THE DISTRIBUTION OF CARBOHYDRATES AND THEIR

ASSOCIATIONS WITH METAL IONS

IN SELECTED GLEYSOLS

by

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ABSTRACT

Three Gleysols from the Lower Fraser Valley were selected to study the distribution of hexoses: and pentoses and their association with iron, aluminum, magnesium and calcium. A sequential extraction procedure was developed. The various stages of the extraction procedure displayed unique patterns of hexose and pentose distribution. The hexose to pentose distribution ratios showed a greater accumulation of hexoses than pentoses in the upper A horizons but the reverse was observed in the lower horizons. Consistent hexose to pentose ratios of less than 1.0 for the dilute acid extraction were observed. Generally, pentose retention relative to hexoses increased with depth.

Comparative distributions of sugars with the metal ions studied were as follows:

- a) Water extraction indicated that hexoses and calcium formed close associations in the Ap and Ah horizons and pentoses with iron and aluminum formed close associations in the lower horizons.
- b) The dilute acid extraction indicated that pentoses were associated with aluminum and to a lesser extent with iron throughout the profile.
- c) The combined pyrophosphate and potassium

phosphate extraction indicated that iron and aluminum were closely related with pentoses or hexoses in the Ap and Ah horizons whereas pentoses appeared to form close relationships with iron, magnesium and aluminum in the lower horizons.

- d) The pyrophosphate extraction indicated as relationship between hexoses, iron and aluminum in the upper Ap and Ah horizons while pentoses, magnesium and aluminum appeared to be associated in the lower horizons.
- e) The potassium sulphate extraction indicated that pentoses or hexoses were closely related to aluminum in the Ap and Ah horizons but pentoses appeared to be associated with magnesium and aluminum in the lower horizons.
- f) The Chelex-100 extraction indicated a relationship of hexoses with aluminum throughout the profile.

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INTRODUCTION

Carbohydrates, and in particular polysaccharides, play an important role in soils by influencing
their physical conditions, affecting cation exchange
capacity, the retention of anions, complexing of metals,
and generally affecting the mineralization of associated
elements by acting as an energy source for heterotrophic
organisms.

Although the main stimulus for investigation of carbohydrates has originated from their influence on the soil physical condition, the tendency has been to study this group of organic substances mainly from an analytical point of view. It is the purpose of this study to:

- (1) develop a method of extraction which selectively and progressively extracts organic matter and associated cations.
- (2) describe the soil as a whole entity by way of distribution of the constituents.
- (3) demonstrate differentiation of profile characteristics by way of different carbohydrate and ionic association trends.

In order to achieve this a sequential procedure of extraction was utilized.

On survey of the literature, it was felt that no reference was made to the influence of ions and other organic constituents on the present carbohydrate

status of the soils. It therefore became apparent that a holistic approach was a more realistic one in achieving the goals of study on carbohydrates. To pay attention to a group of compounds without acknowledgement of associated constituents is to ignore relevant material in the continuum of soils. However, due to the limited time available, the scope of the study was confined to a few associations of carbohydrates with metal ions. Gleysols were chosen for the study because very little information has been compiled on this group of soils, and because of the widespread distribution of poorly drained soils.

LITERATURE REVIEW

INTRODUCTION

Carbohydrates are by far the largest and not the least important food source of living organisms. They are photosynthesized by plants via the utilization of carbon dioxide and water through a transfer of energy from sunlight. A simplified expression of this reaction is:

 $nCO_2 + nH_2O \rightarrow (HCHO)_n + nO_2$

On incorporation of carbohydrate substances into the soil, they become an integral part of the soil organic matter, forming complexes with various materials. original forms of the carbohydrates are rapidly changed on incorporation and many new polysaccharides are resynthesized by soil micro-organisms. Therefore, these polysaccharides are quite different from those of plants and animal material and should be studied in this light. Although much is known about the nature and function of many polysaccharides synthesized by individual organisms, very little is known about the polysaccharides that are synthesized in the soil environment, which by itself is quite unique and distinct in that it integrates a great variety of biological forms. It is indeed, very surprising that there is such neglect, because soils not only support the majority of higher plants, but also they are the chief habitat for micro-organisms. The quantities of carbohydrate substances added to soils

as plant residues and those synthesized by the indigenous micro-organisms are enormous.

Shorey and Lathrop (59), first reported the presence of sugar in soil. The evolution of furfural on heating the soil in hydrochloric acid was regarded as evidence for the presence of pentosans, amounts of furfural being considered proportional to the amounts of pentosan or pentose. Subsequently, several reports were made of isolation of various carbohydrates from soils.

Carbohydrates constitute 5 to 20% of the soil organic matter (23, 27, 71, 78). Because a large proportion of these carbohydrates can only be recovered after harsh chemical treatment, their isolation intact is extremely difficult. Clearly, the great complexity which does exist in the soil causes soil scientists to query the forms in which they occur, their amounts, modes of isolation and characterization. To date, only a few of the answers are known. With the use of more advanced chemical, physical, physico-chemical and biological techniques, a lot more questions may eventually be answered.

CLASSIFICATION AND COMPOSITION

1. CLASSIFICATION: (23)

Even though a variety of organisms contributes to the content of carbohydrates in soils, only a limited

number of them have been positively identified. Continually, the chemical and biological processes of degradation and synthesis alter their structures. Furthermore, destruction and modification during hydrolysis and extraction are liable to occur. A small amount of monosaccharides have been found in the free state while the rest have been found in the bound state.

The following carbohydrates have been isolated from soils: (23)

Monosaccharides:

Hexoses (glucose, galactose, mannose, fructose)

Pentoses (arabinose, xylose, ribose, fucose, rhamnose)

Disaccharides (sucrose, cellobiose, gentiobiose)

Oligosaccharides (cellotriose)

Polysaccharides (cellulose, hemicellulose)

Amino Sugars (glucosamine, galactosamine,

N-acetyl-D-glucosamine)

Sugar Alcohols (mannitol, inositol)

Sugar Acids (galacturonic, glucuronic)

Methylated sugars (2-0-methyl-D-xylose,

2-0-methyl-D-arabinose, 2-0-methyl

rhamnose, 4-0-methyl galactose)

2. COMPOSITION:

The presence of such a wide variety of sugars

makes it difficult to measure the total soil carbo-hydrates. The instability of some of the monomers under harsh conditions (strong acid - 72% H₂SO₄) only aggravates an already crucial situation. There is bound to be some destruction during hydrolysis. Mild methods however, only recover up to 30% of the total soil carbohydrates.

Free monosaccharides constitute less than 1% of the soil carbohydrates, and extracted polysaccharides have rarely accounted for more than 20%. Approximately another 10% may consist of cellulose (28). Swincer et al. (71), have reported an extraction method that enables almost complete extraction of carbohydrates from soil. They contend that material removed by their vigorous extraction procedure is similar to that of materials removed by less efficient methods.

In some mineral and organic soils, glucose constitutes a major percentage (42 - 54%) of the total sugars in all the horizons. Usually, the other sugars formed in decreasing order are galactose, mannose, arabinose, xylose, rhamnose, fucose and ribose (23). A similar range of sugars has been found by Gupta, Sowden and Stobbe (27), showing a dominance of galactose, glucose and mannose in a podzol, a chernozem and a gleysol. The data is presented in Table 1. Glucose constitutes one-third of the carbohydrates in mineral horizons and one-half in forest litters which have not

TABLE I

DISTRIBUTION OF SUGARS IN HYDROLYSATE OF ORGANIC MATTER

(Gupta, Sowden and Stobbe, 1963)

	Podzol		Chernozemic		Gleysolic	
	Ao	В	Ah	В	Ap	
	% of total sugars					
Galactose	15	16	14	15	15	
Glucose	54	35	36	34	31	
Mannose	15	16	. 16	14	15	
Arabinose	5	9	15	12	15	
Xylose	4	9	8	8	11	
Fucose-Ribose	3	6	3	5	5	
Rhamnose	4	9	8	12	8	
Total sugar mg/g OM	161	80	95	68	156	
OM mg/g soil	. 730	140	62	20	24	

undergone extensive degradation. The relative concentration of the sugar monomers mannose, arabinose, xylose, fucose, ribose and rhamnose and the presence of 2-0-methyl rhamnose and 4-0-methyl galactose in soil indicates their microbial origin (17, 82).

The total sugar content generally decreases rapidly with depth in soils. However, the reverse was observed in a podzol with permafrost (fig. 1, p. 106 - Gupta (23)), and was attributed to churning of organic matter by frost action. Also, in a solonetzic soil profile, the amount of carbohydrate carbon was higher in the B than in the A horizon (31). These differences in the amount of total carbohydrates among various soils can be attributed to differences in vegetation, degree of organic accumulation in different horizons, microflora, and analytical artifacts.

Free carbohydrates are present only in small quantities in soils and tend to be in a steady state which depends on factors such as moisture, temperature, reaction (pH), texture, organic matter content, microflora, and vegetation in and on the soil.

Soil cellulose may contain 65% - 82% glucose and thus the material is primarily a glucose polymer (33). In contrast, "hemicelluloses", the group of polysaccharides found in cell walls of plants in association with lignin as an amorphous phase enveloping the cellulose strands, consists of varying amounts of D-xylose,

L-arabinose, D-glucuronic acid, 4-0-methyl-D-glucuronic acid, D-and-L-galactose D-mannose, L-rhamnose, and L-fucose (79). There are as yet no satisfactory methods available for separation and purification of these materials.

Easily hydrolysed carbohydrates, referred to in soil science literature as "polysaccharides", extracted by mild treatment involving hot water, dilute acids, alkalis and buffers, constitute a large percentage of plant material, but only a small percentage of soil organic matter. They are frequently insoluble in water and non-reducing and are made up of monosaccharide units linked together. At least two -OH groups in each sugar unit are used for linkage to other sugars. With end units only one -OH is involved in bonding. The polymers are usually produced from bacteria, both as capsular and extracellular polysaccharides (79). It is very difficult to isolate individual polysaccharides because of the continuous process of synthesis and degradation of polysaccharides that occurs in soils, thereby resulting in a considerable variation in the amount of polymers of differing sizes and aomposition.

It appears that soils rich in organic matter contain large amounts of polysaccharides. The polysaccharides isolated from the upper mineral horizon beneath mor have been found to be smaller in quantity and of lower molecular weight than the polysaccharides isolated

from a mull (6). The presence of small amounts of nitrogen and ribose suggest the possibility that soil polysaccharides are of microbial origin (21). According to Keefer and Mortensen (35), soil polysaccharides undergo continuous degradation and resynthesis and are products of microbial activity. Furthermore, that they are not derived directly from higher plants is substantiated by the low proportion of D-xylose and D-glucose (82). The presence of mannose, rhamnose and hexosamine also indicates their microbial origin.

Uronic acids, chiefly D-glucuronic, D-galacturonic and D-mannuronic acids, which are widely distributed in the plant kingdom, probably result from the oxidation of the primary alcoholic group to a carboxyl group.

Their presence in soils was first reported by Shorey and Martin (61). The uronic acid values reported by workers for mineral and organic soils ranged from 0 - 12 to 8 - 47 mg. per gram of organic matter respectively.

The occurrence of glucosamine and galactosamine (8, 9, 26, 62, 65, 66) has been reported and one instance of N-acetyl-D glucosamine (64), has been reported. The former two amino sugars account for nearly all of the hexosamine in soil hydrolysates.

Glucosamine (chitosamine) is found in chitin

(a skeletal polysaccharide of crustacea and fungi) and

in mucoproteins and mucopolysaccharides and the latter

in chondroitin sulphuric acid of cartilage and tendons.

Amino sugar nitrogen constitutes from 1 to 11% (8, 26, 65, 68), of the total nitrogen present in soils. Soil type (62, 63), depth in soil profile (62, 67), and different crop rotations (26, 66), are some of the factors that affect the total quantity of amino sugars.

The origin of glucosamine in soils is uncertain although microbial origin is indicated (77). It is known that mucopolysaccharides of many micro-organisms contain galactosamine. Glucosamine could also originate from chitin.

THE SOURCE, SYNTHESIS, TRANSFORMATION AND DEGRADATION OF SOIL CARBOHYDRATES

1. SOURCE AND SYNTHESIS:

Undoubtedly, the major portion of soil carbohydrates is derived primarily from plant materials,
while a significant portion also originates from animal
residues and from microbial residues. Vegetation contributes carbohydrates in the form of mono-, oligo-,
and polysaccharides (chiefly cellulose and hemicellulose).
The structural classes represented in soil organic matter
include glucosides, neutral polysaccharides and acid
sugars (23). However, it is quite obvious that very few
unchanged plant polysaccharides occur in soil at any
specific time. On addition, plant materials are rapidly
decomposed by the soil microbes (12, 24, 69). Even
cellulose, which is a chemically resistant polymer, is

quite rapidly degraded by the complex of endo- and exoenzymes possessed by soil fungi, bacteria and actino-mycetes (2). Other plant polysaccharides such as starch, hemicellulose, pectic substances, gums and mucilages, are less stable, suggesting that most polysaccharides found in soils are of microbial origin. Mucilaginous secretions by the root caps of many plants are a possible exception.

Some evidence that polysaccharides isolated from soils are of microbial origin has been given in the previous section. However, the strongest and most direct evidence has been presented by Mortensen and Keefer (35), using glucose and alfalfa tissue substrates labelled with radioactive carbon. After incubation, they examined the distribution of radioactivity among different sugars in a polysaccharide extracted from the soil. All sugars isolated were found to be labelled, even though there was considerable variation in specific activity. The changes in the specific activity of the different sugars with time indicated that not only soil polysaccharides, but also their component sugars undergo continual degradation and resynthesis. It therefore follows that non-carbohydrate materials may sometimes act as precursors for soil polysaccharides. The sugars examined were glucose, arabinose, galactose, fucose, mannose, rhamnose, xylose and uronic acids.

Mehta et al. (49), claim that soil bacteria are

capable of producing polysaccharides containing all the sugars found in soils except arabinose and galactose. The latter are probably derived from other sources. Very little is known about the polysaccharides produced by most of the soil bacteria (39, 53). Consequently, it is difficult to establish the origin of some of the carbohydrates containing nucleic and uronic acids.

Animals contribute a small portion of the soil carbohydrates in the form of glycogen, snail galactan, nucleic acids, chitin (52), and the polysaccharides containing nitrogen and sulphur. The latter ones are of immense importance to animals and consist of hyaluronic acid (mucoitin), chondroitin sulfate, heparin, and heparin sulphate.

Studies with pur cultures are not likely to throw much light on the situation in the soil itself. The complexity of the natural soil environment ensures that at least the exocellular polysaccharides produced within it differ considerably from those obtained in the laboratory from either pure or mixed cultures even with the use of the same carbon substrates.

Tissues of recently dead microbial cells, particularly the cell walls, probably contribute to many of the polysaccharides that persist in the soil (53, 82). It has been established that hexosamines and other sugars are important constituents of the polymers which make up the cell walls of bacteria (53), and actino-

mycetes (64), while chitin, a polysaccharide composed at least predominantly of N-acetyl-glucosamine residues (22), is the main constituent of fungal cell walls (64).

The composition of available plant substrates influences the kind of polysaccharides synthesized in the soils, but the nature of this effect is not understood. The kind of crop growing on the soil would be expected to influence the microbes in the rhizosphere.

2. TRANSFORMATIONS AND DEGRADATIONS:

According to Reese (53), factors affecting the transformations of soil polysaccharides are the kind of sugar(s), modification of the sugar moiety, linkage types, extent of branching and the orientation of chains which influence the amorphous to crystalline superstructure. Soil polysaccharides, he claims, are essentially mixtures of the components of the organisms present in the soil so analysis of these organisms will greatly simplify and promote the identification of the soil polysaccharides. It is obvious though, that the situation is not as simple as this.

Fungal walls consist of thin chitinous (or less often, cellulosic) substance variably thickened with p-glucans, d-mannans, and heterogeneous glycans. Bacteria contain various modifications of the insoluble chitinous wall (eg. muramic acid) and in addition, capsular material outside of the wall. The rate of

decomposition is greatest for the simple polymers of the common sugars, the most rapid rates being those of glucans, xylans, and mannans of higher plants. As the polymers increase in complexity, they become more resistant to enzymic hydrolysis. Consequently, the residual soil polysaccharide is mainly derived from more complex cell walls of micro-organisms. Materials isolated by Whistler and Kirby (82), were found to be more resistant to decomposition than many plant polysaccharides. Also, in pure culture, certain soil micro-organisms (eg. Chromobacterium violaceun and Azotobacter indicus) produced polysaccharides which were not rapidly attacked when added to soils (41, 42, 44, 45). However, the polysaccharides produced by many other soil micro-organisms suffer extremely rapid degradation, up to 75% of their carbon being converted to CO2 after incubation for four weeks (44, 45). contrast, only about 10% of the carbon of the polysaccharide from Azotobacter indicus was lost in the same time. Roughly, the order of increasing stability of plant carbohydrates is the following: reducing sugars, non-reducing carbohydrates, pectin, hemicelluloses and celluloses.

At the end of a growing season, a considerable amount of higher plant polysaccharide returns to the soil. It is rapidly converted to microbial tissue, the efficiency of conversion being very low, and the total

organic matter in the soil drops quickly. involves the action of both bacteria and fungi fixing themselves to the substrate or penetrating it. addition, some exo-enzymes may diffuse to the substrate. The strong adsorptive action of clay would tend to minimize the amount of enzymes diffusing to any great distance away from their original site of production. Some enzymes are constitutive, i.e. produced regardless of the substrate being consumed, while others are inductive, <u>i.e.</u> induced by the presence of the substrate. Complex polysaccharide digestion proceeds in both ways, but the ability to produce the required enzymes is limited to a relatively few organisms. These, then, become dominant members of the microflora. Often dead cells may be digested by their own intracellular enzymes. Such autolysis has not been evaluated, yet it suggests that many organisms which have the ability to synthesize a polysaccharide also produce the enzymes for its digestion.

An initial fragmentation of the insoluble substrate may be a general prerequisite to the action of hydrolytic enzymes. In complex systems composed of several structural components, the removal of an encrusting substance or of side groups from a polymer chain may be required.

Several enzymes, including amylase, cellulase, hemicellulase, polygalacturanase, and invertase have

been found to play a role in the transformation of various carbohydrates (49). Phytase (32), xylanase (61), and glucose oxidase also have been detected and these too may play a role in the transformation and synthesis of carbohydrates. A look at the types of enzymes shows basically what changes are brought about. The endo-polysaccharases (activity in random fashion) produce a wide variety of products. The rate of hydrolysis by these enzymes increases with degree of polymerization. In contrast, the exopolysaccharases, removing one monomer or dimer at a time from the nonreducing end of the polymer chain are consequently slower acting. These are important in the degradation of polymers only after the endo-enzymes have brought about a considerable increase in the number of chains. On these short chains, their action decreases rapidly from tetramer to trimer to dimer.

Finally, the glycosidases represent the terminal enzymes in the process of hydrolyzing polysaccharides to simple sugars. The type of glycosidase involved depends on the nature of the aglycone, that is, whether it is another sugar unit or alkyl or arylunit.

Undoubtedly, some changes can take place without the intervention of micro-organisms but humification
can only reach completion via their mediation. It
appears that the final stage of humus formation pro-

ceeds mainly by physico-chemical processes, without the participation of micro-organisms (34). It appears that carbohydrates can ultimately reappear as aromatic substances via the shikimic acid pathway. It is evident that carbohydrates decomposed earlier are sources of structural units in molecules of humus substances (amino acids, proteins and polyphenolic substances) through diverse transformations during metabolism and resynthesis by micro-organisms.

The carbohydrate by-products of microbial reactions can be protected from degradation, by four mechanisms:

(i) Living Tissue:

Living tissue is essentially resistant to degradation. Large amounts of carbohydrates are contained in this biomass and thereby protected from degradation. After death this material becomes relatively less resistant to attack.

(ii) Adsorption:

Resistance to breakdown may be enhanced by adsorption to clays (38, 57). Lynch and Cotnoir (18, 38), found that adsorption to clays decreases rate of breakdown.

Adsorption of ¹⁴C - labelled polysaccharide by kaolinite saturated with different exchange cations was varied with the type of cation present (57). Their effects decreased in order Fe³⁺, Al³⁺, H⁺, Ca²⁺, Mg²⁺, Na⁺. Hence, exchangeable cations will influence the degree of decomposition. Also reactions with metals (e.g. copper) (43), confer resistance on plant gums and capsular polysaccharides of soil bacteria, to microbial degradation.

(iii) Condensation:

Processes similar to those described for browning reactions in food products, involving the condensation of carboxyl groups with amino-derivatives, may also explain the stability of some carbohydrate materials in soil (34).

(iv) Inaccessibility:

Some polysaccharides are probably shielded from attack by being present in parts of the soil that are inaccessible to soil organisms, e.g. interlamellar spaces (55).

The influence of soil type and such environmental variables as soil temperature, moisture content, aeration, pH, nutrient status and agronomic treatment, upon decomposition and synthesis of soil carbohydrates is poorly understood (73). These factors probably operate in much the same way as they do with organic materials. Highly aerobic conditions favour rapid metabolic activity and the conversion of the large part of the organic materials to ${\rm CO_2}$, whereas under anaerobic conditions, decomposition is much slower.

It has been demonstrated (30), that aggregatestabilizing materials (presumably carbohydrates) are
synthesized in sucrose-amended soil under both aerobic
and anaerobic conditions but deteriorate rapidly with
oxygen, and while oxygen is excluded aggregation persists. Decomposition would also be facilitated by
cultivation since soil aeration is improved. This
decomposition may involve increased production of
relevant enzymes as a result of general stimulation
of the microbial population, and the exposure of previously inaccessible polysaccharides (55).

The metabolic rate of the microbial population as a whole increases with temperature up to about 37° C. It has been suggested (54), that the microbial population at low temperatures produces more aggregating substances (probably polysaccharides) whereas high temperatures favour rapid decomposition of the soil binding agents. In spite of the various protective mechanisms, most of the polysaccharides end up as CO₂, and the high level of carbohydrate present in soils can largely be attributed to an adequate supply of plant

materials. Consequently, the timing of addition has some bearing on the composition and the quantity of the resultant polysaccharides.

In spite of the various transformations that take place, the indication is that the differences in the polysaccharides from different Great Soil Groups in their monosaccharide composition are very small (37). The paucity of studies in this area prohibits further conclusive generalization.

THE SIGNIFICANCE OF SOIL CARBOHYDRATES.

Undoubtedly, the study of soil carbohydrates has been mainly stimulated by the indications of their influence on the physical conditions of the soil. All the roles they play in the soil include their effect on the cation exchange capacity (due to the uronic acid units), the retention of anions (due to amino groups, but only in acidic soils), and generally, carbon metabolism, thereby affecting mineralization of associated elements involving stimulation of biological activity (for example, by acting as an energy source for heterotrophs) and finally complexing of metals (43).

Several reports (40, 46, 70), indicated that microbially produced gums could bind soil particles into stable aggregates and these reports have attracted the attention of many workers. Recently, several workers (14, 30, 44, 45), have reportedly confirmed

these observations. Further, it is believed (6, 18, 20, 40, 41, 81), that the presence in the soil of organisms that produce aggregate stabilizing gums when cultivated in the laboratory, is enough to suggest that these gums will also occur in soils and accordingly should stabilize natural soil aggregates. These microbial gums have been extracted from a wide variety of soils and it has been deomonstrated that the extracted polysaccharides are able to stabilize soil aggregates (48, 54, 82).

Although the statistically significant correlations were not very close, since methods employed were crude estimates of the polysaccharide and aggregate stabilization, it has been demonstrated that correlations do exist between the polysaccharide content and the degree of aggregation (1, 54, 79, 80). Griffith's (24), review criticized the correlations on the grounds that since methods for the quantitative estimation of microbial polysaccharides in soils have not been developed, it has not been possible to evaluate accurately the contribution of these materials to aggregation. Further, it has been pointed out (71), that probably only certain of the polysaccharides present in soils are responsible for aggregate stabilization. Oades et al. (51), having developed and used quantitative methods of high precision, found that the correlation of aggregate stability with both composition and total amount of neutral sugar constituents was no better than with other organic materials. Others also have found bacterially produced gums differing considerably in their effectiveness (14, 44, 45), and that some plant products such as cellulose exert no influence on aggregate stability (25).

a soil under old pasture was similar to the composition of polysaccharides in the same soil type which had been under a wheat fallow for 40 years, there were differences between the treatments with respect to the amounts of carbohydrates present in the soil and ease with which they were extracted (72). Therefore, the distribution of the polysaccharides might be important since only a portion of a polymer may be effective.

Mehta et al. (48), postulated that if the polysaccharides are the essential cements in the aggregates, periodate or acid hydrolysis should dis-rupt their molecules and thereby decrease aggregation. The periodate oxidises sugars containing cis-glycol groups, then the partly oxidised polymers are readily degraded in alkaline solution into various non-polymeric fragments. Polymers cleaved in this way can no longer act as bridges between the soil particles forming an aggregate (73).

On addition of dextrans or soil polysaccharides to the dispersed soil, the artificial aggregates formed

lost their stability when treated with dilute (0.01 M) sodium periodate and sodium borate (pH 9.6) (48). However, on examination of natural soil aggregates, (Mehta et al. (43)), the treatment was found to be ineffective and concluded that other stabilizing agents were involved. In soils of lower organic matter content, periodate treatment has effected a marked decrease in aggregate stability (23). Also, others have shown that soils incubated with additional organic matter showed increased aggregate stability which was largely associated with periodate-sensitive materials (30, 79).

eability of beds of aggregates could be reduced by periodate treatment. Williams et al. (83) showed that the principal factor controlling penetration of polyvinyl alcohol (PVA) molecules is probably the size of the soil pores in relation to the size of the polymers. PVA adsorption along the pore walls is essentially an irreversible process. The adsorbed molecules hinder or prevent the movement of other molecules in solution. In a similar manner, naturally occurring organic matter, e.g. carbohydrates can reduce the extent of adsorption of other substances and vice versa. The water potential gradient was also an important factor. The degree of branching and cross-linking modifies the ease of adsorption.

Mucilages and gums can undesirably reduce the size of soil pores in fine textured soils (56). In such soils, microbial production of gums and mucilages in the few pores may narrow the pores to a point where their permeability is reduced appreciably (3). This could also occur in submerged soils whereby the important coarse pores may have their permeability reduced (3). Therefore, it can be concluded that polysaccharides do influence aggregate stability probably mostly in cultivated soils of relatively low organic matter content, but where other stabilizing agents exist, polysaccharides may be of little additional benefit or even hinder permeability.

Fulvic acid has frequently been shown to contain a polysaccharide component. Kononova (34), states that the fulvic acids, on complexing cations of the alkali and alkaline earth metals play an important part in the formation of soils, especially of the podzolic and chernozemic groups. It is not known to what extent the polysaccharide component is responsible for this effect.

Studies by McKeague (47), have shown that without organic matter, the soil has little capacity for reduction. High amounts of solubilized iron found in reduced soils with high organic matter content, were consistent with the commonly observed maximum bleaching of gleying soils just below the

horizon of organic accumulation. Presumably, reducing carbohydrates play an important part in light of the above discussion. Several sugars normally produced in the soil may inhibit the precipitation of phosphorus by iron and aluminum (2), and favour the leaching of these sesquioxides from the upper to the lower horizons os some soils.

The mineralization of carbohydrates containing phosphorus and nitrogen, supply inorganic phosphorus and nitrogen to the plant. In addition, energy and carbon for various fungi, bacteria and actinomycetes are supplied by carbohydrates. Undoubtedly, investigation of this category of soil organic matter is necessary to acquaint soil scientists with the processes of soil formation and the status of soils in general.

AND METHODS OF ANALYSIS:

Most competent reviews of these facets of carbohydrate studies have been published by Swincer et al. (73), and Gupta (23). To avoid redundancy, the entire spectrum is not presented here. Of substantial consequence though, is the approach of Swincer et al. (73), to an ideal extractant, which must meet the following criteria. The ideal extractant must:

a) be non-degradative.

- b) give a sufficiently complete extraction for materials to be representative of the total.
- c) be equally effective for all soils.
- d) extract selectively carbohydrate materials. They recognised that no extractant has been able to meet these criteria. The main aim of most studies has: been to isolate from the soil, a sample of relatively pure polysaccharide material for characterization. Accordingly, Swincer et al. (73), proceeded to a three stage chemical extraction procedure preceded by ultrasonic dispersion. This has been the first step from a naive approach to a more realistic one. The complexity of the types of organic substances and their chemical, physical and physico-chemical associations with other organic and inorganic material's, obviously necessitates gradual chemical extraction aided by mechanical dispersion. The exploratory procedures have ably demonstrated this and serve as a guideline for developing a more meaningful extraction procedure. In addition to extracting carbohydrate materials to completion, it is also necessary to identify and classify carbohydrates and other simultaneously extractable materials in order to interpret possible associations between these constituents. In developing an extraction procedure the following facets should be taken into account:

l. The ease of solubility in mild extractants.

This gives an indication of the ease of release of organic substances under mild conditions of chemical change. The ease of release is dependent on the amount of energy involved in binding the soil constituents.

2. The solubility of organic matter by different types of extractants and combinations of extractants.

This gives an indication of the mechanisms involved in binding and partially the ease of release of organic substances.

3. Gradual dispersion of soil aggregates.

The greater the degree of dispersion necessary to release a soil constituent, the more tightly it is bound and/or the more centrally it is located within an aggregate.

4. Sequence of extractants.

Extracting sequentially attempts to obtain distinct homogeneous extracts which permit clarification of binding mechanisms of different types involving different constituents. The larger the number of identifiably different extractants, the

greater the clarification. Both structural and energy relationships can be investi-

5. Identification of extractable materials.

Other simultaneously extractable materials must be recognized as possibly contributing to the process of aggregation.

6. Residue Composition.

All materials must be accounted for and those in the residue may exhibit specific and different properties from the others previously extracted. Also extraction of homogeneous constituents to completion is difficult.

7. Extraction yields.

The relative amounts of different extracted materials indicate the relative contributions to the aggregation process.

8. Polymer degradation.

Although polymer degradation is probably unavoidable, the extent of degradation should be minimized.

Extractants commonly found useful for isolation of polysaccharide from soils include water, aqueous buffers and complexing agents, dilute mineral acids and alkalis.

WATER:

This has been a popular extractant because of the simplicity of the extraction, relatively low simultaneous extraction of humic materials and the ease of subsequent purification (8, 75). Hot water extraction has proven more efficient but autohydrolysis occurs with temperatures above 70 degrees centigrade (73).

AQUEOUS BUFFERS AND COMPLEXING AGENTS:

Sodium pyrophosphate and sodium EDTA, have been used as complexing agents (5, 6), and as buffer solutions (6). These afford minimum alteration of the polysaccharides. Schweitzer's reagent (Cupra-ammonium hydroxide) is considered effective for extracting cellulose (28).

DILUTE MINERAL ACIDS AND CATION EXCHANGE RESINS:

Dilute mineral acids are far more effective for extracting carbohydrate substances than humic materials. Repeated treatments with strong solutions apparently hydrolyses bonds between polysaccharides and humic materials. Extractants used include dilute (0.1N) hydrochloric acid (7), hydrofluoric acid (71), amberlite IR - 120, cation exchange resin (5).

ALKALIS:

Dilute sodium hydroxide has not only been the classical extractant for soil organic matter but frequently has proven to be the most potent of all the extractants. Large amounts of humic materials have

always been extracted simultaneously with carbohydrates. Further, it is claimed by many workers that autooxidation or hydrolysis of polymers occurs with this alkaline treatment.

SEQUENTIAL EXTRACTION:

Swincer et al. (71), employed the following chemical extraction sequence after ultrasonic dispersion:

- 1) 1N HCl at 20° centigrade
- 2) 0.5N NaOH
- 3) Acetic anhydride with 2.5% conc. ${\rm H_2SO_4}$ at 60° centigrade
- 4) Final hydrolysis with 72% ${\rm H_2SO_4}$

A "light fraction" obtained by a densimetric flotation procedure (19), contained from 10 - 50% of the total soil carbohydrate. Of the remainder, 10 - 20% occurs in the acid extract, 30 - 50% in the sodium hydroxide extract, 20 - 30% in the acetic anhydride extract and 10 - 20% may be left in the final residue.

The sequential approach has the advantage of progressive removal of material which eliminates further recombination of each type of material extracted. This multiple approach also has the advantage of selectivity of material in a specific order which leads to better understanding of the binding processes involved for each fraction. There are limitations to this selectivity because of incompleteness of treatment

at any one stage being dependent on the bonds being broken. The specific compounds being removed at one stage may contain bonds which might escape attack by an extractant employed at a later stage. This results in the incompleteness of the later stage. The sequential approach also gives an almost complete extraction because of the diversity of extractants used.

MATERIALS AND METHODS

For the study of carbohydrates in poorly-drained soils, soil horizons were sampled in three soils representing three series in the gleysolic order at the beginning of the summer in mildly rainy conditions. The samples were air-dried and crushed to pass through a 2 mm. sieve. It is not known to what extent drying and crushing affect the distribution of carbohydrate fractions, although some influence on levels of water soluble constituents is likely. Drying is, however, necessary to minimize differences between samples arising only from differences in field moisture conditions at the time of sampling. A classification of the soils investigated along with the topographical features of the sampling sites are given in the table The horizon samples were used to study carbohydrate distribution according to the extraction procedure presented in a later section, and for characterization by standard methods indicated below.

Particle size distribution was determined by the hydrometer method, following destruction of the organic matter with hydrogen peroxide (15). Soil pH was measured in a 0.01M CaCl₂ suspension (ratio 1:2.5) using a Beckman Zeromatic pH meter. Total carbon content was determined with a Leco Induction Furnace and Carbon Analyzer (4), and total nitrogen by a

semi-micro-kjeldahl procedure (10). Cation exchange capacity and exchangeable cations were determined by the neutral 1.0N ammonium acetate procedure (13).

Hexose and pentose contents in soil extracts were determined as follows: aliquots of soil extracts were hydrolyzed by treatment in 1N ${\rm H_2SO}_{\Lambda}$ and autoclaving for one hour at 121° centigrade and 15 P.S.I. (16). The hydrolysate was then applied to a chromatographic column of Ion Retardation resin AG-11A8 (Bio-Rad, Richmond, California) previously washed with $0.25 \text{M NH}_{\Delta}\text{Cl}$ (pH 8.0) followed by distilled water. column was eluted with distilled water and the eluate collected for estimation of hexose and pentose contents. Hexose and pentose contents were estimated by the anthrone (11), and the aniline acetate (76), colorimetric procedures respectively. The amounts of hexoses and pentoses are determined as glucose and xylose equivalents respectively and are not absolute quantities.

Extracted cations were in all cases determined by atomic absorption spectroscopy using a Perkin-Elmer Model 303 spectrophotometer.

EXTRACTION OF CARBOHYDRATE FRACTIONS

An extraction procedure was developed, based on a series of extractrants of increasing solubilizing strength, accompanied by progressively greater dispersion of soil aggregates, that would yield a series

of polysaccharide fractions with concurrently released cations. Details of the procedure employed are given below, followed by a discussion of the rationale for the methods selected. A flow sheet for the extraction procedure is presented as Fig. 1.

Stage A: 10 gm. soil samples were shaken for 1 minute with 50 ml. of water in 100 ml. plastic centrifuge tubes. Tubes were then centrifuged at 2500 g. (R.C.F.) for 10 minutes. The supernatant solution was decanted, the floating material was filtered off, and the filtrate saved for analysis as Fraction A. Stage B: The residue from stage A was then extracted four times with 50 ml. of $0.1N H_2SO_A$ each time by dispersing the suspension in a water bath for 15 minutes. Dispersion in the water bath involved the use of the DisOntegrator Ultrasonic cleaner supplied by Ultrasonic Industries Inc., N.Y., with system No. Forty and Generator Model No. G.-40 Cl-P with power output 80 watts. The tank had a half gallon capacity. The tubes were then centrifuged at 2500 g. (R.C.F.) for 15 minutes each time, the supernatant solution decanted off and saved for analysis as Fraction B.

Stage C: The residue from stage B was then
extracted with 50 ml. of distilled water dis-

persing the suspension in a water bath for 15 minutes. The tubes were then centrifuged for 15 minutes and the solution decanted off and saved for analysis as $\underline{\text{Fraction}}$ $\underline{\text{C}}$.

Stage D: The residue from stage C was then extracted three times with 50 ml. of 0.05M

Na4P2O7 + 1N K2SO4 (pH 7.0) by dispersing the suspension in a water bath for 15 minutes each time. The tubes were then centrifuged at 6000 g. (R.C.F.) for 15 minutes. The The supernatant solution was then decanted off and saved for analysis as Fraction D.

Stage F: The residue from stage D was then extracted twice with 200 ml. of 0.05N NaOH by shaking in an end-to-end shaker for one hour each time. The tubes (500 ml.) were then centrifuged for 15 minutes at 6000 g. (R.C.F.) and the supernatant decanted off and saved for analysis as Fraction F.

Stage G: The residue from stage F was then hydrolyzed with 25 ml. of 72% $\mathrm{H_2SO_4}$ for 15 minutes and the hydrolysate diluted to 1N $\mathrm{H_2SO_4}$ with distilled water. The diluted hydrolysate was then refluxed for 16 hours after which it was allowed to cool. Then the hydrolysate was filtered and saved for analysis as Fraction G.

Stage DD, DDD and DS were not in the main sequence of extraction. The fractions were obtained as follows:

Stage DD: The residue from stage C was extracted with 0.05M ${\rm Na_4}^{\rm P}{_2}^{\rm O}{_7}$ (pH 7.0) in a similar manner as in stage D to give <u>Fraction DD</u>.

Stage DS: The residue from stage C was ex-

tracted with 1N K₂SO₄ in a similar manner as in stage D to give <u>Fraction DS</u>.

Stage DDD: The residue from stage C was extracted with 50 ml. of distilled water and 1.76 gm. of Chelex-100 resin in the sodium form by shaking the suspension for 15 hours in an end-to-end shaker. The supernatant solution obtained after centrifugation at 6000 g.

(R.C.F.) for 15 minutes, was decanted off and

Initially, in developing the extraction procedure, no attempt was made to extract carbohydrate material free of other organic constituents. The approach entailed the removal of carbohydrates with successive treatments so that location and ease of access played an important role in determining the amounts of material extracted. Presumably, the more easily accessible materials would be extracted first, for they are less likely to be tightly bound than those within the aggregates because of their more recent time

save for analysis as Fraction DDD.

of deposition. This approach can be considered to represent at least a recent record of the state of carbohydrates in the horizon and when related to a profile, gives some record of the carbohydrate distribution within. In order to differentiate between the different binding mechanisms, it is also necessary to extract carbohydrate materials in a selective manner, in this case by using different types and strengths of extracting solutions. The various stages were rationalized as follows:

Stage A: With as many binding agents as exist in the soil, it is to be expected that the freely water soluble portion of carbohydrates would not be substantial. The amount of material extracted would mainly be dependent on the amount of water used and the degree of dispersion. Since the soil had been dried before extraction, only a very limited comparison is in order. This can only represent partially what would occur on the wetting and drying of the soil in the field. importance would conceivably include its influence on immediate energy availability to organisms even though this may not be the only energy source available at the time. This portion can also indicate the amounts available for leaching to lower horizons during changing wet and dry conditions. The carbohydrates may or may not be bound to ions in the soil and may also indicate an equilibrium state between soluble and

insoluble material.

Stage B: The dilute acid extractant was used to solubilize material at a slightly lower pH than the soil pH and should represent part of the normal fulvic acid fraction obtained after acidification of the sodium hydroxide extract. These fractions would not be directly comparable because of the limited degree of dispersion in the dilute acid extraction compared with that achieved with sodium hydroxide in the conventional extraction. It is possible that the slightly more acid conditions can solubilize carbohydrates which had precipitated out under higher pH conditions or were bound through acid soluble components. The extent of hydrolysis should not be very great due to the dilution of the extractant and the low temperature during extraction.

Stage C: This stage was simply intended to remove excess acid and the remainder of carbohydrates extracted by the dilute acid.

Stage D: The sodium pyrophosphate should remove ions involved in bridge-bonding of carbohydrate material to clays and other organic matter, thereby releasing them usually into the solution. The concentrated salt should provide a homo-ionic surface of potassium, especially for the clay and other organic materials, thereby minimizing bridging through other ions. The combination of effects is designed to obtain

some indication of the .ions and the carbohydrate materials that were involved in the bridging (adsorption) mechanism by associating those constituents released simultaneously.

Stage F: The powerful dispersing effect of this alkali has been demonstrated frequently and was used to extract more strongly bound material and material which was excluded from extraction due to incomplete dispersion of the sample.

Stage G: This hydrolysis was used to measure the remainder of carbohydrates that were too large to dissolve in the solutions or that were too tightly bound to be extracted by any of the previous extractants.

Stage DD: A comparison can therefore be made between the ions and carbohydrate materials released by these two methods (D and DD) of extraction as some different bridges may be expected to persist or sever in the presence of the complexing agent alone.

Stage DS: A similar comparison with stage D can be made and this should indicate the effect of providing opportunity for a homo-ionic surface without any aid from complexation. This should indicate the strengths of the ionic bridges with respect to those possibly formed through potassium.

Stage DDD: A comparison can be made between the solid and liquid forms of chelating (complexing)

agents even though their functional groups are different. This could indicate how the degree of penetrability affects the dissolution of bridge bonds.

On the other hand, the difference in functional groups
can indicate their susceptibility to dissolution
because of their different binding strengths.

Flow Sheet: Sequential Extraction Procedure

```
10 grms
  2 mm SOIL
WATER (1 x 50 mls)
       - shake for 1 min.
       - centrifuge - 2500 g for 10 mins.
       - filter off light fraction + Decant + Fraction A
STAGE A
  Residue
0 | \text{IN } \text{H}_2 \text{SO}_{\text{H}} \text{ (4. x 50 mls)}
       - disperse (4 x 15 mls)
       - centrifuge - 2500 g for 15 mins.
       - Decant + Fraction B
STÄGE B
  Residue
H_2O (1 x 50 mls)
       - as in B + Fraction C
STAGE C
  Residue
                                                        TNK2SO4
                                                           - as in D - Fraction DS
                                                        *STAGE DS
0.05 MNa P207
+ INK2SO4 (PH7 = 7.0) (3 x 50 mls)
       - disperse (3 x 15 mins.)
       - centrifuge - 6000 g for 15 mins.
       - Decant - Fraction D
STAGE D
  Residue
                                                     0|04 \text{ MNa}_{4}P_{2}O_{7} \text{ (pH = 7.0)}
                                                            - as in D → Fraction DD
                                                     *STAGE DD
0.05 NNaOH (2 x 200 mls)
       - shake for 1 hr.
       - centrifuge - 6000 g for 15 mins.
- Decant + Fraction F
                                                  CHELEX-100 (Na)
STAGE F
                                                      (1.76 \text{ gms} + 50 \text{ mls H}_20)
                                                     - shake for 15 hrs.
- centrifuge - 6000 g for 15 mins.
  Residue
                                                     → Decant → Fraction DDD
                                                 *STAGE DDD
Final Hydrolysis
       - 25 mls of 72% (15 mins.)
       - dilute to INH SO<sub>4</sub> - reflux for 16 hrs.
       - filter + Decant + Fraction G
STAGE G
```

Not part of main sequence.

RESULTS AND DISCUSSION

GENERAL CHARACTERISTICS OF SAMPLES

The tabulated physico-chemical properties of the samples are presented in Tables IIA and IIB.

Sample pH ranged from 4.30 to 5.75. There was a general increase in pH with depth in each soil profile. The clay content ranged from 31.9 to 59.5 percent and appeared to be highest in Ap and Ah horizons with the exception of the Cg horizon of the Langley series which was highest for that soil. The silt and sand contents ranged from 39.2 to 61.2, and 0.2 to 9.3 percent respectively.

The total carbon and nitrogen contents ranged from 0.1 to 9.9 and 0.01 to 0.58 percent respectively. The total carbon and nitrogen generally decreased with depth, with the exception of the Ah₂ horizon of the Hazelwood series which was higher in the upper adjacent horizon but not as high as the Ah₁ horizon.

The carbon to nitrogen ratios ranged from 8.3 to 19.0 and did not show remarkable uniformity with depth. The total cation exchange capacity decreased with depth with the appropriate indication of a clay size particle accumulation in the Btg horizon of the Langley series. There was a general increase in exchangeable calcium and magnesium contents with depth, the former showing a greater increase.

The yields of hexoses and pentoses released at each stage of the extraction sequence expressed as $\mu g/g$ of soil and as percentages of the total extractable hexoses and pentoses are presented in Table III and IV.

HEXOSE DISTRIBUTION

Stage A. Water Soluble Hexoses

The water soluble hexoses extracted ranged from zero to 2,475 μ g/g soil whereas the percentages of the total extractable hexoses for the corresponding horizons ranged from zero to 4.7%.

There was a general decrease in the amount of hexoses with depth, correlating with the total carbon contents, except in the Hazelwood series where there was an increase in the hexoses to the Ah₂ horizon, then a decrease in the Cg horizon. There was a similar correlation between the percentages of the hexoses. Probably, the most notable feature of this stage was that the Btg horizon yielded no water soluble hexoses.

Stage B. Hexoses Soluble in Dilute Acid (0.1N H₂SO₄)

The amounts of hexoses soluble in dilute acid ranged from 9 to 4,950 $\mu g/g$ of soil amounting to 0.18 to 20.8% of the total extractable hexoses.

In the Langley series, the highest amounts of hexoses were observed in the Ah and Ap horizons corresponding with the total carbon contents. However,

TABLE IIA

PHYSICAL CHARACTERISTICS OF SAMPLES

Soil Sub groups	Series	Topography	Horizons	Depth (ins)	Sand (%)	Silt (%)	Clay (%)
Humic	Langley	Level to	Ap	0 - 9	5.3	45.4	49.3
Eluviated Gleysol		depressional	Ah	9 - 20	5.0	44.6	50.4
Gieysoi			Aeg	26 - 35	2.0	53.6	44.4
			Btg	35 - 45	2.8	49.8	47.4
		•	Cg	52+	0.5	46.0	53.5
Orthic Gleysol	Hatzic	Level	Ар	0 - 8	1.3	39.2	59 . 5.
			Bg	8 - 20	0.3	48.6	51.1
			Cg	28+	0.2	55.8	44.0
Orthic	Hazelwood	Level to	Ah _l	0 - 10	0.8	61.2	38.0
Humic Gleysol		depressional	Bg	15 - 23	1.5	53.3	45.2
OTC 3 D T			*Ah ₂	23 - 32	0.3	45.8	53.9
			.Cg	36+	9.3	58.8	31.9

^{*}According to the current Canadian classification ${\rm Ah}_2$ should read ${\rm Ah}_{\rm b}{}^{\bullet}$

TABLE IIB

CHEMICAL CHARACTERISTICS OF HORIZON SAMPLES

Series	**	7.7	Total	Total	C/N	Exchangea	C.E.C.			
	Horizon	pН	© C (%)	N (%)	ratio	Na	K	Ca	Mg	(me/100g)
Langley	. Ap	4.35	9.9	0.58	17.1	0.4	0.4	3.4	7.3	58.0
	Al	4.35	9.5	0.50	19.0	0.3	042	3.1	6.4	48.6
	Aeg	5.10	0.4	0.04	9.8	0.6	10.2	8.3	5.8	24.2
	. Btg	5.60	0.2	0.02	12.0	0.8	0.3	11.6	7.3	39.4
	Cg	5.75	0.1	0.01	10.7	0.7	0.5	9.8	6.4	28.8
									•	
Hatzic	Аp	4.30	4.6	0.49	9.4	0.2	0.4	2.1	5.2	49.0
	Bg	4.65	0.8	0.10	8.3	0.1	0.1	3.7	7.3	36.4
	Cg	4.70	0.5	0.06	8.8	0.2	0.1	6.9	9.4	19.8
Haz el wood	Ah _l	4.40	4.5	0.42	10.7	0.1	0.1	1.3	5.3	37.3
	Bg	4.30	1.9	0.15	12.4	0.1	0.3	2.1	5.0	35.7
	Ah ₂	4.50	2.6	0.23	11.4	0.2	0.2	4.6	6.4	36.2
	Cg	4.60	0.3	0.03	9.7	0.2	0.2	6.3	6.3	24.3

TABLE III

HEXOSE DISTRIBUTION IN FRACTIONS (µg hexose/g soil)

(figures in parenthesis represent fraction - hexose as percent or total extractable hexose)

Series Horizons			Langle	У		•	Hatzio	:		Н	iaz elw ood	•	
Stages	Ap	Ah	Aeg	Btg	Cg	Ap	Bg	Cg	Ah _l	Вg	Ah ₂	Cg	_
Α '	2,475	641	230	< 1	45	396	90	< 1	45	90	117	23	
A	(4.7)	(1.2)	(3.7)	(<0.1)	(3.5)	(1.4)	(1.3)	(<0.1)	(0.2)	(1.1)	(1.7)	(0.5)	
В	3,060 (5.8)	4,950 (9.3)	450 (7.3)	9.0 (0.2)	270 (20.8)	896 (2.5)	540 (7.9)	360 (7.8)	1,584 (5.5)	180 (2.3)	468 (6.8)	360 (8.7)	
C	138 (0.4)	180 (0.3)	< 1 (<0.1)	< 1 (<0.1)	158 (12.1)	170 (0.5)	135 (2.0)	180 (3.9)	54 (0.2)	432 (5.4)	90 (1.3)	113 (2.7)	
D	8,100 (15.2)	6,500 (12.1)	9 (0.2)	< 1 (<0.1)	168	2,228	1,202 (17.5)	405 (8.7)	1,225	432 (5.4)	108 (1.6)	90 (2.2)	47.
DD	5,700 (10.7)	5,400 (10.1)	450 (7.3)	135 (2.7)	405 (31.0)	2,228 (6.2)	567 (8.3)	630 (13.6)	3,375 (11.8)	635 (7.9)	567 (8.2)	270 (6.5)	
DS	920 (1.7)	2,700 (5.0)	108 (1.7)	540 (10.7)	360 (29.0)	2,835 (7.9)	1,215 (17.7)	600 (13.0)	783 (2.7)	270 (3.4)	24	338 (8.1)	
DDD	801 (1.5)	781 (1.8)	248 (4.0)	189 (3.8)	203 (16.5)	1,418 (3.9)	216	180 (3.9)	1,134 (4.0)	473 (5.9)	365 (5.3)	113 (2.7)	
F	17,370 (32.7)	16,200 (30.2)	1,368 (22.0)	1,800 (35.7)	216 (16.6)	13,230 (37.7)	1,215	600 (12.7)	8,675 (30.2)	4,050 (50.6)	1,890 (27.5)	900 (21.6)	
G	24,375 (45.8)	26,250 (49.0)	3,712 (59.8)	3,094 (61.4)	475 (36.6)	19,125 (53.1)	4,320 (62.9)	3,400 (73.4)	15,000 (52.2)	2,625 (32.8)	3,750 (54.5)	2,500 (60.1)	
Total* Extractable Hexoses	53,118	53,570	6,210	5,038	1,298	36,045	6,867	4,630	28,733	8,006	6,882	4,160	

 $^{^{*}}$ Total extractable hexoses represents the sum of fractions A, B, C, D, F and G.

the Ah horizon yielded a much higher quantity than did the Ap horizon. The Aeg, Btg and Cg followed the same pattern as the water soluble hexoses. The ratio of acid extractable to water extractable hexoses was substantially higher in the Ah than the Ap horizon (7.7 and 1.2 respectively). The Aeg horizon yielded twice the amount of hexoses while the Cg horizon yielded six times as much hexoses obtained by water extraction. Again, the Btg horizon failed to yield a sizeable portion of its hexose content.

In the Hatzic series, there was a general decrease in the amounts of hexoses extracted but there was an increase in the percentages with depth of profile. The hexoses yielded in stage A and stage B paralleled each other. This may indicate similar types of hexose-containing substances being solubilized by a stronger agent with more insonation or that the reagent had no effect and the increased dispersion was responsible for the increased yield of water soluble hexoses. Also this may indicate the removal of some ions by the acid extractant.

In the Hazelwood series, there is also a general decrease in the amounts of hexoses extracted with depth, with the exception of the lower yield from the Bg horizon. Again, there is the increase in percentage of hexoses with depth, excepting the Bg horizon. The yield of hexoses in stage B do not parallel those

of stage A, except with regard to the high levels obtained from the Ah₁ horizon. The increases in yield over stage A in each horizon are at least five fold except the Bg horizon which is only two-fold.

Generally, the indication is that the dilute acid effectively severed bonds yielding sizeable portions of the hexoses contained in the respective horizons, especially the Cg horizons.

Stage D. Hexoses Soluble in 0.05M
$$Na_4^P_2^O_7$$
 + $IN K_2^{SO_4}$ (pH 7.0)

The yields of extractable hexoses for this extraction ranged from zero to 8,100 μ g/g of soil and the percentages of total extractable hexoses ranged from zero to 17.5.

In the Langley series, the Ap and Ah horizons yielded copious quantities of hexoses whereas the Aeg and Btg horizons yielded negligible amounts. The percentages of hexoses in the Ap, Ah and Cg horizons respectively were 15.2, 12.1 and 12.9, indicating similar equilibrium conditions.

The Hatzic series showed a general decrease in the amount of hexoses extracted with depth but the percentage of hexoses extracted was far greater in the Bg horizon than in the Ap and Cg horizons. The results indicate that this extractant severed more bonds of this type in the Bg horizon than it did in the other

horizons. Therefore, this type of bond is probably more typical of the Bg horizon than the others.

The Hazelwood series showed a general increase in yield of hexoses. The low - high - low - high alternating pattern of the hexoses yielded from the Ah, Bg, Ah₂, Cg horizons was the opposite of the high - low - high - low alternating pattern of the total carbon for the corresponding horizons. The percentage yield in the Bg horizon was again the highest. The indication is that the Bg horizon appears to be opposite to that of the Btg horizon. A possible explanation lies in the difference in clay contents between the Btg and Bg horizons.

Stage DD. Hexoses Soluble in 0.05M Na₄P₂O₇

(pH 7.0)

A greater degree of dispersion of the samples was displayed in this stage than any of the other parallel stages. The amounts of hexoses extracted ranged from 135 to 5,700 μ g/g of soil while the percentages of total extractable hexoses ranged from 2.7 to 31.0.

In the Langley series, the yield of hexoses was again very high as in stage D and similar percentages were obtained for the Ap and the Ah horizons.

The Aeg, Btg and Cg horizons yielded very much higher amounts of hexoses than stage D in a somewhat parallel

pattern. Accordingly, the dilute buffer solution extracted 31 percent of the total hexoses from the Cg horizon. According to the above, it follows that the buffered salt solution was more effective in the Ap and Ah horizons while the buffer solution was most effective in the lower horizon in releasing hexoses.

In the Hatzic series, the yields of hexoses showed a steep decline from the Ap to the Bg horizon and a slight increase in the Cg horizon. However, the percentages showed a continued increase with depth of profile.

In the Hazelwood series, the amounts of hexoses extracted showed a similar decrease with depth as in stage D but the amounts of extracted hexoses were all higher. Correspondingly, the percentages were higher but the high - low - high - low pattern of the total carbon was followed.

It is evident that the amounts of hexoses extracted generally decrease with depth but the percentages do not follow similar patterns in all cases.

Stage DS. Hexoses Soluble in 1N K_2SO_4

This extraction always yielded the clearest solution of the parallel D stages. The amounts of hexoses extracted ranged from 24 to 2,835 μ g/g of soil while the percentages of total extractable hexoses ranged from 0.4 to 29.0.

The Langley series showed a low - high - low -

high - low sequence in the amounts of hexoses extracted from the Ap, Ah, Aeg, Btg and Cg horizons respectively. The quantities extracted from the upper Ah and Ap horizons are far less than that extracted in the stages D and DD, with an anomalously low amount from the Ap The Aeg horizon yielded an intermediate amount, the Btg a greater amount and the Cg an intermediate quantity of hexoses when compared with their counterparts in stages D and DD. The percentages of total extractable hexoses showed a general increase with depth but the Aeq horizons were the same as the Ap horizons, indicating a similar equilibrium condition. Of note is the fact that the pyrophosphate buffer alone and the concentrated salt solution alone yielded similar percentages of hexoses while the combined extractants in the Cg horizon yielded less.

The Hatzic series showed a general decrease with depth in profile in the amounts of hexoses extracted but the yields were slightly higher than those in stage D. This indicates that potassium sulphate is most effective alone in this soil, especially on the Bg horizon. The ${\rm K_2SO_4}$ -sensitive material in the Bg horizon is probably quite different from that which is extracted by the pyrophosphate buffer solution alone.

The Hazelwood series showed a decrease from the Ah to the Ah₂ horizon in the amounts of hexoses extracted, then a substantial increase in the Cg hor-

izon. The percentages of hexoses showed a similar pattern as in stage D, that is, opposite to the carbon pattern, although the amounts were not as high.

Stage DDD. Hexoses Soluble in Chelex-100 Na

The amounts of hexoses extracted ranged from 113 to 1,418 μ g/g of soil and the percentages of total extractable hexoses ranged from 1.5 to 16.5.

In the Langley series, the amounts of hexoses extracted showed a general decreasing trend with depth and the values were less than for any other comparable stages. The values did not parallel those of stage DD and at best only partially similar chelating solubility properties can be presumed. The percentages of hexoses showed an increasing trend with depth which is a different pattern from stage DD. In any case, the percentage for the Cg horizon showed that both chelating agents were efficient for this horizon.

In the Hatzic series, the amounts of hexoses extracted showed a general decrease with depth but the percentages of hexoses were fairly similar indicating an equilibrium condition throughout the profile for this extractable material.

In the Hazelwood series, the amounts of hexoses extracted showed a general decrease with depth, however, the percentage values for the Ah₁ and Cg horizons were half those for the Bg and Ah₂ horizons. This could indicate some accumulation in the former horizons.

Stage F. 0.05N NaOH Soluble Hexoses

The amounts of hexoses extracted ranged from 2,160 to 17,370 μ g/g of soil and the percentages of total extractable hexoses ranged from 12.7 to 50.6.

In the Langley series, the NaOH extractable hexoses showed a general decrease with depth except that the Aeg horizon indicated some depletion and the Btg horizon some accumulation. The only other stage where a similar effect was shown was with the $1 \text{N K}_2 \text{SO}_4$ solution. The percentage values accentuated this effect to a greater degree.

In the Hatzic series, the NaOH extractable hexoses showed a general decrease with depth as did the percentage values.

In the Hazelwood series, the NaOH extractable hexoses showed a 50 percent decrease per horizon with depth. However, the percentage values showed some accumulation in the Bg horizon.

Stage G. Final Hydrolysis

A large portion of the hexoses for most horizons was shown to be very tightly bound. The easily
extractable hexoses removed in the previous stages can
therefore be labelled the labile portion and this
stage represents the stable polysaccharides. It might
be suggested that this large portion of stable material
is partially responsible for the basic stability of
microaggregates while the various labile fractions

modify them dynamically into various larger aggrequates.

In the Langley series, there is a general decrease in amounts of stable hexoses with depth while the percentages show a gradual increase from the Ap to the Btg horizon and a fall in the Cg horizon. The indication is that even though larger quantities of stable hexoses reside in the upper horizons, greater percentages accumulate in the lower Btg horizons. The stabilization of a larger percentage of hexoses in the Btg horizon could partially be accounted for by a clay accumulation. The distribution of cellulose cannot explain these trends.

In the Hatzic series, the amounts of hexoses again show a general decrease with depth. The percentages show the reverse indicating a greater stabilization at the lower depths of hexoses.

In the Hazelwood series, the amounts of hexoses show a similar alternating high — low — high — low sequence as the total carbon. However, the percentages show an increasing trend from the Bg to the Cg horizon with the ${\rm Ah}_1$ horizon having a similar value as the ${\rm Ah}_2$ horizon.

TOTAL EXTRACTABLE HEXOSES

The total extractable hexoses consisting of stages A, B, D, F and G in the Ap and Ah horizons, were very much higher than the rest. In general, it can be

said that the organic A horizons contain the greatest amounts of hexoses but the lower horizons have a larger proportion of their hexoses stabilized. In the buried Ah₂ horizon, the absence of continued surficial addition of organic material has changed its character in that even though it has a higher total carbon content than the Bg horizon, the total extractable hexoses are less, whereas amount of hexoses fixed is greater in the Ah₂ horizon than in the Bg horizon. Further investigation of the above observation concerning the Ah₂ horizon may lead to a method of its characterization and differentiation.

It should be noted that the nature of the extraction procedure gives reason to believe that most, if not all, of the carbohydrates were extracted from the soil.

PENTOSE DISTRIBUTION

Stage A. Water Soluble Pentoses

The water soluble pentoses ranged from 80 to $608~\mu g/g$ of soil and the percentages of total extractable pentoses ranged from 0.3 to 4.7.

In the Langley series, similar quantities were extracted from the Ap and Cg horizons and from the Ah, Aeg and Btg horizons. The Cg horizon yielded the highest percentage while the Ah and the Btg were relatively low.

Series Horizons			Langl	ey			Hatzio	2		F	łaz elw ood	1	
Stages	Ар	Ah	Aeg	Btg	Cg	Ap	Bg	Cg	Ah	Bg	Ah ₂	Cg	
A	450 (1.1)	113	169 (1.3)	113	450 (4.7)	90 (0.3)	80	225 (1.6)	585 (2.3)	608 (3.4)	608 (4.0)	563 (3.7)	
В	5,940 (15.5)	5,062 (14.2)	2,430 (18.5)	3,330 (21.9)	3,600 (37.9)	2,925 (9.2)	2,925	3,038	3,735 (14.9)	3,060 (17.3)	3,060	2,925 (19.2)	_
С	50 (0.1)	< 1 (<0.1)	160 (1.2)	100 (0.7)	57 (0.6)	40 (0.1)	40 (0.4)	5 > (0.4)	240 (0.9)	245 (1.4)	210 (1.3)	100	
D	4,275 (11.1)	4,005 (11.2)	450 (3.4)	338	1,125 (11.8)	1,800 (5.7)	405 (3.7)	1,350 (9.8)	2,025 (8.0)	1,755 (9.9)	1,013 (6.7)	2,025 (13.3)	5 7.
DD	3,960 (10.3)	3,960 (10.4)	338 (2.7)	608 (4.0)	720 (7.6)	1,800 (5.7)	405 (3.7)	540 (3.9)	2,093 (8.3)	1,857 (10.5)	1,958 (12.9)	1,688 (11.1)	
DS	10,125 (26.3)	10,125	900 (6.9)	1,575 (10.3)	2,700 (28.4)	720 (2.3)	900 (8.1)	450 (3.3)	2,093	1,620 (9.2)	1,620 (10.7)	1,654 (11.0)	
DDD	563 (1.5)	675 (1.9)	450 (3.4)	113 (0.7)	450 (4.7)	788 (2.5)	101	225 (1.6)	551 (2.2)	743 (4.2)	585 (3.9)	563 (3.7)	
F	13,050 (33.9)	11,880	1,800 (13.7)	3,600 (23.6)	1,350 (14.2)	11,880	1,440 (13.1)	2,880 (20.8)	6,413 (25.5)	4,725 (26.7)	3,915 (25.8)	4,050 (26.5)	
G	15,000 (39.0)	14,875 (41.8)	8,250 (62.8)	7,500 (49.2)	2,813 (29.6)	15,125 (47.5)	6,125 (55.6)	6,563 (47.5)	12,093 (48.1)	7,188 (40.7)	5,438 (35.8)	5,438 (35.8)	
* Total Extractable Pentoses	38,444	35,620	13,146	15,250	9,512	31,860	11,015	13,820	25,158	17,682	15,178	15,263	

 $[\]mbox{\ensuremath{^{\star}}}$ Total extractable pentoses represents the sum of fractions A, B, C, D, F and G.

In the Hatzic series, the yields of pentoses showed a general increase with depth as did the percentage yields.

In the Hazelwood series, similar amounts were extracted fro the horizons but there was an increase in percentage yields with depth. No clear overall trends were shown by the yields of pentoses but the percentage values showed and increasing trend with depth.

Stage B. Pentoses Soluble in Dilute Acid

The pentose yields ranged from 2,430 to $5,940~\mu g/g$ of soil and percentages of total extractable pentoses from 9.2 to 26.6.

In the Langley series, the amounts of pentoses extracted decreased from the Ap to the Aeg horizon and then increased further to the Cg horizon showing some depletion in the Aeg horizon and accumulation in the lower Btg and Cg horizons. The percentage values showed a general increase in depth from the Ah horizon, but the Ap horizon was slightly higher than the Ah horizon. The indication is that although there is a depletion in quantities in the Aeg area, this portion of the total pentoses which is soluble in dilute acid increases with depth.

In the Hatzic series, the amounts of pentoses extracted increased very little with depth whereas the percentages, although increasing with depth, showed

some accumulation in the Bg horizon.

In the Hazelwood series, the amounts of pentoses extracted showed a decrease with depth while the percentages showed an increase.

The general trends were, even though there might be depletions and accumulations in the quantities of pentoses, that this portion of the pentoses always showed a general increase with depth.

Stage D. Pentoses Soluble in 0.05M Na₄P₂O₇ +

$1N K_2 SO_4 (pH 7.0)$

The amounts of pentoses extracted ranged from 338 to 4,275 μ g/g of soil and the percentages of total extractable pentoses from 2.2 to 13.3.

In the Langley series, the amounts of pentoses extracted and the percentages of pentoses showed not only a decrease down to the Btg horizon but an accumulation in the Cg horizon. Similar trends were observed in the Hatzic series.

In the Hazelwood series, the yields of pentoses showed the same trend as the above. The percentages also showed the same pattern, with the Ah₂ horizon having the lowest value.

Stage DD. Pentoses Soluble in 0.05M Na₄P₂O₇

(pH 7.0)

The yields of pentoses ranged from 338 to $3,960~\mu g/g$ of soil and the percentages of total ex-

tractable pentoses ranged from 2.7 to 12.9.

In the Langley series, the amounts of pentoses extracted decreased down to the Aeg horizon, and then increased to the Cg horizon. The values showed that less pentoses were released by pyrophosphate alone, except in the Aeg and the Btg horizons whose yields were greater. The percentages showed a similar pattern.

In the Hatzic series, similar quantities were released from the Ap and Bg horizons but the Cg horizon yielded less with the pyrophosphate alone.

In the Hazelwood series, the amounts of pentoses extracted followed the high - low - high - low
sequence of the total carbon whereas the percentages
showed a general increase with depth.

Stage DS. Pentoses Soluble in $1N K_2 SO_4$

The yields of pentoses ranged from 450 to $10,125~\mu g/g$ of soil and the percentages of extractable pentoses ranged from 2.3 to 28.4.

In the Langley series, the yields and percentages of pentoses showed a decrease down to the Aeg horizon, then an increase to the Cg horizon. However, the values were about twice as great as those for the buffer solutions of stages D and DD.

In the Hatzic series, the yields and percentages of pentoses showed an increase down to the Bg horizon, then a decrease in the Cg horizon. This situation was opposite to that determined for both of

the buffer solutions and the values for the Ap and Cq horizons were less.

In the Hazelwood series, the yields of pentoses showed a slight decrease with depth whereas the percentages showed an increase.

Stage DDD. Pentoses Soluble in Chelex-100 Na⁺

The amounts of pentoses extracted ranged from 101 to 788 μ g/g of soil and the percentages of total extractable pentoses ranged from 0.7 to 4.7.

In the Langley series, the amounts of pentoses extracted were relatively similar, except for the Btg horizon which was far lower. The values were lower than those of the pyrophosphate buffer alone in all of the horizons except the Aeg horizon. The percentage values showed a general increase with depth except for the Btg horizon which indicated that the materials were not susceptible to chelating agents in the solid form.

In the Hatzic series, the amounts of pentoses extracted and percentages showed a similar pattern as for the pyrophosphate buffer alone, though the values were less.

In the Hazelwood series, the amounts of pentoses extracted showed a similar pattern to the pyrophosphate buffer, whereas the percentage values showed a slightly different pattern.

Stage F. Pentoses Soluble in 0.05N NaOH

The NaOH soluble pentoses ranged from 1,350 to 13,050 µg/g of soil and the percentages of extractable pentoses ranged from 13.1 to 37.3.

In the Langley series, the yields of pentoses and percentages showed large amounts residing in the Ap and Ah horizons with a depletion in the Aeg horizon and an accumulation in the Btg horizon.

In the Hatzic series, high amounts in the Ap horizon and low amounts extracted in the Bg horizon, compared with the Cg horizon, followed the patterns of both pyrophosphate buffer solutions.

In the Hazelwood series, the amounts of pentoses extracted showed a decreasing trend with depth
whereas the percentage values were quite similar showing some equilibrium throughout the profile.

Stage G. Final Hydrolysis

In the Langley series, similar amounts and percentages of extractable pentoses for the organic Ah and Ap horizons, smaller than for the eluviated horizon, indicated that the pentoses were strongly bound in the Aeg horizon. The Btg and Cg values were less. However, the stable pentoses showed a decrease with depth.

In the Hatzic series, the yields of pentoses showed a depletion in the Bg horizon yet the percentage values showed a relative accumulation of stable forms

of pentoses in this horizon.

In the Hazelwood series, the yields of pentoses and percentages showed a decrease with depth down
to the Ah₂ horizon and further, an increase in the Cg
horizon. The general pattern indicated appeared to
be the reverse of that obtained for the distribution
of hexoses.

TOTAL EXTRACTABLE PENTOSES

The Langley series yielded very high values for the A horizons, slight depletion in the Aeg horizon and an accumulation in the Btg horizon in a general decrease with depth.

In the Hatzic series, a pentose 'sink' in the Bg with comparable amounts in the Ap and Cg horizons were observed.

In the Hazelwood series, a general slight decrease with depth, with quite similar values in the Ah, and Cg horizons were observed.

Results for stage C were ignored in both hexoses and pentoses distribution analysis because they were thought to be insignificant.

HEXOSE:PENTOSE RATIOS (Table V)

A comparison of hexoses and pentoses was made to indicate trends of relative sugar accumulations according to depth of profile. The ratios varied from

less that 0.01 to 5.70 for individual extractions.

In the Langley series, most of the values showed a decrease of hexoses in relation to pentoses. The highly organic Ap and Ah horizons had notably higher ratios than the Aeg, Btg and Cg horizons. Most of the ratios for Ap and Ah horizons were greater than one except for 0.1N $\rm H_2SO_4$ and $\rm 1N~K_2SO_4$ extracts, indicating that amounts of hexoses were generally greater than pentoses.

In the Hatzic series, there was a similar decrease with depth except with the Chelex-100 and pyrophosphate buffer solutions (with and without ${\rm K_2SO_4}$) where there was an accumulation in the Bg horizon of hexoses as opposed to pentoses. Again, in the 0.1N ${\rm H_2SO_4}$ extract, the values were below one, indicating that hexoses were generally greater than pentoses.

In the Hazelwood series, the decrease with depth was very clearly shown. All the ratios for the Ah₁ horizon except for 0.1N H₂SO₄ (with and without pyrophosphate) were above one, indicating a generally greater occurrence of hexoses than pentoses.

These observations may indicate that:

1) microbial degradation of pentoses occurs less rapidly, that is pentose containing carbohydrates are bound more tightly, thus preventing rapid degradation. 2) that hexoses are converted to pentoses
to a greater extent than the reverse.

However, the equilibrium that may exist between hexoses
and pentoses in soil aggregation favours the retention
of pentoses, especially in the lower horizons.

TABLE V
HEXOSE/PENTOSE RATIOS

Series Horizons Stages	Langley					Hatzic			Hazelwood			
	Ap	Al	Aeg	Btg	Cg	Ap	Bg	Cg	Ah	Bg	Ah ₂	Cg
Stage A	5.50	5.70	1.36	sî	0.1	4.40	1.10	*	1.08	0.15	0.19	0.04
Stage B	0.52	0.98	0.19	0.01	0.08	0.31	0.19	0.12	0.42	0.06	0.15	0.12
Stag e D	1.89	1.63	0.02	*	0.15	1.24	2.97	0.30	0.61	0.25	0.11	0.04
Stage DD	1.44	1.46	1.33	0.22	0.56	1.24	1.40	1.17	1.47	0.34	0.27	0.1ô
Stage DS	0.09	0.27	0.12	0.34	0.13	3.94	1.35	1.33	0.37	0.17	0.02	0.02
Stage DDD	1.42	1.45	0.55	0.68	0.45	1.80	2.13	0.80	2.06	0.64	0.62	0.20
Stage F	1.33	1.36	0.76	0.50	0.16	1.11	0.84	0.21	1.35	0.86	0.45	0.22
Stage G	1.63	1.77	0.45	0.41	0.17	1.26	0.71	0.52	1.24	0.37	0.69	0.42

^{*} Hexoses extracted were too low to determine ratio.

COMPARATIVE DISTRIBUTION OF HEXOSES AND PENTOSES WITH IRON, ALUMINUM, CALCIUM AND MAGNESIUM

LANGLEY SERIES:

Stage A. Water Extraction

The results showed an increase from zero p.p.m. in the Ap horizon to 2069 p.p.m. of iron plus aluminum extracted in the Cg horizon. Only the A horizons yielded any calcium while no magnesium was extracted. The indication is that in the Ap and Ah horizons, the hexoses and pentoses are associated with calcium whereas in the Aeg and Cg horizons, they are mainly associated with aluminum. In the Btg horizon, they are mainly associated with iron. The decrease in the sugar to metal ratio with depth further indicated a greater degree of association with depth of profile. The major associations in the upper A horizons consist of calcium and iron with hexoses as compared to aluminum with pentoses in the lower horizons.

Stage B. 0.1N H₂SO₄ Extraction

The results for the iron plus aluminum extracted showed a general decrease with depth although
some eluviation of the Aeg horizon and accumulation
in the Btg horizon is indicated. The calcium plus
magnesium amounts extracted showed a general increase
with depth and similar eluviation and accumulation in
the Aeg and Btg horizons respectively. The majority

Stages	Horizons	Ca (ppm)	Mg (ppm)	Ca. + Mg (ppm)	Fe (ppm)	Al (ppm)	Fe + Al (ppm)	Fe + Al + Ca + Mg (ppm)	Hex oses + Pentoses (µg/g)	Sugars Fe + Al	Sugars Total Metals
	Аp	70	•	70	_	_	1	70	2,925	2,935.00	384.40
Stage	Ah	60	-	60	5	-	5	65	736	147.20	9.20
Δ	Aeg	10	-	10	150	341	491	501	398	0.81	0.80
Α	Btg Cg	-	-	-	320 520	50 1,549	370 2,069	370 2,069	113 495	0.30 0.24	0.30 0.24
								•			
Stage	Ap Ah	164 1,440	7,200 4,880	7,364 6,240	28,000 24,00	73,171 73,171	101,171 97,171	108,535 103,411	9,000 10,013	0.09 0.10	0.08 0.10
	Aeg	760	13,200	13,960	15,000	24,390	39,390	53,350	2,880	0.07	0.05
В	Btg	960	20,400	21,360	26,000	31,707	57,707	79,067	3,339	0.06	0.04
	Cg	1,740	15,600	17,340	20,500	30,487	50,987	68,327	3,870	0.08	0.06
Stree	ĄΦ	-	24	24	7,800	3,122	10,922	10,946	12,375	1.13	1.13
Stage	Ah	160	2,400	2,560	9,000	3,951	12,951	15,511	10,505	0.81	0.68
D	Aeg	280	4,800	5,080	190	366	556	7,346	459	0.20	0.06
	Btg Cg	80 	9,600 84,000	9,680 84,000	1,120 2,660	4,220 7,317	5,440 9,977	15,120 93,777	338 1,293	0.06 0.13	0.02 0.01
	Ap	_	24	24	18,000	10,829	28,829	28,853	0.660	0.20	0.22
Otage	Ah	-	-	-	9,000	3,950	12,950	12,951	9,660 9,040	0.34 0.70	0.33 0.70
	Aeg	40	3,120	3,160	190	366	556	3,716	799	1.42	0.21
DO	Btg	40	8,160	8,200	1,220	4,220	5,440	13,640	742	0.14	0.05
	Cg	-	4,800	4,800	2,660	7,310	9,970	19,777	1,125	0.11	0.88
Stage	Αp	40	2,400	2,400	2,160	14,634	16,794	19,234	11,045	0,66	0.57
	Ah Ana	40 140	2,400	2,440	2,000	24,390	26,390	28,830	11,825	0.49	0.45
DC	Aeg Btg	200	4,800 6,000	4,940 6.200	80 30	4,878	4,908	9,808	1,008	0.21	0.10
	Cg	100	4,800	6,200 4,900	860	2,439 10,956	2,519 11,816	8,719 16,716	2,115 3,060	0.84 0.26	0.24 0.18
	Аp	_	_	_	510	512	1,022	1,022	1,362	1.38	1.33
Stage	Ah	_	_	-	775	683	1,458	1,458	1,656	1.14	1.14
	Aeg	-	-	-	5	61	66	66	698	10.57	10.57
PDD	Btg	-	-	-	220	293	513	513	302	0.59	0.59
	CR	-	-	-	1,190	1,707	2,897	2,897	652	0.23	0.23
HATZIC											
····	Аp	300	60 .	360	50	49	99	459	486	0.90	1,06
Stage	Bg	40	-	40	65	11	175	215	170	0.97	0.79
Α	Cg	20	600	620	340	79	419	1,039	225	0.54	0.22
	Аp	920	4,800	5,720	25,000	48,780	73,780	79,500	13,821	0.05	0.05
Stage	Вg	1,600	9,600	11,200	32,000	36,385	68,585	79,785	3,465	0.05	0.03
В	Cg	1,640	10,800	12,440	20,000	25,610	45,610	58,050	3,398	0.08	0.05
	۸	_	5 1:00	E 1100	2 600	1 427	r 007	10 1177	4 028	0.00	0.30
Stage	Ap Bg	186	5,400 1,800	5,400 1,980	3,600 1,755	1,427 1,427	5,027 3,182	10,477 5,162	4,028 1,606	0.80 0.50	0.39 0.31
D	Cg	390	3,900	4,290	280	780	1,060	5,350	1,755	1.66	0.33
			000	000	C 1150		72 500	12 500	h 020	0.33	0.20
Stage	Ap Bg	-	900 1,170	900 1,170	6,450 6,150	6,000 11,780	12,600 17,930	13,500 19,100	4,028 972	0.32 0.05	0.30 0.05
DĐ	Cg	-	720	720	5,900	13,414	19,314	20,034	1,170	0.06	0.06
	An	30	1,170	1,200	870	7,317	8,187	9,387	3,535	0.43	0.38
Stage	Ap Bg	60	1,800	1,860	795	10,975	11,770	13,630	2,115	0.18	0.16
DS	Cg	Ř٨	2,400	2,480	1,150	14,634	15,784	18,264	1,050	0.07	0.05
	Ap	-	· -	-	1,300	2,012	3,312	3,312	2;205	0.57	0.57
Stage DDD	Bg	-	-	-	870	2,207	3,077	3,077	317	0.10	0.10
טטט	Cg	-	-	-	65	159	224	2,230	405	0.18	0.18
HAZ ELW OOI)										
	Ah ₁	180	*	180	5		5	185	630	126.00	3,41
Stage	Bg	90	-	90	20	50	70	160	698	9.96	4.36
Α	Ah ₂	30	-	30	160	396	550	580	725	1.32	1.25
	Cg	10	-	10	370	732	1,102	1,112	585	0.53	0.53
S+300	Ahı	1,040	1,560	2,600	22,000	41,463	63,463	73,108	5,319	0.08	0.97
Stage	Bg	800	3,600	4,400	30,000	31,707	61,707	66,107	3,140	0.05	0.05
В	Ah ₂	164	7,200	7,364	60,000	29,268	89,268	96,632	3,528	0.04	0.04 0.04
	Cg	1,480	12,600	14,080	60,000	21,951	81,951	96,031	3,285	0.04	0.04
Stage	Ah ₁	360	900	1,260	1,095	439	1,534	2,794	3,250	2.12	1.16
~	Bg	270	630	1,000	1,200	622	1,822	2,822	2,187	1.20	0.80
D	Ah ₂	120 30	360 900	480 930	300 2,085	1,024 3,549	1,324 5,634	1,804 6,564	1,120 2,115	0.85 0.38	0.62 0.32
	Cg										
Stage	Ah ₁	30	450 580	480 540	5,400 5,250	3,366	8,766	9,246	5,468 2.490	0.67 0.22	0.59 0.21
	Bg Ab	-	540 180	540 180	5,250 3,300	6,000 4,610	11,250 7,910	11,790 8,090	2,490 2,525	0.22	0.21
DD	Ah ₂ Cg	-	540	540	4,500	4,683	9 '783	9,723	1,958	0.21	0.20
		300	450	750	1,710	10,980	12,690	13,440	2,875	0.23	0.21
Stage	Ah Bg	300	450 450	750 750	1,710	10,980	12,690	12,930	1,890	0.23	0.15
DS	Ah ₂	300	180	480	1,275	7,317	8,592	9,072	1,644	0.19	0.13
	Cg	300	900	1,200	900	9,150	10,050	11,250	1,990	0.20	0.18
	Ah ₁	-	30	30	1,640	2,512	4,152	4,182	1,685	0.41	0.40
Stage	Bg Bg	-	210	210	3,450	12,804	16,254	16,464	1,215	0.08	0.07
Ctage	-0										
DOD	Ah ₂	-	60	60	2,225	7,927	10,152	10,212	950 675	0.09 0.10	0.09

of sugars to metals ratios are below 1. This may indicate a closer association between the sugars and metals. The Btg horizon shows the most association while the Ah and Ap horizons show the least.

Although the pentoses show closer relation—ships throughout the profile in the upper horizons, magnesium and aluminum were closely associated with hexoses. Throughout the profile, aluminum and iron were closely associated with pentoses, though aluminum was associated to a greater extent.

Stage D. 0.5M $Na_4P_2O_7 + 1N K_2SO_4$ (pH 7.0)

Extraction

The yields of iron plus aluminum in the Ap and Ah horizons were the highest with lower values in the Aeg, Btg and Cg horizons which increased respectively. The increase of calcium and magnesium values with depth were again demonstrated. Magnesium and iron were high in the Ap and Ah horizons while magnesium and aluminum were high in the others. The high values of hexoses in the Ap and Ah horizons along with high values of iron and aluminum indicated a closer relationship of these constituents. In the Aeg, Btg and Cg horizons, the closest relationships were shown between magnesium and pentoses and to a lesser extent, pentoses and aluminum.

Stage DD. 0.05M $Na_4P_2O_7$ (pH 7.0) Extraction Although the values and trends were quite

similar for aluminum and iron, those for calcium and magnesium were different from those of Stage D. The values were neither as high nor was the continuous decrease still present. The Ah and Cg horizons show the most prominent decreases in calcium plus magnesium amounts extracted. However, the magnesium remained high in comparison with calcium.

The result was a general decrease in hexose values but an increase in pentose values in the Aeg, Btg and Cg horizons. A further look indicated that the closer relationships in the Ap and Ah horizons were also between iron and hexoses whereas the Aeg, Btg and Cg horizons showed considerably closer relationship between magnesium and pentoses. However, in the Cg horizon considerable association between aluminum and pentoses was observed and the Aeg horizon between hexoses and magnesium.

Also, the potassium sulphate in stage D released more magnesium and pentoses than the pyrophosphate alone.

Stage DS. 1N K₂SO₄ Extraction

This extract released more aluminum and less iron than it did in the presence of pyrophosphate, except in the Btg horizon where the aluminum released was less. It also released greater amounts of calcium in the Ap, Cg and Ah horizons, greater amounts of magnesium only in the Ap horizon, and lesser amounts

in the Btg and Cg horizons, than it did in the presence of pyrophosphate. The results indicated that there were close relationships between pentoses and aluminum in the Ap, Ah and Cg horizons, and similarly close associations between pentoses and aluminum and magnesium in the Aeg horizon. However, pentoses and magnesium were closely related in the Btg horizon. Also, there was considerable association between pentoses and magnesium in the Cg horizon.

Stage DDD. Chelex-100 (Na Form) Extraction

Similar trends as in stage D and stage DD were demonstrated except that iron and aluminum were extracted to the same extents. In the Cg horizon, more aluminum than iron was extracted and throughout the profile, neither calcium nor magnesium was extracted.

These results were deceiving because the Chelex-100 is a solid, so it may have removed all the calcium and magnesium out of the solution and therefore could not be analyzed. All of the Chelex-100 extracts showed this feature, therefore, it is most likely that this occurred. Further, this extract was probably not as complete as the pyrophosphate because of the limited penetrating ability of the extractant. Another interesting feature was that although the hexose/pentose ratios for the Ap and Ah horizons were quite similar, similar amounts of iron and aluminum were extracted from both horizons.

HATZIC SERIES

Stage A. Water Extraction

There was a general decrease in calcium and an increase of magnesium, iron and aluminum amounts extracted with depth of profile. The most aluminum and the least magnesium were released from the Btg The closest relationships observed were hexoses with calcium in the Ap horizon, both hexoses and pentoses with aluminum in the Bg horizon, and primarily pentoses with magnesium in the Cg horizon. Also, there was some association between pentoses and aluminum in the Cg horizon. Basically, the same trend was seen with calcium and hexoses being associated in the Ap horizon while pentoses were associated with magnesium and aluminum in the lower horizons. Also, there is a general increase of association of total extractable sugars with metals indicating an increase of retention with depth.

Stage B. 0.1N H₂SO₄ Extraction

A similar pattern of the sugar to metal ratios, all of which were below 0.1, indicated close associations between the sugars and the metals, with the Bg horizon showing the closest. In an overall decreasing trend with depth, the Ap horizon released considerably more aluminum than iron, whereas there were similar values for the Bg and Cg horizons respectively.

Both calcium and magnesium yields showed a general increase with depth, though magnesium values were considerably greater. The increasing closeness of pentose associations with aluminum and magnesium with depth did not overshadow the association with iron. These results were quite similar for the Langley horizons.

Stage D. 0.05M Na₄P₂O₇ + 1N K₂SO₄ (pH 7.0) Extraction

High yields of magnesium, iron and aluminum respectively, throughout the profile did not follow the same pattern as the Langley series, iron plus aluminum extracted decreased with depth. The lower magnesium value for the Btq horizon and the high magnesium value for the Ap horizon were conspicuous. However, magnesium and iron were still closely associated, mostly with hexoses, in the Ap horizon. Bg horizon showed a relative accumulation of hexoses. associated with magnesium, iron and aluminum to a similar extent. This was opposite to the Langley series for the Btg horizon. However, the Cg horizon showed a similar accumulation of pentoses, as did the Langley series, being primarily associated with mag-A uniform sugar to metal ratio between 0.3 nesium. and 0.4 indicated some equilibrium common to all of the horizons.

Stage DD. 0.05M $Na_4P_2O_7$ (pH 7.0) Extraction

A very slight decrease with depth in the aluminum yields and a much greater increase for yields of aluminum plus iron were observed. High values of aluminum and iron were seen throughout the profile but a small amount of magnesium and the absence of calcium were conspicuous. There was a closer relationship between hexoses and aluminum than either pentoses with aluminum or hexoses with iron, especially in the lower horizons. Magnesium values did not indicate a considerable degree of association with the sugars. However, there was a slight accumulation of magnesium in the Bg horizon.

Stage DS. 1N K₂SO₄ Extraction

There was an increase of extractable iron,
magnesium and calcium with depth, although calcium
values were very low. Although aluminum values were
far higher than iron values, they also showed an
increase with depth. The Ap horizon showed a close
relationship between hexoses and aluminum, however
this relationship was shown to a lesser extent in the
Bg and Cg horizons. Magnesium and iron associations
were also substantial. No explanation is given for
the completely different trend shown in this extraction.

Stage DDD. Chelex-100 (Na Form) Extraction

The absence of calcium and magnesium was still evident. There was a decrease of extractable aluminum

and iron with depth. This was different from the pyrophosphate buffer extraction in that although a slight accumulation was seen for aluminum in the Bg horizon, the definite increase with depth was not evident. The hexoses and aluminum relationship was the closest throughout the profile. The sugar to metal ratios showed a similar trend, though much higher values were obtained than for the pyrophosphate extraction.

HAZELWOOD SERIES

Stage A. Water Extraction

A decrease in extractable calcium with depth and the complete absence of extractable magnesium were observed. An increase of extractable aluminum and iron with depth was also seen. The decrease of sugar to metal ratios indicated a greater degree of association between the sugars and aluminum than with iron associations with depth. However, hexoses and pentoses appeared equally closely related with the metals in the Ah₁ horizon. Pentoses and aluminum showed closer relationships in the lower horizons, especially in the Cg. The association between pentoses and aluminum was always prominent in the lower horizons.

Stage B. 0.1N H_2SO_4 Extraction

There was a decrease of the extractable calcium values from the Ah_1 to the Ah_2 horizon, whereas

there was an increase of extractable magnesium values with depth down to the Cg horizon. However, calcium plus magnesium yields showed an increase with depth. The aluminum values showed a decrease but iron values showed an increase respectively with depth. The Ah horizons showed a slightly greater iron plus aluminum yields than the immediately underlying horizons. There was a decrease in the sugar to metal ratios with depth, indicating that, although the Ah horizons yielded higher amounts of extractable sugars than the immediately underlying horizons, there was a closer association with depth. The Ah, horizon showed closest association between pentoses and aluminum, and the Ah2, Bg and Cg horizons showed close associations between pentoses and iron. The closest association throughout the soil was between pentoses and iron which was much greater than between pentoses and aluminum.

Stage D. 0.05M $Na_4P_2O_7 + 1N K_2SO_4$ (pH 7.0) Extraction

A decrease of extractable calcium with depth and of extractable magnesium down to the Ah_2 horizon was observed. However, the calcium plus magnesium yields showed a decrease down to the Ah_2 horizon. The Ah horizons yielded lower values than the underlying horizons for iron plus aluminum yields. However, the metals showed a decrease down to the Ah_2 horizon.

Iron and pentoses in the Ah₁ and Bg horizons and aluminum with pentoses in the Ah₂ and Cg horizons were the dominant associations. A decrease of sugar to metal ratios with depth indicated that there was an increasing association, especially between aluminum and pentoses with depth.

Stage DD. 0.05M $Na_4P_2O_7$ (pH 7.0) Extraction

The increase of extractable magnesium values, though less than in the presence of $K_2 SO_4$, was similarly observed down to the Ah_2 horizon. Calcium yields were conspicuously low. Iron values decreased down to the Ah_2 horizon, while aluminum values showed an accumulation in the Bg horizon within a slight increasing pattern with depth. However, as with the pyrophosphate plus $K_2 SO_4$ extraction, the Ah horizons showed lower values than the immediately underlying horizons. A similar trend of accumulated metals was evident. The Ah horizons showed higher sugar to metal ratios and indicated relationships of less closeness than underlying horizons.

Stage DS. 1N K_2SO_4 Extraction

Equal yields of calcium were observed through—out the profile and a decrease of extractable magnesium down to the Ah_2 horizon. Calcium plus magnesium yields were equal for the Ah_1 and Bg horizons with a decrease in the Ah_2 horizon and the greatest amount was in the

Cg horizon. Aluminum values decreased down to the Ah₂ horizon while similar values for the Bg and Ah₂ horizons were seen in the generally decreasing trend with depth. The iron plus aluminum yields showed a decrease down to the Ah₂ horizon. Again the sugar to metal ratios showed greater values in the Ah horizons indicating greater degrees of association in the underlying horizons. Pentoses and aluminum were associated to the greatest extent throughout the profile.

Stage DDD. Chelex-100 (Na Form) Extraction

Again no calcium was extracted but unlike the other series, a small amount of magnesium was observed. The aluminum values for the Ah horizons were less than the underlying horizons. These patterns do not coincide with those of the pyrophosphate buffer solution. Again, aluminum and hexoses were most closely related in the Ah₁ horizon but aluminum and pentoses were closely associated in the lower horizons. A similar trend for the sugar to metal ratios indicated association to a greater extent in the lower horizons except in the Cg horizon.

SUMMARY AND CONCLUSIONS

Three Gleysols were selected, yielding samples of twelve horizons, which were used to develop the extraction procedure and to study the distribution of hexoses and pentoses and their associations with iron, aluminum, calcium and magnesium.

A summary of the stages of extraction is as follows:

A. Water Soluble Extraction

Percentages of total extractable hexoses and hexose values showed a general decrease with depth in the Langley and Hatzic series. Hazelwood series showed an increase from the Ah₁ to Ah₂ horizons, then decreased in the Cg horizon. No clear overall trend was shown by total pentoses. However, percentages of total extractable pentoses show a general increase with depth.

B. 0.1N H₂SO₄ Extraction

Yields of hexoses were greater than those for Stage A. Ap and Ah horizons yielded very high values in a decreasing trend while percentages of total extractable hexoses tended to increase with depth.

Yields of pentoses were greater than those of Stage A. Although there were accumulations and depletions within the profile, there was an overall, though sometimes slight, decrease of extracted pentoses with depth while

percentages of total extractable pentoses showed an increase.

D. 0.05M $Na_4P_2O_7 + 1N K_2SO_4$ (pH 7.0) Extraction

Although the values for the Ap and Ah horizons were generally higher, percentages of the total extractable hexoses were higher in the Bg horizon than in the Btg horizon. There appeared to be a general decrease in yields with depth, and accumulations in the Bg and Cg horizons were indicated by the percentage values. Both the yields of pentoses and percentage values showed a decrease to the B horizons, then an increase to the Cg horizons.

DD. 0.05M $Na_4P_2O_7$ (pH 7.0) Extraction

This stage displayed greater dispersion than any of the other parallel stages. According to the yields of hexoses obtained for the Langley series, the D extraction was more efficient for the Ap and Ah horizons while the DD extraction was more efficient in the lower horizons. Generally, there was a decrease of extractable hexoses with depth of profile but percentages of total extractable hexoses did not reveal a definite pattern. According to the yields of pentoses, similar amounts or less pentoses were extracted generally except for the Btg horizon.

DS. 1N K₂SO₄ Extraction

This stage always yielded the most clear solution of the parallel stages. The yields of hexoses

were high in the Ap and Ah horizons of the Langley series but greater in the Ap horizon of the Hatzic series. The patterns were all unique to each series and did not show a definite difference between stages D and DD. A general decrease of amounts of pentoses and an increase of percentages of total extractable pentoses with depth was observed. The patterns were again unique for the extraction and again did not show a definite difference between stages D and DD.

DDD. Chelex-100 (Na + Form) Extraction

With respect to chelating properties, both hexoses and pentoses extracted were only able to indicate, at best, only partly similar chelating properties.

F. 0.05N NaOH Extraction

There was a general decrease in hexose yields and percentages of total extractable hexoses with depth of profile with indications of depletions and accumulations with depth. There were no new patterns of yields of pentoses and percentages of total extractable pentoses for the horizons.

G. Final Hydrolysis

Yields of hexoses showed a general decrease with depth but the percentages of total extractable hexoses indicated that the Bg, Btg and Aeg horizons had large amounts of hexoses fixed. The yields of pentoses showed a similar pattern, though there were

marked differences.

The total extractable hexoses, comprising stages A, B, D, F and G showed that the organic A horizons contained the largest amounts of hexoses, while the lower horizons although having lesser amounts, had most of their hexoses fixed.

The total extractable pentoses showed that the organic A horizons contained the largest amounts, with a pentose 'sink' in the B horizons, in a general decreasing pattern with depth.

The hexose/pentose distribution showed a greater accumulation of hexoses than pentoses in the upper A horizons but the reverse in the lower horizons. Of note, is the consistent ratio of less than one for the dilute acid treatment. The general indication is that there is greater retention of pentoses than hexoses with increase in depth of profile.

The comparative distribution of sugars with ions showed the following:

Stage A

The water soluble extraction showed (if valid associations can be made) that hexoses and calcium were associated in Ap and Ah horizons, and the pentoses with iron, aluminum and magnesium in order of decreasing importance, formed the closest associations

in the lower horizons.

Stage B

The dilute acid extraction showed a relationship of pentoses with aluminum and to a lesser extent, iron, throughout the profile.

Stage D

The pyrophosphate and potassium sulphate extraction showed that pentoses or hexoses formed close associations with iron and aluminum in the Ap and Ah horizons. Pentoses appeared to be associated with iron, magnesium and aluminum in the lower horizons.

Stage DD

The pyrophosphate extraction showed close relationships of hexoses with iron and aluminum in the upper Ah and Ap horizons while pentoses were associated with magnesium and aluminum in the lower horizons.

Stage DS

The potassium sulphate extraction showed that pentoses or hexoses were closely associated with aluminum in the Ap, Ah and Cg horizons while pentoses were closely related with magnesium and aluminum in the other horizons.

Stage DDD

The Chelex-100 extraction showed that hexoses were associated with aluminum throughout the soil.

Neither calcium nor magnesium was measureable.

The sequential procedure of extraction might have achieved desired purposes of selectively and progressively extracting organic matter and associated ions in order that the distribution of carbohydrates and ionic constituents can demonstrate profile differentiation. The significance of various distributions lies in the trends which show characteristic patterns. No pretense is given to the exact meaning or implications of these trends but various suggestions have been made in the pertinent sections. As an example, the trends of carbohydrate depletion in the Aeg horizon and fixation of carbohydrates in the B horizons are indicative of a characteristic process which does not necessarily include only carbohydrate and ionic materials but possibly other constituents.

The extraction procedure is biased towards the progressive removal of constituents by solubilization of material in the outer locations with the help of mild mechanical dispersion. It can be argued that complete dispersion facilitates the complete extraction of similar constituents simultaneously. However, this excludes most relevant information concerning the proximity of constituents and possibly information concerning the formation of associations. The greatest

difficulty lies in precisely locating these constituents. However, for the present, an approximation provided by gradual dispersion can suffice.

A study of all compounds, organic and inorganic, remaining after various stages of extraction can be made by completely dispersing each residue and studying for persisting organo-metallic complexation. Leaching studies without dispersion can also provide information as to the processes involved in redistribution of organic and inorganic constituents in the soil by using various solubilizing agents.

Of some significance is the indication by partial study of the distribution of individual sugars, that there are marked differences between extracts. Such information can provide knowledge of the relative distribution of the individual sugars which can be related in part to microbial activity and mineral retention. Similar data concerning all organic constituents would lead inevitably to a greater understanding of the processes which occur in profile differentiation.

LITERATURE CITED

- 1. Acton, C.J., Rennie, D.A., and Paul, E.A. (1963).

 The relationship of polysaccharides to soil

 aggregation. Can. J. Soil Sci. 43, 201-209.
- 2. Alexander, M. (1961). In "Introduction to Soil Microbiology". Wiley, New York.
- 3. Allison, L.E. (1947). Effect of microorganisms on permeability of soil under prolonged submergence. Soil Sci. 63, 439.
- 4. Allison, L.E., Bollen, W.B., Moodie, C.D. (1965).

 In "Methods of Soil Analysis". Part II.

 Agron. No. 9. Am. Soc. of Agron. Madison,

 Wisconsin. 1346.
- 5. Barker, S.A., et al. (1967). Studies on soil polysaccharides. Carbohyd. Res. 5, 13-24.
- 6. Bernier, B. (1958). <u>Characterization of poly-saccharides isolated from forest soils</u>.

 Biochem. J. 70, 590.
- 7. Black, W.A.P., et al, (1957). A preliminary investigation of the chemical composition of Sphagnum moss and peat. J. Appl. Chem. London. 5, 484.
- 8. Bremner, J.M., and Shaw, K. (1957). The mineralization of some nitrogenous materials in soil. J. Sci. Food Agr. 8, 341-347.
- 9. Bremner, J.M. (1958). Amino-sugars in soil.

 J. Sci. Food Agr. 9, 528-532.
- 10. Bremner, J.M. (1965). In "Methods of Soil Analysis". Part II. Agron. No. 9. Am. Soc. of Agron. Madison, Wisconsin. 1346.
- 11. Brink, R. H. Jr., Dubach, P., and Lynch, D.L.

 (1960). Measurement of carbohydrates in soil

 hydrolysates with anthrone. Soil Sci. 89,

 157-166.

- 12. Burges, N.A. (1967). In "Soil Biology". Academic Press, New York. 479.
- 13. Chapman, H.D. (1965). In "Methods of Soil Analysis".

 Part II. Agron. No. 9. Am. Soc. of Agron.

 Madison, Wisconsin. 894.
- 14. Clapp, C.E., Davis, R.J., and Waugaman, S.H. (1962).

 The effect of rhizobial polysaccharides on
 aggregate stability. Soil Sci. Soc. Am. Proc.
 26, 466-469.
- 15. Day, P.R. (1965). In "Methods of Soil Analysis".

 Part I. Agron. 9. Am. Soc. of Agron. Madison,
 Wisconsin.
- 16. Dormaar, J.F., and Lynch, D.L. (1962). Amendments to the determination of uronic acids in soils with carbazole. Soil Sci.Soc. Am. Proc. 26, 251-256.
- 17. Evans, L.T. (1959). The use of chelating reagents and alkaline solutions in soil organic-matter extraction. J. Soil Sci. 10, 110.
- 18. Finch, P., Hayes, M.H.B., Stacey, M. (1967). Studies on soil polysaccharides and on their interaction with clay preparations. Trans. Meet.

 Commns. II and IV. Int. Soc. Soil Sci. (1966).

 19-32.
- 19. Ford, G.W., et al. (1969). Separation of the light fraction from soils by ultrasonic dispersion in halogenated hydrocarbons containing a surfactant. Soil Sci. 20, 291.
- 20. Forsyth, W.G.C., and Webley, D.M. (1947). Studies on the more soluble complexes of soil organic matter. I. A method of fractionation. Biochem. J. 41, 176-181.
- 21. Forsyth, W.G.C. (1950). Studies on the more soluble complexes of soil organic matter.

 II. The composition of the soluble polysaccharide fraction. Biochem. J. 46, 141.

- 22. Foster, A.B., and Webber, J.M. (1960). Chitin.

 Adv. Carbohyd. Chem. 15, 371-393.
- 23. Greenland, D.J., et al. (1962). Organic materials which stabilize natural soil aggregates. Soil Sci. Soc. Am. Proc. 26, 366-371.
- 24. Griffiths, E. (1965). Microorganisms and soil structure. Biol. Rev. 40, 129-142.
- 25. Griffiths, E., and Jones, D. (1965). Microbio-logical aspects of soil structure. Plant Soil. 23, 17.
- 26. Gupta, U.C. (1962). The carbohydrate fraction of soil organic matter. Soils and Fert. 25, 255-257.
- 27. Gupta, U.C., et al. (1963). The characterization of carbohydrate constituents from different soil profiles. Soil Sci. Soc. Am. Proc. 27, 380.
- 28. Gupta, U.C., and Sowden, F.J. (1964). <u>Isolation</u> and characterization of cellulose from soil organic matter. Soil Sci. 97, 328-333.
- 29. Gupta, U.C. (1967). In "Soil Biochemistry".

 Arnold, London. 991.
- 30. Harris, R.F., et al. (1964). Mechanisms involved in soil aggregate stabilization by fungi and bacteria. Soil Sci. Soc. Am. Proc. 28, 529-532.
- 31. Ishikawa, H., Schubert, W.J., and Nord, F.F. (1963).

 Investigations on lignins and ligninification.

 XXVII. The enzymatic degradation of softwood

 lignin by White-Rot fungi. Arch. Biochem.,

 Biophys. 100, 131.
- 32. Jackman, R.H., and Black, C.A. (1952). Phytase activity in soils. Soil Sci. 73, 117.
- 33. Jorgensen, J.R. (1961). "Studies on the Nature and Occurrence of Organic Acids in Soils".

 Ph.D. Thesis, Univ. Minnesota, Minneapolis.

- 34. Keefer, R.F., and Mortensen, J.L. (1963). <u>Bio-synthesis of soil polysaccharides. I. Glucose and alfalfa tissue substrates</u>. Soil Sci. Soc. Am. Proc. 27, 156-160.
- 35. Kononova, M.M. (1966). In "Soil Organic Matter".

 2nd ed. Permagon Press, Oxford.
- 36. Lindquist, I. (1968). On the extraction and fractionation of humic acids. Lantbrukshog-skolans Annaler. 34, 379-389.
- 37. Lowe, L.E., and Turnbull, M.E. (1968). Removal of interference in the determination of uronic acids in soil hydrolysates. J. Soil Sci. 106, 312.
- 38. Lynch, D.L., et al. (1957). The determination of polyuronides in soils with carbazole.

 Soil Sci. Soc. Am. Proc. 21, 160-162.
- 39. Lynch, D.L. (1960). The effect of rose bengal on polysaccharide production in soils. Can.

 J. Microbiol. 6, 673.
- 40. Martin, J.P. (1945a). Microorganisms and soil aggregation. I. Origin and nature of some of the aggregating substances. Soil Sci. 59, 163.
- 41. Martin, J.P. (1945b). <u>Some observations on the synthesis of polysaccharides by soil bacteria</u>.

 J. Bacteriol. 50, 349.
- 42. Martin, J.P. (1946). Microorganisms and soil aggregation. II. Influence of bacterial polysaccharides on soil structure. Soil Sci. 61, 157-166.
- 43. Martin, J.P., and Craggs, B.A. (1946). <u>Influence</u>
 of temperature and moisture on the soilaggregate effect of organic residues. J. Am.
 Soc. Agron. 38, 332.

- 44. Martin, J.P., and Richards, S.J. (1963).

 Decomposition and binding action of polysaccharide from Chromobacterium violaceum
 in soil. J. Bacteriol. 85, 1288.
- 45. Martin, J.P., et al. (1966). Decomposition of iron, aluminum, zinc and copper salts or complexes of some microbial and plant polysaccharides in soil. Soil Sci. Soc. Am. Proc. 30, 196.
- 46. McCalla, T.M. (1943). Changes in the physical properties of straw during the early stages of decomposition. Soil Sci. Soc. Am. Proc. 7, 258.
- 47. McKeague, J.A. (1968). Metal: organic ratios in horizons. Can. J. Soil Sci. 48, 27.
- 48. Mehta, N.C., et al. (1960). Role of polysaccharides in soil aggregation. J. Sci. Food Agr. 11, 40-47.
- 49. Mehta, N.C., Dubach, P., and Deuel, H. (1961).

 <u>Carbohydrates in soil</u>. Adv. Carbohyd. Chem.

 16, 335-355.
- 50. Mortensen, J.L. (1960). Physico-chemical properties of a soil polysaccharide. Trans.

 Intern. Congr. Soil Sci. 7th. Madison. (1960).
 98-104.
- 51. Oades, J.M. (1967). <u>Carbohydrates in some Austral-ian soils</u>. Aust. J. Soil Res. 5, 103-115.
- 52. Percival, E. (1962). In "Structural Carbohydrate Chemistry". 2nd ed. J. Garnet Miller Ltd., London.
- 53. Reese, E.T. (1968). Microbial transformation of soil polysaccharides. Pont. Acad. Sci. Script. Var. 32, 535-577.

- 54. Rennie, D.A., Truog, E., and Allen, O.N. (1954).

 Soil aggregation as influenced by microbial gums, level of fertility and kind of crop.

 Soil Sci. Soc. Am. Proc. 18, 399-403.
- 55. Rovira, A.D., and Greacen, E.L. (1957). The effect of aggregate disruption on the activity of microorganisms in the soil. Aust. J. Agr. Rès. 8, 659.
- 56. Russell, E.W. (1961). In "Soil Conditions and Plant Growth" 9th ed. Wiley, New York.
- 57. Saini, G.R., and MacLean, A.A. (1966). Adsorption-flocculation reactions of soil polysaccharides with kaolinite. Soil Sci. Soc. Am. Proc. 30, 697-699.
- 58. Schnitzer, M., and Delong, W.A. (1955).

 Investigations on the mobilization and transport of iron in forest soils. II. Soil Sci.

 Soc. Am. Proc. 19, 363-368.
- 59. Shorey, E.C. (1913). Some organic soil constituents. U.S.D.A. Bureau of Soils Bull. 88.
- 60. Shorey, E.C. (1930). Some methods for detecting differences in soil organic matter. U.S.D.A. Tech. Bull. 211.
- 61. Sorenson, H. (1955). Xylanase in the soil and the rumen. Nature. 176, 74-75.
- 62. Sowden, F. J. (1959). <u>Investigations on the amounts of hexosamines found in various soils and methods for their determination</u>. Soil Sci. 88, 138-143.
- 63. Sowden, F.J., and Ivanson, K.C. (1962). Methods for analysis of carbohydrate materials in soil. II. Soil Sci. 94, 340-344.
- 64. Stacey, M., and Barker, S.A. (1960). In "Poly-saccharides of Microorganisms". Oxford Univ. Press, London.

- 65. Stevenson, F.J. (1954). <u>Ion exchange chromatography of the amino acids in soil hydrolysates</u>. Soil Sci. Soc. Am. Proc. 18, 373.
- 66. Stevenson, F.J. (1956). <u>Isolation and identification of some amino compounds in soils</u>.

 Soil Sci. Soc. Am. Proc. 20, 201.
- 67. Stevenson, F.J. (1957). <u>Investigations of amino-polysaccharides</u>. <u>I. Colorimetric determination of hexosamines in soil hydrolysates</u>. Soil Sci. 83, 113-122.
- 68. Stevenson, F.J. (1957). <u>Investigations of aminopolysaccharides in soils</u>. <u>II. Distribution of hexosamines in some soil profiles</u>. Soil Sci. 84, 99-106.
- 69. Stevenson, I.L. (1964). In "Chemistry of Soil".

 2nd ed. Reinhold, New York. 242.
- 70. Swaby, R.J. (1949). The relationship between microorganisms and soil aggregation. J. Gen. Microbiol. 3, 326.
- 71. Swincer, G.D., Oades, J.M., and Greenland, D.J.

 (1968a). Studies on Soil polysaccharides.

 I. The isolation of polysaccharides from soil.

 Aust. J. Soil Res. 6, 211-214.
- 72. Swincer, G.D., et al. (1968b). ... II. The composition and properties of polysaccharides
 in soils under pasture and under a fallowwheat rotation. Aust. J. Soil Res. 6, 225-235.
- 73. Swincer, G.D., et al. (1969). The extraction, characterization and significance of soil polysaccharides. Adv. Agron. 21, 195.
- 74. Takizima, Y. (1961). Soil Sci. Plant Nutr. (Japan). 7, 30. In "Soil Biochemistry". Arnold, London. 991.

- 75. Thomas, R.L., et al. (1967). Fractionation and characterization of a soil polysaccharide extract. Soil Sci. Soc. Am. Proc. 31, 568.
- 76. Tracey, M.V. (1950). A colorimetric method for the determination of pentoses in the presence of hexoses and uronic acids. J. Biochem. 47, 433.
- 77. Tropsch, H. (1921). Brenstoff-Chem. 2, 316.
 In "Soil Biochemistry". Arnold, London. 991.
- 78. Waksman, S.A. (1938). <u>Humus</u>. 2nd ed. Williams and Wilkins Co., Baltimore.
- 79. Watson, J.H., and Stojanovic, B. J. (1965).

 Synthesis and binding of soil aggregates as affected by microflora and its metabolic products. Soil Sci. 100, 57-62.
- 80. Webber, L. R. (1965). Soil polysaccharides and aggregation in crop sequences. Soil Sci. Soc. Am. Proc. 29, 39-42.
- 81. Webley, D.M., et al. (1965). A study of polysac-charide-producing organisms occurring in the root region of certain pasture grasses. Soil Sci. 16, 149.
- 82. Whistler, R.L., and Kirby, K.W. (1956).

 Composition and behaviour of soil polysaccharides. J. Am. Chem. Soc. 78, 1755-1759.
- 83. Williams, B.G., Greenland, D.J. and Quirk, J.P. (1966). The adsorption of polyvinyl alcohol by natural soil aggregates. Aust. J. Soil Res. 4, 131-143.
- 84. Williams, B.G., et al. (1967). The effect of polyvinyl alcohol on nitrogen surface area and pore structure of soils. Aust. J. Soil Res. 5, 77-83.