

Lipids and Fatty Acid Composition of Riso 1508 and Normal Barley

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ABSTRACT

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The crude lipid contents of Riso 1508 and Bonanza barleys were determined by six procedures. After purification, Riso contained 100, 65, 84, 88, 116, and 76% more lipids than did Bonanza for procedures 1-6 respectively. Riso averaged more palmitic and oleic but less linoleic acids than did Bonanza. The neutral lipids formed 74.9 and 65.2%, the glycolipids 7.3 and 25.6%, and the phospholipids 17.8 and 9.2% in Riso and Bonanza, respectively. In Riso, the embryo (including the scutellum) formed, on the average, 5.3% of the total seed weight, compared to 3.0% in Bonanza and 3.5% in Fairfield. The embryo contributed 23.8% of the total lipids in Riso, compared to 22.3-22.6% in Bonanza and Fairfield, when total lipids were

determined by procedure 6 (gas chromatography). With ether as the solvent, the contribution of embryo to total lipids was 28.0% for Riso and 37.3% for Fairfield. The rest of the lipids were present in the endosperm fraction. The data suggested that an increased embryo size in Riso may not be totally responsible for a higher lipid content in this barley compared to that in Bonanza and Fairfield. Compared to the endosperm, the embryo in all three varieties contained two to three times more linolenic acid but less linoleic acid, the major fatty acid. Riso contained about two and one-half times more lipoxygenase activity than did Bonanza.

In previous publications (Bhatty et al 1974, 1975, 1979) we have suggested that digestible energy of barley can be substantially improved, particularly for monogastric animals, by increasing the lipid content of barley from the present average of about 2.0% (Bhatty et al 1974, Price 1972) to 3.0 or possibly 4.0%. The latter value increases the lipid content of barley to close to that of corn, an ideal feed grain. Such an objective requires a genotype of barley having a high lipid content that is stable and genetically controlled.

Parsons and Price (1974) screened the USDA World Barley Collection and found three lines of barley with lipid content from 4.0-4.6%; line CI 12116 had 35% more lipids than did the check cultivar Prilar. However, the lipid content of CI 12116 grown under Canadian conditions was similar to that in local barleys (Fedak and de la Roche 1977). In 1976 Munck (1976) and later Tallberg (1977) reported that the lipid content of the barley mutant Riso 1508 was 4.1%, 66% higher than that of Bomi, from which the mutant was obtained by treatment with a chemical mutagen ethyleneimine. More recently, Shewry et al (1979) and Welch (1978) also reported higher levels of lipids in Riso 1508 and Hipoly barleys. Our data on Riso 1508 obtained from different sources and grown under Saskatchewan conditions further confirmed the high lipid content of Riso 1508 (Bhatty and Rossnagel 1979). Thus the mutant may provide a gene source for increasing the lipid content of Canadian barleys.

In this paper, we report the crude and purified lipids, lipid classes, fatty acid composition, distribution of lipids in seed, and lipoxygenase activity of Riso 1508 (a two-row barley), Bonanza (a six-row malting barley), and Fairfield (a two-row feed barley with normal lipid content) to complement data previously published (Bhatty and Rossnagel 1979). One objective was to obtain

information on the lipids of Riso 1508 compared to those of regular Canadian barleys.

MATERIALS AND METHODS

Riso 1508 (Riso) was grown in 1978 and 1979 in observation nurseries at Saskatoon from seed that originally came from H. Doll of Denmark. Bonanza barley of the 1978 crop was purchased from a local seed company. Fairfield was of the 1979 crop grown in experimental plots at the University of Saskatchewan, Saskatoon. The seed was stored at 4°C and, when needed, ground to meal in a micro Beating mill (Canadian Laboratory Supplies, Toronto) with 1.0-mm screen. Except where indicated, all data were obtained from Riso and Bonanza, both of the 1978 crop, and are means of duplicate determinations.

Crude lipids were determined by six procedures. Procedure 1 was a 24-hr Goldfish extraction with chloroform/methanol (2:1, v/v) based on the procedure of Folch et al (1957). The meal-to-solvent ratio was 1:20. Procedure 2 was extraction with benzene/ethanol (4:1, v/v) for 18 hr under the same conditions. Procedure 3 was that of Melton et al (1979). Procedure 4 was that of Bligh and Dyer (1959). In procedure 5, 2 g of meal was extracted with 40 ml of petroleum ether for 5 hr in a Goldfish extraction apparatus. Procedure 6 was a gas-liquid chromatography method in which oil was transmethylated without prior extraction, using 2% H₂SO₄ in methanol (Welch 1977). The crude lipids obtained from the first five extraction procedures were freed of solvent, dried at 50°C under vacuum for 30 min to remove traces of moisture, and weighed.

Purification of Crude Lipids

The crude lipid extracts were freed of solvents. The residue was transferred to 100 ml of chloroform/methanol (2:1, v/v) and mixed

with 0.2 volumes of 0.04% calcium chloride (Folch et al 1957). The mixture was allowed to separate for 1 hr at 4°C, after which the chloroform layer was removed. An aliquot was dried to determine the lipid content. The rest was freed of chloroform in a rotary evaporator. The residue was dissolved in diethyl ether; the insoluble materials were removed by filtration through GF/C microfiber filter paper. The clear ether filtrate was dried and the contents weighed. A nitrogen atmosphere was maintained throughout the extraction and purification of the lipids.

Fatty Acid Composition

Fatty acid composition was determined with a Hewlett-Packard 5710 gas chromatograph equipped with 3385 A Automation system. Methyl esters were prepared by adding about 10 mg of the purified lipids to 1.0 ml of hexane containing 0.5 ml (2.0 mg) of heptadecanoic acid as internal standard, 1.5 ml of sodium methylate (200 mg dissolved in 15 ml of methanol/benzene/petroleum ether, 10:2.5:2.5, v/v), one drop of bromophenol blue, and 0.4 ml of 1.0*N* HCl. The mixture was neutralized to blue color with sodium carbonate. After allowing the phases to separate, 1–2 µl of the clear benzene/petroleum ether layer was injected onto a glass column (150 cm × 2.0 mm, id) packed with GP 3% SP-2310/2% SP-2300 on 100/120 Chromosorb WAW (lot No. F 15270) obtained from Supelco Inc., Bellefonte, PA. The carrier gas (nitrogen) flow rate was 40 ml/min and the operating temperatures were: oven, 175°C; injector port and detector, 250°C each. Fatty acid composition was calculated as:

$$Y = \frac{\text{area } Y \times \text{response } Y \times \text{amount of IS} \times \text{DF}}{\text{area IS} \times \text{response IS}}$$

where Y is the fatty acid, IS the internal standard, and DF the dilution factor. Each fatty acid was expressed as percent of the total fatty acids.

Silicic Acid Column Chromatography

The crude lipid was fractionated into neutral lipids (NL), glycolipids (GL), and phospholipids (PL) on silicic acid column chromatography. The crude lipid was extracted by homogenization according to the solvent ratio in procedure 1. The homogenate was filtered and freed of the chloroform/methanol solvent, and the residue was taken up in diethylether. The ether extract was filtered through glass fiber filter paper, the filtrate reduced in volume, and 2–3 ml of samples applied onto a 15.0 × 2.2 cm glass column packed with silicic acid (Sigma, 60–200 mesh) washed according to Hirsch and Ahrens (1958). The NL, GL, and PL fractions were eluted with four column volumes each of ether, acetone, and methanol,

respectively. The eluted fractions were evaporated to dryness and weighed. For fatty acid and phosphorus analyses, the fractions were dissolved in small volumes of the eluting solvents and an aliquot taken for the preparation of methyl esters as described earlier. Another aliquot was taken for the determination of total phosphorus (Chen et al 1956).

Distribution of Lipids in Embryo and Endosperm

The barley was dehulled by treatment with 50% sulfuric acid (Essery et al 1956), dried, and its moisture content determined. The dehulled seed was soaked in water for 4 hr at room temperature. The embryo portion of grain containing the scutellum was manually excised from the endosperm. The embryo and endosperm fractions were dried under vacuum for 2 hr at 50°C. They were then ground and their moisture content determined by drying at 50°C for 16 hr. The ground fractions were used for the determination of lipids by procedure 5 and for fatty acid analysis (Welch 1977).

Lipoxygenase Activity

One gram of meal was extracted at 4°C with 100 ml of 0.2*M* sodium phosphate buffer, pH 6.8, for 30 min and the extract centrifuged at 12,000 × *g* for 30 min. The supernatant was made up to 100 ml with the buffer and used as enzyme source. The lipoxygenase activity (LA) was assayed at 25°C with a substrate of linolenic acid, at a concentration of 1.5 mM, that had been dissolved in 50 µl of Tween 20 and to which 0.1*N* KOH had been added until the mixture was clear (ca 5 ml). The assay mixture contained 0.3 ml of the substrate, 2.7 ml of the phosphate buffer, and 10 µl of the enzyme preparation. After the addition of the enzyme, the absorbance of the mixture was read at 234 nm on a Perkin-Elmer recording spectrophotometer against an appropriate blank over a 7-min period. The absorbance of the product, hydrogen peroxide, was linear up to this time. One unit of LA is defined as an increase in absorbance of 0.1/min/g of dry meal under the assay conditions.

RESULTS AND DISCUSSION

Six procedures, most of them commonly used for the extraction of lipids from plant materials, were selected for the determination of lipids from barley. The aim was not to select a method of extraction per se but to establish the lipid content of Riso barley by using different methods. The extraction conditions for each extraction procedure were first fully established; the repeatability of the six procedures varied between 96 and 99%.

The crude lipids determined by six procedures varied from 4.0 to 7.4% for Riso and from 2.0 to 6.5% for Bonanza (Table I). The difference in crude lipid contents between the two cultivars were 14, 48, 93, 75, 100, and 76% for procedures 1–6, respectively. These differences were partly caused by the solvent system, the extraction conditions, and the degree of purification of the crude lipid achieved with each of the six extraction procedures. Solvent systems used in procedures 1–4 were more polar than the petroleum ether used in procedure 5, and hence their extracts contained crude lipids and nonlipid materials. Similarly, chloroform/methanol, 2:1, was more polar than the benzene/

TABLE I
Crude and Purified Lipid Contents (%) of Riso and Bonanza Barleys
Extracted with Different Procedures (dry basis)

Procedure Number	Solvent	Cultivar	Purified Lipids		
			Crude Lipids	Calcium Chloride	Ether
1	Chloroform/methanol 2:1, v/v	Riso	7.4	4.8	4.8
		Bonanza	6.5	4.3	2.4
2	Benzene/ethanol 4:1, v/v	Riso	6.4	5.3	4.3
		Bonanza	4.3	3.0	2.6
3	Chloroform/methanol/water 1:1:0.5, v/v	Riso	5.6	4.7	4.6
		Bonanza	2.9	2.6	2.5
4	Chloroform/methanol/water 1:2:0.8 and 2:2:1.8, v/v	Riso	4.2	4.9	4.7
		Bonanza	2.4	2.8	2.5
5	Petroleum ether	Riso	4.0	3.9	3.9
		Bonanza	2.0	2.1	1.8
6	Sulfuric acid/methanol 2:100, v/v	Riso	4.4
		Bonanza	2.5

TABLE II
Fatty Acid^a (%) Composition of Riso and Bonanza Barleys

Fatty Acids	Cultivar	
	Riso	Bonanza
In purified lipids, % of total fatty acid		
Palmitic	20.6	19.1
Stearic	1.0	1.2
Oleic	19.1	14.1
Linoleic	53.5	59.8
Linolenic	5.7	5.9
In purified lipids, g/100 g		
	63.4	60.3
In meal, mg/100 g		
	2,824	1,423

^aMean of six procedures.

absolute ethanol azeotrope. While procedures 1 and 2 yielded only crude lipids, procedure 3 partially purified the lipids by concentrating them in the chloroform layer and the nonlipids in the methanol/water layer. Procedure 3 had the further advantage of removing proteins from the extract by zinc acetate added to the monophasic layer. Direct extraction of the meal with petroleum ether (procedure 5) yielded a preponderance of free or neutral lipids, whereas estimation of total oil by gas-liquid chromatography (procedure 6) gave a measure of total long chain fatty acids.

Therefore, for a meaningful comparison of the lipid contents of the two barley varieties, purification of the crude lipids extracted by the six procedures was necessary. A two-step purification procedure was used. In step one, the nonlipid contaminants were removed with calcium chloride. The procedure reduced the crude lipid content in Riso by 35, 17, 16, 0, and 2.5% in methods 1–5, respectively. The corresponding values for Bonanza were 34, 30, 10, 0, and 0% for the same methods. Thus, as expected, crude lipids extracted by procedures 1 to 3 were substantially purified and lipids extracted by procedures 4 and 5 were purified very little or none. Further purification with diethylether reduced the lipid content of Bonanza by 44% in procedure 1 and of Riso by 19% in procedure 2. The purification in the other three procedures was small or insignificant. Data in Table I show that the varieties contained substantially different quantities of the purified lipids, the difference being 100% in procedure 1, 65% in 2, 84% in 3, 88% in 4, 116% in 5, and 76% (the same as for the crude lipids) in procedure 6. Thus, regardless of the solvent system used, the lipid content of Riso was considerably higher than that of Bonanza. The magnitude of the difference obtained with each of the six extraction procedures was greater than in data reported by us previously (Bhatty and Rosnagel 1979) between Riso and Canadian barleys (3.4 vs 2.1%) and by Munck (1976) between Riso and its parent line Bomi (4.1 vs 2.7%). In 1979, we planted Riso and Fairfield barleys at three different locations in Saskatchewan and at one location in Manitoba to further assess lipid content. In these, the lipid content of Riso determined by procedure 5 was higher than that of Fairfield by 69, 84, 85, and 88%. Thus our present data and those presented previously (Bhatty and Rosnagel 1979) conclusively show that the lipid content of Riso is stable and genetically controlled.

Table II shows the mean fatty acid composition of purified lipids obtained from the two varieties by the six procedures. (Only minor differences were found among the procedures.) The major fatty acids of both the barleys were linoleic, palmitic, and oleic. The minor fatty acids were linolenic and stearic, as has been well established previously (Fedak and de la Roche 1977, Parsons and

Price 1974, Price and Parsons 1975). Both the varieties contained trace amounts of myristic acid (C14:0); no attempt was made to quantitate this fatty acid. Variations in the fatty acid composition of European (Welch 1978) and Canadian (Fedak and de la Roche 1977) barleys have been reported. Because linoleic is one of the essential fatty acids, its concentration in barley is of nutritional significance.

Fatty acid composition of cereals, particularly those of the embryo and endosperm fractions, has been reported to be influenced by extraction procedures mainly because of the presence of different lipid classes in polar and nonpolar solvents (Jellum 1971, Weihrauch and Mathews 1977). This effect may, however, be minimal in our data (Table II) because the crude lipids extracted by the various procedures were subjected to similar purification. The mean fatty acid values show that Riso contained more palmitic and oleic acids but less linoleic acid than did Bonanza. The stearic and linolenic acid compositions of the varieties were identical. Welch (1978) reported that Riso 1508 grown in the United Kingdom contained 53.7% linoleic, 23.5% palmitic, 16.8% oleic, 5.2% linolenic, and 0.6% stearic. These values are generally similar to those of Riso obtained in the present study, the mean corresponding values being 53.5, 20.6, 19.1, 5.7, and 1.0%, respectively.

Amounts of fatty acids per 100 g of purified lipids were higher for procedures 1, 2, and 5 than for procedures 3 and 4 and may partly reflect the purity of the lipids, the lipid classes present, or the partial loss of lipids during purification. For instance, when these values were calculated from the fatty acid composition of crude lipids, they were lower for procedures 1 and 2 but higher for the other three procedures. Nevertheless, the mean values in Table II show that Riso contained more fatty acid per unit weight of lipids than did Bonanza (63.4 vs 60.3%). Price and Parsons (1975) reported a fatty acid percentage of 74 for barley, a value not obtained in any of the six procedures in the present study, and 91 for corn. This value may also be affected by the lipid class; the neutral lipids are the major (92%) lipids present in corn.

Fatty acid content expressed as mg/100 g of meal generally reflects the differences in the lipid contents of the two varieties given in Table II. Riso contained 98% more fatty acid per 100 g of meal than did Bonanza barley. Gross energy values of the two varieties determined by bomb calorimetry were 4,246 and 4,070 kcal/kg, respectively, for Riso and Bonanza, a difference of 4%. Welch (1978) reports a similar range in gross energy value of 100 genotypes of barley varying in oil from 1.9 to 4.1%.

Neutral or free lipids formed about 69% of the total lipids applied to the silicic acid column, and both the varieties contained almost equal amounts of this fraction (Table III). The NL were completely free of phosphorus, which was present mostly in the other two

TABLE III
Percent of Total Lipids in Lipid Classes of Riso and Bonanza Barleys

Lipid Class	Cultivar	
	Riso	Bonanza
Neutral	69.4 (74.9) ^a	68.9 (65.2)
Conjugated		
Glycolipids	6.8 (7.3)	27.0 (25.6)
Phospholipids	16.5 (17.8)	9.7 (9.2)
Recovery, %	92.7	105.6

^aResults adjusted to 100% recovery are shown in parentheses.

TABLE IV
Fatty Acid Composition^a of Lipid Classes^b of Riso and Bonanza Barleys

Fatty Acid	Riso			Bonanza		
	NL	GL	PL	NL	GL	PL
Palmitic	20.8	23.9	19.3	17.8	17.4	24.4
Stearic	1.1	2.5	1.5	1.1	5.5	1.3
Oleic	20.1	12.3	14.2	16.0	6.9	9.5
Linoleic	52.8	53.5	57.3	58.9	63.2	59.2
Linolenic	5.2	7.8	7.7	6.1	6.9	5.5

^aPercent of total fatty acids in each lipid class.

^bNL = neutral lipids, GL = glycolipids, PL = phospholipids.

TABLE V
Distribution of Lipids in Seed Fractions of Riso, Bonanza, and Fairfield Barleys

	Cultivars				
	Procedure 6			Procedure 5	
	Riso 1979	Bonanza 1978	Fairfield 1979	Riso 1979	Fairfield 1979
Percent of seed weight ^a					
Embryo	5.7	3.0	3.4	4.9	3.6
Endosperm	93.1	97.3	94.7	87.7	95.1
Lipid content, %					
Seed					
Embryo	4.1	2.4	2.5	3.7	2.0
Endosperm	17.4	18.0	15.2	20.9 ^b	20.5 ^b
Embryo	3.4	1.9	1.9	3.0	1.3
Relative distribution of lipids, % of total					
Embryo	23.8	22.6	22.3	28.0	37.3
Endosperm	76.2	77.4	77.7	72.0	62.7

^aTotals do not equal 100%.

^bSingle determination.

TABLE VI
Fatty Acid Composition^a of Seed, Embryo, and Endosperm of Riso, Bonanza, and Fairfield Barleys

Fatty Acid	Riso			Bonanza			Fairfield		
	Seed	Embryo	Endosperm	Seed	Embryo	Endosperm	Seed	Embryo	Endosperm
Palmitic	22.8	21.9	23.1	22.7	19.3	23.9	22.1	20.0	22.0
Stearic	1.0	0.4	1.1	1.1	0.4	1.4	1.8	0.6	2.2
Oleic	18.3	15.3	19.2	12.3	17.5	10.5	12.2	14.5	11.4
Linoleic	53.1	51.0	53.2	59.3	52.8	60.0	59.4	56.0	61.0
Linolenic	4.8	11.4	3.4	4.7	10.0	4.1	4.5	9.0	3.5

^aPercent of total fatty acids in each fraction.

fractions. The NL fraction was therefore not contaminated with conjugated or polar lipid fractions. However, the latter fractions varied considerably between the two varieties. Riso contained less GL but more PL than did Bonanza. The recovery of the eluted fractions was not the same in each variety in spite of repeated washings of the silica gel. Expression of the data at 100% recovery showed Riso lipids to be 74.9% NL and Bonanza's 65.2% NL. On this basis, 25.1% of Riso lipids (7.3% GL, 17.8% PL) and 34.8% of Bonanza lipids (25.6% GL, 9.2% PL) were conjugated lipids. Thus Bonanza barley contained considerably more GL than did Riso. Barley lipids have been reported to be 75% NL, 9% GL, and 20% PL (Price and Parsons 1974). Weihrauch and Mathews (1977) reported literature values for lipid classes in different cereals. However, these values may not be comparable because different extraction and separation procedures were employed and varieties within a cereal species may vary widely in lipid content and classes.

Palmitic, oleic, and linoleic were the major fatty acids of the three lipid classes of Riso and Bonanza (Table IV). In both the varieties, the NL fraction contained more oleic acid than did the GL or the PL fraction, the difference being greater in Bonanza than in Riso. The GL and PL fractions both contained more linoleic acid than did the NL fraction; the highest amount of palmitic acid was found in the GL fraction for Riso and the PL fraction for Bonanza. Data reported by Price and Parsons (1974) also showed the GL fraction to contain more linoleic and the PL fraction more palmitic acid than the NL fraction contained.

In barley, lipids are present mainly in the endosperm and embryo portions of the seed. Macleod and White (1961) reported that the embryo fraction formed less than 3% of the grain weight in Proctor barley (six-row malting) but contained 30% of the total grain lipid (ether extract). A higher lipid content in Riso 1508 than in Bomi has been ascribed to an enlarged embryo (Munck 1976). Tallberg (1977) reported that the embryo (and scutellum) made up 6.9% of the dry weight of mature seed in Riso 1508 compared to 3.6% in Bomi, an increase of 92%. Our data (Table V) show the average embryo weight in Riso to be 5.3% of the seed weight, in Bonanza, 3.0%, and in Fairfield, 3.5%. Thus the ratio between the embryo and endosperm weights in Riso and the other two varieties was generally similar to the one reported earlier (Tallberg 1977). Unfortunately, the embryo and endosperms weights rarely totaled 100%, partly because of handling loss and loss of volatile matter on drying the samples.

The embryo isolated from Riso contained 17.4% lipids determined by procedure 6, somewhat lower than in Bonanza (18.0%) but higher than in Fairfield barley (15.2%). Riso endosperm contained almost double the lipids present in the endosperm of Bonanza or Fairfield barleys determined by the same procedure. The fraction weight and lipid content data were used to calculate the relative distribution of lipids in the embryo and endosperm fractions of the three varieties. With procedure 6 the embryo contributed 23.8% of the total lipids in Riso, 22.6% in Bonanza, and 22.3% in Fairfield. Because of the fewer number of observations, a test of significance was difficult to apply to the data. Nevertheless, Riso embryo contained only 15% more oil than did Bonanza embryo and 11% more oil than did Fairfield embryo. The endosperm contributed 76.2% of the total lipids in Riso, compared to 77.4 and 77.7% in Bonanza and Fairfield, respectively. Thus, using procedure 6, which measures long chain fatty acids, Riso embryo contained slightly more lipids than did the embryos of

Bonanza or Fairfield barleys. Little or no difference was found in the lipid contents of Riso and Bonanza or Riso and Fairfield endosperm fractions. Both Macleod and White (1961) and Tallberg (1977) employed ether extraction to determine lipid distribution in barley. This solvent may be suitable for neutral lipids but may extract only a portion of the polar lipids. If the predominant type of lipid in barley embryo is the NL or triglycerides, as is the case with corn (Jellum 1971), then such a solvent system may give lower values for endosperm lipids. The gas-liquid chromatography method used in the present study does not discriminate between the lipid classes. However, to study solvent effect on lipid distribution, embryo and endosperm fractions obtained from Riso and Fairfield were extracted with petroleum ether (procedure 5). Because of the small quantities of the embryo material, only single extractions were made. Table V shows that with ether as solvent, the embryo fraction contained almost the same lipid content in both the varieties, higher than that of the embryo fractions of the same varieties and of Bonanza determined by procedure 6. Because of the higher lipid content of the embryo fractions by procedure 5, the contribution to total lipids by embryo lipids was 28.0% in Riso and 37.3% in Fairfield, a difference of 33%. These values are closer to embryo lipid data (30%) reported by Macleod and White (1961). Relative lipid distribution of the endosperm fractions determined by procedure 5 was 72.0% in Riso and 62.7% in Fairfield, a difference of about 15%. The data lead us to conclude that a larger embryo in Riso may not be totally responsible for a higher lipid content in this barley compared to those in Bonanza and Fairfield barleys.

The comparative fatty acid compositions of the whole seed, the embryo, and of endosperm are shown in Table VI. The major difference was in the fatty acid composition of the embryo; in all three varieties it contained two to three times as much linolenic acid as the other two fractions contained. In all three varieties, the endosperm had higher percentages of palmitic acid and linoleic acid than did the embryo. The percentages of oleic acid were higher in the embryo than in the endosperm in Bonanza and Fairfield but not in Riso.

Riso contained two-and-one-half times more (2,360 vs 1,000 units) LA than did Bonanza. Lipoygenase forms hydroperoxides from unsaturated fatty acids; these affect grain storage and have other undesirable effects. Lulai and Baker (1976) found 92% of the LA of barley localized in the germ fraction of grain. A higher LA of Riso compared to Bonanza could possibly result from its larger embryo size. However, the significance of the difference in the LA of the two varieties cannot be explained.

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