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Decomposition of Lignin and Holocellulose of *Pinus roxburghii* Sar. (Pinaceae) Needle Leaves, Twigs and Barks by Fungal Isolates from Virgin Forest Ecosystem of Doddabetta belt of Nilgiris

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ABSTRACT

Decomposition of lignin and holocellulose study was conducted in *Pinus roxburghii* Sar. (Pinaceae) needle leaves, twigs and barks from the virgin forest ecosystem of Doddabetta belt of Nilgiris during monsoon periods June to November 2014. Four lignin and holocellulose degrading fungi which were dominating the *P. roxburghii* forest ecosystem were identified, *viz*., *Amanita muscaria, Coprinus micaceus, Cortinarius collinitus* and *Tricholoma album.* The spores collected from the fruit bodies / the mycelia from the degrading material were inoculated individually to the experimental needle leaves, twigs and barks to analyse the decomposition potentials of lignin and holocellulose. The percentage of degradation of lignin, holocellulose and hot water soluble content varied with the samples and as well as the fungal isolates. However the degradation is a long process which certainly adds nutrients to the virgin forest ecosystem.

Keywords: Decomposition, Holocellulose, Lignin, *Pinus roxburghii*

INTRODUCTION

Lignocellulose is the predominant component of woody plant and dead plant materials, and the most abundant biomass on earth. Lignin and holocellulose in the biomass structure are major energy sources available to decomposer organisms constituting 70-80% of fresh organic material (Swift *et al*., 1979). Lignin is a recalcitrant plant polymer and its mineralization by white rot basidiomycetes plays a major role in carbon recycling (Martinez *et al*., 2005). White rot fungi are wood degrading organisms capable of decomposing all wood polymers, lignin, cellulose and hemicelluloses (Hakala, 2007). Holocellulose, a polysaccharide containing cellulose and hemicellulose (Pettersen, 1984) is a major component of wood suitable for fungal growth. Polysaccharide content generally ranges between 60 and 80% (w/w) in hardwood (Willfor *et al*., 2005). However, decomposition rate of cellulose is higher than that of lignin (Fioretto *et al*., 2005). White-rot fungi belong to the basidiomycetes and their activity is usually related to the moisture content of wood (Blanchette, 1995). The decaying fungi belong to saprophyte fungal organisms, since they live on dead or residual vegetation, decomposing them into simpler molecular compounds (Dubeux *et al*., 2006; Ohkuma *et al.,* 2001). The use of saprophytic fungi can be expected to

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accelerate the decay of waste. Laboratory screening revealed that four fungi from the natural *Pinus* forest of Nilgiris which are capable of degrading lignin as well as holocellulose was identified and their lignin and holocellulse decomposition potentials in *Pinus roxburghii* Sar. needle leaves, twigs and barks was worked out in this study.

MATERIAL AND METHODS

Leaves, twigs and barks of *Pinus roxburghii* (Three-leaved pine):

Sample materials in the form of needle leaves, twigs and barks were collected from the natural virgin forest of Doddabetta belt of Nilgiris, Tamilnadu, India.

Source of fungal materials:

Four genera of fungi were identified for their association with decomposition of needle leaves, twigs and barks of *P. roxburghii* material. They were *Amanita muscaria, Coprinus micaceus, Cortinarius collinitus* and *Tricholoma album* (Figure-1a-d). The fungal fruit body was collected in sterile plastic bags and brought to the laboratory and inoculated in the malt-extract-agar medium and the grown mycelia was used for further experimental work. They were cultured in lignocelluloses agar (LCA) modified by Miura and Kudo (1970). LCA contains glucose 0.1%, KH_2PO_4 0.1%, $MgSO_4$ $7H_2O$ 0.02%, KCl 0.02%, $NaNO_3$ 0.2%, yeast extract 0.02% and agar 1.3% (w/v) LCA modified by Miura and Kudo (1970) does not contain lignin or other recalcitrant compounds.

Decomposition test based on holocellulose and lignin content:

The needle leaves, twig and bark samples of *P. roxburghii* (Figure-1b 3, 4 and 5) was mixed with a 1% malt extract in distilled water to a water content of about 65% and packed in plastic bags (2.5 g for each sample pack with 3 replicates, then total sample were 32 plastic bags). They were then sterilized in autoclave at a temperature of 121° C and pressure of 1.5 atm., for 20 min. After the medium is cooled, it was then inoculated with the test fungi for 90 days to analyse the decomposition (Fig. c 6, 7 and 8). For control, samples were made of same materials with no fungal inoculation or treatment.

Determination of holocellulose content:

Holocellulose content was determined with reference to ASTMD 1104-56 (1978). 1 g of sample was placed in an Erlenmeger flask (250 ml) and 150 ml of distilled water was added. While slowly shaking, 1 g of $NaClO_2$ and 0.2 ml of acetic acid were added and the flask was covered with glass and boiled at 70 to 80° C for 60 min. Again 1 g of $NaClO_2$ and 0.2 ml of acetic acid were added and boiled. After cooling, the sample was filtered using a filter flask and washed with hot water until free of acid. Then, the insoluble portion was dried in an order at 105° C for 4 hr, cooled in a desiccator and weighed repeatedly until a constant weight was obtained. Holocellulose content was calculated as follows:

Holocelulose content (%) = (oven dried weight of holocellulose/Oven dried weight of initial sample) x 100

Determination of Lignin Content:

TAPPI Standard method (2009) T 222 OS-74 procedure was followed for the determination of lignin content. 1 g of air-dried sample was weighed and transferred to a 50 ml beaker, then 10 ml of 72% sulphuric acid was added with a pipette and the mixture was stirred with a glass rod. The mixture was moved quantitatively with a wash bottle of 500 ml and diluted with water until the final volume was 300 ml. The solution was then refluxed for 3 h filtered in a glass filtered and dried in an oven at 105° C for 12 h. The bottle was cooled in a desiccator for 15 min and then weighed. The glass filter containing the lignin was reported as percentage by weight of the dried sample. Lignin content was calculated as follows:

Lignin content % = (oven dried weight of lignin/oven dried weight of initial sample) x 100.

Determination of hot water soluble content:

The soluble of treated *P. roxburghii* needle leaves, twigs and barks were examined with reference to ASTMD1110-87 (2007). Two grams sample was oven-dried and placed into a 250 ml Erlenmeyer flask containing 200 ml of distilled water. A reflux condenser was attached to the flask and the apparatus was placed in a gently boiling water bath for three hours with constant shaking. Special attention was given to insure that the level of the solution in the flask remained below that of the boiling water. Samples were then removed from the water bath and filtered by vacuum suction into a glass

filter of known weight. The residue was washed with hot water before the glass-filter was oven-dried at 103 ± 2°C. The glass-filter was then cooled in a desiccator and weighed until a constant weight was obtained. The following formula was used to obtain the hot water soluble of the sample:

$$\text{Hot water soluble (\%)} = \frac{W_1}{W_1 - W_2} \times 100$$

where W_1 = Weight of oven – Dry test sample (g); W_2 = Weight of oven – Dry sample after extraction with hot water (g)

Statistical Analysis:

All statistical analyses were performed at 0.05% level, using the statistical software (SPSS Inc., Chicago, USA).

RESULTS AND DISCUSSION

Ability of the fungi to degrade lignin:

The lignin content of *P. roxburghii* leaves, twigs and barks are shown in Table-1. For measuring lignin content, *P. roxburghii* fresh needle leaves, twigs and barks collected from the forest soil and incubated with the four fungi for three to six months (90-180 days). The initial lignin content (control) of the needle leaf are 23.5%. After 90 days of treatment with fungi, the lignin content decreased to 17.0 to 19.5%, depending on the fungal genera (Table-1). The genera *Coprinus micaceus* was the most effective (17.0) at 90 days. The initial lignin content (control) of the needle leaves are 22.5%, after 180 days of treatment, the lignin content decreased to 13.8 to 15.5%. However, the genera *Cortinarius collinitus* was the most effective (13.8) at 180 days of degradation.

The initial lignin content (control) of the twigs of *P. roxburghii* was 52.5%. After 90 days of treatment with fungi, the lignin content decrease ranged from 38.2 to 44.3% and after 180 days of degradation the lignin content decrease ranged from 32.8 to 38.0% when compared with the initial content 50.5%. As far as the bark is concerned the initial lignin content (control) was 90.5%, after 90 days of treatment with fungi. The decrease in lignin content ranged from 74.6 to 82.2% and after 180 days of degradation the lignin content decrease ranged from 68.5 to 78.5% as against the initial (control) content 89.0%.

Fungi require a carbon source, macronutrients such as nitrogen, phosphorous and potassium and certain trace elements for their growth. Carbon serves primarily as an energy source for the microorganisms, while a small fraction of the carbon in incorporated into their cells (Tuomela *et al*., 2000).

Biomass including leaf, twigs, bark and other residual materials of forest ecosystem, naturally undergo degradation due to fungal enzymatic action. This causes increase in CO_2 in the

Table-1. Lignin content of *Pinus roxburghii* needle leaves, twigs and barks after treatment with selected fungi

Fungi	Lignin content of Needle leaves (%)		Lignin content of twigs (%)		Lignin content of barks (%)	
	Degradation period		Degradation period		Degradation period	
	90 days	180 days	90 days	180 days	90 days	180 days
Amanita muscaria	18.5 ± 0.20	15.0 ± 0.20	40.5 ± 0.20	36.1 ± 0.10	81.8 ± 0.10	70.1 ± 0.20
Coprinus micaceus	17.0 ± 0.45	15.0 ± 0.15	41.5 ± 0.10	35.5 ± 0.40	80.5 ± 0.25	75.0 ± 0.40
Cortinarius collinitus	18.2 ± 0.15	13.8 ± 1.03	38.2 ± 0.65	32.8 ± 0.15	74.6 ± 0.41	68.5 ± 0.45
Tricholoma album	19.5 ± 0.32	15.5 ± 0.40	44.3 ± 0.40	38.0 ± 0.10	82.2 ± 0.40	78.5 ± 0.20
Control	23.5 ± 0.40	22.5 ± 0.10	52.5 ± 0.15	50.5 ± 0.60	90.5 ± 0.20	89.0 ± 0.10

Values are as Mean ± SD of 3 replicates; Control: Refers to No fungal treatment

environment. Therefore, it would be better for the woody materials to be recycled by biological degradation or removal of lignin (Watanabe *et al*., 2003).

In this study, it was found that lignin content of either leaf, twigs or bark degraded effectively by naturally occurring lignin degrading fungi of *P. roxburghii* forest ecosystem. Lignin a heterogeneous plant cell wall biopolymer consisting of phenyl-propanoid units and the principal source of aromatic compounds found in nature, is extremely resistant to attack by most microorganisms (Tuomela *et al*., 2000; Dekker *et al*., 2002). Lignin is a branched polymer of substituted phenylpropane units joined by carbon and ether linkages. Lignin polymerization pattern and assembly is guided by the orientation of cellulose and the structure of hemicelluloses (Levine *et al*., 2001). However, the decomposition of woody materials could be indicated by measuring the lignin-degrading properties of fungi (Saparrat *et al*., 2008).

Fackler *et al*. (2006) stated that delignification was significant after 3 days of treatment with fungi and the activities of extracellular ligninolytic enzymes (laccase, manganese

Table-2. Holocellulose content of *Pinus roxburghii* needle leaf, twigs and barks after treatment with selected fungi

Fungi	Holocellulose content of needle leaf (%)		Holocellulose content of twigs (%)		Holocellulose content of barks (%)	
	Degradation period		Degradation period		Degradation period	
	90 days	180 days	90 days	180 days	90 days	180 days
Amanita muscaria	17.4 ± 0.20	14.0 ± 0.80	41.5 ± 0.25	38.5 ± 0.45	60.5 ± 0.20	55.1 ± 0.20
Coprinus micaceus	16.2 ± 0.45	14.8 ± 0.40	39.5 ± 0.35	37.7 ± 0.35	60.4 ± 0.65	51.0 ± 0.55
Cortinarius collinitus	18.1 ± 0.30	12.5 ± 0.50	40.0 ± 0.40	36.0 ± 0.40	61.0 ± 0.50	58.4 ± 0.70
Tricholoma album	17.8 ± 0.50	10.8 ± 0.40	39.4 ± 0.10	32.8 ± 0.35	67.5 ± 0.20	60.0 ± 0.40
Control	21.4 ± 0.10	20.2 ± 0.20	45.7 ± 0.40	44.5 ± 0.50	73.5 ± 0.40	72.5 ± 1.00

Values are as Mean ± SD of 3 replicates; Control: Refers to No fungal treatment

Table-3. Weight loss of the treated samples after extraction with hot water for 3 h.

Fungi	Hot water soluble content of Needle leaf (%)		Hot water soluble content of twigs (%)		Hot water soluble content of barks (%)	
	Degradation period		Degradation period		Degradation period	
	90 days	180 days	90 days	180 days	90 days	180 days
Amanita muscaria	34.1 ± 0.40	64.1 ± 0.45	68.5 ± 0.50	78.0 ± 0.20	68.4 ± 0.45	84.0 ± 0.40
Coprinus micaceus	36.3 ± 0.55	72.8 ± 0.05	64.0 ± 0.30	84.5 ± 0.20	64.0 ± 0.60	82.0 ± 0.40
Cortinarius collinitus	30.3 ± 0.50	62.5 ± 0.20	68.2 ± 0.45	82.5 ± 0.35	67.1 ± 0.40	80.1 ± 0.55
Tricholoma album	32.5 ± 0.40	72.1 ± 0.40	62.5 ± 0.45	80.1 ± 0.70	68.5 ± 0.70	79.0 ± 0.20
Control	30.5 ± 0.50	54.0 ± 0.50	54.5 ± 0.40	73.5 ± 0.10	64.5 ± 0.20	76.0 ± 0.40

Values are as Mean ± SD of 3 replicates; Control: Refers to No fungal treatment

peroxidase and/or lignin peroxidase) could be detected in fungal cultures. The present study needle leaves that the reduction of lignin content is 4 to 7% after 90 days and 7 to 9% after 180 days. Those results was much lower than that of Osono *et al*. (2003) who reported that lignin weight loss by Basidiomycetes could range from 23.7 to 39.6%, in *Qnercus* litter after exposure for 3 years.

Vargas-Garcia *et al.* (2007) reported that lignin degradation by *Bacillus licheniformis* activity was reached upto 68%. Osono and Takeda (2006) stated that mass loss of lignin in *Abies* ranged from 4.2 to 36.0% at 20°C and from 0.9 to 13.3% at 10°C and that in *Betula* ranged from 1.9 to 72.8% at 20° C and from 20.4 to 32.9% at 10° C. These variations due to the fungi used to degrade lignin were not as powerful on wood meal as in the previous study which was capable to degrade 14.6 to 24.9% of lignin content on the sample (Djarwanto and Tachibana, 2009). It is to be noted that the fungi involved in the lignin degradation are also different from those who worked on such degradation.

Ability of the fungi to degrade holocellulose:

The holocellulose content of *P. roxburghii* needle leaves, twigs and bark are shown in Table-2. As mentioned in lignin degradation, holocellulose content was measured after incubation of four fungi for 90 to 180 days. The initial holocellulose content of *P. roxburghii* needle leaves was 21.4%. After 90 days of treatment, the holocellulose content decreased to 16.2 to 18.1%, depending on the fungal genera. The genera *Coprinus micaceus* was most effective (16.2) at 90 days and (12.5) *Cortinarius collinitus* on 180 days (Figure-1c). The initial holocellulose content of the needle leaf are 20.2%, after 180 days of treatment, the lignin content decreased to 10.8 to 14.8% of degradation.

The initial holocellulose content of the twigs was 45.7%. After 90 days of treatment with fungi, the holocellulose content decrease ranged from 39.4 to 41.5% and the genera *Tricholoma album* was most effective (39.4) in degradation at 90 and 180 days (Figure-1d). The initial holocellulose content of twigs was 44.5%. After 180 days of treatment with fungi, the decrease in holocellulose content ranged from 32.8% to 38.5%. The initial holocellulose content of bark was 73.5%. After 90 days of treatment, the decrease in holocellulose content ranged from 60.4 to 61.0% and the genera *Coprinus micaceus* was most effective (60.4) in degradation (Figure-1b). After 180 days of treatment the holocellulose content decrease ranged from 51.0 to 60.0% when compared with initial level of 72.5% and the genera *Coprinus micaceus* was more effective (51.0%).

The holocellulose content of needle leaves, twigs and barks of *P. roxburghii* also decreased depending on the fungal genera inoculated with the materials. Besides lignin, the main components of organic matter are holocellulose, protein and lipids. Lignocellulose contain around 40% cellulose, 20% hemicellulose and 20-30% lignin (Tuomela *et al*., 2000). Bark accounts for 10-20% of woody plants and is composed of a variety of biopolymers including polysaccharides, lignin, suberin and tannins (Vane *et al*., 2006). Holocellulose is the main polymeric component of the plant cell wall, the most abundant polysaccharide and an important renewable resource. In the present study the holocellulose content of needle leaves, twigs and barks of *P. roxburghii* decreased due to the degradation of fungi inoculated, depending on the fungi the degradation percentage varied between 3.0 to 5.0% after 90 days and 5.0 to 9.0% after 180 days as reported by Blanchette (1995). The fungal capability in degrading holocellulose of *A. mangium* leaves and twigs 0.8% to 11.0% (Djarwanto and Tachibana, 2009). Osono and Takeda (2006) stated that mass loss of holocellulose or often called as total carbohydrate in *Abies* ranged from 2.5 to 16.6% at 20° C and from 0.0 to 13.2% at 10° C and that in *Betula* ranged from 2.3 to 66.8% at 20° C and from 6.4 to 48.1% at 10° C.

Hot water soluble content of treated needle leaves, twigs and barks:

The volume of the soluble content of *P. roxburghii* needle leaves, twigs and barks in hot water is shown in Table-3. It was found that the duration of inoculation in all the samples tested in this study increased in solubility in hot water. The value of soluble content varied depending on the fungi inoculated. For needle leaves, the value of soluble content was 30.3 to 36.3% as against the control sample 30.5% in 90 days. At 180 days, the value of soluble content was 62.5 to 72.8% when compared with control 54.0%.

Figure-1. Lignin, holocellulose degradation by macrofungi of *Pinus roxburghii forest* ecosystem (a-*Amanita muscaria; b-coprinus micaseus; c-Cortinarius collinitus and d-Tricholoma album*)

a-Amanita muscaria

b-Coprinus micaceus

c-Cortinarius collinitus

d-Tricholoma album

In case of twigs, the percentage was between 62.5 to 68.5 as against 54.5 for control in 90 days. In 180 days the results are between 78.0 to 84.5% when compared with control 73.5%. For bark samples the percentage of soluble content was 64.0 to 68.5% (Control 64.5%) in 90 days. At 180 days the value of soluble content was 79.0 to 84.0% as against the control 76.0%.

In this study the hot-water solubility of treated samples increased significantly with incubation time meaning that some amount of lignocellulose content was degraded, presumably supported by the monosaccharides in *P. roxburghii* sample like xylose, mannose and glucose (Pinto *et al*., 2005) which are soluble in water, besides the degradation of cellulose containing polymers and polysaccharides into simpler components like monomers through fungal activity (M. N. Abubacker and B. Kirthiga, 2015; Blanchette *et al*., 1994 and PaPanda et al, 2012). In both 90 days and 180 days of incubation, the hot water soluble content of all treated samples increased. Perhaps the soluble matter was consumed for energy by the fungi, since lignin and holocellulose content were less decreased.

CONCLUSION

The rate of degradation of *P. roxburghii* needle leaves, twigs and barks varied depending on the fungal genera inoculated. An increase in incubation time tended reducing both lignin and holocellulose content. However, the reduction rate was not significant, therefore,

more time is needed to degrade lignin rather than other components in the sample. This report will help to gain the insight of lignin and holocellulose degradation in the early stage in the natural forest ecosystem.

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Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

References

1. ASTMD1104-56, Method of test of holocellulose in wood. http://www.astm.org/standards/D1104.htm (1978).
2. ASTMD1110-87, Standard Test Methods for water solubility of wood. American Standard Test Methods. ASTM International, West Conshohocken, PA (2007).
3. Blanchette, R. A., Obst, J. R. and Timell, T. E., Biodegradation of compression wood and tension wood by white and brown rot fungi. *Holzforschung.* **48**: 34-42 (1994).
4. Blanchette, R. A., Degradation of lignocellulose complex in wood. *Canadian Journal of Botany.* **73**: 999-1010 (1995).
5. Dekker, R. F. H., Barbosa, A. M. and Sargent, K., The effect of lignin-related compounds on the growth and production of laccases by the ascomycete *Botryosphaeria* sp. *Enz. Microbiol. Technol.* **30**: 374-380 (2002).
6. Djarwanto and Tachibana, S., Screening of fungi capable of degrading lignocelluloses from plantation forests. *Pak. J. Biol. Sci.* **12**: 669-675 (2009).
7. Dubeux, Jr. J. C. B., Sollenberger, L. E., Interrante, S. M., Vendramini, J. M. B. and Stewart, Jr. R. L., Litter decomposition and mineralization in bahia grass pastures managed at different intensities. *Crop Science*. **46**: 1305-1310 (2006).
8. Fackler, K. C., Gradinger, C., Hinterstoisser, B., Messner, K. and Schwanninger, M., Lignin degradation by white rot fungi on spruce wood shavings during short-time solid-state fermentations monitored by near infrared spectroscopy. *Enzyme Microbiol. Technol.* **39**: 1476-1483 (2006).
9. Fioretto, A., Di Nardo, C., Papa, S. and Fuggi, A., Lignin and cellulose degradation and nitrogen dynamics during decomposition of three leaf litter species in a mediterranean ecosystem. *Soil Biology and Biochemistry*. **37**: 1083-1091 (2005).
10. Hakala, T. K., Characterization of the lignin - modifying enzymes of the selective white-rot fungus *Physisporinus rivulosus*. Helsinki University Printing House, Helsinki, Finland. pp. 60 (2007).
11. Levine, L. H., Heyenga, A. G., Levine, H. G., Choi, J. W., Davin, L. B., Krihorian, A. D. and Lewis, N. G., Cell wall architecture and lignin composition of wheat developed in microgravity environment. *Physiochemistry*. **57**: 835-846 (2001).
12. Martinez, A. T., Speranza, M., Ruiz-Duenas, F. J., Ferreira, P. and Camarero, S., Biodegradation of lignocellulosic: Microbiol, chemical and enzymatic aspects of fungal attack to lignin. *International Journal of Microbiology*. **8**: 195-204 (2005).
13. Miura, K. and Kudo, M., An agar-medium for aquatic hypomycetes. Transactions of the Mycological Society of Japan. **11**: 116-118 (1970).
14. M. N. Abubacker and B. Kirthiga. Bioremediation potential of textile *Aspergillus flavus* teak 07 Against textile dye and their toxicity assessment *Biolife* 3(1); 54-60 (2015).
15. Ohkuma, M., Maeda, Y., Johjima, T. and Kudo, T., Lignin degradation and roles of white rot fungi: Study on an efficient symbiotic system in fungus growing termites and its application to bioremediation. *Riken Rev. Foe. Econ. Sci. Res.* **42**: 39-42 (2001).
16. Osono, T., Fukesawa, Y. and Takeda, H., Role of diverse fungi in larch needle-litter decomposition. *Mycologia*. **95**: 820-826 (2003).

17. Osono, T. and Takeda, H., Fungal decomposition of *Abies* needle and *Betula* leaf litter. *Mycologia*. **98**: 172-179 (2006).
18. PaPanda, S.S, Sahoo, K, Muduli,S.D, Sahoo,G, Ahemad, M.D.J, Nayak, B.B and Dhal, N.K chromium tolerant indigenous fungal strains from Industrial effluents of Anugul district, Odisha, India. Biolife. 2(2), 634-640 (2012).
19. Petterson, R. C., The chemical composition of wood. In: The chemistry of solid wood. Rowell, R. M. (Ed.), Americal Chemical Society, Washington, DC, pp. 57-126 (1984).
20. Pinto, P. C., Evtuguin, D. V. and Neto, C. P., Structure of hardwood gucuronoxylans: Modifications and impact on pulp retention during wood kraft pulping. *Carbohyd. Polym*. **60**: 489-497 (2005).
21. Saparrat, M. C. N., Rocca, M., Aulicino, M., Arambarri, A. M. and Balatti, P. A., *Celtis tala* and *Scutia buxifolia* leaf litter decomposition by selected fungi in relation to their physical and chemical properties and lignocellulolytic enzyme activity. *Eur. J. Soil Biol.* **44**: 400-407 (2008).
22. Swift, M. J., Heal, O. M. and Anderson, J. M., Decomposition in terrestrial ecosystems. *Journal of Ecology*. **34**: 45-50 (1979).
23. TAPPI Standard Methods, Fibrous Materials and Pulp Testing. Technical Association of Pulp and Paper Industry, Atlanta (2009).
24. Tuomela, M., Vikman, M., Hatakka, A. and Itavaara, M., Biodegradation of lignin in a compost environment: A review. *Bioresour. Technol.* **72**: 169-183 (2000).
25. Vane, C. H., Drag, T. C. and Snape, C. E., Bark decay by the white-rot fungus *Lentinula edodes,* Polysaccharide loss, lignin resistance and the unmasking of suberin. *Int. Biodeter. Biodegradat.* **57**: 14-23 (2006).
26. Vargas-Garcia, M. C., Suarez-Estrella, F., Lopez, M. J. and Moreno, J., *In vitro* studies on lignocelluloses degradation by microibial strains isolated from composting process. *Int. Biodeter. Biodegradat.* **59**: 322-328 (2007).
27. Watanabe, T., Watanabe, Y. and Nakamura, K., Biodegradation of wood and dual cultures of selected two fungi determined by chopstick method. *J. Biosci. Bioeng.* **95**: 623-626 (2003).
28. Willfor, S. A., Sundberg, A., Pranovich, A. and Holmbom, B., Polysaccharides in some industrially important hardwood species. *Wood Science Technology*. **39**: 601-617 (2005).

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