PHYSIOLOGICAL DIFFERENCES IN GERMINATING WHEAT SEEDS DUE TO SEED SOURCE

SHAHBAZ AHMAD* AND FLOYD E. BOLTON

Crop Science Department,
Oregon State University, Corvallis, OR 97331, U.S.A.

Abstract

Seedlots of 18 winter wheat varieties produced at 2 distinctly different locations, were compared for physiological differences during germination. Varieties were significantly different (P = 0.01) from each other for seedling axes weight, alpha-amylase and glutamine synthetase activities and soluble protein of seedling axes and endosperm. Corvallis source seed was superior to Moro source seed for seedling axes weight and endosperm soluble protein while the rverse was true for alpha-amylase and glutamine synthetase activities, and seedling axes soluble protein. Enzyme activity increased in Moro source seed due to environmental stress but Corvallis source seed was better for total and soluble reserves in the endosperm. Higher quantity of total and soluble protein in the endosperm over-shadowed the benefit of better enzyme activity for seedling growth. Both genotype and environment affected the seed physiological properties. The interaction between varieties and seed source was significant.

Introduction

The source of seed affects seedling shoot dry weight (Quinby et al., 1962; DasGupta & Austenson, 1973 a, b). Both genotype and environment affect the protein content of the seeds (Ries & Everson, 1973). Seed protein content differences are depicted in the seedling development differences (Ching & Rynd, 1978). The objective of this study was to discern the physiological differences in germinating wheat seeds from two seed sources which might indicate the mechanism of subsequent growth stimulation and yield increase.

Materials and Methods

Uniform seed lots of 18 winter wheat varieties were produced at Corvallis and Moro, during the 1984-85 crop season. The locations represent two different growing environments in Oregon, USA. Corvallis is wet (1000+ mm) with a relatively mild winter, while Moro is dry (290 mm) with cold winters.

Seeds from both sources were sized to achieve uniformity by passing all the seedlots through a no. 8 (8/64" x 3/4" slot) screen and holding on a no. 7 (7/64" x 3/4" slot) screen. Their weight and protein content (Micro-kjeldahl N x 5.7) are summarized in Table 1.

^{*}Present Address: Wheat Programme, National Agricultural Research Centre, Islamabad, Pakistan.

Table 1. Seed weight a	ınd p	percent protein of 18 w	vinter wheat varieties from
	,,	two seed sources.	

	1000 Seed Weight (gms)			Percent Protein		
Varietie	Corvallis	Moro		Corvallis	Moro	
	Source	Source	S.E.	Source	Source	S.E.
Stephens	46.63	43.04	1.27	11.23	10.32	0.32
Hill-81	38.21	34.61	1.27	12.71	9.52	1.13
Lewjain	41.81	38.64	1.12	13.11	8.78	1.53
Nugaines	42.92	38.41	1.59	10.89	8.89	0.71
Daws	44.00	41.43	0.91	11.57	8.55	1.07
Luke	41.36	35.02	2.24	12.54	9.46	1.09
Sprague	38.58	30.30	2.93	13.05	9.75	1.17
Dusty	36.70	37.30	0.21	12.37	8.04	1.53
Oveson	47.34	40.88	2.28	12.03	8.61	1.21
Malcolm	48.60	44.18	1.56	11.06	9.52	0.54
Kharkof	43.79	32.80	3.89	12.88	10.89	0.70
Wanser	43.55	35.56	2.82	11.34	9.98	0.48
Hatton	39.65	36.14	1.24	11.86	10.03	0.65
Weston	50.98	44.41	2.32	12.94	11.23	0.60
Bezostaya	48.43	40.78	2.70	13.34	10.66	0.95
Moro	36.93	31.52	1.91	12.71	8.49	1.49
Faro	36.71	33.35	1.19	12.43	9.98	0.87
Tres	41.85	38.03	1.35	11.80	8.04	1.33
S.E.	1.01	0.98		0.18	0.22	

Thirty seeds from each seed lot were germinated in the plastic germination boxes for 48 h at 20°C constant temperature in the dark. Two replications of 10 seedlings each were separated into groups of seedling axes and endosperms in a moist chamber and each group was collected on ice. Fresh weight of 10 seedling axes was determined. After wieghing, the seedling axes and endosperms were placed in 10 ml of respective grinding buffers.

The enzyme extraction was conducted at 0 to 3°C. The seedling axes were washed with ice-cold grinding buffer containing 0.1 M HEPES, 4 mM Mg-acetate, 0.1 M sucrose, and 10 mM mercaptoethanol. The material was ground with a mortar and pestle in 10 ml of grinding buffer. The slurry was centrifuged at 30,000 g for 10 min, and the cell-free supernatant was used as the enzyme preparation.

Already separated, two replications of 10 endosperms each were ground in 10 ml of 0.2% Ca-acetate (pH 6.0) with a mortar and pestle. The slurry was incubated at 70°C for

20 min., with occasional stirring to extract alpha-amylase selectively (Briggs, 1973). The slurry was then centrifuged at 30,000 g for 10 min., and the supernatant was used as enzyme preparation. The activity of alpha-amylase was assayed by the reduction of soluble starch substrate in 0.5 ml of a 10 fold dilution of enzyme preparation at 20°C for 1 to 5 min (Mitchell, 1972). The soluble protein content of both the enzyme preparations was determined by the Bradford (1976) method.

Results and Discussions

Although the seed lots used were of uniform size, there were large differences in 1000-seed weight and seed protein among the wheat varieties within a seed source. Seed weight and seed protein was higher for Corvallis grown seed compared to Moro grown seed in all the varieties (Table 1). In the absence of size differences among the seed lots, the 1000-seed weight variability are essentially differences in seed density.

Varieties included in this study differ from one another for percent protein (Table 1). The glutamine synthetase activity was comparable between the high and low protein seeds during the first two days of germination (Ching & Rynd 1978). The study was done on the seeds germinated for 2 days to avoid the time course differences.

Seedling size: Seeds from Corvallis source produced significantly (P = 0.01) larger seedlings than the seeds from Moro source after 2 days of germination (Table 2). As the seeds from Corvallis source are more dense and protein percentage was also higher, more food reserves are available to the developing seedling axis. The increased growth in case of Corvallis seed source may be attributed to the better availability of food reserves. The increased growth rate apparently is not directly related with genetic capability as the two groups belong to the same cultivars. The difference may be due to availability of substrate and its transfer from the endosperm to seedling axis.

Enzyme Activity: Two enzymes glutamine synthetase and alpha-amylase were assayed in the eighteen winter wheat varieties from two different sources to discern if any differential enzyme synthesis was involved. Glutamine synthetase is known to participate in the synthesis of a variety of key metabolites (Greenbern, 1969). The glutamine synthetase activity was higher in Moro grown seeds. Thus quantitative differences in enzyme activity did occur in seeds of some genetic make-up. Environment had an effect on the glutamine synthetase activity in the seed.

Alpha amylase is an important starch degrading enzyme in the endosperm of cereal grains. The reaction products provide substrate and an energy source for the embryo during germination. The activity of alpha-amylase was significantly higher in the endosperms of Moro grown seeds. The difference in activity indicates the amount of en-

Table 2. Means of 18 winter wheat varieties from 2 seed sources for embryo weight (Emb. Wt.), alpha-amylase activity (AA), glutamine synthetase activity (GS), endosperm soluble protein (End. P) and embryo soluble protein (Emb. P)

Varieties	Emb. Wt	AA	GS	End. P	Emb. P	
	(mg)	(µ mole/min)		(μg/10 Seeds)		
Stephens	177.25	3.45	320.34	60.02	52.09	
Hill-81	108.75	3.44	332.04	52.43	41.23	
Lewjain	138.50	3.63	270.87	61.45	43.04	
Nugaines	143.50	3.38	321.52	62.25	39.16	
Daws	176.50	2.82	341.87	65.38	41.59	
Luke	129.25	3.08	320.24	61.52	39.43	
Sprague	150.50	2.48	291.99	61.79	37.24	
Dusty	150.75	1.86	320.91	59.25	35.86	
Oveson	176.00	2.39	343.63	62.61	40.68	
Malcolm	166.00	3.00	314.06	65.70	44.02	
Kharkof	166.50	2.64	227.93	56.97	44.20	
Wanser	149.25	2.07	307.63	58.66	37.79	
Hatton	108.75	1.95	318.08	57.45	32.84	
Weston	148.25	2.13	263.04	70.86	40.72	
Bezostaya	147.00	3.11	252.39	65.36	38.97	
Moro	148.75	3.93	268.46	57.20	37.79	
Faro	152.75	4.31	281.92	50.34	34.86	
Tres	153.25	2.94	312.94	56.72	36.20	
LSD 0.01	19.58	0.83	47.54	7.42	7.46	
Seed source						
Corvallis	156.81	2.37	223.48	73.72	43.79	
Moro	142.25	3.47	377.57	46.95	53.95	
LSD 0.01	4.74	0.32	20.93	3.33	1.55	

zyme present in the endosperm. The synthesis of hydrolases was greater and faster hydrolysis of reserves in the storage site occurred in Moro grown seeds. The increased alpha-amylase activity in the Moro grown seeds was not the result of different gene products because the cultivars in both the seed sources were the same.

Varieties differed significantly form each other for glutamine synthetase and alphaamylase activity. Quantitative differences in the amount of alpha-amylase produced by different wheat varieties were observed by Gale & Marshall (1975).

Soluble Protein Content: The soluble protein content was significantly higher in the endosperms of Corvallis grown seeds. The larger increase in the size and soluble protein

in high protein seeds indicates a high protein synthesizing ability which probably is attributed to more available substrate (Ching & Rynd, 1978). The soluble protein content in the seedling axis was better in the Moro grown seeds. This soluble protein fraction consists mainly of enzymes and protein factors for various metabolic processes of growth and development of seedling axes (Ching, 1972). This difference is probably due to better enzyme activity in the Moro grown seeds.

References

- Bradford, M.M. 1976. A rapid and sensitive method for the quantification of microgram quantities of protein utilizing the principle of protein-dye binding. *Anal. Biochem.*, 72: 248-254.
- Briggs, D.E. 1973. Hormones and carbohydrate metabolism in germinating cereal grains. In: *Biosynthesis and its control in plants*. (Ed.) B.V. Milborrow. Academic Press, N.Y. pp. 219-277.
- Ching, T.M. 1972. Metabolism of germinating seeds. In: Seed Biology. (Ed.) T.T. Kozlowski, Vol. II. Academic Press, N.Y. pp. 109-213.
- Ching, T.M. and L. Rynd. 1978. Development differences in embryos of high and low protein wheat seeds during germination. *Plant Physiol.*, 62: 866-870.
- DasGupta, P.R. and H.M. Austenson. 1973 a. Relation between estimates of seed vigor and field performance in wheat. Can. J. Plant Sci., 5: 43-46.
- DasGupta, P.R. and H.M. Austenson. 1973, b. Analysis of interrelationships among seedling vigor, field emergence and yield in wheat. Agron. J., 65: 417-422.
- Gale, M.D., and G.A. Marshall. 1975. The nature and genetic control of gibberellic insensitivity in dwarf wheat grain. Heridity, 35: 55.
- Greenberg, D.M. 1969. In: Metabolic Pathways. Vol. 3: 302-306 (Ed.). Greenberg, D.M. Academic Press, N.Y.
- Mitchell, E.D. 1972. Homogeneous alpha-amylase from malted barley. Phytochemistry, 11: 1663-1676.
- Quinby, J.R., L.R. Reitz, and H.H. Lande. 1962. Effect of source of seed on productivity of hard red winter wheat. Crop. Sci., 2: 201-203.
- Ries, S.K., and E.H. Everson. 1973. Protein content and seed size relationships with seedling vigor of wheat cultivars. Agron. J., 65: 884-886.

(Received for publication 30 June 1987)