IMPROVING STIFFNESS OF LIGNOCELLULOSICS THROUGH CELL WALL MODIFICATION WITH CHITOSAN-MELAMINE CO-POLYMERS*

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ABSTRACT

Chemical modification with chitin- and chitosan-hexamethyl methylol melamine (HMMM) co-polymers was investigated for improving the stiffness of lignocellulosic materials. Chitin and chitosan were converted by chemical means to low molecular weight oligosaccharides with molecular weight profiles suitable for penetration of lignocellulosic cell walls. The oligomers were reacted under controlled conditions with hexamethyl methylol melamine (HMMM) to produce aqueous formulations of oligosaccharide bonded to HMMM, the "pre-polymers". The chitosan oligomers reacted with HMMM to produce, on condensation polymerisation, a water-insoluble polymer in high yield (69%), whereas the chitin oligomer HMMM condensation reaction gave poor co-polymer yields (28–34%). The yield of co-polymer from the condensation polymerisation reaction was critical to the success of the cell wall modification in improving stiffness.

Pinus radiata D. Don veneers were treated with chitin and chitosan oligomer HMMM formulations to average dry weight percentage gains of 69% and 57% respectively. No improvement in veneer stiffness was obtained with the chitin oligomer HMMM treatment, whereas the chitosan oligomer HMMM treatment resulted in an average veneer stiffness enhancement of 20%. There was a linear relationship between the level of stiffness improvement and the degree of co-polymerisation of the oligomers with HMMM. A threshold of greater than approx. 30% co-polymer yield was necessary before any improvement in veneer stiffness was observed. Polysaccharides with a β-(1→4) configuration, such as chitosan, therefore offer potential for lignocellulosic stiffness property modification.

Keywords: wood; veneers; modulus of elasticity; chitosan; melamine; polymers.

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INTRODUCTION

Wood is a natural, accessible, and generally easy-to-use material for construction and engineering products. The properties of wood materials which are important in such applications are strength, stiffness, and stability with certain performance criteria required for utility. These properties are related to the wood physical and supra-molecular structure, and in particular to the cell wall structure, density, and also to chemical bond strength within and between the molecular structures of the cell walls.

Much of the currently New Zealand plantation-grown *P. radiata* wood material has an average modulus of elasticity (MoE) of approx. 7 GPa, with a high degree of variability (Kininmonth & Whitehouse 1991). In the manufacture of laminated veneer lumber (LVL) the average stiffness desirable for a market-competitive product is 14 GPa. In order to produce high-performance laminated veneer lumber from *P. radiata* from the New Zealand resource, a manufacturer would be required to use high-stiffness veneers for the outer laminates, leaving low-stiffness *P. radiata* for the less-demanding inner veneers to give an average 14 GPa stiffness. While this appears to be a ready solution to the problem, other problems such as differential gluability and stability (Marra 1992) can result when mixing veneers from different species. The impact of these problems on product quality has generally discounted this approach.

An alternative approach has been to modify the stiffness properties of *P. radiata* veneers through physical and chemical manipulation of the base wood material. Veneer stiffness can be enhanced by several techniques including mechanical compression alone (Li & Cown 2002) or used in combination with heating or steaming (Navi & Girardet 2000; Rowell *et al.* 2000) and treatment with resins (Inoue *et al.* 1993; D.Cown, M.Hedley, J.Li, C.McIntosh, D.Gaunt, R.Franich, & D.Page unpubl. data) to fix the compression. Using compression is known to cause deformation of the cell wall (Ando & Onda 1999) and shearing of the lignocellulosic polymers when the load is applied, resulting in bringing the polymers in the wood supra-molecular structure into closer proximity and consequently tighter and/or more dense inter-molecular hydrogen bonding. Li & Cown (2002) obtained average stiffness improvements of 60% over that of the controls by compression of *P. radiata* veneers to 40% strain. The main disadvantage of use of compression alone for increasing material stiffness has been the propensity of the material to reverse the deformation, to "spring back", and thereby lose stiffness as a result of adsorption of atmospheric moisture.

Heat and steam treatments in combination with compression can permanently fix compression and prevent "spring back", possibly through a mechanism of lignocellulosic de-polymerisation and subsequent condensation and cross-linking of wood components (Rowell *et al.* 2000). The lignin-carbohydrate complexes of

the cell wall provide stiffness of the lignocellulose wall composite through both covalent and hydrogen bonding (Koshijima & Watanabe 2003). Modification of these complexes through heat and compression can cause self-adhesion or self-crosslinking in the cell-wall microfibrils and matrix (Inoue *et al.* 1993; Dwianto *et al.* 1999; Navi & Girardet 2000; Rowell *et al.* 2000).

Cown and co-workers (unpubl. data) have shown that compression of *P. radiata* veneers to 40% strain and subsequent fixing of the compression with a low-molecular weight phenol formaldehyde (PF) resin polymerised *in situ* resulted in a stiffness increase of over 100%. However, a serious disadvantage of the chemically modified veneer was poor gluability using conventional wood-working adhesives. This was most likely because the non-polar nature of the introduced polymer was incompatible with the polar nature of wood-working adhesives (Marra 1992). Because of its poor gluability, the compressed veneer composite had little value for the manufacture of high-performance laminated veneer lumber (Cown unpubl. data).

As an alternative approach to lignocellulose stiffness property modification, treatment of *P. radiata* veneers with polar carbohydrate biopolymers was hypothesised to have the potential to improve stiffness through "intensification" of hydrogen bonding within the composite structure. By creating linkages between the cellulose fibrils and the matrix of lignin-carbohydrate complexes, the extent to which these wood components can slip in response to an applied force could be limited. This effect has already been demonstrated with the Indurite™ process for wood hardening (Fibre7 2004; Franich & Anderson 1998). This process, achieved by condensation polymerisation of maltodextrin (oligomers of starch) with a methylol melamine, was found to improve *P. radiata* lignocellulose material stiffness by an average of 12% in addition to wood hardening at weight percentage gain (wpg) of approx. 40%. An estimated 5–10% of this gain was deposited in the cell wall because of the low molecular weight of part of the maltodextrin components which could diffuse into the wall spaces. In contrast to the phenol formaldehyde resin treatment result, the Indurite™ processed veneers showed significantly improved gluability (Franich & Anderson 1998), probably also due to the increase in hydroxyl group, and therefore hydrogen bond density, at the glued surface. Therefore, modification of *P. radiata* veneer stiffness by deposition of carbohydrate-derived polymers into the cell wall had potential for manufacturing of laminated veneer lumber.

Starch is a polymer of α-1→4-linked glucose units and has tensile stiffness of 1 GPa while cellulose, consisting of β-1→4 linked glucose units, has a tensile stiffness of 138 GPa (Nishino *et al.* 1999). Theoretically, therefore, by substitution of cellulose oligomers for the maltodextrin component of the Indurite™ composition and treatment of veneers with this to form a co-polymer composite within the cell

wall, an enhanced stiffness would result from limiting the stretching of the modified veneer material when a bending force is applied. However, preparation of large quantities of β-1→4 glucan oligomers by hydrolysis of cellulose is not straightforward (Isogai & Usuda 1991). Alternative β-1→4 linked polysaccharides, chitin (poly-*N*-acetylglucosamine), and chitosan (deacetylated chitin) have superior stiffness (41 GPa and 65 GPa respectively (Nishino *et al.* 1999)) to that of starch. Conversion of chitin and chitosan polymers to low molecular weight oligomers can be carried out using enzymatic and acid-catalysed hydrolysis (Yalpani & Pantaleone 1994; Blumberg *et al.* 1982), and by nitrous acid deaminative polymerisation, a process specifically for chitosan (Allan & Peyron 1997). Therefore, it was hypothesised that treating lignocellulose material with oligomers of chitin and chitosan and co-polymerising these with methylol melamine at equivalent treatment loadings to that of the Indurite™ process, should, in theory, lead to greater average stiffness improvement than that achieved using maltodextrins as components in a co-polymer containing melamine cross-linker.

EXPERIMENTAL

Materials

Chitin and chitosan were purchased from Sigma-Aldrich. Hexamethyl methylol melamine (HMMM) was supplied by BASF. *Pinus radiata* veneer (3 mm thick at 12% moisture content) was used as the lignocellulosic material and was obtained from Carter Holt Harvey Ltd, Rotorua, New Zealand.

Chemical Hydrolysis of Polysaccharides

Chitin oligomers were prepared (Blumberg *et al.* 1982) by stirring chitin (500 g, added in portions over 45 min.) suspended in concentrated HCl (37%, 1.6 L) at 40°C for 2 hr. The hydrolysis mixture was then neutralised with 30% aqueous NaOH and filtered on a Buchner funnel to removed precipitated NaCl. The filter cake was washed with water (100 ml) and washings were combined with the filtrate. The filtrate was concentrated to dryness by evaporation under reduced pressure to give a product consisting of chitin oligomers and residual NaCl. The chitin oligomers were dissolved in 9:1 MeOH:water (3.5 L) and decanted from insoluble NaCl. The supernatant was concentrated to dryness by evaporation under reduced pressure and the product redissolved in 9:1 MeOH:water (100 ml). The product was decanted from additional insoluble NaCl and concentrated to dryness to give chitin oligosaccharides (330 g), which were largely free of NaCl, although the residual salt concentration was not determined.

Chitosan (100 g) was suspended in 0.1 mol L^{-1} HCl (2 L) which had been heated to 50°C, $NaNO_2$ (13.8 g, 0.2 mol) in water (40 ml) was added dropwise, and the

mixture was stirred at 50°C for 4 hr. The mixture was neutralised with Amberlite IRA-400 ion-exchange resin (OH^- form). The aqueous solution was decanted and filtered from the resin, the resin was washed with water, and the filtrate and washings were combined and concentrated to dryness by evaporation under reduced pressure. The product was dried overnight in a vacuum oven at 35°C to give chitosan oligosaccharides (100 g).

Electrospray Mass Spectroscopy

Direct infusion electrospray ionisation mass spectroscopy (ESI MS) spectra were obtained on a Thermofinnigan LCQ DECA XP mass spectrometer operating in positive ion mode using 0.1 mg/ml aqueous solutions of chitin and chitosan oligosaccharides. The chitosan oligomer solutions were acidified to pH 4 to assist ionisation.

Condensation of Chitin/Chitosan Oligomers with HMMM

Chitin and chitosan oligomers were reacted with HMMM by stirring aqueous solutions at 50°C for 1–4 hr in the presence of toluene-*p*-sulphonic acid (0.2%) and boric acid (0.45%) as catalysts. The proportions by mass of the various components of the pre-polymerisation formulation were water (56%), chitin/chitosan oligomers (28%), HMMM (9%), and ethanol (6%). The success of the pre-polymerisation reaction was evaluated by determining the degree of co-polymerisation achieved, as indicated by the yield of water-insoluble product formed on heating.

Determination of Degree of Co-polymerisation

Chitin/chitosan oligomer HMMM "pre-polymer" formulations were heated to dryness at 65–70°C for 2 days. This material (1 g) was accurately weighed into pre-weighed test-tubes, water was added (10 ml), and the mixture was agitated in an ultrasonic bath at 50°C for 30 minutes. The contents of the test-tubes were transferred to centrifuge tubes and centrifuged at 3000 rpm for 15 minutes. The supernatant was decanted and the insoluble polymer oven-dried and weighed. The mass of water-insoluble material as a percentage of the original mass was calculated to give a measure of the degree of co-polymerisation.

Veneer Treatments

Pre-conditioned (12% moisture content) and pre-weighed *P. radiata* veneers (10 replicates per treatment) were treated with chitin oligomer HMMM and chitosan oligomer HMMM formulations containing approx. 30% solids. Veneer samples treated with water were used as controls. The "full cell" treatment process involved an initial 30-minute period of vacuum of approx. –85 kPa and flooding of the

treatment vessel with formulation, followed by a 1-hr period of pressure of approx. 1400 kPa. Samples were removed from the treatment vessel, their surfaces were wiped free of excess formulation, and they were weighed. Wet weight percentage gain was calculated. Samples were dried at approx. 65°C for 2 days in an oven containing beakers of water in an attempt to simulate kiln-drying conditions. The samples were conditioned as described below before they were weighed and dry weight percentage gain was calculated.

Stiffness Measurements

Pinus radiata veneer samples before and after treatment were conditioned at 23°C and 50% relative humidity for 1 week prior to stiffness measurement. Width and three thickness measurements were taken for each sample. The width was measured using a sliding calliper and the thickness using a vernier gauge (both accurate to 0.01 mm) through the middle of the sample. Stiffness or modulus of elasticity (MoE) was determined on the veneer samples before and after treatment using non-destructive three-point static bending. The span was set at 100 mm and the load head speed at 10 mm/min. The MoE perpendicular to the plane of the veneer in bending was calculated using the following equation (BS5669 1989):

$$\text{MoE (GPa)} = \frac{Y^3 \Delta W}{4000 B T^3 \Delta S}$$

where Y = the span (mm)

ΔW = the increment of load on the straight line portion of the load deflection curve, 50 N for the original veneer, 200 N for treated veneer

B = the sample width (mm)

T = the mean sample thickness (mm)

ΔS = the increment of deflection (mm).

Microscopy of Chitin Oligomer HMMM and Chitosan Oligomer HMMM Co-polymer Modified Veneers

Cross-sections of veneer specimens were cut with a razor blade by hand and examined and photographed using light microscopy (Zeiss Photomicroscope) and confocal microscopy (Leica TCS/NT confocal laser scanning microscope).

RESULTS AND DISCUSSION

Chemical Hydrolysis and Characterisation

The electrospray mass spectrum of the chitin oligomers prepared by chitin hydrolysis showed ions associated with sodium adducts of oligomers with 1 to 6 *N*-acetylglucosamine units, indicating the chitin oligomer preparation contained

monomers to hexamers. The electrospray mass spectrum of the chitosan oligomers prepared by nitrous acid deaminative depolymersiation indicated the presence of oligomers of 1 to 6 glucosamine units. Larger oligomers may also have been present but were not detectable by ESI MS. Overall, the nitrous acid depolymerisation reaction to produce chitosan oligomers was a simpler procedure, and more amenable to scale-up than the chitin hydrolysis, as no de-salting by differential solubility was required.

Pre-polymerisation of Chitin/Chitosan Oligomers with HMMM

HMMM was reacted with the chitin oligomers to produce a "pre-polymer" product that was not homogeneous; the formulation contained some unreacted HMMM which was immiscible with the aqueous layer. The yield of water-insoluble polymer produced by heating of this formulation was 28–34%. This was a lower yield than that (60–80%) obtained from co-polymerisation of maltodextrin with HMMM (Franich & Anderson 1998). Varying the reaction temperature (65° and 80°C) and reaction time (1 to 24 hr) did not improve the yield of water-insoluble polymer obtained on heating. In contrast, the reaction of chitosan oligomers with HMMM at 50°C for 1 hr produced a homogeneous formulation which on heating gave an average 69% yield of water-insoluble co-polymer.

Acid-hydrolysis of chitin and chitosan produced water-soluble oligosaccharides. In order to polymerise these after the veneer treatment, the oligosaccharides were co-polymerised with HMMM through methylene ether linkages achieved by thermal condensation reactions between the methylol groups of HMMM and alcohol groups of the oligosaccharide. These reactions occurred on drying of the treated veneers. HMMM is insoluble in water, and therefore it was necessary to "pre-polymerise" HMMM with the oligosaccharides to form water-soluble "pre-polymers" with sufficiently low molecular weights to enable cell wall penetration. This pre-polymerisation synthesis approach worked well with maltodextrin in the commercial Indurite™ process (Franich & Anderson 1998) carried out on 20 kL scales.

The success of the pre-polymerisation reaction is critical to the formation of oligosaccharide HMMM co-polymers, within the lignocellulosic cell wall, capable of enhancing the stiffness of the material. As the chitin oligomers on co-polymerisation with HMMM gave only a 30% yield of polymer approximately, it was considered that use of this product would not result in significant enhancement of veneer stiffness. In contrast, the yield (69%) obtained from the co-polymerisation of chitosan and HMMM was similar to that achieved with maltodextrins and was considered sufficient to be useful for lignocellulosic veneer modification to attempt to improve veneer stiffness.

Veneer Treatments

Treatment of veneers with formulations (approx. 30% solids) of chitin and chitosan oligomer HMMM "pre-polymers" resulted in average dry weight percentage gains of 69% and 57% respectively (Table 1). A high degree of variability was observed in both the wet and dry weight percentage gains as indicated by the large standard deviations (Table 1). This was probably due to different proportions of earlywood and latewood in the replicate samples resulting in variable formulation uptake.

TABLE 1–Wet and dry weight percentage gains of veneer samples treated with chitin and chitosan oligomers HMMM formulations.

Treatment	Wet wpg (%)		Dry wpg (%)	
	Average	SD	Average	SD
Water	162	15	2	0.6
Chitin	180	26	69	10
Chitosan	168	14	57	5

Stiffness Assessment

The average percentage improvements in stiffness (MoE) of the *P. radiata* veneers on treatment with the oligosaccharide HMMM "pre-polymers" and subsequent polymerisation are summarised in Table 2. Veneers treated with chitin oligomer HMMM formulation showed no useful improvement in stiffness compared with controls (water treated). The poor degree of co-polymerisation achieved between chitin oligomers and HMMM is the most likely explanation for the poor performance of these treatments despite an increase in the material's density. In contrast, treatment of veneers with the chitosan oligomers HMMM formulation resulted in average stiffness improvements of 20% (Table 2). There was, however, considerable variability in the percentage stiffness improvements as illustrated by the high standard deviations. This variability in stiffness performance did not correlate with the variability in the veneer treatments (i.e., dry weight percentage gain values).

Effective co-polymerisation of the chitin or the chitosan oligomers with HMMM was critical to the success of the treatment in improving veneer stiffness. This is

TABLE 2–Average stiffness (MoE) enhancement for *P. radiata* veneers treated with chitin and chitosan HMMM co-polymer.

Treatment	Average MoE increase (%)	SD
Water	–2	7
Chitin	0.5	5
Chitosan	20	10

evident from the stiffness data obtained from blending chitin oligomer HMMM and chitosan oligomer HMMM formulations and co-polymerising in veneers. The results are summarised in Fig. 1. A minimum level of cross-linking (i.e., >30%) of the components to form a polymer within the cell wall was required before any stiffness improvement was observed. Although stiffness improvements were significantly less than the 100% improvement which was the industry target, the relationship between the degree of co-polymerisation and veneer improvement (Fig. 1) suggests there is potential to realise greater stiffness improvements by achieving high co-polymer yield within the lignocellulosic material.

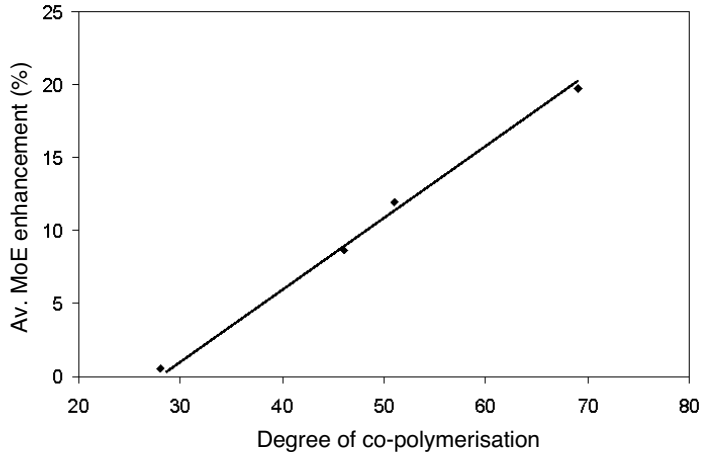

FIG. 1–Linear relationship (r^2=0.997) between average veneer stiffness enhancement (MoE %) and degree of chitin/chitosan HMMM co-polymerisation.

Furthermore, the significant veneer stiffness enhancement achieved by substituting a β-(1→4) oligosaccharide with a degree of polymerisation similar to that of the α-(1→4) maltodextrin used in the Indurite™ process, has shown that the configuration and tensile modulus of the lignocellulose-modifying carbohydrate can influence the overall stiffness of the derived composite. Optimisation of the formulation composition and the pre-polymerisation reaction, use of alternative co-monomers for condensation polymerisation reactions with chitosan oligomers, or substitution of these with β-(1→4) glucan (cellulose) oligomers may provide pathways towards better carbohydrate oligosaccharide co-polymerisation and therefore further enhancement of lignocellulose material stiffness.

Microscopy of Chitin and Chitosan Oligomer HMMM Co-polymer Lignocellulose Composite

Confocal fluorescence micrographs of *P. radiata* veneers treated with water (control), chitin, and chitosan oligomer HMMM co-polymer are shown in Fig. 2.

A light micrograph of the chitosan oligomer HMMM co-polymer within the cell lumens and walls is given in Fig. 3. Whereas the chitin oligomer HMMM co-polymer appeared to be distributed mainly within cell lumens (Fig. 2), the chitosan oligomer HMMM co-polymer appeared to be concentrated in the S_2 layer and more towards the S_3 layer, in addition to lumen filling. This observation implies that increasing the hydrogen bond density with the chitosan oligomer HMMM co-polymer in the S_2/S_3 layer in the lignocellulose wall enhanced the composite stiffness.

The accumulation of the chitosan oligomer HMMM "pre-polymer" within the S_2/S_3 region of the lignocellulose wall could be due to the primary amino groups bonding to acidic components in the wall in this region. In contrast, the neutral and less-polar (due to the 2-acetamido group) chitin oligomer HMMM "pre-polymer" appeared to be distributed within the lignocellulose lumen, where there would be expected to be little influence on cell wall modification and mechanical properties.

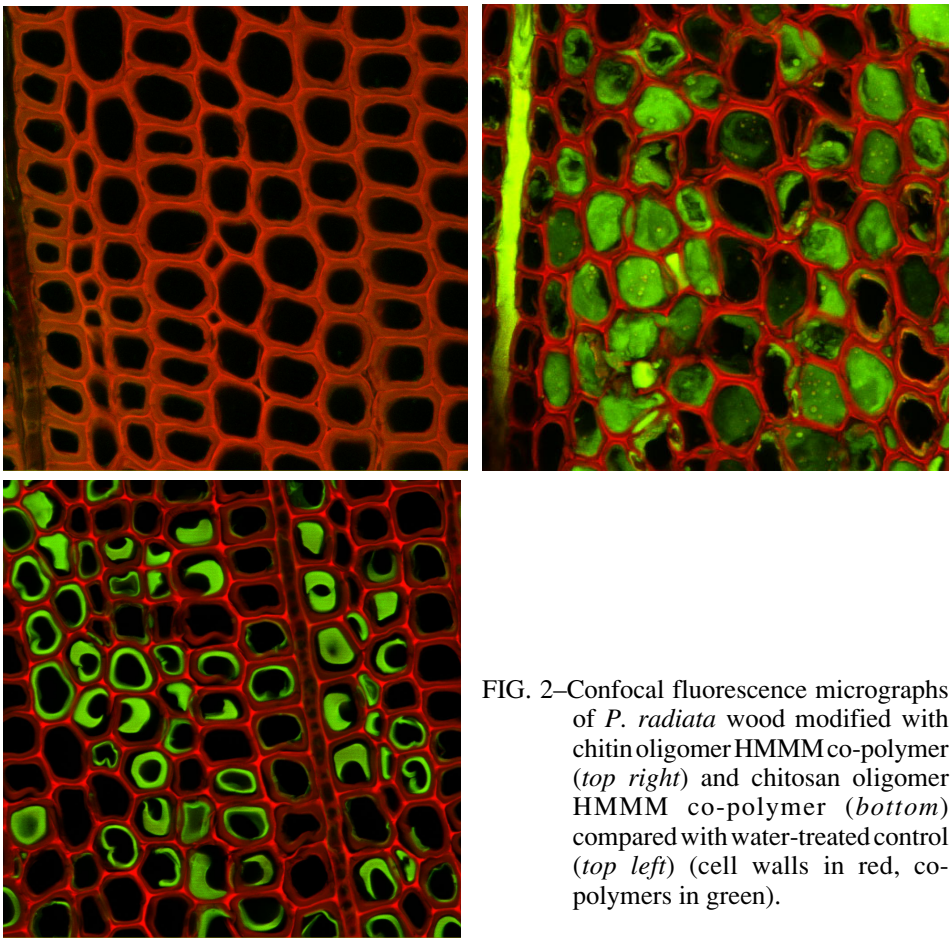

FIG. 2–Confocal fluorescence micrographs of *P. radiata* wood modified with chitin oligomer HMMM co-polymer (*top right*) and chitosan oligomer HMMM co-polymer (*bottom*) compared with water-treated control (*top left*) (cell walls in red, co-polymers in green).

FIG. 3–Light microscope image of chitosan oligomer HMMM co-polymer modified *P. radiata* wood (cell walls in blue, chitosan oligomer HMMM co-polymer in yellow).

CONCLUSIONS

Chitin and chitosan oligosaccharides with degrees of polymerisation ranging from one to six sugar units can be produced by acid-catalysed hydrolysis and nitrous acid deaminative depolymerisation methods respectively, providing a simple route to chitin and chitosan oligomers. HMMM was successfully used to co-polymerise with chitosan oligomers, but was less effective with chitin oligomers. The yield of co-polymer of chitosan oligosaccharides and HMMM within the lignocellulosic material on drying was found to be critical to improving the stiffness of the treated veneers. A threshold of >30% co-polymer yield was required before any stiffness improvement was observed. Interpretation of the microscopy results suggests that the way in which the chitosan or chitin HMMM co-polymers are distributed within the wood cell is likely to be a contributing factor in the stiffness performance of the treated veneers.

These experiments carried out using the β-(1→4) oligosaccharides have shown the potential for lignocellulosic material stiffness improvements and indicated possible alternatives to the previously-used phenol-formaldehyde resins for wood material modification. Optimisation of the chitosan HMMM pre-polymerisation reaction and improved co-polymerisation of the oligosaccharides, which could include use of alternative co-monomers for use with chitosan or other alternative β-(1→4) oligosaccharides, are further aspects for research in this area.

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REFERENCES

ALLAN, G.G.; PEYRON, M. 1997: Depolymerisation of chitosan by means of nitrous acid. *In* Muzzarelli R.A.A.; Peter, M.G. (Ed.) "Chitin Handbook". European Chitin Society, Atec, Grottammare, Italy.

ANDO, K.; ONDA, H. 1999: Mechanism for deformation of wood as a honeycomb structure. I: Effect of anatomy on the initial deformation process during radial compression. *Journal of Wood Science 45*: 120–126.

BLUMBERG, K.; LINIERE, F.; PUSTILNIK, L.; BUSH, A. 1982: Fractionation of oligosaccharides containing *N*-acetyl amino sugars by reverse-phase high-pressure liquid chromatography. *Analytical Biochemistry 119*: 407–412.

BS5669 1989: Particleboard. Part 1 Methods of sampling, conditioning and test. British Standards Institution, London, United Kingdom.

DWIANTO, W.; MOROOKA, T.; NORIMOTO, M.; KITAJIMA, T. 1999: Stress relaxation of sugi (*Cryptomeria japonica* D. Don) wood in radial compression under high temperature steam. *Holzforschung 53*: 541–546.

FIBRE7 2004: "Fibre7: Creating Hardwood Forever" [Online]. Available: www.fibre7.com [7 June 2005].

FRANICH, R.; ANDERSON, K. 1998: Densification of lignocellulosic material. *United States Patent 5770319*.

INOUE, M.; OGATA, A.; KAWAI, S.; ROWELL, R.M.; NORIMOTO, M. 1993: Fixation of compressed wood using melamine-formaldehyde resin. *Wood and Fibre Science 25*: 404–410.

ISOGAI, A.; USUDA, M. 1991: Preparation of low-molecular weight celluloses using phosphoric acid. *Mokuzai Gakkaishi 37*: 339–344.

KININMONTH, J.A.; WHITEHOUSE, L.J. 1991: "Properties and Uses of New Zealand Radiata Pine: Wood Properties" Vol. 1. Forest Research Institute, Rotorua, New Zealand.

KOSHIJIMA, T.; WATANABE, T. 2003: "Association between Lignin and Carbohydrates in Wood and other Plant Tissues". Springer-Verlag, Berlin, Germany.

LI, J.; COWN, D.J. 2002: Enhancement of radiata pine mechanical properties by thermal compression. Paper presented at the 9th APCChE Congress and CHEMECA 2002, 29 September–3 October, Christchurch, New Zealand.

MARRA, A.A. 1992: "Technology of Wood Bonding: Principles and Practice". Van Nostrand Reinhold, New York, United States of America.

NAVI, P.; GIRARDET, F. 2000: Effects of thermal-hydro-mechanical treatment on the structure and properties of wood. *Holzforschung 54*: 287–293.

NISHINO, T.; MATSUI, R.; NAKAME, K. 1999: Elastic modulus of the crystalline regions of chitin and chitosan. *Journal of Polymer Science: Part B: Polymer Physics 37*: 1191–1196.

ROWELL, R.; LANGE, S.; DAVIS, M. 2000: Steam stabilisation of aspen fibreboards. Paper presented at the Fifth Pacific Rim Bio-based Composites Symposium, 10–13 December, Canberra, Australia.

YALPANI, M.; PANTALEONE, D. 1994: An examination of the unusual susceptibilities of aminoglycans to enzymatic hydrolysis. *Carbohydrate Research 256*: 159–175.