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PROGRESS IN BREEDING FOR PROTEIN IN WHEAT: STATUS OF PROTEIN QUALITY IMPROVEMENTS IN WHEAT

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16. Abstracts

~~This~~ paper presents the current status of protein quality improvement in wheat. The author discusses the progress being made on the following topics: genetic variation found for protein and lysine in wheat; sources of high protein and high lysine in the breeding program; identification of best recipient genotypes; current hybrid populations of special interest; biological evaluations to identify improved protein sources and plant physiology studies.

The nitrate reductase step in the metabolism of nitrogen is thought to be the rate limiting reaction for the eventual synthesis of protein. Lines and varieties are being identified for nitrate reductase activity. All high protein lines to date have been found to be high in nitrate reductase activity. Fortunately, a recent break through now permits the quantitative measurement of this rate-limiting enzyme in live tissue. In addition, factors can be accurately identified affecting the process of nitrate reduction. Leaf protease levels are being studied to identify varieties capable of translocating larger quantities of amino nitrogen to the grain for protein systhesis.

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PROGRESS IN BREEDING FOR PROTEIN IN WHEAT

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Status of Protein Quality Improvement in Wheat

Paul J. Mattern

University of Nebraska

To be presented Thursday, December 7, 1972,

at

International Symposium on Protein Quality in Maize

El Batan, Mexico

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STATUS OF PROTEIN QUALITY IMPROVEMENT IN WHEAT

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J. W. Schmidt, L. Klepper, and R. L. Ulmer

SUMMARY

Research on the improvement of wheat protein content has been conducted at the University of Nebraska since 1954. The research is cooperative between the Agricultural Research Service, U. S. Department of Agriculture, and the Agronomy Department, University of Nebraska at Lincoln. In 1966 the Agency for International Development (AID) initiated a project at Nebraska for the "Improvement of Nutritional Quality of Wheat through Increased Protein and Improved Amino Acid Balance." The initial thrust was to screen 16,000 entries in the USDA World Wheat Collection for lysine and protein differences and to cross identified material to good agronomic wheats. In order to identify winter wheat recipient genotypes for possible new genes for protein and lysine an International Winter Wheat Performance Nursery (IWWPN) was developed as a part of the AID-funded Nebraska protein research effort. Lines identified from laboratory screening activities with improved protein and lysine potential have been crossed to best possible recipient parental types.

GENETIC VARIATION FOUND FOR PROTEIN AND LYSINE IN WHEAT

Laboratory analyses of 12,500 common wheats, from the USDA World Collection of Wheats, ranged in protein content from 7 to 22 percent (Figure 1). The mean was 12.9 percent. The genetic component of this variation is estimated at 4 or 5 percentage points.

The range in lysine/protein content on these same samples was 2.2 to 4.2 percent (Figure 2). The genetic component of lysine appears to be 0.5 percent. It is this portion which should be useful for breeding for improved lysine content.

Protein content and lysine level in wheat are negatively correlated. In an analysis of 4100 wheats from the World Collection as protein increased from 10 to 20 percent mean lysine/protein dropped from 3.2 to 2.7 percent (Figure 3). Only 18 percent of the variation in lysine was associated with variation in protein content. However, there was a strong positive relationship between protein content and lysine on a sample basis. Mean lysine values on a dry weight basis increased from 0.33 to 0.53 percent as the grain protein increased from 10 to 20 percent.

As stated above, environmentally induced protein variation is negatively correlated with lysine. However, we have found genetically high protein wheat to be equal to or higher than lysine/protein content of ordinary wheats in the same environment. Of even greater interest is the observation that the negative correlation between protein and lysine appears to become non-significa at higher levels of protein.

SOURCES OF HIGH PROTEIN AND HIGH LYSINE IN THE BREEDING PROGRAM

Atlas 66, a soft red winter wheat variety, has been utilized extensively as a source of high protein in our breeding program. This genetic source originally came from the South American variety Frondoso. Additional genetic sources of higher protein in wheat have also been identified in Table 1. Those of outstanding promise are NB 542437, SD 69103, C.I. 7337, C.I. 6225, and P.I. 176217. The last named, Nap Hal, is of particular interest because it is also the most promising genetic source of high lysine discovered in our screening efforts.

IDENTIFICATION OF BEST RECIPIENT GENOTYPES

Our need to identify good agronomic winter wheat genotypes with wide adaptation led to the establishment of the International Winter Wheat Performance Nursery (IWWPN) in 1969.

The purposes for the IWWPN were to:

1. Test the adaptation of winter wheat varieties under a range of latitudes, daylengths, fertility conditions, water management, and disease complexes.

2. Identify superior winter varieties to serve as recipient genotypes for high protein and high lysine genes.

3. Test the degree of expression and stability of the high protein and high lysine traits in an array of environments.

These purposes are achieved best by continued testing with new varieties from the various winter wheat projects of the world. Therefore, candidate varieties are and will be solicited for future testing.

In accordance with the stated purposes of this nursery, three types of candidate varieties are especially desired. These are:

1. New commercial or promising experimental varieties with desirable combinations of high yield potential, acceptable winterhardiness, straw strength, disease and insect resistance, and protein quality and quantity.

2. Lines with improved nutritional value from higher protein quantity and/or quality even though deficient in some of the traits listed above.

3. Germplasm lines with especially valuable traits useful in breeding programs.

There has been excellent phenotypic expression of the high protein trait in the 1st and 2nd International Winter Wheat Performance Nurseries. Atlas 66 NB 67730 and Purdue 4930A6-28-2-1, all possessing similar genes for high grain protein, were consistently and significantly higher in protein than other varieties at most nursery sites. Atlas 66 and NB 67730 are compared with Bezostaia, Lancer, and Gaines at three test sites in Table 2.

The protein advantage of the high protein varieties over other varieties in the nursery persisted at sites at which nursery mean protein and yield levels were high as well as where they were low.

Table 2. Phenotypic expression of the high protein trait at 3 sites of the International Winter Wheat Performance Nursery in 1970.

Variety	Grain protein content at:		
	Stillwater, Oklahoma	Martonvasar, Hungary	Cambridge, England
	%	%	%
Nursery $\bar{x}$ protein	17.8	15.8	12.5
Bezostaia	16.5	14.3	12.3
Lancer	16.2	14.6	12.1
Gaines	16.5	14.1	10.6
Atlas 66	20.6	19.4	13.5
NB 67730	20.9	18.4	14.2
Nursery $\bar{x}$ yield (q/ha)	25.4	32.7	31.7

From Proceedings 1st International Winter Wheat Conference, Ankara, Turkey, June 5 - 10, 1972.

The IWWPN has identified cultivars which have superior yield potential over a wide range of winter wheat growing environments. During 1969-1971 the cultivars Bezostaia, Timwin and Blueboy have ranked high. In 1971 NS 611 (Sava), a new entry from Yugoslavia, ranked first (Table 3).

Cultivars identified with wide adaptation are used in crosses with lines possessing improved protein and/or lysine content.

The IWWPN, grown at 23 sites in 16 countries in 1969, has increased in size to 44 sites in 27 countries in 1972.

The IWWPN offers a unique opportunity as a source of seed samples to determine the stability of genes for high protein and lysine over a wide array of environmental conditions. We have never failed to receive samples from any nursery which was harvested during the past four years. Only four nursery sites were not analyzed during this time. Two had sustained insect damage, one was badly shriveled and one had been treated with a chemical which interfered with the amino acid determination.

CURRENT HYBRID POPULATIONS OF SPECIAL INTEREST

Atlas 66 was crossed with Nap Hal to determine if the genes associated with the high protein trait of these varieties were different. Also there was interest to check the heritability of the higher lysine content of Nap Hal. Protein and lysine data were obtained from the F_2 progeny bulk rows (F_3 and F_4 generations) grown at Yuma, Arizona, in 1971 and 1972, respectively. The frequency distributions for protein appear in Figures 4 and 5.

The parent varieties produced grain with similar protein content. This was considerably higher than the protein of Lerma Rojo 64 and Triumph 64 grown in the same experiments. There was distinct transgressive segregation for both high and low protein among the F_2 progeny rows for both years which we interpret as evidence for different protein genes in the parent varieties. Therefore, it appears possible to increase the protein of wheat beyond the Atlas 66 level.

Nap Hal parent rows were significantly higher in lysine content, adjusted to a common protein level, than the Atlas 66 parent rows, Lerma Rojo 64 and Triumph 64 (Figures 6 and 7). There was evidence of segregation for lysine level in the F_2 progeny rows with apparent recovery of parental levels of lysine in some rows during both test years.

Nap Hal was also crossed to C.I. 13449 (a Norin 10-Brevor Sel 14 derived line) thought to be higher in lysine. The frequency distribution for F_2 progeny bulk rows (F_3 generation) for protein and adjusted lysine are shown in Figures 8 and 9, respectively. The protein values for the progeny fell between the parental values as one would expect. The lysine values (Figure 9) for both parents were similar and higher than normal varieties Triumph 64 and Atlas 66. Of particular interest was the apparent transgressive segregation taking place to produce progeny with lysine values higher than the parents.

It appears that improvement of both protein and lysine content will be possil with the genetic sources identified to date.

BIOLOGICAL EVALUATIONS TO IDENTIFY IMPROVED PROTEIN SOURCES

A small animal laboratory has been established in the Department of Foods and Nutrition in cooperation with the Department of Agronomy at the University of Nebraska. The objectives of the laboratory are as follows:

1. To set up a mass testing operation for biological evaluation of various wheat materials as sources of protein.
2. To attempt to isolate and define causes of variability among different wheats in relation to protein value.
3. To lay a foundation for establishment of general guidelines for predicting the value of wheat materials as sources of protein.

The results of mouse feeding trials, involving high protein and/or lysin wheat varieties as a protein source, will be correlated with data from human feeding trials using both adults and adolescents.

Correlative studies on high protein lines have been completed for the PPDR Index *in vitro* laboratory method and mouse feeding. The high protein lines were derived from Atlas 66 and were found to have similar feeding potential.

PLANT PHYSIOLOGY STUDIES

We follow the team approach in our research effort to improve the nutritional value of wheat protein. Research in plant physiology is an integral part of our work effort. The nitrate reductase step in the metabolism of nitrogen is thought to be the rate limiting reaction for the eventual synthesis of protein. Presently we are screening and identifying lines and varieties for nitrate reductase activity. All high protein lines to date have been found to be high in nitrate reductase activity. Fortunately, a recent breakthrough now permits the quantitative measurement

of this rate-limiting enzyme in live leaf tissue. In addition, we can now accurately identify factors affecting the process of nitrate reduction. Leaf protease levels are also thought to be important and are being studied to identify varieties capable of translocating larger quantities of amino nitrogen to the grain for protein synthesis. We are also using an *in vivo* method to evaluate nitrogen fertilization practices for maximum and economical increases of yield and grain protein.

Table 1. Useful germplasm for the nutritional improvement of wheat.

Variety	C.I. or Sel. number	Source	Growth habit	Useful trait
Atlas 50	12534	Beltsville	I	High protein
Atlas 66	12561	"	I	" "
Atlas-derived lines	---	Nebraska	W	" "
Aniversario	12578	Argentina	S	" "
Aniversario-derived lines	---	Nebraska	W	" "
Male Fertility Restorer	NB542437	"	W	" "
Hume^2 x Nb^4 - Agrus-Tc^7	SD69103	So. Dakota	W	" "
Nap Hal	176217	India	S	High protein, high lysine
April Bearded	7337	England	S	" " " "
Hybrid English	6225	"	W	" " " "
Pearl	3285	Sweden	S	Probable high lysine
22A	5484	USSR	S	" " "
Fultz x Hungarian	11849	USA	W	" " "
Fultz-Hungarian x Minturki-Fultz	12756	"	W	" " "
Norin 10-Brevor Sel. 14-derived line	13447	Washington	W	" " "
" "	13449	"	W	" " "

Taken from Proceedings 1st International Winter Wheat Conference, Ankara, Turkey, June 5 - 10, 1972. Nebraska Agricultural Experiment Station MP-28, 1972.

Table 3. Top ranking cultivars for yield from International Winter Wheat Performance Nurseries grown in 1969, 1970, and 1971.

Cultivar	Year	No. of Sites	Grain Yield (q/ha)	Rank
Bezostaia	1969	16	45.2	1
"	1970	31	39.5	1
"	1971	26	40.4	2
Timwin	1969	16	39.9	5
"	1970	31	35.4	2
"	1971	26	39.8	4
Blueboy	1969	16	43.5	2
"	1970	31	29.0	18*
"	1971	26	40.3	3
NS 611 (Sava)	1971	26	41.5	1

* Seed with low germination.

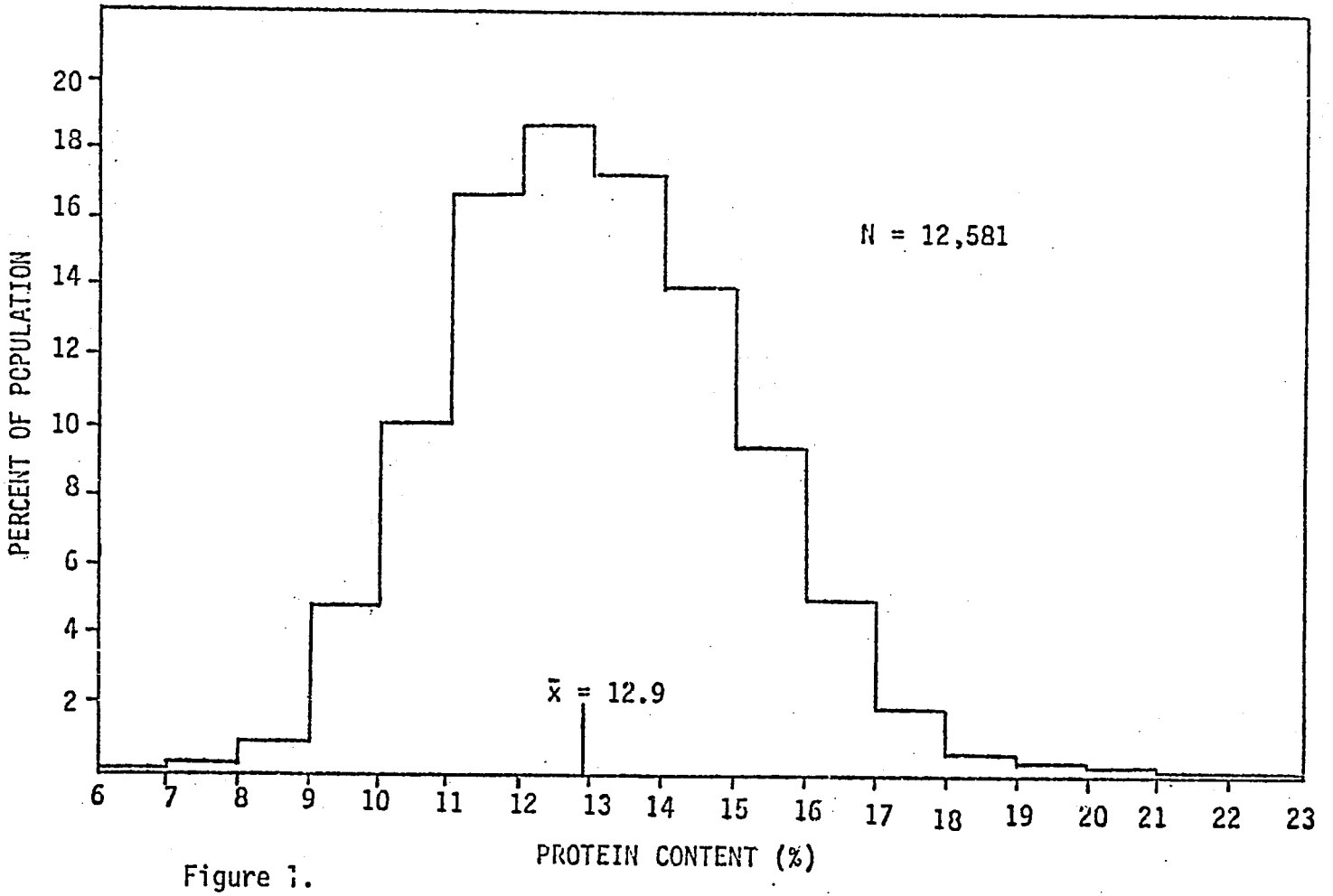


Figure 1.
Frequency distribution for grain protein content among 12,581 wheats in the USDA World Collection.

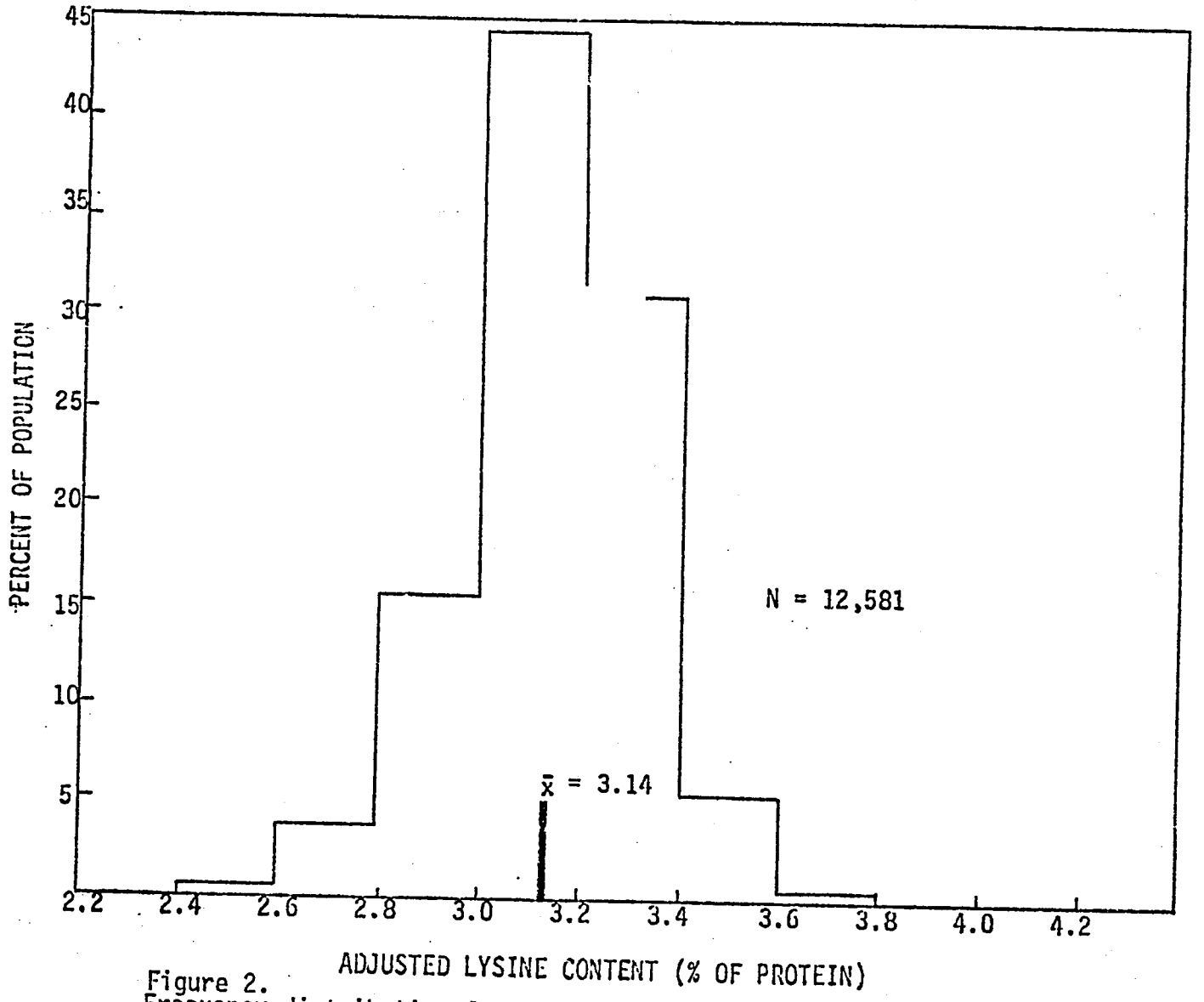
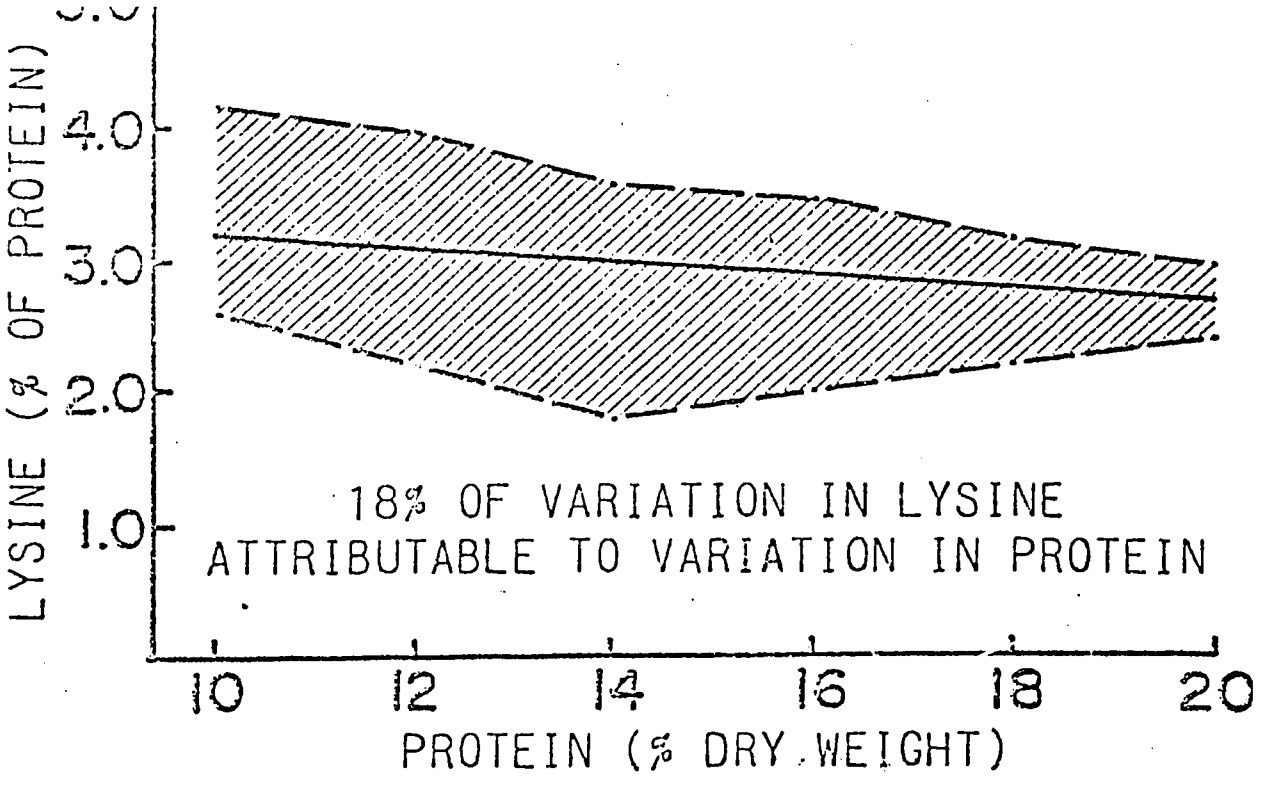


Figure 2.
Frequency distribution for lysine adjusted to a common protein level among 12,581 wheats in the USDA World Collection.



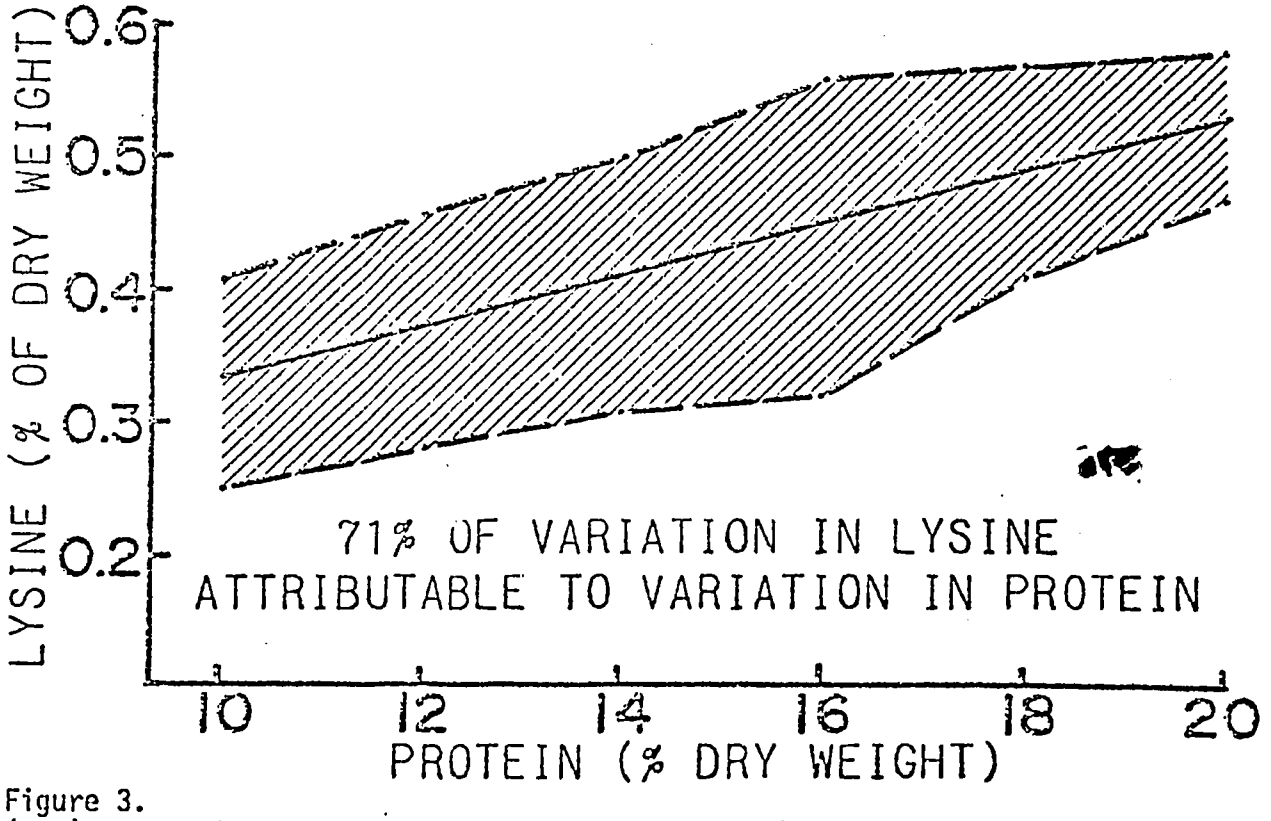


Figure 3.
(Top) Regression of lysine/protein on protein. (Bottom) Regression of lysine/dr
weight on protein. These regressions based on 4100 wheats from the World Collec
tion. Shaded area indicates dispersal of actual values about the regression lir

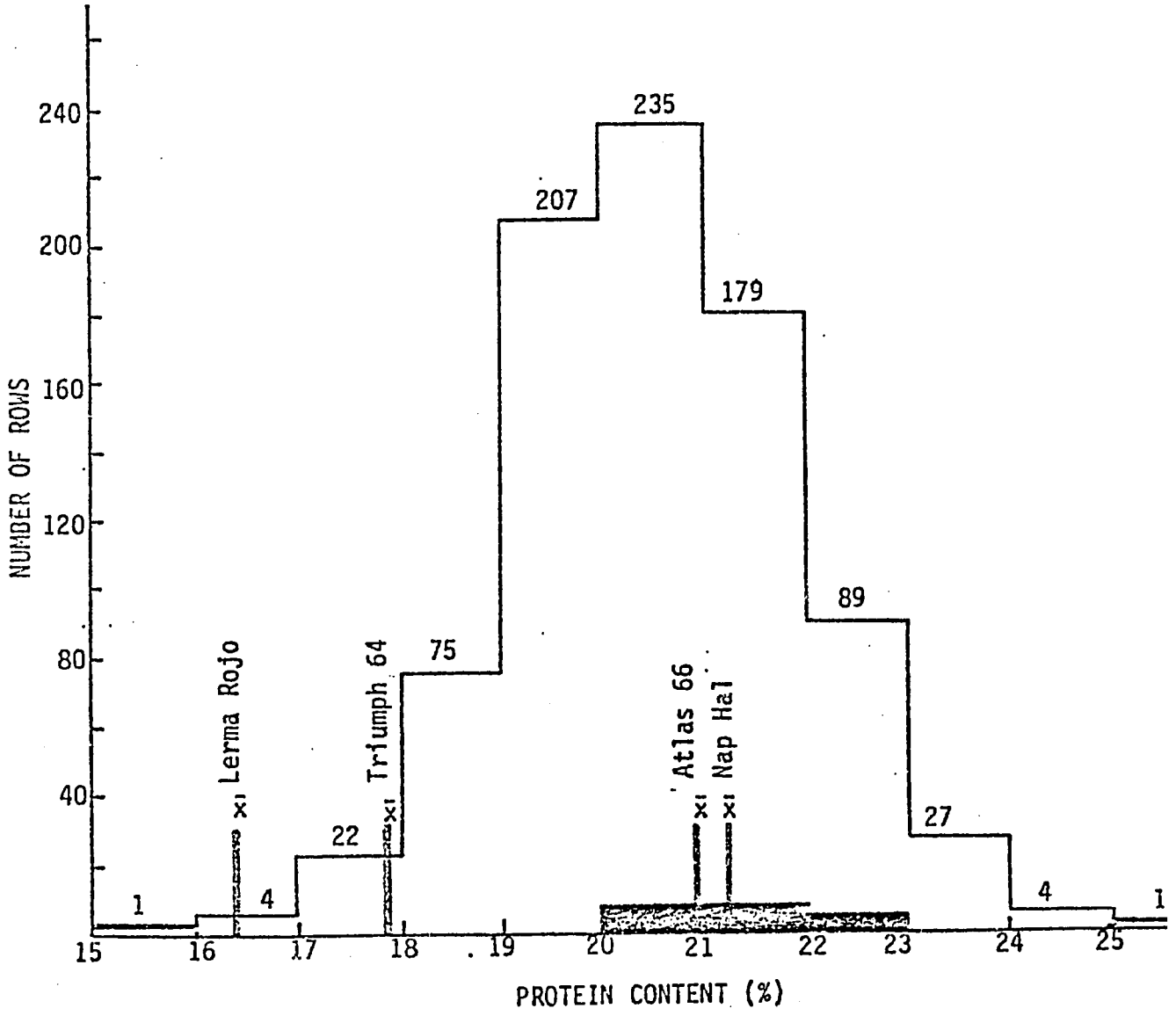


Figure 4.
Protein frequency distribution for F_2 progeny bulk rows (F_3 generation) from a Nap Hal x Atlas 66 cross grown in Yuma, Arizona in 1971.

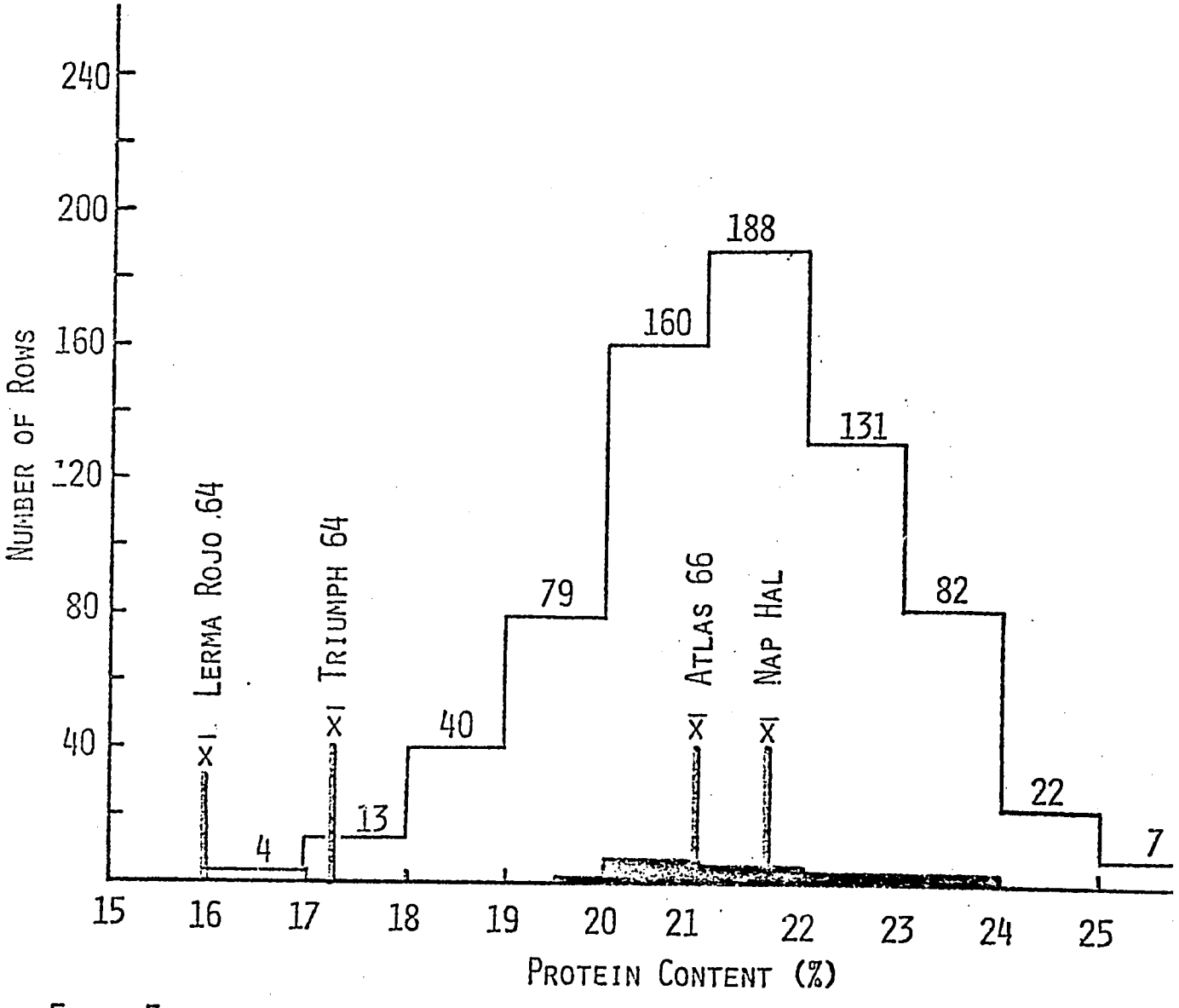


Figure 5.
Protein frequency distribution for F_2 progeny bulk rows (F_4 generati from a Nap Hal x Atlas 66 cross grown at Yuma, Arizona in 1972.

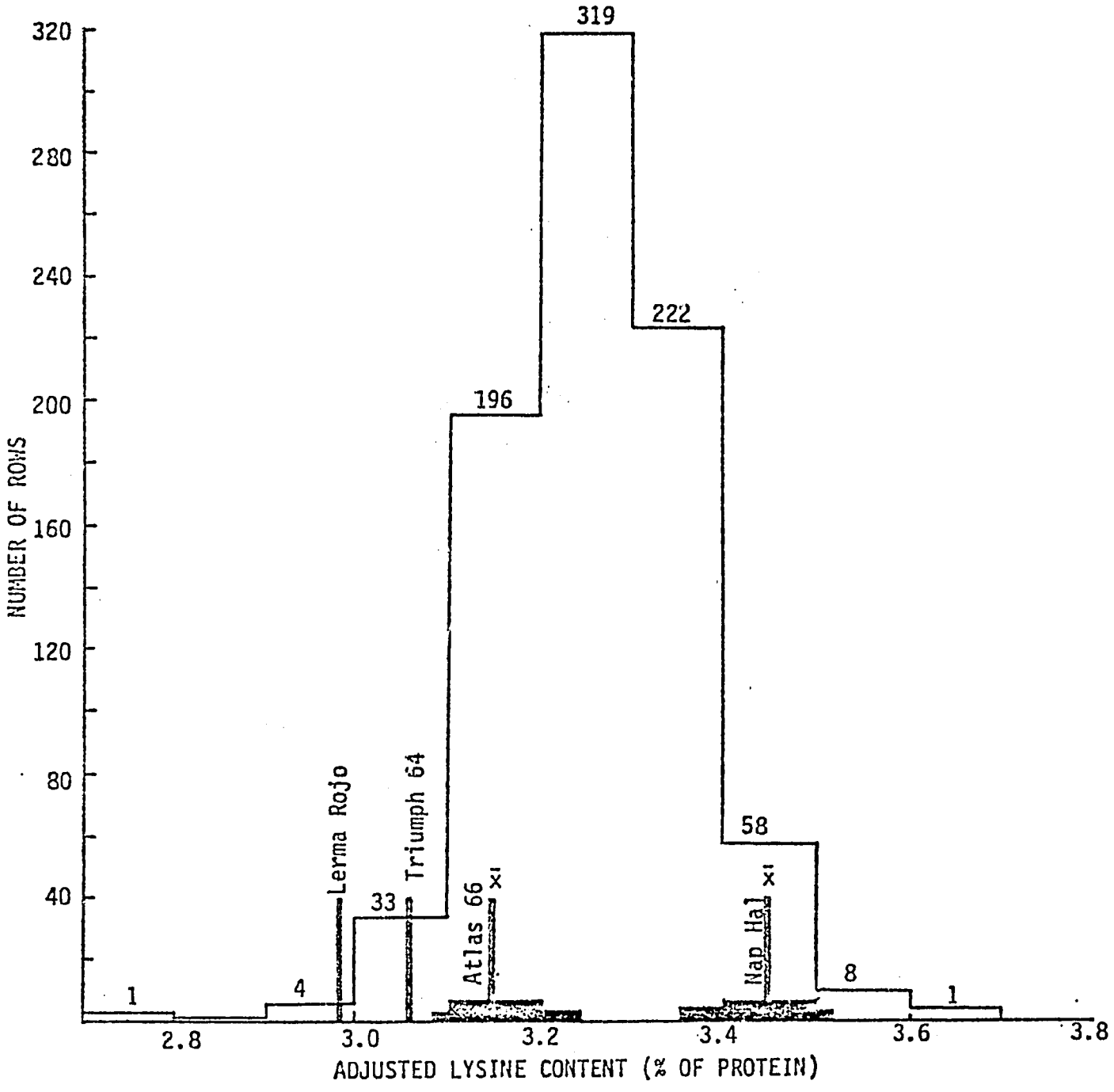


Figure 6.
Frequency distribution for adjusted lysine among F_2 progeny bulk rows (F_3 generation from a Nap Hal x Atlas 66 cross grown at Yuma, Arizona in 1971.

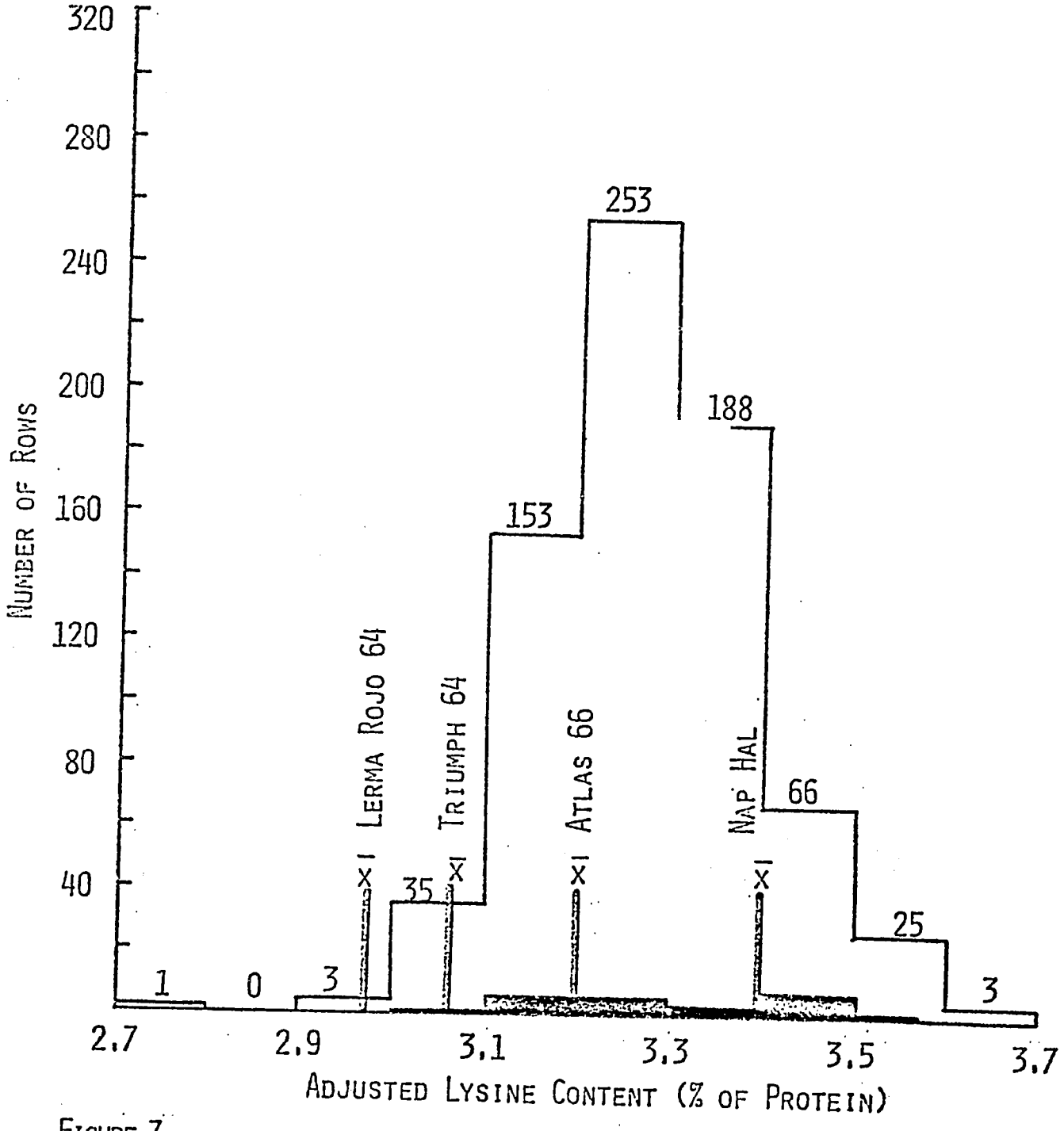


Figure 7.
Frequency distribution for adjusted lysine among F_2 progeny bulk rows (F_4 generation) from a Nap Hal x Atlas 66 cross grown at Yuma, Arizona in 1972.

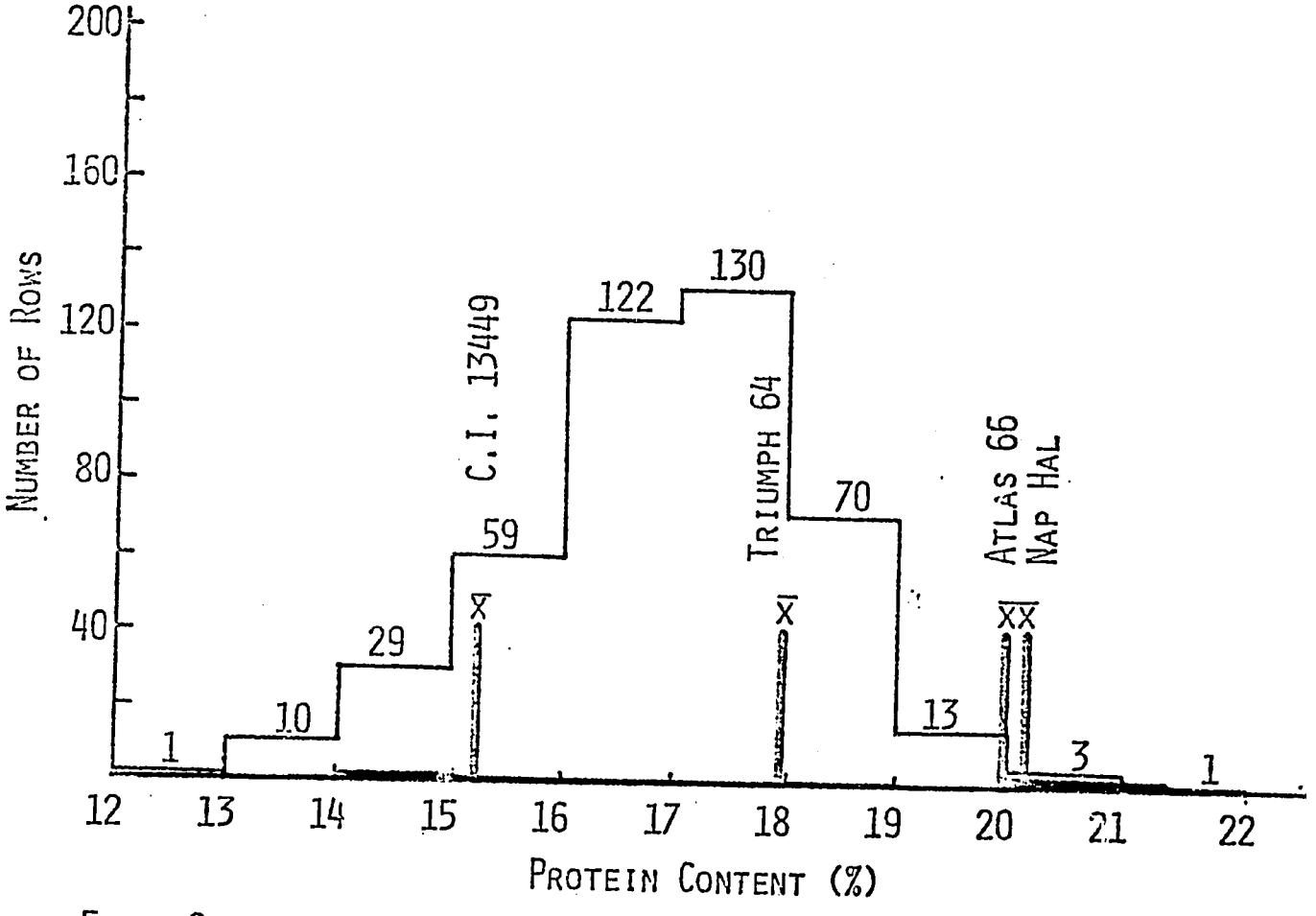


Figure 8.
Protein frequency distribution for F_2 progeny bulk rows (F_3 genera from a Nap Hal x C.I. 13449 cross grown at Yuma, Arizona in 1972.

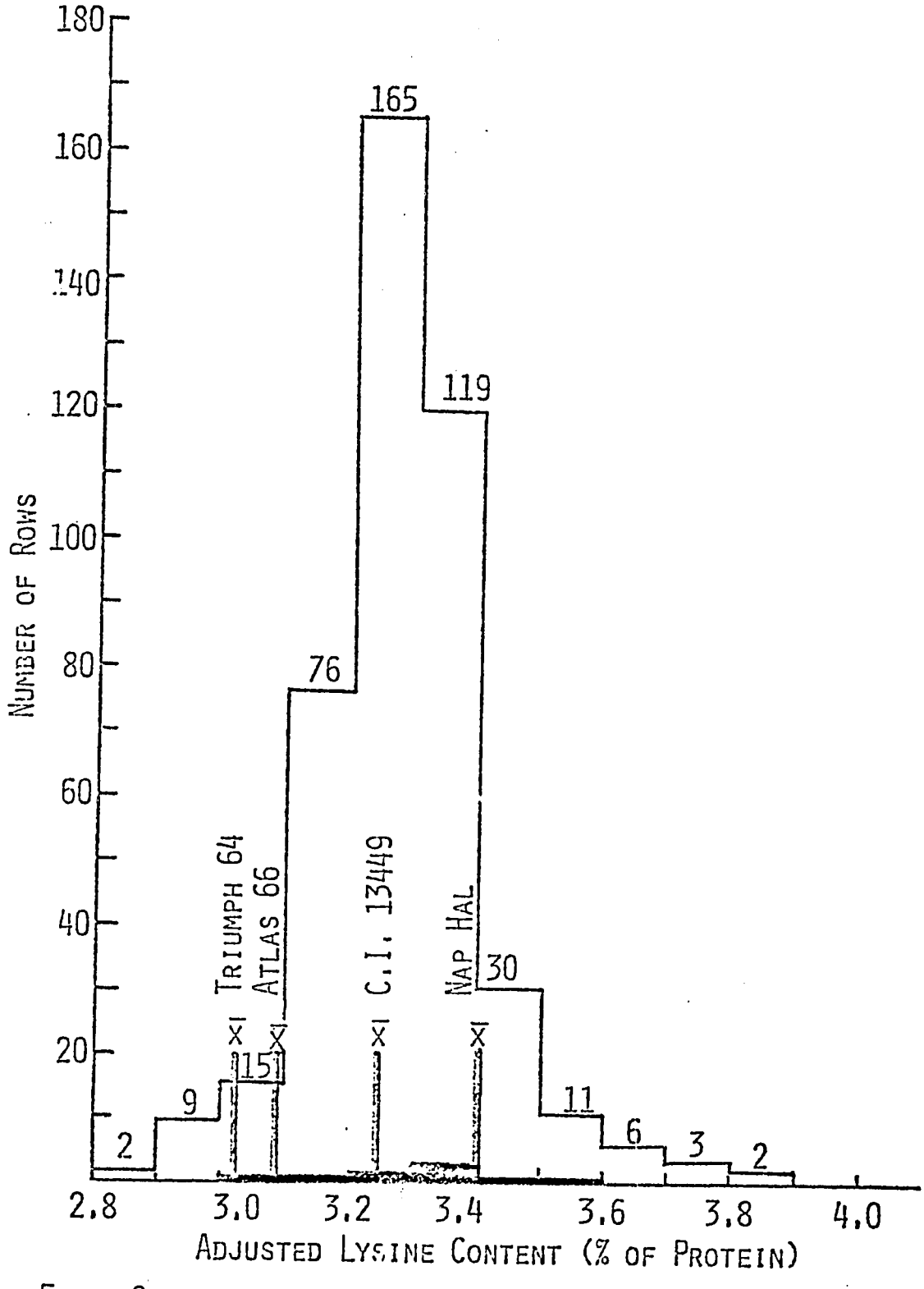


Figure 9.
Frequency distribution for adjusted lysine among F_2 progeny bulk rows (F_3 generation) from a Nap Hal x C.I. 13449 cross grown at Yuma, Arizona in 1972.

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