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Amino Acid Composition of the Endosperm Free Amino Acids and Proteins from a Maturing Common Wheat and Its Extracted Tetraploid

The compositions of the total amino acids and the free amino acids within the endosperm of the common wheat (*Triticum aestivum* L. em. Thell) cultivar Thatcher were compared with those of its AABB extracted tetraploid Tetrathatcher at intervals during kernel development. Rapid changes occurred in the compositions of the total amino acids within the endosperms of both wheats during development. The levels of free amino acids within the endosperm decreased with increasing maturity. At comparable stages of kernel development the total amino acids and free amino acids were of similar composition in both wheats.

The baking quality of synthetic and natural AABBDD hexaploid wheats is generally superior to the baking quality of extracted and natural AABB tetraploid wheats (Kerber and Tipples, 1969). This has been attributed to quantitative (Dronzek et al., 1970) and qualitative (Boyd and Lee, 1967; Boyd et al., 1969; Bietz and Wall, 1972; Orth and Bushuk, 1973a,b, 1974) differences in their gluten proteins. Dronzek et al. (1970) found that removal of the D-genome did not significantly alter the amino acid composition of wheat flour or the amino acid composition of the soluble Osborne protein fractions. The present investigation was undertaken to ascertain whether the D-genome has a significant effect on the amino acid composition of the free amino acids and proteins in wheat endosperm during kernel maturation.

EXPERIMENTAL SECTION

Plant Material. The common wheat (*Triticum aestivum* L. em Thell) variety Thatcher and the AABB tetraploid (Tetrathatcher) derived by Kaltsikes et al. (1968) were grown in a controlled environment chamber (21 °C, 16 h light). Prior to planting, chromosome counts were performed on the germinated Tetrathatcher seeds to ensure that only 28-chromosome plants were grown. Grain development was determined by noting the date of anthesis for each head. Each sample consisted of 12 heads from 12 different plants selected at the same stage of development. Both wheats were fully mature 49 days after anthesis.

Endosperm tissue was excised from immature seeds by hand dissection, freeze-dried, and ground in a Wiley mill to pass through a no. 60 sieve. Flour from the mature samples was obtained by grinding in a Brabender Quadrumat Junior Mill.

Amino Acid Analyses. Endosperm samples were hydrolyzed as described by Orth et al. (1974) and analyzed according to the method of Spackman et al. (1958) on a Beckman 121 automatic amino acid analyzer. Precision was better than ±3% for all amino acids listed except methionine which was partially converted to the sulfoxide during hydrolysis. Cysteine, cystine, and tryptophan were destroyed during hydrolysis.

Kjeldahl nitrogen accounted for by the amino acid analyses ranged from 80 to 92%. Recoveries were lowest for the immature samples, probably because they contained substantially more nonprotein nitrogen (Hoseney et al., 1966). For the purpose of calculating amino acid distribution, the nitrogen recovered from the chromatographic columns was used to estimate the amino acid nitrogen content of the samples. The results were adjusted to a common recovery (90%) in order to eliminate the effect of varying nonprotein nitrogen levels.

Free Amino Acid Analyses. Free amino acid extractions were performed and the free amino acids separated on the Beckman 121 automatic amino acid analyzer as described previously (Dexter and Dronzek, 1975).

Several samples of varying maturity from the two wheats were analyzed in duplicate. The standard deviations were less than 3 and 10% for major and minor components, respectively.

The concentrations of the free amino acids were expressed as grams of nitrogen per 100 g of amino acid nitrogen computed from the total amino acid analyses of the endosperms.

RESULTS AND DISCUSSION

Nitrogen Content. The total nitrogen content of the endosperms from Thatcher and Tetrathatcher decreased on a dry matter basis during the early stages of kernel development and increased as the kernels neared full maturity (Figure 1). Similar results were reported by Jennings and Morton (1963) for maturing Australian

Table I. Total Amino Acids in the Endosperm from a Hexaploid Wheat (C.V. Thatcher) and Its Extracted Tetraploid (Tetrathatcher) during Development (g of N per 100 g of Recovered N Adjusted to 90% Recovery)[a]

Amino acids	Thatcher at days after anthesis					Tetrathatcher at days after anthesis				
	9	14	21	30	49	9	14	21	30	49
Lys	6.1	4.2	3.0	2.5	2.1	7.5	4.3	2.9	2.1	2.0
His	3.2	3.3	3.4	3.3	3.3	3.5	3.1	3.2	3.2	3.2
NH_3	15.6	17.9	20.3	21.0	20.6	12.9	17.0	20.9	21.5	20.8
Arg	7.8	6.9	7.6	5.9	6.3	10.1	6.8	6.0	5.5	6.4
Asp	5.2	3.3	2.9	2.7	2.4	6.2	4.3	2.9	2.5	2.3
Thr	2.4	2.2	1.9	1.8	1.8	2.3	2.0	1.8	1.7	1.7
Ser	4.1	4.1	3.8	3.3	3.6	4.4	4.2	3.6	3.3	3.5
Glu	10.8	16.1	19.0	20.5	21.3	10.0	15.4	19.3	20.6	22.2
Pro	5.3	6.9	7.5	8.7	9.1	3.9	6.2	8.3	9.0	8.8
Gly	5.9	5.2	4.1	4.0	3.7	5.9	5.7	4.2	3.6	3.5
Ala	9.5	6.0	3.3	2.6	2.4	8.6	7.0	3.6	3.5	2.4
Val	3.4	3.1	2.7	2.8	2.7	3.6	3.2	2.7	2.5	2.5
Met	1.3	1.0	0.8	0.8	0.8	1.1	1.0	0.7	0.7	0.7
Ile	2.2	2.2	2.2	2.2	2.1	2.4	2.3	2.2	2.3	2.1
Leu	3.9	4.2	4.1	4.2	4.2	4.4	4.2	4.1	4.2	4.1
Tyr	1.2	1.3	1.2	1.2	1.2	1.4	1.0	1.1	1.2	1.3
Phe	1.9	2.3	2.2	2.5	2.6	2.0	2.4	2.5	2.7	2.7

[a] Tryptophan, cysteine, and cystine were not determined.

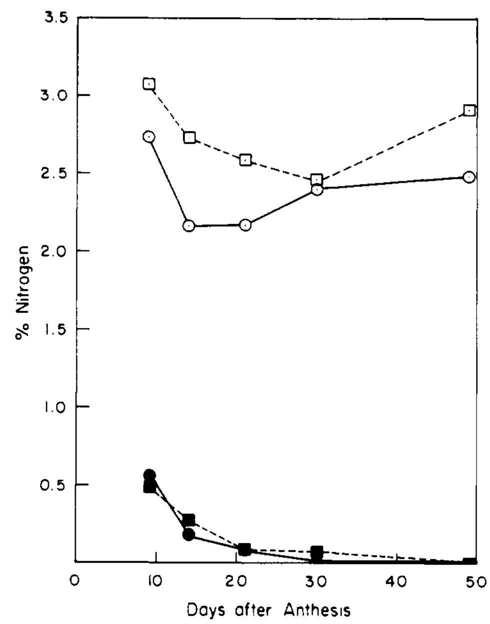

Figure 1. The total nitrogen content (⊙,⊡) and free amino acid nitrogen (•,▪) of developing endosperms from Thatcher (⊙,•) and Tetrathatcher (□,▪). (Results are expressed as percent nitrogen on a dry matter basis.)

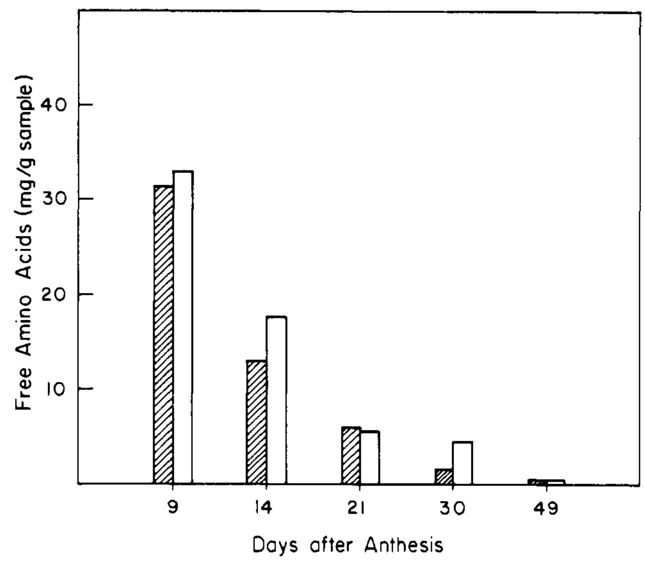

Figure 2. Free amino acid content of developing endosperms from Thatcher (▨) and Tetrathatcher (□).

wheats. They attributed this initial decrease in nitrogen content to rapid starch synthesis during the early stages of kernel development. The nitrogen content of Tetrathatcher was greater than the nitrogen content of Thatcher throughout development.

Free amino acid nitrogen decreased rapidly during development in both Thatcher and Tetrathatcher on a dry matter basis (Figure 1). At maturity the amount of free amino acid nitrogen was very low. The level of free amino acid nitrogen was similar in each wheat at comparable stages of maturity.

Total Amino Acid Composition. Rapid Changes occurred in the amino acid composition of the endosperms from Thatcher and Tetrathatcher during the first 30 days after anthesis (Table I). The largest changes were a decrease in the concentrations of lysine, aspartic acid, glycine, and alanine concomitant with an increase in the concentrations of glutamic acid and proline. Minor changes were a decrease in the concentrations of arginine, threonine, serine, and valine and an increase in phenylalanine. At comparable stages of development the amino acid compositions of Thatcher and Tetrathatcher were essentially the same. These results were in close agreement to previous studies of maturing Australian wheats (Jennings and Morton, 1963), hard red winter wheats (Hoseney et al., 1966), and a durum wheat (Dexter and Dronzek, 1975).

Free Amino Acids. The amount of endosperm free amino acids in Thatcher and Tetrathatcher during development decreased rapidly (Figure 2). This was largely a reflection of the rapid increase in endosperm dry matter during kernel development. The actual amount of free amino acids present on a per kernel basis did not decrease greatly until after 21 days after anthesis.

The amino acids found in largest concentrations within the free amino acid pools of both Thatcher and Tetrathatcher were serine, asparagine, glutamic acid, glutamine, proline, alanine, and glycine (Table II). This was in general agreement with the semiquantitative results of Hoseney and Finney (1967) for maturing hard red winter wheats. In general the differences between the composition of the free amino acids of the two wheats were not

Table II. Free Amino Acids in the Endosperm from a Hexaploid Wheat (C.V. Thatcher) and Its Extracted Tetraploid (Tetrathatcher) during Development (g of N per 100 g of Total Amino Acid Nitrogen from the Endosperm)[a]

Amino acids	Thatcher at days after anthesis					Tetrathatcher at days after anthesis				
	9	14	21	30	49	9	14	21	30	49
γ-Abu	0.98	0.80	0.09	0.02	Trace	0.40	0.47	0.21	0.37	Trace
Trp	0.06	Trace	0.06	0.08	0.03	0.04	0.05	0.03	0.08	Trace
Lys	0.30	0.23	0.12	0.04	Trace	0.30	0.10	0.13	0.13	Trace
His	0.11	0.04	0.02	0.02	Trace	0.05	0.04	0.03	0.05	Trace
Arg	0.18	0.05	0.08	0.03	0.02	0.18	0.03	0.04	0.03	0.03
Asp	0.19	0.22	0.16	0.12	0.01	0.47	0.10	0.07	0.12	Trace
Thr	0.38	0.16	0.07	0.02	Trace	0.30	0.12	0.04	0.06	Trace
Ser	1.6	0.84	0.32	0.07	Trace	1.3	0.74	0.23	0.17	Trace
Asp	1.7	0.29	0.32	0.03	0.02	1.2	0.88	0.23	0.38	0.04
Glu	0.81	0.55	0.86	0.09	0.01	1.1	0.27	0.13	0.04	0.01
Gln	2.7	0.29	0.37	0.10	0.01	2.0	0.91	0.28	0.72	0.02
Pro	1.5	0.05	0.04	0.01	0.01	1.5	0.18	0.05	0.04	0.03
Gly	1.5	1.4	0.25	0.06	Trace	1.0	1.3	0.51	0.15	Trace
Ala	5.9	2.3	0.73	0.07	Trace	3.2	3.2	0.80	0.35	0.01
Val	0.36	0.10	0.05	0.02	Trace	0.15	0.17	0.05	0.06	Trace
Met	0.26	0.05	0.05	0.01	Trace	0.09	0.13	0.05	0.05	Trace
Ile	0.11	0.05	0.04	0.02	Trace	0.05	0.04	0.02	0.02	Trace
Leu	0.14	0.05	0.05	0.02	Trace	0.10	0.06	0.03	0.05	Trace
Tyr	0.05	0.02	0.02	0.01	Trace	0.06	0.02	0.01	0.02	Trace
Phe	0.06	0.02	0.04	0.01	Trace	0.07	0.03	0.01	0.03	Trace
NH_3	1.8	1.2	6.77	0.14	0.02	2.0	1.2	0.64	0.30	0.07

[a] Cysteine and cystine were not determined.

large. In both Thatcher and Tetrathatcher the level of free serine declined steadily to maturity. Asparagine was a major component within the free amino acid pools of both wheats, although it was present in greater proportions in Tetrathatcher both 14 and 30 days after anthesis. Free glutamic acid retained a relatively constant level in Thatcher up to 21 days after anthesis and then declined rapidly. In Tetrathatcher, however, it had declined to a low level as early as 14 days after anthesis. The level of free glutamine decreased rapidly between 9 and 14 days after anthesis in Thatcher. This decrease was more gradual in Tetrathatcher. Unlike Thatcher, the level of free glutamine in Tetrathatcher increased between 21 days after anthesis and 30 days after anthesis. Proline was a major component in the free amino acid pools of both wheats 9 days after anthesis, but had declined to a very low level 5 days later. The level of free glycine dropped rapidly from 14 days after anthesis to maturity in both Thatcher and Tetrathatcher. The largest component within the free amino acid pools of both wheats early in the development process was alanine. γ-Aminobutyric acid (γ-Abu), a nonprotein amino acid, was found in substantial amounts in the free amino acid pools of both wheats, although Tetrathatcher contained a greater proportion of this amino acid from 21 days through 30 days after anthesis. The remaining free amino acids were found in smaller amounts in both wheats and generally declined during endosperm development.

CONCLUSION

Our results showed that the D-genome had very little effect on the amino acid composition of the free amino acids and proteins of wheat endosperm during kernel development. Although rapid changes occurred in the amino acid composition of the free amino acids and protein within the endosperm of both wheats during development, the amino acid compositions of the free amino acids and protein were similar at comparable stages of kernel development.

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