Technical

A Comparison of the Lipids of Triticale, Wheat and Rye Grown under Similar Ecological Conditions

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ABSTRACT

Samples of triticale, wheat and rye grown in the same season at the same location in southern United States were analyzed for amounts and types of lipids and for fatty acid composition. Selected kernels of the 3 cereals were separated into anatomical parts (bran, endosperm and germ) and analyzed for amounts and types of lipids, and the anatomical parts of triticale were analyzed for fatty acid composition. The triticale contained several times more phosphatidylcholine and phosphatidylethanolamine than did the wheat and rye. Marked differences in lipid composition were found in the bran, endosperm and germ fractions; and some differences were found among corresponding fractions of the 3 cereals. The bran fractions were rich in steryl esters and contained no glycolipids or phospholipids. Fatty acid compositions of the lipids from the anatomical parts of triticale exhibited some difference.

INTRODUCTION

Triticale, a cross of wheat (usually *Triticum durum*) and rye (*Secale cereal*), is the first man-made cereal. This intergeneric hybrid is believed to possess greater environmental adaptability than either of its parents, thus permitting the growing of a high-protein crop on marginal land (1). Although most reports on the composition of triticale have been concerned with the protein and carbohydrate fractions (2-9), some lipid analyses have been reported. Chung and Tsen (10) determined types and amounts of lipids in triticales grown in Mexico and Kansas and compared these with the lipids from samples of wheat and rye grown in different areas of Kansas. Lorenz and Maga (11) reported on the major fatty acids in the lipids of triticale and wheat flours from grains produced in Colorado. Price and Parsons (12) analyzed samples of wheat, rye and triticale for glycolipids and phospholipids and for the fatty acid composition of the total lipids. Apparently, neither lipid analyses of the anatomical parts of triticale (bran, endosperm and germ) nor analyses of the whole-grain and anatomical parts of triticale, wheat and rye grown at the same location under similar agricultural practices and environmental conditions have been reported.

MATERIALS AND METHODS

The 3 cereal samples were supplied by Alabama Agricultural and Mechanical University, Huntsville, AL, from crops grown in Spring 1976 near Normal, AL, under similar agricultural conditions and practices. The varieties were triticale, AM 2147; wheat, Arthur 71; and rye, Abruzzi. Moisture contents, determined by the air-oven method (13), were 16.1% for the triticale, 17.2% for the wheat and 15.8% for the rye. The whole-grain samples were stored in the laboratory at -20 C until analyzed.

[1] One of the facilities of the Southern Region, Science and Education Administration, U.S. Department of Agriculture.

Milling and Lipid Extraction

Whole-grain kernels were ground in a Wiley mill immediately preceding extraction. The extractions were conducted at 40 C with 20-g samples. In the first stage, 400 ml of 80% 1-butanol (reagent grade) and 20% water, weight basis, was mixed with the ground sample for 2 hr under nitrogen. The lipid-containing alcohol-water solution was separated from the solids by filtration through a fritted-glass filter (fine, 4-5.5 μm pore size). The solids were washed back into the extraction flask with 160 ml of 80% 1-butanol, and the extraction procedure was repeated. After the second filtration, the solids were washed with 40 ml of 80% 1-butanol. The filtrates and wash solution were combined, 196 ml of water was added to ensure an azeotropic distillation and the solvents were distilled off at ca. 125 mm until an estimated 2-3 ml of solvent remained. The concentrated lipids were extracted 4 times with 10-ml portions of hot chloroform. The extracts were filtered through a fine, fritted-glass filter, and the solvent was removed by passing a stream of nitrogen over the solution at 1 atm pressure and room temperature. The lipids were stored under nitrogen at -20 C until analyzed.

Dissection and Extraction

For separation into anatomical parts (endosperm, bran and germ), 50 of the largest and best-quality kernels of each cereal were hand picked. The kernels were soaked in cold, deionized water for 1 hr before manual dissection with a number 11 surgeon's knife under a stereoscopic dissecting microscope. The water treatment caused the parts to swell and become softer and more pliable. Germ and endosperm fractions were dissected fairly cleanly from the surrounding tissues, but the bran fraction was cut away less cleanly and may have contained aleurone cells and outer parts of the endosperm. Cutting away the bran in the crease area of the triticale kernels was most difficult. The fractions from each cereal were stored under nitrogen in a dessicator at -20 C. Lipid extraction was begun after a constant dry weight had been attained.

Each of the 9 fractions was ground with a mortar and pestle and washed into a 50-ml, round-bottom flask with 30 ml of 80% 1-butanol. A Teflon-coated, magnetic stirring bar was added, and the air over the solution was displaced with nitrogen. Mixing and extraction were conducted for 10 hr at 40 C. The solids and liquid were separated with a fine, fritted-glass filter. The solids were washed back into the extraction flask with 30 ml of 80% 1-butanol, and the mixing and extraction were continued for another 6 hr. Solids and liquid again were separated by filtration, and the solids were washed with additional solvent. The solvent was removed from the combined liquid fractions by distillation under reduced pressure and stripping with nitrogen. The residue was dissolved in warm chloroform, and the mixture

TABLE I

Lipid Content of the Cereals and Distribution of Mass and Lipids among the Anatomical Parts[a]

Cereal	Total lipids (%)	Distribution of mass (%)			Distribution of lipids (%)		
		Bran	Endosperm	Germ	Bran	Endosperm	Germ
Triticale	3.00	4.58	91.2	4.19	2.86	55.2	41.9
Wheat	3.31	4.01	92.0	4.02	3.39	62.7	33.8
Rye	2.99	4.48	91.1	4.36	3.20	62.3	34.5

[a]Calculated on dry-weight basis.

was filtered. The chloroform was evaporated with a stream of nitrogen. The lipid residue was stored under nitrogen at -20 C.

One set of anatomical parts of triticale was extracted in a similar manner with hexane, except that the chloroform extraction was omitted.

Analysis

The proportions of 11 of the major lipids previously shown to be present in cereal grains (10,12,14) were determined by quantitative thin layer chromatography (TLC). Silica gel plates (precoated, Silica Gel 60, No. 5763, EM Laboratories, Inc.) were employed. All spots were identified with standards obtained from Applied Science Laboratories, Inc., State College, PA. Chloroform was the developing solvent for the neutral lipids; chloroform/methanol/water (75:25:4, v/v), for the glycolipids; and chloroform/methanol/water/aqueous ammonia (65:35:4:0.2, v/v), for the phospholipids. After development and drying of the plates, the spots were visualized by spraying with an aqueous solution containing 3% cupric acetate and 8% orthophosphoric acid and charring at 175 C for 15 min (15). Individual spots were quantified with a scanning densitometer. Calibration curves were developed for the quantifications.

Fatty acid composition of the lipids from the whole grain and fractions were determined by gas liquid chromatography (GLC). The fatty acids were converted into methyl esters by saponification and treatment with boron trifluoride/methanol, according to Metcalfe (16). The gas liquid chromatograph (Barber-Coleman, Model 20) was equipped with a tritium detector and a stainless-steel column (1/8 in. x 15 ft) packed with Gas Chrom P, 80-100 mesh, coated with 12% DEGS (stabilized).

RESULTS AND DISCUSSION

The total lipids extractable with 80% 1-butanol (Table I) were somewhat higher for the wheat than for the rye and triticale, but the differences among the 3 cereals are not significant. The values are within the generally accepted ranges (10,12). Water-saturated 1-butanol (ca. 80% 1-butanol) is the most effective solvent for extracting lipids from ground cereals. Under similar conditions, hexane extracts from triticale only ca. 3/4 this amount of lipids (17). However, 80% 1-butanol does not extract all lipids. Subjecting the meal residue from the 80% 1-butanol extraction to an acid hydrolysis yields ca. 0.5% fatty material, calculated on the basis of the original weight of ground triticale (17).

The proportion of bran, endosperm and germ in each of the 3 cereals was practically constant. The absolute differences in the percentage of an anatomical part, such as bran, need to be considered in light of the *manual* separation of bran and endosperm.

The distribution of lipids among the anatomical parts of the cereals (Table I) was less uniform than was the distribution of the mass. The calculated proportion of lipid in the bran fractions is based on a lipid content of 1.65% for the triticale, 2.42% for the wheat and 1.05 for the rye. For each cereal, over half of the total lipids extracted came from the endosperm fraction (Table I), even though the lipid content per unit weight of fraction was low—1.60% for the triticale, 1.96% for the wheat, and 2.45% for the rye. The proportion of lipid (33.8 to 41.9%) in the germ fractions was calculated from lipid contents of 26.4% for the triticale, 24.2% for the wheat and 20.1% for the rye.

The lipids from the whole-grain cereals were analyzed only for certain components for which good standards were available (Table II). Consequently, the total percentages are below 100 for each cereal. Most of the lipids not identified and listed were of the polar type. For each cereal the so-called "neutral lipids" (glycerides, free fatty acids and steryl esters) amounted to slightly over 1/3 of the total lipids. The neutral lipid content of 38.0% for triticale was only slightly above that for wheat (35.0%) and rye (34.9%). Differences were greater among the polar lipids. The triticale contained several times more phosphatidylcholine (PC)

TABLE II

Composition of Lipids from Whole-grain Cereals

Lipid	Composition (wt %)		
	Triticale	Wheat	Rye
Monoglycerides	2.40	5.70	3.71
Diglycerides	5.50	2.85	1.14
Triglycerides	22.6	20.1	23.1
Free fatty acids	5.00	3.50	5.10
Steryl esters	2.50	2.90	1.85
Monogalactosyl diglycerides	2.50	2.93	3.73
Digalactosyl diglycerides	8.00	12.00	11.10
Lysolecithin	2.80	4.80	4.00
Phosphatidylcholine	4.30	1.20	0.75
Phosphatidylethanolamine	5.90	0.48	0.12
Phosphatidylinositol	1.70	1.90	2.75

TABLE III

Fatty Acid Composition of Lipids from Whole-grain Cereals[a]

Fatty acid	Composition (wt %)[b]		
	Triticale	Wheat	Rye
C_{14}	0.38	0.25	0.12
C_{15}	0.46	0.26	0.22
C_{16}	20.1	17.8	14.8
$C_{16:1}$	1.42	0.26	1.18
C_{18}	0.94	1.23	0.78
$C_{18:1}$	11.0	16.5	17.1
$C_{18:2}$	59.0	56.5	57.7
$C_{18:3}$	5.26	6.56	7.36
$C_{20:1}$	1.43	0.70	0.71

[a]Some fatty acids present in trace amounts are not included.
[b]Calculated as methyl esters.

TABLE IV

Lipid Composition of Anatomical Parts of Cereals (wt %)[a]

Component	Triticale			Wheat			Rye		
	Bran	Endosperm	Germ	Bran	Endosperm	Germ	Bran	Endosperm	Germ
Monoglycerides	6.60	5.00	-	6.60	6.60	-	10.0	2.66	-
Diglycerides	8.70	5.70	1.30	3.90	3.50	2.50	10.6	1.60	3.20
Triglycerides	14.7	14.7	17.3	14.3	11.7	19.0	6.8	11.2	21.3
Free fatty acids	11.3	5.93	6.67	14.5	3.53	8.33	9.87	3.67	9.20
Steryl esters	30.0	2.67	5.0	23.5	0.67	7.67	14.3	2.52	4.06
Monoagalactosyldiglycerides	–[b]	4.93	-	-	7.06	-	-	5.2	-
Digalactosyldiglycerides	-	9.93	-	-	15.3	-	-	13.3	-
Lysolecithin	-	2.40	-	-	1.28	-	-	6.44	-
Phosphatidylcholine	-	3.64	5.2	-	2.50	4.10	-	0.60	6.62
Phosphatidylethanolamine	-	15.2	2.2	-	20.1	1.8	-	16.0	1.00
Phosphatidylinositol	-	3.24	8.80	-	2.44	7.92	-	-	9.92

[a]Lipids analyzed only for components listed.
[b]–Means not detected.

and phosphatidylethanolamine (PE) than did the wheat and rye.

Practically, the fatty acid compositions of the lipids from the whole-grain were the same for the 3 cereals (Table III). The largest deviations found were the relatively high content of palmitic acid and low content of oleic acid in the triticale lipids. Over 95% of the total fatty acids from each cereal consisted of palmitic, oleic, linoleic and linolenic. As noted in Table III, some fatty acids present in trace amounts were not included. These minor fatty acids are included in Table V.

Marked differences in lipid composition were found in the anatomical parts (Table IV). No monoglycerides were found among the lipids from the germ fractions of the grains. At least, they were not measurable by the TLC techniques used. The diglyceride content of the bran, endosperm and germ fraction of triticale and rye varied considerably, whereas the diglyceride contents in the lipids from the 3 fractions of wheat were more uniform. Free fatty acids and steryl esters were highest in the lipids from the bran fractions. Steryl esters accounted for 30% of the bran lipids from tricale and 14.3% of the bran lipids from rye. Although steryl esters were the largest component of the bran lipids, their concentration varied considerably among the 3 cereals.

Monogalactosyl diglycerides, digalactosyl diglycerides and lysolecithin were found only in the endosperm fractions of the 3 cereals. In the endosperm lipids from each cereal, the proportion of digalactosyl diglycerides was about double that of the monogalactosyl diglycerides.

None of the bran fractions contained phospholipids, and the endosperm of rye contained no phosphatidylinositol. PE was the major lipid in the endosperm of each of the 3 cereals and amounted to 20.1% of the lipids in the endosperm of wheat. In the germ fraction, the ratio of PC to PE exceeded 2:1.

Marked differences were observed in the fatty acid composition of lipids from the anatomical parts of triticale extracted with 80% 1-butanol (Table V). The bran lipids contained higher proportions of most of the shorter chain fatty acids, including lauric, myristic, and palmitic. Stearic acid in the bran lipids amounted to 10.2%, but only 2.00% and 1.11% in the endosperm and germ lipids, respectively. On the other hand, the bran lipids contained only 31.8% linoleic acid, whereas the endosperm and germ lipids contained 60.0% and 62.4%, respectively. The fatty acids of the bran lipids were 61.5% unsaturated, and those from the germ lipids were 87.0% unsaturated.

TABLE V

Fatty Acid Composition of Lipids from Anatomical Parts of Triticale[a]

Fatty acid	Extracted with 80% 1-butanol			Extracted with petroleum ether		
	Bran	Endosperm	Germ	Bran	Endosperm	Germ
C_{12}	2.32	0.38	0.09	-	-	0.42
ECL 13.58	0.23	0.51	-	-	-	-
C_{14}	2.01	0.17	0.29	-	-	-
ECL 14.70	0.74	0.16	-	-	-	-
C_{15}	0.72	0.83	0.23	-	-	0.20
$C_{14:2}$	0.24	0.05	0.27	-	-	-
C_{16}	20.7	18.2	10.9	23.2	14.6	19.1
$C_{16:1}$	2.80	0.84	0.76	-	0.45	1.46
C_{17}	0.87	0.16	0.07	-	-	-
$C_{16:2}$	0.53	0.04	-	-	-	-
$C_{17:1}$	0.53	0.11	0.07	-	-	-
C_{18}	10.2	2.00	1.11	1.21	1.14	1.66
$C_{18:1}$	21.2	12.5	16.2	23.2	15.1	17.7
$C_{18:2}$	31.8	60.0	62.4	50.0	65.0	52.2
ECL 19.73	0.11	–[b]	-	-	-	-
C_{20}	0.62	0.20	0.17	-	-	-
$C_{18:3}$	3.17	3.24	6.54	2.44	3.72	5.62
$C_{20:1}$	1.23	0.59	0.83	-	-	1.67

[a]Calculated as methyl esters.
[b]–Means not detected.

Extraction of the component parts of the triticale with hexane yielded lipids having a substantially different fatty acid pattern from that obtained with 80% 1-butanol. The hexane extracted only ca. 74% of the lipids obtained by extraction with 80% 1-butanol followed by extraction of the butanol water solubles with chloroform. The hexane extractables are less tightly bound in the tissues and are called "free lipids" by some investigators. The hexane extractables were essentially free of compounds containing odd-carbon fatty acids, such as C_{15} and C_{17}. Others have demonstrated that hexane extracts a considerably lower proportion of polar lipids than does water-saturated 1-butanol (10).

ACKNOWLEDGMENT

B. Singh, Alabama Agricultural and Mechanical University, Huntsville, AL, provided the cereal samples.

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[Received March 10, 1980]

⁂Quality of Oil from Acid Delintered Cottonseed

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ABSTRACT

Oils were extracted experimentally by direct solvent and prepress solvent processes from saw delintered cottonseed as well as from some of the same seed delintered with dilute sulfuric acid. Oils were refined, bleached and deodorized. Deodorized oils were stored in an air oven at 60 C for 30-35 days. Approximately twice a week, stored samples were analyzed for peroxide value. No significant differences in rate of increase of peroxide value were found among the oils.

INTRODUCTION

Delintering of cottonseed with saw or abrasive delintering machines is a standard procedure in cottonseed oil mills. Delintering is done primarily to decrease losses of oil and kernels in hulls and to decrease operating problems associated with linters on seed (1). However, the linters have values of their own (2).

In recent years, some segments of the cottonseed industry have been considering alternatives to mechanical delintering. One is acid delintering, such as that applied to cotton planting seed. A previous paper described economic evaluation of gaseous HCl and dilute sulfuric acid delintering applied to oil mill use (3). Preliminary investigations of qualitites of oil and meal from the dilute sulfuric acid process were made. This paper reports the results of those studies.

Cotton Inc. developed the dilute sulfuric acid process for planting seed after several years of work and publicized it during 1975-77 (4-8). The process is now in use by several commercial seed companies (J.K. Jones, personal communication).

In this process, seed is wetted with sulfuric acid (10% concentration) with a pickup of ca. 10% of the weight of seed (6). Wetted seed is passed through a rotary dryer which removes the water from the acid, leaving concentrated acid on the linter fibers. Under the influence of acid and dryer heat of ca. 52 C maximum (125 F), the linters become brittle. Tumbling action of the dryer and a subsequent rotary abrader remove embrittled linters and suspend loose linters in the drying air stream. Lesser degrees of delintering than are required for planting seed may be suitable for oil mill purposes. These would be achieved by lower amounts and/or concentrations of acid.

Iron salts are known to increase the rate of oxidative deterioration of soybean oils (9) and of other unsaturated vegetable oils (10). Because of the contact between sulfuric acid and iron in the acid delintering machinery, presence of iron salts in the seed, which might find their way into oil, seemed likely. Therefore, measurement of the oxidative stability of the oils was desired.

Oven storage, in combination with organoleptic evaluations and peroxide determinations may be employed to measure oxidative rancidity tendencies of oils (11), and this method was selected for use.

MATERIALS AND METHODS

This study compared oils extracted from the same lot of seed, half of it saw delintered and the other half acid delintered. Seed was collected in an oil mill after cleaning and first cut saw delintering. Half of it was given a second delintering cut. The other half was transported to a commercial planting seed plant where it was acid delintered. After delintering, the seed was treated with anhydrous ammonia to neutralize residual acid. Both the oil mill and the seed plant were in Mississippi.

Both types of seed were hulled and separated in pilot plant machinery in College Station, TX. Material balances were made on this operation with accumulation and weigh-