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RECENT ASPECTS OF THE CHEMISTRY OF DISACCHARIDES

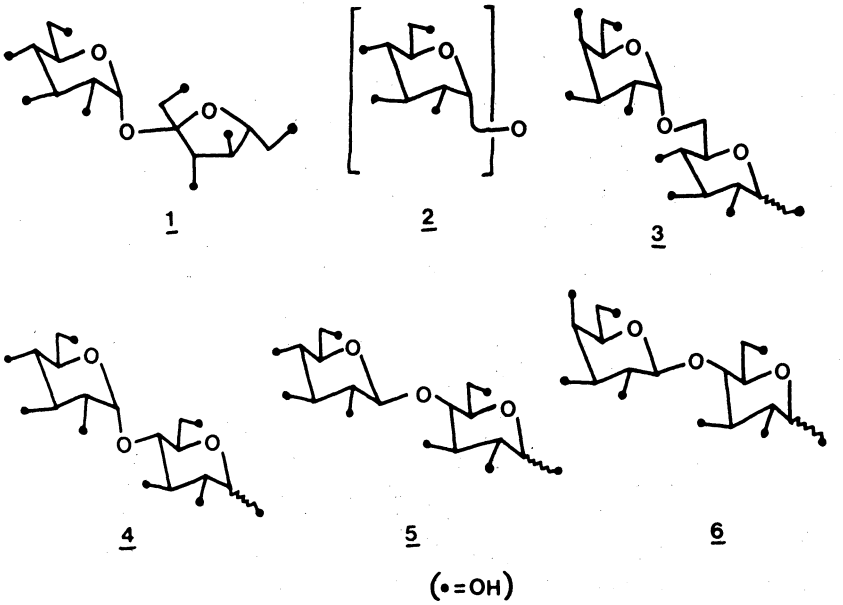
Leslie Hough and Anthony C. Richardson

Department of Chemistry, Queen Elizabeth College, Kensington, London W8 7AH U.K.

ABSTRACT - Our interest has focussed on stereoselective chemical manipulations of readily available disaccharides either by replacement of specific hydroxyl groups, for example, by halogenation, or by selective esterification of the more reactive hydroxyl groups. In the latter, the unreacted hydroxyl groups are subsequently modified. Another approach has been to prepare selected sulphonate esters by stereospecific reactions followed by the introduction of new substituents by nucleophilic displacement of the sulphonyloxy groups. The observed stereoselectivity in the latter nucleophilic substitutions and also that encountered in the halogenations, using either sulphuryl chloride-pyridine or mesyl chloride-N,N-dimethylformamide, was in general accordance with the empirical bimolecular, nucleophilic, transition state theory.

The rapid progress made in these studies of the complex chemistry of disaccharides has been due entirely to the application of mass spectrometry, ^{1}H n.m.r. and ^{13}C n.m.r. to the structure determination of the large variety of products encountered.

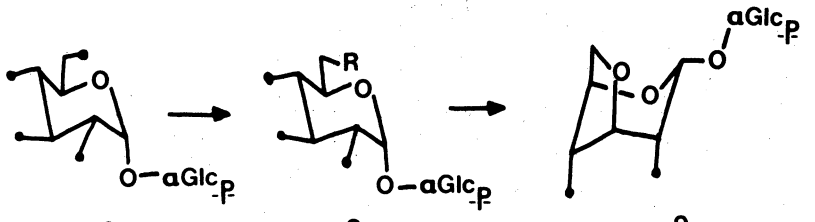
The chemistry of the readily available disaccharides, namely sucrose (1), α,α-trehalose (2), melibiose (3), maltose (4), cellobiose (5) and lactose (6), have been explored with a view to their economic conversion into compounds of either chemical or biological utility. In



contrast to petrochemicals, these derivatives, in common with other natural carbohydrates, are replenishable and consequently likely to assume increasing importance for commercial exploitation as the fossilised materials become increasingly scarce and expensive. At the present time sucrose is available on a larger scale than any other pure and crystalline organic substance. We have explored the chemistry of this group of disaccharides with emphasis upon their unique properties, in each case dependent upon their individual stereochemistry and conformations.

The simplest disaccharide is α,α-trehalose (2; α-D-glucopyranosyl α-D-glucopyranose).

Because of its two-fold axis of symmetry, it was thought that the reactions of α,α-trehalose would be simple, resembling those of methyl α-D-glucopyranoside (Ref. 1). Indeed, symmetrical derivatives of α,α-trehalose are readily available, in both the gluco and galacto series, by conventional methods, including amino, halogeno (e.g. 7) and deoxy derivatives at C-2, C-3, C-4 and C-6 (Ref. 1 to 10). However, as the chemistry was probed, unsymmetrical derivatives gradually emerged, putting a subtle and complex interpretation upon the chemical behaviour by distinguishing chemically between the two glucopyranosyl units of each trehalose molecule (Ref. 4, 11 and 14). As a simple illustration, α,α-trehalose (2) can selectively be mono-substituted at C-6, either directly (Ref. 14) or via the 6-tosylate



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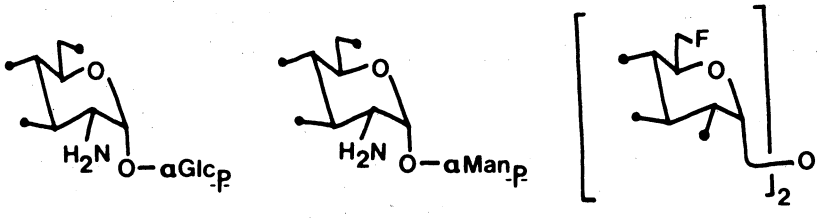


10 11 7

(• =OH)

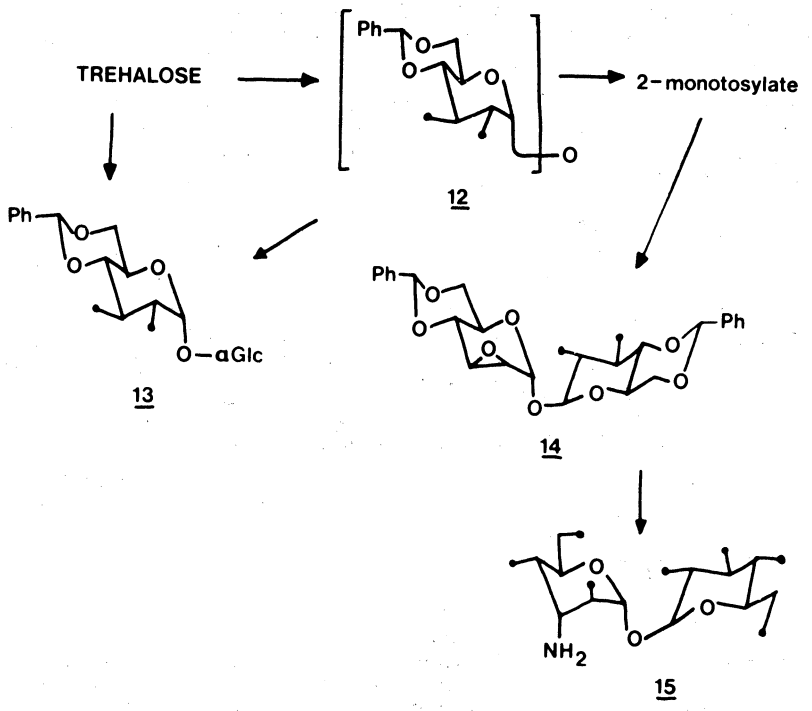
(Ref. 1 and 10) to give the 6-deoxy-6-iodo derivative (8; R=I) from which the 6-deoxy (8;R=H) and 3,6-anhydro derivatives (9) are readily prepared (Ref. 12, 13 and 14).

It is appropriate to recall that α,α-trehalose (2) occurs as the blood sugar of insects and in fungi and yeasts as a reserve carbohydrate. Since it is metabolised by hydrolysis with the enzyme trehalase to D-glucose, inhibitors of this hydrolytic enzyme could possibly function as insecticides or fungicides. In this connection, two unsymmetrical trehalose derivatives (Ref. 15 and 16), namely trehalosamine (10) and its D-manno epimer (11) from streptomyces broths, have antibiotic activity. Apart from 6,6'-dideoxy-6,6'-difluoro-α,α-trehalose (7) (Ref. 6), none of the symmetrical derivatives have shown significant activity against insect and mammalian trehalases. Non-symmetrical amino- and fluoro- derivatives, in which one of the α-D-glucopyranosyl units remains intact, might therefore prove more effective as inhibitors of trehalase.

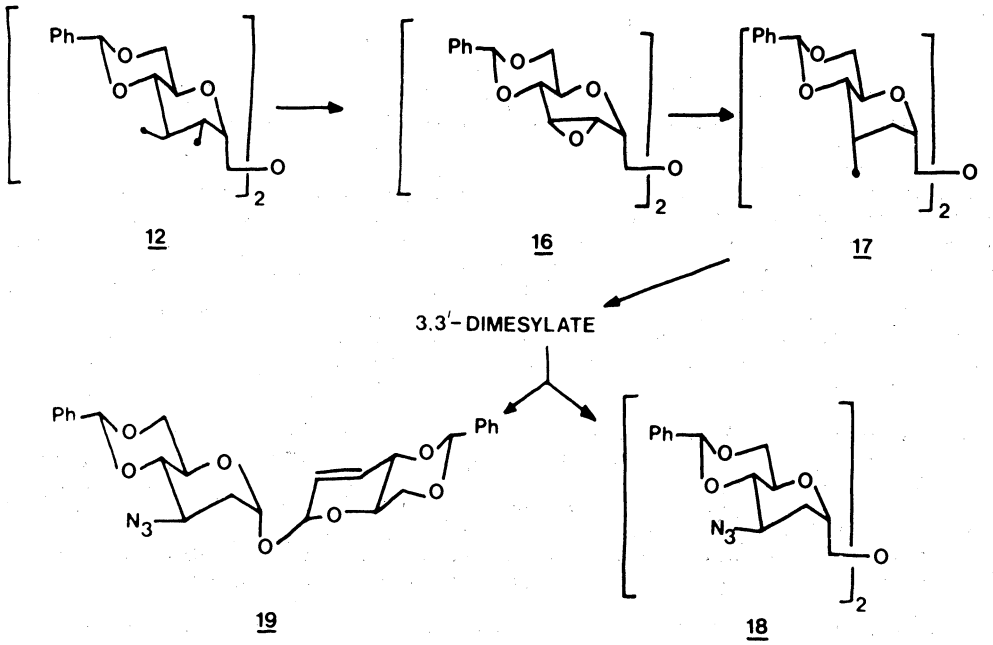
An important route to non-symmetrical derivatives came from the observation that the water-insoluble 4,6:4',6'-di-O-benzylidene acetal (12) as its tetrabenzoate, can be methanolysed selectively to give the water soluble mono-4,6-O-benzylidene derivative (13) in good yield (Ref. 17). The method has been improved by Lee (Ref. 18) using a direct benzylidenation reaction on trehalose (2). Benzoylation of the monoacetal 13 and subsequent inversion of configuration at C-4 by nucleophilic substitution of the derived 4,6-dimesylate with benzoate gave, after removal of the protective groups, α-D-galactopyranosyl α-D-glucopyranose.

Selective tosylation of the dibenzylidene acetal 12 gave the 2-tosylate from which the mono-2,3-epoxide (14; D-manno-D-gluco) is readily accessible (Ref. 19). Ring opening of this epoxide (14) with azide (Ref. 7) followed by removal of the benzylidene group and reduction, afforded the 3-amino-3-deoxy derivative (15; D-altro-D-gluco).

Another example of the formation of an unsymmetrical derivative arose when the dibenzylidene acetal (12) was converted sequentially into its 2,3;2',3'-tetratosylate, then 2,3;2',3'-di-epoxide (16; D-allo-D-allo), and finally into its 2,2'-dideoxy derivative (17; D-ribo-D-ribo)

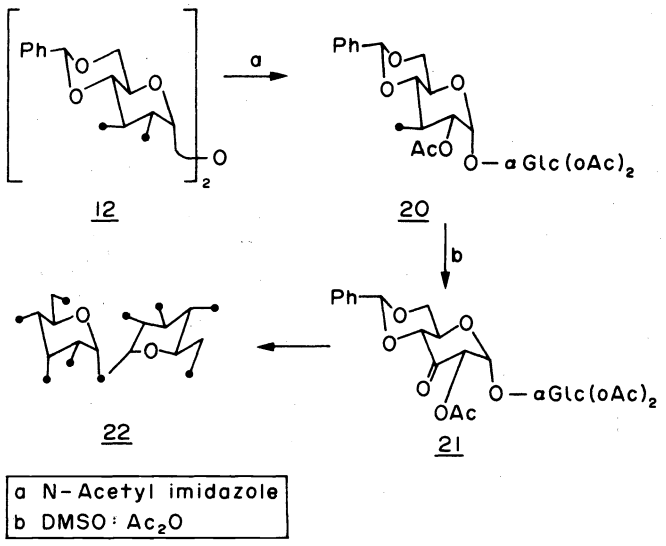


which upon mesylation followed by treatment with sodium azide in H.M.P.T. gave two products, the expected 3,3'-diazide (18; D-arabino-D-arabino) together with the unsymmetrical 3-azido-2'-ene (19; D-arabino-D-erythro) as a result of elimination competing with nucleophilic substitution (Ref. 4).

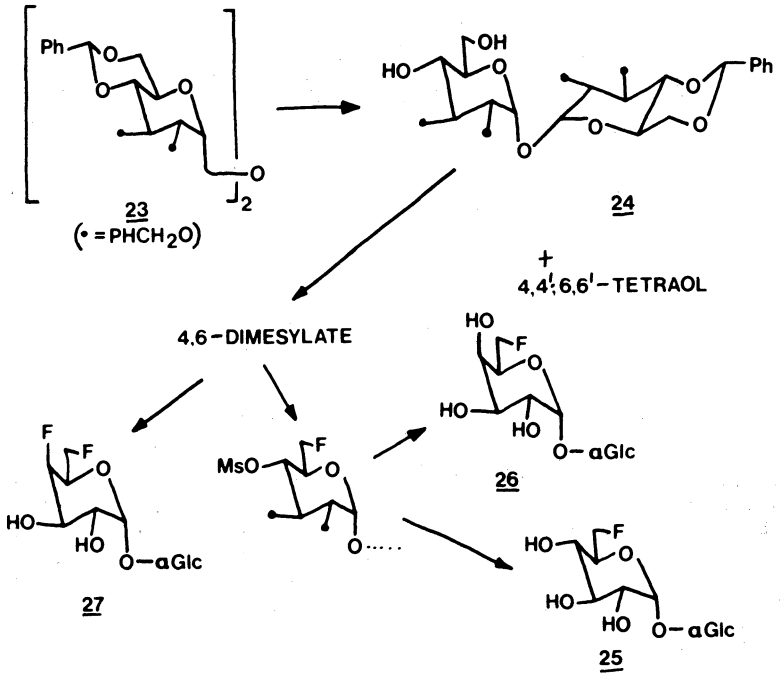


Bar-Guilloux et al (Ref. 20) observed that the dibenzylidene acetal (12) undergoes selective esterification with N-acetyl imidazole to give the unsymmetrical 2,2',3'-triacetate (20) which on oxidation and stereoselective reduction of the resulting 3-ulose (21) afforded α-D-allopyranosyl α-D-glucopyranoside (22). Unsymmetrical analogues of trehalose have also been synthesised by the French group (Ref. 20) by condensation of 2,3,4,6-tetra-O-acetyl-D-

glucopyranose with the appropriate benzoylated pyranosyl bromide. By this method they isolated α-D-glucopyranosyl α-D-xylopyranoside, α-D-allopyranosyl α-D-glucopyranoside and α-D-glucopyranosyl α-D-mannopyranoside; the latter was a competitive inhibitor of cockroach trehalase.

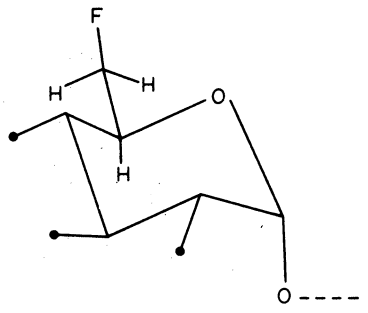


The selective methanolysis of the tetrabenzyl ether of the 4,6;4',6'-dibenzylidene acetal (23) gave the 4,6-diol (24) which was exploited for the synthesis of 6-deoxy-6-fluoro- (25) and 4,6-dideoxy-4,6-difluoro-trehalose and α-D-galactopyranosyl α-D-glucopyranoside and its 4-deoxy-4-fluoro-, 6-deoxy-6-fluoro (26) and 4,6-dideoxy-4,6-difluoro (27) derivatives by nucleophilic substitution reactions using tetra-N-butyl ammonium fluoride (Ref. 21), Characterisation of the products was facilitated by the use of ^{19}F n.m.r. spectra, in which it was of interest to note that the coupling constants favoured the rotamer about the $-CH-CH_2F$ bond in 25 with the C-6—F bond anti-periplanar to the axial C-5—H bond (25a).

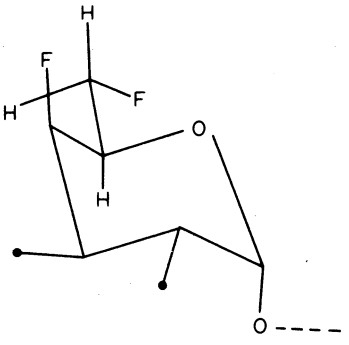


On the other hand, the data for the galacto derivatives 26 and 27 indicated the gauche form (27a) which undoubtedly arises from the influence of the axial group at C-4. The 4,6-diol (24) also served as a precursor of 4,6-diacetamido-4,6-dideoxy-trehalose (29) via the 4,6-dichloride (28; galacto-gluco), and of 4-amino-4,6-dideoxy-6-fluoro-α-D-galactopyranosyl

α-D-glucopyranoside HCl (30). In contrast to the previous ^{19}F n.m.r. results, the spectrum

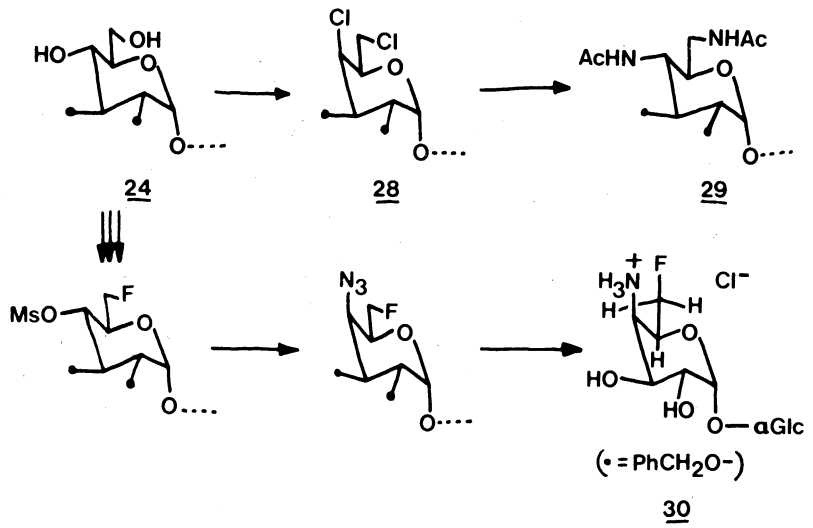


25a



27a

of the latter (30) suggested that C-6—F bond is antiperiplanar to the C-5—H bond due to the parallel alignment of the opposing C-F and C-NH_3^+ dipoles at C-6 and C-4 respectively (Ref. 21).



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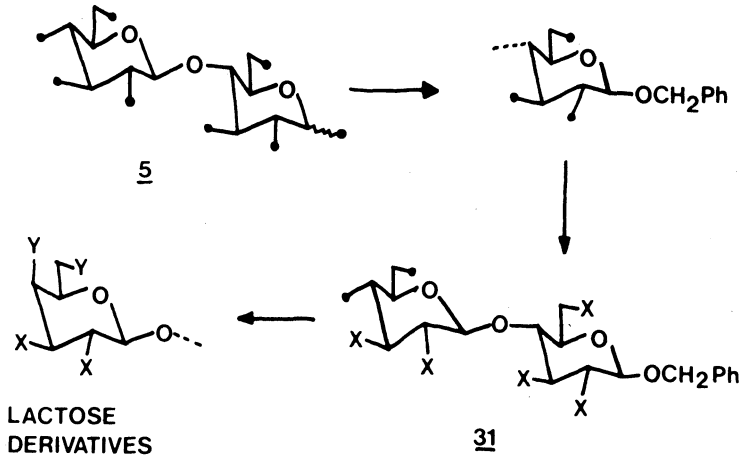
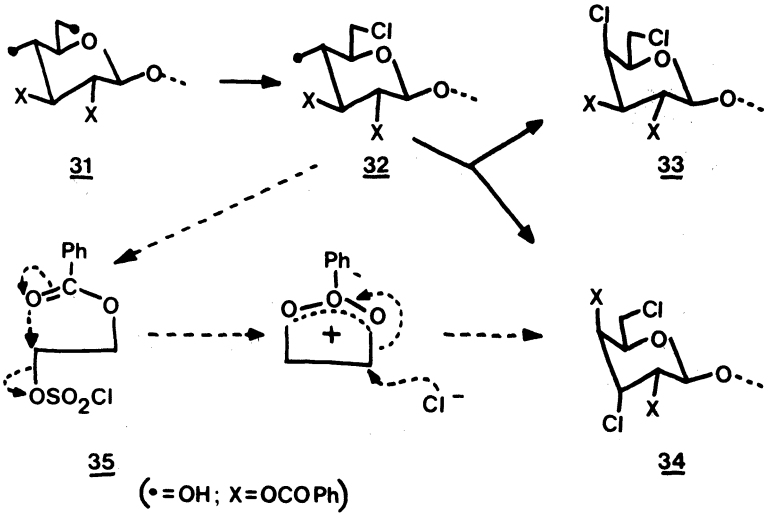
In an assay for activity against trehalase, an enzyme isolated from the flight muscle of the green bottle fly, Lucilia sericata, 6-deoxy-6-fluoro-trehalose (25) acted as a competitive reversible inhibitor with an affinity some 1.4 times that of the natural substrate trehalose (Ref. 22).

Amino-, chloro- and fluoro-lactoses, with these substituents at C-4' and C-6', have been synthesised from cellobiose (5), using benzyl β-cellobioside 2,2',3,3',6-penta-benzoate (31) as the key intermediate since it is readily available via the 4,6-benzylidene acetal (Ref.23). Nucleophilic displacements of the 4',6'-disulphonate of 31 occurred with inversion of configuration at C-4' (i.e. cellobiose ⟶ lactose) but the rate was abnormally slow when compared with maltosides, an effect that we attribute to unfavourable stereoelectronic interactions between the incoming nucleophile and the β-anomeric substituent at C-1'. Reaction of the pentabenzoate 31 with sulphuryl chloride in pyridine gave the 6'-chloride (32) initially, then the expected 4',6'-dichloride (33) together with the 3',6'-dichloride (34 (D-gulo-D-gluco) as a consequence of participation by the neighbouring 3-benzoate group



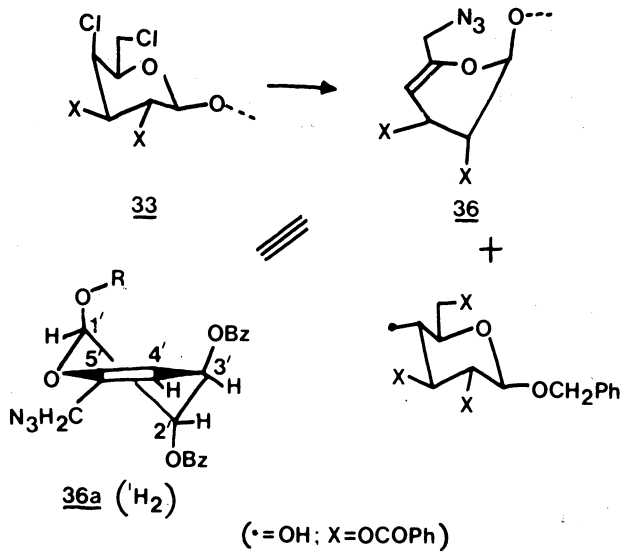
in the displacement of the 4'-chloro-sulphonyloxy group (35). When the 4',6'-dichloro-lactoside (33) was treated with sodium azide, no 4',6-diazide was detected but elimination

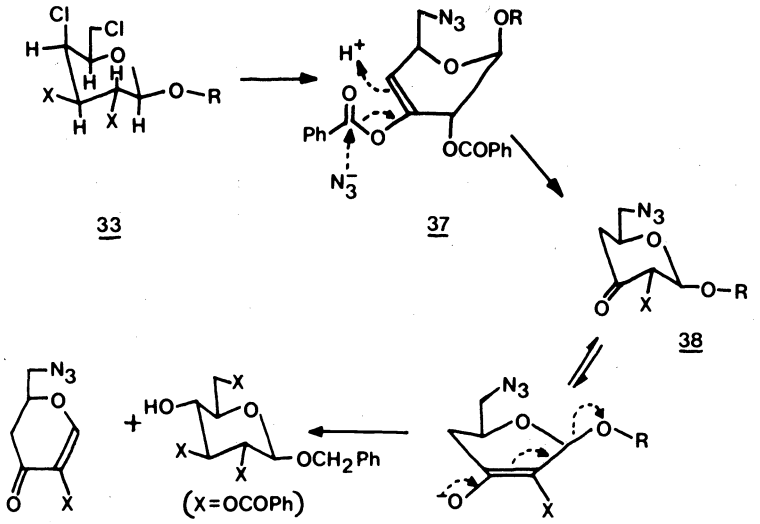


of the axial 4'-chloro group predominated to give the 6'-azido-4'-ene (36; $^{1}H_{2}$ conformation 36a), together with a substantial amount of benzyl tri-O-benzoyl-β-D-glucopyranoside (Ref. 23). The latter could arise from the intermediary vinylic benzoate (37) with the formation of the 3'-ulosyl derivative (38) which would then undergo β-elimination with cleavage of the inter-glycosidic linkage.

In a similar sequence of reactions (Ref. 24) methyl β-lactoside (39) was transformed into 4',6'-diacetamido-4',6'-dideoxycellibiose hexaacetate (40). As before, the nucleophilic substitution position to give the 4',6'-diazide (41; 36% yield) was accompanied by elimination of the axial 4'-methylsulphonyloxy group to give methyl 4-O-(6-azido-2,3-di-O-benzoyl-4,6-dideoxy-α-L-threo-hex-4-enopyranosyl)-2,3,6-tri-O-benzoyl-β-D-glucopyranoside (42; 35% yield) and by interglycosidic cleavage, methyl 2,3,6-tri-O-benzoyl-β-D-glucopyranoside (10% yield). On the basis of ^{1}H n.m.r. data, the hex-4-enopyranosyl ring of 42 was assigned the $^{1}H_{2}$ conformation (36a) in which the large substituents at C-1', C-2' and C-3' are held in axial or quasi-axial orientations (Ref. 24). In addition to the anomeric effect, Ferrier and Sankey (25) showed that pyranoids containing an endocyclic double bond allylic ester group favour quasi-axial orientations.

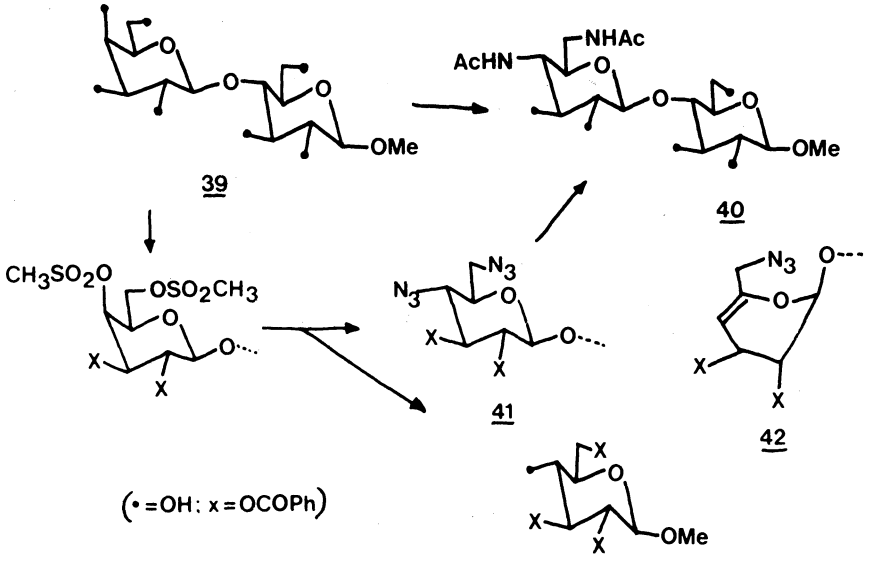
A detailed study (Ref. 26) of the selective benzoylation of methyl β-lactoside (39) revealed that the order of reactivity of the hydroxyls is 6' > 3' > 6 > 2 > 2',4' > 3. This approach

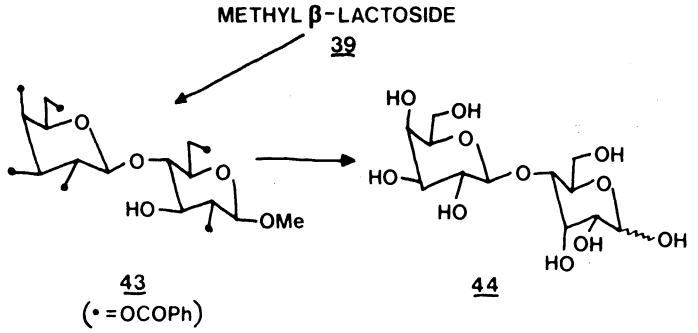


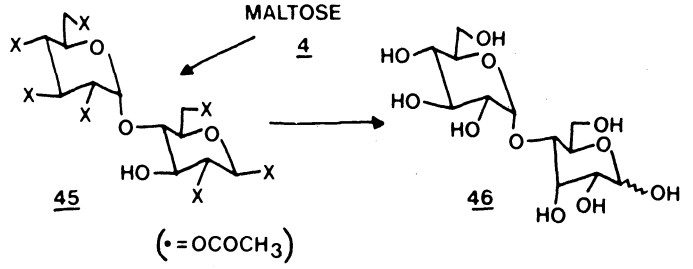


yields specifically blocked synthetic intermediates which would otherwise be available only after a long sequence of reactions. In an alternative approach, Chiba, Hoga and Tojima (27) studied chemical modifications of 1,6-anhydro derivatives of disaccharides, including the selective benzoylation of the secondary hydroxyls in 1,6-anhydro-4,6-O-benzylidene-β-lactose.

Lactose (6) was transformed into the 3-epimer, namely 4-O-β-D-galactopyranosyl-D-allopyranose (44) by exploiting the selective hexabenzoylation (43; 33% yield) of methyl β-lactoside (39) (Ref. 28). The hydroxyl groups at C-3 of lactose (6), maltose (4) and methyl β-maltoside are similarly resistant to esterification (Ref. 29 and 30). The slow esterification of this hydroxyl group in maltose has been correlated with the strong intramolecular hydrogen bond between the 3-hydroxyl and the 2'-hydroxyl groups; but the related behaviour in lactose, which cannot form such a hydrogen bond, suggests a more profound explanation (Ref. 30). The maltose heptaacetate (45) was utilised in the synthesis of 4-O-α-D-gluco-pyranosyl-D-allopyranose (46) and the related 3-chloro analogue (47), 3-azido and 3-oxo (48) derivatives (Ref. 30); the latter was readily epimerised to the 2-axial epimer (49; D-gluco-D-arabino).

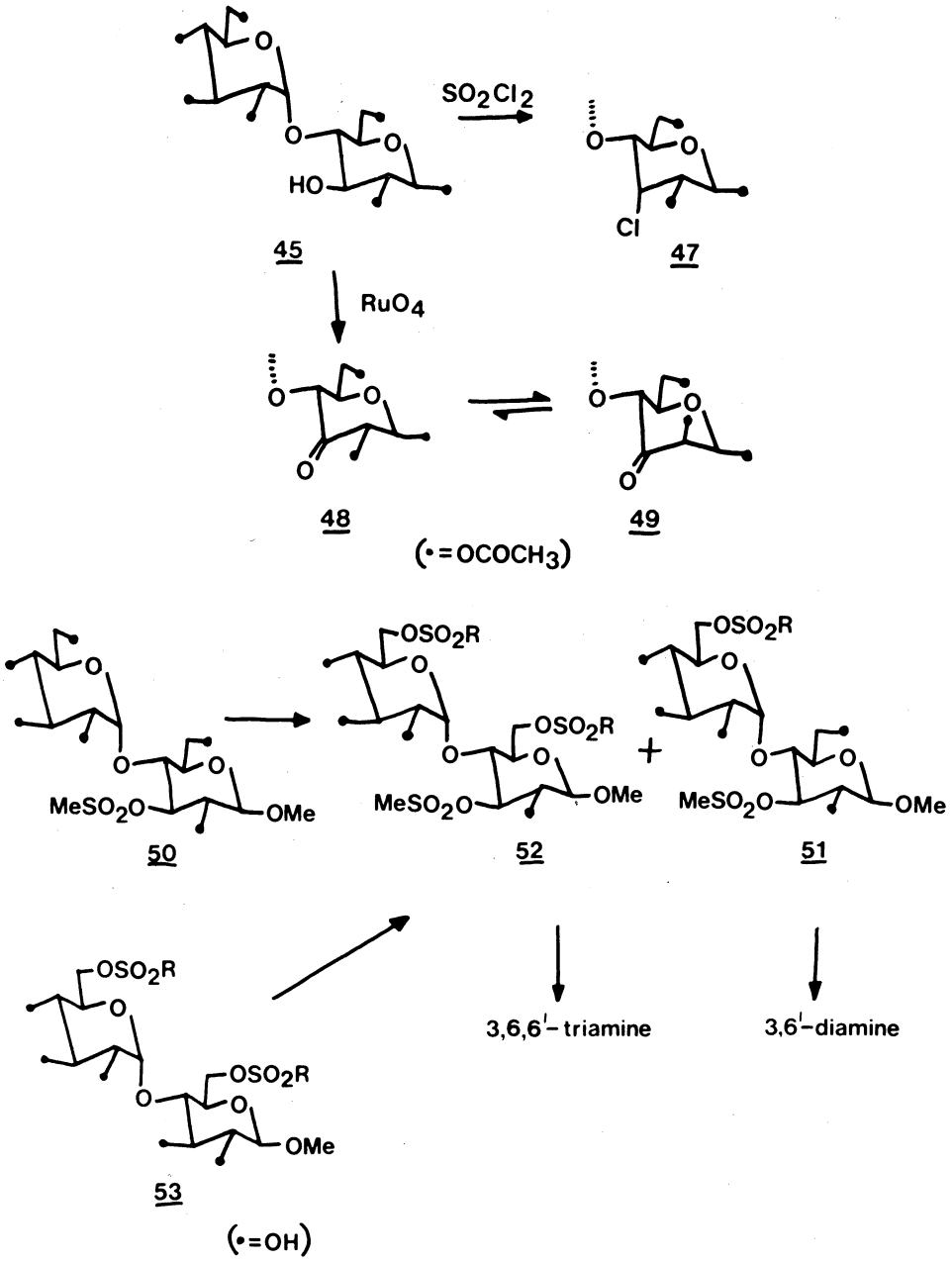




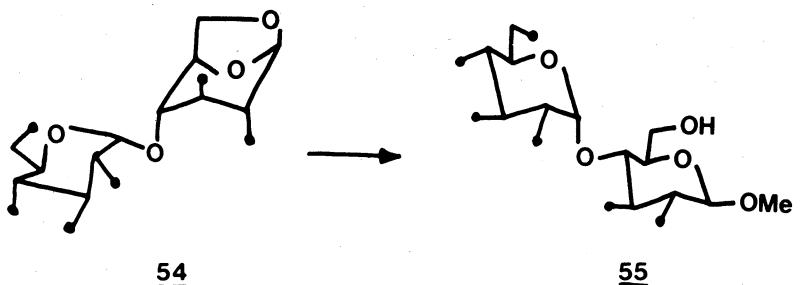


Using a similar approach 3,6'-disulphonate (51) and 3,6,6'-trisulphonate (52) esters were synthesised from methyl 3-O-mesyl-β-maltoside (50) by selective primary tosylation; subsequent azide substitution followed by reduction gave the corresponding 3',6- and 3,6,6'-amines (Ref. 31). The 3,6,6'-trisulphonate (52) is more readily prepared by selection tetra-O-acetylation of methyl 6,6'-di-O-tosyl β-maltoside (53) and subsequent mesylation at C-3 (Ref. 32). 6-Substituted derivatives of maltose can be prepared by ring-opening of 1,6-anhydro-β-maltose hexaacetate (54) to give methyl β-maltoside hexaacetate (55) with a free 6-hydroxy group (Ref. 30 and 33).

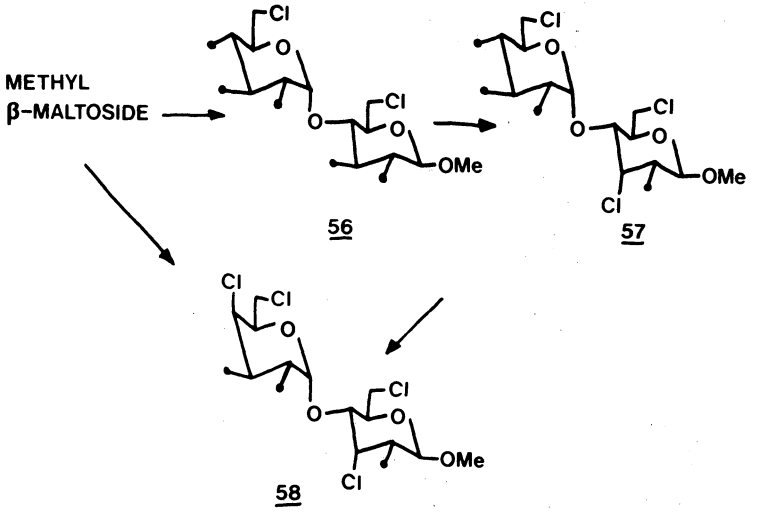
Studies on the selective replacement of hydroxyl substituents in disaccharides by chloro groups has given a variety of useful precursors of amino-, deoxy- and other derivatives. Thus mesyl chloride in conjunction with N,N-dimethylformamide (D.M.F.) reacted with methyl β-maltoside and benzyl β-cellobioside to give products beyond the stage of primary substitution (Ref. 34). For methyl β-maltoside, the first-formed 6,6'-dichloride (56) was transformed into the 3,6,6'-trichloride (57; D-gluco-D-allo), and subsequently into 3,4', 6,6'-tetrachloride (58; D-galacto-D-allo.



The reaction with benzyl β-cellobioside was more complex, with the introduction of chloro substituents at C-6, C-6', C-3, C-3' and C-4'. In each case, chlorination at secondary positions occurred with inversion of chirality and were consistent with substitution at

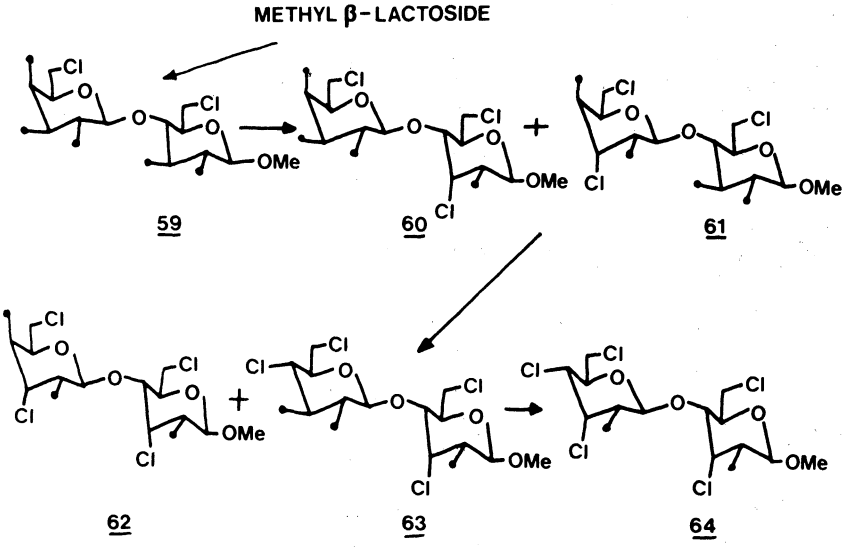


positions that are favourable to S_N2 type transition states, according to the general theory proposed by Richardson (Ref. 35). Under mild conditions methyl β-lactoside gave the expected 6,6'-dichloride (59) but at 94° for 9 days a mixture of at least ten products



56 57 58

resulted, from which the 3,6,6'- and 3',6,6'-trichlorides (60 ; 61), the 3,3',6,6'- and 3,4',6,6'-tetrachlorides (62 ; 63) and the 3,3',4',6,6'-pentachloride (64) were isolated (Ref. 28). It is noteworthy that the displacement occurred at C-3' of the lactoside despite the vic-axial group at C-4' which should have hindered the reaction; however, the high reactivity of the 3'-hydroxyl group to esterification could lead to an abnormally high concentration of the intermediary 3'-O-forminium ester ($R\text{-}O\text{-}CH=\overset{+}{N}\text{-}Me_2Cl^-$) and hence substitution.



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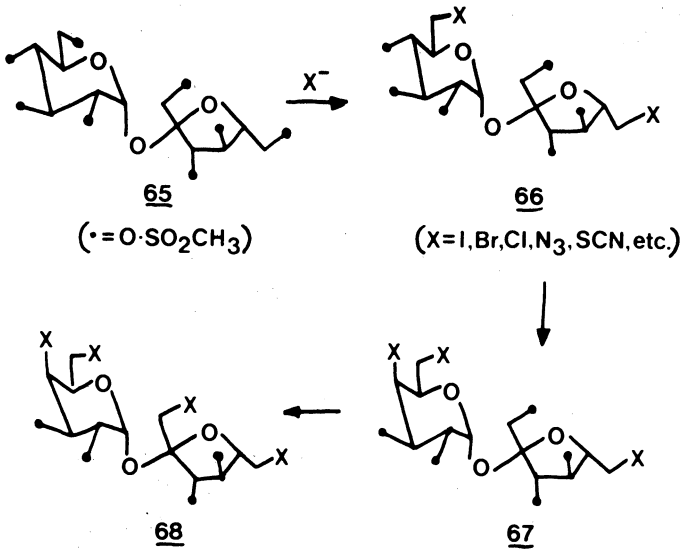
The 3,6,6'-trichloro derivative (57) prepared from methyl β-maltoside was converted by hydrogenolysis into the 3,6,6'-trideoxy derivative, a potential precursor of the amicetin type of nucleoside antibiotics. (Ref. 32). The 3,4',6,6'-tetrachloride (58) was also obtained in 48% yield by reaction of methyl β-maltoside with sulphuryl chloride in pyridine (Ref. 36).

The chemistry of sucrose (1), often termed "Sucrochemistry", has played a special role in our studies in view of its ubiquity and great importance in commerce. In the past, progress was slow in view of the complexities associated with the chemistry of this unique molecule, but many of the problems have now been overcome with the emergence of a profile of chemical reactivity. Substantial progress can now be expected with wider applications in sugar and associated technologies. In common with the aforementioned disaccharides, sucrose and its derivatives show stereoselectivity in many of their reactions, which is fortunate, since a large number of partially substituted derivatives are in theory possible. Thus, whilst only one octa-derivative of sucrose can exist, such as the octamesylate (65), there are 70 possible tetra-derivatives and 56 alternatives in the case of both tri- and penta- derivatives. Our studies (Ref. 37 and 38) have shown that of the eight hydroxyls of sucrose four can be replaced in a selective manner, and pure 6,6'-di-,4,6,6'-tri- and

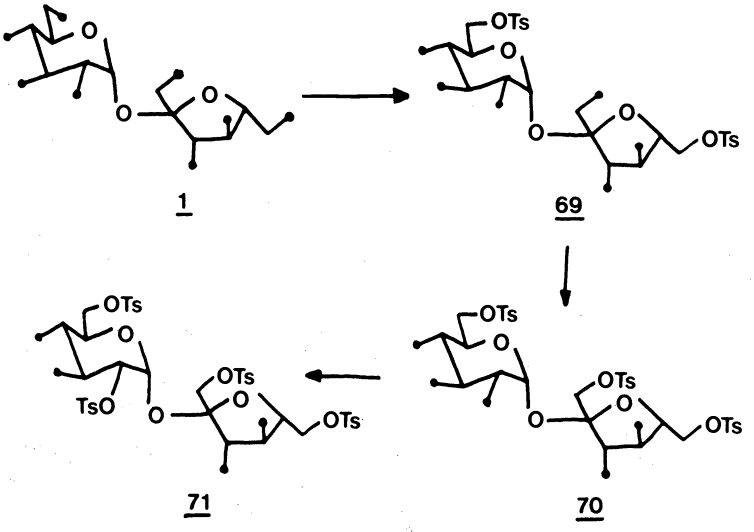
1',4,6,6'-tetra-substituted products isolated, dependent upon the reactants and the reaction conditions. For example, when sucrose octamesylate (65) is treated with nucleophiles such as iodide, bromide, chloride, azide, etc. in an aprotic solvent such as D.M.F. or hexamethylphosphoric triamide (H.M.P.T.), certain mesyloxy substituents undergo nucleophilic substitution to give products ranging from 6 and 6'-mono-derivatives to 1',4,6,6'-tetra-derivatives (68). It was established that the order of replacement is 6~6'> 4> 1'>>



all other positions and that 6,6'-di-substituted (66) and 4,6,6'-tri-substituted (67) products could be readily isolated by careful control of the reaction conditions.

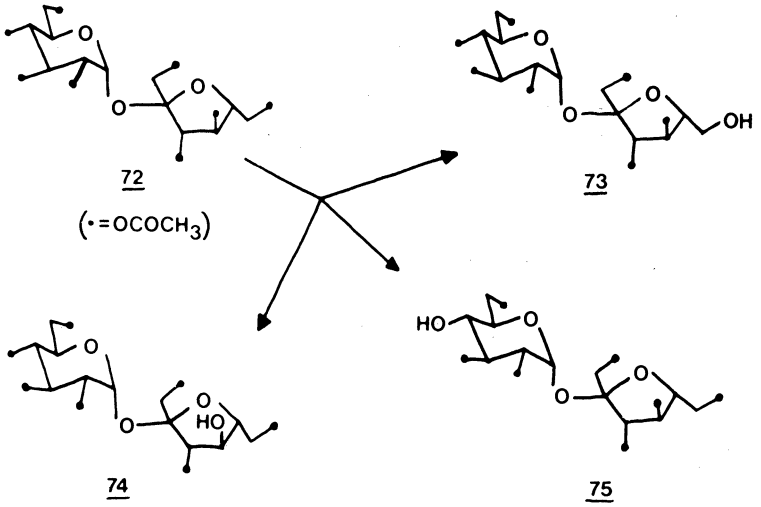
The observed stereo-selectivity (Ref. 37 and 38) is in accord with the bimolecular transition state theory (Ref. 39), the substituents at the least hindered 6- and 6'- primary positions reacting fastest and the substituent at the crowded neo-pentyl type, 1'-primary position reacting slower than that at the more favoured, but secondary, 4-position. In accord with this mechanism, inversion of configuration is observed when substitution occurs at a chiral centre, in this case at C-4. By this device, 4-sulphonate esters of sucrose are converted into β-D-fructofuranosyl α-D-galactopyranoside ('galacto-sucrose') and its derivatives (67 and 68) (Ref. 40 and 41). It is of interest to note that galactosucrose is not sweet (Ref. 42), due to hydrogen bonding by the axial 4-hydroxyl group to the ring-oxygen (see later).

By selective esterification, partially substituted sulphonate esters of sucrose can be prepared by using limited quantities of toluene-p-sulphonyl ('tosyl') chloride in pyridine and subsequent purification by column chromatography. This approach has afforded the 6,6'-ditosylate (69) (Ref. 37), 1',6,6'-tritosylate (70) (Ref. 43) and 1',2,6,6'-tetra-tosylate (71) (Ref. 44). Advantage has been taken of the increased selectivity of bulkier sulphonyl halides (Ref. 45), such as triisopropylbenzene sulphonyl chloride (tripsyl



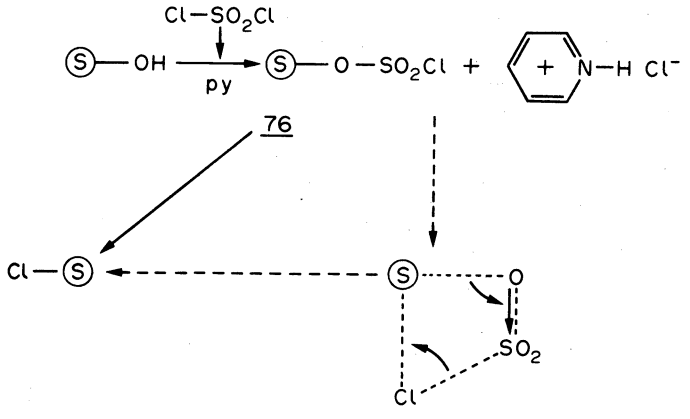
chloride) and mesitylene sulphonyl chloride, to isolate the corresponding crystalline 1',6,6'-trisulphonate esters directly in good yield without the use of column chromatography (Refs. 46 and 47). Nucleophilic substitution of the sulphonyloxy substituents in these partially substituted esters readily yields 6,6'-di- and 1',6,6'-tri-substituted sucrose derivatives, such as the corresponding chlorides and azides, and hence amines.

An alternative approach was to examine the selective de-esterification of sucrose octa-acetate (72) in chloroform with alumina, when three hepta-acetates were obtained with free hydroxyls at C-6', C-4' and C-4 respectively (73, 74 and 75) (Ref. 48). Since acetyl groups readily migrate (via a 4,6-ortho-ester intermediate) from the 4- to the 6- position of the D-gluco-

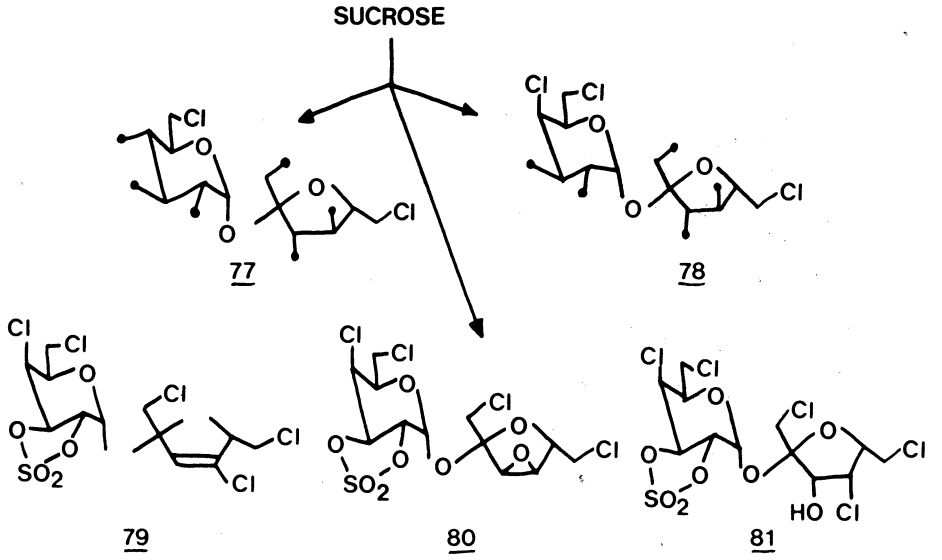


pyranosyl ring, and less readily from 4' and 6', we (48) suggested, with supporting evidence that the initial de-acetylation products were the two heptaacetates, one with the 6-hydroxyl and the other with the 6'-hydroxyl free, the final products resulting from acetyl migration.

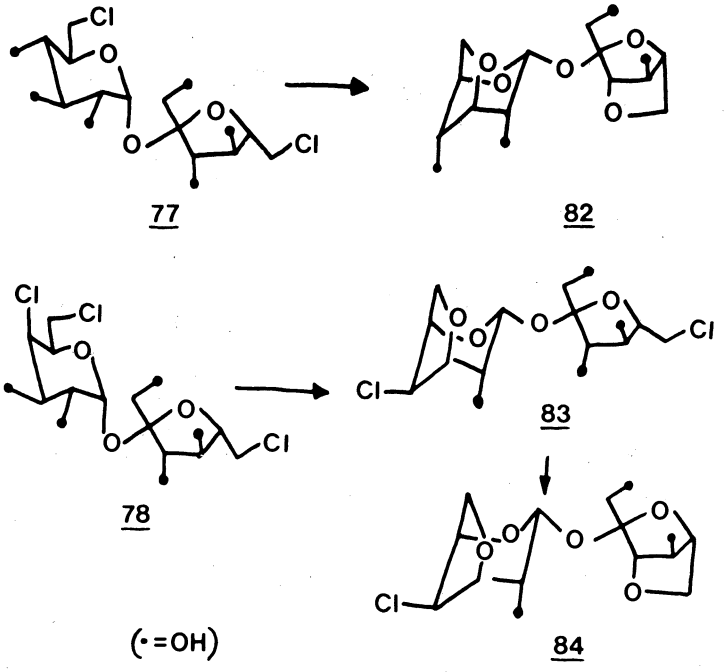
The replacement of selected hydroxyls in sucrose by chloride is conveniently achieved by reaction with either sulphuryl chloride in pyridine (Ref. 49) or mesyl chloride - D.M.F. (Ref. 50 and 51). The former reaction involves the initial formation of a chlorosulphate ester (76) which can then undergo an intramolecular nucleophilic substitution with the introduction of chloride. By careful control of the reaction conditions of sucrose with sulphuryl



chloride in pyridine, 6,6'-dichloro-6,6'-dideoxy-sucrose (77) and 4,6,6'-trichloro-4, 6,6'-trideoxy-'galacto'sucrose (78) can be obtained directly, after de-chlorosulphonation, without recourse to preparative chromatography (Ref. 52). Again the least crowded, primary 6- and 6'- positions react preferentially and the favourable secondary 4-position is more reactive than the more hindered primary 1'-position. Parolis (Ref. 53) has also isolated the trichloro-sucrose (78) directly but as the crystalline 1',2,3,3',4'-penta-chlorosulphate. Under more forcing conditions, extensive reaction of sucrose occurs with sulphuryl chloride to give three 4,6-dichloro-4,6-dideoxy-α-D-glucopyranosyl 2,3-sulphate derivatives (79, 80 and 81) each differing in the modifications to the fructofuranosyl ring (Ref. 54). The chloro-

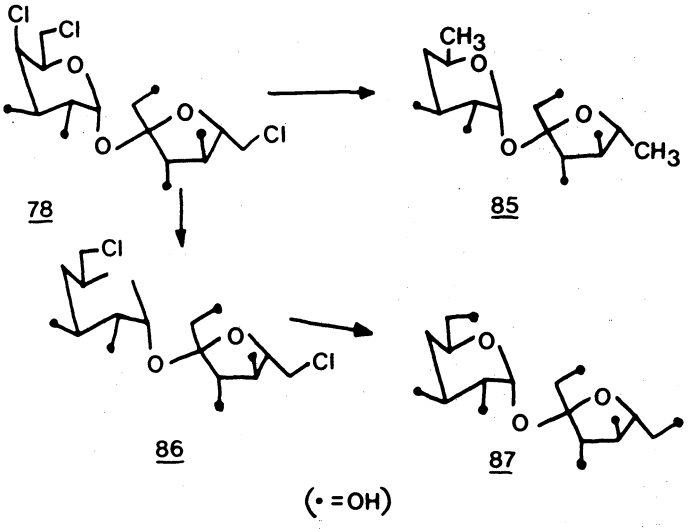


sucroses 77 and 78 are readily converted into anhydrides (Ref. 52) and deoxy-derivatives (Ref. 55). Thus the 6,6--dichloride (77) was smoothly converted into the 3,6;3',6'-dianhydro derivative (82) when treated with methanolic sodium methoxide, whereas, by contrast, the 4,6,6'-trichloride (78) gave initially the 3,6-anhydro-4,6'-dichloride (83), due to rate-enhancement by the axial 4-chloro group moving to the more favourable equatorial position ($^4C_1 \rightarrow {}^1C_4$), followed by the accumulation of the final product, the 4-chloride of the 3,6;3',6'-dianhydro derivative (84) (Ref. 52).

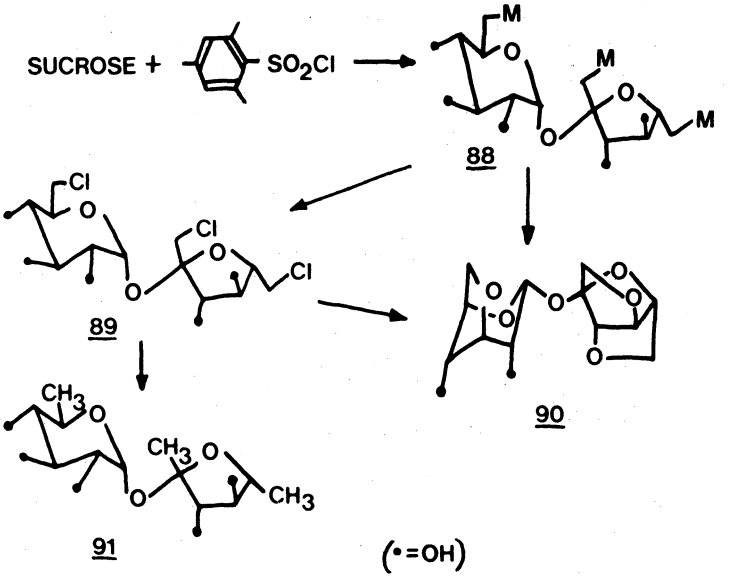


The 4,6,6'-trichloride (78) was converted into 4,6,6'-trideoxy-sucrose (85) by catalytic, reductive dehalogenation in the presence of potassium hydroxide (Ref. 55) whereas, as anticipated from the results of Lawton, Wood, Szarek and Jones (Ref. 56), in the presence of triethylamine, dechlorination occurred exclusively at the 4-position giving the 4-deoxy-6,6'-dichloride (86). Nucleophilic replacement of the chloro-substituents of the latter (86) by benzoate then gave 4-deoxy-sucrose (87) (Ref. 55). 1',6,6'-Trichloro-1',6,6'-trideoxy-sucrose (89) was conveniently synthesised from the 1',6,6'-trimesitylenesulphonyl-sucrose (88), and then converted as above into 1',4'; 3,6; 3',6'-trianhydro-sucrose (90) and 1',6,6'-tri-

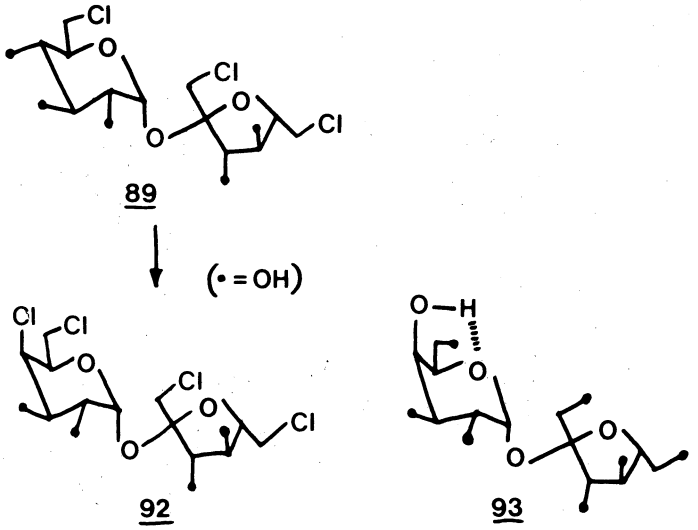
deoxy-sucrose (91) (Ref. 47). Reaction of the 1',6,6'-trichloride (89) with sulphuryl



chloride in pyridine afforded, as predicted, 1',4,6,6'-tetrachloro-1',4,6,6'-tetradeoxy-galacto-sucrose (92). One of the objectives in our chemical studies on sucrose has been to enhance its natural sweetness. We have been greatly encouraged by the surprising discovery



that this tetra-chloride (92) is intensely sweet, comparable to saccharin but with a pleasant after-taste (Ref. 57). The apparently contrary loss of sweetness in galacto-sucrose (93) has been attributed to the axial 4-hydroxyl group being masked from the taste buds by an intra-molecular hydrogen bond to the ring-oxygen (Ref. 42), which cannot of course occur in the sweet 1',4,6,6'-tetrachloride analogue (92). The considerable enhancement of the sweetness of sucrose by derivatisation, not observed hitherto in this disaccharide nor in any other carbohydrate, is clearly of importance not only in dietetics in the development of alternative sweeteners to sucrose but in relation to theories of sweetness.



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