

***Phanerochaete chrysosporium* Burds INOCULATION TO IMPROVE THE PHYSICAL PROPERTIES OF KAPOK PULP**

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Abstract

White rot fungus are wood degrading organism able to decompose wood polymers; lignin, cellulose and hemicelluloses. Some species of white rot fungi decompose wood lignin over wood polysaccharides (e.g. cellulose), make it preferable for biopulping applications. A study of *Phanerochaete chrysosporium* Burds fungi inoculation to kapok chips was conducted to investigate the characteristics for the wood chips to be used as pulp and paper raw materials, as degradation level influences pulp characteristic. *Phanerochaete chrysosporium* fungi was incubated at PDA for 10 days, then inoculated to kapok chips for 20, 30, and 40 days. Fiber morphology was analyzed furthermore 16% active alkali of kraft process was conducted for 1, 1.5, and 2 hours cooking times. The results showed that range of tear strength 4.23 mNm²/g up to 6.35 mNm²/g, tensile strength 35 N m/g until 39.9 N m/g and bursting strength 1,5 kPa up to 2,01 kPa.

Key words : Biopulping, *Phanerochaete chrysosporium*, *Ceiba pentandra*, pulp and paper

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I. Introduction

Biopulping, a process of pretreating chips with fungus before pulping, leads to a significant energy savings and improves sheet strength. Among wood decay fungi, there are two broad classification, white-rot and brown-rot. These classifications are generally based on observations of wood after an extensive decay. Most white-rot species have the ability to degrade and mineralize lignin. In contrast, most brown-rot species only modify lignin and

do not mineralize it. Lignin degradation and mineralization is an almost exclusive domain of white-rot fungi. The preference of selective white-rot fungi to degrade the lignin compound of wood in over cellulose and hemicelluloses could be utilized of biokraft fiber. It has been demonstrated that the pre-treatment of wood chips with selective white-rot fungi leads to energy savings of about 30% (Scott et al. 2002). Furthermore, selective white-rot fungi are able to remove lignin from wood surface and

shavings (Fackler et al. 2002, 2003). Although simple classification of white versus brown-rot does not completely describe the diversity of action of all fungi (Hibbett and Thorn, 2001), it has provided guidance in selection of species for application in the pulp and paper industry. Since some species of white-rot fungi selectively remove lignin and leave cellulose intact, they and their enzymatic system have been the subject of research focusing on producing pulp and paper (Young and Akhtar, 1998).

Phanerochaete chrysosporium Burds. is the most intensively studied wood-decaying white-rot fungus and efficiently degrades lignin (Ander and Eriksson, 1977). Taxonomically it belongs to the Corticiaceae. It is a resupinate or crust fungus and is thermophilic, having an optimum temperature about 40° C. LiP was first found in *P. chrysosporium* cultures by two research groups (Tien and Kirk, 1983; Glenn et al. 1983). Depending on culture conditions, it produces variable amounts of LiP and MnP isoenzymes, but not laccase. Thus it can be considered as an atypical white-rot fungus (Hatakka, 1994). The presence of 10 LiP-encoding genes, five MnP-encoding genes and the absence of laccase-encoding genes was verified

when the entire genome of that fungus was sequenced (Martínez et al. 2004). Veratryl (3,4- dimethoxybenzyl) alcohol (VA) was the first low-MW compound found in the liquid culture of *P. chrysosporium* and may be associated with lignin degradation (Lundquist and Kirk, 1978). Many biochemical and physiological reactions related to lignin degradation were first found in cultures of *P. chrysosporium* followed with other finding of it in cultures of other fungi.

During biopulping, a white-rot fungus, *P.chrysosporium*, was grown on wood chips for 10; 20 and 40 days. When these chips are used to produce biokraft pulp, the refining requires 30-40% less energy and the sheet strength properties are improved.

2. Material and Methods

2.1. Inoculums preparation

Potato Dextro Agar (PDA) media was prepared for fungi culture. The pottery-plates were inoculated with *P. chrysosporium* which was incubated at 36°C for 10 days without agitation. The mycelia were separated by blending aseptically in a blender using distilled water. By adding sterile water, 200 ml suspension was prepared with 10 mg m⁻¹ concentration on dry weight basis.

The suspension was used to treat wood chips for cooking.

2.2. Wood material

Wood sample of approximately 3x3x0.2 cm were obtained from 25-year old debarked stems of Kapok (*Ceiba pentandra*) from Gantiwarno; Klaten; Central Java; Indonesia. The sample were dried (35°C) until it reached a constant weight, then it was submerged in distilled water for 24 h and autoclaved at 121°C for 20 min. One part of kapok chips was treated with the above-mentioned fungi prior to cooking and the rest of the chips were cooked directly (without fungal treatment) for control. Each flask was inoculated with mycelium suspension of *P. chrysosporium* without any addition of supplementary nutrients. The rate of mycelium application was 0.5% on dry weight basis of chips. After receiving inoculate, the flasks were shaken vigorously for a uniform mixing. Each flask with sufficient aeration was sealed and placed in an incubator maintaining the temperature at 36°C for different time periods (20;30; 40 days). After the specified period of incubation (20;30 and 40 days), each flask was autoclaved at 121°C for 20 min to stop further growth of fungi.

2.3. Kraft Pulping

Fungally-pretreated wood chips by the kraft method. The cooking liquor contained 25% Na₂S (unbleach) and 16% NaOH. The pulping time was 1;1.2 and 2 h with the maximum temperature of 170°C and the pressure is 1 MPa.

2.4. Pulp Test

All procedures and analyses were done according to the relevant SNI standard as follows: refining pulp with SNI 14-0490-1989-A, papermaking with SNI 14-0489-1989-A, Tensile Strength Test with SNI 14-0437-1989, Tearing Test with SNI 14-0436-1989 and Bursting Test with SNI 14-0489-1989.

3. Result and Discussion

Relationship between hand sheet properties and the energy saving is of great importance in papermaking. Thirty samples were tested for their paper properties. All kapok samples were pulped at cooking liquor containing 25% Na₂S (unbleached) and 16% NaOH with different cooking times. The pulping time was 1; 1.2 and 2 h with a maximum temperature of 170°C and 1 MPa of pressure.

After 20, 30 and 40 days growth of *P.chrysosporium*, the samples were used to prepare handsets, which were analyzed for their physical properties (Fig.1).

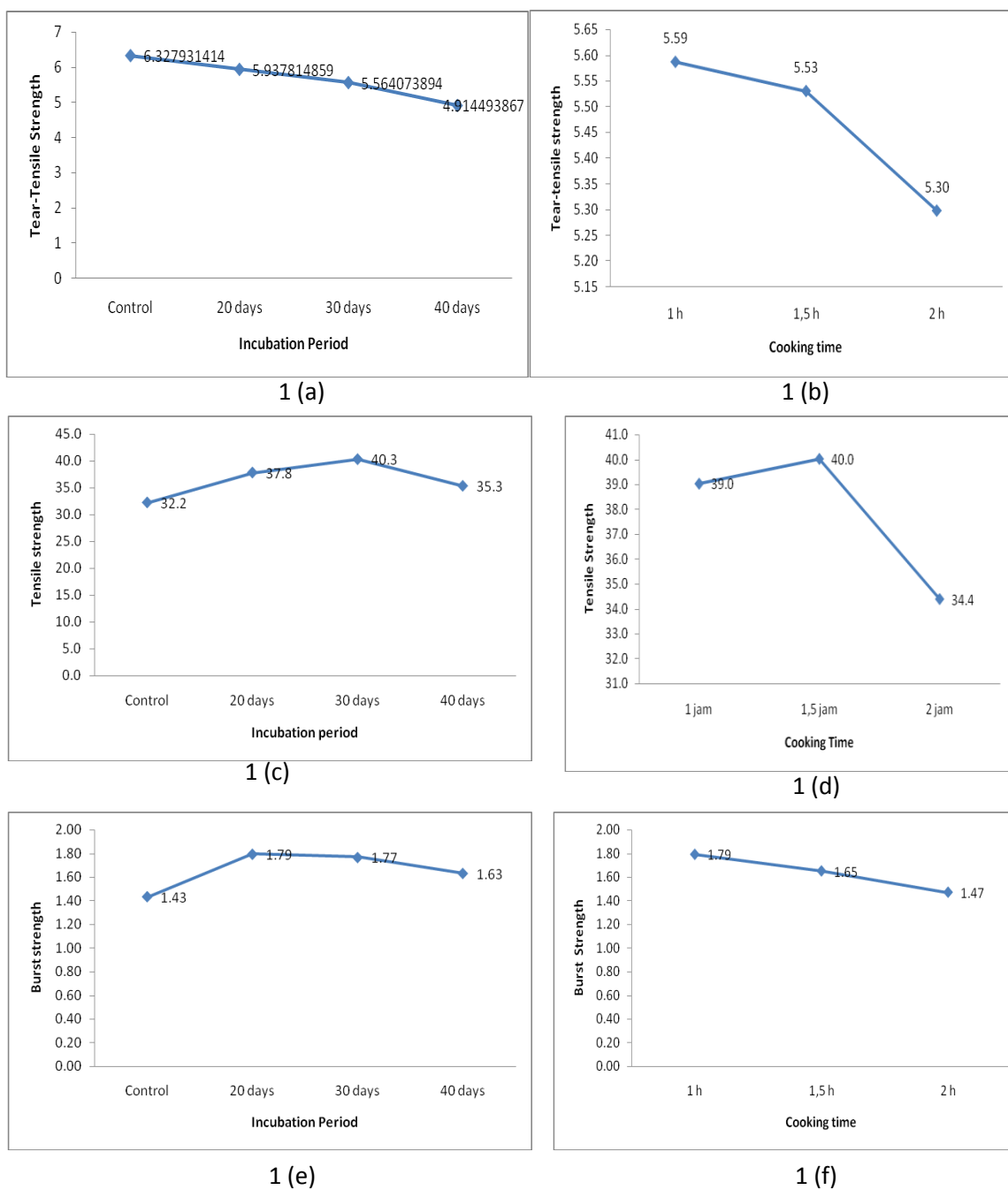


Fig. 1. (a) tear-tensile strength (mN m²/g) and incubation period, (b) tear-tensile strength (mN m²/g) and cooking time, (c) tensile-strength (N m/g) and incubation period, (d) tensile-strength (N m/g) and cooking time, (e) burst strength (kPa) and incubation period, (f) burst strength (kPa) and cooking time

The tear-tensile strength of the fibre pulp may not be the most important property in its use, for example, as a component in printing and writing grades of paper. Nevertheless, the tear-tensile relationships have been chosen to illustrate the influence of species and pulping method on the papermaking characteristics of the pulps. It serves the purpose of placing the non-wood-based papers on the evaluation scale used for the paper properties of wood-based pulps (Rydholm, 1965). The tear-tensile strength is properties of paper sheet that is influenced by individual fibre and bonding of fibre (Haygreen and Bowyer, 1996). Tear-tensile strength depends of fibre length.

The result shows that the tear-tensile strength of kapok pulp in 20, 30, and 40 days lower than control but the tear-tensile strength in 20 and 30 days were higher than standard of bleaching kraft pulp (SNI 14-0698-1989). The standard is 5.0 mN m²/g. In 40 days incubation lower than standard is 4.9 mN m²/g. Kapok pulp in all of cooking time higher than standard.

From the analysis of tensile-strength, treated pulps showed an increase from control in its pulping cooking time and incubation period. That means

biopulping process can increase tensile strength. The increase of tensile-strength because the fibre from biopulping process is clear from lignin so the bonding of celluloses more strength (Casey, 1996). The burst-strength of kapok pulp is lower than standard of pulp (SNI 14-0698-1989). The result of burst-strength of kapok pulp lower than 2 (SNI standard), that is because the fiber begin to be degraded by enzyme activity of *P. chrysosporium*.

4. Conclusion

From the handsheets analysis, we conclude that the pre-treatment of kapok chips with *P. Chrysosporium* has a positive effect on some strength properties of the pulps (e.g. tear-strength and tensile strength) but has a negative impact on other properties (bursts- strength). It is possible that the small size of kapok fibre particles used in this experiment is responsible to be the negative factor for biopulping. Thus, studies are in progress to overcome this problem.

References

- Ander, P., Eriksson, K.E., 1977. Selective degradation of wood components by white-rot fungi. *Physiologia Plantarum*. 41, 239-248.
- Fackler, K., Wulz, P., Schild, D., Schwanninger, M., Hinterstoisser, B., Lamaipis, P., et al., 2002. Effect of biopulping on the surface properties of wood. In: Seventh European Workshop on Lignocellulosics and Pulp. p. 289-92.
- Fackler, K., Schwanninger, M., Hinterstoisser, B., Messner, K., 2003. Biomodification of spruce wood by *Ceriporiopsis subvermispora*: comparison of the effects of three different strains. In: 12th ISWPC, International Symposium on Wood and Pulp Chemistry. University of Wisconsin-Madison, Department of Forest Ecology and Management. p. 291-4.
- Glenn, J.K., Morgan, M.A., Mayfield, M.B., Kuwahara, M., Gold, M.H., 1983. An extracellular HO-requiring enzyme preparation involved in lignin biodegradation by the white-rot basidiomycete *Phanerochaete chrysosporium*. *Biochem Biophys Res Commun*. 114:1077- 1083.
- Hatakka, A.I., 1994. Lignin-modifying enzymes from selected white-rot fungi - production and role in lignin degradation. *FEMS Microbiol Rev*. 13:125-135.
- Hibbett, D.S., Thorn, R.G., 2001. Basidiomycetes: Homobasidiomycetes, in *The Micota VII Part B: Systematics and Evolusion*, McLaughlin, and Lemke, Editors. Springer-Verlag:Berlin.
- Haygreen, J.G. and J.L. Bowyer., 1989. *Hasil Hutan dan Ilmu Kayu, Introduction*. Gadjah Mada University Press. Yogyakarta.
- Lundquist, K., Kirk, T.K., 1978. *De novo* synthesis and decomposition of veratryl alcohol by a lignin-degrading basidiomycete. *Phytochemistry*. 17:1676-1676.
- Martínez, D., Larrondo, L.F., Putnam, N., Gelpke, M.D.S., Huang, K., Chapman, J., Helfenbein, K.G., Ramaiya, P., Detter, J.C., Larimer, F., Coutinho, P.M., Henrissat, B., Berka, R., Cullen, D., Rokhsar, D., 2004. Genome sequence of the lignocellulose degrading fungus *Phanerochaete chrysosporium* strain RP78. *Nat Biotechnol*. 22:695-700.
- Rydholm, S.A., 1965. *Pulping Processes*. Wiley, New York. Preprint 1985 (facsimile edition) Robert E. Krieger, Malabar, FL. p. 1270.
- Scott, G.M., Akhtar, M., Swaney, R.E., Houtman, C.J., 2002. Recent developments in biopulping technology at Madison, WI. In: Lantto R, Viikari L, editors. *Biotechnology in the pulp and paper industry*. Amsterdam: Elsevier Science B.V.
- Tien, M., Kirk, T.K., 1983. Lignin-degrading enzyme from the hymenomycete *Phanerochaete chrysosporium* Burds. *Science*. 221:661-662.
- Young, R., Akhtar, M., 1998. *Environmentally Friendly Technologies for The Pulp and Paper Industry*. John Wiley & Sons:New York.