RICE LIPIDS[1]

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ABSTRACT

Types and amounts of lipids are high in the bran and low in the endosperm, the two rice kernel parts of dietary significance. The main lipid class is triglyceride in the bran, free fatty acid in the endosperm, and almost exclusively a monoacyl lipid in the rice starch. Bran lipids are classified in groups of glycerolipids, sterol lipids, and sphingolipids, according to their structural and metabolic relationships. In the glycerolipids group, the diglyceridic species in triglyceride and glycerophospholipid are generally similar but differ from the glyceroglycolipids. In the sterol lipid group, component species are similar in free sterols and sterol esters and in sterylglycosides and acylsterylglycosides but differ in the neutral and polar sterol lipids. A new compound, steryloligohexoside (oligohexosylsterol) is reported. In the sphingolipid group, ceramide and sphingoglycolipid component species differ, although the sphingoglycolipid classes of monohexosylceramides, dihexosylceramides and trihexosylceramides show similarities.

Rice plants are threshed to produce paddy rice (rough rice), which is dehulled to brown rice. The brown rice is then separated by a polishing machine into bran (the outer part of grain that consists of pericarp, tegmen, aleurone layer and embryo) and polished rice (milled rice or white rice, which is the inner part of the grain that corresponds substantially to endosperm). Bran is used for feeds, oil products, pharmaceuticals, and chemicals; polished rice is a staple food and is the base of fermented, confectionary and other products.

This review deals with general aspects of lipids in commercial bran and the starchy endosperm and with food-chemical or biochemical profiles of lipids, especially in bran because it is a major source of rice lipids. Most of the data reported represent work done in our laboratories. Comprehensive reviews on rice lipids by Juliano (1,2), Weber (3), Nechaev and Sandler (4) and Morrison (5) also describe varieties, properties and compositions of lipids in rice grain.

RICE LIPIDS

Contents

The chemical composition of rice grain is shown in Table I. The 2.3% lipid content in brown rice is slightly lower than the 2.4–3.9% reported by Juliano (2). The value seems to differ with variety, degree of kernel maturation and growth conditions of the rice, and the procedure used to extract the lipids. The lipid contents of 18.3% in bran and of 0.8% in polished rice are a little lower and higher, respectively, than those of 17.5–21.7% and 0.3–0.6% reported previously (2). Lipid values in bran and polished rice are affected significantly by variations in the degree of polishing; clean, perfect mechanical separation of bran and polished rice from brown rice is difficult. The pericarp, tegmen, aleurone layer, embryo (germ), and parts of the endosperm are removed gradually by polishing,

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but part of the germ often adheres to white rice even after 20% polishing. Admixture of the aleurone layer and the starchy endosperm with bran and adhesion of the embryo to the endosperm considerably influence the lipid contents of bran and white rice, respectively.

Isolation and Fractionation

Polished rice and especially bran were steamed immediately after preparation to inactivate the lipid-degrading enzymes such as lipase, phospholipase, glycolipase, lipoxygenase, and oxidase. Total lipid in the steamed material was extracted with organic solvents, immediately or after freeze-storage. The total lipid was then fractionated into neutral lipid, glycolipid, and phospholipid by column chromatography on a silicic acid column (7,8). The procedure for isolation and fractionation of rice lipids is outlined in Fig. 1; yields of the fractions are not always reproducible. Glycolipids and phospholipids are sometimes called *en bloc* polar lipids in opposition to neutral lipids.

Distribution in Grain

Neither the types nor the quantities of lipids are distributed uniformly in bran and the endosperm. Approximate ratios of neutral lipid and polar lipid, for

TABLE I
Composition of Rice[a]

Material	Moisture (%)	Protein (%)	Lipid (%)	Sugar Substance (%)	Fiber (%)	Ash (%)	Degree of Polishing (%)
Brown rice	15.5	7.4	2.3	72.5	1.0	1.3	0
Rice bran	13.5	13.2	18.3	38.3	7.8	8.9	...
Polished rice	15.5	6.2	0.8	76.6	0.3	0.6	8–10

[a]Source: Bureau of Science and Technology (6).

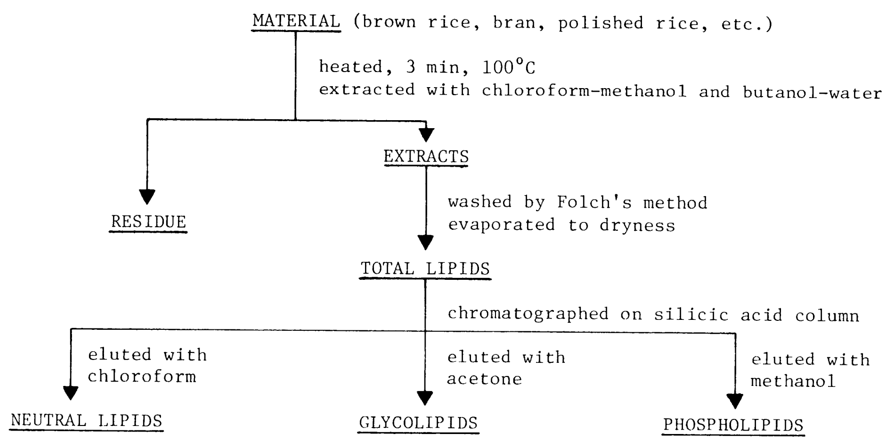


Fig. 1. Isolation and fractionation of lipids from rice.

instance, were 90.6:9.4 in bran (9), 49:51 in endosperm (10) and 37:63 in endospermic starch (11). Thus, bran is rich predominantly in neutral lipid (actually storage lipid), but the endosperm, especially the starch, contains considerable amounts of neutral lipid and also polar lipid (functional lipid). According to thin-layer chromatograms on silicic acid, more types of lipid classes are in starchy endosperm and particularly in bran than in starch. The principal lipid class is triglyceride in bran and free fatty acid in the endosperm and starch (9–11).

Purified rice starch contains almost exclusively a monoacyl type of lipids (single carbon-chain lipids) such as free fatty acid, lysophosphatidylcholine, lysophosphatidylethanolamine, monoglycosylmonoglyceride, and diglycosylmonoglyceride (11). The monoacyl lipids that correspond to the so-called fat-by-hydrolysis (12) are believed to be included in the amylose structure of starch (5,13,14) and thus constitute internal lipids. Partially purified rice starch contains single carbon-chain lipids and some non-monoacyl lipids (12,15,16). These probably interact with protein outside the amylose structure and might be called external or surface lipids. The internal and the external lipids are assumed to be related because of the physicochemical properties of commercial starch or starchy foods and the metabolism of starch in living cells.

Classification of Bran Lipids

Rice bran lipids can be classified by structure, in three main groups. The glycerolipid group includes glyceride, glycerophospholipid, and glyceroglycolipid; all have glycerol and a fatty acid as common components. The sterol lipid group includes free sterol, sterol ester, sterylglycoside, and acylsterylglycoside; all contain sterol. The sphingolipid group includes ceramide and hexosylceramide (ceramidehexoside); sphingosine and fatty acid are the common components. Structural analogies and metabolic relationships are known to exist among the lipid classes in each of the groups.

MOLECULAR SPECIES AND STRUCTURE OF GLYCEROLIPIDS

The main glycerolipids in rice bran are triglyceride, glycerophospholipid, and glycoglycerolipid. These lipids commonly contain a diglyceridic radical on which fatty acids and molecular species were determined in establishing relationships among the glycerolipid classes.

Tryglycerides

Triglyceride isolated from the neutral lipid fraction by silicic acid column chromatography (17) was examined for total and positional composition of fatty acid constituents and for species of triglyceride molecules (18,19). The principal fatty acids are linoleic, oleic, and palmitic acids. Positions C-1, C-2, and C-3 on the glycerol moiety are rich in unsaturated fatty acids; C-2 contains no saturated acids. Among the 12 major molecular species, trienoic and tetraenoic species dominate (9). If the isomeric species with the same fatty acids at C-1 and C-3 are postulated to exist in a 1:1 ratio, 1,2-diglyceridic species of triglyceride would be arranged as shown in the extreme left column of Table II. In practice, trienoic and particularly dienoic species dominate and would be preferential precursors in biosynthesis of triglycerides in rice bran.

TABLE II
Composition of Molecular Species of Glycerolipids in Rice Bran[a]

Double Bond No.	Diglyceride Species C-1	Diglyceride Species C-2	Triglyceride (%)	Phospholipid: Phosphatidyl-choline (%)	Phospholipid: Phosphatidyl-ethanolamine (%)	Phospholipid: Phosphatidyl-inositol (%)	Glycolipid: Monoglycosyl-diglyceride (%)	Glycolipid: Diglycosyl-diglyceride (%)	Glycolipid: Sulfoquinovosyl-diglyceride (%)
0	16:0	16:0	...	...	...	...	...	...	37.9
1	16:0	18:1	16.9	18.5	16.1	29.4	4.9	7.8	6.8
	18:1	16:0	...	...	...	...	...	...	11.9
2	16:0	18:2	20.7	20.3	38.3	37.9	13.9	8.3	9.2
	18:1	18:1	23.4	19.6	6.5	8.1	6.3	6.1	1.7
	18:2	16:0	...	...	...	...	...	...	18.9
3	16:0	18:3	2.0	...	...	...	11.9	9.6	2.3
	18:1	18:2	12.5	18.8	14.2	14.4	11.6	8.3	2.6
	18:2	18:1	12.5	10.1	7.6	4.5	3.0	4.6	1.5
	18:3	16:0	...	...	...	...	...	...	4.1
4	18:1	18:3	...	...	...	...	6.6	8.7	...
	18:2	18:2	13.0	12.7	17.3	5.7	14.8	16.3	1.1
	18:3	18:1	...	...	...	...	8.7	6.2	...
5	18:2	18:3	...	...	...	...	8.3	9.2	2.0
	18:3	18:2	...	...	...	...	6.9	5.4	...
6	18:3	18:3	...	...	...	...	3.1	9.5	...

[a]Source: Fujino and colleagues (20,21).

Glycerophospholipids

Rice grain contains more than ten classes of glycerophospholipids, of which phosphatidylcholine, phosphatidylethanolamine, and phosphatidylinositol are major (12,22,23). A thin-layer silicic acid chromatogram of phospholipids in rice bran is depicted in Fig. 2. The three major lipids were isolated separately from the phospholipid fraction by silicic acid column chromatography (24) for determination of the whole and positional compositions of the fatty acids and of the diglyceridic molecular species (8). Linoleic, oleic, and palmitic are the chief fatty acids. Position C-1 of the three major glycerophospholipids contains saturated and unsaturated acids; C-2 is rich in unsaturated fatty acids. Rice has six major molecular species of bran phospholipids (8), as shown in Table II. The main species of phosphatidylcholine and phosphatidylethanolamine are dienoic and trienoic; diglyceridic patterns of the two lipids and of the triglyceride are similar. The chief species of phosphatidylinositol are dienoic and monoenoic; the diglyceridic pattern is less unsaturated than those of triglyceride, phosphatidylcholine, and phosphatidylethanolamine. Differences in species in phosphatidylinositol and the three glycerolipids may relate to the fact that acidic phospholipids such as phosphatidylinositol, phosphatidylserine, and phosphatidylglycerol include CDP-1,2-diglyceride as a direct precursor, but

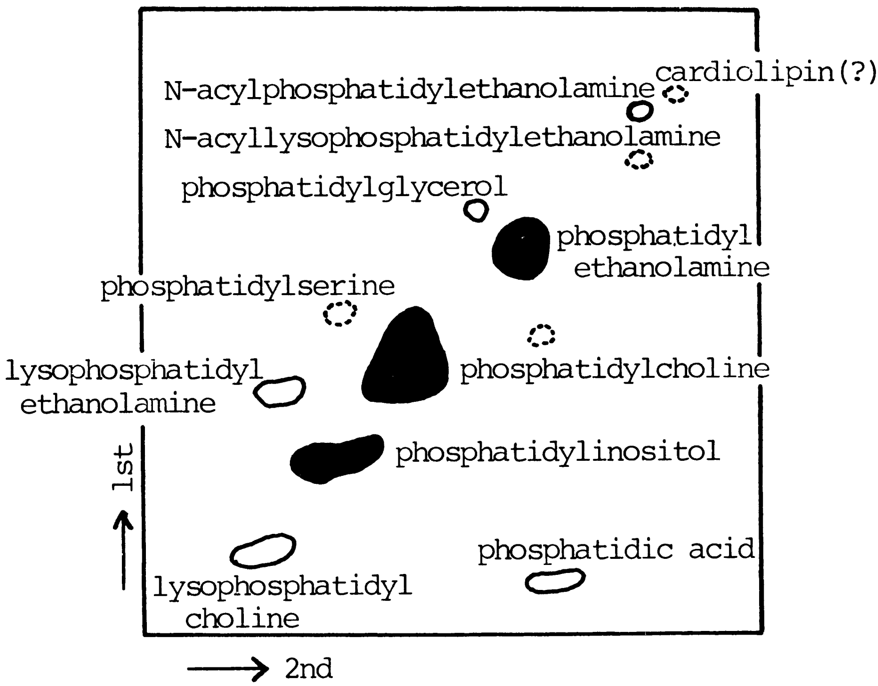


Fig. 2. Thin-layer chromatogram of rice bran phospholipids (8). Plate: Wako silica gel B5. Development: 1st, chloroform-methanol-ammonia (65:35:5, by volume); 2nd, chloroform-acetone-methanol-acetic acid-water (10:4:2:2:1, by volume). Detection: molybdenum blue reagent.

nonacidic glycerolipids such as triglyceride, phosphatidylcholine, and phosphatidylethanolamine are biosynthesized directly from 1,2-diglyceride.

Glycoglycerolipids

Rice grain contains several glycoglycerolipids, of which diglycosyldiglyceride is the main component (12,25–27). A thin-layer silicic acid chromatogram of the glycolipid fraction from rice bran is shown in Fig. 3. The three well-known glycoglycerolipids (monoglycosyldiglyceride, diglycosyldiglyceride and sulfoquinovosyldiglyceride) were separated from the glycolipid fraction by silicic acid column chromatography (29). The former two are nonsulfonic lipids and the latter is a sulfonic one, a type of monoglycosyl lipid usually found in photosynthetic tissues. The glycoglycerolipids were analyzed to determine fatty acid composition and diglyceridic species (28). The main component fatty acids usually are palmitic, oleic, linoleic, and linolenic acids. In monoglycosyldiglyceride and diglycosyldiglyceride, C-1 positions contain the saturated acids in addition to unsaturated ones; C-2 positions are rich in unsaturated fatty acids. In

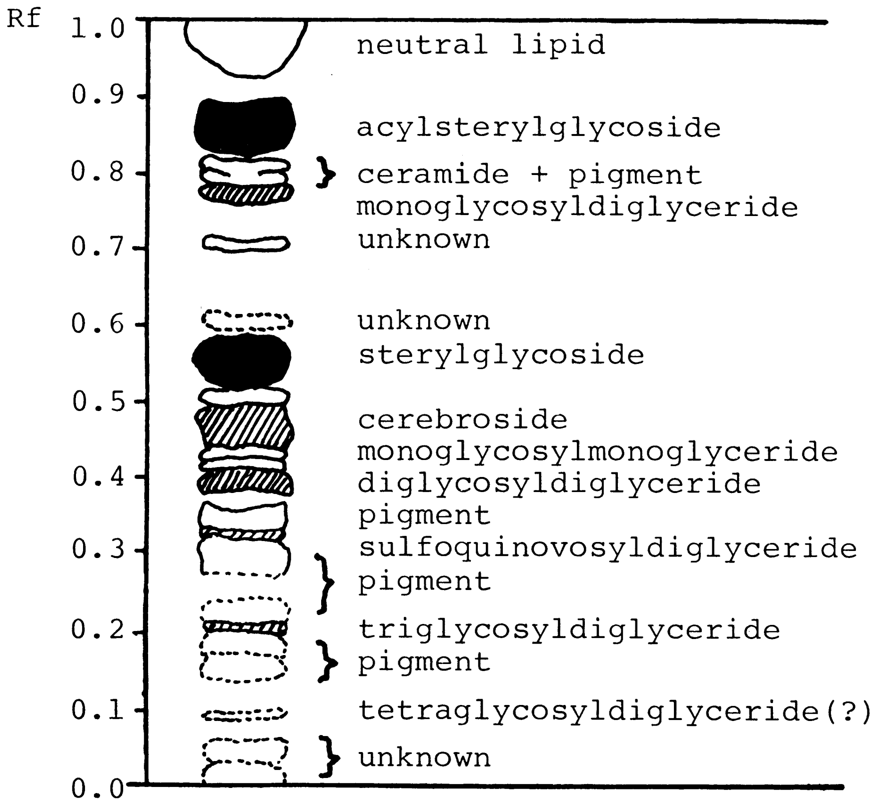


Fig. 3. Thin-layer chromatogram of rice bran glycolipids (28). Plate: Wako silica gel B5. Development: chloroform-methanol-acetic acid-water (35:10:1:1, by vol.). Detection: anthrone reagent.

sulfoquinovosyldiglyceride, however, C-1 and C-2 positions are both rich in saturated acids.

The 16 major molecular species of bran glycoglycerolipids (28) are shown in Table II. The main species of monoglycosyldiglyceride and diglycosyldiglyceride are tetraenoic and trienoic; diglyceridic patterns of the two lipids are much more unsaturated than those of triglyceride and glycerophospholipids. The chief species of sulfoquinovosyldiglyceride are nonenoic (saturated), monoenoic, and dienoic; the diglyceridic pattern is much less unsaturated than those of triglyceride, glycerophospholipid, and nonsulfonic glycoglycerolipid. This suggests that, in biosynthesis of glycoglycerolipid in rice bran, nonsulfonic lipids require highly unsaturated diglycerides as precursors and sulfonic lipids require lowly unsaturated diglycerides.

Nonsulfonic Glycoglycerolipids

Monoglycosylmonoglyceride, monoglycosyldiglyceride, diglycosylmonoglyceride, diglycosyldiglyceride, triglycosyldiglyceride and probably tetraglycosyldiglyceride (Fig. 3) are nonsulfonic glycoglycerolipids in rice grain (12,28). Component sugars of these lipids were galactose and glucose in every case. Generally, the ratio of galactose and glucose decreased as the number of hexoses increased in the lipid molecule. The data suggest that varieties of glycoglycerolipids shown in Fig. 4 can exist in rice bran. Isolation of each lipid, anomeric structures of the hexose units, and combinative positions of adjacent hexoses have not yet been established for the oligohexosyldiglyceride.

Recently, bran monohexosyldiglycerides were separated into galactosyl and glucosyl lipids and characterized (30); the structures are depicted in Fig. 5. Monogalactosyldiglyceride is considerably unsaturated, with linoleic acid as the major component, and monoglucosyldiglyceride is relatively saturated, with palmitic acid as the principal component. It is interesting that fatty acid composition of the monoglucosyldiglyceride resembles that of sulfoquinovosyldiglyceride, since sulfoquinovose is a glucose analogue, 6-sulfo-6-deoxy-glucose.

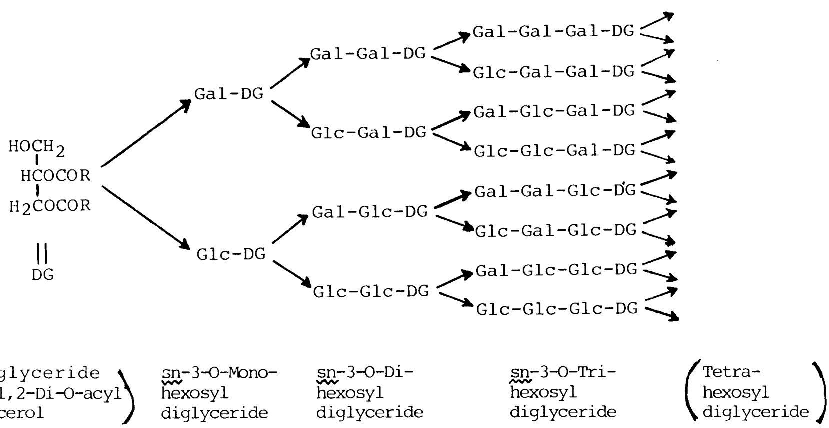


Fig. 4. Possible varieties of glycoglycerolipids in rice bran (Gal = galactose; Glc = glucose).

Interrelationships

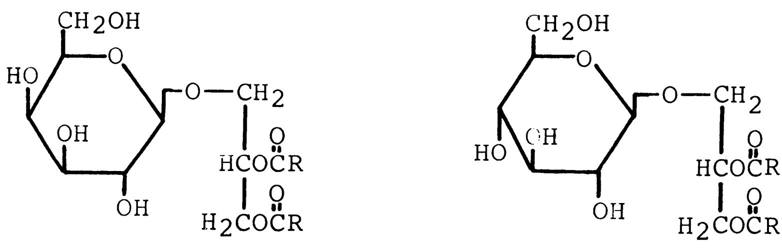
Generally, diglyceridic species (Table II) of rice bran glycerolipids, triglyceride, phosphatidylcholine, phosphatidylethanolamine, and phosphatidylinositol are moderately unsaturated; monoglycosyldiglyceride (to be precise, monogalactosyldiglyceride) and diglycosyldiglyceride are highly unsaturated; and sulfoquinovosyldiglyceride (and monoglucosyldiglyceride) are lowly unsaturated. The findings imply that, among the diglyceride precursors, relatively saturated diglycerides may convert to sulfoquinovosyldiglyceride and monoglucosyldiglyceride, moderately unsaturated ones may convert to triglyceride and glycerophospholipid, and highly unsaturated diglycerides may convert to monogalactosyldiglyceride and diglycosyldiglyceride.

CONSTITUTION OF STEROL LIPIDS

The main classes of sterol lipids in rice bran are free 4-demethylsterol, free 4-monomethylsterol, free 4,4-dimethylsterol (triterpene alcohol), sterol ester, sterylglycoside, and acylsterylglycoside (31). Silicic acid column chromatography separately isolates the former four from the neutral lipid fraction (17), and the latter two from the glycolipid fraction (29). These lipids were analyzed for sterols, fatty acids, and sugars, to deduce relationships among the sterol lipid classes.

Fatty Acids

The major component fatty acids of sterol ester were linoleic, oleic, and several other acids; those of acylsterylglycoside were linoleic, palmitic, and several others; the patterns differ somewhat in the two classes. In acyl composition, the relationships of sterol ester and acylsterylglycoside are only limited. Small amounts of C_{15} and C_{17} acids detected in sterol ester of brown rice are not conclusively characteristic constituents of sterol ester in the rice grain (32).



3-O-β-D-Galactopyranosyl-sn-1,2-diacylglycerol

Monogalactosyldiglyceride

3-O-β-D-Glucopyranosyl-sn-1,2-diacylglycerol

Monoglucosyldiglyceride

Fig. 5. Structure of monohexosyldiglycerides in rice bran (R = long-chain alkyl group).

Sugars

The constituent saccharide of both sterylglycoside and acylsterylglycoside is glucose (31), implying that the two glycolipids are metabolically related. The component sugar of free and esterified sterylglycoside in cereals generally is reported as glucose only (33–36), but a small quantity of mannose can be detected in sterylglycoside of brown rice (37).

Sterols

Component 4-demethylsterols of both the sterol ester and free demethylsterol fractions are cholesterol, campesterol, stigmasterol, β-sitosterol, Δ^5-avenasterol, Δ^7-stigmastenol, and Δ^7-avenasterol (31,38). The most abundant constituent was β-sitosterol. The patterns in the two classes are similar, but the patterns of 4-demethylsterols of the neutral sterol lipid, sterol ester, and free demethylsterol fraction were not like those of the polar sterol lipids, sterylglycoside, and acylsterylglycoside. Constituent 4-monomethylsterols of both sterol ester and free monomethylsterol fractions include obtusifoliol, gramisterol, and citrostadienol. Gramisterol is a major component and the patterns are remarkably similar in the two classes. Component 4,4-dimethylsterols of both sterol ester and free dimethylsterol fractions include cycloartanol, cycloartenol, 24-methylenecycloartanol, and cyclobranol; 24-methylenecycloartanol is the principal component and the patterns are similar in the two classes. Neither 4-monomethylsterol nor 4,4-dimethylsterol are components of sterylglycoside and acylsterylglycoside.

These findings suggest close relationships between classes of neutral sterol lipids and between those of polar sterol lipids and less intimate relationships between neutral sterol and polar sterol lipids.

Interrelationships

A review of the types of demethylsterols and methylsterols suggests that sterols in rice bran may be formed by squalene through 4,4-dimethylsterols and 4-monomethylsterols to 4-demethylsterols, as in general plants (20). Consideration of those findings and of the patterns of the component fatty acids, sugars, and sterols suggests the possibility of metabolic and constitutional relationships among sterol lipid classes in rice bran as depicted in Fig. 6.

Novel Sterylglycosides

Careful thin-layer silicic acid chromatography of the alkali-stable glycolipid fraction revealed the usual sterylglycosides (monohexosylsterols) and also oligohexosylsterols; the latter were suggested but were not characterized in plant

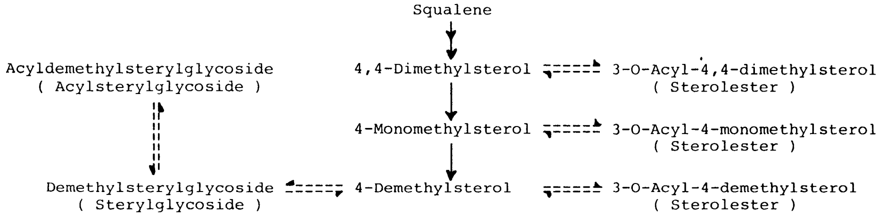


Fig. 6. Interrelationships of sterol lipid classes in rice bran (20,21).

tissue cultures (39). These novel sterylglycosides consist of dihexosylsterol, trihexosylsterol, tetrahexosylsterol, and probably pentahexosylsterol (40). The common components are β-sitosterol, stigmasterol, and campesterol as sterols and only glucose as hexose; interlinkages of sugars are mainly β1→4 in the diglucosylsterol. The general formula for oliglucosylsterols and the structure for diglucosylsterols are shown in Fig. 7. No oligohexosylsterols have been detected in nature, except for 3-0-gentiobiosyl-α-spinasterol in green tea (41).

CHARACTERISTICS OF SPHINGOLIPIDS

Main classes of sphingolipids in rice grain and rice bran are ceramide, monohexosyl, dihexosyl, trihexosyl, and probably tetrahexosyl ceramide (25,42–45). Ceramide belongs to the neutral sphingolipids and the glycosylceramides to polar sphingolipid. Only sugar-containing sphingolipids are found in rice bran as polar; neither sphingophospholipid (such as sphingomyelin) nor phosphoglycosphingolipid (such as phytoglycolipid) have been reported. Bran sphingolipids isolated from the alkali-stable glycolipid fraction by silicic acid column chromatography (29) were analyzed for the component sphingosines,[3] fatty acids, and sugars to study relationships among sphingolipid classes.

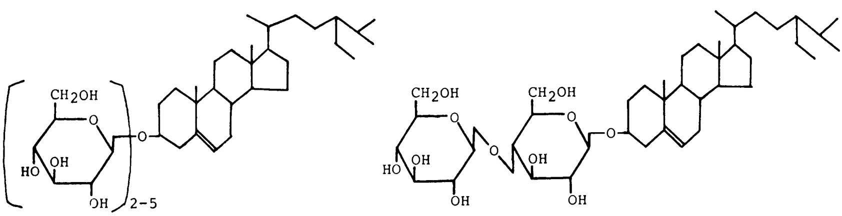
Fatty Acids

The major acid group of both ceramide and glycosphingolipid classes is 2-hydroxy one, of which 2-hydroxytetracosanoic, 2-hydroxypentacosanoic, 2-hydroxyhexacosanoic, etc., are the principal acids in ceramide; 2-hydroxyeicosanoic, 2-hydroxydocosanoic, 2-hydroxytetracosanoic, etc., are the main acids in glycosphingolipid classes (44,45). Thus, the component acid patterns of ceramide and glycosphingolipid classes differ, although they are somewhat similar in the glycosphingolipid classes.

Sugars

Component saccharides of the glycosphingolipid classes are glucose, mannose, and galactose; glucose predominates (44,45). The patterns are similar.

[3]Sphingosine is used as a comprehensive term for sphinganine analogues or long-chain bases.



3-0-Oligoglucosyl-β-sitosterol

D-Glucopyranosyl-(β1→4)-D-glucopyranosyl-(β1→3)-β-sitosterol

Oligoglucosylsterol

Diglucosylsterol (Cellobiosylsterol)

Fig. 7. Structure of novel sterylglycosides in rice bran.

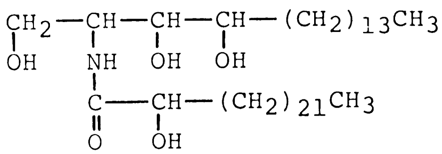
Thus, glycosphingolipids in rice bran are practically monoglucosylceramide and oligoglucosylceramides in which anomeric structures of glucose units and the combinative positions of the adjacent glucose units remain to be elucidated.

Sphingosines

The predominant constituent long-chain base is phytosphingosine (4-hydroxyoctadecasphinganine) in ceramide; it is octadecasphinga-4,8-dienine, 4-hydroxyoctadecasphing-8-enine, etc. (44,45), in the glycosphingolipid classes. Thus, the component base patterns in the ceramide and glycosphingolipid classes differ, although they are considerably similar in the glycosphingolipid classes. Octadecasphing-4-enine, which was reported to be the main component base of monohexosylceramide (42), could possibly be octadecasphinga-4,8-dienine. Both sphingosines belong to the sphingenine series and often behave similarly in chromatography.

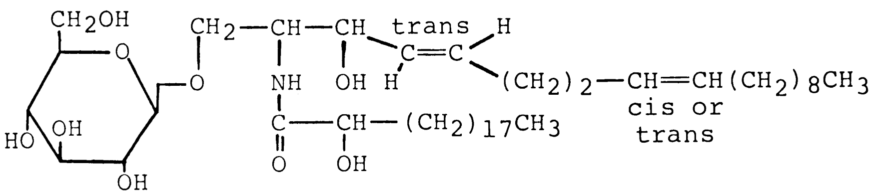
Interrelationships

Principal molecular species of representative sphingolipids in rice bran, ceramide, and monohexosylceramide (ceramidemonohexoside), as deduced from the analytical data of the components, are shown in Fig. 8. Ceramide species of the two sphingolipid classes differ. Generally, molecular species of rice bran ceramide differ from the ceramide moiety of glycosphingolipid classes, among which component patterns are considerably alike. This implies that the biosynthetic pathway of ceramide may differ from those of glycosphingolipid classes or that particular species of ceramide may become precursors in the formation of glycosphingolipids.



N-2-HYDROXYTETRACOSANOYL-4-HYDROXYSPHINGANINE

CERAMIDE



N-2-HYDROXYEICOSANOYL-1-O-β-D-GLUCOPYRANOSYL-OCTADECASPHINGA-4-TRANS-8-CIS/TRANS-DIENINE

MONOHEXOSYLCERAMIDE (CERAMIDEMONOHEXOSIDE, PHYTOCEREBROSIDE)

Fig. 8. Main molecular species of sphingolipids in rice bran.

CONCLUSION

Many chemical studies have been performed on rice lipids. Some discrepancies probably can be attributed to varietal differences, degree of maturity, growth conditions, and analytical techniques. Consequently, authoritative results require analyses of numerous samples by several methods.

Studies on rice lipids have been done largely on the rice grain or on rice bran; little is known about the other parts of rice plant such as hull, leaf, stem, and root. This is justified because the rice grain and the bran are used widely as foods, feeds, oil materials, and sources of many products. Similarly, other parts of the rice plant are potentially useful for production of feeds, soil fertilizers, and many industrial materials. It is important, therefore, to study the lipids of the rice grain and also those of the other parts of the rice plant. Such studies could provide fundamental structural and biosynthetic information and the basis for much needed renewable organic resources.

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