



Development of "Green" Wood Adhesives for Wood Composite Products

Development of Bio-Modified Chitosan-Based Adhesives for Wood Composites

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Transformative Technologies - Development of "Green" Wood Adhesives for Wood Composite Products

Development of Chitosan-Based Adhesives for Wood Composites

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ABSTRACT

Chitosan is an amino polysaccharide obtained from the deacetylation of chitin, which is naturally occurring in the shells of a large number of marine crustaceans. Chitosan is soluble in weakly acidic aqueous solutions and possesses adhesive properties. Chitosan has received much attention for medical and industrial applications; however, only limited studies have been conducted on the application of chitosan as a wood adhesive, because its bonding properties on wood are poor. To improve the adhesive quality of chitosan resin, an innovative study on chitosan adhesives has been conducted to use selected fungal species to modify chitosan and improve its bonding properties, to synthesize non-formaldehyde resins with the fungus-modified chitosan, and to enhance urea-formaldehyde (UF) and phenol-formaldehyde (PF) resin performance with the fungus-modified chitosan.

The bonding properties of wood composites made with these chitosan-based green wood adhesives were significantly improved, in terms of lap-shear strength. Unmodified chitosan solution was not compatible with ammonium lignosulfonate, liquid PF resin, soybean resin, powder PF resin, or soybean flour, but was compatible with UF resin, polyvinyl acetate (PVA) resin, and phenol. With the addition of chitosan in UF and PVA resins, both the dry and wet shear strengths of plywood panels were improved, compared with those of panels bonded with the control UF and PVA resins, i.e. without chitosan. A number of chitosan and chitosan-reinforced UF resins were prepared as a binder for particleboard panel manufacturing. Six (6) types of particleboard panels with different levels of resin loadings and press conditions were manufactured. The resulting boards were tested to evaluate the bond quality of the chitosan and chitosan-reinforced UF resins. The test results showed that particleboard panels with good visual quality could be produced with all formulations of chitosan-UF adhesives, even with resin systems made with 1% of chitosan resin only. All chitosan resins used alone or added to UF resins yielded panels with better internal bond (IB) strength than those made with the UF control resin. The panels made with 1% chitosan resin plus 66% UF resin in a 1:1 ratio yielded panels with the highest IB strength and the best overall mechanical properties.

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1. OBJECTIVES

The objective of this research was to explore the feasibility of using chitosan-based wood adhesives as “green” binders in the manufacture of wood composites such as plywood and particleboard panels.

2. INTRODUCTION

Chitosan is an amino polysaccharide obtained from the deacetylation of chitin which is naturally occurring and found in large amounts in the shells of marine crustaceans such as crabs and shrimps, and in the cell walls of fungi. The chemical structure of chitosan consists of β -1,4-linked D-glucosamine residues with a number of randomly located N-acetyl-glucosamine units. Chitosan is soluble in weakly acidic aqueous solutions, in which it becomes a cationic polyelectrolyte that can interact with negatively charged molecules. In other words, chitosan possesses some adhesive properties. Chitosan has received much attention as a potential polysaccharide resource in various fields, and has been studied extensively for medical and industrial applications. However, only a few studies have been conducted on the application of chitosan in the manufacture of wood adhesives.

An earlier study at FPInnovations has shown that when chitosan alone was used as a wood adhesive, its wood bonding properties were poor. Subsequently, an innovative feasibility study on chitosan adhesives was conducted on the use of selected fungal species to modify chitosan and improve its bonding properties. In this study, a non-formaldehyde resin was synthesized and traditional urea-formaldehyde (UF) and phenol-formaldehyde (PF) resins were enhanced with fungus-modified chitosan. The resulting chitosan-based wood adhesives were evaluated for bonding quality through manufacturing and testing of wood composites, such as plywood and particleboard panels.

3. TECHNICAL TEAM

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4. MATERIALS AND METHODS

4.1. Chitosan Characterization

4.1.1. Preparation of Chitosan Solutions

The chitosan used in this study was purchased from Marinard Biotech in Rivière-au-Renard, Québec. The degree of deacetylation was 86.6%. The characteristics of this product are presented in Table 1.

Table 1 - Characteristics of chitosan used in the studies

Parameters	Characteristics
Appearance	Off-white
Form	Flake
Moisture	5.6%
Ash	0.197%
Protein	< 0.2%
Insoluble matter	< 1%
Degree of deacetylation	86.6%
Toxic heavy metals (As, Cd, Hg, Pb)	< 5 ppm
Viscosity (1% wt in 1% acetic acid)	307 cps
Microbial contamination	890 CFU/g

Chitosan appears as white flakes, as shown in Figure 1. It is soluble in dilute acids such as acetic acid or formic acid. The bonding strength of chitosan alone can be enhanced with lactobionic acid and p-azidebenzoic acid. However, different concentrations of these acids will affect the pH, solubility, and viscosity of the chitosan solutions.

In most studies recorded in the literature, chitosan is usually dissolved in a 1% (w/v) acetic acid solution. In order to find the optimal concentration of acetic acid to dissolve chitosan, a series of diluted solutions of acetic acid containing 1%, 2%, 4%, 6%, 8% and 10% (v/v) of 1N acetic acid solution was prepared. The concentrations of chitosan in these acid solutions were of 1%, 2%, and 4% (w/v); the solutions were stirred with or without heating for 30 min.

**Figure 1. Appearance of pure chitosan**

4.1.2. Compatibility of Chitosan with Other Resins

Chitosan solutions can be used alone as a wood adhesive or combined with other types of synthesized resins to improve their performance, such as water resistance, bonding strength, or formaldehyde emissions. In this study, the following resin formulations were tested to evaluate chitosan's compatibility with other resins or components:

- 1) Chitosan solution + ammonium lignosulfonate from the chemical pulping process
- 2) Chitosan solution + liquid PF resin
- 3) Chitosan solution + liquid UF resin
- 4) Chitosan solution + liquid PVA resin
- 5) Chitosan solution + liquid soybean resin
- 6) Chitosan solution + powdered PF resin
- 7) Chitosan solution + phenol crystals
- 8) Chitosan solution + soybean flour

In Formulations 1 to 5, 100 mL of chitosan solution (2% chitosan in 10% 1N acetic acid solution) were mixed with 50 mL of lignosulfonate or commercial resins specified. For Formulations 6 to 8, 150 mL of chitosan solution (2% chitosan in 10% 1N acetic acid solution) were mixed with 10 g of the powdered component. All mixtures were stirred at room temperature for 30 minutes without heating.

4.2. Fungal Modification of Chitosan

4.2.1. Fungal Species Selection and Chitosan Modification

Three (3) fungal species were selected to modify chitosan: *Trichoderma harzianum* Rifai (isolate FTK160D), *Trichoderma viride* Pers.:Fr. (isolate FTK161D), and *Gliocladium roseum* Bainier (isolate FTK321U). These fungal isolates came from the Culture Collection of Wood-Inhabiting Fungi at FPInnovations, Québec, Canada. The cultures were maintained in a liquid nitrogen reservoir for cryopreservation at -198°C until use.

The fungal cultures were first grown on a 2% malt extract agar medium in Petri plates, at 25°C, for one week. Mycelia plugs (5 mm in diameter) were cut from each fungal colony and five of them were then transferred to separate flasks. Each flask contained 500 mL of a specific sterile medium consisting of 10 g of KNO₃, 5 g of KH₂PO₄, 2.5 g of MgSO₄·7H₂O, 3.3 mg of FeCl₃·6H₂O, 20 g of chitosan, 10 g of polyvinylpyrrolidone, and 150 mL of V8 juice; the volume was adjusted to 1 liter with distilled water. The pH of the medium was 6.0. The contents of each flask were cultured on a shaker (125/rpm) at 25°C, for 21 days (Figure 2).

After incubation, the fungal cultures were first filtered through a layer of cheese cloth to remove mycelia plugs and fungal residue, and then through a No.1 filter to remove the liquid medium. The slurry of chitosan was washed with sterile distilled water and filtered twice, to get rid of any remaining medium. The final slurry of chitosan was heated to dryness at 50°C for three (3) days and crushed into a powder, ready for use. The color of the modified chitosan varied with the fungal species used. The chitosan modified by *Gliocladium roseum* was yellowish (Figure 3), that modified by *Trichoderma harzianum* was light brown (Figure 4), whereas that modified by *Trichoderma viride* was a darker brown (Figure 5).



Figure 2. Fungal modification of chitosan by a fermentation process



Figure 3. Appearance of chitosan modified by fungal species 321U



Figure 4. Appearance of chitosan modified by fungal species 160D



Figure 5. Appearance of chitosan modified by fungal species 161D

4.2.2. Preparation of Fungus-Modified Chitosan Resins

Three (3) groups of adhesives were prepared with fungus-modified chitosan, i.e., adhesives made of fungus-modified chitosan alone, phenol-formaldehyde (PF) resins reinforced with fungus-modified chitosan, and urea-formaldehyde (UF) resins reinforced with fungus-modified chitosan. The procedures for preparing each group of adhesives are outlined below:

- (1) Resins prepared from fungus-modified chitosan alone: three (3) grams of each fungus-modified chitosan powder were weighed, and then added to 100 mL of a solution containing 1% (wt) acetic acid. The reaction mixture was stirred at room temperature for one (1) hour until all of the chitosan powder was dissolved (pH 4.75).
- (2) Chitosan-PF resins: a commercial PF resin (non-volatile content around 57% w/w, and pH around 11.3) was mixed with 10 wt% of each fungus-modified chitosan powder by slowly adding the chitosan to the resin and stirring at room temperature, until all of the chitosan powder was well dispersed.

- (3) Chitosan-UF resins: a commercial UF resin (non-volatile content around 60% w/w) was mixed with 5 wt% of each fungus-modified chitosan powder by slowly adding the chitosan to the resin and dispersing it in a same way as that used in the preparation of chitosan-PF resin.

A control set of resins made with unmodified chitosan was also prepared following the same procedures as those used in preparing the resins with fungus-modified chitosan. All of the prepared resins were stored at room temperature for a week, until used in composite panel manufacturing.

4.2.3. Evaluating Bonding Quality of Fungus-Modified Chitosan Resins via Plywood Manufacturing

The bonding quality of the resulting fungus-modified chitosan resins was evaluated through plywood panel manufacturing and testing. Two 2-ply plywood panels were prepared with each resin system and sixteen (16) specimens were cut from these two plywood panels. The resulting plywood panels were tested for lap-shear strength and the results were expressed as the average of the values obtained for the sixteen (16) specimens tested.

4.3. Enzyme Modification of Chitosan

4.3.1. Evaluation of Chitosan Degrading Enzymes Produced by Different Fungi

The fungal species, *Trichoderma harzianum* (160D), *Trichoderma viride* (161D) and *Gliocladium roseum* (321U), were cultivated on a 2% malt extract agar medium (MEA), and grown at 25°C for 7 days. Three (3) agar discs (5-mm in diameter) were punched from the edge of each fungal colony and inoculated at equal distance from each other, in a 9-cm diameter plate (3 plates per fungus) containing a selective medium for chitosan enzymes. The medium contained 0.1% K₂HPO₄, 0.01% MgSO₄·7H₂O, 0.1% NaCl, 0.7% (NH₄)₂SO₄, 0.05% yeast extract, 1% chitosan (4% chitosan dissolved in 10% 1N acetic acid solution), and 1.5% agar. They were grown at 25°C in darkness for seven (7) and fourteen (14) days. The diameters of the fungal colonies and of the clear zones formed around the fungal colonies were measured.

4.3.2. Enzyme Modification

For enzyme modification of chitosan, two (2) fungal species, *Trichoderma viride* (161D) and *Gliocladium roseum* (321U) were used. The culture medium and culturing conditions were the same as those described above for the fungal modification of chitosan. After filtration of the fungal mycelia and spores, the culture filtrate was passed through a No.1 filter; this was used as a crude enzyme solution and kept at 4°C until use. A 2% solution of chitosan (w/v) was added to this crude enzyme solution in a flask and fermented on a shaker (125/rpm) at 25°C for 24 hours. After fermentation, the chitosan was filtered through a layer of cheese cloth, and then dissolved in an 18% solution of acetic acid to yield a clear resin solution. The chitosan solution modified by the enzymes produced by *G. roseum* was yellowish (Figure 6), whereas that modified by the enzymes produced by *T. viride* was brownish (Figure 7). The color of the enzyme-modified chitosans matched the colors of the chitosans modified with the fungal mycelia of each of these two (2) species.



Figure 6. Chitosan modified by enzyme extracted from fungal strain 321U



Figure 7. Chitosan modified by enzyme extracted from fungal strain 161D

4.3.3. Evaluating Bond Quality of Enzyme-Modified Chitosan Adhesives via Plywood Manufacturing

The following adhesive formulations were prepared with original chitosan, enzyme-modified chitosan, and commercial resins.

- A) Chitosan resin (2% solution): chitosan powder (2% w/v) was added to a 10% 1N acetic acid solution. The solution was stirred for 1 hour at room temperature until all the chitosan had dissolved (Figure 8), and then stored for 24 hours before it was used (Figure 9, A).

- B) Chitosan resin (4% solution): chitosan powder (4% w/v) was added to a 10% 1N acetic acid solution. The solution was stirred for 1 hour at room temperature until all the chitosan had dissolved, and then stored for 24 hours before it was used (Figure 9, B).
- C) Chitosan resin (2% solution) + tyrosinase: 200 mL of chitosan resin solution (2%) were mixed with 20 mg of tyrosinase (50KU, Sigma), stirred for 1 hour at room temperature, and allowed to react for 24 hours before use (Figure 9, C).
- D) Chitosan resin (2% solution) + phenol (2%) + laccase: 200 mL of chitosan resin solution (2%) were mixed with 4 g of phenol (90%, Sigma), and stirred for 30 hours at room temperature until all the phenol had dissolved. Then, 1g of laccase (Sigma) was added; the solution was stirred for 1 hour at room temperature, and allowed to react for 24 hours before use (Figure 9, D).
- E) Chitosan modified by enzyme from fungal strain 321U (2%): the enzyme-modified chitosan powder (2% w/v) was added to an 18% 1N acetic acid solution. The solution was stirred for 1 hour at room temperature until all the chitosan had dissolved, and stored for 24 hours before it was used (Figure 9, E).
- F) Chitosan modified by enzyme from fungal strain 161D (2%): the enzyme-modified chitosan powder (2% w/v) was added to an 18% 1N acetic acid solution, stirred for 1 hour at room temperature until all the chitosan had dissolved, and stored for 24 hours before use (Figure 9, F).
- G) Chitosan resin (2% solution) + commercial urea-formaldehyde (UF) resin (2:1): 200 mL of chitosan resin (2%) were mixed with 100 mL of a commercial UF resin, stirred for 1 hour at room temperature, and stored for 24 hours before use (Figure 10, G).
- H) Chitosan resin (2% solution) + commercial urea-formaldehyde (UF) resin (1:2): 100 mL of chitosan resin (2%) were mixed with 200 mL of a commercial UF resin, stirred for 1 hour at room temperature, and stored for 24 hours before use (Figure 10, H).
- I) Chitosan resin (2% solution) + commercial polyvinyl acetate (PVA) resin (2:1): 200 mL of chitosan resin (2%) were mixed with 100 mL of a commercial PVA resin, stirred for 1 hour at room temperature, and stored for 24 hours before use (Figure 10, I).
- J) UF resin control (Figure 10, J).
- K) PVA resin control (Figure 10, K).
- L) PF resin control (Figure 10, L).

For each resin, two (2) pieces of yellow birch veneer strips (1.5 mm thick x 148 mm wide x 313 mm long) were coated with resin by brushing. Based on the resin solids content, different spread rates were applied to make 2-ply plywood panels. Table 2 gives the relevant parameters for the different panels produced. The plywood panel manufacturing conditions are summarized in Table 3. The two (2) plies were stacked together after a proper open assembly time and then hot-pressed at 140°C for 3 min under a pressure of 1500 kPa.

After manufacturing, all panels were conditioned at 21°C and 20% relative humidity (RH) until equilibrium moisture content (EMC) was reached. These 2-ply plywood panels were then cut into the

specimen size (25 mm wide x 80 mm long) required for lap-shear testing, as illustrated in Figure 11. The lap-shear strengths of these specimens were determined using a MTS Alliance RT/50 testing machine with a crosshead speed of 1 mm per min, according to CSA Standard Test Method O112.0-M, under both dry and wet test conditions. To perform the test under wet conditions, the specimens bonded with the different resins were soaked in tap water for 48 hours at room temperature, and then tested when the specimens were still wet (CSA O112.5-M, CSA O112.6M, CSA O112.8-M). Twelve (12) specimens cut from the two (2) plywood panels were tested for each resin system. The shear strength values reported is the average of the values obtained for the twelve (12) specimens tested for a particular resin system.

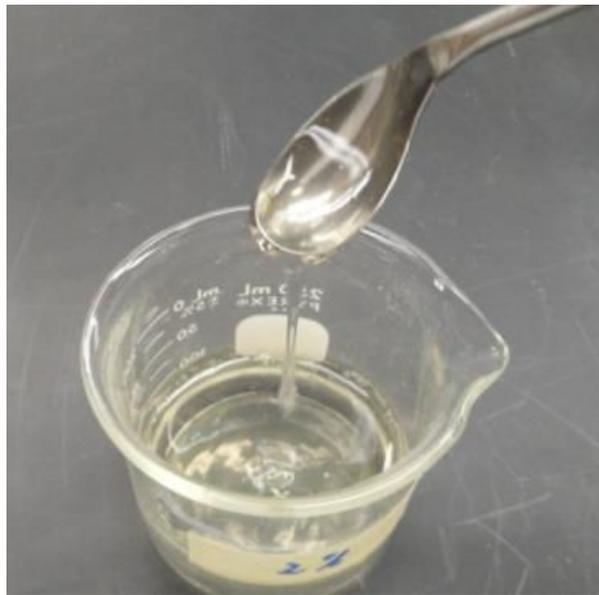


Figure 8. Preparation of chitosan (alone) resin

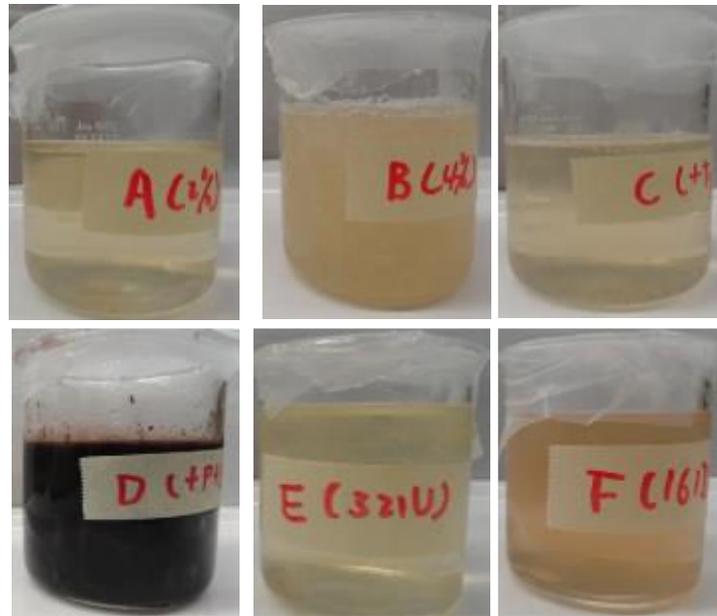


Figure 9. Appearance of chitosan and bio-modified chitosan resins A to F



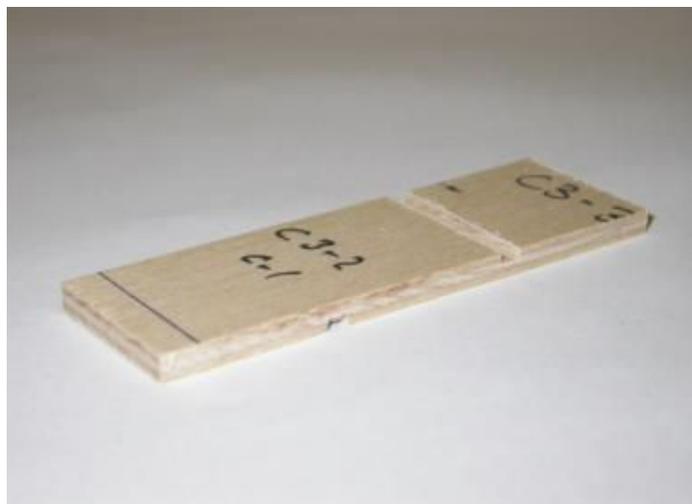
Figure 10. Appearance of three chitosan-based resins G to I and three commercial resins J to L

Table 2 - Information on glue application for 2-ply plywood panels made with different types of resins

Panel ID	Veneer Mass (g)	Glue Mass on Face Veneer (g)	Average Liquid Glue Spread per Panel per Glue Line (g/m ²)	Average Solid Glue Spread per Panel per Glue Line (g/m ²)	Open Assembly Time (min)	Closed Assembly Time (min)
A1-1	51.28	10.28	434.33	8.69	9.20	21.15
A1-2	51.40	10.80	445.34	8.91	9.20	21.15
A2-1	51.22	14.18	635.74	12.71	9.10	7.00
A2-2	51.20	6.95	286.89	5.74	9.10	7.00
B1-1	53.53	5.55	236.59	9.46	5.40	18.47
B1-2	52.30	5.28	236.81	9.47	5.20	18.47
B2-1	52.97	10.29	452.90	18.12	5.19	7.01
B2-2	53.05	10.80	457.21	18.29	5.03	7.01
C1-1	60.51	10.18	447.07	8.94	4.35	22.50
C1-2	59.99	10.72	447.72	8.95	4.35	22.50
C2-1	60.40	19.71	870.18	17.40	5.12	7.50
C2-2	59.58	20.35	862.62	17.25	5.04	7.10
D1-1	51.77	4.86	212.85	8.51	4.05	19.54
D1-2	51.74	4.89	210.91	8.44	4.00	19.54
D2-1	52.05	9.87	414.26	16.57	5.02	7.00
D2-2	52.27	10.22	430.45	17.22	4.95	7.00
E1-1	51.73	9.27	408.00	8.16	5.30	25.37
E1-2	51.93	10.03	436.06	8.72	5.10	25.37
E2-1	51.31	14.6	635.96	12.72	4.35	7.00
E2-2	52.01	14.18	615.88	12.32	4.15	7.00
F1-1	52.48	10.17	428.94	8.58	5.62	23.15
F1-2	52.37	9.85	420.52	8.41	5.08	23.15
F2-1	51.69	4.97	217.60	4.35	5.05	4.00
F2-2	51.82	6.01	232.49	4.65	4.01	4.00
G1-1	53.13	4.76	102.75	23.98	3.58	21.37
G1-2	53.60	4.97	107.29	25.03	4.20	21.37
G2-1	51.68	5.01	219.54	51.23	5.10	7.00
G2-2	51.55	5.09	228.18	53.24	5.04	7.00
H1-1	52.55	5.93	128.01	57.18	4.32	19.51
H1-2	52.67	5.76	124.34	55.54	4.08	19.51
H2-1	51.56	5.54	244.15	109.05	5.17	7.18
H2-2	52.38	5.46	240.91	107.61	5.07	7.08
I1-1	53.89	4.90	105.78	17.63	4.30	16.3
I1-2	60.22	4.98	107.50	17.92	4.30	16.3
I2-1	60.15	6.52	301.36	50.23	4.00	7.15
I2-2	58.97	6.60	289.27	48.21	4.00	7.15
J1-1	51.55	3.42	73.83	48.73	6.00	16.3
J1-2	51.92	4.20	90.67	59.84	6.12	16.3
J2-1	50.63	7.47	161.26	106.43	5.13	6.00
J2-2	51.16	7.38	159.31	105.15	5.20	6.00
K1-1	53.43	4.63	99.95	49.97	6.11	15.18
K1-2	53.34	4.90	105.78	52.89	6.11	15.18
K2-1	53.38	9.83	212.20	106.10	5.08	5.53
K2-2	53.33	9.65	208.32	104.16	5.05	5.53
L1-1	53.08	4.92	106.21	53.10	6.11	14.37
L1-2	54.19	4.86	104.91	52.46	5.05	14.37
L2-1	53.39	9.60	207.24	103.62	4.98	6.12
L2-2	53.26	9.36	202.06	101.03	5.00	6.12

Table 3 - Manufacturing parameters for 2-ply plywood panels made with different types of resins

Face veneer dimension	148 mm x 313 mm x 1.5 mm
Target glue spread on face veneer	7.7 to 8.3 g
Glue application method	3" roller with a 1/8"-thick foam
Area for applying pressure	185296 mm ²
Gauge pressure on Viceroy 24 in x 24 in	367 psi
Press time	3 min + 20 sec. degas

**Figure 11. Plywood specimen for shear test**

4.4. Preparation of UF-Chitosan Resins by Addition of Chitosan Solution during the Resin Synthesis Process

In the first part of the study, it was found that the performance of wood composites made from UF resins could be improved by post-addition of fungus-modified chitosan to some commercial resins. The objective of the second part of the study was to try to incorporate unmodified or fungus-modified chitosan into urea-formaldehyde (UF) resin by adding 2% chitosan solutions during the UF synthesis process.

4.4.1. Materials

Three 2% chitosan solutions in 1% acetic acid (2:1, w/w) were prepared as follows:

- R01: unmodified chitosan
- R02: fungus-modified chitosan, 321U
- R03: fungus-modified chitosan, 161D

Other raw materials used in resin preparation included:

- Formaldehyde (F), 37%, pH 2.79

- Urea
- Triethanolamine (TEA)
- Sodium chloride (NaCl)
- Ammonium chloride (NH₄Cl), 20% solution (used for adjusting pH)
- Sodium hydroxide (NaOH), 25% solution (used for adjusting pH)

4.4.2. Addition of Chitosan Solutions during UF Resin Synthesis Process

Different UF-chitosan resins were synthesized as described below:

- 1) The control UF resin, i.e., without addition of chitosan (coded control resin C01) was prepared as shown in Scheme-A in Figure 12.
- 2) Different UF/chitosan resins were produced by adding unmodified chitosan solution R01 at the initial condensation stage of the UF resin synthesis, (Scheme-B in Figure 12) at a ratio of 2:1, w/w, UF: chitosan solution.
- 3) Evaluation of the feasibility of adding unmodified or modified chitosan solutions (coded R01, R02, R03) at different condensation stages of the UF resin synthesis process is described in the scheme shown in Figure 13.
- 4) Exploratory synthesis of UF resin with addition of modified chitosan (R03) at the initial condensation stage of the UF resin synthesis process, at a ratio of 2:1, w/w, UF: chitosan solution, is illustrated in the scheme shown in Figure 14.

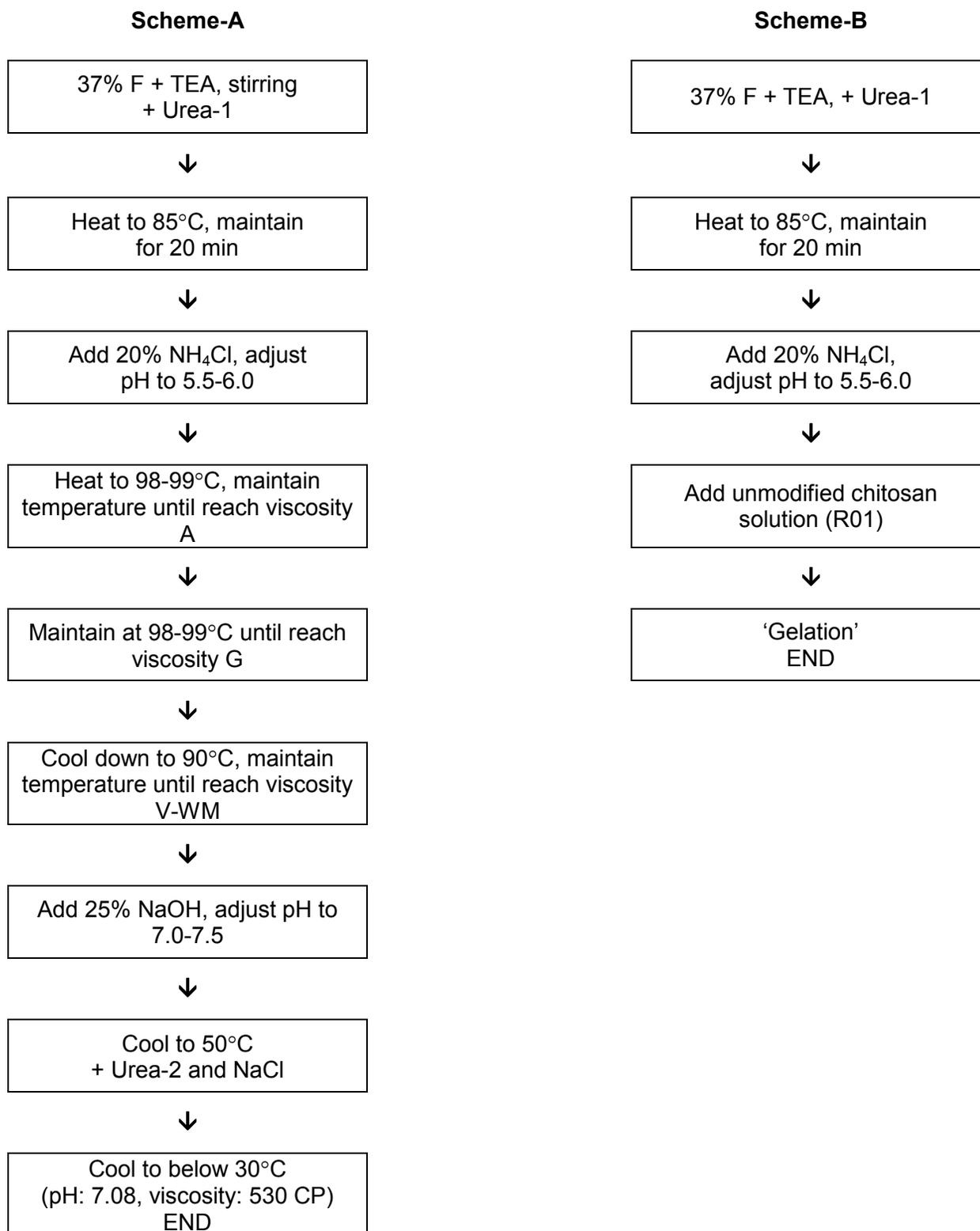


Figure 12. Synthesis of control UF resin (Scheme-A) and UF resins with the addition of unmodified chitosan (Scheme-B)

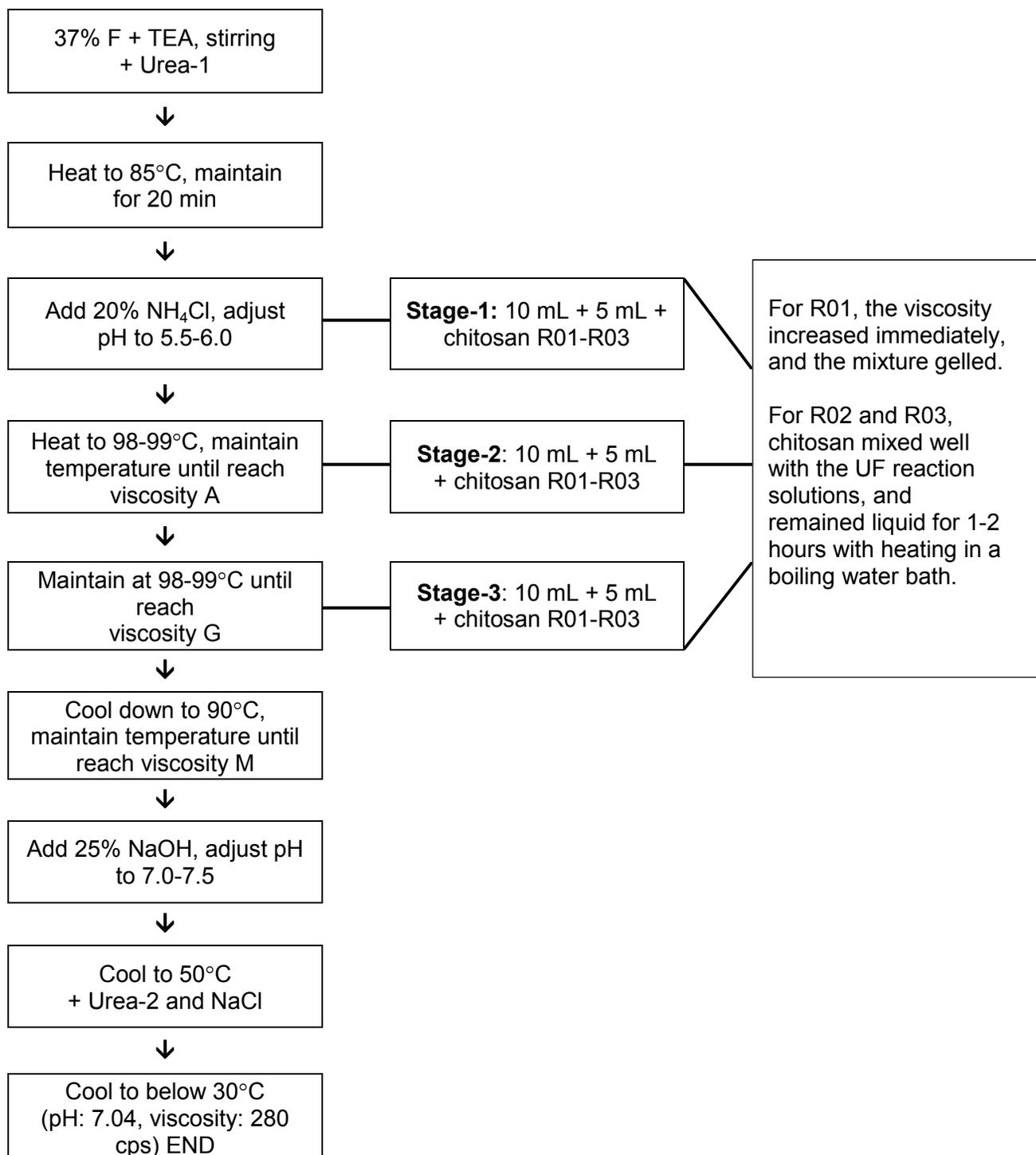


Figure 13. Scheme for evaluating the feasibility of adding chitosan solutions to the UF reaction mixture at three different condensation stages of the UF resin synthesis process

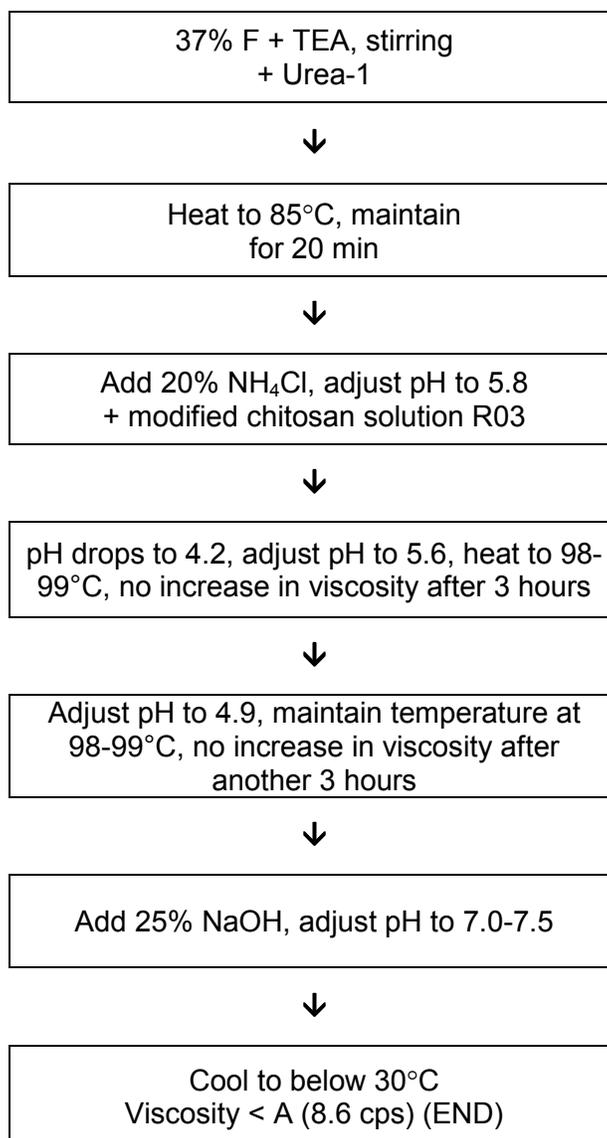


Figure 14. Scheme for synthesis of UF resin with addition of bio-modified chitosan solution R03

4.4.3. Evaluating Bond Quality of UF/Chitosan Resins Prepared by Addition of Chitosan Solutions during the Resin Synthesis Process, via Plywood Manufacturing

Two-ply plywood panels were prepared with the different resins synthesized as described above: 1) control without chitosan, 2) control with unmodified chitosan, and 3) addition of modified chitosan during UF resin synthesis. The resin codes and their descriptions are given below. The resin properties are listed in Table 4.

- UFControl (C1): F/U1 = 2:1
F/(U1+U2) = 1.12
No chitosan was added.
- UFControl (C2): F/U1 = 2:1

$F/(U1+U2) = 1.12$
Chitosan fine powder was post-blended.

- UFR1A: $F/U1 = 2:1$
 $F/(U1+U2) = 1.12$
Unmodified chitosan was added at viscosity G, during synthesis process.
- UFR1B: $F/U1 = 4:1$
 $F/(U1+U2) = 2:1$
 $F/(U1+U2+U3) = 1.12$
Unmodified chitosan was added with U1 at pH 7.2, during synthesis process.
- UFR1C: $F/U1 = 4:1$
 $F/(U1+U2) = 2:1$
 $F/(U1+U2+U3) = 1.12$
Unmodified chitosan was added at pH 2.9 and reacted with formaldehyde first at 60°C for 20 min.
- UFR2: $F/U1 = 2:1$
 $F/(U1+U2) = 1.12$
Fungus-modified chitosan was added at viscosity G.

Table 4 - Resin codes and their properties

UF Code	Solid Content (%)	Viscosity (cP)	pH	Gel Time (sec.)
UFControl (C1)	54.8	282	7.52	119
UFR1A	55.4	316	7.58	136
UFR1B	54.9	336	8.02	140
			7.44	133
UFR1C	54.6	565	8.08	>600
			7.58	390
UFR2	55.5	262	7.19	132

4.5. Formulating UF/Chitosan Resins via Post-Blending

4.5.1. Resin Formulation

A number of UF/chitosan resins were formulated by incorporating the unmodified chitosan (in solution or powder form) into a commercial urea-formaldehyde (UF) resin via post-blending. The commercial UF resin had 66% solids content and was designed for particleboard manufacturing. Information on the UF/chitosan formulations is given in Table 5; the mixing/application procedure is outlined immediately below the table.

Table 5 - Adhesive formulations for particleboard manufacturing

Formulation No.	Solids Content of Mixture (%wt.)	Ratio of Chitosan to UF by Weight	Original Chitosan Solution (%wt.)	Original UF Resin (%wt.)
1	1	1 : 0	1	-
2	44.67	1 : 2	2	66
3	-	2 : 2	2	66
4	-	0.09 : 5.91	100 (powder)	66
5	50.00	1 : 3	2	66
6	66.00	0 : 1	-	66

Formulation #1: Prepared 1% chitosan solution and used it as is.

Formulation #2: Mixed 1 part of 2% chitosan solution with 2 parts of commercial UF resin.

Formulation #3: First loaded 2 parts of 1% chitosan onto the wood furnish, dried the furnish to the target moisture content according to panel manufacturing specifications, and then applied 2 parts of commercial UF resin to the furnish.

Formulation #4: First loaded 0.0896 parts of chitosan powder onto the wood furnish and then applied commercial UF resin at 5.91%, based on resin solids weight.

Formulation #5: Mixed 1 part of 2% chitosan solution and 3 parts of commercial UF resin, and then applied it to the wood furnish.

Formulation #6: UF resin control applied as is.

4.5.2. Evaluating Bond Quality of UF/Unmodified Chitosan Resins via Particleboard Manufacturing

SPF (spruce, pine, fir) wood particles were obtained from a local particleboard mill. To evaluate the performance of the UF/chitosan resin formulations described above, six (6) types of particleboard panels were manufactured, as listed in Table 6 below. A 24 in x 24 in Viceroy Standard Press was used to press the boards. Detailed information on board manufacturing conditions is presented in Table 7.

Table 6 - Chitosan-based adhesives used in the manufacturing of particleboard panels

Panel Code	Adhesives	Resin Solids (%wt.)	Adhesive Loading (%wt.)	Furnish Moisture Content with Resin (%wt.)	Furnish Moisture Content after Drying (%wt.)
2	1% chitosan solution	1.0	1.0	99.0	6.1
3	Mixed 1 part of 2% chitosan solution and 2 parts of commercial UF resin	44.67	6.0 (0.0896 for chitosan & 5.91 for UF)	7.5	4.8
4	First loaded 2 parts of 1% chitosan solution onto furnish, dried, and then added 2 parts of commercial UF resin	1 for chitosan solution; 66 for UF	6.0 (0.0896 for chitosan & 5.91for UF)	11.9	6.1
5	First loaded 0.0896 parts of chitosan powder onto furnish and then added 5.91 parts of UF resin, based on resin solids	100 for chitosan and 66 for UF	6.0 (0.0896 for chitosan powder & 5.91for UF)	3.0	6.1
6	Mixed 1 part of 2% chitosan solution and 3 parts of commercial UF resin	50.0	6.0 (0.06 for chitosan, 5.94 for UF)	6.0	4.8
11	UF resin control	66.0	6.0	3.1	6.1

Table 7 - General particleboard manufacturing conditions

Board dimension	20 in x 20 in x 1/2 in (510 mm x 510 mm x 12.7 mm)
Board construction	Homogenous
Wood species	Commercial mixed species
Support	Caul plates on the bottom and on the top
Target mat MC	5 - 6% (after blending or after drying)
Wax emulsion	0.5% (solids on a dry wood basis)
Resin	Outlined in Table 6
Catalyst	0.5% ammonium chloride for UF resin (solids on resin solids)
Board target density	688 kg/m ³ (43 lb/ft ³) (OD basis)
Press temperature	185°C (surface of platen)
Total press time	180 seconds (daylight to daylight)
Press closing time	20 seconds
Degas time	20 seconds
Replicate	2

All particleboard panels were conditioned at 65% RH/23°C for more than three (3) weeks until equilibrium moisture content (EMC) was reached. The panels were then evaluated for internal bond (IB) strength, modulus of rupture (MOR), modulus of elasticity (MOE), and thickness swelling (TS)/water absorption (WA) after soaking for 24 hours in water, according to the ASTM D1037-06 standard.

To measure the IB strength, eighteen specimens were sampled from each board, and the average density of all specimens measured was used as an estimated board density. Eight (8) specimens out of the eighteen (18) having a density close to the average board density were selected to measure the IB strength.

5. RESULTS AND DISCUSSION

5.1. Solubility, pH, and Viscosity of Chitosan Solutions

The test results showed that chitosan did not dissolve in a solution containing 1% of 1N acetic acid (v/v), and that it only partially dissolved in solutions containing between 2% and 4% of 1N acetic acid to form a milky solution. However, a solution containing 6% of 1N acetic acid could be used to completely dissolve 2% chitosan, while a solution containing 8% of 1N acetic acid could be used to completely dissolve 4% chitosan. A clear and transparent chitosan solution could be produced using a solution containing 10% of 1N acetic acid. Applying heat did not help to dissolve chitosan, but gentle stirring was required. Vigorous stirring produced many small bubbles in the solution, and these bubbles required at least 12 hours to settle out.

The pH and viscosity of the chitosan solutions made with 10% 1N acetic acid increased with an increase in the chitosan content of the solution. The 1% chitosan solution had a pH of 3.97 and a viscosity of 300 cps, the 2% chitosan solution had a pH of 4.77 and a viscosity of 2300 cps, and the 4% chitosan solution had a pH of 5.53 with an immeasurably high viscosity.

5.2. Compatibility of Chitosan with Other Resin Materials

The results obtained when testing the compatibility of chitosan solutions with other resin materials showed that chitosan resin was not compatible with ammonium lignosulfonate solution, liquid PF resin, soybean resin, powdered PF resin, or soybean flour. When mixed with ammonium lignosulfonate solution or liquid PF resin, the chitosan precipitated out, whereas chitosan solution and soybean resin solution separated into two (2) distinct layers when allowed to settle after they were mixed together. Finally, PF powder or soybean flour could not be dissolved in chitosan resin solutions. However, chitosan solution was compatible with UF resin, PVA resin, or phenol crystals. The chitosan solution could be mixed well with UF resin or PVA resin and produced a creamy liquid; it also dissolved phenol crystals to yield a clear, transparent mixture before it was oxidized by laccase.

5.3. Bond Quality of Fungus-Modified Chitosan Adhesives in Plywood Manufacturing

The lap-shear strengths of the plywood panels made with fungus-modified and unmodified chitosan (alone) adhesives are shown in Figure 15. Under the dry test conditions, plywood panels made with unmodified chitosan adhesive (C) had a lap-shear strength of 1100 kPa. However, the shear strengths of panels made with fungus-modified chitosan adhesives were significantly higher: 1318 kPa was observed for the resin with the chitosan modified by *Gliocladium roseum* (321U), 1698 kPa with the chitosan modified by *Trichoderma viride* (161D), and 1892 kPa with the resin prepared from the chitosan modified by *Trichoderma harzianum* (160D). Under the wet test conditions, the shear strengths of the panels were generally reduced compared with the results obtained for the same panels tested under dry conditions. However, the plywood panels made with the chitosan modified by *T.*

harzianum (160D) showed superior bonding properties compared to the panels made with the other adhesives.

The lap-shear strengths of the plywood panels made with UF adhesives reinforced with fungus-modified chitosan and unmodified chitosan are illustrated in Figure 16. The plywood panels made with a commercial UF adhesive had a lap-shear strength of 1129 kPa, which was similar to results obtained for the panels made with the unmodified chitosan adhesive. The unmodified chitosan improved the bonding strength of the UF adhesive (1298 kPa). The shear strengths of samples made with UF adhesives reinforced with the fungus-modified chitosan were greatly increased: 1655 kPa was observed for the resin reinforced by the chitosan modified with *G. roseum* (321U), 1818 kPa for that reinforced by the chitosan modified with *T. viride* (161D), and 1407 kPa for that reinforced by the chitosan modified with *T. harzianum* (160D). In this set of tests, plywood samples made with the UF adhesives with chitosan modified by *T. viride* (161D) had bonding properties superior to those of the other adhesives.

The lap-shear strengths of plywood samples made with PF adhesives enhanced with fungus-modified chitosan and unmodified chitosan are shown in Figure 17. The plywood samples bonded with a commercial PF adhesive had a lap-shear strength of 2633 kPa, which is higher than the results obtained for the panels made with the unmodified chitosan adhesives. The unmodified chitosan and chitosan modified by *T. viride* (161D) did not increase the bonding strength of the PF adhesive. The shear strengths of plywood panels made with the PF adhesive enhanced with the chitosan modified by *G. roseum* (321U) and *T. harzianum* (160D) were of 3128 kPa and 2998 kPa, respectively. In this set of tests, plywood panels made with a PF adhesive enhanced with the chitosan modified by *G. roseum* (321U) had superior bonding properties compared to the other adhesives.

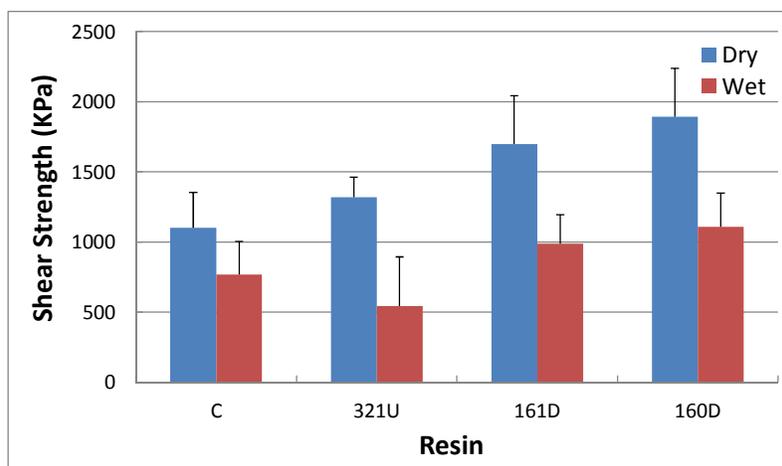


Figure 15. Shear strength of plywood panels made with adhesives containing fungus-modified chitosan

(C = adhesive made of unmodified chitosan; 321U = adhesive made of chitosan modified by fungal species *Gliocladium roseum* (321U); 160D = adhesive made of chitosan modified by fungal species *Trichoderma harzianum* (160D); 161D = adhesive made of chitosan modified by fungal species *Trichoderma viride* (161D). Dry = dry test; Wet = tested after soaking specimens in water for 48 hours. Data were gathered from an average of 8 specimens for fungus-modified chitosan adhesives and 16 specimens for unmodified control chitosan adhesive per treatment)

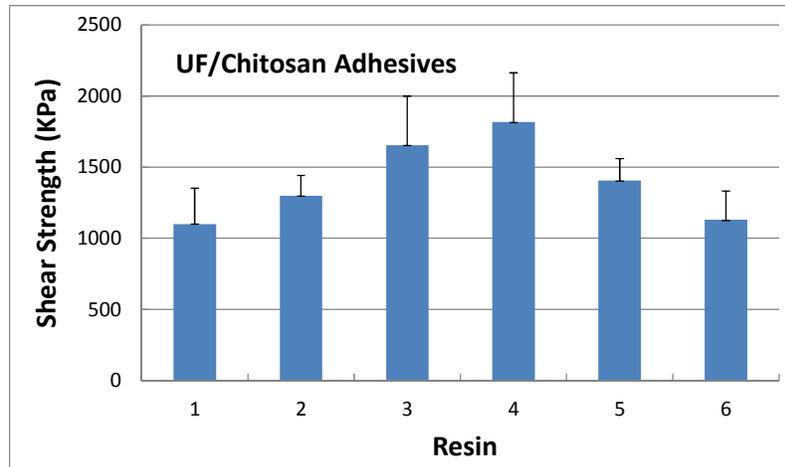


Figure 16. Shear strength of plywood panels made with UF adhesives enhanced with fungus-modified chitosan

(1 = adhesive made with unmodified chitosan; 2 = UF adhesive reinforced with unmodified chitosan; 3 = UF adhesive enhanced with chitosan modified by fungal species *Gliocladium roseum* (321U); 4 = UF adhesive enhanced with chitosan modified by fungal species *Trichoderma viride* (161D); 5 = UF adhesive enhanced with chitosan modified by fungal species *Trichoderma harzianum* (160D); 6 = commercial UF adhesive. Data were gathered from an average of 16 specimens per treatment)

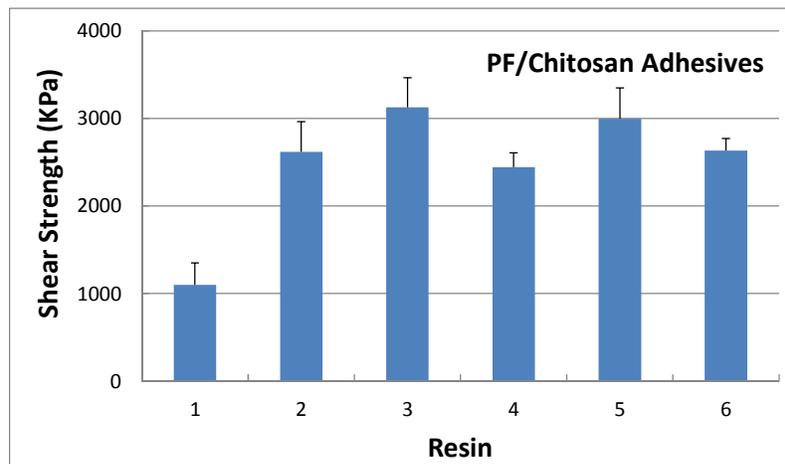


Figure 17. Shear strength of plywood panels made with reinforced PF adhesives containing fungus-modified chitosan

(1 = adhesive made with unmodified chitosan; 2 = PF adhesive enhanced with unmodified chitosan; 3 = PF adhesive enhanced with chitosan modified by fungal species *Gliocladium roseum* (321U); 4 = PF adhesive enhanced with chitosan modified by fungal species *Trichoderma viride* (161D); 5 = PF adhesive enhanced with chitosan modified by fungal species *Trichoderma harzianum* (160D); 6 = PF commercial adhesive. Data were gathered from an average of 16 specimens per treatment)

5.4. Bond Quality of Enzyme-Modified Chitosan Adhesives in Plywood Manufacturing

The dry shear test results of 2-ply plywood panels made with twelve (12) different types of chitosan-based resins are given in Table 8. The findings can be summarized as follows:

- 1) 2% chitosan resin (A) was superior to UF resin (J), but weaker than PVA (K) and PF (L) resins.
- 2) 4% chitosan resin (B) was similar to PF resin (L).
- 3) 2% chitosan resin plus tyrosinase (C) was superior to 2% chitosan resin alone (A) at a lower loading level (9 g/m²).
- 4) 2% chitosan resin plus phenol (2%) and laccase (D) was similar to PVA resin (K) at a higher loading level (17 g/m²).
- 5) Chitosan modified with enzymes extracted from either 321U (E) or from 161D (F) was weaker than 2% chitosan resin alone (A), due to over-degradation.
- 6) 2% chitosan resin plus UF resin at a ratio of either 2:1 (G) or 1:2 (H) were similar (G is slightly better than H) to PF resin (L).
- 7) 2% chitosan resin plus PVA resin at a ratio of 2:1 (I) was similar to PVA resin at a higher loading level (48 g/m²).

The wet shear strengths of the 2-ply plywood panels made from the twelve (12) different types of chitosan-based resins are given in Table 9. The results can be summarized as follows:

- 1) 2% chitosan resin (A) and 4% chitosan resin (B) were similar; both were superior to UF resin (J), similar to PVA resin (K), and weaker than PF resin (L).
- 2) 2% chitosan resin plus tyrosinase (C) did not improve the bond quality for 2% chitosan resin alone (A).
- 3) 2% chitosan resin plus phenol (2%) and laccase (D) was similar to PF resin (L), at a higher loading level (17 g/m²).
- 4) 2% chitosan resin plus UF resin at ratios of either 2:1 (G) or 1:2 (H) were similar (H was slightly better than G); both were better than UF (J), PVA (K), and PF (L);
- 5) 2% chitosan resin plus PVA resin at a ratio of 2:1 (I) was superior to UF (J) and PVA (K) resins, but similar to PF resin (L).

The dry and wet strengths of the 2-ply plywood panels are illustrated in Figures 18 and 19 for two different resin spread rates. For all 2-ply plywood panels, the dry strength was found to be much higher than wet strength. With the addition of chitosan to the UF resin (G and H), both the dry and wet strengths of the plywood panels were improved, compared to panels made with the control UF resin (J). Addition of chitosan to PVA resin (I) also improved the wet strength of the panels, compared to those made with the control PVA resin (K).

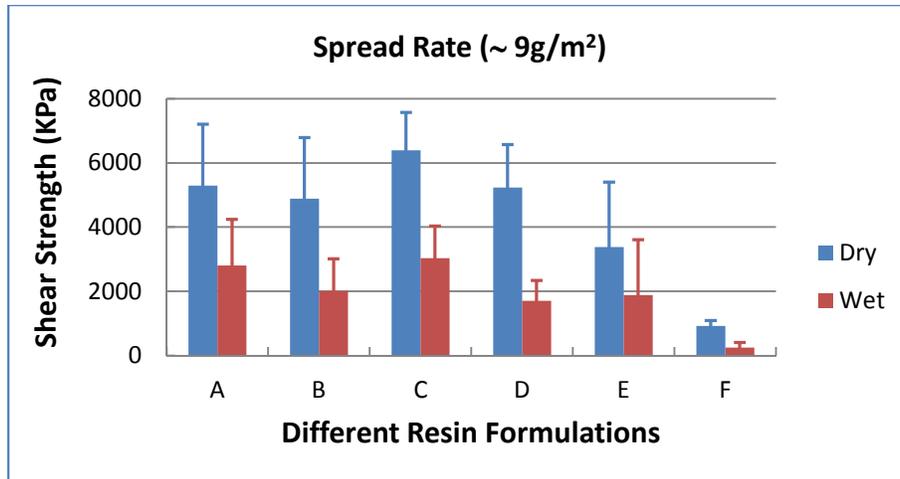


Figure 18. Shear strength of 2-ply plywood panels made with chitosan-based formulations, with a spread rate of about 9 g/m²

(A: 2% chitosan; B: 4% chitosan; C: chitosan (2%) + tyrosinase; D: chitosan (2%) + phenol (2%) + laccase; E: chitosan (2%) modified with crude enzyme extract from 321U; F: chitosan (2%) modified with crude enzyme extract from 161D)

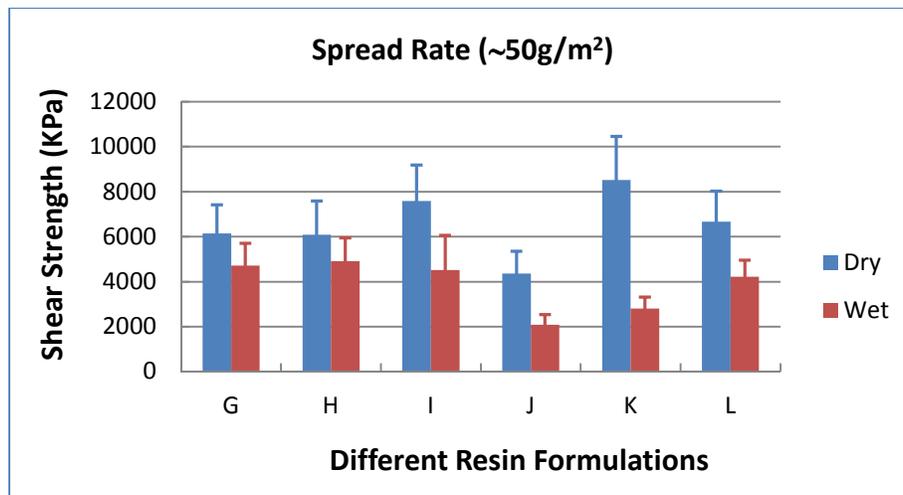


Figure 19. Shear strength of 2-ply plywood panels made with chitosan-based formulations with a spread rate of about 50g/m²

(G: 2% chitosan + UF(2:1); H: 2% chitosan + UF(1:2); I: 2% chitosan + PVA (2:1); J: UF alone; K: PVA alone; L: PF alone)

Table 8 - Lap-shear test results of 2-ply plywood panels made with different types of chitosan-based resins (dry strength)

Resin Type	Resin Content (g/mL)	Resin Spread Rate (g/m ²)	Lap-Shear Strength		Wood Failure	
			kPa	SD	%	SD
Chitosan (2%)	0.02	8.7	5881	2298	19	28
		8.9	4696	1540	15	27
		12.7	4768	1655	18	27
		5.7	4442	691	7	23
Chitosan (4%)	0.04	9.5	4520	1424	12	6
		9.5	5244	2398	22	29
		18.1	6478	1034	10	19
		18.3	6799	1501	43	41
Chitosan (2%) + tyrosinase	0.02	8.9	6211	980	0	0
		9.0	6584	1375	2	4
		17.4	2249	553	0	0
		17.3	4311	2361	0	0
Chitosan (2%) + phenol (2%) + laccase	0.02 + 0.02	8.5	3676	1234	6	14
		8.4	6799	1444	6	7
		16.6	9251	1975	43	30
		17.2	-	-	-	-
Chitosan (2%) modified with enzyme (321U)	0.02	8.2	3632	1923	8	23
		8.7	3115	2130	8	23
		12.7	3614	1402	0	0
		12.3	3577	1220	3	6
Chitosan (2%) modified with enzyme (161D)	0.02	8.6	1463	330	0	0
		8.4	373	0	0	0
		4.4	1900	533	0	0
		4.6	2061	659	0	0
Chitosan (2%) + UF resin (2:1)	0.013+0.233	24.0	6809	1678	15	19
		25.0	5632	750	49	38
		51.2	6187	1501	90	14
		53.2	6106	1037	88	19
Chitosan (2%) + UF resin (1:2)	0.007+0.447	57.2	6547	1894	75	45
		55.5	5616	1104	93	26
		109.1	5235	1494	61	49
		107.6	5490	1059	100	0
Chitosan (2%) + PVA resin (2:1)	0.013+0.167	17.6	3438	1285	8	29
		17.9	2622	1234	0	0
		50.2	6807	1797	93	11
		48.2	8360	1412	53	36
UF resin control	0.66	48.7	4231	965	14	25
		59.8	4479	1031	21	34
		106.4	4168	1555	4	9
		105.1	3591	1103	0	0
PVA resin control	0.5	50.0	8312	2199	62	42
		52.9	8722	1675	100	0
		106.1	9291	1353	100	0
		104.2	7985	1509	100	0
PF resin control	0.5	53.1	6689	1854	100	0
		52.5	6648	849	100	0
		103.6	6600	1416	100	0
		101.0	5910	1141	100	0

Note: Each strength value listed is the average of the results obtained on 12 test specimens.

Table 9 - Lap-shear test results of 2-ply plywood panels made with different types of chitosan-based resins (wet strength)

Resin Type	Resin Content (g/mL)	Spread Rate (g/m ²)	Lap-Shear Strength	
			(kPa)	(SD)
Chitosan (2%)	0.02	8.7	3466	1794
		8.9	2135	1086
		12.7	2022	654
		5.7	1994	425
Chitosan (4%)	0.04	9.5	1591	793
		9.5	2435	1208
		18.1	2645	336
		18.3	3216	629
Chitosan (2%) + tyrosinase	0.02	8.9	2752	989
		9.0	3317	1001
		17.4	651	463
		17.3	2671	1276
Chitosan (2%) + phenol (2%) + laccase	0.02 + 0.02	8.5	1126	324
		8.4	2280	953
		16.6	5165	862
		17.2	4491	877
Chitosan (2%) modified with enzyme (321U)	0.02	8.2	2518	2078
		8.7	1234	1385
		12.7	1454	959
		12.3	2448	1563
Chitosan (2%) modified with enzyme (161D)	0.02	8.6	189	46
		8.4	308	262
		4.4	55	29
		4.6	419	137
Chitosan (2%) + UF resin (2:1)	0.013+0.233	24.0	3782	1208
		25.0	4167	574
		51.2	5088	981
		53.2	4353	1000
Chitosan (2%) + UF resin (1:2)	0.007+0.447	57.2	5456	1246
		55.5	4375	810
		109.1	5184	1255
		107.6	4511	861
Chitosan (2%) + PVA resin (2:1)	0.013+0.167	17.6	1071	418
		17.9	1315	964
		50.2	4996	2383
		48.2	4051	678
UF resin control	0.66	48.7	1929	486
		59.8	2228	416
		106.4	1599	640
		105.1	2311	881
PVA resin control	0.5	50.0	2807	609
		52.9	2799	407
		106.1	2694	293
		104.2	2849	322
PF resin control	0.5	53.1	4152	837
		52.5	4282	644
		103.6	4604	868
		101.0	4036	1221

Note: Each strength value listed is the average of the test results obtained for 12 test specimens after soaking for 48-hour in water.

5.5. Production of Chitosan Degrading Enzymes by Different Fungal Species

The chitosan-enriched agar medium appeared as a yellowish opaque medium. When a fungus is capable of producing chitosanase to degrade the chitosan in the medium, it can form a clear zone on the medium surrounding the fungal colonies (Figure 20). The greater the distance between the edges of the mycelia growth and of the clear zone, the stronger the chitosanase released by the fungus. On the chitosan-enriched agar medium, among the three (3) fungal species tested, *Gliocladium roseum* grew faster than the other two (2) species; *Trichoderma viride* grew at a moderate rate, and *Trichoderma harzianum* grew more slowly (Table 10). However, when measuring the distance between the edges of the mycelia growth and the clear zone, it was found that *T. viride* produced the largest clear zone, followed by *T. harzianum* (Table 11). *G. roseum* also produced chitosanase, but the enzyme did not migrate away from the mycelia and the distance between the edges of the mycelia growth and the clear zone was very short. This phenomenon may be due to the presence of different chitosanases or a difference in the degrading patterns involved in this species, compared to the other two fungi. In general, chitosan degrading enzymes produced by *T. viride* are stronger than the enzymes produced by the other two (2) fungal species.



Figure 20. Chitosanase test on chitosan-enriched agar medium

Table 10 - Fungal growth and clear zone formation on a chitosan-enriched agar medium

Fungal Code	Fungal Species	Fungal Colony Diameter (mm)				Clear Zone Diameter (mm)			
		7 Days		14 Days		7 Days		14 Days	
		Average	SD	Average	SD	Average	SD	Average	SD
161D	<i>Trichoderma viride</i>	10.7	0.5	20.8	0.7	18.4	0.9	31.3	3.1
160D	<i>Trichoderma harzianum</i>	12.3	0.9	13.4	0.9	20.1	1.5	22.0	1.9
321U	<i>Gliocladium roseum</i>	38.1	0.8	40.0	0.0	38.7	1.0	40.3	0.7

Table 11 - Chitosanase release from fungi on a chitosan-enriched agar medium

Fungal Code	Fungal Species	Distance between Edges of Mycelia Growth and Clear Zone (mm)			
		7 Days		14 Days	
		Average	SD	Average	SD
161D	<i>Trichoderma viride</i>	7.8	0.7	10.6	2.9
160D	<i>Trichoderma harzianum</i>	7.8	1.0	8.6	2.1
321U	<i>Gliocladium roseum</i>	0.6	0.9	0.3	0.7

5.6. Preparation of Chitosan-UF Resins by Addition of Chitosan during the Synthesis Process

5.6.1. Synthesis of Control UF Resin

The control UF resin (Resin C01) was synthesized following the procedure given in Scheme-A in Figure 12. The resins produced were characterized by their pH, viscosity, and solids contents. The test results are outlined below:

- pH: 7.08 (measured immediately at the end of the synthesis process)
6.95 (measured after two weeks of storage at room temperature)
- Viscosity: 530 cps (measured immediately at the end of the synthesis process)
230 cps (measured after two weeks of storage at room temperature)
- Solids content: 55.8%

5.6.2. Synthesis of UF Resin with Unmodified Chitosan Solution (R01)

The UF resin containing unmodified chitosan was synthesized by adding the chitosan solution (R01) into the mixture of formaldehyde and urea (2:1, v/v, UF/chitosan solution), after reaction at pH 7.2, at 85°C for 20 min, and then adjusting the pH to around 5.6 (Scheme-B in Figure 12). However, this procedure was not suitable, because gelation of the reaction mixture took place very soon after addition of the unmodified chitosan solution (Figure 21).



Figure 21. Gelation of unmodified chitosan solution with the mixture of formaldehyde and urea at an early stage of the UF synthesis process

5.6.3. Compatibility of Chitosan Solutions with 37% Formaldehyde Solution and Water

Because of the unsuccessful synthesis of UF resin with unmodified chitosan (R01), the compatibility of chitosan solutions with a 37% formaldehyde solution (pH 2.79) and distilled water was examined (Figure 22). The test results showed that when the unmodified chitosan solution was added to the 37% formaldehyde solution, a transparent gel substance was formed. This shows that an unmodified chitosan solution cannot be well mixed with a 37% formaldehyde solution, and perhaps that a reaction between the unmodified chitosan and formaldehyde is taking place. This result can explain the gelation observed in the synthesis of UF resin with unmodified chitosan. The unmodified chitosan solution could be mixed well with distilled water, but the viscosity of the diluted solution was still very high. When the two modified chitosan solutions (R02 and R03) were tested, it was found that both solutions could be mixed very well with 37% formaldehyde solution or distilled water.

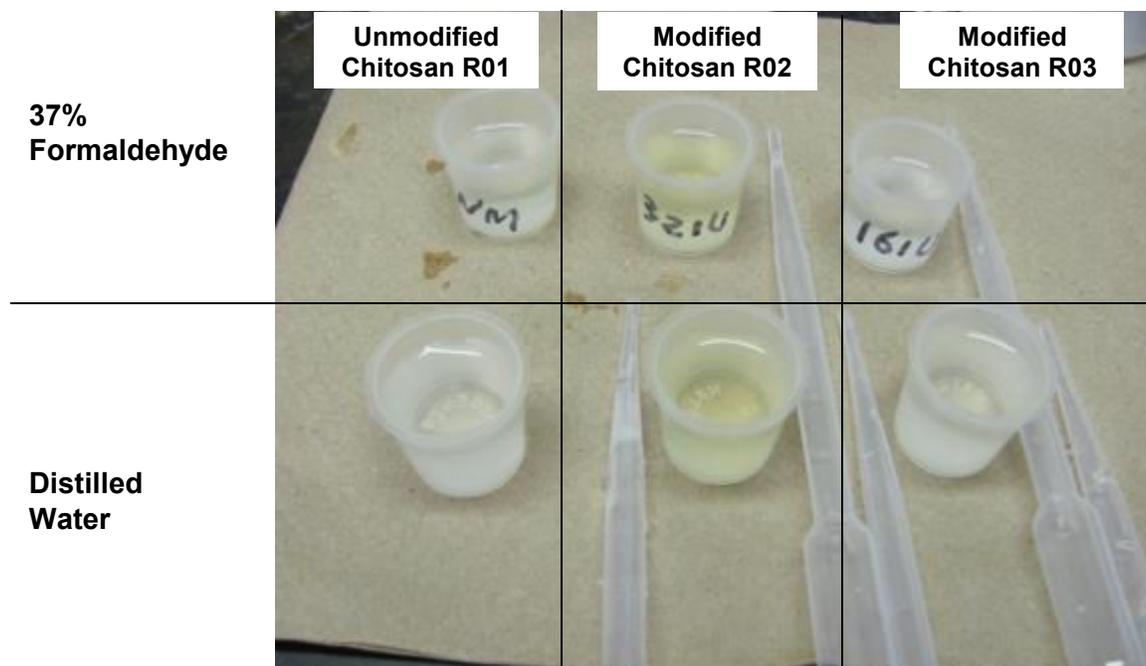


Figure 22. Observation of compatibility of chitosan solutions with a 37% formaldehyde solution and distilled water

5.6.4. Compatibility of Chitosan Solutions with UF Reaction Mixtures at Three Different Condensation Stages of the UF Synthesis Process

Based on the tests above and observations made during the resin synthesis, another batch of UF control resin was synthesized to determine the compatibility of chitosan solutions with the UF reaction mixture at three different stages of the UF condensation process. During the condensation process, three (3) 10-mL samples of the UF reaction mixture were taken out at different stages (Stage-1, Stage-2, and Stage-3 as shown in the Scheme in Figure 13); these were added to 5 mL of chitosan solution (R01, R02, or R03), the mixtures shaken vigorously, and observed for compatibility and viscosity change.

For the unmodified chitosan solution (R01), the viscosity of the mixture increased immediately after adding the chitosan solution to the UF reaction mixture, and the mixture formed a soft, but uncured gel-like substance, at all three stages of the UF condensation process. On the other hand, the two modified chitosan solutions (R02 and R03) could be well mixed with the UF reaction solutions, at all three stages. The mixtures had not gelled even after 1-2 hours of continuous heating in a boiling water bath.

Based on these preliminary observations, it seems that the 2% unmodified chitosan solution is not suitable for addition during the UF synthesis process, whereas the modified chitosan solutions could be added to the UF reaction mixture during the process.

5.6.5. Synthesis of UF Resin with Modified Chitosan Solution R03

The test results for the pH and viscosity of the chitosan solutions and the UF/chitosan resins are given in Table 12 and Table 13, respectively. Since the modified chitosan solutions could be well mixed with the UF reaction mixture, an additional synthesis was conducted to observe the trend in the variation of pH and viscosity following the addition of the modified chitosan R03, during the initial condensation

stage (Stage-1) of the UF synthesis process (Figure 14). At the initial condensation stage, the pH was controlled at 5.8. After the 2% modified chitosan solution (in 1% acetic acid) was added, the pH dropped to 4.2, and then was adjusted to 5.6 using 25% NaOH. The condensation conditions were maintained at pH 5.6 and a temperature of 98-99°C for three hours: no increase in the viscosity was observed. Since UF condensation is known to take place more quickly at a lower pH, and thus should result in a faster increase in the viscosity, the pH of the reaction mixture was adjusted to 4.9 with 20% NH₄Cl. However, the viscosity did not increase after another three hours of reaction at 98-99°C. The viscosity was of only 8.6 cps after six hours of reaction (three hours at pH 5.6 and three hours at pH 4.9). The fact that the viscosity did not increase during the UF condensation process might be mainly due to dilution, i.e. water was introduced with the addition of the modified chitosan solution to the UF reaction mixture, which reduced the UF concentration significantly and, therefore, decreased the reaction rate of the condensation process. Thus, the absence of an increase in viscosity during the UF synthesis process could be due to a significantly reduced reaction rate.

Table 12 - pH and viscosity of the three chitosan solutions

Characterization	R01	R02	R03	1% Acetic Acid
pH	4.92	5.72	5.06	2.81
Viscosity (CP, 25°C)	1880	1.4	1.6	-

Table 13 - pH and viscosity of UF/chitosan resins

Characterization	UF (C01)	UF-R01	UF-R02	UF-R03	UF-1% Acetic Acid
pH	6.95	5.33	5.75	5.37	3.71
Viscosity (CP, 25°C)	230	900	18.6	17.5	17.0

It was observed that the viscosity of the 2% modified chitosan solutions R02 and R03 (in 1% acetic acid) was very low (< 2 cps) (Table 12). After adding one part of R02 or R03 to two parts of UF resin, the viscosity of the reaction mixture decreased significantly from 230 to about 18 cps (Table 13). This was similar to adding 1% acetic acid to the UF resin. The viscosity of the 2% unmodified chitosan solution R01 (in 1% acetic acid) was very high (1880 cps) (Table 12). After adding one part of R01 to two parts of UF resin, the viscosity increased from 230 to about 900 cps (Table 13).

When the R-01 solution was added, the viscosity of the UF-R01 reaction mixture increased to about 900 cps immediately after mixing and continued to increase steadily even during the viscosity measurements. After two hours at room temperature, the resin became thicker and, in the next 48 hours, it slowly transformed into a soft gel. Therefore, it seems that unmodified chitosan solution could be used as a UF resin tackifier.

5.6.6. Gel Time of UF Resin (C01) Post-Blended with Unmodified Chitosan (R01)

A batch of post-blended UF-R01 resin (10.0 g of UF resin C01 mixed with 5.0 g of R01 solution in a glass vial) was heated in a boiling water bath to observe its gel time. It was found that the UF-R01 resin became a white paste-like material at about 10 minutes of heating, but did not transform into a cured hard state even after 20 minutes of heating. For comparison purposes, 10.0 g of UF resin C01 was mixed with 0.7 g of 20% NH₄Cl (a conventional hardener for UF resin) and was then heated under the

same conditions. The UF resin cured to form a hard material in 2-3 minutes. These observations indicate that gelation of UF resin C01 blended with R01 (2:1 w/w) did not occur or was very slow at 100°C. A higher concentration of acetic acid solution and/or the use of NH₄Cl as a resin hardening agent might be necessary to achieve good gelation (curing) with this resin system.

5.6.7. Lap-Shear Tests of Plywood Bonded with UF-Chitosan Resins Produced by Addition of the Chitosan during the Resin Synthesis Process

The test results for the lap-shear strength of plywood panels bonded with these UF-chitosan resins are summarized in Table 14, and illustrated in Figures 23 and 24. As observed before, all the plywood panels bonded with 0.5% chitosan-modified UF resin achieved through post-blending of unmodified chitosan (C2) showed average peak stress values that were 19% higher than those obtained for the panels made with the UF control resin. For the panels bonded with the resins made by addition of unmodified chitosan during the resin synthesis process, no improvement was observed with the R1A and R1B resins, whereas a negative impact (27% lower than the control) was observed with the R1C resin. Addition of fungus-modified chitosan during the synthesis process did not improve the resulting UF resins bonding performance; the panels produced with these resin systems displayed a 16% lower peak stress value, compared with panels produced with the control resin.

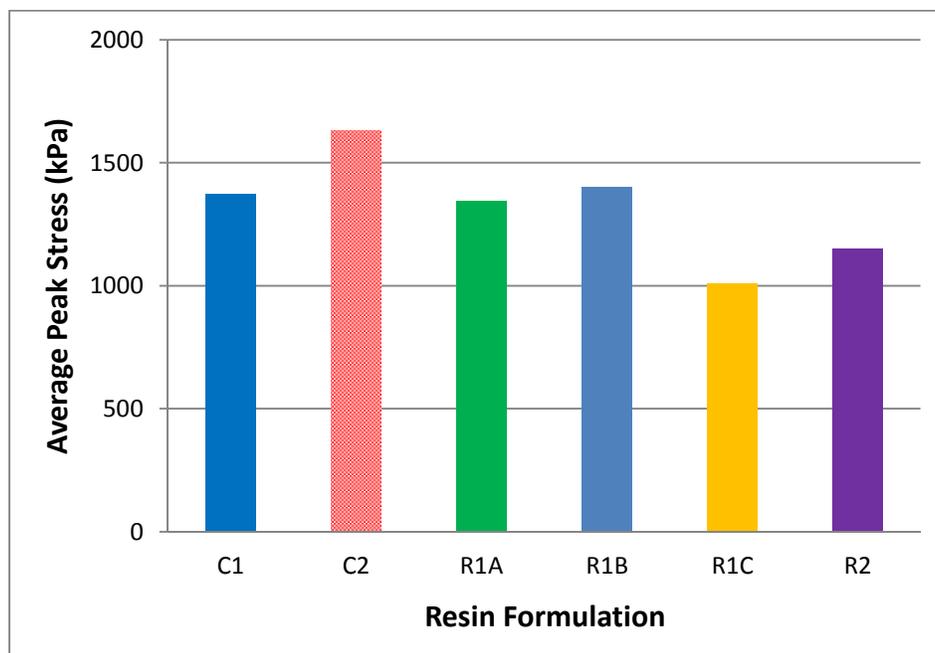


Figure 23. Average peak stress of four panels made with each resin formulation

(C1: UF control, F/U1 = 2:1, F/(U1+U2) = 1.12, and no chitosan added; C2: UF control, F/U1 = 2:1, F/(U1+U2) = 1.12, and chitosan fine powder added via post-blending; R1A: F/U1 = 2:1, unmodified chitosan added at viscosity G, and F/(U1+U2) = 1.12; R1B: F/U1 = 4:1, unmodified chitosan added with U1 at pH 7.2, F/(U1+U2) = 2:1, and F/(U1+U2+U3) = 1.12; R1C: unmodified chitosan added at pH 2.9, F/U1 = 4:1, F/(U1+U2) = 2:1, and F/(U1+U2+U3) = 1.12; R2: F/U1 = 2:1, fungus-modified chitosan added at viscosity G, and F/(U1+U2) = 1.12)

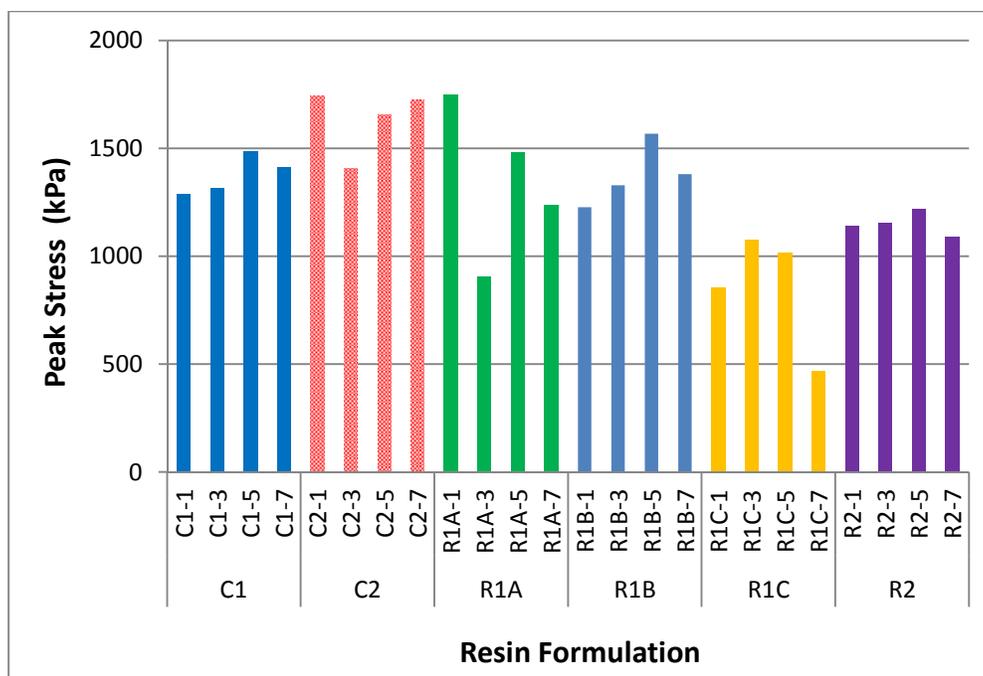


Figure 24. Peak stress of individual panel replicates made with each resin formulation

(C1: UF control, F/U1 = 2:1, F/(U1+U2) = 1.12, and no chitosan added; C2: UF control, F/U1 = 2:1, F/(U1+U2) = 1.12, and chitosan fine powder added via post-blending; R1A: F/U1 = 2:1, unmodified chitosan added at viscosity G, and F/(U1+U2) = 1.12; R1B: F/U1 = 4:1, unmodified chitosan added with U1 at pH 7.2, F/(U1+U2) = 2:1, and F/(U1+U2+U3) = 1.12; R1C: unmodified chitosan added at pH 2.9, F/U1 = 4:1, F/(U1+U2) = 2:1, and F/(U1+U2+U3) = 1.12; R2: F/U1 = 2:1, fungus-modified chitosan added at viscosity G, and F/(U1+U2) = 1.12)

Table 14 - Peak stress values of panels in lap-shear test

Specimen	Peak Stress (kPa)	Standard Deviation (kPa)	Difference
UF Control (C1)	1373	392	-
C2	1633	324	19%
R1A	1343	365	-2%
R1B	1400	393	2%
R1C	1009	388	-27%
R2	1149	415	-16%

5.7. Evaluation of Particleboard Panels Manufactured with Chitosan-UF Resins

The physical and mechanical properties of particleboard panels made with various chitosan-UF resins are presented in Table 15. The test results indicate that all formulations of chitosan-UF adhesives could produce boards with good visual quality, even those made with only 1% of chitosan resin, as shown in Figures 25-27.

All chitosan and chitosan-based UF resins yielded panels with better IB strength than the panels made with the UF control resin (Panel 11). The panels made with 1% chitosan resin plus 66% UF resin in a 1:1 ratio (Panel 4) had the highest IB strength among all of the panels tested.

The panels made with 1% chitosan resin alone (Panel 2) had a lower MOR value than the panels made with the UF control resin, whereas all the other panels manufactured with chitosan-UF resins had a similar or higher MOR than the panels made with the UF control resin. Again, the panels made with 1% chitosan resin plus 66% UF resin in a 1:1 ratio (Panel 4) had the highest MOR value.

The MOR properties of panels made with several resin systems could not be measured; however, among the panels that could be tested, those produced with 1% chitosan resin plus 66% UF resin in a 1:1 ratio (Panel 4) had the highest MOE value.

The panels made with 1% chitosan resin alone (Panel 2) had higher thickness swelling (TS) and water absorption (WA) than panels made with the UF control resin (Panel 11), whereas all the other panels prepared with chitosan-UF resins had a similar or lower TS and WA than the panels made with the UF control resin. The panels made with 0.0896 parts of chitosan powder plus 5.91 parts of UF resin (Panel 5) had the lowest TS and WA values.

Comparing all of the physical and mechanical properties of the particleboard panels made with these six (6) chitosan-UF resin systems, it was found that the panels made with 1% chitosan resin plus 66% UF resin in a 1:1 ratio (Panel 4) showed the best overall properties, followed by those made with 0.0896 parts of chitosan powder plus 5.91 parts of UF resin (Panel 5).

It should be noted that all of the particleboard panels tested, including the control panel bonded with a commercial UF resin, displayed poorer mechanical and physical properties than expected, which may likely be attributed to the lower resin content used for the homogeneous construction, in manufacturing the panels. However, the test results still showed that chitosan can be used to enhance UF resin performance in the manufacture of particleboard panels.

Table 15 - Physical and mechanical properties of particleboard panels made with chitosan-UF resins

Panel ID*	Board Moisture Content (%)	IB (MPa)	Area of Failure in IB Test	MOR (MPa)	MOE (MPa)	24-h TS (%)	24-h WA (%)
2	8.8	0.061	100% Face	2.73	1069	92.9	115.1
3	8.5	0.065	100% Core	7.08	-	77.1	100.7
4	8.4	0.111	100% Core	7.86	2279	64.7	92.0
5	8.5	0.080	100% Core	7.08	2217	59.1	82.4
6	8.8	0.097	100% Core	5.08	-	66.8	87.9
11	8.6	0.049	100% Core	5.78	-	76.4	100.2

*2 = 1% chitosan solution; 3 = 1 part 2% chitosan solution and 2 parts commercial UF resin; 4 = loading of 2 parts 1% chitosan solution onto furnish, drying, and then addition of 2 parts of commercial UF resin; 5 = loading of 0.0896 parts chitosan powder onto furnish and then addition of 5.91% UF resin based on resin solid; 6 = mixing of 1 part 2% chitosan solution and 3 parts of commercial UF resin, followed by application to furnish; 11 = UF resin control.



Figure 25. Particleboard made with 1% chitosan adhesive



Figure 26. Particleboard made with 1% chitosan resin and 66% UF resin in 1:1 ratio

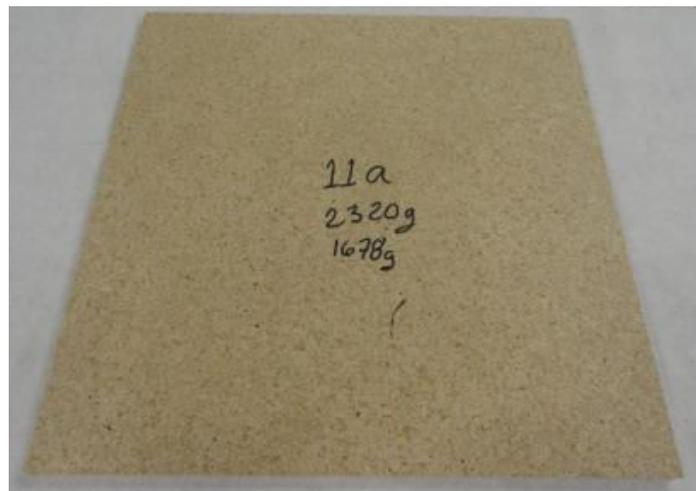


Figure 27. Particleboard made with 66% UF adhesive

6. CONCLUSIONS

Three (3) fungal species, *Gliocladium roseum* (321U), *Trichoderma viride* (161D) and *Trichoderma harzianum* (160D), were selected to modify chitosan. The lap-shear test results of plywood panels produced with the various resins showed that the panels bonded with the fungus-modified chitosan adhesives had significantly higher bonding strength than those bonded with the unmodified chitosan adhesives, in terms of both dry and wet shear strengths. The bonding properties of the adhesives made with the chitosan modified by *T. harzianum* had superior bonding properties compared to the other adhesives. Based on this work, a patent entitled “Fungal Modified Chitosan Adhesives and Wood Composites Made from the Adhesives” has been granted by the USA Patent Office (Patent No.: US 8,562,731 B2) and the Chinese Patent Office (Patent No.: ZL 2011 8 0012458.3).

The unmodified chitosan solution was not compatible with ammonium lignosulfonate solution, liquid PF resin, soybean resin, PF powder, or soybean flour, but was compatible with UF resin, PVA resin, and phenol crystals. Compared to the control UF and PVA resins, both dry and wet shear strengths of the plywood panels produced were improved with the addition of chitosan.

A series of tests were conducted to synthesize UF resin with biologically modified and unmodified chitosan. Significant progress has been made in the incorporation of chitosan into UF resin via in-situ polymerization. These new resin formulations were evaluated for performance through manufacturing and testing of plywood panels. The results showed that, among all chitosan-reinforced UF resin formulations, post-blending of unmodified chitosan resulted in the best bonding quality. No improvement in the bond quality was observed with the addition of unmodified chitosan during the resin synthesis process. The addition of fungus-modified chitosan during the resin synthesis process did not seem to improve the UF resin bonding performance either.

A number of chitosan and chitosan-reinforced UF resins were prepared and used as a binder for particleboard panel manufacturing. Six (6) types of particleboard panels produced with different levels of resin loading and under different pressing conditions were manufactured and evaluated to determine the bond quality of the chitosan and chitosan-reinforced UF resins. The test results showed that all formulations of chitosan-UF adhesives could produce particleboard panels with good visual properties, even those made with 1% of chitosan resin alone. Particleboard panels produced with all of the chitosan resins, alone or added to UF resin, had a better IB strength than the panels made with the UF control resin. The panels made with 1% chitosan resin plus 66% UF resin in a 1:1 ratio had the highest IB strength and best overall mechanical properties.

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