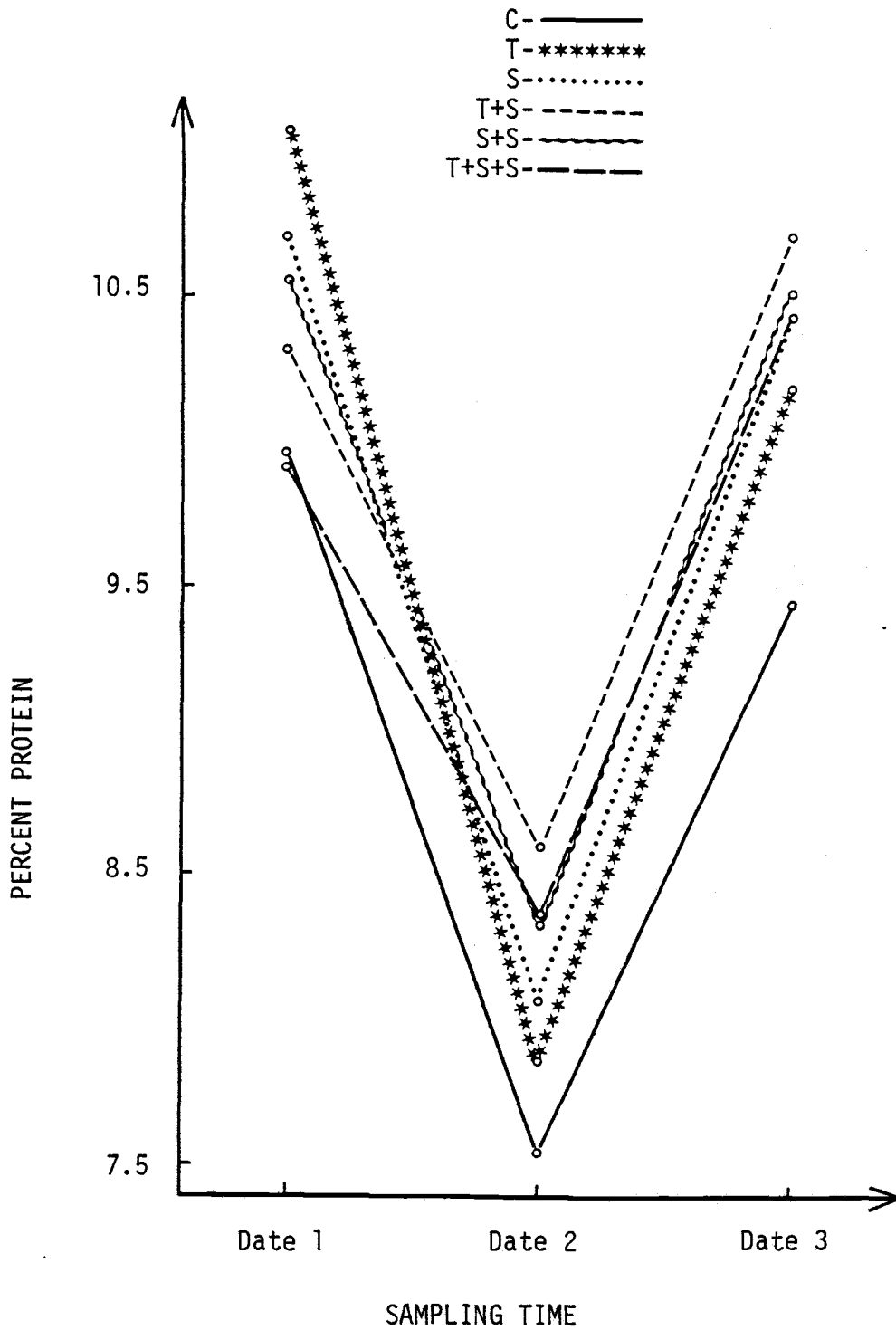


Figure 2. Nitrogen treatment means of immature spikes ,

Wasco 1979-1980



no variety effect is evident by date three which corresponded to three weeks post-anthesis. Also, the relative position of the treatments is generally the same across locations (See Figures 1 and 2). For example, in comparing a topdressing to a topdressing and a single spray (which was applied at sample date two), the topdressing is significantly higher at date one, but the effect of the single spray seems to invert this relationship making the T+S treatment superior by date three.

It seems reasonable to conclude that a marked nitrogen treatment effect is evident by three weeks after anthesis. These results seem even more substantial taken in light of the reduced number of protein determinations due to the time consuming procedure for micro-Kjeldahl analysis. Only five of the ten varieties were sampled, and only two of the four replications were used for each variety at the two locations.

### III. Main Trial, Hyslop 1979-1980

The analysis of variance for yield, hardness, protein, and height is presented in Table 9. The analysis for lodging is not shown because of the high CV of 81 percent and the ineffective reduction of error from the analysis of covariance. These results could either have been due to difficulty in accurately measuring lodging or the efficiency of the blocking in eliminating this effect suggested by the significant replication effect for all traits. Differential response across replications was noted which seemed to be a result of soil fertility and the impaired drainage in the field, but the reasonable CV's indicate

Table 9. Analysis of variance for main trial, Hyslop 1979-1980.

Source of variation	df	Observed M.S.			
		Yield	Hardness	Protein	Height
Variety	9	502765.0**	8506.24**	33.937**	8548.43**
Replication	3	206163.0**	878.29**	7.442*	922.36**
Main plot error	27	20517.2	117.92	1.979	54.07
Nitrogen treatment	5	194463.0**	94.11*	22.025**	1059.92**
Comparison 1	1	9238.00	81.43	27.36**	175.10**
Comparison 2	1	13286.03	192.06*	1.28	330.63**
Comparison 3	1	389952.00**	55.43	67.63**	1240.33**
Variety x nitrogen treatment	45	9706.7*	59.92*	.463*	40.43*
Subplot error	150	6750.0	38.52	.282	23.99
Coefficient of variation		12.6%	9.3%	4.7%	4.1%

Comparison 1=all top vs half and half, Comparison 2=split vs single, Comparison 3=control vs nitrogen

\*,\*\* indicate probability of Type I error at the .05 and .01 level respectively

that none of these difficulties substantially affected the precision of measurements.

A highly significant varietal response was recorded for all measured traits. This would indicate that the cultivars chosen for testing differed enough to make the other results meaningful. However, it must be stressed that a fixed effects model for analysis of variance has to be employed, and all conclusions will directly apply just to these cultivars. Varietal means are listed in Table 10 with only the standard error for each trait since it was not the intent of this experiment to make a primary objective of comparing response of cultivars to the parameters measured.

One of the main objectives of this study was to look at the response of nitrogen treatments and any variety x nitrogen treatment interaction. From examining Table 9, it can be seen that there was a significant subplot effect and interaction for all traits measured. However, from a brief examination of the results of the three planned orthogonal comparisons, it can be seen that for each factor the nitrogen effect was the result of a different type of response. Therefore, the treatment means along with the standard error, LSD, and HSD are recorded for all readings in Table 11, and the means for yield, hardness, protein, and height within each variety as well as the appropriate statistics for comparison both within and between varieties are presented in Tables 12, 13, 14, and 15 respectively.

The different responses to nitrogen treatments within varieties are noted after perusal of these tables. For example, in Table 12 when comparing the response of Stephens and Wanser, it can be seen that no

Table 10. Hyslop Varietal Means 1979-1980.

Variety	Yield (T/ha)	Hardness *	Protein (%)	Height (cm)
Stephens	6.71	61.1	9.67	103
Wanser	3.47	69.7	11.79	144
Hatton	5.42	97.5	11.15	143
Centurk	4.52	75.3	11.99	130
Kavkaz	6.90	56.3	11.78	136
Vorochilovskaja	5.50	82.8	12.77	111
Pumafen/Lilifen	6.10	25.0	12.91	134
GK-Protein	5.61	66.9	12.63	102
NE 7060	4.95	63.3	14.16	113
NE 95021	3.60	69.3	12.36	91
Standard error	0.24	2.2	0.29	2

\* values < 50 grade soft.  
 values > 50 grade hard.

Table 11. Hyslop nitrogen treatment means 1979-1980.

Nitrogen treatment	Yield (T/ha)	Hardness *	Protein (%)	Height (cm)
C	4.55	67.8	10.93	116
T	5.87	64.7	11.46	127
S <sup>2</sup>	4.79	65.9	12.59	118
T+S <sup>2</sup>	5.68	65.8	12.47	126
S <sup>1</sup> +S <sup>2</sup>	5.01	68.9	12.78	115
T+S <sup>1</sup> +S <sup>2</sup>	5.76	67.2	12.48	123
Standard error	0.10	1.0	0.08	1
LSD <sup>#</sup>	0.29	2.7	0.23	2
HSD <sup>#</sup>	0.52	4.0	0.34	3

\* values < 50 grade soft.  
values > 50 grade hard.

# .05 significance level



Table 12. Yield means for nitrogen treatments within varieties,  
Hyslop 1979-1980.

Variety	(T/ha)					
	Nitrogen treatment					
	C	T	S <sup>2</sup>	T+S <sup>2</sup>	S <sup>1</sup> +S <sup>2</sup>	T+S <sup>1</sup> +S <sup>2</sup>
Stephens	6.01	7.27	5.60	7.04	6.13	8.19
Wanser	3.00	3.52	3.43	3.60	3.38	3.92
Hatton	4.86	5.52	5.02	5.75	5.15	6.24
Centurk	3.90	4.21	4.39	5.33	4.57	4.71
Kavkaz	6.28	7.78	6.09	6.83	6.81	7.59
Vorochilovskaja	4.73	6.75	4.69	6.26	5.21	5.35
Pumafen/Lilifen	5.15	7.08	5.68	6.69	5.43	6.59
GK-Protein	4.46	6.55	5.08	6.14	5.60	5.85
NE 7060	4.08	6.15	4.48	5.19	4.42	5.38
NE 95021	3.03	3.89	3.49	4.00	3.41	3.77

For same variety:

SE = 0.33

LSD = 0.92

HSD = 1.34

For different varieties:

SE = 0.38

LSD = 1.08

HSD = 1.60

(values are for .05 significance level)

Table 13. Hardness means for nitrogen treatments within varieties, Hyslop 1979-1980

Variety	Nitrogen treatment*					
	C	T	S <sup>2</sup>	T+S <sup>2</sup>	S <sup>1</sup> +S <sup>2</sup>	T+S <sup>1</sup> +S <sup>2</sup>
Stephens	64.2	58.6	65.4	56.0	65.8	56.7
Wanser	68.4	68.8	62.7	68.9	73.7	75.9
Hatton	104.1	92.0	95.5	93.9	99.1	100.7
Centurk	77.6	67.6	71.4	74.1	83.4	77.9
Kavkaz	51.4	56.0	53.7	60.0	57.6	59.2
Vorochilovskaja	87.9	80.8	82.6	82.3	83.5	79.5
Pumafen/Lilifen	24.4	25.6	22.6	25.4	25.8	26.2
GK-Protein	67.1	70.6	63.6	68.5	63.7	68.1
NE 7060	67.6	63.1	65.2	63.3	63.5	57.4
NE 95021	65.5	64.3	76.7	65.8	73.5	70.1

For same variety:

SE = 3.1  
 LSD = 8.6  
 HSD = 12.5

For different varieties:

SE = 3.6  
 LSD = 10.1  
 HSD = 14.5

(values are for .05 significance level)

\* values < 50 grade soft.  
 values > 50 grade hard.

Table 14. Protein means for nitrogen treatments within varieties, Hyslop 1979-1980.

Variety	Nitrogen treatment (%)					
	C	T	S <sup>2</sup>	T+S <sup>2</sup>	S <sup>1</sup> +S <sup>2</sup>	T+S <sup>1</sup> +S <sup>2</sup>
Stephens	8.58	9.39	10.11	9.85	10.40	9.70
Wanser	10.90	10.92	12.32	12.25	12.34	12.01
Hatton	9.95	10.90	11.66	12.02	11.45	10.91
Centurk	10.41	11.52	12.67	12.67	12.35	12.32
Kavkaz	10.76	11.51	12.10	12.34	11.86	12.10
Vorochilovskaja	11.62	11.38	13.67	12.78	13.79	13.39
Pumafen/Lilifen	11.60	11.63	13.65	13.12	13.77	13.67
GK-Protein	11.18	12.22	12.77	12.68	13.60	13.31
NE 7060	12.81	13.72	14.34	14.48	14.96	14.66
NE 95021	11.54	11.45	12.65	12.58	13.28	12.71

For same variety:

SE = 0.27  
LSD = 0.74  
HSD = 1.07

For different varieties:

SE = 0.38  
LSD = 1.06  
HSD = 1.51

(values are for .05 significance level)

Table 15. Height means for nitrogen treatments within varieties, Hyslop 1979-1980.

Variety	Nitrogen treatments (cm)					
	C	T	S <sup>2</sup>	T+S <sup>2</sup>	S <sup>1</sup> +S <sup>2</sup>	T+S <sup>1</sup> +S <sup>2</sup>
Stephens	98	109	99	109	98	108
Wanser	141	150	141	146	135	150
Hatton	133	153	135	154	139	146
Centurk	115	141	130	139	123	134
Kavkaz	135	144	133	138	130	136
Vorochilovskaja	106	118	111	115	106	109
Pumafen/Lilifen	130	136	133	139	129	135
GK-Protein	100	105	100	108	95	105
NE 7060	110	118	109	116	111	115
NE 95021	89	96	90	95	86	91

For same variety:

SE = 2  
LSD = 7  
HSD = 10

For different varieties:

SE = 3  
LSD = 8  
HSD = 11

(values are for .05 significance level)

significant differences are found for Wanser, but there are important differences for Stephens.

#### IV. Main Trial, Wasco 1979-1980

The results of analysis of variance for yield, hardness, protein, and height are shown in Table 16. The analysis for shattering is not reported since the CV was 40.5 percent and analysis of covariance proved ineffective. For example, the subplot mean square error for yield was 5873.9, and analysis of covariance with shattering as the covariate reduced this to 5770.7. Unlike the Hyslop location, there were no significant replication effects which reflected the homogeneous nature of the response at Wasco.

As at Hyslop, a highly significant main plot effect was observed. Table 17 lists the varietal means and the standard error for the four factors. The comments mentioned above concerning the value of these observations also apply to the data from Wasco. One additional point that should be made is that the range of values for Wasco was considerably smaller. This is probably due to the nature of the dryland situation with the reduction in available moisture (See Table 1).

Tables 18, 19, 20, 21, and 22 present the nitrogen treatment means and the yield, hardness, protein, and height means for nitrogen treatments within varieties respectively. Wasco data again showed a significant response for nitrogen with the exception of hardness (See Table 16). However, no variety x nitrogen treatment interaction was

Table 16. Analysis of variance for main trial, Wasco 1979-1980.

Source of variation	df	Observed M.S.			
		Yield	Hardness	Protein	Height
Variety	9	299676.0**	10727.10**	20.727**	3504.22**
Replication	3	11949.4	255.23	.334	39.55
Main plot error	27	8303.5	100.79	.651	17.87
Nitrogen treatment	5	59630.4**	72.18	20.378**	84.19**
Comparison 1	1	98496.0**	--	.246	192.60**
Comparison 2	1	9501.8	--	.004	75.63**
Comparison 3	1	105900.4**	--	49.996**	54.19*
Variety x nitrogen treatment	45	7051.1	102.11*	.467	7.47
Subplot error	150	5873.9	67.86	.383	9.20
Coefficient of variation		11.5%	11.1%	7.0%	3.0%

Comparison 1=soil vs foliar, Comparison 2=split vs single, Comparison 3=control vs nitrogen  
 \*,\*\* indicate probability of Type I error at the .05 and .01 level respectively

Table 17. Wasco varietal means 1979-1980.

Variety	Yield (T/ha)	Hardness *	Protein (%)	Height (cm)
Stephens	5.13	42.9	8.50	89
Wanser	3.01	79.1	9.75	117
Hatton	3.40	100.8	8.06	113
Centurk	3.07	76.0	9.76	113
Kavkaz	4.18	59.8	8.70	110
Vorochilovskaja	4.33	84.8	9.52	101
Pumafen/Lilifen	3.71	36.0	10.54	110
GK-Protein	4.25	84.6	9.91	94
NE 7060	3.62	89.5	10.69	103
NE 95021	3.83	87.8	10.67	79
Standard error	0.11	2.0	0.13	1

\* values < 50 grade soft.  
 values > 50 grade hard.

Table 18. Wasco nitrogen treatment means 1979-1980.

Nitrogen treatment	Yield (T/ha)	Hardness *	Protein (%)	Height (cm)
C	3.58	75.8	8.59	102
T	4.05	74.8	9.34	104
S <sup>2</sup>	3.61	74.5	9.41	102
T+S <sup>2</sup>	4.04	71.9	10.45	105
S <sup>1</sup> +S <sup>2</sup>	3.79	74.3	9.45	102
T+S <sup>1</sup> +S <sup>2</sup>	4.05	73.5	10.45	103
Standard error	0.07	1.3	0.10	0.5
LSD#	0.19	3.6	0.27	1
HSD#	0.28	5.2	0.39	2

\* values < 50 grade soft.  
values > 50 grade hard.

# .05 significance level



Table 19. Yield means for nitrogen treatments within varieties,  
Wasco 1979-1980

Variety	(T/ha)					
	Nitrogen treatment					
	C	T	S <sup>2</sup>	T+S <sup>2</sup>	S <sup>1</sup> +S <sup>2</sup>	T+S <sup>1</sup> +S <sup>2</sup>
Stephens	4.70	5.65	5.15	5.35	5.25	4.71
Wanser	2.79	3.34	2.70	2.96	2.98	3.31
Hatton	2.98	3.26	3.24	3.54	3.49	3.86
Centurk	2.19	3.73	2.75	3.18	2.96	3.63
Kavkaz	4.13	4.26	3.83	4.19	4.10	4.58
Vorochilovskaja	4.25	4.52	4.17	4.43	4.05	4.58
Pumafen/Lilifen	3.34	3.71	3.30	4.19	3.92	3.81
GK-Protein	3.95	4.39	4.04	4.45	4.15	4.55
NE 7060	3.57	3.68	3.42	3.99	3.28	3.78
NE 95021	3.92	3.97	3.55	4.14	3.68	3.69

For same variety:

SE = 0.22

LSD = 0.62

HSD = 0.89

For different varieties:

SE = 0.23

LSD = 0.64

HSD = 0.92

(values are for .05 significance level)

Table 20. Hardness means for nitrogen treatments within varieties, Wasco 1979-1980.

Variety	Nitrogen treatment*					
	C	T	S <sup>2</sup>	T+S <sup>2</sup>	S <sup>1</sup> +S <sup>2</sup>	T+S <sup>1</sup> +S <sup>2</sup>
Stephens	39.9	40.7	45.6	40.1	44.4	46.5
Wanser	85.2	79.7	77.5	75.8	82.8	73.6
Hatton	109.9	94.0	101.1	94.5	109.7	96.0
Centurk	69.3	85.2	72.6	66.0	81.2	81.5
Kavkaz	63.5	55.1	59.7	55.1	66.5	59.0
Vorochilovskaja	80.7	90.0	90.4	86.4	78.6	83.0
Pumafen/Lilifen	39.7	35.7	39.0	33.3	35.1	33.5
GK-Protein	83.1	87.2	84.2	82.6	83.4	87.2
NE 7060	94.9	93.7	89.1	93.8	79.9	85.7
NE 95021	92.2	87.1	86.0	91.4	81.3	88.7

For same variety:

SE = 4.1

LSD = 11.5

HSD = 16.6

For different varieties:

SE = 4.0

LSD = 11.3

HSD = 16.2

(values are for .05 significance level)

\* values < 50 grade soft.  
 values > 50 grade hard.

Table 21. Protein means for nitrogen treatments within varieties, Wasco 1979-1980

Variety	Nitrogen treatment (%)					
	C	T	S <sup>2</sup>	T+S <sup>2</sup>	S <sup>1</sup> +S <sup>2</sup>	T+S <sup>1</sup> +S <sup>2</sup>
Stephens	7.76	8.23	8.74	8.72	8.47	9.08
Wanser	8.13	9.68	9.91	11.08	8.71	11.02
Hatton	6.84	8.47	7.78	8.73	7.38	9.14
Centurk	8.65	9.29	9.39	11.00	10.03	10.23
Kavkaz	7.93	8.32	8.33	9.76	8.37	9.50
Vorochilovskaja	8.59	9.26	9.40	10.12	9.39	10.39
Pumafen/Lilifen	9.78	10.33	9.97	11.47	10.40	11.33
GK-Protein	9.19	9.30	9.71	10.62	10.13	10.52
NE 7060	9.68	10.40	10.41	11.34	10.74	11.57
NE 95021	9.36	10.10	10.47	11.68	10.93	11.47

For same variety:

SE = 0.31

LSD = 0.86

HSD = 1.25

For different varieties:

SE = 0.32

LSD = 0.92

HSD = 1.32

(values are for .05 significance level)

Table 22. Height means for nitrogen treatments within varieties,  
Wasco 1979-1980.

Variety	Nitrogen treatments (cm)					
	C	T	S <sup>2</sup>	T+S <sup>2</sup>	S <sup>1</sup> +S <sup>2</sup>	T+S <sup>1</sup> +S <sup>2</sup>
Stephens	89	90	90	90	88	86
Wanser	116	120	116	121	113	116
Hatton	111	114	113	114	113	111
Centurk	111	115	111	115	111	111
Kavkaz	109	111	108	113	109	109
Vorochilovskaja	99	101	100	101	100	104
Pumafen/Lilifen	106	111	109	114	109	113
GK-Protein	94	95	93	96	94	95
NE 7060	104	105	101	105	101	101
NE 95021	79	81	79	80	79	79

For same variety:

SE = 2  
LSD = 4  
HSD = 6

For different varieties:

SE = 2  
LSD = 5  
HSD = 7

(values are for .05 significance level)

significant at Wasco except for hardness. This would indicate a similar varietal response for the other traits in a dryland situation, but hardness apparently has a very different response pattern where the average effect would tend to negate the differences for nitrogen (See Table 20). Using the mean yields for Stephens and the Russian cultivar Vorochilovskaja as an example (Table 19), there are significant treatment effects for Stephens, but there are none for Vorochilovskaja.

The results of the orthogonal contrasts also are important in making conclusions about the observed significant effects for nitrogen treatments. Table 16 shows that different contrasts are significant for each of the traits measured. As at Hyslop, this suggests that the nature of the nitrogen response is not the same for the parameters measured.

#### V. Primary and Secondary Tillers, 1979-1980

The partial analyses of variance for primary and secondary spike type at the Wasco and Hyslop locations are presented in Tables 23 and 24 respectively. The response at each location is different reflecting the contrasting growing conditions at each site for the 1979-1980 crop season.

At Wasco where the environment was optimal for this crop year, there was no response between spike type for either protein or hardness. Therefore, differences were not found for seed from primary and secondary tillers. A lack of response was also recorded for all interaction terms.

Table 23. Partial analysis of variance for spike type, Wasco  
1979-1980.

Source of variation	df	Observed M.S.	
		Protein	Hardness
Spike type	1	.1712	27.337
Spike type x nitrogen treatment	5	.2837	40.503
Spike type x variety	4	.2886	15.086
Spike type x variety x nitrogen treatment	20	.2030	40.892
Sub-subplot error	89	.1692	43.097
Coefficient of variation		3.9%	9.1%

Table 24. Partial analysis of variance for spike type, Hyslop  
1979-1980.

Source of variation	df	Observed M.S.	
		Protein	Hardness
Spike type	1	1.3605*	2056.86**
Spike type x nitrogen treatment	5	.1550	13.17
Spike type x variety	4	3.2460**	131.65**
Spike type x variety x nitrogen treatment	20	.2397	15.95
Sub-subplot error	90	.2488	35.58
Coefficient of variation		4.5%	9.5%

\*,\*\* indicate probability of Type I error at the .05 and .01  
level respectively

Table 25. Protein means for spike type within varieties, Hyslop 1979-1980.

Spike type	V a r i e t y (%)				
	Stephens	Wanser	Vorochi- lovskaja	GK-Protein	NE 95021
Main	9.55	11.40	12.73	12.47	12.40
Secondary	10.25	10.91	13.19	12.86	12.09

For same variety: SE = 0.10, LSD = 0.28 (.05 significance level)

Table 26. Hardness means for spike type within varieties, Hyslop 1979-1980.

Spike type	V a r i e t y *				
	Stephens	Wanser	Vorochi- lovskaja	GK-Protein	NE 95021
Main	56.1	64.5	76.4	64.1	66.9
Secondary	55.5	59.7	69.7	55.3	58.5

For same variety: SE = 1.2, LSD = 3.4 (.05 significance level)

\* values below 50 grade soft, values exceeding 50 grade hard

However, at the Hyslop site there were significant effects for spike type with both protein and hardness measurements. Also, for each trait a significant spike type x variety interaction was observed. The protein and hardness means for spike type within varieties are listed in Tables 25 and 26 respectively. It can be seen that the differences in levels of these two traits change substantially for different cultivars. For example, Stephens has secondary tillers that are significantly higher in protein, whereas Wanser produces secondary tillers significantly lower in protein.

At both locations, there were no significant effects for any interaction terms including nitrogen treatment. This observation confirms the result of the 1978-1979 data which indicated no preferential uptake of nitrogen by different types of seed.

## VI. Greenhouse Experiment

The analysis of variance for recovery of N<sub>15</sub> in the greenhouse experiment is presented in Table 27. It should be stressed that the two growth chambers utilized in this experiment were manufactured in the same year by the same company, and they were monitored several times daily to insure that no fluctuations beyond the limits mentioned in the methods section occurred in either chamber throughout the period following foliar application. Therefore, a 2x2x2 factorial analysis was employed with one degree of freedom associated with each treatment effect and interaction.



Table 27. Analysis of variance for recovery of N15

Source of variation	df	Observed MS
Light	1	5565.13*
Variety	1	13612.50**
Fertilizer	1	2628.13
Light x Variety	1	2.00
Light x Fertilizer	1	171.13
Variety x Fertilizer	1	32.00
Light x Variety x Fertilizer	1	3960.50
Error	21	1189.44
Coefficient of Variation		15.7%

\*,\*\* indicate the probability of Type I error at the .05 and .01 level respectively

A highly significant effect for varieties was found. The variety Stephens had an average recovery of 48.1 percent, and Centurk's average recovery rate was 39.9 percent. Therefore, under optimal conditions for nitrogen uptake, a difference in cultivars was evident. Also, a significant response for light duration was observed with a nitrogen recovery of 41.4 and 46.6 percent for the 12 and 18 hour photoperiod respectively. None of the other sources of variation proved significant. The effect of varying the fertility level after vernalization in particular did not give a significant response.

The recovery rate did vary considerably for all samples. The lowest recovery rate of 29.2 percent was observed for one sample of Centurk under 12 hours of light, and the highest recovery rate of 61.4 percent was found for one plant of Stephens under 18 hours of light. However, the accuracy of the recovery data was reflected by the close agreement of all control plants. The average N15 content of controls was 0.376 percent in both chambers (0.366 percent is the standard for atmospheric nitrogen). Also, two groups of controls were used; one group was from plants in the same pot with a treated plant, and the other was from pots that did not contain a treated plant. No difference was found between the two groups which indicated the success of the method of foliar application for affecting only the designated plant. Finally, the controls confirmed the accuracy of the data since they did not show evidence of contamination from the ammonium distillation and tube digestion.

## DISCUSSION

The objectives of this study were (1) to decide what kind of response could be expected from foliar fertilization for protein, yield, and hardness and (2) to determine how observed responses might be manifested by (a) examining immature spike samples taken over the time of foliar applications, (b) comparing the protein content of grain from primary and secondary tillers and of seeds produced at different positions within the spikelet, and (c) determining the uptake of urea from foliar applications by using N15 treated plants that were in a controlled environment. The results obtained from the two years of this study permit preliminary conclusions to be drawn concerning these objectives. Hopefully, they will also address the broader issue of what is feasible in terms of crop management practices in order to permit expanded hard red winter wheat production in the Pacific Northwest.

Protein response was the principle measurement because of the necessity of achieving sufficient protein levels for any potential hard red wheat cultivar. The two locations in 1978-1979 and the site at Wasco in 1979-1980 showed a lack of response to a single spray applied after anthesis (See Tables 5, 6, and 18) in comparison with an adequate topdressing program. However, in comparing the effect of a single spray and a topdressing application, T vs. S2, at Hyslop in 1979-1980 (Table 11), a significant response was observed. This finding would seem to reflect the conflicting reports in the literature on the advantage of a single foliar spray for elevating protein (Reeves, 1954; Juarez and

Swanson, 1956; Finney et al, 1957; Jain et al, 1971). It appears that protein response is highly dependent on particular environments of different cropping situations. The importance of environment for grain protein content has been emphasized by other researchers (Kramer, 1979).

Also, if the comparison is made in terms of the difference from the control, then highly significant increases can be reported for this study at both locations in 1979-1980 when more accurate testing was accomplished. Protein content was increased by over two percent for a number of cultivars after foliar application of urea. For example, Vorochilovskaja at Hyslop in 1979-1980 had protein levels of 11.62 percent in the control and 13.79 percent in the S1+S2 treatment. These increases would be in line with the results reported by Finney et al (1957) and Pushman and Bingham (1976).

The alternative of combining foliar and topdressing treatments seems very promising. The comparison of all topdressing vs. half foliar and half topdressed was highly significant at Hyslop in 1979-1980 (Table 9), and the significant difference between treatments T and T+S2 at Wasco in 1979-1980 (Table 18) points out this possibility. Few researchers to date have examined combinations of foliar and soil applications, but the results of this study showed that this is probably the more interesting prospect for practical utilization of foliar feeding.

Any protein response has to be considered in light of the effect on yield. Yield was not increased by foliar sprays at Wasco in 1979-1980 with treatments T, T+S2, and T+S1+S2 producing 4.05, 4.04, and 4.05 T/ha respectively (Table 18). However, although the yields were equivalent,

the T+S2 treatment had an average protein increase of 1.11 percent. The 1979-1980 data at Hyslop was in close agreement with yield differences of the same three treatments mentioned above being non-significant, but T+S2 produced grain with 1.01 percent higher protein content (Table 11). Delaying nitrogen applications until anthesis seems to be detrimental to yield when all additional nitrogen is applied through the foliage. Both the Hyslop and Wasco locations in 1979-1980 showed decreases in yield when comparing either S2 or S1+S2 with any of the topdressing treatments. This is apparent from the highly significant contrast for comparison one at Wasco. A lack of response to foliar sprays has been frequently reported for yield when application is made after the heading stage (Reeves, 1954; Finney et al, 1957; Pushman and Bingham, 1976). This has been the case for other forms of nitrogen applied after this point of plant development as well (Thorne, 1962; Langer and Liew, 1973).

Varietal response might be of interest as discussed in the results section. If it is possible to obtain yields of 8.19 T/ha with the cultivar Stephens from the T+S1+S2 treatment compared to the topdressed treatment which yielded 7.27 T/ha, then there could be an advantage for foliar applications. In any case, the general conclusion would be that yield was not enhanced by urea sprays.

Hardness results show little advantage for any nitrogen applications. The means of all treatments are similar, and no treatments exceeded the control at either location in 1979-1980. This is contrary to the conclusions of Sadaphal and Das (1966) when they mentioned there was a decrease in yellowberry for the urea spray plots.

None of the other major studies of foliar urea applications have looked into any response for hardness. A significant effect for comparison two at Hyslop was obtained which contrasted single and split applications. However, the means for these treatments do not differ enough to substantially change the milling characteristics.

The protein analysis of immature spikes seems to be in general agreement with the results of the main trials. Other than the research of Singh and Seth (1978), there have been no reports of the effect of urea sprays on vegetative plant parts. Their experiment showed lower nitrogen content of vegetative parts at the end of the season, but higher grain protein content. This indicates a higher rate of translocation of nitrogen for foliar sprays which is suggested by the results of this study as well.

The importance of sampling at the same growth stage is seen by the more accurate measurement of treatment effects at Hyslop. At Hyslop, the highest protein treatment at three weeks post-anthesis was also the highest at maturity. In addition, the high response at harvest of the group of treatments S2, T+S2, S1+S2, and T+S1+S2 were the same as those identified during the flowering period. Although the differences at Wasco were smaller, the highest treatment at three weeks after anthesis was again the highest treatment at maturity for protein determination. Also, the ranking was the same for five of the six treatments with only the S1+S2 treatment changing position by harvest. These results suggest that nitrogen treatment effects could possibly be measured after anthesis since differences that show up early were corresponding to those identified later.

Results of the two years of data on tiller type are somewhat contradictory because each location-year allows different conclusions. For example, Wasco was the only site that did not have a significant mean square value for spike type, and Hyslop 1979-1980 was the only experiment to have a significant interaction for spike type x variety. Since there were no interaction terms including nitrogen treatment for Hyslop 1978-1979 and 1979-1980 or for Wasco 1979-1980, it is possible to conclude that nitrogen applications did not change the difference in protein or hardness levels. Both years at Hyslop, there were significant effects for spike type which confirms other work (McNeal and Davis, 1966). Also, the significant variety x spike type interactions for 1979-1980 confirm that the difference between main and secondary tillers is not always in the same direction. Wasco had no significant effects possibly as a consequence of the exceptional growing season that occurred. Apparently, the type of environment greatly influences whether or not any differences will be manifested for tillers that develop at different stages as seen by the lack of consistency for the three trials.

The analysis of central and lateral seeds within the spike for the 1978-1979 crop season also showed no preferential nitrogen uptake. Although one published report did suggest that differences in uptake for central and lateral seeds does exist (Ries et al, 1976), the significance of these results is difficult to assess. Also, as noted with the analysis for primary and secondary tillers, the particular environment could greatly influence the results.

The data on N15 recovery for the greenhouse experiment was

strikingly contrary to the only other foliar study with labelled urea in the literature. However, the study of Alkier et al (1972) did not sufficiently explain the methods of analysis to make an adequate comparison. Also, these researchers found one percent uptake of foliar applied urea vs. 30 percent uptake for soil applied urea, but the protein response was equivalent for the two treatments. The explanation offered was that the foliar spray could have washed off the foliage and then have been absorbed by the roots. This does not seem possible given the immediate metabolism of urea found by other researchers (R. Gill, personal communication).

An uptake of 39.9 percent nitrogen for Centurk and 48.1 percent for Stephens was reported in this study under optimal conditions. Because each plant had only one tiller and all liquid urea was painted on the foliage, these values can be considered as maximum values. Under field conditions, the uptake could be reduced, but in any case, these results confirm that the response for protein noted with the yield trial data can be explained by actual uptake of the applied nitrogen rather than solely by some secondary effect on the nitrogen metabolic pathway.

Also, the results of the greenhouse experiment support the conclusions of Van Vuurde and Tonneyck (1978). They found that different levels of light intensity did influence the response for dry matter accumulation and that soil fertility levels were not important for assimilation. A significant response for light duration and no significant response for soil fertility was found in this study. This supports the hypothesis of Van Vuurde and Tonneyck (1978) that the rate of photosynthesis at the time of foliar application will greatly affect



the efficiency of utilization. It might be more effective to apply urea sprays during periods when less cloud cover is present.

## CONCLUSIONS

The purpose of this study was to measure the effectiveness of foliar urea application and to explain how any potential response could occur. Eleven cultivars of winter wheat were planted in yield trials for four location-years, nitrogen treatments were applied, and measurements were made for protein, yield, and hardness. In addition, the protein and/or hardness values after different nitrogen applications were recorded for (1) immature spikes at three dates during the period of flowering, (2) seed from primary and secondary tillers, and (3) central and lateral seeds within the spikelet. Finally, a greenhouse experiment was conducted to measure the uptake potential of foliar urea treatment under differing light duration and soil fertility levels and for two different cultivars.

The following conclusions were made upon completion of this study:

(1) Foliar application increased protein by more than two percent above the control and over one percent above the standard topdressing treatment. For example, cultivar NE 7060 at the Hyslop site in 1979-1980 produced grain with a protein content of 12.81 percent for the control, 13.72 percent for the topdressing treatment, and 14.96 percent for a foliar application. Split applications did not significantly alter these results.

(2) Generally, no increase in yield occurred over the control with foliar applications, and yield was significantly below the standard topdressing treatment. However, replacing half of the topdressed

nitrogen with a foliar spray or adding an additional increment of nitrogen to a standard topdressing gave equal yields while increasing protein.

(3) Foliar applications did not significantly alter kernel hardness.

(4) Significant cultivar x nitrogen treatment interactions were found, and different wheat cultivars did not have the same response to foliar fertilization for protein, yield, and hardness.

(5) Following urea sprays applied at the heading stage, differences in nitrogen treatments were distinguished by three weeks after anthesis at both locations. The ranking of treatments from immature spike analysis corresponded to that observed at harvest.

(6) Nitrogen treatments did not change the protein or hardness levels disproportionately for seed from primary and secondary tillers or for seed from central and lateral florets within the spike.

(7) A potential uptake of between 29.2 and 61.4 percent of foliar applications of urea was determined with N15 labelling in a greenhouse experiment. A significant effect for light duration was observed with recovery values of 41.4 and 46.6 percent for 12 and 18 hours of light respectively. The cultivars Stephens and Centurk had a significant difference in uptake of 48.1 and 39.9 percent recovery respectively.

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## APPENDICES

## APPENDIX 1: Variety Descriptions

Stephens: An awned, standard height, soft white winter wheat from the cross Nord Desprez/Pullman Selection 101; released in 1977 by Oregon State; with medium to high tillering levels, moderate head fertility, a high seed weight; high yielding for Pacific Northwest conditions.

Wanser: An awned, tall, hard red winter wheat from the 1952 cross Burt/Itana; released in 1965 by Washington; with brown glumes, an oblong, dense spike, erect to inclined; susceptible to all rusts; standard hard red winter variety for Pacific Northwest.

Centurk: An awned, medium to tall, hard red winter wheat from the 1959 cross Kenya 58/2/Newthatch/3/Hope/2\*Turkey/4/Cheyenne/5/Parker; released by the Nebraska AES and ARS in 1971; high yielding over a range of environments; excellent milling and baking qualities; with white glumes, an oblong to fusiform spike, erect to inclined.

Kavkaz: An awnless, medium to tall, hard red winter wheat from the cross Lutescens 314-h-147/Bezostaja 1; released in 1971 by Russia; large, cylindrical, white spikes, poor tillering, with large seed of good milling and baking qualities.

Hatton: An awned, medium to tall, hard red winter wheat with grain yields higher than Wanser, from the cross 2\*McCall/Koeltz; released in 1979 by Washington; with similar plant and head type as Wanser, but more

erect.

GK-Protein: An awned, standard height, hard red winter wheat from the ezostaja 1/Firello; released in 1978 by Hungary; with white chaff; two to three days earlier than Bezostaja; good milling and baking quality.

Vorochilovskaja: An awned, standard to medium height, hard red winter wheat, from the cross Kanred/Fulcaster; a recent release from Russia; with white glumes, early maturity, good resistance to strip rust, moderate head fertility.

Pumafen/Lilifen: An awned, medium height, soft red facultative wheat, crossed in 1969 in Davis, California, selected as an F6 by Oregon State, and retained as a parent line; with brown chaff, good disease resistance; good head fertility.

Centurk/Ciano: An awned, medium to tall, hard red winter wheat; a winter x spring cross made by CIMMYT in the late 1960's and retained as a parent line; with white chaff, susceptible to strip rust; high in protein.

NE 7060: An awned, medium to tall, hard red winter wheat, from the 1970 cross Favorite/5/Cirpiz/4/Jang Kwang/2/Atlas 66/Comanche/3/Velvet made at Nebraska for inclusion in the protein improvement germplasm; white chaff, moderate winter hardiness, susceptible to strip rust.

NE 95021: An awned, short, hard red winter wheat, from the 1972 cross Atlas 66/Nap Hal/2/TX62A2522-1-4 made at Nebraska for inclusion in the protein improvement germplasm; white chaff, fair winter hardiness, moderately late, very susceptible to strip rust, high in protein.

## APPENDIX 2: Techniques for Protein Determination

Micro-Kjeldahl Procedure

1. Samples were ground to pass a 40 mesh screen after having been dried for three days at 44 degrees Centigrade. After grinding, the samples were placed in a desiccator container and left overnight.
2. 100 mg of sample were weighed on a Zig Zig cigarette paper with a Mettler balance and then placed in a graduated Folin-Wu tube. A 108 Lee powder measuring scoop of Kjeldahl salt-catalyst was then added to each tube along with four ml of concentrated sulfuric acid. The salt catalyst consisted of potassium sulfate, cupric sulfate, and selenium at a ratio of 100:10:1 by weight.
3. A small glass funnel (25 mm in diameter) was placed in the mouth of each tube, and the tubes were transferred to an aluminum heating block with asbestos shields that was preheated to 300 degrees Centigrade.
4. Tubes were removed from the heating block approximately one hour past the time of clearing and then allowed to cool. All tubes were then diluted to 50 ml volume, stoppered, mixed by inversion, and topped to 50 ml.
5. The Bremner and Edwards method for steam distillation was used when 10 ml of each sample solution was made alkaline with three ml of 10 N NaOH.
6. Five ml of standard Boric Acid-Indicator Solution were placed in a

50 ml Erlenmeyer flask, and the distillate was added to the 35 ml mark. The indicator solution was made by adding 40 g of reagent grade boric acid in 1400 ml of distilled water to 400 ml of 95% ethanol and 40 ml of Mixed Indicator Solution (1000 ml 95% ethanol, 0.66 g of bromocresol green, and 0.33 g of methyl red). 0.05 N NaOH was added to the Boric Acid-Indicator Solution until a color change from pink to pale green was just detectable when one ml of the solution was treated with one ml of distilled water.

7. Distilled water was introduced into the distiller and allowed to pass through all glassware between distillation of each sample. The 35 ml of distillate and indicator were then stoppered and later titrated back to acid with 0.01 N HCL. This volume was recorded in ml and the percent protein was calculated by the following formula:

$$\% \text{ protein} = \frac{5.7 \times 70.035(\text{ml of sample} - \text{ml of blank})}{(\text{total sample wt} - \text{cigarette paper wt})}$$

#### InfraAlyzer 400 Calibration

1. A set of standards were analyzed for percent protein with the micro-Kjeldahl procedure described above.
2. These standards were then run through the InfraAlyzer, and F values were entered in the machine until the readings were in agreement with the micro-Kjeldahl values. F values are a series of constants stored in the machine that are used to determine percent constituent by solving the equation:

$$\% \text{ constituent} = F_1 \log R_1 + F_2 \log R_2 + \dots + F_n \log R_n + F_0$$

where  $F$  is a constant from regression,  $R$  is the near infrared reflectance at a given wavelength, and  $F_0$  is the intercept value.

3. Occasional adjustment for bias was performed by running a random sample of entries with the micro-Kjeldahl method and comparing the values for the two methods.  $F$  values used for this study were as follows: (1) soft wheat---- $F_0 = 14.37$ ,  $F_{10} = 523.9$ ,  $F_{14} = -394$ ,  $F_{20} = -170.1$ ; (2) hard wheat---- $F_0 = 14.1$  and the rest were identical to those of the soft wheat program. All the other possible  $F$  values were zero.