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Cellular and thylakoid-membrane glycolipids of *Chlamydomonas reinhardtii* 137^{+}

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Abstract The glycolipids of the green alga *Chlamydomonas reinhardtii* 137^{+} have been quantitated in the phototrophically-cultured cell and in its thylakoid membrane. Three lipids, the galactolipids monogalactosyldiacylglycerol (MGDG) and digalactosyldiacylglycerol (DGDG), and the sulfated glycolipid sulfoquinovosyldiacylglycerol (SL), constitute the total glycolipid complement and some 70–80% of the total polar lipid at both levels. About 70% of the alga's sulfolipid, but only about half of its galactolipid, is localized in the thylakoid. The three cellular and thylakoid glycolipids all contain prominent hexadecanoic and octadecanoic fatty acids. Quantitatively, each lipid has a distinctive acyl profile, making SL the most highly saturated and MGDG the least saturated glycolipid. Differences between the fatty acid profiles of each corresponding cellular and thylakoid glycolipid indicate that discrimination of the glycolipid species assembled into photosynthetic membrane takes place during thylakoid membrane biogenesis.—**Janero, D. R., and R. Barrnett.** Cellular and thylakoid-membrane glycolipids of *Chlamydomonas reinhardtii* 137^{+}. *J. Lipid Res.* 1981. **22:** 1119–1125.

Supplementary key words fatty acids · monogalactosyldiacylglycerol · digalactosyldiacylglycerol · sulfoquinovosyldiacylglycerol

Glycolipid and phospholipid appear to be the two exclusive types of polar glycerolipid in most higher green plants (1). Three characteristic glycolipids, the two uncharged galactolipids MGDG and DGDG and the acidic, sulfated lipid SL, often comprise over 80% of the total polar lipid complement along with such phospholipids as PG, PE, and PC. From more limited subcellular evidence, the three glycolipids are also the major polar lipids of the chloroplast photosynthetic membrane (thylakoid) to the virtual exclusion of other glycerolipids except PG.

Certain fatty acids are characteristically and even exclusively esterified to particular green-plant polar lipids (2). Stereospecific acyl-group analyses have highlighted the disposition of some fatty acids to appear at one or the other esterified glycerol carbons (3). Only in spinach tissue has compelling evidence been obtained (4) that the distribution of phospholipids and glycolipids among intracellular and even intraorganellar green-plant membranes can vary considerably in amount and in the types and intramolecular arrangements of esterified fatty acids.

Mainly because of technical factors, most details of plant and, especially, of thylakoid lipid biochemistry have come from higher green plants. The few analytical studies on the lipids of lower green plants, particularly green algal phytoflagellates, have been concerned mainly with gross cellular lipid or fatty acid profiles of either green tissue or etiolated mutants (5). From these studies, one can conclude merely that lower green plants frequently contain the major glycolipids, phospholipids, and fatty acids of higher green plant tissue. Information on the thylakoid membrane lipids of the green phytoflagellates is the least authoritative, relying on assumptions, for example, that cellular fatty acid profiles can be held reflective of the large amounts of photosynthetic membrane in the cell, or that differences in fatty acid profiles between green cells and their etiolated counterparts are indicative of thylakoid lipid biochemistry. The most direct approach, isolation of reasonably pure and homogeneous thylakoid membrane followed by determination of its lipid composition, has rarely been carried out on lower green plants and phytoflagellates.

As part of our studies designed to augment the limited quantitative data on the lipids of the lower green plants, we have analyzed the polar glycerolipids of the single-celled green alga *Chlamydomonas reinhardtii* 137^{+} (wild-type). Advantage was taken of the ability to culture the plant material axenically in defined minimal medium and to obtain from it thylakoid fractions of analytical quality. This report details the

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Abbreviations: MGDG, monogalactosyldiacylglycerol; DGDG, digalactosyldiacylglycerol; SL, sulfoquinovosyldiacylglycerol; PC, phosphatidylcholine; DGTS, diacylglyceryl-trimethylhomoserine; TLC, thin-layer chromatography; GLC, gas–liquid chromatography; PE, phosphatidylethanolamine; PG, phosphatidylglycerol.

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glycolipid biochemistry of phototrophic *C. reinhardtii* 137^+ and of its thylakoid and the fatty acids esterified to the respective glycolipids at both levels. A companion report (6) contains a molecular analysis of the cell's phospholipid biochemistry.

EXPERIMENTAL PROCEDURES

Cell culture and fractionation

Wild-type *Chlamydomonas reinhardtii*, strain 137^+, was grown phototrophically and asynchronously in axenic, log-phase culture as described (7). The minimal medium of Sager and Granick (8) was used. Cells were harvested by low-speed centrifugation.

An analytical fraction of thylakoid membranes was purified from *Chlamydomonas* homogenates according to Chua and Bennoun (9). In brief,[2] the thylakoid fraction represents over 75% of the 95% of chlorophyll recovered in the procedure and is purified ~2.7-fold over the starting homogenate. Chlorophyll *a*-to-chlorophyll *b* ratios for the cells as a whole and for the isolated membrane are identical, 2.1 ± 0.1 (S.D.; n = 5). Analyses of the thylakoid fraction by isopycnic sucrose density gradient centrifugation and electron microscopy support results from marker enzyme[3] assays that the fraction is virtually homogeneous, the only detectable contamination some 3–4% of the total cellular mitochondrial population.

Lipid purification, chromatography, and quantitation

Algal lipids were extracted and purified by a modified Bligh-Dyer method (10). Lipid recovery was quantitative as determined by the identical amounts of saponifiable materials produced from direct alkaline treatment (11) of cells labeled to constant specific lipid radioactivity with [^{3}H]acetate and from the lipid extract of a parallel cell sample. No free fatty acid could be detected chemically (12) in the lipid extracts. Lipids in the final chloroform phase were separated by two-dimensional thin-layer chromatography (TLC) on 0.25 mm layers of silica gel Type-60 F-254 with fluorescent indicator (Merck, Darmstadt, Germany) in the solvents described (13). The resolved lipids were visualized nondestructively under ultraviolet light, and, when necessary, complete elution of lipid from adsorbent was achieved by extracting the gel (10). All glycerolipids were quantitated by a hydroxamate method (14); in addition, glycolipids were estimated by sugar analysis (15), and phospholipids by phosphate determination (16) with results identical to those of the hydroxamate ester analysis. Mild base saponification (11) was used to deacylate the lipids.

Identification of the glycolipids

The glycolipids were identified by a number of chemical and chromatographic criteria. Only three cellular and thylakoid-membrane polar lipids give a positive reaction to TLC detection reagents for sugar (17), contain chemically-assayable sugar (15), and do not incorporate label from $^{32}PO_4^{3-}$ in vivo. The ratio of sugar as determined by microanalysis (15) to lipid ester as determined by a hydroxamate method (14) is 1:1 in the lipid so identified as DGDG, but 0.5:1 in the other two galactolipids, presumably MGDG and SL (cf. 1, 2). Only one of the three is radiolabeled in vivo by $^{35}SO_4^{2-}$, contains sulfur as determined chemically (18), and gives a positive response to a chromatographic spray reagent for sulfur detection ("Sulfvis"; Supelco, Bellefonte, PA); this lipid was identified as SL, and the third sugar-containing lipid was taken as MGDG. On paper (19) and in TLC (13) in the solvent systems described, each intact lipid as identified comigrates with the appropriate lipid standard commercially obtained (Sigma, St. Louis, MO; Serdary Labs, London, Ontario; Nu-Chek Prep, Elysian, MN), as do the corresponding water-soluble deacylation products prepared by mild alkaline hydrolysis (11). Additionally, the R_f values obtained for all the glycolipids so identified correlate well with published migration patterns in the TLC solvents employed (13).

Fatty acid ester preparation and analysis

Lipids were transesterified in 0.5 N sodium methoxide (20), and the fatty acid methyl esters produced were fractionated into subclasses based on unsaturation by argentation TLC (21). Quantitative ester recovery was demonstrated by the 98% recovery of radioactivity from di[1-^{14}C]palmitoyl phosphatidylcholine (New England Nuclear, Boston, MA) added as internal standard to a sample of algal lipid carried through the methyl ester preparation. The methyl esters were separated by gas–liquid chromatography (GLC) on a 10% stabilized diethylene glycol succinate glass column (Supelco, Bellefonte, PA) in an HP 5830A gas chromatograph (Hewlett-Packard, Chicago,

[2] Janero, D. R., and R. Barrnett. Submitted for publication.

[3] The marker enzymes assayed include: 5′-nucleotidase (EC 3.1.3.5), antimycin A-sensitive and insensitive NADH cytochrome c reductases (EC 1.6.2.2), nucleoside (inosine-5′) diphosphatase (EC 3.6.1.6), and cytochrome c oxidase (EC 1.9.3.1). In brief (Janero and Barrnett, submitted for publication), recoveries of marker enzyme activities were >98% with respect to starting homogenate, and the only marker detected in the thylakoid fraction was ~4% of the total homogenate cytochrome c oxidase activity.

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IL) operated isothermally at 200°C with carrier nitrogen flow at 25 ± 1 ml/min. Detector response was calibrated with standard fatty acid methyl ester mixtures (Supelco; Analabs, North Haven, CT; Alltech, Arlington Heights, IL); all analyses were carried out well within the linear response range. Identification of fatty acids was the result of combined information from several sources, principally retention times of known commercial ester standards and mathematical analyses of retention time-chain length relationships under the conditions employed (cf. 22). Quantitation of peak areas on resulting chromatograms was by computer integration, and conversion of relative ester areas to mole-percent composition was based on response factors obtained with the quantitative standards (22).

Statistical evaluations

Assessment of the significance of the difference between two means was made with a Student *t*-test (23); the significance level was set at the 95th confidence interval, $P < 0.05$ indicating a statistically significant difference.

Miscellaneous

Chlorophyll was quantitated spectrophotometrically in 80% acetone extracts using Arnon's coefficients (24). Cell number was determined by replicate hemacytometer counting of algae fixed in 0.25% glutaraldehyde (final concentration).

RESULTS

Glycolipids of *C. reinhardtii* 137$^+$

The individual glycolipids of the phototrophic alga and of thylakoid membrane purified therefrom are quantitated in **Table 1.** Three lipids, MGDG, DGDG, and SL, constitute all the glycolipid in the cells and thylakoid membrane. These three lipids taken together represent some 70% of total-algal and 80% of total-thylakoid polar lipid; the remainder was phospholipid and ether lipid (diacylglyceryl-trimethylhomoserine (DGTS) in roughly equal proportion.[2] The galactolipids MGDG and DGDG, which comprise more than 85% of the total-algal and thylakoid glycolipid masses, are found, respectively, in a ratio of ~3:1 in the cell and ~2.5:1 in the membrane. The sulfated glycolipid constitutes ~7% of the total polar lipid and ~10% of the glycolipid of the alga and makes a slightly greater contribution to these lipid classes in the thylakoid. At the cellular level, the concentration of SL is exceeded by phospholipid and ether lipid, but the content of SL in the thylakoid is greater than the membrane's phospholipid and ether lipid contents. The general predominance of glycolipid over other polar lipid types is accentuated in the thylakoid, due especially to the membrane's relatively low concentration of phospholipid.

As an indication of the subcellular glycolipid distribution, the mass of each glycolipid in the thylakoid as standardized to chlorophyll mass was divided by the mass of each in the cell so standardized. The resulting mass ratios (Table 1) should reflect the fraction of each glycolipid found in the alga's thylakoid, on the reasonable assumption that cellular chlorophyll is localized exclusively to the green lamellae. The ratios for MDGD, DGDG, and SL (0.49, 0.56, and 0.71, respectively) indicate that about half of the total algal galactolipid, but over 70% of algal SL, is localized in the photosynthetic lamellae. On average, then, most cellular glycolipid is associated with the thylakoid, but no glycolipid is exclusive to that structure.

Fatty acids of cellular and thylakoid glycolipids

The heterogeneity of each *Chlamydomonas* glycolipid and the extent to which, at the molecular level,

TABLE 1. Quantitation of glycolipids in *C. reinhardtii* 137$^+$

Lipid	μg/mg Chlorophyll		% of Total Polar Lipid		Mass Ratio TM/WC
	WC	TM	WC	TM	
MGDG	1291 ± 58	634 ± 22	45 ± 2.7	50 ± 1.5	0.49 ± 0.02
DGDG	443 ± 18	249 ± 11	15 ± 1.0	18 ± 0.7	0.56 ± 0.02
SL	206 ± 17	146 ± 5	7 ± 0.3	11 ± 0.3	0.71 ± 0.04
Phospholipid	391 ± 30	119 ± 4	14 ± 1.0	9 ± 0.3	
Ether lipid	344 ± 25	131 ± 6	13 ± 0.8	10 ± 0.4	

The mass of each cellular (WC) and thylakoid-membrane (TM) glycolipid as standardized to chlorophyll mass and the ratio for each glycolipid of its thylakoid and cellular masses so standardized are tabulated. The percent each glycolipid contributes to the total polar lipid complement of the cell and the thylakoid membrane is computed. For completeness, mass and percent-contribution values of the total phospholipid and total ether lipid at the cellular and thylakoid levels are also included and represent the sums of the various individual lipids of each type resolved by TLC (13). All values are means ± S.D.; n ≥ 6.

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TABLE 2. Major fatty acids of *C. reinhardtii* 137⁺ glycolipids

Fatty Acid	Mole Percent of Total Fatty Acids					
	MGDG		DGDG		SL	
	WC	TM	WC	TM	WC	TM
14:0	<1.0	1.0 ± 0.1	<1.0	1.5 ± 0.1	2.2 ± 0.2	2.4 ± 0.3[a]
14:1	<1.0	<1.0	<1.0	1.4 ± 0.1	1.7 ± 0.1	1.9 ± 0.1[a]
14:2	<1.0	<1.0	<1.0	<1.0	1.7 ± 0.2	1.8 ± 0.2[a]
16:0	1.1 ± 0.2	3.0 ± 0.1	22.4 ± 0.7	24.3 ± 1.6[a]	53.5 ± 3.9	48.1 ± 0.9[a]
16:1	<1.0	1.8 ± 0.1	2.3 ± 0.2	2.7 ± 0.1	2.8 ± 0.1	3.3 ± 0.1
16:2	1.3 ± 0.2	1.9 ± 0.1	3.1 ± 0.1	3.4 ± 0.1[a]	3.0 ± 0.1	2.1 ± 0.1
16:3	1.7 ± 0.1	2.0 ± 0.1	5.6 ± 0.1	6.5 ± 0.1	1.5 ± 0.1	2.4 ± 0.2
18:0	2.7 ± 0.1	4.7 ± 0.3	3.0 ± 0.2	3.1 ± 0.2[a]	5.4 ± 0.4	5.1 ± 0.3[a]
18:1	13.6 ± 0.2	15.2 ± 0.3	13.4 ± 0.4	10.6 ± 0.5	4.0 ± 0.2	4.5 ± 0.1
18:2	25.4 ± 0.4	23.2 ± 0.4	12.3 ± 0.3	12.1 ± 0.4[a]	4.7 ± 0.1	4.2 ± 0.2
18:3	49.1 ± 1.1	39.7 ± 0.7	25.6 ± 1.0	24.1 ± 0.6[a]	9.2 ± 0.5	9.5 ± 0.2[a]
18:4	<1.0	1.4 ± 0.1	1.1 ± 0.1	1.1 ± 0.1[a]	1.7 ± 0.1	3.1 ± 0.1
20:1	<1.0	<1.0	<1.0	<1.0	1.4 ± 0.1	1.3 ± 0.1[a]
20:2	<1.0	<1.0	1.1 ± 0.1	<1.0	1.8 ± 0.1	1.6 ± 0.1[a]
20:3	<1.0	<1.0	1.2 ± 0.2	<1.0	<1.0	1.0 ± 0.1
20:4	<1.0	<1.0	1.1 ± 0.1	1.2 ± 0.2[a]	1.4 ± 0.1	2.8 ± 0.2
22:1	<1.0	<1.0	<1.0	<1.0	1.0 ± 0.2	<1.0
22:2	<1.0	<1.0	1.9 ± 0.1	<1.0	2.6 ± 0.2	1.2 ± 0.1

[a] Means not significantly different ($P \geq 0.05$; Student's *t*-test) between WC and TM.

Fatty acids comprising >1.0 mole percent of each cellular (WC) and thylakoid-membrane (TM) glycolipid are quantitated. For each lipid, the sum of the tabulated mole percents is always >90%. All values are the mean mole percent ± S.D. for eight determinations. For each lipid, the difference between the respective cellular and thylakoid-membrane means of any fatty acid is significantly different ($P < 0.05$; Student's *t*-test) unless indicated.

the cellular glycolipids are reflective of those in the thylakoid membrane were evaluated through comparative analyses of fatty acid methyl ester derivatives prepared from the algal and lamellar glycolipids (**Table 2**). While a range of acyl lengths from C-14[4] to C-24 is encountered in every glycolipid, 80–90% of the esterified fatty acids are in the C-16 and C-18 series. Relative to the other glycolipids, MGDG (whether from the cell or the thylakoid) has the narrowest range of acyl lengths, with negligible contributions outside the hexadecanoic and especially octadecanoic families. Contrary to what is seen in the galactolipids, C-16 acids predominate over C-18 groups in SL. Both thylakoid (TM) galactolipids contain greater amounts of C-14 and C-16, but less C-18, acids than do their respective algal counterparts (WC).

MGDG is the most highly unsaturated glycolipid whether from the alga as a whole or from the thylakoid, and SL is the least unsaturated at both levels. Monogalactolipid is significantly (over 5-fold) more unsaturated than digalactolipid, the latter containing about one-third saturated acids. Although the unsaturated-to-saturated fatty acid ratios of cellular and thylakoid DGDG are the same (~2.4), thylakoid MGDG is about twofold more saturated than it is from the cell as a whole. However, thylakoid SL displays greater unsaturation over its cellular counterpart. No *trans*-unsaturated acids were detected in any glycolipid by methyl-ester argentation TLC.

[4] Two shorthand notations for fatty acids are used throughout. The number of carbon atoms in a fatty acid family is designated numerically (e.g., C-16 denotes the hexadecanoic series). Individual fatty acids are denoted by two numbers separated by a colon. The first number signifies the carbon chain length; the second, the number of unsaturated bonds.

Differences in chain length and saturation among the different glycolipids and with their cellular or subcellular source are based upon their individual fatty acids. C-18 unsaturated groups, notably 18:1, 18:2, and 18:3, comprise about 85% of the cellular and thylakoid MGDG acyl chains. In DGDG and SL, however, their contributions to the total fatty acid complements decrease to ~45% and ~17%, respectively. The saturated acid 16:0 is only ~2% of the total MGDG acids, but more than 20% of the DGDG acids and about 50% of the SL fatty acids, irrespective of whether the lipids are from the cell or the thylakoid.

Statistically significant differences between the fatty acid complements of each respective cellular and thylakoid glycolipid are most extensive in MGDG; thylakoid MGDG contains the higher proportions of all major acids except 18:2 and 18:3. Cellular DGDG has somewhat greater amounts of 18:1, 20:2, 20:3, and 22:2 than does thylakoid DGDG; the latter, how-

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ever has more 14:0 and 14:1. Differences between SL of the whole alga and SL of its photosynthetic lamellae include lower proportions of 16:1, 16:3, 18:1, and 20:4, but greater amounts of 16:2 and 18:2, at the cellular level.

DISCUSSION

Studies of green-plant lipids have concentrated largely on those few tissues, such as *Spinacia oleracea*, for which fractionation procedures are sufficiently sophisticated to enable isolation of thylakoid and even some non-green organelles in reasonably homogeneous form. Technical limitations of cellular fractionation have tended to restrict study of the lipids of lower green plants to the tissue as a whole. In this regard, phytoflagellates have been employed mainly to monitor changes in gross fatty acid composition either upon etiolation (25) or upon alteration of growth conditions (26). The unicellular green alga *Chlamydomonas reinhardtii* 137^+ (wild-type) represents a singularly attractive material for more detailed investigation of lower green-plant lipid biochemistry, especially since it contains large amounts of thylakoid which can be purified rather easily in high yield. Subcellular fractionation facilitates direct biochemical study of this important energy-transducing structure in the unique plant organelle, the chloroplast.

Previous biochemical investigation of the lipids of *Chlamydomonas* have been concerned with mixotrophic (i.e., light-grown in acetate-supplemented media) or heterotrophic (i.e., etiolated and dark-grown in acetate-supplemented media) mutants: the arginine-requiring strain ss (27), the streptomycin-resistant, arginine-requiring strains sr_3 and sr_{35} (27), and the yellow mutant y-1 (7, 25, 28, 29). The glycolipids detected in the phototrophic wild-type alga, strain 137^+, also appear in these mutants as the exclusive cellular glycolipids, together comprising some 70–80% of total-algal polar lipid in all the *Chlamydomonas* strains studied so far. The non-etiolated mutants, however, generally contain larger proportions of DGDG than does strain 137^+, making their MGDG-to-DGDG ratio 2:1 or less, as opposed to ~3:1 in the wild-type strain. Conversely, the mutants have some twofold less SL than the wild-type, although in all five *Chlamydomonas* strains SL is the glycolipid present in least amount.

Comparisons of the fatty acids esterified to individual glycolipids from wild-type and mutant *Chlamydomonas* cells can be made only to a limited degree because of lack of data on the y-1 strain. Information about the ss and sr mutants (27) encompasses only the C-16 and C-18 acids, the dominant acyl components of all three glycolipids, as in the wild-type analyzed here. Other qualitative similarities among strains 137^+, ss, sr_3, and sr_{35} include large amounts of 18:3 in MGDG and of 16:0 in SL, making these the least saturated and most saturated glycolipids, respectively.

MGDG, DGDG, and SL have been detected in crude photosystem particles prepared from strain sr_3 (30) and in a green membrane pellet from strain 137^+ (31). The lack of characterization of these *Chlamydomonas* subcellular fractions makes the few quantitative conclusions advanced in these reports uncertain. Only one study (28) other than the present investigation has utilized a photosynthetic membrane fraction of analytical quality to gain subcellular information about *Chlamydomonas* lipids. Radiochemical quantitation of the lipid components of thylakoids prepared from non-etiolated y-1 cells has detected MGDG, DGDG, and SL in the membrane at proportions similar to those we have found for the photosynthetic lamellae of wild-type 137^+. From the distribution of radioactivity assimilated during long-term labeling and associated with each cellular and thylakoid glycolipid as standardized to chlorophyll, dePetrocellis, Siekevitz, and Palade (28) concluded that no glycolipid is exclusive to the y-1 thylakoid, a finding supported by our data on strain $137.^+$ Their determination that ~75% of cellular MGDG, but only ~50% of cellular DGDG and SL, is localized in thylakoid membrane contrasts with our result that SL is the only glycolipid predominantly (~70% of that in the alga) associated with thylakoid. The inability at present to obtain fractions of any intracellular membrane other than thylakoid precludes direct study or definitive localization of extra-thylakoid glycolipid in any *Chlamydomonas* strain; perhaps, as for spinach, significant amounts are associated with the double-membrane chloroplast envelope (4).

It remains to be determined to what extent the differences in glycolipid biochemistry among the five *Chlamydomonas* strains (137^+, y-1, ss, sr_3, and sr_{35}) reflect lipid/fatty acid-synthesizing capabilities, the activity of specific acyltransferases, or the presence of an external carbon source (acetate) during the mixotrophic culture of the mutants. Despite uncertainties as to the physiological basis for such biochemical differences, the quantitative prominence of glycolipid in all these *Chlamydomonas* strains and, for strains 137^+ and y-1, in the thylakoid as well, is striking. Additionally, the contrasting fatty acid complements between each respective cellular and thylakoid-membrane glycolipid which we have detailed in the wild-type provide the first evidence that discrimination of glycolipid type at the molecular level occurs in

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Chlamydomonas with respect to (at least) the alga's photosynthetic membrane.

More general comparison of our data on phototrophic, wild-type *Chlamydomonas* glycolipids with the glycolipids of some dozen other green plants (32) reinforces the general quantitative importance of glycoglycerolipid in green plant tissue. Some higher plants, additionally, contain low levels of two other glycolipids, trigalactosyl diglyceride and tetragalactosyl diglyceride (33), not detected in *Chlamydomonas*. Lack of detailed analysis of the lipid composition of a wide variety of green plant tissues and, especially, their purified thylakoid and the fragmentary nature of most of the data at hand preclude quantitative generalization over all green plants concerning the biochemistry and function of the glycoglycerolipids. The ubiquity of MGDG, DGDG, and SL must yet be reconciled with often large quantitative variations among species, algal strains, and even intraorganellar membranes (4) in the amounts of individual fatty acids associated with any particular glycolipid. Such variety of acyl groups suggests that the predominance of glycolipids in green plants may not be as important functionally as the ability of the cell to biogenetically establish and maintain particular glycolipid species in the membranes that contain them. The biogenetic discrimination would lend to each membrane the biophysical characteristics of its glycolipid acyl groups. Experimental support for the suggestion comes from recent analyses on model membrane systems (34) which indicate that glycolipid imparts to the thylakoid membrane bilayer an unsaturated character essential to thylakoid fluidity.

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