Extracellular Laccase Activity among *Ganoderma* and *Coriolopsis* Species Grown on Lignocellulosic Wastes

Mei-Ling Han,[a,#] Jing Yang,[a,#] Jian-Jun Ma,[a,b] Chun-Rui Wang,[a] Si-Yu Chen,[a] Ming-Xin Xu,[a] Qi-Yue Yang,[a] Lu-Sen Bian,[c] Xun-You Yan,[a,d] and Qi An [a,d,*]

The extensive application of laccase in various fields depends on the supply of a large amount of laccase with high activity and low cost. Interest on the screening of fungal strains suitable for obtaining massive high-activity laccase in solid-state fermentation with suitable low-cost lignocellulosic wastes has increased. The present work determined the laccase secretion from different species belonging to genera *Ganoderma* and *Coriolopsis* fermented on different lignocellulosic wastes. Maximal laccase activity was obtained from *Ganoderma lingzhi* Han 1345 grown on rice straw for 9 days, and indicated the capacity of secreting laccase of *G. lingzhi* Han 1345 was superior to that of *Coriolopsis trogii* Han 1211, *C. strumosa* Han 1356, and *G. applanatum* Han 1578. The presence of cottonseed hull was more favorable for *Coriolopsis* species secreting laccase, while *Ganoderma* species were more preferred to secrete laccase on rice straw. Further, the consistent substrate preference in laccase production of different species in the same genus was first exhibited. The results are useful to screen and obtain new species with superior capacity of secreting laccase and suitable low-cost lignocellulosic wastes as fermentation substrate for further industrial application of laccase.

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Contact information: a: College of Life Science, Langfang Normal University, Langfang 065000, Hebei, China; b: Forestry Engineering and Biodiversity Conservation Technology Research and Development Center in Langfang City, Langfang 065000, Hebei, China; c: Experimental Centre of Forestry in North China, Chinese Academy of Forestry, Beijing 102300, China; d: Technical Innovation Center for Utilization of Edible and Medicinal Fungi in Hebei Province, Langfang 065000, Hebei, China;
 # Mei-Ling Han and Jing Yang contributed equally to this article;
** Corresponding author: fungiqian@163.com*

INTRODUCTION

Lignocellulosic biomass, considered as the most abundant and renewable resource on earth, is mainly composed of cellulose (30 to 50% of dry matter weight), hemicellulose (20 to 40% of dry matter weight), and lignin (15 to 25% of dry matter weight), in addition to a small amount of structural proteins, lipids, and ash. Certainly, a part of it is usually considered as wastes, like corncob, leaves, sweet sorghum bagasse, coffee shell, corn stover, wheat straw, and sugar cane (Zhou *et al*. 2014; Sun *et al*. 2015; Moreira *et al*. 2016; Huang *et al*. 2019; Karagoz *et al*. 2019; Kumar *et al*. 2021). Generally, the main way of disposing of lignocellulosic wastes is to discard it to rot or burn it directly. Lignocellulose contains a variety of structural units, such as five-carbon sugars, six-carbon sugars, and aromatic compounds. The chemical diversity of these structural components allows production of different chemical products from lignocellulose. Another application, of course, is to grow fungi to produce various types of bioactive substances (Atilano-Camino *et al.* 2020; Gaikwad and Meshram 2020; Backes *et al*. 2022). Laccase is a biological active substance that has been widely studied by researchers because it is an ancient enzyme and has many applications (Unuofin *et al*. 2019; Zhou *et al*. 2021; Khatami *et al*. 2022).

Laccase (benzenediol: oxygen oxidoreductases, EC 1.10.3.2) belonging to a group of enzymes called blue multi-copper oxidases, is capable of oxidizing phenols and aromatic amines by reducing molecular oxygen to water (Hou *et al*. 2004). Laccase is widely distributed in nature, such as various plants, fungi, bacteria, and some insects (Unuofin *et al*. 2019; Zerva *et al*. 2019; Khatami *et al*. 2022). Laccase is one of the most important ligninolytic enzymes involved in lignin degradation (An *et al*. 2021a,b). Recent studies have indicated that laccases display broad substrate specificity and play an important role in the global carbon cycle (Wang *et al*. 2019; Teigiserova *et al*. 2021). Moreover, laccases are applied in many biotechnological processes and applications, including biodegradation, bioremediation, biosensors, biopharmaceuticals, decolorization and detoxification of textile effluents, biobleaching, and biopulping of paper industry, food chemistry and biofuels (Minussi *et al*. 2002; Sharma *et al*. 2019; Wang *et al*. 2019; Huang e*t al*. 2020; Coria-Oriundo *et al.* 2021; Shokri *et al.* 2021; Sun *et al.* 2021a; Backes *et al*. 2022). Certainly, low-cost and high-activity are important characteristics for wide application of laccase in various aspects of industrial and environmental technology areas. Thus, obtaining low-cost laccase with high activity and high yield has attracted the attention of many researchers (Akpinar and Urek 2017; Chenthamarakshan *et al*. 2017).

White-rot fungi (Basidiomycetes) are of great interest because of their excellent laccase producing ability. *Ganoderma* and *Coriolopsis* spp. are white-rot fungi belonging to the family Polyporaceae. They typically grow on angiosperm wood and fallen angiosperm wood, respectively. Additionally, *Ganoderma* and *Coriolopsis* spp. are efficient lignin-degrading fungi that are able to colonize different lignocellulosic wastes as substrates. Submerged fermentation is suitable for fungi-producing laccase in an enzyme preparation factory due to the convenience of controlling fermentation conditions, while the main disadvantage of submerged fermentation is that the enzyme is diluted due to high water content. The method of solid-state fermentation (SSF) contains little or no free water, avoids the problem of enzyme dilution, and is more similar to the habitat of fungi living in the wild (Steudler and Bley 2015). The SSF is typically lignocellulosic wastes that contain rich nutrients for microorganism growth and enzyme production (Hatvani and Mécs 2001). Currently, numerous types of lignocellulosic wastes, *e.g.*, sugarcane bagasse, wheat bran, oil palm trunk, rice husks, and straw, have been used for producing enzymes under solid-state fermentation (Singh *et al*. 2013; Karp *et al*. 2015; Thakur and Gupte 2015; Postemsky *et al*. 2017; Soccol *et al*. 2017; An *et al.* 2020; Han *et al*. 2021a, 2021c). Consequently, screening of the appropriate fungal species and lignocellulosic wastes for producing laccase is advisable. In the present study, four newly isolated strains of the wild white-rot fungi were obtained from their native habitats in northern and central China and examined for their ability to produce laccase. Furthermore, the capacity of secreting laccase of fungal strains collected from Central China and Northern China was also investigated for the first time. Four kinds of lignocellulosic wastes, *Populus beijingensis*, rice straw, cottonseed hull and corncob, were used as the only source of carbon, nitrogen, and mineral elements for fungi grown. The new isolated strains had the potential to serve as the alternative laccase-producing fungi, and lay a foundation for the optimization of the fermentation conditions for obtaining low cost laccase with high activity and the industrial application of laccase.

EXPERIMENTAL

Materials

Microorganisms

Two *Ganoderma* sp. strains Han 1345 and Han 1578, and two *Coriolopsis* sp. strains Han 1211 and Han 1356 were used in the present study. Among them, Han 1345 and Han 1578 were collected in Ma'an Mountain Forest Park of Hubei Province of central China and Wulingshan National Nature Reserve of Hebei Province of northern China, respectively. Another two strains, Han 1211 and Han 1356 were collected from Langfang Normal University of Hebei Province of northern China and the Wuhan Botanical Garden of the Chinese Academy of Sciences, Hubei Province, China, respectively. All strains were isolated and purified on complete yeast medium (CYM), which was composed of glucose 20 g/L, peptone 2 g/L, yeast extract 2 g/L, $MgSO_4 \cdot 7H_2O$ 0.5 g/L, $K_2HPO_4 \cdot 3H_2O$ 1 g/L, KH_2PO_4 0.46 g/L, and agar 15 g/L. All strains were maintained in 2% (w/v) malt extract agar (MEA) plates at the Experimental Centre of Forestry in North China, Chinese Academy of Forestry.

Lignocellulosic wastes

Lignocellulosic wastes used in the present study were *Populus beijingensis*, rice straw, cottonseed hull, and corncob. Among them, *Populus beijingensis* was obtained from Langfang City, while rice straw, cottonseed hull, and corncob were kindly provided by farmers from Chengde City. To obtain lignocellulosic particles with particle size of 20- to 60-mesh, all lignocellulosic wastes were air-dried, then ground.

Methods

Culture conditions of fungi and inoculum preparation

A liquid medium contained per liter: 20 g glucose, 2 g peptone, 2 g yeast extract, 0.5 g $MgSO_4 \cdot 7H_2O$, 1 g $K_2HPO_4 \cdot 3H_2O$, and 0.46 g KH_2PO_4, was used for growing mycelium pellets of fungi. All cultures were inoculated with five mycelial plugs (5 mm diameter) taken from the periphery of a colony grown on CYM at 26 °C for 7 days. The cultures were incubated at 26 °C for 7 days on a rotary shaker at 150 rpm. Then, mycelial suspensions were prepared with modular homogenizer S10 (Ningbo Xinzhi Biotechnology Co., Ltd., Ningbo, China) to homogenize under 8,000 rpm for 3 min. These homogenized mycelial suspensions were used as the inoculum.

Solid-state fermentation for laccase production

For the production of laccase through SSF, two *Ganoderma* sp. strains Han 1345 and Han 1578, and two *Coriolopsis* sp. strains Han 1211 and Han 1356 were inoculated separately on the substrate containing single lignocellulosic waste. A total of 3 g of lignocellulosic waste was taken into a 250-mL flasks and 9 mL of deionized water was added to moisten the lignocellulosic waste and then autoclaved (121 °C, 30 min). Then, 3 mL of homogenized inoculum was added into each Erlenmeyer flask and kept for 1 to 10 days at 26 °C under constant agitation at 150 rpm.

Sampling, extraction and assay of laccase

To obtain the crude enzyme, 100 mL of acetate-sodium acetate buffer (50 mM, pH 5.5) was added to solid lignocellulosic wastes. Then, all flasks were transferred and agitated on a rotary shaker at 120 rpm for 4 h at 10 °C (An *et al.* 2021; Han *et al*. 2021a,c).

Cell free supernatants, considered as a crude laccase, were obtained by filtering the liquid containing lignocellulosic waste through a filter paper and centrifuging the culture liquid at 4 °C, 12,000 rpm for 20 min. Supernatant obtained after centrifugation was determined for the laccase.

Quantitative estimation of laccase activity was determined by observing the change of substrate in absorbance at 415 nm ($\mathcal{E}_{415} = 3.16 \times 10^4$ M^{-1} cm^{-1}). The substrate used for laccase was 2,2-azino-bis-[3-ethylbenzthiazoline-6-sulfonate] (ABTS). Laccase activity was assessed by the reaction mixture containing 1 mM ABTS and 50 mM acetate-sodium acetate buffer (pH 4.2) (Han *et al.* 2021a, 2021c) and detected by an iMark™ microplate absorbance reader (Bio-Rad, Hercules, CA, USA). One unit of laccase activity was defined as the amount of crude enzyme required to transform 1 μmol of ABTS per minute into product.

Statistical analysis

The presented data was exhibited as the mean ± standard error (SE) of three replicates (n = 3). Significant differences in laccase activity between the lignocellulosic wastes and fungi were assessed using two-way analysis of variance (ANOVA) with Tukey's honest significance difference (HSD) *post hoc* test by SPSS 22.0 (IBM Corp., version 22.0, Armonk, NY, USA).

Identification of the Ganoderma sp. *strains* and *Coriolopsis* sp. *strains*

Ganoderma sp. strains and *Coriolopsis* sp. strains were identified by amplification of the ITS rRNA gene. The mycelium was scraped with a surgical blade from the surface of the Petri dishes cultivated *Ganoderma* sp. strains and *Coriolopsis* sp. strains for 7 days. Then, the total genomic DNA was extracted using the cetyltrimethylammonium bromide (CTAB) rapid plant genome extraction kit-DN14 (Aidlab Biotechnologies Co., Ltd., Beijing, China) according to the instructions (Han *et al.* 2016, 2021b). ITS5 and ITS4 primers were used for polymerase chain reaction (PCR) amplification in a thermocycler. Then, the PCR products were sent to the Beijing Genomics Institute (Beijing, China) for sequencing. The obtained sequence was queried against ITS sequences available in GenBank, using BLAST. Finally, all new ITS sequences of Han 1211, Han 1345, Han 1356, and Han 1578 were submitted to GenBank and corresponding returned GenBank numbers were ON262337, ON262338, ON262339, and ON262340, respectively.

RESULTS AND DISCUSSION

Identification of the Selected Fungal Strain

The taxonomic status of strains Han 1211, Han 1345, Han 1356, and Han 1578 identified by ITS sequence was *Coriolopsis trogii*, *Ganoderma lingzhi*, *Coriolopsis strumosa*, and *Ganoderma applanatum*, respectively.

Statistical Analysis

As shown in Table 1, it was not difficult to find that species, lignocellulosic wastes, and the interaction of species, and lignocellulosic wastes were significantly affecting the laccase activity ($P < 0.001$) during the whole fermentation process (Table 1).

Table 1. Two-way ANOVA to Analyze Effects of Different Species, Lignocellulosic Wastes, and the Interactions of Different Species and Lignocellulosic Wastes on Laccase Activity

Incubation Period (d)	Species	Lignocellulosic Wastes	Species × Lignocellulosic Wastes
1	633.247***	970.201***	309.245***
2	1393.826***	534.477***	187.359***
3	1771.505***	34.003***	369.116***
4	421.389***	131.934***	181.812***
5	450.288***	578.698***	218.652***
6	1120.100***	667.071***	633.307***
7	748.106***	225.705***	243.628***
8	2255.378***	606.250***	941.625***
9	8447.980***	3794.090***	3572.711***
10	3222.439***	1820.813***	1499.181***
*Note: df = 3, 3, 9; ***P < 0.001. The values are the F-value of SPSS analysis.			

Effect of Different Lignocellulosic Wastes on Laccase Activity

Certain lignocellulosic wastes contain natural inducers that have the potential ability to enhance laccase productivity, diminish the cost of production, and reduce pollution in solid-state fermentation (Wang *et al*. 2019). To select lignocellulosic wastes for producing laccase by SSF, lignocellulosic wastes should be a suitable support for fungi growth. There have been few studies on laccase production of the genus *Coriolopsis* grown on lignocellulosic wastes that indicate newly isolated *Coriolopsis sanguinaria* belonging to genus of *Coriolopsis* was an excellent laccase-producing strain (An *et al*. 2021b). Hence, assessment the effect of different lignocellulosic wastes to different newly isolated fungi strains secreting laccase was a key and sustainable task.

The laccase activity of *Coriolopsis trogii* Han 1211 stimulated by cottonseed hull, rice straw, corncob, and *Populus beijingensis* at the 1st day was 28.03 ± 2.09 U/L, 8.34 ± 0.92 U/L, 1.61 ± 0.17 U/L, and 7.84 ± 0.52 U/L, respectively (Fig. 1). Clearly, the existence of cottonseed hull was conducive to accelerate the production of laccase by *C. trogii* Han 1211. Similarly, An *et al*. (2020) compared the laccase activity of different *Pleurotus ostreatus* and *Flammulina velutipes* strains on poplar wood, corncob, and cottonseed hull, and indicated that the presence of cottonseed hull was beneficial to secrete laccase *via* testing *P. ostreatus* and *F. velutipes* strains in submerged fermentation. Maximum laccase activity of *C. trogii* Han 1211 grown on cottonseed hull, rice straw, corncob, and *Populus beijingensis* was 223.03 ± 11.51 U/L at 8th day, 118.24 ± 3.48 U/L at 3rd day, 151.50 ± 3.32 U/L at 3rd day, and 75.85 ± 3.62 U/L at 8th day (Table 2). Thus, the presence of cottonseed hull was helpful for improving laccase activity of *C. trogii* Han 1211 and the existence of *Populus beijingensis* was disadvantaged for *C. trogii* Han 1211 secreting laccase. The trend of laccase activity of *C. trogii* Han 1211 on cottonseed hull was difference from that on rice straw, corncob, and *Populus beijingensis* due to the peak number of laccase activity (Fig. 1). In contrast, rice straw and corncob contributed to produce laccase in a short time, although the highest laccase activities on rice straw and corncob were not the highest of the four tested lignocellulosic wastes. Furthermore, the most cost-effective method of producing laccase using *C. Trogii* Han 1211 on the four lignocellulosic wastes was to perform the fermentation process on corn cob for 3 days.

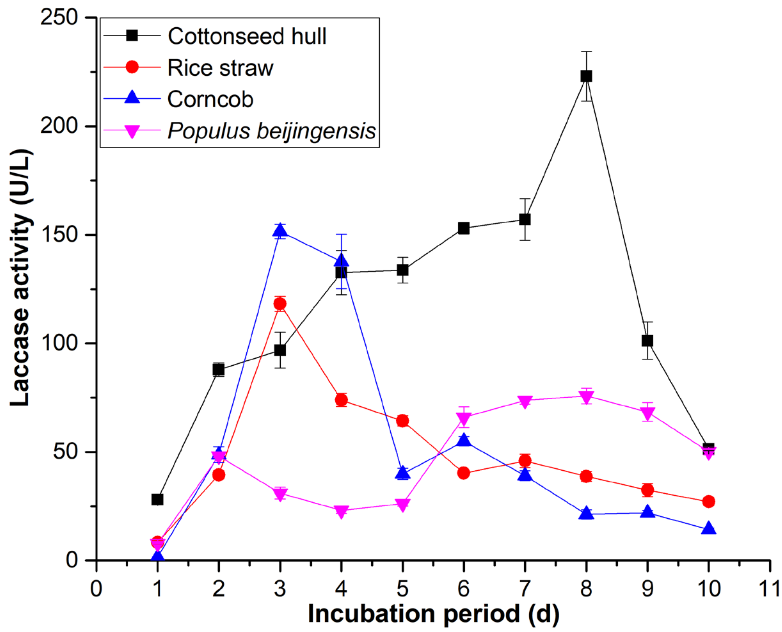

Fig. 1. Time course of laccase activity of *Coriolopsis trogii* Han 1211 grown on cottonseed hull, rice straw, corncob, and *Populus beijingensis* by solid-state fermentation

Table 2. Maximum Laccase Activity and Corresponding Occurrence Time of *Coriolopsis* sp. and *Ganoderma* sp. Strains on Different Lignocellulosic Wastes

Maximum Laccase Activity (U/L)	Lignocellulosic Wastes	Different Species	Time (Day)
223.03 ± 11.51	Cottonseed hull	*Coriolopsis trogii* Han 1211	8th
118.24 ± 3.48	Rice straw	*Coriolopsis trogii* Han 1211	3rd
151.50 ± 3.32	Corncob	*Coriolopsis trogii* Han 1211	3rd
75.85 ± 3.62	*Populus beijingensis*	*Coriolopsis trogii* Han 1211	8th
84.09 ± 7.69	Cottonseed hull	*Ganoderma lingzhi* Han 1345	5th
745.83 ± 9.00	Rice straw	*Ganoderma lingzhi* Han 1345	9th
333.74 ± 11.41	Corncob	*Ganoderma lingzhi* Han 1345	8th
119.05 ± 4.98	*Populus beijingensis*	*Ganoderma lingzhi* Han 1345	4th
203.64 ± 5.89	Cottonseed hull	*Coriolopsis strumosa* Han 1356	5th
122.51 ± 8.21	Rice straw	*Coriolopsis strumosa* Han 1356	7th
116.94 ± 2.09	Corncob	*Coriolopsis strumosa* Han 1356	6th
60.08 ± 2.60	*Populus beijingensis*	*Coriolopsis strumosa* Han 1356	3th
41.09 ± 3.60	Cottonseed hull	*Ganoderma applanatum* Han 1578	2th
53.04 ± 1.31	Rice straw	*Ganoderma applanatum* Han 1578	8th
35.16 ± 2.93	Corncob	*Ganoderma applanatum* Han 1578	6th
14.67 ± 0.63	*Populus beijingensis*	*Ganoderma applanatum* Han 1578	7th
All data are expressed as the mean value ± standard deviation (SD) (n = 3)			

At the 1st day, laccase activity of *Ganoderma lingzhi* Han 1345 on cottonseed hull, rice straw, corncob, and *Populus beijingensis* was 7.03 ± 0.63 U/L, 0 ± 0 U/L, 0.30 ± 0 U/L, and 13.66 ± 0.46 U/L, respectively (Fig. 2). Therefore, *Populus beijingensis* was helpful for improving laccase activity at the beginning of fermentation. Maximum level of laccase activity of *G. lingzhi* Han 1345 on rice straw was 745.83 ± 8.99 U/L at the 9th day, nearly 8.87-fold, 2.23-fold, and 6.26-fold higher than that on cottonseed hull (84.09 ± 7.69 U/L, at 5th day), corncob (333.74 ± 11.41 U/L, at 8th day), and *Populus beijingensis* (119.05 ± 4.98 U/L, at 4th day) (Table 2). Clearly, the presence of rice straw was conducive to improving laccase activity of *G. lingzhi* Han 1345, but it was not helpful for obtaining high laccase activity in a shorten time. At the end of solid-state fermentation, the laccase activity of *G. lingzhi* Han 1345 on rice straw and corncob was 573.34 ± 17.33 U/L and 212.48 ± 13.39 U/L, while the laccase activity detected on cottonseed hull and *Populus beijingensis* was 50.53 ± 2.18 U/L and 0 ± 0 U/L. This showed *G. lingzhi* Han 1345 could secret stable and high-level activity of laccase when it was stimulated by rice straw and corncob (Fig. 2). An *et al*. (2021a) evaluated the laccase activity of *G. lingzhi* Han 500 on cottonseed hull, *Toona sinensis*, *Sophora japonica*, *Salix babylonica*, and straw of *Oryza sativa*, and found that the maximum laccase activity was 788.73 ± 6.12 U/L measured on cottonseed hull at 8th day. However, the maximum laccase activity of *G. lingzhi* Han 1345 on cottonseed hull was merely 84.09 ± 7.69 U/L at 5th day in the present work. Actually, *G. lingzhi* Han 1345 was more preferred to produce laccase on rice straw. This also reflects the specificity of enzyme production and substrate preference of different strains belonging to one species. Of course, *G. lingzhi* Han 500 in the previous study was collected from northern China and *G. lingzhi* Han 1345 in the present study was collected from central China. The difference of habitat environment between central China and northern China may also be one of the reasons for the difference in enzyme production and substrate preference.

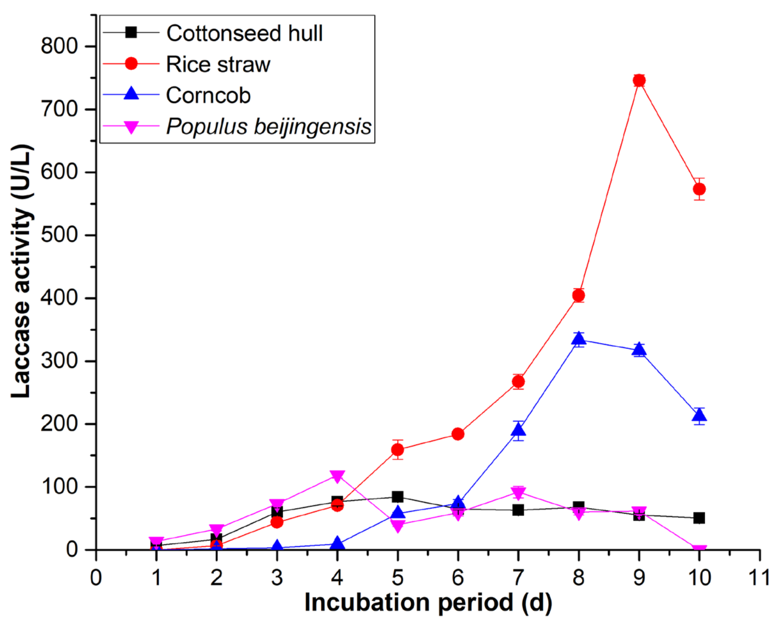

Fig. 2. Time course of laccase activity of *Ganoderma lingzhi* Han 1345 grown on cottonseed hull, rice straw, corncob, and *Populus beijingensis* by solid-state fermentation

No laccase activity of *Coriolopsis strumosa* Han 1356 was detected in all four tested lignocellulosic wastes on the first day (Fig. 3). Laccase activity on cottonseed hull, rice straw, corncob, and *Populus beijingensis* was first detected on day 2 (9.24 ± 0.76 U/L), day 3 (40.18 ± 0.76 U/L), day 3 (16.68 ± 1.77 U/L), and day 2 (42.70 ± 3.60 U/L). Based on this, it seems likely that *Populus beijingensis* was suitable for *C. strumosa* Han 1356 secreting laccase. However, maximum laccase activity detected on cottonseed hull was

203.64 ± 5.89 U/L for 5 days, nearly 1.66-fold, 1.74-fold, and 3.39-fold higher than that on rice straw (122.51 ± 8.21 U/L, 7 days), corncob (116.94 ± 2.09 U/L, 6 days), and *Populus beijingensis* (60.08 ± 2.60 U/L, 3 days), respectively (Table 2). Clearly, the presence of cottonseed hull was conducive to improve laccase activity of *C. strumosa* Han 1356 in a shorten time.

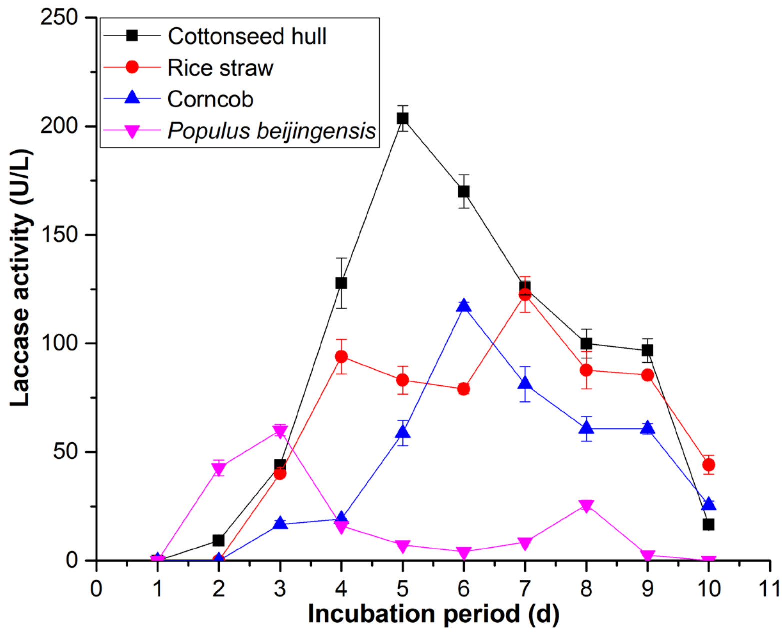

Fig. 3. Time course of laccase activity of *Coriolopsis strumosa* Han 1356 grown on cottonseed hull, rice straw, corncob, and *Populus beijingensis* by solid-state fermentation

At the 1st day, laccase activity of *Ganoderma applanatum* Han 1578 on cottonseed hull was 19.09 ± 0.76 U/L, and was undetected on rice straw, corncob, and *Populus beijingensis* (Fig. 4). Maximum laccase activity from cottonseed hull was 41.09 ± 3.60 U/L at the 2nd day, nearly 7.30-fold, 9.30-fold, and 9.30-fold on rice straw (5.63 ± 0.35 U/L), corncob (4.42 ± 0.46 U/L), and *Populus beijingensis* (4.42 ± 0.17 U/L), respectively. To some extent, cottonseed hull was truly effective in promoting rapid production of laccase from *G. applanatum* Han 1578. Maximum level of laccase activity on rice straw, corncob, and *Populus beijingensis* and was 53.04 ± 1.31 U/L at 8th day, 35.16 ± 2.93 U/L at 6th day, and 14.67 ± 0.63 U/L at 7th day (Table 2). Furthermore, the laccase activity detected on cottonseed hull, rice straw, corncob, and *Populus beijingensis* was 1.61 ± 0.17 U/L, 33.96 ± 1.49 U/L, 14.77 ± 0.60 U/L, and 4.52 ± 0.30 U/L. Thus, rice straw was conducive to improve laccase activity of *G. applanatum* Han 1578, but it was not helpful for obtaining high laccase activity in a short time.

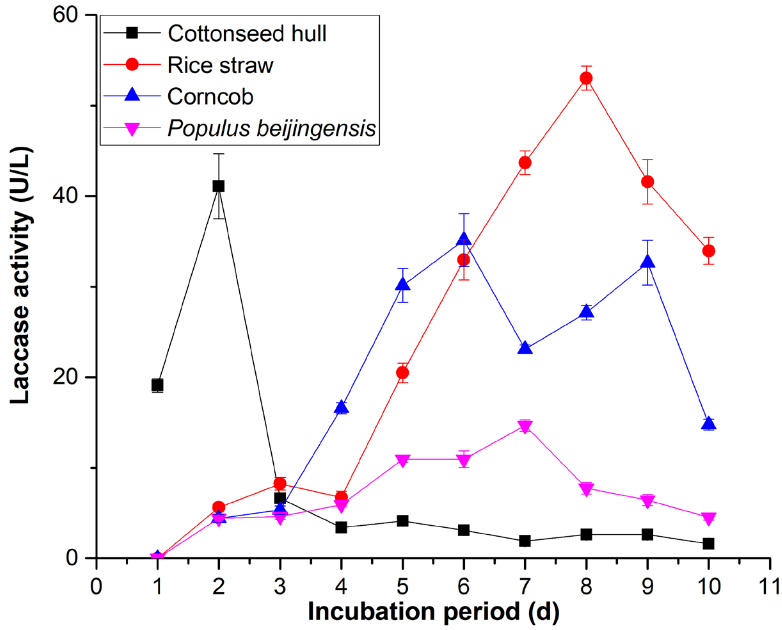

Fig. 4. Time course of laccase activity of *Ganoderma applanatum* Han 1578 grown on cottonseed hull, rice straw, corncob, and *Populus beijingensis* by solid-state fermentation

Effect of Fungi in Same or Different Genus on Laccase Activity

Previous study has shown that the laccase activity of fungi can vary greatly under identical culture conditions among both species and strains of the same species (Elisashvili *et al*. 2008). Commonly, species is a classification noun, but strains can be used to refer to fungi of the same or different species. White-rot fungi (species or strains of Basidiomycota) have the ability to degrade lignocellulosic wastes with high efficiency because they could secret various extracellular enzymes (Kuhar *et al*. 2015; Rodrigues *et al*. 2019). Recently, there has been increased research attention on screening fungal or bacterial strains suitable for obtaining massive high-activity laccase in solid-state fermentation (Al-balawi *et al*. 2017; Sun *et al*. 2017; Boruah *et al*. 2019; Wang *et al*. 2019; Yasar *et al*. 2019; Dullah *et al*. 2021; Sun *et al*. 2021b). In recent years, novel fungi with the ability to produce ligninolytic enzymes have been isolated from different natural habitats. Meanwhile, the capacity of secreting laccase from different species belonging to the same genus was compared. Huang *et al*. (2019) compared the differences in degrading crop straw from different edible fungi due to their differences in the ability of secreting lignocellulolytic enzymes. An *et al*. (2021a) analyzed the laccase activity of *Pleurotus ostreatus* CY 568 and *Ganoderma lingzhi* Han 500, and found that the laccase activity of newly isolated *G. lingzhi* Han 500 was higher than that of other *Ganoderma* strains in previous studies. Research on laccase of genus *Coriolopsis* has rarely been reported, and the only relevant studies focused on recombinant expression and heterologous expression of laccase (Songulashvili *et al*. 2016; Avelar *et al*. 2017; Glazunova *et al*. 2018). Based on this, screening new strains with the ability of producing laccase enzyme is of great value for obtaining high activity and high yield of laccase. Furthermore, the laccase production of fungal strains collected from central China and northern China was also investigated for the first time.

Laccase activity of *Coriolopsis trogii* Han 1211, *Ganoderma lingzhi* Han 1345, *Coriolopsis strumosa* Han 1356, and *Ganoderma applanatum* Han 1578 detected on cottonseed hull was 28.03 ± 2.09 U/L, 7.03 ± 2.09 U/L, 0 ± 0 U/L, and 19.09 ± 0.76 U/L at the first day (Figs. 1 through 4). Similarly, the value of laccase activity from *C. trogii* Han 1211, *G. lingzhi* Han 1345, *C. strumosa* Han 1356, and *G. applanatum* Han 1578 on rice straw was 8.34 ± 0.92 U/L, 0 ± 0 U/L, 0 ± 0 U/L, and 0 ± 0 U/L for the first day. Laccase activity of *C. trogii* Han 1211, *G. lingzhi* Han 1345, *C. strumosa* Han 1356, and *G. applanatum* Han 1578 on corncob was 1.61 ± 0.17 U/L, 0.30 ± 0 U/L, 0 ± 0 U/L, and 0 ± 0 U/L at the 1st day. Based on the above description, the capacity of secreting laccase of *C. trogii* Han 1211 was superior to other three strains in the early stages of fermentation. However, laccase activity of *C. trogii* Han 1211, *G. lingzhi* Han 1345, *C. strumosa* Han 1356, and *G. applanatum* Han 1578 on *Populus beijingensis* was 7.84 ± 0.52 U/L, 13.66 ± 0.46 U/L, 0 ± 0 U/L, and 0 ± 0 U/L. Thus, *G. lingzhi* Han 1345 was more in favor of secreting laccase than other tested strains when *Populus beijingensis* as a substrate at the beginning of fermentation. In terms of laccase activity on cottonseed hull, the maximum value from *C. trogii* Han 1211 was 223.03 ± 11.51 U/L for the 8th day, nearly 2.65-fold, 1.10-fold, and 5.43-fold higher than that from *G. lingzhi* Han 1345 (84.09 ± 7.69 U/L, for 5th day), *C. strumosa* Han 1356 (203.64 ± 5.89 U/L, for 5th day), and *G. applanatum* Han 1578 (41.09 ± 3.60 U/L, for 2th day), respectively. Furthermore, *C. trogii* Han 1211 and *C. strumosa* Han 1356 belonged to the genus of *Coriolopsis*. Hence, the presence of cottonseed hull was conducive to secret laccase of the genus of *Coriolopsis* (Figs. 1 through 4). Maximum laccase activity of *G. lingzhi* Han 1345 on rice straw was 745.83 ± 9.00 U/L at the 9th day, nearly 6.31-fold, 6.09-fold, and 14.06-fold higher than that from *C. trogii* Han 1211 (118.24 ± 3.48 U/L, 3rd day), *C. strumosa* Han 1356 (122.51 ± 8.21 U/L, 7th day), and *G. applanatum* Han 1578 (53.04 ± 1.31 U/L, 8th day). Clearly, rice straw was helpful for *G. lingzhi* Han 1345 secreting laccase. The value of laccase activity obtained from *C. trogii* Han 1211 and *C. strumosa* Han 1356 was similar, but *C. trogii* Han 1211 reached the maximum laccase activity faster. In terms of laccase activity on corncob, maximum laccase activity of *C. trogii* Han 1211, *G. lingzhi* Han 1345, *C. strumosa* Han 1356, and *G. applanatum* Han 1578 was 151.50 ± 3.32 U/L for 3rd day, 333.74 ± 11.41 U/L for 8th day, 116.94 ± 2.09 U/L for 6th day, and 35.16 ± 2.93 U/L for 6th day. The phenomenon of laccase activity was similar to that on rice straw. In other words, corncob was helpful for *G. lingzhi* Han 1345 secreting laccase, and the maximum value of laccase activity obtained from *C. trogii* Han 1211 was close to that from *C. strumosa* Han 1356. Maximum laccase activity from *C. trogii* Han 1211, *G. lingzhi* Han 1345, *C. strumosa* Han 1356, and *G. applanatum* Han 1578 on *Populus beijingensis* was 75.85 ± 3.62 U/L (8th day), 119.05 ± 4.98 U/L (4th day), 60.08 ± 2.60 U/L (3rd day), and 14.67 ± 0.63 U/L (7th day). Thus, the ability of secreting laccase of *G. lingzhi* Han 1345 was superior to other strains during fermentation on *Populus beijingensis*.

Qiu *et al*. (2014) investigated the effect of Kudzu vine root for *Funalia trogii* secreting laccase and found the laccase activity was 42.5 IU/g *via* solid-state fermentation with Kudzu vine root. The maximum value of laccase activity of *Coriolopsis rigida* was 3 × 10^5 nkat/L *via* fermentation with barley bran and proved that *C. rigida* was an excellent producer for secreting laccase (Gomez *et al*. 2005; Alcantara *et al*. 2007). Maximum laccase activity of *C. trogii* Han 751 on stalk of *Sorghum bicolor* was 799.03 ± 40.89 U/L (An *et al*. 2021b). It was noteworthy that two strains of genus *Coriolopsis*, *C. trogii* Han 1211 and *C. strumosa* Han 1356, had a preference on cottonseed hull for secreting laccase. Additionally, two strains of genus *Ganoderma*, *G. lingzhi* Han 1345 and *G. applanatum*

Han 1578, showed the consistency of secreting laccase on rice straw. An *et al*. (2016) analyzed the laccase activity from the Genus *Flammulina* (Agaricomycetes) and indicated different fungi preferred different substrate for secreting laccase. However, strains belonging to *Ganoderma* or *Coriolopsis* used in the present study showed a consistent preference for laccase secretion on lignocellulosic wastes. Maximum laccase production of *G. lingzhi* Han 500 on cottonseed hull was higher than that on *Salix babylonica*, corncob, *Populus beijingensis*, *Toona sinensis*, and straw of *Oryza sativa* (An *et al*. 2021a). While the maximum laccase of *G. lingzhi* Han 1345 and *G. applanatum* Han 1578 on rice straw was higher than that on cottonseed hull, it showed the consistency of secreting laccase on cottonseed hull, corncob, and *Populus beijingensis*. Overall, the capacity of secreting laccase of *G. lingzhi* Han 1345 was superior to that of *C. trogii* Han 1211, *C. strumosa* Han 1356, and *G. applanatum* Han 1578.

CONCLUSIONS

1. Different from previous studies, the present study found that two strains belonging to *Ganoderma* showed a consistent preference for laccase secretion in the presence of lignocellulosic wastes, and two strains belonging to *Coriolopsis* showed a consistent preference for laccase secretion with lignocellulosic wastes.
2. The existence of cottonseed hull was conducive to secret laccase of *Coriolopsis trogii* Han 1211 and *C. strumosa* Han 1356. Additionally, cottonseed hull was beneficial to improve laccase activity of *C. strumosa* Han 1356 in a short time.
3. *Ganoderma lingzhi* Han 1345 and *G. applanatum* Han 1578 were more preferred to secret laccase on rice straw.
4. The capacity of secreting laccase of *G. lingzhi* Han 1345 was superior to that of *C. trogii* Han 1211, *C. strumosa* Han 1356, and *G. applanatum* Han 1578.

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