

## Mould resistance of the bamboo *Thyrostachys siamensis* treated with chitosan

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### ABSTRACT

In this study, mould resistance of the bamboo species *Thyrostachys siamensis* treated with chitosan was tested under laboratory and praxis condition. In the laboratory experiment, bamboo specimens were treated with various solutions of both low molecular weight chitosan (LMW) and medium molecular weight chitosan (MMW) at concentrations of 1, 2 and 3% and Chitosan-copper complexes (CC) at concentrations of 2, 4 and 6%. Mould growth on the specimens was evaluated 1, 2, 4 and 8 weeks after they were exposed to the inoculation with a conidia mixture of six moulds isolated from bamboos. In field test, bamboo samples were treated with the effective formulations from the laboratory experiment. Evaluation of mould growth on the samples was done 1, 2, 4 and 8 weeks after exposure at the storage site of Bamboo company, Binh Duong. The results showed that treatments with chitosan (MMW) at the concentration of more than or equal to 3% and Chitosan-copper complexes at the concentration of more than or equal to 4% completely inhibited mould growth on the bamboo *T. siamensis*.

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## 1. Introduction

Bamboo is one of the important vegetative resources after plantation wood and is a major raw material for the forest product industry. In Vietnam, bamboo has become the main material for industrial manufacturing of round and laminated bamboo furniture and parquet. The bamboo *Thyrostachys siamensis* with its Vietnamese name "Tam Vong" is one of the most common species growing mainly as a forest and also largely cultivated in the provinces Binh Thuan, Gia Lai, Kong Tum, Lam Dong and Tay Ninh. The culms are the main raw material of many bamboo companies in South Vietnam for furniture for exportation.

Bamboo has low natural durability against fungi and insects compared with wood. In gen-

eral, several fungi from the groups of moulds, ascomycetes and basidiomycetes colonise the culms of bamboos. Exposed bamboo is especially affected by moulds during storage, processing, transport in containers and its final use (Liese & Tang, 2014). For protection of bamboo against moulds and other fungi, sodium pentachlorophenol had been widely used due to its effectiveness and relative low cost. However, the chemical is banned due to its high toxicity and the public concerns on the environment. Thus, bamboo manufacturers have extreme problems in protecting bamboo for local use and export. Since bamboo countries export large quantities of bamboo culms and utilities in containers, the damage due to mould growth at port arrival has become quite serious. Manufacturers need cost-effective and also environment-friendly treatment

methods for moist bamboo during its susceptible phase.

Chitosan is the de-acylated form of chitin produced commercially from shellfish. Chitosan can be used in a wide range of industries, due to its high degree of biocompatibility and its ability to offer an environmentally friendly alternative to current applications (Kumar, 2000; Chirkov, 2002). Chitosan-copper complex is produced from chitosan and copper salts. Recently, Chitosan and Chitosan-copper complex have been considered as interesting environmentally friendly material for wood, bamboo preservation. It is used as a potential wood preservative alone or in combination with other biocides for protection of wood and bamboo against fungi and mould (Sun et al., 2007, Gorgija et al., 2014; Gamal et al., 2016). The effectiveness of the chitosan mentioned in these researches led us to investigate mould resistance of the bamboo species *Thyrostachys siamensis* treated with various chitosan solutions of different molecular weight and concentration.

## 2. Materials and Methods

### 2.1. Laboratory experiment

Mature 3-year-old bamboo culms of *T. siamensis* were collected from a plantation of Bamboo Nature company in Binh Thuan province. From fresh culms, samples of 60 mm length were taken halfway between the internodes and split lengthwise.

Solutions of both low molecular weight chitosan (LMW) and medium molecular weight chitosan (MMW) and chitosan-Cu (II) complexes (CC) with ratio 1:1 at the concentrations of 1, 2 and 3% were applied.

Three specimens were dipped for 10 min in the respective treatment solution and placed in a small plastic box (12 x 12 x 6 cm). They were exposed to artificial infection with a water-based mixture of conidia of six moulds *A. niger*, *A. flavus*, *A. oryzae*, *Aspergillus* sp., *Paecilomyces variotii*, and *Penicillium* sp. by using a small brush. These six moulds (Figure 1) were isolated from natural growth on bamboos and were identified by DNA-IIS sequencing at the Center of Wood Biology, Hamburg University. The exposure was done in an incubation room at 30°C and 75% RH. The development of mould growth on the surface of the specimens was assessed after 1,

2, 4 and 8 weeks according to the rating method based on the BSI 2005 (Table 1).

### 2.2. Experiment for field tests

Samples were prepared from *T. siamensis*, as of 1000 mm length. The effective chitosan formulas from the laboratory experiments were applied. The bamboo samples were dipped for 15 min in the treatment solutions, then bundled and placed on supports over wet soil ground. After one day of exposure to natural infection, the samples were covered with a plastic sheet to avoid sunlight and drying. The test was carried out in a raw material storage area in the factory of the Bamboo Nature company, Binh Duong province. The field tests were carried out in three periods, each of 8 weeks during the rainy season. The temperature during exposure was about 28°C and the relative humidity was between 80 and 90%. The development of mould growth on the surface of the samples was assessed after 8 weeks.

## 3. Results and Discussion

### 3.1. Laboratory test

Based on the rating system (Table 1) and average mold covering for 3 replicates of each treatment, the results of the laboratory test for *T. siamensis* are presented in Table 2. The samples treated with 3%, chitosan MMW, 4% and 6% Chitosan-copper complexes (CC) did not show any infection, whereas water-treated control samples, chitosan LMW 1% and 2% had the highest infection rate. Medium molecular weight chitosan (MMW) and Chitosan-copper complexes showed better antifungal property than low molecular weight chitosan (LMW). Effect of medium molecular weight chitosan is improved by increasing its concentration from 1% to 3%. This confirms with previous studies of the antifungal effect of chitosan by Larnoy et al. (2006) and Gorgij et al. (2014), and our results are also in agreement with Kobayashi & Furukawa (1995) and Mekahlia & Bouzid (2009) who mentioned that Chitosan-copper complexes prevented mould.

For the laboratory experiments, the specimens were infected only once. Under field conditions with larger samples, bamboo would be exposed to permanent infection pressure from the surrounding air, so that the applied concentrations might not meet those conditions. Therefore, the effec-

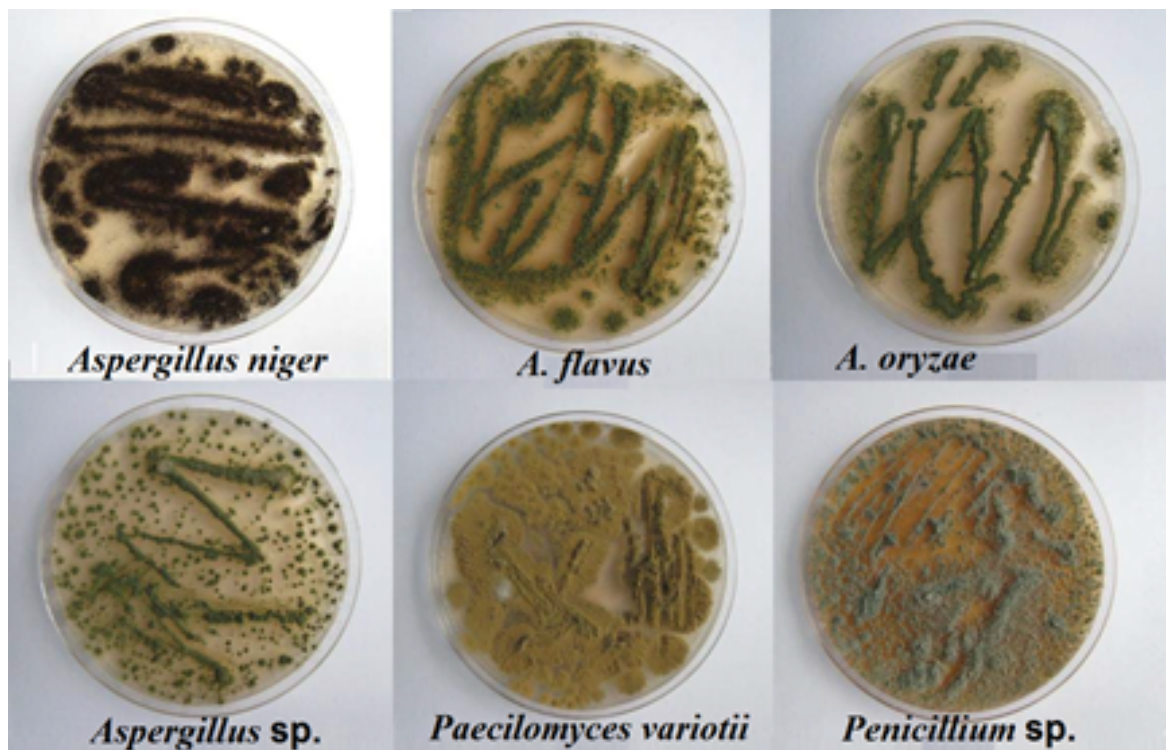






Figure 1. Moulds for testing.

Table 1. Standard method for rating the infection on the surface

Rating	Definition	Samples
1	No growth	
2	Slightly overgrown	
3	Moderately overgrown	
4	Severely overgrown	

**Table 2.** Infection value of treated bamboo *T. siamensis* in the laboratory test

Treatment solution <sup>1</sup>	Exposure time			
	After 1 week	After 2 weeks	After 3 weeks	After 4 weeks
MMW 1%	0	1	2	3
MMW 2%	0	0	1	1
MMW 3%	0	0	0	0
LMW 1%	1	3	4	4
LMW 2%	0	3	3	4
LMW 3%	0	2	2	2
CC 2%	1	3	4	4
CC 4%	0	0	0	0
CC 6%	0	0	0	0
Control	1	4	4	4

<sup>1</sup>MMW: Medium molecular weight chitosan; LMW: Low molecular weight chitosan; CC: Chitosan-copper complexes.

**Table 3.** Infection value of treated bamboo *T. siamensis* in the laboratory test

Treatment solution <sup>1</sup>	Test period	Exposure time			
		After 1 week	After 2 weeks	After 3 weeks	After 4 weeks
MMW 2%	I	0	0	2	3
	II	1	3	4	3
	III	0	1	2	3
MMW 3%	I	0	0	0	0
	II	0	0	0	0
	III	0	0	0	0
CC 4%	I	0	0	0	0
	II	0	1	1	1
	III	0	0	0	0
CC 6%	I	0	0	0	0
	II	0	0	0	0
	III	0	0	0	0
Control	I	1	1	3	4
	II	4	4	4	4
	III	2	3	3	4

<sup>1</sup>MMW: Medium molecular weight chitosan; CC: Chitosan-copper complexes.

tive treatment solution of 2% and 3% MMW, 4% and 6% CC were further investigated in field trials.

### 3.2. Field test

Results of the field experiment are summarized in Table 3. Differences occurred in moulding between exposure periods. In most treatments, the samples from the second period were more quickly overgrown by moulds due to the high relative humidity of about 90%.

Generally, results of this field test are similar to the laboratory experiments with smaller samples. Treatments with 3% medium molecular weight chitosan (MMW), 4% and 6% Chitosan-

copper complexes (CC) completely inhibited mould growth on the bamboo *T. siamensis*.

This investigation has shown that treatment of the bamboo *T. siamensis* with medium molecular weight chitosan and Chitosan-copper complexes can be protected from moulding. However, there are significant differences in efficacy of antimould treatments for the bamboo species (Schmidt et al., 2011). A previous study of Sun et al., 2012 indicated that Moso bamboo treated with Chitosan-copper complexes not being effective against moulds such as *Trichoderma viride* and *Aspergillus niger*. Hence, further experiments regarding mould susceptibility of different bamboo species may be of interest.

#### 4. Conclusions

Treatment of the bamboo *T. siamensis* by medium molecular weight chitosan and Chitosan-copper complexes could completely prevent moulding during the exposure period of at least eight weeks, whereas the bamboo *T. siamensis* treated with low molecular weight chitosan was not effective against moulds. Treatments with medium molecular weight chitosan at the concentration of more than or equal to 3% and Chitosan-copper complexes at the concentration of more than or equal to 4% completely inhibited mould growth on the bamboo species.

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