PROTEIN AND AMINO ACIDS CONTENTS, NITRATE REDUCTASE ACTIVITY, AND STOMATAL RESISTANCE OF ANEUPLOID WHEAT (TRITICUM AESTIVUM L.) VARIETY "CHINESE SPRING"

BY

CHANG-CHI CHU

B.S., TAIWAN CHUNG-HSING UNIVERSITY, 1954

A MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

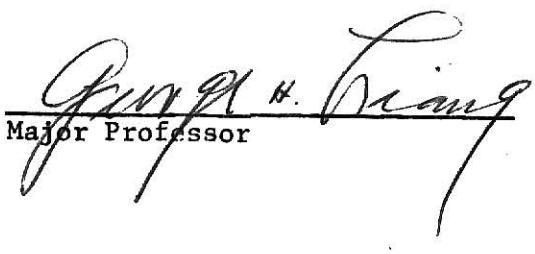
MASTER OF SCIENCE

Department of Agronomy

KANSAS STATE UNIVERSITY
Manhattan, Kansas

1973

Approved by:



Major Professor

THE FOLLOWING PAGE IS CUT OFF

THIS IS AS RECEIVED FROM THE CUSTOMER

TABLE OF CONTENTS

LIST OF TABLES

INTRODUCTION

The development of monosomic and tetrasomic stocks for each of 21 chromosomes of hexaploid wheat (26) provided a simple and effective way to study many characteristics of wheat in relation to specific chromosomes. However, only a few of the physiological aspects have been associated with individual chromosomes in wheat.

Physiological properties of plants are the expression of their respective genes but are subjected to the influence of environment. Ample evidence shows that genetic expressions of certain physiological properties might be masked by environmental effects. Therefore, controlled conditions seem to be needed to reduce the interaction between the environmental factors and genetic make-up of the plant.

The present report concerns the content of soluble protein, total free amino acids, nitrate reductase activity, and stomatal resistance of monosomic and tetrasomic lines of Chinese Spring wheat leaves. Plants were grown in a growth chamber, in 1972-73 to single out the chromosomes related to these physiological properties.

REVIEW OF LITERATURE

Photoperiodic response of the 'Hope' variety was found due to a major gene for sensitivity on 4B, and minor genes for sensitivity on 1A and 6B, and genes for insensitivity on 3B and 7D (10). Also, homoeologus group 5, particularly 5A and 5D, was important in genetic control of vernalization in Hope (9). It was found that delayed heading in 'Chinese Spring' mono-5D x 'Yaqui-53' hybrid and significant early heading in Chinese Spring mono-5B x Yaqui 53 hybrid indicated presence of the recessive gene Sg_1 and dominant gene Sg_5 for growth habit on these chromosomes, respectively (27). 'Marquillo-Tinstein' was found to carry genes controlling dwarfism on chromosomes 1A, 2A, 2B, 2D, and 3D, whereas, the variety 'Marquillo-Kenya Farmer' possessed dwarfing genes on chromosome 2A, 4B, and 2D (24). The major difference between the two dwarfs appeared due to 4B. Recently, Morris et al. located a dwarfing gene in 'Tom Thumb' on chromosome 4A (22).

Genetic expression of some physiological characters might be masked by the environmental factors. The famous tri-local experiments seemed to prove that climate was by far the decisive factor in nitrogen content of wheat grain. Wheat grown on the same soil had a protein content of 18% in Kansas, 13% in California, and 11% in Maryland (16). Leaf area per plant, total numbers of leaves, and total plant dry weight of spring wheat increased with day length over 8-24 hours (6). Soluble protein content of the crown of winter wheat was increased by decreasing photoperiod as well as by decreasing temperature (23), and total soluble protein of wheat leaves decreased drastically with increasing water stress (28). Nitrate reductase activity of wheat leaves was found to vary greatly with water content and substrate

in the leaf, and photoperiod in which the plant grew (1). Even the light quality had significantly affected the enzyme activity (8). Stomata opening and hence the diffusive capacity has been shown to be extremely sensitive to its environment (19).

MATERIALS AND METHODS

Chinese Spring 1A, 1B, 6B, and 5D monosomic and tetrasomic, and normal lines were used to study soluble protein, total free amino acids, and nitrate reductase activity in leaf tissue, and all 21 monosomic and tetrasomic, and normal lines were used to study stomatal resistance.

Normal lines of Chinese Spring were obtained from segregation of monosomic lines, which occurs at about the rate of 25% normal to 75% monosomic progeny (30). Segregation of monosomic progeny was confirmed by microscopic study of irregular behavior of chromosomes at metaphase I in the pollen mother cell. Nullisomes which were normally expected from monosomic-selfed progeny were not isolated in this experiment, perhaps because of their low occurrence.

Plants were grown in 15-cm plastic pots. Soil used in the pots was mechanically mixed at the ratio of 2 clay:1 sand:1 peat moss. All plants were subjected to similar conditions in the growth chamber. Lighting of about 1,250 ft-c at plant height as provided by sixteen 160-watt fluorescent lamps and six 300-watt incadescent lamps. Photoperiod was maintained at 14 hours with 10 hours of darkness. Temperatures were $25^{\circ}C$ - $15^{\circ}C$ for day and night, respectively.

Soluble protein and total free amino acids were assayed by removing blades from the plants and cutting them into 1-cm sections. One gm subsample of the leaf tissue was homogenized in 20 ml of 0.1 M Tris in an ice bath. The extractant was filtered and centrifuged under 10,800 g at $2^{\circ}C$ for 20 minutes. The supernatant was filtered through glass wool and 2 ml was transferred to a 25 ml volumetric flask and diluted to the volume.

An aliquot was used for the assay of soluble protein by the methods of Lowry (18) and Miller (20). The procedure was as follows:

1. To 1.0 ml of diluted leaf extract add 1.0 ml of alkaline copper reagent, mix, and allow to stand at room temperature for 10 minutes.

2. Add 3.0 ml of diluted Folin reagent rapidly to the above sample, mix, and heat sample tube at 50°C for 10 minutes in a water bath.

3. Cool the sample tube to room temperature and read absorbance at 650 nm against a reagent blank in which distilled water replaces the leaf extract.

4. Prepare protein standards with bovine serum albumin at concentrations of 0.08 to 0.20 mg per ml.

Assay of total free amino acids was carried out by the method of Rosen (25).

1. Pipet a 2 ml aliquot of the diluted leaf extract into a sample tube. Prepare a sample blank by substituting water for the leaf extract.

2. Add 1 ml of ninhydrin reagent to each tube and mix the contents by shaking them gently.

3. Heat the tubes in a boiling water bath for exactly 10 minutes. Cool the sample tubes in a water bath.

4. Add 5 ml of color diluent to each tube and mix the contents by inverting the tubes.

5. Prepare amino acids standards with alanine at concentration of 0.02 to 0.10 mg per ml.

6. Measure absorbance of samples and standards against blanks at 570 nm.

The assay of nitrate reductase activity was done by a modified method based on that of Jaworski (13):

1. Remove leaf material from the central section of flag and second leaves of plants, weigh to 0.1 gm and immediately keep in a cool place.

2. Cut the weighed leaf sample into 1 cm pieces and place them into the prepared incubation medium.

3. Invert the capped test tubes and shake vigorously for a few seconds and be sure the leaf pieces are submerged in the medium.

4. Immediately place the sample tube in a water bath for incubation of 1 hour at 25°C.

5. Add 1 ml of 1% sulfanilic acid in 3 M HCl to the incubated sample tube followed immediately by 1 ml of 0.02% N-1-napthyl ethylene diamide HCl solution to complete the color reaction.

6. Let tube set for 20 minutes to develop the color.

7. Make up the blank exactly like the enzyme assay only without the plant material.

8. Prepare the standards in the range of 0.1 to 1.0 µmoles NO_2^- as KNO_2 per ml.

9. Read the absorbance at 540 nm.

10. The incubation medium consists of the following components in each tube:

Double distilled H_2O	1.25 ml
Chloramphenicol (0.5 mg/ml)	2 drops
0.2 M Phosphate buffer, pH 7.5	2.50 ml
100% propanol	0.25 ml
0.1 M KNO_3	1.00 ml

Determination of stomatal resistance of plants was carried out by a stomatal diffusion porometer designed and described by Kanemasu et al (14). The central section of the flag, second, and third leaves was fitted to the opening in the baseplate of the vapor cup with upper surface of the leaf faced to the sensor. The time lapse is measured for the ammeter reading to change from 2 to 6 μa which corresponds to a change in relative humidity from approximately 18 to 20%. The time lapse for the meter reading to increase over the specified limits is measured with a stop watch.

RESULTS AND DISCUSSION

Contents of soluble protein and total free amino acids of leaves of the tested lines are compared within each group in Table 1. Statistical analyses indicated that chromosome 1A might have a dosage effect on soluble protein content of the leaves as shown by the 1% significant F-test. This seems to indicate that chromosome 1A influenced the soluble protein content in leaves.

No significant differences in total free amino acids in the leaves were found among the tested lines. Results seem to indicate that protein synthesis might not be as efficient in the leaves of monosomic 1A as the normal line, and even less efficient than in the corresponding tetrasomic lines.

The possible role of chromosome 1A in protein synthesis in wheat leaves is supported by the nitrate reductase activity analyses. Table 2 shows that nitrate reductase activity was not significantly different among the four homoeologous groups. There is evidence that nitrate reductase activity is related to leaf protein of wheat (1,4). But this relationship was not established in the present study. It seems possible that the conversion of amino acids into protein in the leaf might slow down due to deficiency of a 1A chromosome as in Chinese Spring monosomic 1A.

Crosby reported that monosomic 1A, 1B, 6B, and 5D are the nucleolar-organizing chromosomes (3). Since major RNA components of ribosomes are synthesized in the nucleolus (21), and trisome 6 and triploid maize had higher RNA content than other trisomics and diploid (17), it seems reasonable to assume that chromosome 1A might be involved in the protein synthesis in the leaf.

Table 1. Soluble protein and total free amino acids content in leaves of aneuploid wheat (Triticum aestivum L.) variety Chinese Spring at heading stage.

Lines	Soluble protein (mg/gm dry wt)	F-test	Free amino acids (mg/gm dry wt)	F-test
CS Mono 1A	141.88	**	45.71	N.S.
CS Normal	165.21		60.30	
CS Tetra 1A	171.93		61.32	
CS Mono 1B	165.89	N.S.	54.02	N.S.
CS Normal	165.21		60.30	
CS Tetra 1B	168.57		55.22	
CS Mono 6B	182.99	N.S.	72.03	N.S.
CS Normal	165.21		60.30	
CS Tetra 6B	162.30		59.88	
CS Mono 5D	167.79	N.S.	55.67	N.S.
CS Normal	165.21		60.30	
CS Tetra 5D	178.03		52.19	

** Significant at 1% level, LSD (.05) = 13.95

Table 2. Nitrate reductase activity in leaves of aneuploid wheat (Triticum aestivum L.) variety Chinese Spring at heading stage.

Lines	NRA(1)[1/] (µmoles NO_2/hr/gm)	F-test	NRA(2)[1/] (µmoles NO_2/hr/gm)	F-test	NRA(1+2)[1/] (µmoles NO_2/hr/gm)	F-test
CS Mono 1A	9.143	N.S.	9.105	N.S.	9.124	N.S.
CS Normal	9.463		9.923		9.693	
CS Tetra 1A	9.956		8.070		9.013	
CS Mono 1B	11.083	N.S.	11.937	N.S.	11.510	N.S.
CS Normal	9.463		9.923		9.693	
CS Tetra 1B	9.976		12.379		11.178	
CS Mono 6B	12.073	N.S.	11.242	N.S.	11.658	N.S.
CS Normal	9.463		9.923		9.693	
CS Tetra 6B	9.880		8.173		9.027	
CS Mono 5D	7.943	N.S.	8.492	N.S.	8.217	N.S.
CS Normal	9.463		9.923		9.693	
CS Tetra 5D	7.266		6.499		6.883	

[1/] NRA(1), NRA(2), and NRA(1+2) are nitrate reductase activity of flag leaf, second leaf, and average of flag and second leaves, respectively.

Tables 1 and 2 show that amino acids and nitrate reductase were not different significantly among tested lines. That might be explained by the disproportionate replication of ribosomal RNA genes in the nucleolus organizer region of the single dose in the cell, as shown in F_1 Drosophila melanogaster (29). Therefore, nitrate reductase activity and amino acids syntheses might be partially compensated by a higher number of ribosomal RNA genes per nucleolar-organizing chromosome due to deficiency of one dose. Tartof (29) suggested that there might be mechanism in the cell capable of sensing a deficiency in the number of ribosomal RNA genes.

Stomatal resistance was varied greatly in all tested lines and the results were not statistically significant among the comparisons except 7D group. In reviewing the literature, it was found that many factors are involved in the opening of stomata. Environmental factors that can affect stomata are light and carbon dioxide, transmission of stimulus, temperature, water supply, and relative humidity (11,19), and mechanical shock (31). Internal factors that affect stomata are endogenous rhythms and metabolites (11), and recently discovered substances like abscisic acid (12,15), potassium concentration in the leaf (5,7) and interaction between abscisic acid and kinetin (2). The complexity of the interactions among the listed factors complicated this line of study. The multiple factors involved in the stomata control might cause in the great variation in stomatal resistance in this study. It also shows that the heritability of stomatal resistance character of wheat might be low.

The significant F-test of stomatal resistance in 7D lines needs to be carefully considered.

Table 3. Stomatal resistance in leaves of aneuploid wheat (Triticum aestivum L.) variety Chinese Spring at heading stage.

Lines	SR(1)[1/] (Sec)	F-test	SR(2)[1/] (Sec)	F-test	SR(3)[1/] (Sec)	F-test	SR(1+2)[1/] (Sec)	F-test	SR(1+2+3)[1/] (Sec)	F-test
CS Mono 1A	15.0	N.S.	17.6	N.S.	20.8	N.S.	16.3	N.S.	17.8	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 1A	27.8		37.5		37.5		32.7		34.3	
CS Mono 2A	18.4	N.S.	33.0	N.S.	33.8	N.S.	25.7	N.S.	28.4	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 2A	27.6		39.2		26.1		33.4		31.0	
CS Mono 3A	25.4	N.S.	38.9	N.S.	38.2	N.S.	32.1	N.S.	34.2	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 3A	25.4		33.6		34.8		29.5		31.3	
CS Mono 4A	20.4	N.S.	26.0	N.S.	29.7	N.S.	23.2	N.S.	25.4	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 4A	38.6		53.6		72.9		46.1		55.0	
CS Mono 5A	18.9	N.S.	24.6	N.S.	28.7	N.S.	21.8	N.S.	24.1	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 5A	23.3		26.2		19.9		24.8		23.2	
CS Mono 6A	20.3	N.S.	27.4	N.S.	22.7	N.S.	23.9	N.S.	23.5	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 6A	24.8		19.6		27.9		22.2		24.1	
CS Mono 7A	18.3	N.S.	21.5	N.S.	23.2	N.S.	19.9	N.S.	21.0	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 7A	27.8		23.0		24.3		25.4		25.0	

(Continued)

Lines	SR(1)[1/] (Sec)	F-test	SR(2)[1/] (Sec)	F-test	SR(3)[1/] (Sec)	F-test	SR(1+2)[1/] (Sec)	F-test	SR(1+2+3)[1/] (Sec)	F-test
CS Mono 1B	30.4	N.S.	23.1	N.S.	28.9	N.S.	26.8	N.S.	27.5	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 1B	20.2		29.3		28.1		24.8		25.9	
CS Mono 2B	20.6	N.S.	19.4	N.S.	32.1	N.S.	20.0	N.S.	24.1	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 2B	27.8		30.6		32.3		29.2		30.2	
CS Mono 3B	16.7	N.S.	20.9	N.S.	29.9	N.S.	18.8	N.S.	22.5	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 3B	23.1		22.0		27.9		22.5		24.3	
CS Mono 4B	18.0	N.S.	24.8	N.S.	22.3	N.S.	21.4	N.S.	21.7	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 4B	16.9		20.0		28.6		18.5		21.9	
CS Mono 5B	29.5	N.S.	29.4	N.S.	46.4	N.S.	29.4	N.S.	35.1	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 5B	23.7		24.5		30.5		24.1		26.3	
CS Mono 6B	13.4	N.S.	17.2	N.S.	24.5	N.S.	15.3	N.S.	18.4	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 6B	32.4		47.3		46.0		39.8		41.9	
CS Mono 7B	20.2	N.S.	28.0	N.S.	32.8	N.S.	24.1	N.S.	27.0	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 7B	27.1		22.3		29.7		24.7		26.4	

(Continued)

Lines	SR(1)[1/] (Sec)	F-test	SR(2)[1/] (Sec)	F-test	SR(3)[1/] (Sec)	F-test	SR(1+2)[1/] (Sec)	F-test	SR(1+2+3)[1/] (Sec)	F-test
CS Mono 1D	28.5	N.S.	28.4	N.S.	35.5	N.S.	28.4	N.S.	30.8	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 1D	28.7		34.8		40.7		31.8		34.7	
CS Mono 2D	26.7	N.S.	39.0	N.S.	40.8	N.S.	32.8	N.S.	35.5	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 2D	50.6		50.8		65.1		50.7		55.5	
CS Mono 3D	33.9	N.S.	40.3	N.S.	52.2	N.S.	37.1	N.S.	42.1	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 3D	21.8		21.6		24.2		21.7		22.5	
CS Mono 4D	24.3	N.S.	48.4	N.S.	47.8	N.S.	36.4	N.S.	40.2	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 4D	23.4		28.7		21.7		26.1		24.6	
CS Mono 5D	21.6	N.S.	30.3	N.S.	29.1	N.S.	26.0	N.S.	27.0	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 5D	19.8		29.8		29.4		24.8		26.3	
CS Mono 6D	53.4	N.S.	50.8	N.S.	59.2	N.S.	52.1	N.S.	54.5	N.S.
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 6D	22.9		23.0		23.7		23.0		23.2	
CS Mono 7D	63.4	**	80.2	**	74.9	**	71.8	**	72.9	**
CS Normal	30.7		38.9		43.3		34.8		37.6	
CS Tetra 7D	30.6		37.8		46.5		34.2		38.3	

[1/] SR(1), SR(2), SR(3), SR(1+2), SR(1+2+3) are stomatal resistance determination of flag, second, third, average of flag and second, and average of flag, second, and third leaves, respectively.

SUMMARY AND CONCLUSION

Chinese Spring 1A, 1B, 6B, and 5D monosomic and tetrasomic, and normal lines of wheat were used to study soluble protein, total free amino acids, and nitrate reductase activity, and all 21 monosomic and tetrasomic, and normal lines were used to study stomatal resistance of wheat leaves under controlled conditions.

Content of soluble protein in wheat leaves differed significantly between monosomic and normal, and between monosomic and tetrasomic lines of chromosome 1A, but contents of amino acids and nitrate reductase activity did not. Conversion of amino acids into protein might be slowed down by the deficiency of one dose of chromosome 1A. The non-significant differences in amino acids contents and nitrate reductase activity among the tested lines is interpreted as disproportionate replication of ribosomal RNA genes on the single dose of certain nucleolar organizers.

Stomatal resistance of leaves did not differ among lines of Chinese Spring except 7D lines, indicating the complexity of interaction between genes and environment. It also shows that the heritability of this character might be low.

ACKNOWLEDGEMENTS

The author wishes to express his sincere appreciation to Dr. George H. Liang, major professor, for his direction of the experiments and preparing the thesis.

Appreciation is also extended to Dr. Gary M. Paulsen, Dr. K. C. Lee, Dr. Edward T. Kanemasu, Dr. Elmer G. Heyne, Dr. Howard L. Mitchell and Dr. Richard L. Vanderlip for their criticism and assistance.

LITERATURE CITED

1. Beevers, L. and R. H. Hageman. 1969. Nitrate reduction in higher plants. Ann. Rev. Plant Physiol., 20:495-522.

2. Cooper, M. J., J. Digby, and P. J. Cooper. 1972. Effects of plant hormones on the stomate of barley: a study of the interaction between abscisic acid and kinetin. Planta (Berl.), 105:43-49.

3. Crosby, A. R. 1957. Nucleolar activity of lagging chromosomes in whest. Am. J. Bot., 44:813-822.

4. Croy, L. I. and R. H. Hageman. 1970. Relationship of nitrate reductase activity to grain protein production in wheat. Crop Sci., 10:280-285.

5. Fischer, R. A. 1971. Role of potassium in stomatal opening in the leaf of Vicia faba. Plant Physio., 47:555-558.

6. Friend, D. J. C., V. A. Helson, and J. E. Fisher. 1967. Effect of day-length on the growth of wheat. Can. J. Bot., 45:117-131.

7. Graham, R. D. and A. Ulrich. 1972. Potassium deficiency-induced changes in stomatal behavior, leaf water potentials, and root system permeability in Beta vulgaris L. Plant Physiol., 49:105-109.

8. Harper, J. E. and G. M. Paulsen. 1968. Influence of intensity, quality, and duration of light on nitrogen reduction and assimilation in wheat. Crop Sci., 8:537-539.

9. Halloran, G. M. and C. W. Boydell. 1967. Wheat chromosomes with genes for vernalization response. Can. J. Genet. Cytol., 9:632-638.

10. Halloran, G. M. and C. W. Boydell. 1967. Wheat chromosomes with genes for photoperiodic response. Can. J. Genet. Cytol., 9:394-398.

11. Heath, O. V. S. and T. A. Mansfield. 1969. The movements of stomata (in M. B. Wilkins edited "The physiology of plant growth and development", McGraw-Hill Pub. Co., pp. 303-326.)

12. Horton, R. E. 1971. Stomatal opening: the role of abscisic acid. Can. J. Bot., 49:583-585.

13. Jaworski, E. G. 1971. Nitrate reductase assay in intact plant tissues. Biochem. Biophys. Res. Comm., 43:1274-1279.

14. Kanemasu, E. T., G. W. Thurtell and C. B. Tanner. 1969. Design, calibration and field use of a stomatal diffusion porometer. Plant Physiol., 44:881-885.

15. Kriedemann, P. E., B.R. Loveys, G. L. Fuller, and A. C. Leopold. 1972. Abscisic acid and stomatal regulation. Plant Physiol., 49:842-847.

16. LeClerc, J. A. and P. A. Yoder. 1912. Tri-local soil-exchange experiments with wheat. 8th Int. Cong. Appl. Chem., 26:137-150. Reviewed in "Wheat and wheat production", edited by Quisenberry, K. S., and L. R. Reitz. 1967. P. 134. ASA.

17. Lin, M. 1955. Chromosomal control of nuclear composition in maize. Chromosoma, Bd. 7, S., 340-370.

18. Lowey, O. H., N. J. Roseborough, A. L. Farr, and R. J. Randall. 1951. Protein measurement with the Folin phenol reagent. J. Biol. Chem., 193: 265-275.

19. Meidner, H. and T. A. Mansfield. 1968. "Physiology of stomata", McGraw-Hill Pub. Co.

20. Miller, G. L. 1959. Protein determination for large number of samples. Anal. Chem., 31:964.

21. Miller, L. and D. D. Brown. 1969. Variation in the activity of nucleolar organizers and their ribosomal gene content. Chromosoma (Berl.), 28:430-444.

22. Morris, R., J. W. Schmidt, and V. A. Johnson. 1972. Chromosomal location of a dwarfing gene in 'Tom Thumb' wheat derivative by monosomic analysis. Crop Sci., 12:247-249.

23. Paulsen, G. M. 1968. Effect of photoperiod and temperature on cold hardening in winter wheat. Crop Sci. 8:29-32.

24. Piech, J. and L. E. Evans. 1968. Monosomic analysis of temperature sensitive dwarfism in common wheat. Can. J. Genet. Cytolo., 10:125-130.

25. Rosen, H. 1957. A modified ninhydrin colorimetric analysis for amino acids. Arch. Biochem. Biophys., 67:10-15.

26. Sears. E. R. 1954. The aneuploids of common wheat. Missouri Agr. Exp. Sta. Columbia Res. Bull., 572.

27. Singh, M. P. 1967. Monosomic analysis in wheat. Heredity, 22:591-596.

28. Stutte, C. A., G. W. Todd 1969. Some enzyme and protein changes associated with water stress in wheat leaves. Crop Sci., 9:510-512.

29. Tartof, K. D. 1971. Increasing the multiplicity of ribosomal RNA genes in Drosophila melanogaster. Science, 171:294-297.

30. Tsunewaki, K. 1964. The transmission of the monosomic condition in a wheat variety, Chinese Spring. II. A critical analysis of nine years records. Jan. J. Genetics, 38:270-281. Reviewed in "Wheat and wheat production", edited by Quisenberry, K. S., and L. R. Reitz, 1967, p. 45., ASA.

31. Williams, W. T. 1949. The sensitivity of stomata to mechanical shock. Ann. Bot. (London) N.S. 13:309-327.

PROTEIN AND AMINO ACIDS CONTENTS, NITRATE REDUCTASE ACTIVITY, AND STOMATAL RESISTANCE OF ANEUPLOID WHEAT (TRITICUM AESTIVUM L.) VARIETY "CHINESE SPRING"

BY

CHANG-CHI CHU

B.S., TAIWAN CHUNG-HSING UNIVERSITY, 1954

AN ABSTRACT OF A MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Agronomy

KANSAS STATE UNIVERSITY
Manhattan, Kansas

1973

Chinese Spring 1A, 1B, 6B, and 5D monosomic and tetrasomic, and normal lines were used to study the soluble protein, total free amino acids, and nitrate reductase activity, and all 21 monosomic and tetrasomic, and normal lines were used to study the stomatal resistance of wheat leaf under controlled conditions.

Content of soluble protein in wheat leaves differed significantly among monosomic, normal, and tetrasomic lines of 1A, but the amino acids and nitrate reductase activity did not. Conversion of amino acids into protein might be slowed down by the deficiency of chromosome 1A as in the monosomic line of 1A.

Stomatal resistance of leaves did not differ among lines of Chinese Spring, except for the 7D monosomic line, indicating the complexity of interaction between genes and environment. It also shows that the heritability of this character might be low.