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Preliminary communication

Synthesis of a new class of model glycolipids

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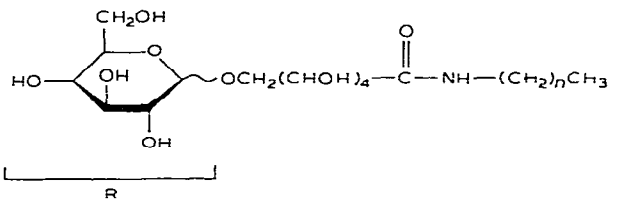
Recent awareness of the importance of carbohydrate-containing molecules to membrane structure and function has promoted the synthesis of glycolipids[1–11], in both naturally occurring and in model form Among their many applications, glycolipids have been investigated as detergents for the solubilization of membrane components[12] and as receptors for such carbohydrate-binding proteins as lectins[1] Regardless of the use of such compounds it would be advantageous to have procedures whereby relatively large amounts of pure glycolipids could be obtained simply and quickly Most synthetic procedures are tedious and involve long sequences of reactions that do not always yield reasonable quantities of pure compounds We now report the preparation of a new class of model glycolipids (**1**) that may be prepared readily in gram quantities in chromatographically-pure form



R = α-D-galactopyranosyl , β-D-galactopyranosyl, α-D-glucopyranosyl , or β-D-glucopyranosyl

n = 7, 9, 11, 13 or 15

1

Originally, *N*, *N'*-dicyclohexylcarbodiimide (DCC) was added to catalyze the formation of *N*-substituted aldonamides from aldonic acids and amines However, the DCC was found to make a minor contribution to the condensation reaction between acid and amine The coupling reaction most likely proceeds *via* the lactone[10], and aldonic acids are readily converted into lactones, even on dissolution in non-aqueous solvents Nevertheless addition of the carbodiimide ensures that the coupling reactions go to completion when free acids are used as reactants

Preparation of all compounds in the scheme followed the same procedure, except that the *N*-alkylmaltobionamide glycolipids were recrystallized from ethanol rather than from methanol Occasionally, the product appeared as a gel during recrystallization However, repeated recrystallization provided an analytically pure, crystalline product Those compounds that assumed a gel-like appearance during purification included the melibionamides, cellobionamides, and gentiobionamides, the maltobionamides and lactobionamides generally formed crystals more readily Some glycolipids crystallized better at 4° than at 25°

Purity of the glycolipids was readily monitored by t l c , the R_F values of the C_{12} alkyl derivatives in 9 6 3 1 1-butanol—acetic acid—diethyl ether—water were melibionamide (0 40), cellobionamide (0 48), and maltobionamide (0 50) Shortening the alkyl chain lowers the R_F values, whereas lengthening the chains increases the values The glycolipids were found to be ninhydrin- and aldose-positive to spray reagents only after acid hydrolysis The glycolipids showed the characteristic amide II (1550) and amide I (1655 cm^{-1}) bands in the i r

This synthetic procedure provides several advantages over methods for preparatio of other model glycolipids Synthesis and purification requires only a few days Products are obtained in high yields as pure solids, and gram quantities may be obtained with a variety of sugars, linkages, and alkyl chain-lengths The availability of sufficient quantities of these pure, well characterized, model substances should facilitate studies on the role and function of glycolipids in biological systems

As with naturally occurring glycolipids, these model substances possess a hydrophobic sugar portion and a non-polar, alkyl chain In common with glycosphingolipids, the synthetic glycolipids also contain an amide linkage The synthetic glycolipids have several interesting and useful properties They form micelles that are specifically precipitated by appropriate lectins Precipitin curves between the glycolipids and several lectins have been obtained and in all cases, precipitation is initiated at the critical micelle concentration and may be inhibited completely by specific sugars These glycolipids act as detergents in causing lysis of red blood cells The following example typifies the general procedure

Materials — Ninhydrin diphenylamine, 1-octylamine, 1-decylamine, 1-dodecylamine, 1-tetradecylamine, and *N,N'*-dicyclohexylcarbodiimide were obtained from Aldrich Chemical Company (Milwaukee, Wisconsin) Melibiose, lactose, maltose, cellobiose, and gentiobiose were purchased from Pfanstiehl Chemicals (Waukegan, Illinois) Amberlite cation-exchange resin, IR-120 C P , was obtained from Mallinkrodt (St Louis, Missouri) Precoated silica gel G-60 t l c plates were obtained from Brinkmann Instruments, Inc (Des Plaines, Illinois)

Free reducing sugars were detected on t l c plates by spraying with diphenylamine (2 3%) in butanol followed by heating at 120° Glycolipids were detected by spraying chromatograms with a solution containing 20 mL of diphenylamine (10% in ethanol), concentrated hydrochloric acid (100 mL), and acetic acid (80 mL), t l c plates were covered with a glass plate and heated for 30 min at 120°

Free amines were detected by spraying plates with ninhydrin (0 2%) in ethanol Amide linkages were hydrolyzed by spraying t.l c plates with M hydrochloric acid, after which time the plates were covered and heated for 20 min at 120° Ninhydrin spray was used for detection of amines liberated

N-*Dodecyllactobionamide* – Lactose was oxidized to potassium lactobionate[13] and then converted into the free acid by treatment with Amberlite IR-120 (H^+) and the aqueous solution was evaporated and dried *in vacuo* at 40° Repeated evaporation from methanol and ethanol[14] converted the acid into lactobiono-1,5-lactone Lactobiono-1,5-lactone (1 g, 2 8 mmol) and *n*-dodecylamine (0 6 g, 2 3 mmol) were dissolved in methanol (10 mL) by gentle heating and the mixture was stirred overnight at room temperature The resultant precipitate was filtered off, washed with methanol, and air dried Repeated recrystallization from methanol gave *N*-dodecyllactobionamide as a crystalline, white solid that gave a single spot (R_F 0 34) in t.l c , yield 90%, m p 144–145°

Anal Calc for $C_{24}H_{47}NO_{11}$ C, 54 8, H, 9 08, N, 2 53, Found C, 54 50, H, 8 76, N, 2 53

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