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Flavonoid Biocides: Wood Preservatives Based on Condensed Tannins

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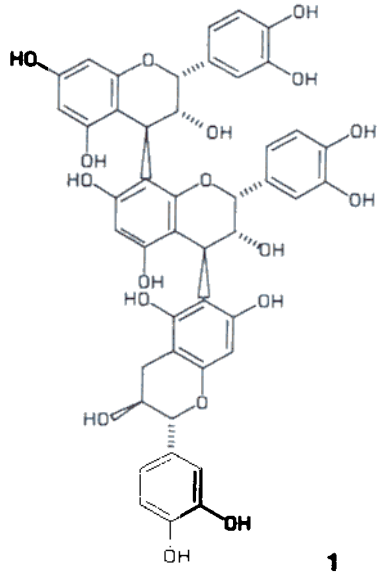
Flavonoid Biocides: Wood Preservatives Based on Condensed Tannins

Summary

The condensed tannins are natural wood preservatives found in high concentrations in the bark and wood of some tree species. Condensed tannin-containing bark extracts from loblolly pine *(Pinus taeda)* were evaluated as wood preservatives using standard methods. Bark extracts by themselves did not cause any reduction in weight loss of pressure-treated wood blocks at the retentions tested. However, they do have efficacy as wood preservatives when complexed with copper (II) ions. The best experimental wood preservative formulation was a dual treatment using a sulphited bark extract first, followed by a $CuCl_2$ treatment. At some retentions, this method yielded wood blocks with greater resistance to decay by *Coriolus versicolor* than pentachlorophenol. A single stage treatment of extract plus copper using an aqueous ammoniacal solvent was also successful but not as effective as the dual treatment.

Introduction

Condensed tannins are hydroxylated aromatic polymers based on a 15-carbon flavan monomer unit. These polymeric proanthocyanidins are found in large quantities in tree barks, seed coats and other plant protective tissues. A generalized structure for the 3,3′,4′,5,7-pentahydroxy (procyanidin-type) condensed tannin found in loblolly pine bark is shown as structure 1 (Hemingway *et al.* 1982; Hemingway *et al.* 1983). Two different types of interflavanoid bonds are present, (4ß–>8) and 4ß–>6), in relative proportions of about 3 to 1, respectively. In an acetone/water extract these tannins have a number average molecular weight of 2,500, to 3,000 (about 9 flavanoid units) and dispersivity ranging from 2 to 4 (Karchesy and Hemingway 1980; Williams *et al.* 1983). The upper units of the tannin have an epicatechin structure with 2,3-*cis* stereochemistry, while the lower terminal unit is derived from catechin. This general structure is applicable to tannins that can be extracted from most species of the Pinaceae (Samejima and Yoshimoto 1982).



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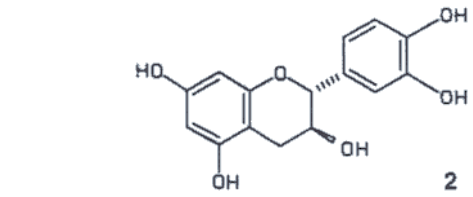
Condensed tannins are natural preservatives and antifungal agents (Zucker 1983). This is the reason for their presence in the outer tissues of plants – to help protect against attack by a variety of pathogens including fungi. Most plant-pathogenic fungi excrete extracellular enzymes such as cellulases and lignases to break down plant tissues. Condensed tannins most likely act as inhibitors of these enzymes by complexing with these proteins to block their action.

The practical use of tannin-containing extracts from various woods and barks as preservatives has a long history. For example, the extract from the heartwood of *Acacia catechu* has been used in India as a preservative for fabrics and fishing nets. This extract is termed "catechu" and has been found to contain a large pro-

portion of low molecular weight flavonoids, catechin (2) in particular (Howes 1953).



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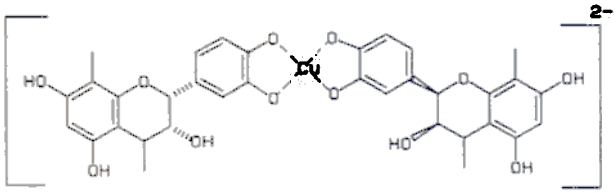

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The ability of condensed tannins to complex with metal ions is well known, presumably giving a chelated structure utilizing the catechol rings of the phenolic monomers (3). This chelating property is utilized in the work described here to formulate copper/tannin wood preservatives. Copper metal and ions are toxic to most lignocellulose destroying organisms. However, to make the metal useful as a practical wood preservative, it is usually complexed with an organic ligand which also has some toxicity (e.g. copper 8-quinolinolate), or fixed in an inorganic system such as CCA. In one study, fish nets treated with tannins were exposed to Cu^{++} solutions, resulting in a copper/tannin complex having good preservative qualities (Cecily and Kunjappan 1973). Cotton duck was treated with solutions of catechu or an unspecified-source tannin, followed by immersion in various metal salt solutions (Furry and Humfield 1941). The catechu plus $CuSO_4/NH_4OH$ treatment resulted in an entirely decay resistant fabric at a treatment level lower that used for sodium pentachlorophenates or metal naphthenates to achieve the same level of protection. Tannin, without fixation and augmentation by a metal ion, was not as effective.

In this paper we describe the behavior of condensed tannin-containing loblolly pine bark extracts and related compounds as wood preservatives, alone and after complexation with Cu(II) ions.

Materials and Methods

1. Preparation of Loblolly Pine Bark Extracts

Two different samples of bark were used for preparation of extracts. One sample of southern pine bark was taken directly off the transfer chain from the debarker at Boise Cascade Corporation's plywood mill at Oakdale, Louisiana. This bark was obtained from logs of comparatively old, large-diameter trees that had been attacked by southern pine beetles. The second sample of bark was obtained by hand-peeling pole-sized logs from trees cut from the Kisatchie National Forest in Central Louisiana. Both bark samples were air-dried and then reduced to a fine particle size by, first passing the bark through a garden mulcher, and then refining it twice in a Sprout-Waldrin disk refiner fitted with breaker plates. The particle size distribution of the material used for extraction is shown in Table 1.

Table 1. Particle Size Distribution of Bark Used in Extraction

Retained on Screen Mesh Size	Percent of Bark Weight Retained on Screen
4	0.7
4–8	17.4
8–16	26.4
16–20	11.4
20–35	14.3
35–45	8.1
>45	21.6

The ground bark was extracted in lots of approximately 30 lbs in a 40 gallon stainless steel tank fitted with stainless steel steam-heating and cold water-cooling coils. The temperature was controlled using an immersion temperature probe connected to control valves for steam and cold water. For the sulfite extractions, the bark was extracted with 4% sodium sulfite and 0.4% sodium carbonate (on dry bark weight basis) at a liquor to bark ratio of 7:1 for 2 hours at 95–100°C using about 1 hour to reach temperature. The suspension was stirred intermittently and the liquor adjusted to a constant volume by periodic addition of water. At the end of the extraction time, the suspension was allowed to cool, adjusted to constant volume, and the liquor drained through a fiberglass mat in the base of the tank. The wet pulp was pressed to recover as much of the liquor as possible, typically 55–65% of the liquor added. The volume of recovered liquor was measured.

The extract liquor was filtered a second time through either sharkskin or a mat of fine glass wool to remove small amounts of fine particulates. To obtain an estimate of extract yield, aliquots of 100 or 200 ml were freeze-dried to establish the solids content of the recovered liquor and the total yield of dissolved solids was corrected by dividing by the proportion of liquor recovered and assuming that the bark pulp could be dewatered to 50% moisture content in a commerical process. Total extract yield varied from 16.3% for the bark from the plywood mill (LPBSE1) to 25.1% for the bark obtained from the pole-sized logs (LPBSE2) (Table 2). The extracts obtained from the bark from the plywood mill were combined to give approximately 100 gallons of extract that was dried by a hot-pan evaporation method.

The extract obtained from bark of the pole-sized logs was dried in a vacuum-pan evaporator. Yields for reducing sugars after hydrolysis (measured by quantitative liquid chromatography methods after Klason hydrolysis conditions) and Stiasny phenol content are shown in Table 2. Approximately 8–10% of the extract was accounted for as recovered sodium and sulfur (Table 2). Elemental analyses were provided by Galbraith Laboratories.*

The acetone-water extract (LPBAWE) was made from bark obtained from the plywood mill by extraction at 50°C for 2 hours with acetone/water (1:1 v/v) at a liquor to bark ratio of 9:1. The slurry was stirred frequently and, after the appropriate extraction time, the solution was adjusted to a constant volume and cooled. Approximately 57 liters of extract was recovered from a total liquor volume of 93 liters. It was filtered through sharkskin to remove fine

* Mention of trade names does not constitute endorsement by the US Department of Agriculture.

Table 2. Properties of Extracts Obtained From Loblolly Pine Bark

Property	LPBSE1[1)]	LPBSE2[1)]	LPBAWE[2)]
Extract Yield	16.3	25.1	8.8
Stiasny Polyphenols	59	81	83
Carbohydrates after Acid Hydrolysis			
Glucose	6.6	4.2	
Xylose	1.7	2.6	
Galactose	3.5	2.4	
Arabinose	1.7	1.7	
Mannose	1.2	1.9	
Total Carbohydrate	14.8	12.7	
Inorganics			
Sodium	5.8	4.5	
Sulfur	3.8	3.1	

[1)] Sulphited extract of loblolly pine bark
[2)] Acetone/water extract of loblolly pine bark

particulates. The acetone was removed under reduced pressure on a Bucchi evaporator and the final suspension in water amounted to 26.3 liters at 2.63% solids content. The projected extract yield considering the pulp could be dewatered to 50% moisture content amounted to only 8.8% of the bark dry weight. The Stiasny polyphenol content of this extract was 82%. Because of the low extract yield, the composition of the extract was not examined further.

2. Preparation of Treatment Formulations and Treated Wood Samples

Treatment solutions were formulated in water except for the pentachlorophenol standard for which ethanol was used. Concentrations in the solvent are given within Tables 3 to 6. Ammoniacal copper/bark extract formulations were prepared using the minimum amount of concentrated NH_4OH necessary to keep the components in solution. Two molar ratios of tannin monomer unit to Cu(II) were used – 2:1 and 1:1. They were prepared by making an ammoniacal solution of $CuSO_4$ and adding it to an appropriate amount of the aqueous solution of the bark extract to give the correct ratio of components. A 75% tannin content in the extracts was assumed. The amount of concentrated NH_4OH in the final solution was 20% for the acetone/water bark and 10% for the sulphited extract. The sulphited extract used in these ammoniacal solutions was Extract II. In the two-stage treatments the concentrations of the tannin monomers and Cu(II) in their respective solutions was approximately equal.

Treated blocks were prepared using a full-cell method. Wood blocks were submerged in a beaker of the test formulation, exposed to a partial vacuum of 25 mm Hg for 30 min, then pressurized at 70–90 psi for 40 min. The blocks were allowed to fix by sealing them in a plastic bag for 7 days, then air-drying for an additional week.

The blocks were weathered and leached according to NWMA standard M–1–81. Briefly, this involves storing the blocks in a forced-air oven at 49°C for 14 days. On nine of these days the blocks are removed and soaked in distilled water for 2 hours.

3. Soil-Block Testing Procedure

Experimental formulations were evaluated against wood decay fungi in accordance with ASTM D1413-81: Standard Method of Testing Wood Preservatives by Laboratory Soil Block Cultures. In this procedure, sets of five 19 mm blocks impregnated with preservative are exposed to actively growing cultures of known wood decay fungi in soil jars. After 3 month incubation, the test blocks are removed from the jars and any adhering fungus removed. The blocks are then air dried, oven dried, and re-weighed. Preservative performance is evaluated by the average percent weight loss calculated for each block set. Two fungi were used in these tests – *Gloeophyllum trabeum* (brown-rot, ATCC #11539) and *Coriolus versicolor* (white-rot, ATCC #12679).

4. Soft-Rot Testing Procedure

Experimental formulations were evaluated for effectiveness against soft-rot fungi using an unsterile soil or "soil burial" test. This method is a modification of ASTM D1413 described above. Treated blocks are incubated in jars of unsterile soil moistened to an appropriate level to encourage the development of naturally occuring soft rot fungi. Soil jars are prepared with the soil moisture content adjusted to 100% of the water holding capacity. Birch blocks (30 × 10 × 6 mm) are treated and prepared as described for ASTM D1413, but are not sterilized. The blocks are added to the jars by pushing them into the soil, lengthwise, until completely covered and then incubated for two months. Preservative performance is evaluated by the average percent weight loss of each block set.

5. Fractionation of Sulphited Bark Extract

The second sulphited bark extract (LPBSE2) was divided into tannin-rich and tannin-poor fractions by dissolving 10 g of the extract in 20 ml of water then adding dropwise 20 ml of 100% ethanol to the stirred aqueous solution. After stirring for 30 min. the suspension was filtered. The filtrate and precipitate were dried and weighed. Yields: filtrate – 6.6 g, precipitate – 3.5 g. Percent formaldehyde reactive components were determined by the Stiasny method, results: filtrate – 90%, precipitate – 67%.

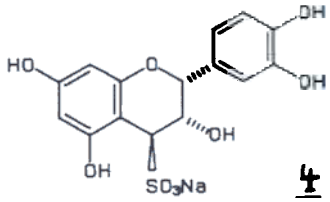
6. Staisny Reaction

The sample to be analysed was first dried under vacuum for at least 24 hours to give an accurate weight. After dissolving the dry sample in 10 ml of water, 1 ml of 10 M HCl and 2 ml of 37% formaldehyde added. The solution was then sealed in a reaction vial and heated in a boiling water bath for 30 minutes. The vial was then opened and the suspension filtered through a sintered glass filter under suction. The resulting precipitate was then washed 5 times with 10 ml aliquots of boiling water, dried under vacuum and the weight determined. The percentage of formaldehyde reactive material in the original sample was calculated by dividing the weight of the precipitate by the weight of the original sample. Due to the poor solubility of acetone/water extract in water, 30% DMSO was used as the reaction solvent with this material. This solvent was shown to have no significant affect on the Stiasny yield when using extracts of known composition.

Results and Discussion

The results of soil block tests conducted on tannin containing wood preservative formulations are summarized in Tables 3 through 6. Two different types of bark extracts were used. Acetone/water extraction gives a tannin preparation with minimal effect on the tannin structure. The content of formaldehyde-reactive components, including tannin, in the extract used in this study was 82%. Extraction with sodium bisulfite gives a modified tannin. Interflavonoid bonds are cleaved and derivatives obtained with sulfonic acid functionality on C–4 of the flavonoid monomer unit. Sodium epicatechin-(4β)-sulfonate (4) and a sulfo-

nated dimer (5) were both isolated from this type of extract and are typical of the type of sulfonated tannin derivatives present (Foo, McGraw, and Hemingway 1983).



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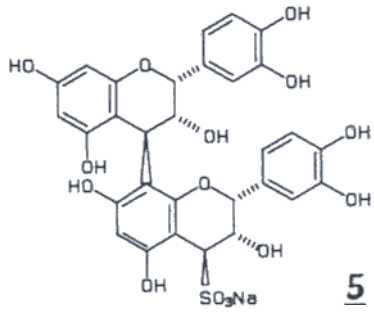

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A problem that immediately became apparent when making solutions containing tannin and copper (II) ions is the insolubility of the resulting complex in any neutral solvent. This is probably the result of the multiple complexation sites on any one tannin molecule. When the metal ion is added, the result is similar to crosslinking with formaldehyde – a complex with a

Table 3. Soil block tests using uncomplexed and copper-complexed, loblolly pine bark acetone/water extract (LPBAWE), loblolly pine bark sulfite extract I (LPBSE 1), and catechin with soft rot fungi

Treatment	Conc. in Soln. (%)		Mean Retention[1]		Mean % Wt. Loss (S.D.)[1]	
	Organic Component	Copper Salt	Organic Component	Copper Salt		
LPBAWE	0.5		3.13		12.6	(1.0)
	1.0		6.48		9.2	(1.7)
	2.0		12.69		10.8	(2.2)
	4.0		26.35		10.5	(1.3)
LPBAWE + $CuCl_2$	0.5	0.25	3.09	1.56	10.2	(2.6)
	1.0	0.5	6.36	3.22	6.6	(0.7)
	2.0	1.0	12.49	6.10	4.1	(2.8)
	4.0	2.0	25.02	10.95	2.0	(1.3)
LPBSE 1	0.5		3.19		9.2	(3.0)
	1.0		6.45		10.3	(2.8)
	2.0		12.82		12.0	(2.3)
	4.0		26.17		12.4	(0.6)
LPBSE 1 + $CuCl_2$	0.5	0.25	3.13	1.53	7.5	(1.3)
	1.0	0.5	6.80	3.22	2.1	(1.3)
	2.0	1.0	12.89	6.20	1.3	(1.8)
	4.0	2.0	26.18	12.18	0.4	(0.8)
Catechin (Ethanol Solvent)	0.5		2.32		13.9	(2.5)
	1.0		5.05		12.4	(0.7)
	2.0		9.53		12.4	(1.9)
	4.0		20.31		12.1	(2.0)
Catechin + $CuCl_2$	0.25	0.125	1.19	0.78	12.1	(1.0)
	0.5	0.25	2.40	1.59	5.5	(1.4)
	1.0	0.5	4.82	3.16	3.1	(1.0)
	2.0	1.0	9.69	6.24	6.1	(2.7)
	4.0	2.0	19.82	12.68	1.0	(1.2)
$CuCl_2$		0.125		0.77	11.4	(0.9)
		0.25		1.64	7.6	(2.9)
		0.5		3.19	6.9	(1.0)
		1.0		7.37	3.3	(1.6)
		2.0		12.71	2.4	(2.3)
Pentachlorophenol	0.5		2.40		5.5	(2.1)
	1.0		5.05		0.3	(0.5)
			9.49		0.6	(1.3)
Controls					11.2	(1.3)

[1] Average of 5 replicates

very high effective molecular weight is formed, too large to be soluble in conventional solvents. This is an advantage when the complex is within the wood since it is not prone to leaching out, but does present formulation and treatment problems.

The simplest solution to the insolubility problem is to form the complex *in situ*, within the wood block. The wood sample is first saturated with the bark extract solution, allowed to dry, then treated with the copper salt. Using this procedure, an initial experiment was done to determine whether the tannin/Cu(II) complex has any suitable wood preservative characteristics. Results of unsterile soil block tests and monoculture soil block tests with *G. trabeum* and *C. versicolor* on acetone/water and sulfited extracts (LPBSE1) of loblolly pine bark, and catechin (2), a simple model of the tannin monomer unit, both alone and complexed with Cu(II) are shown in Tables 3, 4 and 5. Untreated blocks, a pentachlorophenol treatment, and the copper salt solution by itself were included as controls.

The unchelated acetone/water and sulfited bark extracts, as well as the catechin, did not have weight losses significantly different from the untreated controls (Tables 3, 4, and 5). These treatments had no significant wood preservative properties at the retentions tested. However, copper complexes with tannin extracts or catechin when formed *in situ* by double treatment did give much lower weight losses than the untreated controls at some retentions. The brown-rot fungus, *G. trabeum*, is especially sensitive to copper.

Table 4. Soil block tests using uncomplexed and copper-complexed, loblolly pine bark acetone/water extract (LPBAWE), loblolly pine bark sulfite extract I (LPBSE 1), and catechin with the brown rot fungus *Gloeophyllum trabeum*

Treatment	Conc. in Soln. (%)		Mean Retention[1]		Mean % Wt. Loss (S.D.)[1]	
	Organic Component	Copper Salt	Organic Component	Copper Salt		
LPBAWE	0.5		3.08		46.9	(4.5)
	1.0		6.39		52.5	(4.2)
	2.0		12.60		49.4	(9.7)
	4.0		25.13		50.5	(6.8)
LPBAWE + $CuCl_2$	0.5	0.25	3.13	1.59	0.0	
	1.0	0.5	6.28	3.13	0.0	
	2.0	1.0	12.63	6.19	0.0	
	4.0	2.0	24.43	12.57	0.0	
LPBSE 1	0.5		3.01		46.2	(1.9)
	1.0		6.24		43.6	(5.2)
	2.0		12.59		49.4	(5.2)
	4.0		24.20		50.5	(10.2)
LPBSE 1 + $CuCl_2$	0.5	0.25	3.06	1.52	0.8	(0.6)
	1.0	0.5	6.15	3.04	0.0	
	2.0	1.0	12.63	6.19	0.9	(0.6)
	4.0	2.0	24.83	11.98	1.0	(0.3)
Catechin (Ethanol Solvent)	0.5		2.24		46.5	(5.9)
	1.0		4.71		51.0	(5.7)
	2.0		8.80		50.2	(1.9)
	4.0		18.00		42.6	(2.8)
Catechin + $CuCl_2$	0.25	0.125	1.15	0.77	1.0	(0.5)
	0.5	0.25	2.12	1.47	0.0	
	1.0	0.5	4.35	3.01	0.0	
	2.0	1.0	9.15	6.03	0.0	
	4.0	2.0	17.54	11.87	0.0	
$CuCl_2$		0.125		0.71	1.4	(0.5)
		0.25		1.51	0.2	(0.4)
		0.5		3.06	0.0	
		1.0		6.09	0.0	
		2.0		12.68	0.0	
Pentachlorophenol	0.5		2.32		0.0	
	1.0		4.59		0.0	
	2.0		9.73		0.0	
Controls					46.0	4.1)

[1] Average of 5 replicates

All the copper containing formulations, including $CuCl_2$ by itself, completely inhibited the growth of this fungus, even at the lowest retentions tested (Table 4). The significance of the tannin copper complex is most clearly seen in the experiments with *C. versicolor* (Table 5). Here, the complex performed much better than the controls in which only $CuCl_2$ was used as the treatment.

The copper chelated acetone/water at a total retention of about 10 kg/m^3 was equivalent in efficacy to pentachlorophenol at retentions between 5 and 11 kg/m^3 for *C. versicolor* (Table 5) and pentachlorophenol at 2.5 kg/m^3 for the soft-rot fungi (Table 3). Catechin chelated with copper was more effective than the acetone/water tannin preparation against the soft-rot fungi but less effective towards *C. versicolor*. The combination of the sulfited bark extract plus $CuCl_2$ was more effective than either of the other two chelated treatments. Against the soft-rot fungi, the sulfited extract was about twice as effective as the acetone/water extract, both chelated with copper. On the basis of these results further work was done with sulfited bark extracts and is summarized in Table 6. Only the test procedure with *C. versicolor* was used because it was felt this test challenged the test formulation the most.

The sulfited extract used in the above described work was made from bark obtained from trees that had

Table 5. Soil block tests using uncomplexed and copper-complexed, loblolly pine bark acetone/water extract (LPBAWE), loblolly pine bark sulfite extract I (LPBSE 1), and catechin with the white rot Fungus *Coriolus versicolor*

Treatment	Conc. in Soln. (%)		Mean Retention[1]		Mean % Wt. Loss (S.D.)[1]	
	Organic Component	Copper Salt	Organic Component	Copper Salt		
LPBAWE	0.5		3.47		76.5	(2.0)
	1.0		6.97		76.6	(1.1)
	2.0		14.35		59.1	(10.2)
	4.0		28.34		65.4	(2.7)
LPBAWE + $CuCl_2$	0.5	0.25	3.53	1.77	49.0	(10.2)
	1.0	0.5	7.13	3.58	32.7	(9.0)
	2.0	1.0	14.23	7.07	18.5	(3.8)
	4.0	2.0	29.38	13.75	11.5	(2.2)
LPBSE 1	0.5		3.50		>50.0[2]	
	1.0		7.17		58.5	(2.7)
	2.0		14.70		63.2	(5.4)
	4.0		29.48		65.3	(4.4)
LPBSE 1 + $CuCl_2$	0.5	0.25	3.46	1.71	48.5	(11.5)
	1.0	0.5	7.22	3.54	32.9	(10.6)
	2.0	1.0	14.28	6.87	15.2	(2.6)
	4.0	2.0	28.71	13.37	5.0	(1.1)
Catechin (Ethanol Solvent)	0.5		2.71		73.2	(2.0)
	1.0		5.37		73.0	(3.0)
	2.0		10.87		72.2	(4.1)
	4.0		21.86		71.0	(3.4)
Catechin + $CuCl_2$	0.25	0.125	1.32	0.86	61.8	(4.4)
	0.5	0.25	2.66	1.73	71.6	(8.8)
	1.0	0.5	5.42	3.52	71.1	(3.1)
	2.0	1.0	11.03	7.12	63.0	(7.5)
	4.0	2.0	21.83	13.98	31.1	(7.0)
$CuCl_2$		0.125		0.83	55.4	(6.4)
		0.25		1.74	61.1	(5.4)
		0.5		3.45	52.2	(12.0)
		1.0		7.16	62.7	(5.1)
		2.0		14.27	49.3	(9.6)
Pentachlorophenol	0.5		2.74		57.4	(3.8)
	1.0		5.05		46.9	(10.6)
	2.0		11.03		11.4	(4.5)
Controls					>50.0[2]	

[1] Average of 5 replicates
[2] Blocks in these sets could not be separated from the soil

Table 6. Soil block tests using copper-complexed, loblolly pine bark sulfite extract II (LPBSE 2), fractionated and in aqueous ammonia, and copper-complexed, loblolly pine bark acetone/water extract (LPBAWE), in aqueous ammonia with the white rot fungus *C. versicolor*

Treatment	Conc. in Soln. (%)		Mean Retention[1]		Mean % Wt. Loss (S.D.)[1]	
	Organic Component	Copper Salt	Organic Component	Copper Salt		
LPBSE 2 + $CuCl_2$ (Double Treatment)	0.5	0.25	3.09	1.53	15.3	(3.8)
	1.0	0.5	6.26	3.14	2.6	(1.2)
	2.0	1.0	12.34	6.27	1.5	(1.2)
	4.0	2.0	22.93	12.22	0.3	(0.4)
Low Tannin Fract. of LPBSE 2 + $CuCl_2$ (Double Treatment)	0.5	0.25	3.02	1.49	20.8	(8.1)
	1.0	0.5	6.11	3.05	7.9	(3.6)
	2.0	1.0	12.38	6.33	0.4	(0.9)
	4.0	2.0	22.76	12.17	0.1	(0.3)
High Tannin Fract. of LPBSE 2 + $CuCl_2$ (Double Treatment)	0.5	0.25	2.91	1.45	17.8	(5.4)
	1.0	0.5	6.05	3.01	4.8	(3.5)
	2.0	1.0	12.17	6.03	0.1	(0.3)
	4.0	2.0	24.59	11.63	0.0	(0)
2:1 LPBAWE/$CuSO_4$ in 20% NH_4OH	1.0		6.05		44.3	(16.5)
	2.0		12.40		22.4	(6.1)
	3.0		18.08		14.1	(1.4)
	4.0		24.91		8.9	(1.4)
	5.0		30.70		8.5	(1.9)
1:1 LPBAWE/$CuSO_4$ in 20% NH_4OH	1.0		6.16		27.5	(2.4)
	2.0		11.97		19.1	(2.2)
	3.0		18.34		7.7	(1.0)
	4.0		24.15		5.4	(0.9)
	5.0		30.29		1.6	(1.0)
2:1 LPBSE 2/$CuSO_4$ in 10% NH_4OH	1.0		6.00		42.2	(8.0)
	2.0		12.14		25.6	(2.2)
	3.0		18.30		17.5	(2.6)
	4.0		21.67		13.0	(2.9)
	5.0		28.51		9.3	(1.4)
1:1 LPBSE 2/$CuSO_4$ in 10% NH_4OH	1.0		6.09		32.1	(6.2)
	2.0		12.47		13.6	(2.5)
	3.0		18.60		8.1	(2.1)
	4.0		24.87		3.1	(0.8)
	5.0		31.46		1.0	(0.6)
Pentachlorophenol	0.5		2.52		62.3	(6.9)
	1.0		5.05		33.6	(4.0)
	2.0		10.27		7.0	(1.8)
	3.0		15.19		0.9	(1.5)
$CuCl_2$ Only	0.25			1.49	35.3	(6.7)
	0.5			2.97	15.5	(5.9)
	1.0			6.11	11.4	(1.8)
	2.0			12.18	12.1	(3.3)
Controls					>50[2]	

[1] Average of 5 replicates
[2] Blocks in these sets could not be separated from the soil

been killed by Southern pine beetle. To obtain data on an extract from a more typical source, a second sulfited extract was prepared from the bark of freshly harvested pulpwood-sized loblolly pine trees. This extract is referred to here as LPBSE2 and had a higher content of formaldehyde-reactive constituents (Table 2).

A two-stage treatment using this tannin preparation and copper resulted in a treatment with better performance than sulfite extract from beetle-killed trees (LPBSE1) at all retentions. The copper chelated sulfite extract II was even better than pentachlorophenol at a retention of about 10 kg/m^3. Sulfited bark extracts contains some $NaHSO_3$ and carbohydrates. To deter-

mine what effect these components have on the preservative efficacy, the bark extract was fractionated into a high tannin content portion (Stiasny – 90%) and a relatively tannin-poor fraction (Stiasny – 67%) and the two materials tested for preservative efficacy after complexation with copper. Table 6 shows that the high tannin content fraction resulted in a lower weight loss. This result indicates that the sulfited tannin is the component of the extract contributing most to its efficacy as a wood preservative when combined with copper ions.

Formulations were also tested that contain both sulfited tannin and copper in an ammonia solution. A two-stage treatment is not necessary with this type of solution. The ammonia forms a stronger complex with the Cu(II) ions than the catechol-type tannin. The extended tannin/copper complex does not form until the ammonia volatilizes from the solution, in this case during the fixing process. Previously published work indicated that procyanidins exposed to oxygen-free alkaline conditions rearrange to a polymer with fewer aromatic rings in total, but the catechol-derived complexation site is unaffected (Laks and Hemingway 1987a, b; Laks, Hemingway, and Conner 1987). However, we observed substantially reduced performance when the tannin was formulated in ammonia solution (Table 6). This was probably due to the oxygen present in this case, oxidizing the catechol rings which react further to ultimately give moieties incapable of complexation. These types of oxidation reactions have been observed with catechin (Jensen and Pedersen 1983).

Conclusion

Copper complexed, condensed tannin-containing bark extracts have efficacy as wood preservatives when evaluated by standardized wood preservatives laboratory testing procedures. Bark extracts by themselves do not cause any reduction in weight loss of wood blocks at reasonable retentions. The best experimental wood preservative treatment was a dual treatment using a sulphited bark extract first, followed by a $CuCl_2$ treatment. A single stage treatment using a sulphited bark extract with $CuCl_2$ in a dilute aqueous ammoniacal solvent was also effective.

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