Cellulose–Glucomannans and Plant Cell Wall Structure

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Abstract—The plant cell wall is a composite made of cellulose microfibrils associated with matrix components, among which the xylin family is the most represented. The review made of two types of biological models that are cellulosic (Glucomannans) (G), compositional and compartmental models present, both models are consistent with the need to reproduce and dispersion. Models correspond to structural and functional identities. They are permanent constructions and ensure a role of support and protection. The cell wall and caps are characterized by their ordered and ordered structure. Walls showing a chitosan-like pattern (hemicelluloses, typical of which the emerging in the stacks of microfibrils, combination of colors and orientation of microfibrils). The microfibril is imbricated and can be correlated with a preferential localization of GX in cellulosic microfibrils. They are never consolidated and their development is a complete process. Quagmire mucilage presents a typical hemicellulose organization which is relevant to a true chitosan-like chitooligosaccharide. Cytochemical labelling reveals the close association between cellulose and GX, the latter constituting a strong acidic environment around the mesocellulose. The importance of GX in hemicellulose of cellulosic microfibrils is demonstrated by means of in vitro experiments. Spontaneous self-assembly of isolated mucilage components occurs in cellulose and glucuronoxylans in accordant conditions only if GX is present. In the last section of the paper, the intercalation of free-floating sections of rat microfibrils can be reduced. The model of this paper is the most complete key role as well as the composite cellular and individual (as a minor agent) in the construction of lignification (host structure).

Key words: Cellulose–glucuronoxylan composites, wall cellulose, cell wall structure, self-assembly.

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I. Introduction

A. General

Compared to animal cells, plant cells present an important uniqueness in that they possess a cell wall, i.e., a highly organized extracellular compartment. Although present around each cell, the cell wall forms a continuous and dynamic network through the whole plant body. The body of the plant. During evolution of plant species, the majority of terrestrial higher plants have selected cellulose
as the fibrous reinforcing component for their cell wall. The latter can therefore be considered as a composite in which cellulose, like the steel rods in reinforced concrete, is always associated with a ductile matrix which includes hemicelluloses, pectins and lignins. From early studies, many theoretical wall models have been proposed for cell wall at the molecular level (Kemp et al., 1973; Lamport, 1986; Fry, 1986). The recent improvements of biophysical methodologies and the explosion of new ways of targeting the wall polymers have both contributed to the emergence of new structural models that are more consistent with the physical properties of the wall and its components; these models generally concern the elongating cell walls (McCann and Roberts, 1991; Carpita and Gibeaut, 1993).

In the last few years, literature has emphasized the fact that in a great number of cellulosic cell walls, the wall design is basically a helicoidal pattern that is analogous to a cholesteric order (Rolland et al., 1987, 1989; Emmons, 1988; Satist-Juenemaitre, 1992; Neville, 1993; Wohlers-Arts et al., 1993). In this review, we focus on cellulose/glucono­xylan-based cell walls that present a helicoidal structural order and at the same time express a great variability relating to cell differentiation.

Xylan is a generic name for a series of polysaccharides with a backbone of D-xylopyranose. The xyans are the major hemicelluloses; they are the most common in the majority of angiosperms (Aspinal, 1980). They are found in the secondary walls of dicotyledons (oak), where they account for 20-30% of the dry weight of woody tissues. They also occur both in primary and secondary walls of monocotyledons (cereal endosperms, grasses, maize cobs, etc). In grasses, for example, they can make up 20% of the primary wall, but they will not be dealt with here. In gymnosperms, xyans are less abundant (8%). Xyans also constitute the polymer associated with cellulose in wall-like cellulosic mucilages.

B. Scope of the review

We will first define the chemical background of glucuronoxylan (GXs) that are the most representative and establish the possible linkages that they can on two types of biological models that present both a opposite: hard model and consolidation; soft model and dispersion.

Hard models correspond to stony tissues and related structures. They are permanent constructions and ensure a role of support and protection. Stone cells and endocarps are characterized by thick and ordered secondary walls showing a cholesteric-like pattern (helicity structure). Typical defects emerging in the stacks of microfibrils (disclinations, distortions, saddle-like figures) will be analyzed as a diagnostic of an actual liquid crystal behaviour under mechanical constraints. Chemical analysis and cytochemical labelling will permit relation of the helicoidal motion of microfibrils with a preferential localization of GX.

Soft models correspond to cellulosic mucilages. They are never consolidated and their behaviour is a complete dispersion. Quince mucilage presents a typical helicoidal organization which is representative of a true cholesteric liquid crystal state. The importance of GX in helicoidal ordering of cellulose microfibrils can be demonstrated by means of in vitro experiments. This will be completed with new information on the behaviour of cellulose suspensions deprived of GX that are incapable of re-ordering in acellular conditions.

In the last section of the review, a simple hypothesis will be proposed which emphasizes that GX could play a possible role both in the composite cholesteric assembly (twisting agent) and in the hardening by lignification process (host structure).

II. BACKGROUND: CHEMISTRY OF GLUCURONOXYLAN

A. Chemical structure

GXs are composed of a linear backbone of D-xylopyranose linked in 1-4 and substituted in O3 and/or O-3 by side chains that are short and mobile. The latter are mainly units of O-D-glucuronic acid and also some units of O-acetylglucuronic acid. Additional substitutions can also be found such as residues of D-xylopyranose and D-galacto­pyranose. The xylopyranose residues of the backbone can be acetylated in O-2 and/or O-3 (Aspinal, 1970, 1987; Dupont and Selvendran, 1987; Fry, 1988; Jodhule, 1988; Carpita and Gibeaut, 1993; Marcellin, 1992) (Fig. 1).

B. Affinity of glucuronoxylan with cellulose

The degree of substitution determines the solubility of the GX and also affects the capability of the polymer strand to bind to each other and to cellulose by hydrogen bonding. Poorly branched GX presents a close affinity with cellulose and many hydrogen bonds can be established (Lahav and Ray, 1974; McNeil et al., 1975; More et al., 1986; Neville, 1988a; Jodhule, 1991). Moreover, their conformation in triple helices and their juxtapositional linkages constitute twisted ribbon-like structures that are compatible with the flat ribbons of cellulose (Best, 1977; Atkins, 1991).

Early in vitro experiments of co-crystallization of xyloglucan and cellulose had shown a parallel orientation of both molecular types, the xylosans being oriented inside the intermicrofibrillar spaces (Marchessault and Liao, 1975; Marchessault et al., 1964). More recently it has been elegantly shown that the occurrence of xyloans during polymerization and aggregation of bacterial cellulose modifies the native properties of cellulose, thus indicating the existence of a very strong cellulose/xylan association (Attila, 1991).

C. Glucuronoxylan-lignin linkages

Another important point concerns the linkages that can occur between xylan and cellulose leading to the formation of a lignocellulosic complex. The linkages can be of two types: covalent linkages and ester type between the carboxylic groups of the side chains of glucuronic acid and the hydroxyl groups of lignins and covalent linkages of ether type between the hydroxyl groups of sugars and phenol groups of lignins (Das et al., 1981; Brillouet and Hoebler, 1985; Leavy et al., 1983; Fengel and Wegener, 1983).

D. Accessibility to xylanases

The high complexity of xyans is reflected in the battery of enzymes that are required to act in combination for their hydrolysis. A multiplicity of exo- and endoxylanases are secreted by fungi, bacteria, yeasts and many microorganisms of the rumen (Brillouet and Hoebler, 1985). In fact, the action of these enzymes depends on the degree of polymerization, the rate of substitution and the type of linkages existing between the side chains and the main backbone of the xylan. At present, industrial applications for xylanases are more and more evident; they concern cellulosysis, upgrading of animal feeds and the food processing industry. The specificity of xylanases and the possibility of obtaining enzymes that could be suitable for industrial scale applications is the goal of many laboratories (Slade et al., 1989; Debere et al., 1990; Nishitani and Nevis, 1991; Sinner et al., 1991).

Concerning the GX, Nishitani and Nevis (1991) have purified a novel glucuronoxylanase that recognizes glucuronolyl moieties inserted as monomeric side chains along the xylan backbone and hydrolyses the β(1-4) xylosyl linkage of the adjacent unsubstituted xylosyl residues (Fig. 2). This enzyme demonstrated the occurrence of repeating structural units in the GXs and thus appears as a new powerful tool for exploring the structure of one of the major constituents of the cell wall.
inner side of the primary wall according to a relatively uniform and synchronous process. In the case of stone cells of fruits, the future seeder forms isolated nodules that develop within the pulp of the fruit. The first cells engaged in the seeder constitute developing centers by differentiation of continuous cells. As for endocarps, the secondary wall is deposited on the inner side of the primary wall, but the process is less synchronous inside each nodule. For both, an intense activity gives rise to heliocidal patterns, according to a sustained and typical cell wall being extremely reduced at maturity (Reis et al., 1993). Stony cells and stony tissues walls that appear particularly favorable for a study of wall textures.

B. Molecular partners: molecular shape and interrelations

The results of analysis of the wall fractions are very similar for both endocarps and stone cells. The final intensively lignified. The treatment with Na chlorite lignification is maximum in the middle area and the Na chlorite treatment produces the complete solubilization of the cementing material and the dispersion of the cells. The ghosts of secondary walls deprived of lignin remain compact with an undistorted shape and constitute a very homogeneous fraction (Fig. 4).

A subsequent extraction of the wall ghosts with KOH yields a soluble fraction corresponding to 28-32% of the wall dry weight. The chemical analysis of neutral sugars and uronic acids shows that this fraction contains almost pure GX (Fig. 5). Among the neutral sugars, xylose is massively dominant: 95-96% in both endocarps and stone cells. Arabinose, which is often a noticeable component of the xylans is here very low (1-2%). No mannose and a low quantity of glucose (1.5-2.5%) are detected, though mannose or glucuramin could be a major component of the matrix in certain secondary walls. A few unidentified sugars are sometimes detected in very small amounts. They are likely to correspond to acetylated or methylated residues. The amount of uronic acid represents 16-23% of the whole fraction, corresponding to a ratio of one acid to five to seven xylose residues.

Molecular shape of isolated GX can be visualized by electron microscopy using negative staining (Fig. 6). The fractions have a microfibrillar aspect as previously described in wood (Fengel and Wegener, 1984; Vian et al., 1986). The fractions are homogenous, whatever their biological origin and show thin and still rodlets. Their dimensions are mean 30-40 nm in length and 3-3 nm in width. Rodlets are tightly packed along their whole length forming spindles either isolated or joined by their ends. Cytological checking of isolated GX can be made by using specific probes. They can be xylanases labelled with colloidal gold particles (Vian et al., 1986). They can also be antixylan antibodies that are directly revealed onto the fraction by negative staining without any gold labelling (Fig. 6D). The residual fraction following alkali extraction represents 28-35% of the bulk of the dry weight. It is the cellulosic residue. Hydrolysis of this fraction always yields a more than negligible amount of xylose and uronic acid residues, even after additional treatments with alkali. This reveals that GX is tightly bound to the cellulose moiety. This tight association between GX and cellulose microfibrils has been confirmed by a cytochemical approach. For example, the incubation of endocarps with cellulase microfibrils has been observed by negative staining along the isolated groups, revealed a constant labelling along the isolated microfibrils (Reis et al., 1992). The tight interaction between GX and cellulose confirms the occurrence of a between GX and cellulose, confirms the occurrence of a between GX and the microfibrils that is likely to play an important role, both in assembly and consolidation of the wall (see Section IV for detailed discussion).
C. The helicoidal wall pattern and its consolidation

Stone cells and stony tissues represent amplified multilayered structures. The morphogenetic activity of the wall construction is sustained and the layers are progressively counted in tens (Roland et al., 1987). The final number is variable, the maximum being up to 80–100 concentric layers, giving a monotonous and uniform system. Each layer is formed of superimposed series of nested arcs (Fig. 7A). The arc pattern is cancelled or inverted when observed at various angles of a goniometric stage. Each arc is formed of successive sheets of microfibrils arrayed in parallel and in which the microfibril orientation regularly and progressively changes; the visible alternation of dark and light electron density is due to the orientation of the microfibrils related to the sectioning plane. A full 180° rotation constitutes one layer and this repetition corresponds to the basic structural periodicity of the wall, which is typically a helicoidal structure (Bouligand, 1978; Neville and Levy, 1985; Roland et al., 1987; Satiat-Jeunemaitre, 1992). According to the orientation of the section, the width of the arcs is variable.

Characteristic deviations from the helicoidal array occur during the wall construction. They can correspond to variations in the helicoidal rhythm or to variations expressed by intrawall distortions and disclinations in response to topological constraints.

(a) The endocarp of Prunus sativum shows a gradual change from a monotonous to a bimodal helicoidal. The outer part of the tissue is progressively built up, giving rise to monotonous wall pattern as described earlier. Towards the inner part, the regularity disappears and is replaced by series of arcs, the width of which varies periodically (Fig. 7B). There is an alternation of wide and single arcs followed by narrow and repeated arcs (2–4). This corresponds to a bimodal pattern of construction. In many examples of cell walls, the different variations which can occur (sporadic bursts, blockages) prove the flexibility of the helicoidal pattern (Roland et al., 1987; Satiat-Jeunemaitre and Mosiniak, 1992).

(b) The stony tissues reveal the large capacity of the helicoidal morphogenesis to adjust itself under the influence of two topological constraints: the spherical or polyhedral shape of the cell and the numerous pit canals which maintain the symplastic transport and produce a recces during the construction of the wall. The axes of the helicoidal point toward the center of the spherical sphere. The layering around the cell lumen implies that much of the internal stresses are relieved by a lateral flow of the constitutive molecules. Warps, buckles and disclinations are encountered that absorb the stresses and second cause of constraints and defects is the presence of pits. They behave as many gaps open through the layering and the microfibrils are more or less propagated to the neighbouring layers. The emerging defects are distortions and disclinations such as saddle-like disclinations (Fig. 7C), which are relevant to a liquid crystal behaviour. When submitted to strains, actual cholesteric mesophases expose characteristic deviations from the perfect helical array. The defects that develop in the biological structure to relieve the strained body give information on the liquid crystal phase that could operate during lamellogenesis (Mazur et al., 1982, 1989). The endocarp shows a type of spherical twist in which the helicoidal expression is maintained owing to the accumulation of multiple microstructural defects. The occurrence of these defects strongly suggests that the wall ordering might be relevant to a liquid crystal assembly that is later strongly consolidated by lignification (Reis et al., 1992; Roland et al., 1993, discussion in Section IV).

In such constructions, immunocytochemical labelling reveals the overall distribution of GX, without any preferential and differential localization (Fig. 8). The confirms that GX is regularly distributed within the wall thickness of the wall. The lignification of these wall occurs relatively early, before the construction of the secondary wall is over and it proceeds slowly (Smilga, 1953, 1954). As for the lignified cells of wood, lignification starts classically in the middle lamella and then the polyphenolic percolation progresses within the wood, i.e. following the lamellation and helicoidal orders (Fig. 9). The incrustation and reticulation provide a centripetal densification and hardening of the helicoidal pattern.

D. The case of hardwoods

GX is the major hemicellulosic component of the secondary cell walls of dicotyledonous woods (hardwoods). Because of its great technological interest, wood has been extensively analyzed (Timell, 1965; Niedzwipski and Marchessault, 1972; Aspinall, 1980; Roland and Mosiniak, 1983; Fengel and Wegener, 1984 and bibliography therein; Vian et al., 1986; Dupont and Selvendran, 1987; Joseleau, 1991; Vian et al., 1992).

Contrary to stone cells and endocarps, hardwoods constitute a very heterogeneous material, both from biochemical and cytochemical viewpoints. At the ultrastructural level, the texture of secondary walls appear highly defined in space and is variable according to the specimen, depending on species, cell type, age, season and conditions of growth and development. Despite the encountered diversity, it has been proposed that the orientation of the cellulose framework could result in a common mechanism related to GX localization and the building up a basic helicoidal pattern (Rolande et al., 1983; Vian et al., 1986; Neville, 1988a).

In fibers and tracheids, for example, it has been known for long that the structure is typically three-layered (S1–S2–S3 layers). Mild extractions and stereoelectron microscopy have revealed the fine details of fibrillar architecture. It was shown that the classical S1 and S2 (or S2 and S3) layers are connected by transition zones in which a progressive rotation of the microfibrils correspond to a typical semi-helicoidal motion (Neville and Levy, 1985; Roland et al., 1987). Preliminary results that indicated a concentration of GX appearing in transition zones between S sheets (Meier, 1961; Parameswaran and Last, 1982; Fengel and Wegener, 1984) were confirmed in...
of the secondary wall deposition, i.e. in the area where slight oscillations in orientation of the microfibrils intervene (Vian et al., 1992).

Thus, the complex case of secondary walls of wood is interesting since the helicoidal motion is intermittent and not regular. The preferential localization of GX in wall domains where the cellulose changes in orientation leads to the hypothesis that GX could play a key role in the twisted morphogenesis of the cell walls (see Section V for discussion).

Another outstanding feature is the close correspondence between the distribution of the xylan shown with affinity cytochemical labelling and the progression of lignin within the secondary wall (Catesson, 1983; Saka and Goring, 1983; Eriksson et al., 1988; Vian et al., 1992). This leads to a search for possible mechanisms by which localization of polysaccharide polymers like GX and cellulose might mediate localization of lignin (see Section V for discussion).

**IV. SOFT MODEL, ASSEMBLY AND DISPERSION: CELLULOSE MUCILAGES**

Contrary to hard models, the cellulosic mucilages constitute systems totally different in appearance. They are never consolidated (no chemical bonds, no lignin incorporation), they are not permanent since their behavior is a complete dispersion. At first sight it is difficult to consider these mucilages in terms of the 'classical' cell wall. According to Schnepf and Deichgräber (1983) and Abeysekara and Willison (1988) periplasmic mucilage is in 'a broad sense' a form of secondary wall if it contains cellulosic microfibrils. This is typically the case of the seed mucilage of quince: it is a true cellulose/GX composite, it presents a basic helicoidal structure and provides an extracellular matrix built up with the wall synthetic machinery. We will see later that the fluid characteristics of the model confers exceptional potentialities for experimentation that allow a better insight in knowledge of wall morphogenesis.

**A. Origin and chemical characteristics of quince seed mucilage**

An abundant mucilage is produced by the epidermal cells of seeds of quince (Cydonia oblonga L.). It is deposited by the cell into an extended periplast and accumulates beyond the plasma membrane against the primary wall, forming a perimucilage deposit according to the terminology proposed by Abeysekara and Willison (1988). During its development, the perimucilage compresses the protoplasm at the basis of the cell.

When the seeds come into contact with water, the mucilage is released from the epidermis cells. By hydrated mucilage is released from the epidermis cells. By hydrated mucilage is released from the epidermis cells. By hydrated mucilage is released from the epidermis cells. By hydrated mucilage is released from the epidermis cells. By hydrated mucilage is released from the epidermis cells.
and Enmen, 1969; Willison and Abeysekera, 1985; Reis et al., 1991). Figures 13A and B show the microfibrils to be ribbon-like, thin (ca. 2 nm in diameter), very long and rigid (few ends are visible). They are spaced, nonaggregated in bundles and constitute a tangled network, without any visible order.

The chemical composition of the mucilage reveals the occurrence of two major types: cellulose associated with a (4-O-methyl-β-D-glucurono)-α-D-xylan containing an exceptional high content of uronic acid groups (1 per 2 xylose). The 4-O-methyl-α-D-glucopyranosyluronic and α-D-glucopyranosyluronic acid groups are linked to positions 2 of α-(1-4)-β-D-xylan. The O-acetyl groups are linked to the β-D-xyllopyranosyl residues (Lindberg et al., 1990).

B. Cholesteric-like organization

In non-mature fruit, the mucilage stored within the seeds fills a large space between the plasmalemma and the wall of the epidermis cells. It appears as banded material when viewed by bright-field microscopy (Willison and Abeysekera, 1988). By electron microscopy, the banded material is found to be composed of a typical helicoidal array of microfibrils (Fig. 10). In the bulk of the ordered mucilage, one can count several tens of arcs variable in width. These variations in the width of the arcs correspond to a variable number of microfibrillar lamellae per arc (Abeysekera and Willison, 1990).

The helicoidal organization of the mucilage is relevant to a true cholesteric liquid crystalline state (Abeysekera and Willison, 1988). As in any liquid crystalline order, many imperfections are visible. Near typical series of nested arcs, more or less organized domains are found: amorphous blocks without apparent order, blocks with unidirectionally-oriented microfibrils, regions with sinuous arcs (Fig. 10A), local whirling patterns, foliated and spirals, the most organized regions always being within the middle zone of the periplasmic mucilage (Abeysekera and Willison, 1990; Reis et al., 1991). All these aspects indicate the fluidity and the flexibility of the helicoidal construction. The helicoidal pattern is adapted to the shape of the cells and lateral margins near the adjacent radial cell walls often clearly have a sharp saw-tooth appearance (Fig. 10B). These serrated margins correspond to a regular spatial arrangement of the edges of the microfibrillar lamellae, closely pressed against the cell walls.

C. Glucuronoxylan as a coat onto cellulose microfibrils

As seen for the stony tissues, cytochemical labelling reveals the spatial relations which exist between the cellulose and GX. Different tests can be used to detect the components: for example, cellobiohydrolase-gold complex for cellulose (Reis et al., 1991). In the quince mucilage, the gold particles are fully extended, occasionally dispersed in the suspension. An agglomerate of gold particles can be recognized on the surface of the mucilage (Fig. 11). Gold particles are mainly seen aligned along the microfibrils, underlining the arrayed pattern with a regular density. This again reveals the close association between cellulose and GX, the latter constituting a charged coat around each microfibril. Few particles are seen between the microfibrils, revealing a cloud of GX not directly bound to microfibrils but which forms a strong acidic environment around cellulose.

D. Reassociations in acidic conditions and importance of glucuronoxylan

The cholesteric state implies the occurrence of self-assembly processes. Quince mucilage is an exceptional material which allows the isolation of the mucilage components in their native state and to follow their possible capacity for spontaneous reassociation in acidic conditions.

Following the wetting of the mature seeds, the mucilage is hydrated and spewed out. Cellulose microfibrils progressively disperse and become scattered. When the mucilage is totally hydrated, a suspension is obtained that appears isotropic when observed in polarizing microscopy. The mucilage makes a disperse suspension which cannot spontaneously sediment in water under normal gravity. By high centrifugation followed by a gentle dehydration, a concentrated and viscous suspension is obtained which is highly anisotropic (Reis et al., 1991). It shows birefringent domains with a layered organization, indicative of a liquid crystalline order. Figure 12 shows the reassembled suspension by electron microscopy. The whole suspension appears stratified, with juxtaposed domains. Many regions present a liquid crystal-like organization: alignments of microfibrillar subunits, sigmoidal arc patterns, all aspects that are similar to the in situ construction. Some regions appear as blocks of unidirectionally-oriented microfibrils and others as blocks of nonordered material. Within the bulk of the mucilages many nodules are seen, around which order takes place; the system is typically polydomain.

In a liquid crystal, constraining surfaces are necessary to make planar monodomain helicoids. When not suitably constrained, a polydomain cholesteric phase is able to form (Neville, 1988b). Generally, in plant cell walls, synthesized wall polymers are trapped in the periplasm which represents a narrow gap between the cell wall, and the cell wall. These initial constraining surfaces are the requirement for monodomain helicoid assembly. In the enlarged periplasm of quince epicarp cells, the initial constraining forces were weaker. In this case, orderliness seems to begin in the center, independently of plasma membrane and primary cell wall, constituting nodules of physical nucleation rather than layer-by-layer directed assembly (Abeysekera and Willison, 1990). As the secreted components accumulate, the increasing constraining forces give rise to increased constraining surfaces and the whole tends to a monodomain helicoidal construction, with many imperfections. In cellular spontaneous assemblies which constitute an extreme case, constraining cellular surfaces are absent; experimentally induced constraints are the centripetal forces and controlled dehydration which restrict the total volume of the suspension. However, physicochemical conditions are expected compared to the in situ state.

This leads to a typical polydomain construction.

The intervention of GX in the spontaneous reassociation can be shown by experiments in which GX is extracted with trifluoroacetic acid (TFA). Biochemical analyses have allowed us to follow the progressive extraction of the GXs (data not shown). When the uronic acid ratio is less than 10%, the mucilage spontaneously flocculates in the suspension. Figure 13 compares the flocculal and the suspensions state and to follow their possible capacity for spontaneous reassociation in acidic conditions. Hard and soft models show a convergence in composition and in morphogenesis. Both systems are cellulose-
GX based composites with a basic helicoidal structure, but their behaviour and fate are different: support and consolidation on the one hand, hydrophilicity and dispersion on the other hand, depending on the cell differentiation program of each cell.

The morphogenesis of cell wall is a complex, multistep process. It has given rise to an abundant literature that is controversial in certain aspects. It is out of the scope of this paper to go into too many details. The reader is referred to reviews (Delmer, 1987; 1991; Preston, 1988; Stachelin et al., 1988; Hayashi, 1989; Varner and Lin, 1992). The debate is often biased towards the cellulose side in order to explain some of the arising questions, which has led to only partial interpretations. Here, we want to discuss the wall morphogenesis by quantifying the role of noncellulosic polymers, that represent more than 50% of the bulk of the wall.

A consensus exists on what concerns the biosynthesis and crystallization of cellulose. The involvement of terminal complexes (linear terminal complexes or rosettes) has been widely documented from many cell types (Giddings et al., 1980; Montezinos, 1982; Itoh and Brown, 1988; Emmons, 1991). Cellulose is synthesized on the plasma membrane via the cellulose-synthases and forms crystalline microfibrils in the periplasmic zone that are parallel to each other. The discussion is more complex concerning the subsequent orientation of the sheets of microfibrils in hierarchical three-dimensional constructions that represent multiphasic structures. It has often been claimed that this orientation results neither from a directed assembly driven by some type of cellular mechanism. The orientation can be driven by the orientation of the terminal complexes themselves or by terminal complexes directed by underlying cytoskeleton elements (Roberts et al., 1985; Giddings and Skotheim, 1988; Itoh, 1989; Seagull, 1991; Emmons 1988, 1991), a set of microtubule channel model (Neville, 1993). The mechanism, although compatible with many situations, does not explain everything and in particular does not explain how to interact with helicoidal cell walls. As recently discussed by Neve (1993) the existence of helicoidal structures requires somewhere a rotary mechanism likely located in the periplasmic domain where the cellulose microfibrils could be oriented according to self-assembly mechanisms. We want to emphasize that particular point, insinuating the possible role of GX in the step of ordered helicoidal assembly. We will see that GX could also be implicated in subsequent step that can lead to the consolidation of the composite.

A. Glucuronoxylan, a twisting agent in the composite assembly

Our discussion leads to the idea that helicoidal walls, whatever their construction, hard or soft, are the result of a self-assembly process and pass through an actual liquid crystal state, if only briefly. The question remains of what is the exact role of GX. The intervention of GX in the cellulose organization of the cell wall has already been postulated by numerous authors: ‘helper molecule’, ‘twisting molecule’, ‘molecular organizer’ or ‘lubricating layer’ (Nieduszinski and Marchessault, 1972; Vian et al., 1986; Neville, 1988a; Roland et al., 1989; Abeysekera and William, 1990; Reis et al., 1991). To make a long story short, one can try to select from the literature and from the above data the outstanding arguments that emphasize the key role of GX in cholesteric assembly.

Correspondence glucuronoxylan/helicoidal construction. This is shown by in situ labelling experiments. They reveal a regular distribution of GX in monotonous helicoids and the occurrence of preferential distribution of GX in helicoidal regions of intermittent systems (Neville, 1988a; Vian et al., 1987, 1992; Fig. 14), thus indicating that GX could direct the cellulose microfibrils in a helicoidal array.

Capability of spontaneous reassociation of native suspensions. The suspensions of cellulose/GX carry the information necessary to reassociate into a quasi-order in a composite acellular system. This capability manifests only when GX is present. As early as 1950 Mühlethaler had postulated that the solubility of cellulose containing.
mucilage was due to the existence of polyuronic acids and cell wall micelles. 

The cellulosic-GX composite is a charged and highly anisotropic construction. It forms a sort of host structure for lignin precursors. The incrustation by lignin can be compared to a form of intercalation chemistry, which occurs in low-dimensional solids as documented for advanced industrial material (Rouxel et al., 1986 and 1989). In a low-dimensional solid, the surface is considerable and exogenous monomers can be intercalated and activated by the cellulosic-GX composite. This is a possible way in which the cellulosic-GX composite could be formed and the cohesion within the system strengthened (Fig. 15B). In this hypothesis, the GXs have a key-position in the successive steps of the morphogenesis of lignified secondary walls.

### CONCLUDING REMARKS

To build a cell wall is not an easy task for a plant cell. Except cellulosic, the wall is assembled from polymers that are synthesized in the cell, secreted in a soluble form and assembled extracellularly in a composite forming a large and insoluble network. As emphasized recently by Carpita and Gibeaut (1993), the wall construction needs an 'automated machinery consisting of hundreds of enzymes involved in assembly, cross-linking, turn-over of polysaccharides and proteins and formation of lignin.' In this paper, we have focussed on cellulosic-xylan-based cell walls. Though studied from various types of biological specimens—hard or soft or soft model—many of them share in common a helical organization, even if this organization is sometimes difficult to expose. Among the events that lead to the construction of these walls we have focused on those that imply self-assembly processes, i.e. intracellular and extracellular remodeling. We have considered the self-assembly events happen: at a molecular level, at the level of supramolecular assemblies and at the level of lignified cell walls.

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**Fig. 16.** A hypothetical scheme for the construction of cellulosic-glucuronoxylan helicoidal cell wall. **1.** Cellulose polymers: **A.** Xyloglucans; **B.** Cellulose microfibrils. **2.** Cellulose crystallinity; **3.** Helicoidal assembly. **PM** is a periplasmic space; **SV** is a secretory vesicle; **CK** is a cellulose synthetase. **GX** is a glucuronoxylan. **TC** is a cellulose synthetase. **D** is a dormant cell. **4.** Gyrating agents.

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