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Dry-Matter Accumulation and Carbohydrate Composition in Developing Normal- and High-Lysine Sorghum Grain[1]

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Eight sorghum [*Sorghum bicolor* (L.) Moench] varieties (four high lysine and four normal) were evaluated 21, 31, and 61 days after flowering (DAF) for changes in total dry-matter accumulation and carbohydrate composition. The chemically induced high-lysine mutant P-721 opaque was not significantly different from P-721 normal in whole-kernel weight, endosperm weight, germ weight, germ percent, and moisture percent as well as in carbohydrate composition. High-lysine natural mutants from Ethiopia (IS 11758, IS 11167, YM-3) showed lower kernel weight, lower endosperm weight, higher percent germ, and higher percent moisture than normal sorghum varieties at all stages of grain development. More significantly, distinct differences were found in the overall carbohydrate profile and the rate of starch synthesis, as well as the level of total sugars at the various stages of grain development in the Ethiopian high-lysine and Ethiopian normal varieties. Between 21 and 31 DAF, starch synthesis was delayed and a concomitant accumulation of sugars occurred in the high-lysine sorghums. The level of sugars was the highest at the late-dough stage (31 DAF), providing a nutritional basis for the Ethiopian tradition of consuming the high-lysine varieties at the late-dough stage.

Cereals provide most of the carbohydrates and about 50% of the annual human dietary protein requirement. This estimate would be even higher if an assessment was made of how much of the diverse cereals are consumed at their respectively perferred immature stages. In Eastern Africa and specifically in Ethiopia, almost every cereal crop is consumed (green-fresh, roast, or boiled etc.) sometime before maturity. Ear roasts of corn (*Zea mays* L.) or sorghum are special delicacies of their respective seasons. In the United States vegetable corn types are widely used before maturity.

Research on protein and carbohydrate improvements of vegetable corn (Barbosa, 1971; Glover and Crane, 1972) utilizing the opaque-2, sugary-2 and other endosperm mutants indicate that double-mutant combinations such as su_1, o_2; sh_2, o_2; and bt_2, o_2 can improve the nutritional quality of maize at immature stages of development. Tossello (1974) evaluated some endosperm mutants and their double-mutant combinations with opaque-2 for their protein and carbohydrate quality at the immature stages of 21 and 42 days after pollination. He reported that the overall carbohydrate profile and protein yield and quality of the mutants and their double-mutant combinations were superior to the normals both at 21 and 42 days after pollination. The data demonstrated that several of the double-mutant combinations may be of interest for their food value as a fresh product (fresh boiled or roasting ear corn) and possibly as a snack food or in dry cereal breakfast food products.

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While similar genes in sorghum have essentially the same pattern of effects, work on vegetable sorghum types is lacking. Sorghum grain has a wide range of variation in endosperm composition. Waxy, floury, corneous, sugary, and high-lysine types exist, as they do in corn. Several workers (Quinby and Martin, 1954; Creech, 1965; Webster, 1965; Gorbet and Weibel, 1972) have reported on the many carbohydrate mutants affecting starch content and the relative properties of amylose and amylopectin in the starch. Singh and Axtell (1973) reported the protein and carbohydrate composition of mature whole-kernel samples of two high-lysine sorghum lines from Ethiopia. Shannon (1968) suggested that all carbohydrates in maize, whether starch, phytoglycogen, or sucrose, begin to accumulate at about the same physiological age. This age was dependent upon environmental conditions during the first 2 weeks following pollination.

At the present time genes in two naturally occuring high-lysine lines in sorghum, IS-11167 and IS-11758 (Singh and Axtell, 1973), and one induced mutant, P-721 opaque (Mohan and Axtell, 1975), are known to change drastically the relative proportion of certain storage proteins in the sorghum endosperm. The chemical composition and nutritional value of these mutants were shown to be superior to any known sorghum lines. However, the effects of these mutations on absolute and relative changes in protein quality and carbohydrate profile in the endosperm during grain development have never been investigated. Farmers in Wollo, Ethiopia (where the two high-lysine lines originated), indicated that they consumed the grain of these lines at the dough stage as head-roast. They also claimed to use them this way because of the sweet flavor of these lines but make no mention of their nutritional quality.

The overall purpose of this study was to evaluate the nutritional value and carbohydrate profile of high-lysine and normal varieties of sorghum at the milk-dough stage

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when they are consumed by sorghum farmers in Ethiopia. In this paper we report on the dry-matter and carbohydrate levels in the developing sorghum grain. Protein and lysine levels are discussed in a separate paper (Ejeta and Axtell, 1987).

MATERIALS AND METHODS

Eight strains of sorghum were used for this study. These included three naturally occurring high-lysine mutants from Ethiopia, IS-11758, IS-11167, and YM-3, and three normal cultivars, BG-5, BG-6, and BG-10, collected by the authors from Wollo, Ethiopia, in 1973. Also included were the chemically induced, high-lysine mutant P-721 opaque and its normal sib parent P-721 normal. The eight varieties were grown at the Purdue Agronomy Farm in three-row plots that were 0.60 m wide and 7 m long.

Three random heads from each of the eight strains of sorghum were sampled at three different stages of maturity: 21 days after flowering (DAF), 31 DAF, and 61 DAF. At each stage panicle branches were cut from central sections of the triplicate head samples, put in labeled beakers, and immediately frozen in dry ice and rushed to the laboratory to be stored at −20 °C. Fifty kernels were removed from each sample, weighed, and lyophilized in a minilyophilizer (Virtis trap model) for 72 h. Lyophilized samples were weighed and placed in hydrolysis vials, stored in a desiccator, and kept in a cold room (−20 °C). These kernels were later dissected with a scalpel and separated into endosperm and germ fractions. The germ and the endosperm were weighed separately. Before the samples were used for chemical analyses, the following data were collected: (a) 50-kernel weight, (b) 50-kernel endosperm weight, (c) 50-kernel germ weight, (d) percent germ, and (e) percent moisture.

Starch was determined by optical rotation measurement on a calcium chloride solution of the endosperm sample prepared according to the procedure described by Shuman and Plunkett (1964). Amylose was determined by blue color measurement on the same calcium chloride solution prepared for starch.

Total sugars and reducing sugars were determined on an 80% ethyl alcohol extract made at 80 °C. Total sugars were determined by the phenol–sulfuric acid colorimetric method and reducing sugars by the Nelson method. Both methods are described by Hodge and Hofreiter (1964). Sucrose content was obtained by calculating the difference between the total sugars and reducing sugars.

Water-soluble polysaccharide (WSP) concentration was determined on a 10% ethyl alcohol extract made at refrigerator temperature on the residue from the total and reducing sugar extraction. The phenol–sulfuric acid procedure was used to determine water-soluble polysaccharides in this extract.

RESULTS AND DISCUSSION

Total Kernel Weight and Endosperm Weight. Means for 50-kernel weight (Table I) at 21, 31, and 61 DAF were not significantly different between p-721 opaque and P-721 normal varieties. There was, however, a highly significant difference between the Ethiopian high-lysine and Ethiopian normal varieties for the same character in all three stages of grain development. Differences in endosperm weight (Table II) between P-721 opaque and P-721 normal were also not significant at 21 and 31 DAF and only slightly significant at maturity, 61 DAF. The three high-lysine varieties from Ethiopia, because of their dented kernels, showed significantly lower kernel weight and endosperm weight than their normal, plump counterparts. An interesting contrast between P-721 opaque (an induced mutant) and the Ethiopian high-lysine lines (natural mutants) is clearly presented in Tables I and II. Table I shows dry-matter accumulation, as measured by whole-kernel weight of four high-lysine and four normal, low-lysine sorghum lines at three developmental stages. In maize, the effect of opaque-2 (o_2) gene was reported to be decreasing kernel and endosperm weight in mature kernels (Barbosa, 1971; Sreeramulu and Bauman, 1970; Tossello, 1974). Unlike opaque-2 maize, there appears to be no different in dry-matter accumulation between opaque and normal P-721. On the other hand, there is significantly different dry-matter accumulation between normal Ethiopian varieties and the high-lysine Ethiopian varieties collected from the same region (Table I). The difference appears to be due to some limitation on starch synthesis between 31 DAF and 61 DAF in the Ethiopian high-lysine varieties as contrasted to the normals that show a faster rate of carbohydrate accumulation.

Germ Weight and Germ Percent. The least significant difference test (LSD) in Table III shows no difference in germ (embryo) weight between the mean values of high-lysine and normal varieties in all three stages of grain development used for this study. On the other hand, there was a different response for germ percent (calculated on the basis of whole-kernel dry weight) in P-721 and the Ethiopian varieties. P-721 opaque and normal showed no difference in either germ weight or in perm percent. The Ethiopian high lysine (*hl*) varieties, while not showing an absolute increase in germ weight, did, however, produce

Table I. Whole-Grain Weight[a] of High-Lysine and Normal Sorghum Genotypes at 21, 31, and 61 DAF

type	genotype	wt,[b] g 21	31	61
high lysine	P-721 opaque	0.739	1.097	1.330
normal	P-721 normal	0.834	1.115	1.253
	difference	0.095	0.018	0.077
high lysine	IS-11758	0.840[a]	1.057[a]	1.268[a]
	IS-11167	0.772[a]	1.013[a]	1.242[a]
	YM-3	0.683[a]	0.880[a]	1.146[a]
	mean	0.765	0.983	1.218
normal	BG-5	0.851[a]	1.069[a]	1.755[a]
	BG-6	0.833[a]	1.231[a]	1.928[a]
	BG-10	1.031[a]	1.292[a]	1.804[a]
	mean	0.905	1.197	1.829
	difference	0.140*	0.214**	0.611**

[a] 50 kernels (dry basis). [b] Means within a column involving the three high-lysine lines and similarly for the three normal lines followed by a common letter did not differ significantly as determined by Duncan's Multiple Range Test at the 0.01 probability level. Differences between means of high-lysine and normal sorghum lines significant according to an LSD test: *, 5% probability; **, 1% probability.

Table II. Endosperm Weight of High-Lysine and Normal Sorghum Genotypes at 21, 31, and 61 DAF[a]

type	genotype	wt, g 21	31	61
high lysine	P-721 opaque	0.663	0.966	0.961
normal	P-721 normal	0.764	1.028	1.090
	difference	0.101	0.062	0.129*
high lysine	IS-11758	0.676[a]	0.937[a]	1.007[a]
	IS-11167	0.652[a]	0.847[a]	0.961[a]
	YM-3	0.492[b]	0.651[b]	0.795[b]
	mean	0.606	0.812	0.921
normal	BG-5	0.734[ab]	0.976[b]	1.425[c]
	BG-6	0.675[b]	1.137[a]	1.684[a]
	BG-10	0.802[a]	0.967[b]	1.572[b]
	mean	0.737	1.027	1.560
	difference	0.131**	0.215**	0.639**

[a] See footnote of Table I.

Table III. Germ Weight of High-Lysine and Normal Sorghum Genotypes at 21, 31, and 61 DAF[a]

type	genotype	wt, g 21	31	61
high lysine	P-721 opaque	0.114	0.186	0.236
normal	P-721 normal	0.125	0.188	0.185
	difference	0.011	0.002	0.051
high lysine	IS-11758	0.192[a]	0.255[a]	0.349[a]
	IS-11167	0.181[a]	0.257[a]	0.367[a]
	YM-3	0.144[a]	0.206[a]	0.342[a]
	mean	0.172	0.239	0.352
normal	BG-5	0.101[a]	0.140[a]	0.243[a]
	BG-6	0.122[a]	0.192[a]	0.299[a]
	BG-10	0.139[a]	0.196[a]	0.269[a]
	mean	0.121	0.176	0.270
	difference	0.051	0.063	0.082

[a] See footnote of Table I.

Table IV. Germ as Percent of Total Kernel Weight in High-Lysine and Normal Sorghum Varieties at 21, 31, and 61 DAF[a]

type	genotype	wt, g 21	31	61
high lysine	P-721 opaque	15.39	16.99	17.70
normal	P-721 normal	14.96	16.94	14.87
	difference	0.43	0.05	2.83
high lysine	IS-11758	22.91[a]	24.19[a]	27.62[a]
	IS-11167	23.45[a]	25.36[a]	29.57[a]
	YM-3	21.12[a]	23.46[a]	29.89[a]
	mean	22.49	24.34	29.03
normal	BG-5	11.87[a]	13.08[a]	16.91[a]
	BG-6	14.61[a]	15.56[a]	15.51[a]
	BG-10	13.49[a]	15.20[a]	15.84[a]
	mean	13.32	14.61	16.08
	difference	9.17**	9.73**	12.95**

[a] Means within a column involving the three high-lysine lines and similarly for the three normal lines followed by a common letter did not differ significantly as determined by Duncan's Multiple Range Test at the 0.01 probability level. Differences between means of high-lysine and normal sorghum lines significant according to an LSD test: *, 5% probability; **, 1% probability.

Table V. Moisture Percentage of High-Lysine and Normal Sorghum Varieties at 21, 31, and 61 DAF[a]

type	genotype	wt, g 21	31	61
high lysine	P-721 opaque	54.74	44.94	17.48
normal	P-721 normal	50.23	43.36	15.18
	difference	4.51	1.58	2.30
high lysine	IS-11758	65.89[a]	63.18[b]	20.34[a]
	IS-11167	66.05[a]	67.71[a]	15.77[a]
	YM-3	68.64[a]	61.02[b]	20.38[a]
	mean	66.86	63.97	18.83
normal	BG-5	57.61[a]	48.82[b]	12.62[a]
	BG-6	58.63[a]	40.39[b]	15.99[a]
	BG-10	56.13[a]	49.05[a]	12.87[a]
	mean	57.46	46.09	13.83
	difference	9.40**	17.88**	3.16**

[a] See footnote of Table IV.

differences in relative germ size (germ percent) at 21, 31, and 61 DAF. The o_2 gene in corn is also associated with larger germ size (Ruschel, 1972). This suggests that the increase in percent germ of these mutants over the normals is one of the factors responsible for their improved protein quality. In addition, the difference in absolute germ size between the *hl* and normal Ethiopian varieties is also worth noting as the germ size for *hl* varieties was consistently greater than the normal (*HL*) varieties. Overall, both germ weight (Table III) and germ percent (Table IV) for all genotypes increased as the season progressed. The differences in magnitude at each stage of grain development were perhaps a reflection of the differences in the rates of synthesis as indicated by the significant genotype × maturity interaction (statistical table not reported).

Moisture Percent. Mean comparisons in Table V indicated other differences in kernel characteristics between Ethiopian *hl* varieties and P-721 opaque. In all three stages, P-721 opaque showed no significantly higher moisture content than P-721 normal. The Ethiopian high-lysine varieties showed significantly higher moisture percentage than the normal varieties at 21, 31, and 61 DAF. This suggests that in P-721 opaque, which is as plump as the normal and with normal starch synthesis, there is limited empty space for moisture, while the dented kernels of the Ethiopian *hl* varieties, with less dry-matter accumulation than in normal varieties, have room for moisture filling. In corn, o_2 is associated with higher moisture in the kernel at maturity (Lambert et al., 1969; Paez et al., 1969). Tossello (1974) also found this to be true for corn 21 and 42 days after pollination.

Carbohydrate Composition. The carbohydrate composition of the eight genotypes at 31 DAF is shown in Table VI. At this stage of grain development, P-721 opaque and P-721 normal were not significantly different in starch (percent of sample), amylose (percent of starch), total sugars (milligrams/gram of sample), reducing sugars (milligrams/gram of sample), sucrose (milligrams/gram of sample), and water-soluble polysaccharides (WSP) (milligrams/gram of sample). The Ethiopian *hl* varieties showed significantly lower starch content than the normal varieties. Amylose as a percent of starch was significantly lower in the *hl* varieties than in normals. There was a 5-fold increase in total sugars in *hl* varieties (av = 172 mg/g of sample) over the normal (*Hl*) varieties (av = 34 mg/g of sample). Reducing sugars in the *hl* varieties (av = 73 mg/g of sample) were more than 3 times that of the *Hl* varieties (av 26 mg/g of sample). This high level of reducing sugars coupled with the more than 10-fold increase in sucrose of the *hl* varieties (av = 100 mg/g of sample) over the *Hl* varieties (av = 8 mg/g of sample) could be the reason why the sorghum farmers of Wollo, Ethiopia, boast of a special sweet flavor for their high-lysine sorghums.

Data in Table VI show significantly higher content of WSP accompanied by lower starch contents for *hl* varieties than in normal varieties at 31 DAF. If the same kind of relationship exists in mature kernels, it would indicate that there is a reduction in the synthesis of starch from WSP in *hl* varieties. To evaluate this hypothesis, the carbohydrate composition at 21, 31, and 61 DAF of the Ethiopian high-lysine line IS-11758 and its normal counterpart BG-10, as well as P-721 opaque and normal, were examined (Table VII).

Data in Table VII show that the carbohydrate profile of the chemically induced high-lysine mutant P-721 opaque and its normal parent are similar at all three stages of grain development. The percent starch in P-721 opaque was 6% higher at 21 DAF than in P-721 normal, indicating perhaps that starch synthesis was initiated earlier in P-721 opaque than in its normal counterpart. Levels of starch as well as water-soluble polysaccharides at maturity (61 DAF) were similar in both P-721 opaque and normal, supporting the similar dry-matter accumulation in the two lines (Table I).

In contrast, there was a striking difference in the overall carbohydrate composition and the rate of starch synthesis, as well as the level of sugars at the various stages of grain development in the Ethiopian high-lysine and Ethiopian normal varieties. Percent amylose at 31 DAF was low in *hl* varieties but caught up with normals at 61 DAF. As

Table VI. Carbohydrate Composition of Four High-Lysine and Four Normal Sorghum Varieties at 31 DAF[a]

type	genotype	starch, % sample	amylose, % starch	total sugars, mg/g sample	reducing sugars, mg/g sample	sucrose, mg/g sample	WSP, mg/g sample
high lysine	P-721 opaque	69.4	24.9	27.0	23.6	3.4	5.3
normal	P-721 normal	72.4	24.1	19.4	14.7	4.7	4.9
	difference	3.0	0.8	7.6	8.9	1.3	0.4
high lysine	IS-11758	46.5[a]	16.5[a]	184.3[a]	80.3[a]	104.0[a]	8.6[b]
	IS-11167	49.3[a]	19.2[a]	169.0[a]	65.3[b]	103.7[a]	7.6[b]
	YM-3	43.0[b]	16.9[a]	164.7[a]	74.9[a]	93.8[a]	17.9[a]
	mean	46.3	17.5	172.7	73.5	100.5	11.4
normal	BG-5	67.7[a]	22.2[a]	40.2[a]	26.1[b]	14.1[a]	8.4[a]
	BG-6	71.7[a]	23.2[a]	16.7[b]	12.1[c]	4.6[b]	4.7[b]
	BG-10	65.6[b]	22.6[a]	45.7[a]	39.4[a]	6.4[b]	3.6[b]
	mean	68.3	22.7	34.2	25.9	8.4	5.6
	difference	22.0**	5.2*	138.5**	47.6**	92.1**	5.8*

[a] See footnote of Table IV.

Table VII. Carbohydrate Composition of Two High-Lysine and Two Normal Sorghum Varieties at 21, 31, and 61 DAF[a]

maturity	type	genotype	starch, % sample	amylose, % starch	total sugars, mg/g sample	reducing sugars, mg/g sample	sucrose, mg/g sample	WSP, mg/g sample
21 DAF	high lysine	P-721 opaque	70.5	26.2	48.1	17.9	30.2	5.9
	normal	P-721 normal	64.3	26.2	51.6	24.1	27.5	6.9
		difference	6.2	0.0	3.5	6.2	2.7	1.0
	high lysine	IS-11758	46.3	20.2	148.0	48.7	99.3	9.2
	normal	BG-10	57.1	22.1	85.4	34.6	50.8	12.8
		difference	10.8*	1.9	62.6**	14.1*	48.5**	3.6
31 DAF	high lysine	P-721 opaque	69.4	24.9	27.0	23.6	3.4	5.3
	normal	P-721 normal	72.4	24.1	19.4	14.7	4.7	4.9
		difference	3.0	0.8	7.6	8.9	1.3	0.4
	high lysine	IS-11758	46.5	16.5	184.3	80.3	104.0	8.6
	normal	BG-10	65.6	22.6	45.7	39.4	6.3	3.6
		difference	19.1**	6.1*	138.6**	40.9**	97.7**	5.0*
61 DAF	high lysine	P-721 opaque	72.6	18.8	13.3	3.6	9.7	11.4
	normal	P-721 normal	71.1	22.3	12.8	2.7	10.1	10.6
		difference	1.5	3.5	0.5	0.9	0.4	0.8
	high lysine	IS-11758	67.7	20.4	15.3	1.3	14.0	15.7
	normal	BG-10	75.8	21.9	9.6	2.2	7.4	9.1
		difference	8.1*	1.5	5.7	0.9	6.6*	6.6*

[a] Differences between means of high-lysine and normal sorghum lines declared significant according to an LSD test: *, 5% probability; **, 1% probability.

would be expected, starch contents of both the *hl* (46%) and normal (57%) varieties were low at 21 DAF. At 31 DAF, however, the starch content of IS-11758 (*hl*) remained at 46%, whereas in BG-10 (*Hl*), it increased to 65%. Between 31 and 61 DAF starch synthesis rate increased in the *hl* variety IS-11758 as compared to the normal check BG-10. These data suggest that a permanent disruption in the conversion of water-soluble polysaccharides to starch in IS-11758 did not occur. Rather it indicated that there was only a delay in starch synthesis in lieu of a dramatic accumulation of sugars in IS-11758 at the soft dough (21 DAF) and the late dough (31 DAF) stages of grain development. This is unlike high-lysine corn where a significant reduction in starch content was reported (Barbosa, 1971; Tossello, 1974).

The differences in sucrose content (2–17-fold) between 21 and 31 DAF in the high-lysine and normal Ethiopian varieties could be the reason for the claim by farmers in Wollo, Ethiopia, that the *hl* varieties have a sweet taste when consumed at the soft dough or late dough stages. The Ethiopian high-lysine varieties possess a combination of desirable attributes, including higher percent germ (Table IV), higher total content of lysine-rich proteins (Singh and Axtell, 1973), higher tryptophan levels (Hassen et al., 1986), and higher niacin content (Pant, 1975), as well as a very high level of sugars (Table VII) that make these mutants good candidates for specialty food products.

SUMMARY AND CONCLUSIONS

The eight genotypes showed significant differences for 50-kernel weight, 50-kernel endosperm weight, 50-kernel germ weight, germ percent, and moisture percent at all three stages of grain development considered for this study. A striking difference was also noted between the two types of genetic mutants for lysine in sorghum. The high-lysine (*hl*) mutants for Ethiopia showed significantly lower kernel and endosperm weight than the normal varieties in all three stages of grain development, whereas P-721 opaque, an induced mutant, remained equal to its normal sib parent in whole kernel weight at the same stages. The difference appears to be due to a delay in starch synthesis in the Ethiopian high-lysine varieties. In general, germ weight and germ percent for all genotypes increased as the season progressed. The differences in magnitude at each stage of grain development are thought to be a reflection of the differences in the rate of synthesis as indicated by the significant genotype × maturity interaction. The *hl* Ethiopian varieties showed considerably higher germ percent than the normal varieties. These lines also showed significantly higher moisture percentage than normal varieties at 21, 31, and 61 DAF. P-721 opaque did not show significantly different germ size, germ percent, or moisture percent from P-721 normal. As would be expected, moisture level decreased in later stages of grain development for all of the genotypes.

At the milk-dough stage of grain development, 31 DAF, the Ethiopian *hl* varieties showed lower starch (percent of sample) and lower amylose (percent of starch) than the normal varieties. The total sugar level in *hl* lines was 4 times that of the normal varieties at 31 DAF. Also a 3-fold increase in reducing sugars and a 17-fold increase in sucrose occurred in the *hl* varieties when compared with the normal varieties. This could be the reason why sorghum farmers in Ethiopia claim a special sweet flavor for their high-lysine sorghum varieties. This acclaimed taste coupled with the superior protein qualtiy and nutritional value (Ejeta and Axtell, 1987) provide a nutritional basis for the established tradition of consuming the *hl* varieties exclusively at the dough stage in Ethiopia. Further, such combinations of desirable components make the *hl* lines from Ethiopia unique when compared with any of the other known endosperm mutants in corn and sorghum.

Registry No. Starch, 9005-25-8; amylose, 9005-82-7; sucrose, 57-50-1; lysine, 56-87-1.

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Effect of Lipids and Carbohydrates on Thermal Generation of Volatiles from Commercial Zein

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Model systems composed of zein, regular and waxy corn starch, and corn oil were heated in an oven at 120 and 180 °C. The volatile compounds generated were analyzed by gas chromatography (GC) and gas chromatography/mass spectrometry (GC/MS). Maillard reaction products and carotenoid decomposition products increased in the presence of corn oil. Two pyrazines identified from the increased number of volatiles generated at the higher temperature were 2-methyl-3(or 6)-pentylpyrazine and 2,5-dimethyl-3-pentylpyrazine. These two pyrazines were characterized as lipid–protein–carbohydrate interaction products.

Thermal interactions between proteins, carbohydrates, and lipids in foods are complicated by extrusion processing. During extrusion, shear force and pressure, as well as temperature, influence the interactions. Protein–carbohydrate interactions during the heat treatment of foods have been given a great deal of attention, and the mechanism proposed by Hodge (1953) for the amino–carbonyl reaction of carbohydrates with proteins has been widely accepted. However, recent discoveries by Hayashi and Namiki (1980, 1981, 1986) indicate that free-radical products form during the early stage of the browning reaction of sugar–amino compounds. Lipids, especially the polyunsaturated lipids, are a well-known source of free radicals. As part of a larger cooperative effort to increase the current understanding of extrusion processing, the effects of corn oil and corn

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