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Light microscopic analysis of *Tectona grandis* L.f. wood inoculated with *Irpex lacteus* and *Phanerochaete chrysosporium*

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Abstract Felling of immature teak (*Tectona grandis* L.f.) trees or delay in transport of wood logs from felling sites provide platform to microbial attack. Among them, white rot fungi are central driving force that degrades wood and causes severe economic loss. In contrast, *Phanerochaete chrysosporium* and *Irpex lacteus* are more extensively studied for their ability to degrade synthetic dyes and polyaromatic compounds. Therefore, in the present study, both the fungi collected from the Gujarat forest were utilised for in vitro decay test to assess their potential in lignin degradation, extent of cell wall damage and pattern of wood decay in sound blocks of teak. In the early stage of fungal inoculation, there was a negligible amount of weight loss; after 1 month it became rapid and highest weight loss (30.05 % by *P. chrysosporium* and 27.97 % by *I. lacteus*) was observed at the end of 120 days. Mycelial invasion occurred through vessels, from vessels to axial and ray parenchyma and subsequently into xylem fibres. Both the strains showed selective delignification and the first symptom of degradation was defibration, separation of rays, and formation of boreholes on ray cell walls at an advanced stage. Xylem fibres and parenchyma cells lost their integrity and collapsed completely. Among all the cell types, parenchyma cells and fibres were more vulnerable to fungal attack, while vessels were resistant to the activity of lignolytic enzymes.

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Lichtmikroskopische Untersuchung von mit *Irpex lacteus* und *Phanerochaete chrysosporium* befallenem Teakholz (*Tectona grandis* L.f.)

Zusammenfassung Das Fällen junger Teakbäume (*Tectona grandis* L.f.) oder Verzögerungen beim Abtransport der gefällten Holzstämme bietet eine ideale Plattform für mikrobiellen Befall. Vorwiegend Weißfäulepilze sind verantwortlich für die Holzzerstörung, die schwerwiegende wirtschaftliche Schäden verursacht. Von den Weißfäulepilzen *Irpex lacteus* und *Phanerochaete chrysosporium* wurde bisher vorwiegend der Abbau von synthetischen Farbstoffen und polyaromatischen Bestandteilen untersucht. Aus diesem Grunde wurden hier die beiden aus dem Gujarat Forest stammenden Pilze für In-vitro Pilzabbauversuche verwendet, um zu untersuchen, inwieweit diese Lignin abbauen und die Zellwand beschädigen und wie die Holzzerstörung von nicht befallenen Teakholzprüfkörpern abläuft. Zu Beginn der Pilzbeimpfung war der Masseverlust unerheblich; nach einem Monat beschleunigte sich dieser und nach 120 Tagen wurde der höchste Masseverlust (30,05 % bei *Phanerochaete chrysosporium*, 27,97 % *Irpex lacteus*) verzeichnet. Das Myzel befiel erst die Gefäße, dann das axiale und Markstrahl-Parenchym und schließlich die Fasern des Xylems. Beide Stämme zeigten selektive Delignifizierung und die ersten Symptome der Zerstörung waren Zerfaserung, Trennung der Markstrahlen und Durchdringung der Markstrahlzellwände im fortgeschrittenen Stadium. Xylemfasern und Parenchymzellen wurden beschädigt und kollabierten vollständig. Unter all den Zellarten waren Parenchymzellen und Fasern für Pilzbefall mehr anfällig, wohingegen die Gefäße gegenüber lignolytischen Enzymen resistent waren.

1 Introduction

Wood is the natural source of renewable energy and plays an important role in the national economy. The main role of wood is not only to provide energy but it also provides building material and furniture for commodities and as a raw material for paper, paper products, boards and wood products (Plomion et al. 2001). It is also one of the imperative sinks for atmospheric carbon dioxide stored in the form of cell wall material. However, many times living trees as well as wood logs are invaded by microorganisms causing severe economic loss due to structural damage in the wood components of forest trees and butt and root rot diseases of fruit trees (Nicolotti et al. 2010). Among them, white rot fungi are central driving forces that have the ability to degrade lignin selectively or simultaneously along with other cell wall polysaccharides (Blanchette 1984a, b; Maloy and Murray 2001; Schwarze 2007; Koyani et al. 2010; Sanghvi et al. 2013). In contrast, members of ascomycetes and deuteromycetes also attack wood but most of them are unable to degrade lignin, a major and complex biopolymer which is an important constituent of wood cell wall (Blanchette 1984a, b; Adaskaveg et al. 1995; Maloy and Murray 2001; Schwarze 2007; Sanghvi et al. 2013).

Owing to lignin degradation ability, white rot fungi are extensively studied due to their higher potential in the production of ligninolytic enzymes that are used in bioremediation process. Among them, *Irpex lacteus* and *Phanerochaete chrysosporium* are extensively studied by several workers (Datta et al. 1991; Baborová et al. 2006; Gassara et al. 2010; Erkurt 2010; Koyani et al. 2013) but similar information is lacking on their ability to degrade wood, its pattern of wood decay and extent of damage caused. In the forest areas, both fungi are observed frequently on wood logs of different trees including teak. In the present study, several wood rot fungi were collected from naturally infected wood samples from Gujarat Forests. Since, *Irpex lacteus* and *Phanerochaete chrysosporium* are known in the literature for high lignolytic activity, it was expected to damage the wood within a short period.

Tectona wood is prized for its valuable timber industry throughout the world and widely used for heavy construction and fine quality furniture, doors and windows (Nocetti et al. 2011). It is therefore under cultivation in different parts of the tropical countries other than its natural habitat. Various aspects, such as seasonality of vascular cambium (Rao and Dave 1981; Rao and Rajput 1999; Priya and Bhat 1997), wood properties and geographical variations in wood density (Bhat and Priya 2004; Varghese et al. 2000; Macchioni et al. 2007), properties of juvenile wood (Priya and Bhat 1997), province effect on growth ring structure (Nocetti et al. 2011) and effect of insect defoliation and false ring formation (Priya and Bhat 1998; Rajput et al. 2005; Kokutse et al. 2009, 2010) have been studied in detail. Many times teak wood logs stored in the forest depot, saw mills and window frames in old buildings exposed to open environment show fungal fruiting bodies on them. In spite of its high timber value, damage caused by these fungi on teak wood has not been investigated. Since, fruiting bodies of both the fungi are mostly observed on the surface, i.e. sapwood of wood logs (which is less resistant to fungal attack), sapwood is utilised in the present study to investigate the ability of *Irpex lacteus* and *Phanerochaete chrysosporium* in degrading teak wood by in vitro laboratory decay test and to characterise wood decay pattern by anatomical methods using light microscope.

2 Materials and methods

2.1 Source of fungi

Irpex lacteus and *Phanerochaete chrysosporium* used in this study were isolated from the infected teak wood samples collected from Junagarh (Saurashtra region) forest of Gujarat State. Small pieces of infected wood samples were surface sterilized by 0.1 % $HgCl_2$ for 40–45 s with intermediate washing by sterile distilled water. Subsequently, these wood pieces were treated with 70 % ethanol for few seconds and inoculated on 2.5 % Malt Extract Agar (MEA) media. Pure cultures were established by serial transfer and cultures were maintained at 4 °C. Fungal DNA was extracted according to the method described by Möller et al. (1992) and sent to Chromous Biotech Pvt. Ltd., Banglore for PCR sequencing and molecular identification.

2.2 Wood decay test

Cubic wood blocks ($2 \times 2 \times 2$ cm^3) from sound sapwood portion of *Tectona grandis* L.f. were prepared from the stem disc free from knots. Prior to rewetting, some of the wood blocks were marked and weighed prior to and after fungal inoculation to study the weight loss. Marked and unmarked wood blocks were rewetted in water for 24 h to obtain optimum moisture level to facilitate fungal action. Subsequently, these blocks were autoclaved at 120 °C for 30 min; surface sterilized with routine method and kept in an autoclaved Petri dish containing Malt Extract Agar Media. Thereafter, these Petri dishes were inoculated with 15-day-old pure culture of *Irpex lacteus* and *Phanerochaete chrysosporium*. Each Petri dish containing four blocks was incubated for 30, 60, 90 and 120 days at 27 ± 1 °C and 70 % relative humidity. For each incubation period, four wood blocks without fungal inoculation were maintained as control. At the time of harvesting, three treated

blocks and one control block were removed and cleaned with a brush to take out mycelia superficially growing on the external surface of the blocks. This experiment was performed in triplicates for every incubation period.

Unmarked wood blocks were fixed in formaldehyde acetic acid alcohol (Berlyn and Miksche 1976) for histological study while marked wood blocks were oven dried and percent weight loss was calculated as: weight of oven dried wood block after fungal incubation/weight of oven dried original wood block) × 100. After 24 h of fixation these blocks were transferred to 70 % ethanol for further processing and storage.

2.3 Sample processing

Suitably trimmed samples were dehydrated with Tertiary Butyl Alcohol series and processed by routine method of paraffin embedding as described by Berlyn and Miksche (1976). Transverse, radial and longitudinal sections of 12–15 μm thickness were obtained using a Leica Rotary Microtome (RM 2035). Some of the samples were also sectioned directly on the sliding microtome and stained with Safranin-Astra blue combination (Srebotnik and Messener 1994). After dehydration in ethanol-xylene series, the sections were mounted in dibutyl phthalate xylene (DPX). Important results were micro-photographed with a Leica DM 2000 research microscope.

3 Results

3.1 Wood structure

Secondary xylem of *Tectona grandis* L.f. was ring porous with distinct growth rings. Sapwood was white or pale yellow while heartwood was light golden brown in fresh wood and brown to dark brown in dry wood, often with darker streaks. The secondary xylem was composed of vessels, fibres, axial and ray parenchyma cells. Parenchyma formed thin sheaths around the vessels, which were distinct in early wood but visibility disappeared in latewood and may be seen only with a hand lens. Rays were uni-multiseriate with oval to polygonal cluster of ray cells. They were 351–521 (±11.27) μm in height and 56–74 (±2.37) μm wide. Vessels were mostly solitary, oval to oblong in outline with simple perforation plates on the transverse to slightly oblique terminal ends. Vessel elements were measured about 184–278 (±7.70) μm and 50–135 (± 6.86) μm in length and width, respectively. Length of the xylem fibres ranged from 1,075 to 1,364 (±9.59) μm. Axial and ray parenchyma possessed oval to circular simple pits.

3.2 Wood colonisation and weight loss

Both, *Irpex lacteus* and *Phanerochaete chrysoporium* completely ramified over the wood blocks within 10–12 days of their inoculation. Hyphae of both the strains entered into the wood cells through the vessel lumen and vessel associated axial parenchyma. From the vessels, hyphae traversed into the neighbouring rays and gradually extended to all directions including xylem fibres (Fig. 1a) and adjacent axial and ray parenchyma cells. Hyphae traversed from one cell to the next through the pits present on their walls and formed a mycelial mat in the ray cells. When the fungal mycelia moved from one cell to the next through the pits, the mycelia adjust their diameter to the relatively narrow pit diameter. No visual damage was observed in the cell walls within 15 days of incubation.

After 30 days of inoculation, no appreciable weight loss was observed in the wood blocks inoculated with both the strains. Thereafter, weight loss became rapid in the succeeding days and showed 27.97 % by *Irpex lacteus* and 30.05 % by *Phanerochaete chrysosporium* after 120 days (Table 1). However, negligible weight loss of control wood samples was observed after 120 days, which might be associated with the preparation procedure.

3.3 Degradation of cell components

Wood blocks inoculated with *I. lacteus* showed not much appreciable alterations in the cell wall after 30 days except that some of the fibres and ray cells located at the periphery of the block showed separation from the middle lamellae (Fig. 1b). As a result, the portion of secondary wall exposed to ligninolytic enzymes produced by the fungi was stained blue with Astra blue instead of red by safranin (Fig. 1b). Separation of fibres and ray cells was observed only adjacent to the rays and vessel elements. Occurrence of fungal mycelia in the fibre lumen was observed frequently in wood blocks inoculated with both the strains (Fig. 1c). However, wood cells located in the central part of the blocks were intact and not much alteration was observed in cell walls. In contrast, wood blocks inoculated with *P. chrysosporium* showed no visible signs of fungal attack on the cell walls of xylem rays (Fig. 1d) and vessels but fibres showed the first sign of structural alterations, which were typical of selective delignification, i.e. separation of fibres from the middle lamella (Fig. 1e). Similar to former fungal strain, separation of xylem elements adjacent to the rays (Fig. 1d) and vessels was more common in all the treated wood blocks. At this stage, delignification was mostly restricted to the cells located at the external (peripheral) part of the wooden blocks.

After 60 days, there was no pronounced cell wall changes in different cell types of xylem but separation of

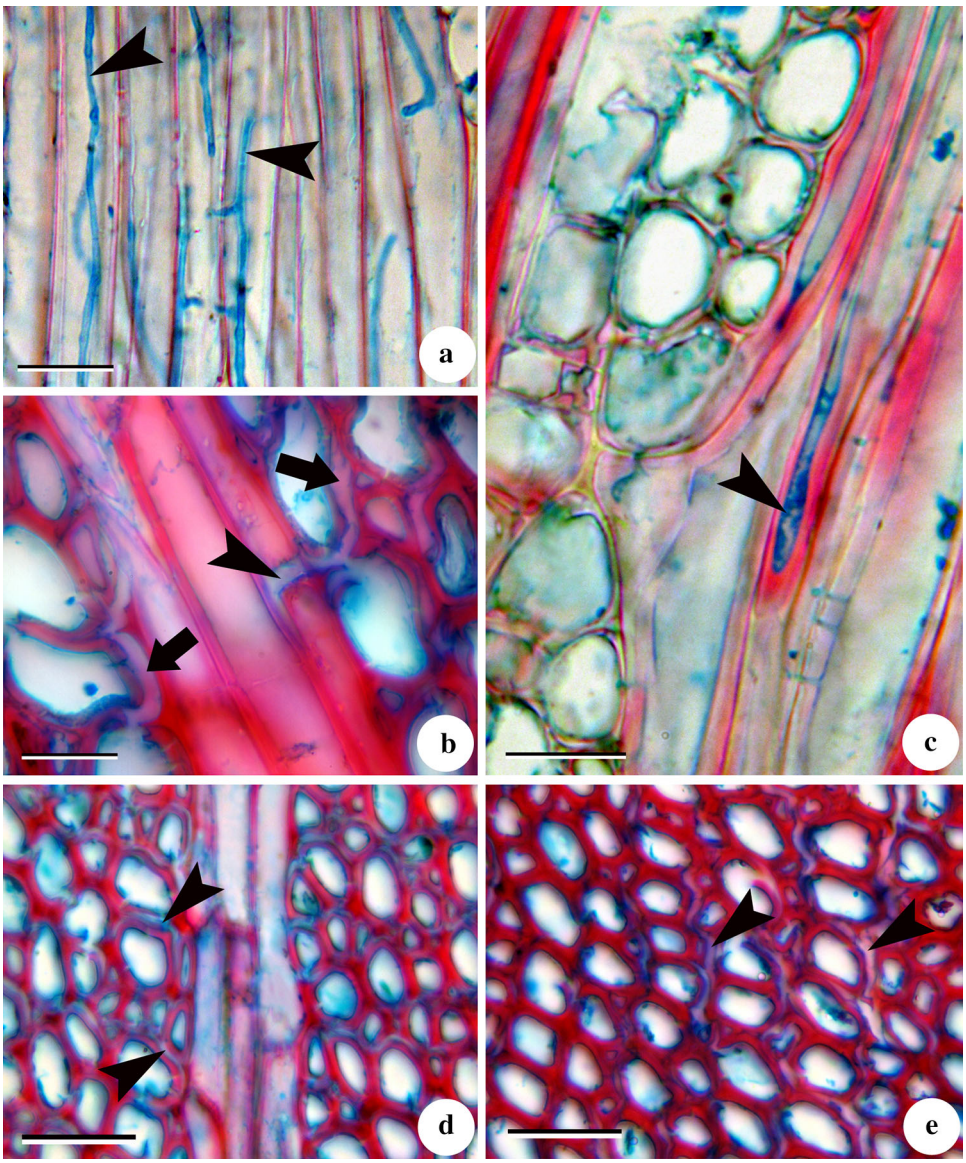

Fig. 1 Longitudinal (**a**, **c**) and transverse (**b**, **d**–**e**) view of teak wood inoculated with *Irpex lacteus* and *Phanerochaete chrysosporium*. **a** Xylem fibres infested by *P. chrysosporium*. Note the fungal hyphae in fibre lumen (*arrowheads*). **b** Xylem invaded by *I. lacteus* showing separation of ray cell walls (*arrowhead*) and xylem fibres adjacent to rays (*arrows*). **c** Lumen of xylem fibre showing hyphae of *I. lacteus* (*arrowheads*). **d** Separation of xylem fibres (*arrowhead*) due to dissolution of middle lamella by the activity of lignolytic enzymes produced by *P. chrysosporium*. **e** Xylem inoculated with *P. chrysosporium* showing separation of fibres (*arrowheads*). *Scale bar* 100 µm; **b**–**e** *scale bar* 75 µm
Abb. 1 Längs- (**a**, **c**) und Querschnitt (**b**, **d**–**e**) von mit *Irpex lacteus* und *Phanerochaete chrysosporium* befallenem Teakholz. **a** Mit *P. chrysosporium* befallene Xylemfasern (Pfeile). Zu beachten sind die Hyphen in den Faserlumen. **b** Mit *I. lacteus* befallenes Xylem, das die Trennung der Markstrahlzellwände (Pfeile) und der an die Markstrahlen angrenzenden Xylemfasern (Richtungspfeile) zeigt. **c** Lumen einer Xylemfaser mit *I. lacteus* Hyphen (Pfeil). **d** Trennung der Xylemfasern (Pfeile) aufgrund des Zerfalls der Mittellamelle durch von *I. lacteus* erzeugte lignolytische Enzyme. **e** Mit *P. chrysosporium* beimpftes Xylem, das die Trennung von Fasern zeigt (Pfeile)

these elements scattered uniformly throughout the blocks. In transverse view, many of the fibres and ray cells showed concentric delignification starting from middle lamellae towards lumen (Fig. 2a, b). As a result, the secondary wall adjacent to the middle lamellae was stained blue with Astra blue instead of red by safranin. Effect of fungal enzymes was predominantly more pronounced at cell corners of ray cells and showed distinct intercellular spaces (Fig. 2a), which consequently separated the ray cells (Fig. 2b). At this stage, ray cell corners became thinner and discoloured due to the absence of lignin and stained blue with Astra blue. Compared to *I. lacteus* (Fig. 2a), blocks inoculated with *P. chrysosporium*, were relatively more delignified and showed cell wall thinning and complete separation of

Table 1 Average percent weight loss of *Tectona* sapwood inoculated with *Irpex lacteus* and *Phanerochaete chrysosporium*
Tab. 1 Mittlerer prozentualer Masseverlust von mit *Irpex lacteus* und *Phanerochaete chrysosporium* beimpftem Teaksplintholz

Decay fungi	% weight loss (60 days)	% weight loss (90 days)	% weight loss (120 days)
Irpex lacteus	9.75 (± 5.23)	15.06 (± 4.84)	27.97 (± 4.97)
Phanerochaete chrysosporium	10.61 (± 3.41)	17.92 (± 4.94)	30.05 (± 5.89)

ray cells (Fig. 2b). Except for cell wall thinning and separation of ray cells, no noticeable sign of cell wall change was observed in the morphology of xylem elements in the transverse and longitudinal plane.

Separation of vessel elements without any significant effect on their wall was recorded only after 90 days of fungal inoculation. Narrow vessels and tracheids also showed separation due to dissolution of middle lamella (Fig. 2c). At this stage, fibres were separated consistently throughout the wood blocks. When compared with other xylem elements, rays became more susceptible to fungal invasion. Pits on the ray and axial parenchyma walls became larger in size and irregular in shape (Fig. 2d). Subsequently, several additional boreholes were formed on the lateral walls (Fig. 2e) due to the activity of fungal enzymes. Though dissolution of middle lamella and separation of cells were common in axial elements, early wood axial parenchyma cells were more vulnerable to enzyme activity as compared to fibres and axial parenchyma of the late wood (Fig. 2f).

Wood blocks exposed to both the fungi for 120 days were severely damaged due to ligninolytic enzymes secreted by the fungal mycelia. Cell walls were completely bleached out and not only stained blue coloured with Astra blue due to loss of lignin but also showed complete disintegration of cells (Fig. 2f). At this stage, xylem fibres were not only separated from each other but also lost their rigidity due to removal of lignin from the walls. Formation of several boreholes on the lateral walls of fibres and extensive thinning of many ray cells resulted in partial or complete disintegration of rays. In case of xylem fibres and axial parenchyma, pits present on their lateral wall also became larger, irregular in size, which measured about 4–6 µm in diameter, sometimes 2–3 such boreholes fused together and formed relatively larger boreholes. Vessels were more resistant in contrast to fibres and axial parenchyma, while rays were the most vulnerable cell types to fungal enzymes. At this stage, vessel walls also showed uneven thinning; therefore, most often vessel walls were broken at the time of sectioning.

4 Discussion

Longevity or natural durability of timber depends upon various factors such as genetic makeup of the species, surrounding environment, moisture content in the atmosphere, growth rate and age of that specific species (Suprapti 2010). Therefore, fortitude of any wood is also determined by its natural durability, they are always susceptible to deterioration due to wood borers, beetles or wood rot fungi. *Tectona* is one of the important timbers known for natural durability and strength. However, rainy season provides ideal situation for different fungi to invade wood logs under natural conditions. During monsoon, high moisture content and relatively low temperature of the surrounding environment/atmosphere favors fungal growth while wood surface provides a suitable platform to establish fungal infection. In the present study, *Irpex lacteus* and *Phanerochaete chrysosporium* isolated from naturally infected wood samples were utilized for the in vitro decay test. Both the fungi belong to basidiomycota and show selective delignification pattern, which was manifested by separation of xylem cells. Anagnost (1998) and Luján Luna et al. (2004) considered this feature as the most reliable indicator of selective delignification. Selective delignification by both fungal isolates may be supported by the staining technique described by Srebotnik and Messener (1994), which contributes to discriminate between simultaneous and selective delignification (Luján Luna et al. 2004). Other signs of simultaneous rot are cell wall thinning, borehole formation, rounded pit erosion and formation of erosion troughs (Anagnost 1998; Luján Luna et al. 2004). All these features are characteristic to both the fungal isolates except for the formation of erosion troughs which was not observed in this study.

Mycelia of both fungal isolates although totally ramified over the wood blocks and all the cells types of secondary xylem were invaded, no appreciable weight loss was observed even after 1 month. It appears that in the early stage of fungal invasion, mycelia survive on the reserved photosynthate (if any) available in the wood cells particularly ray and axial parenchyma. Similar behaviour of wood rot fungi has been correlated with the presence of several low molecular weight compounds present in the wood that might have been consumed first at the initial stage of invasion without damaging cell wall polymers (Fengel and Wegener 1989; Worall et al. 1997). Percent weight loss of wood blocks increased rapidly after 30 days, and showed 27.97 % loss by *Irpex lacteus* and 30.05 % by *Phanerochaete chrysosporium* after 120 days. Rapid increase in rate of weight loss seems to be associated with complete consumption of low molecular weight compounds present in the wood and the degradation potential of fungi. Luján Luna et al. (2004) reported about 50–60 % weight loss of

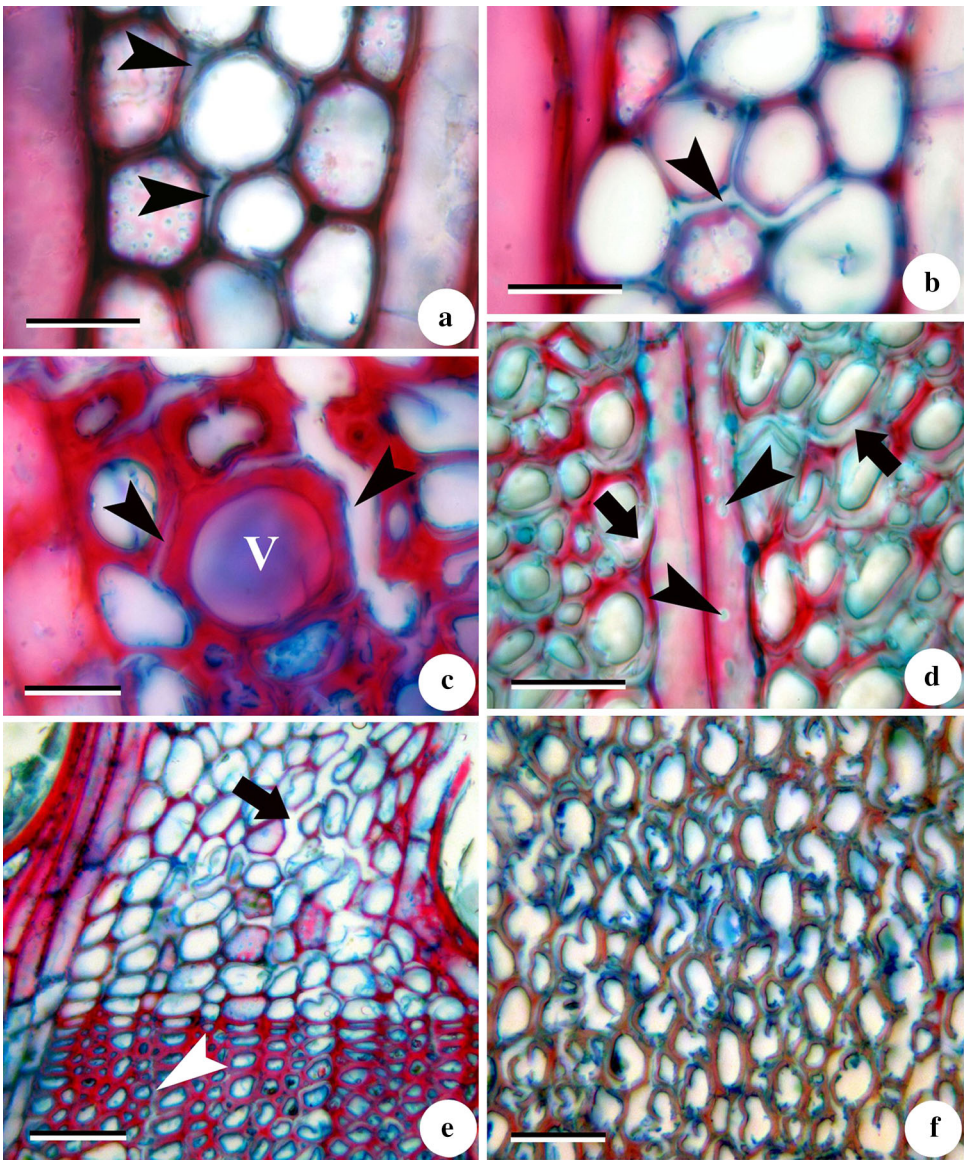

Fig. 2 Tangential longitudinal (**a**, **b**) and transverse (**c**–**f**) view of teak wood inoculated with *Irpex lacteus* and *Phanerochaete chrysosporium*. **a** Separation of rays cells at cell corners (*arrowheads*) in wood blocks inoculated by *I. lacteus*. **b** Separation of ray cells (*arrowhead*) in wood blocks inoculated by *P. chrysosporium*. Note that separation of cells and wall thinning is relatively more as compared to **a**. **c** Dissolution of middle lamella and separation of vessels (*arrowheads*) in wood inoculated with *P. chrysosporium* after 90 days. **d** Separation of fibres adjacent to ray (*arrows*). *Arrowheads* indicate development of additional bore holes on the ray cell walls by *I. lacteus*. **e** Separation of early wood parenchyma (*arrow*) and late wood fibres (*arrowhead*) by enzymatic activity of *P. Chrysosporium*. **f** Severely affected secondary xylem at the end of 120 days of incubation showing loss of cell integrity and deformed xylem cells. **a**–**e** *scale bar* 75 µm; **f** *scale bar* 100 µm

Abb. 2 Längsschnitt tangential (**a**, **b**) und Querschnitt (**c**–**f**) von mit *Irpex lacteus* und *Phanerochaete chrysosporium* beimpftem Teakholz. **a** Trennung der Markstrahlzellen an den Zellecken (Pfeile) in mit *I. lacteus* beimpften Prüfkörpern. **b** Trennung der Markstrahlzellen (Pfeile) in mit *P. chrysosporium* beimpften Prüfkörpern. Zu beachten ist, dass die Zelltrennung und die Wandverdünnung verglichen mit **a** größer sind. **c** Zerfall der Mittellamelle und Trennung der Gefäße (Pfeile) in mit *P. chrysosporium* beimpften Prüfkörpern nach 90 Tagen. **d** Trennung der an die Markstrahlen angrenzenden Fasern (Richtungspfeile). Die Pfeile zeigen die Entwicklung von zusätzlicher Durchdringung der Markstrahlzellwände durch *I. lacteus.* **e** Trennung des Parenchyms von Frühholz- (Richtungspfeil) und Spätholzfasern (Pfeil) durch Enzymaktivität von *P. chrysosporium.* **f** Stark befallenes sekundäres Xylem nach 120-tägigem Befall, das den Verlust der Zellstruktur und verformte Xylemzellen aufweist

poplar wood within 2–5 months by *Pycnoporus sanguineus* and *Gonoderma lucidum*. Therefore, it appears that weight loss differs from fungal species to species or within species and natural durability of the wood.

In the early stages of fungal attack, hyphae enter through the vessel lumen and further invasion is facilitated by ray cells and vessel associated parenchyma cells for wide spread distribution of mycelia. Similar mechanism of

fungal invasion has also been reported in *Ailanthus* (Koyani et al. 2010) and other trees (Rayner and Boddy 1988; Schwarze 2007; Schwarze et al. 2004; Sanghvi et al. 2013). The presence of fungal hyphae in all the cell types coincides with the observations by Luján Luna et al. (2004). Access to adjacent cells is reported to occur through the simple or bordered pits present on the walls which subsequently become larger in diameter. It may also penetrate directly by forming boreholes with the help of specialised cell wall degrading hyphae (Schwarze et al. 2004; Schwarze 2007; Koyani et al. 2010; Sanghvi et al. 2013). In this study, formation of several boreholes is also observed on the walls of xylem fibres, axial and ray parenchyma cells. According to Anagnost (1998) and Luján Luna et al. (2004), fungi that selectively delignify wood may also form boreholes that are similar to simultaneous rot.

Among the different cell types of xylem, ray cells are highly vulnerable and showed significant damage due to fungal activity, while vessels are relatively resistant, except the bleaching effect on the vessel wall pits. The observations here are in agreement with Luján Luna et al. (2004) who reported similar feature in case of populous wood treated with *Pycnoporus sanguineus* and *Gonoderma lucidum*. Similar feature has also been observed in an earlier study on *Ailanthus excelsa* (Koyani et al. 2010) and *Azadirachta indica* (Sanghvi et al. 2013). Blanchette (1988) named various reasons for the persistence of vessels in wood degraded by white rot fungi. Iiyama and Pant (1988) reported higher content of syringyl monomer in fibres and ray parenchyma, while fibre tracheids contain higher guaiacyl monomer content (Faix et al. 1985; Blanchette 1988). Therefore, hardwood is degraded rapidly by white rot fungi due to higher content of syringyl units of lignin than the guaiacyl (Faix et al. 1985; Luján Luna et al. 2004; Koyani et al. 2010). However, further chemical studies are required to confirm this hypothesis of increasing concentration of guaiacyl monomer with persistence of vessels in *Tectona*.

At an advanced stage of decay, completely delignified tissue stained blue with safranin-astra blue combination. Complete removal of lignin from the cell wall results in separation of fibres and ray cell which leads to loss of cell integrity and consequently leads to collapse. Though, vessels are resistant to fungal action; separation of vessels at this stage may be associated with the dissolution of compound middle lamella. Severance of cells in response to complete degradation of middle lamella in *Populous* treated with *G. lucidum* is also reported by Luján Luna et al. (2004).

5 Conclusion

Irpex lacteus and *Phanerochaete chrysosporium* show similar white rot pattern resulting in 27–30 % weight loss after 120 days. As expected, both the fungal strains have higher potential of lignin degradation as is reported for textile dye and polyaromatic compound degradation and may serve as potential species to be utilised in biopulping process. However, further studies on the wood that is used in the paper industry are needed. Fibres and parenchyma cells are more vulnerable to ligninolytic activity of the fungal enzymes and at an advanced stage of wood decay; fibres show complete separation and collapse, while vessels are the most resistant cell type. Rapid weight loss of wood blocks after 30 days may be associated with complete utilization of low molecular weight compounds present in the wood.

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