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Isolation, screening and molecular identification of indigenous Lignocellulolytic bacteria from agricultural soils

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Abstract

Lignocellulosic crop residues are a great potential renewable biomass source, yet the efficient degradation of this material is limited because of the complex structure of cellulose and lignin. In the present study, the aim is to isolate and screen the lignocellulolytic bacteria from various soil habitats of the crop rhizosphere and organic waste. The total of 25 morphologically different bacterial isolates was obtained by serial dilution and spread plate method. The isolates were qualitatively screened on selective media, with the identification of isolates that could produce cellulolytic, ligninolytic and pectinolytic enzymes. The quantitative enzyme assays showed significant differences between the isolates. The activity of endoglucanase varied from 0.020 to 0.616 U mL⁻¹ with PBNB 6 showing the highest activity (0.616 U mL⁻¹). Lignin peroxidase activity ranged from 0.009 to 0.103 U mL⁻¹ and the highest activity was observed in PBNB 22 (0.103 U mL⁻¹). Pectinase activity ranged from 0.022 to 0.030 U mL⁻¹ and the highest activity was observed in PBNB 20 (0.030 U mL⁻¹). Based on the qualitative screening and quantitative enzyme activity, 3 bacteria isolates were selected as the most promising bacteria namely PBNB 6, PBNB 18 and PBNB 22. Among them, isolate PBN6 was identified as *Pseudomonas juntendi* by molecular approach. These results highlight soil biological system as a wide variety of lignocellulolytic bacteria and that these bacteria have the potential for biological degradation of crop residues and sustainable management of biomass.

Keywords: Lignocellulosic biomass, bacterial isolation, screening, cellulolytic bacteria, ligninolytic bacteria, pectinolytic bacteria, endoglucanase, lignin peroxidase, pectinase

Introduction

Agricultural residues are a significant part of renewable biomass and a valuable resource for sustainable waste management and nutrient recycling. The cellulose, hemicellulose and lignin are largely structurally bound to one another and form a complex, relatively resistant matrix of the plant cell-walls, the majority of the residues. The resistance of lignocellulosic materials to decomposition is largely associated with the close association of lignin with structural carbohydrates, which limits the accessibility of microorganisms and their extracellular enzymes to the polysaccharide components (Shinde *et al.*, 2022) [20].

In global level, agricultural residues are produced in significant amounts every year from cereal, pulse and oilseed production. Rice straw, wheat straw, maize stalks, sorghum straw and soybean straw are traditionally regarded as agricultural wastes, but contain significant amounts of organic matter and nutrients. In many areas, these residues are burned in the field, causing significant environmental problems such as greenhouse gas emissions and the loss of valuable plant nutrients, soil health and air pollution. Therefore, it is crucial to develop a technology to effectively utilize and decompose lignocellulosic biomass in an environmentally friendly manner. Crop residues can be sustainably disposed of using microorganisms as biological methods, which can be used to achieve nutrient recycling and soil fertility (Shinde *et al.*, 2022) [20].

The utilization of microbial degradation is an environmentally friendly method for degradation and transformation of lignocellulosic residues. The enzymes include a wide range of exoenzymes such as cellulases, hemicellulases, pectinases and ligninolytic enzymes, working either individually or in combination on various parts of the plant biomass.

While fungi have been extensively studied in lignocellulose decomposition, bacteria have also been found to break down cellulose and hemicellulose and in some instances, lignin, using a variety of enzymatic processes (Chukwuma *et al.*, 2021; Shinde *et al.*, 2022) [4, 20].

Bacteria have several attributes which make them desirable organisms for degradation of the lignocellulose, compared to fungi. They tend to have high growth rates, quick generation time and higher metabolic polymorphism, allowing them to occupy many ecological niches. The enzymes secreted by many bacterial species are extracellular and are stable under a wide range of environmental conditions and can degrade plant biomass of different structures effectively. In addition, bacteria are able to adapt quickly to varying environmental conditions and they can often act synergistically in microbial communities and increase the efficiency of degradation of complex lignocellulosic substrates (Chukwuma *et al.*, 2021) [4].

Lignocellulose-degrading bacteria can be isolated from such environments where plant materials are naturally decomposing such as agricultural soils, crop residues, compost and other organic waste-fields. These environments contain a variety of microbial communities that are able to grow on plant polymers as a carbon and energy source. Thus, isolation of indigenous microorganisms from these habitats is a good strategy for the identification of bacteria with better lignocellulolytic capabilities. Previously, cellulolytic and ligninolytic bacteria have been found in agricultural soils and several authors have pointed out that screening several bacterial isolates is essential to identify isolates with high enzyme-producing potential (Arman Shamshinov *et al.*, 2022) [19].

Selective substrates and indicator compounds may be used to screen microorganisms for their ability to degrade lignin using a qualitative method which is rapid and reliable. Carboxymethyl cellulose (CMC) Agar can be used for the detection of cellulolytic bacteria as clear zones appear after staining. The measurement of the microbes can be carried out with the help of indicator compounds like methylene blue, ABTS or tannic acid for ligninolytic activity while pectinolytic bacteria can be identified by observing substrate degradation in pectin containing media. The advantage of these methods is the ability to select potential enzyme-producing microbes from a large microbial population before detailed biochemical evaluation (Shinde *et al.*, 2022; Hsin *et al.*, 2025) [20, 8]. The qualitative screening is useful for the identification of microorganisms that produce enzymes; the quantitative enzyme assays are essential for assessing the enzymes' true degradative activity. Enzyme activities can be estimated for a precise comparison between isolates and allow selection of highly efficient bacterial strains for further research. Therefore, combining the two types of analyses, qualitative screening and quantitative enzyme analysis, offers an all-encompassing approach to the identification of microorganisms that possess more outstanding lignocellulolytic properties (Hsin *et al.*, 2025) [8].

Although recently, there has been a growing interest in microorganisms that are able to degrade lignocellulose, only a few isolates of bacteria have been reported to have efficient cellulolytic, ligninolytic and pectinolytic activities all at once. Moreover, the diversity of indigenous Lignocellulolytic bacteria in agricultural soils and organic waste sites have not been adequately explored. Hence, the

present study was conducted to isolate bacteria from various soil and decomposing organic matter environment and to assess their lignocellulolytic capability qualitatively and quantitatively. The cellulase, lignin peroxidase and pectinase production capabilities of the isolates were tested for identification of efficient bacteria producing multiple enzymes. The isolates selected offer a valuable microbial resource for future development research work in the conversion of lignocellulosic biomass and sustainable utilization of crop residues.

Materials and Methods

Experimental site

The experiment was conducted in the Department of Plant Pathology, College of Agriculture, Vasant Rao Naik Marathwada Krishi Vidyapeeth (VNMKV), Parbhani, Maharashtra, India. All experimental procedures were carried out under aseptic and controlled laboratory conditions.

Sample collection site

To isolate the bacterial strains, soil samples were collected from a variety of micro-habitats such as the rhizosphere of the crop, sewage waste and decomposing area in Parbhani region. Collection was done under aseptic conditions in pre-sterilized polythene bags with adequate labelling with location details and source information. On arrival and storage, all specimens were placed immediately into the refrigerator at 4 °C and they were immediately processed in the laboratory.

Bacteria isolation and preservation

Bacterial isolation was performed using the method of serial dilution and spread-plating with the samples collected. A 1g quantity of soil was diluted in 9 mL of sterile distilled water to yield a tenfold serial dilution series (10^{-1} to 10^{-9}). Aliquots of 0.1 mL from 10^{-6} and 10^{-7} dilutions were spread evenly on the surface of pre-solidified Nutrient Agar with the use of a sterile spreader in the laminar air flow cabinet. The culture plates were incubated at 28 ± 1 °C for 1 to 2 days. These colonies were picked and subcultured on fresh media to form pure axenic cultures and were distinguished by their contrasting morphology.

Bacterial isolates were purified and cultured in Nutrient Agar slants and maintained at 28 ± 1 °C for 24 hours. The slant cultures were stored at 4 °C as routine working stock and subcultured every 60 days for strain viability for downstream use.

Qualitative screening

Cellulolytic activity

Bacterial screening for endoglucanase activity was performed on CMC minimal agar consisting of $C_4H_{12}N_2O_6$ (5g), KH_2PO_4 (1g), $MgSO_4 \cdot 7H_2O$ (0.5g), Yeast extract (0.1g), $CaCl_2 \cdot 2H_2O$ (0.001g) supplemented with 2% (w/v) low-viscosity CMC and 1.6% (w/v) agar (pH 7 ± 0.2). Spot inoculations were made and kept at 28 ± 1 °C for 48 h. The enzymatic degradation was observed by flooding the plates with 0.1% (w/v) Congo Red and then destaining with 1 M NaCl for 20 min and resulted in the appearance of prominent clear zones around the colonies of positive isolates.

Ligninolytic activity

The isolated bacterial strains were tested for their extracellular lignin-degrading activity by using dye-decolorization and oxidation plate assays. Standard agar media were formulated and each was supplemented with three different substrates: 0.01 (w/v) methylene blue, 0.5 (w/v) tannic acid and 0.1 (w/v) ABTS for the various ligninolytic oxidoreductases. Bacterial cultures were fresh and spot-inoculated on each plate and incubated for up to 96 hours at $28 \pm 1^\circ\text{C}$. No extra chromogenic reagents were used; the production of lignin-modifying enzymes was confirmed by direct chromogenic reactions: the enzymatic degradation of methylene blue caused zones of transparent clearing; tannic acid oxidation resulted in brown rings of pigment and ABTS oxidation in zones of prominent bluish-green clearing around the functional colonies.

Pectinolytic activity

Screening of pectin-degrading bacteria was conducted on PSAM plates consisting of (g L⁻¹): citrus pectin (10.0), K₂HPO₄ (1.0), NaNO₃ (1.0), MgSO₄ · 7H₂O (0.5), KCl (1.0) and agar (15.0), with the pH adjusted to 7.0 ± 0.2 . Bacterial isolates were spot-inoculated and incubated at $28 \pm 1^\circ\text{C}$ for 48–72 h. Plates were flooded with 2% Gram's iodine solution and left for 15 min for enzymatic degradation and the presence of prominent clearing halos around the strains that produce pectinase confirmed the presence of pectinase.

Quantitative enzyme assays**Endoglucanase (CMCase) assay**

The activity of CMCase was measured by the amount of reducing sugars produced from CMC by DNS method. Briefly, 0.5 mL crude enzyme extract was incubated with 0.5 mL 1% (w/v) sodium-CMC solution (50 mM citrate buffer, pH 4.8) at 50°C for 30 min. The reaction was stopped with the DNS reagent, heated for 10 min and then cooled. The absorbance was measured at 540 nm using a blank. Glucose was used as the reference compound. One Unit (U) of activity was defined as 1 μmol of glucose released per minute per milliliter (U mL⁻¹).

$$\text{Endoglucanase Activity (U/mL)} = \frac{\Delta G \times DF}{MW \times t \times V}$$

where:

ΔG = amount of glucose released (μg)

DF = dilution factor of enzyme extract

MW = molecular weight of glucose (180.16 μg/μmol)

t = incubation time (30 min)

V = volume of enzyme used in the assay (0.5 mL)

Lignin peroxidase assay

Lignin peroxidase activity was evaluated by monitoring the decolorization of Methylene Blue at 665nm. The reaction assay contained 0.1M tartrate buffer (pH 3), 0.1 mM dye, crude enzyme and 2.0 mM H₂O₂ in a 3 mL final volume. Absorbance decay was recorded at 30°C for 5 min. Enzyme activity was computed using an extinction coefficient of 74,000 M⁻¹ cm⁻¹, where one Unit (U) represents the transformation of 1 μmol of substrate per minute (U mL⁻¹).

$$\text{LiP Activity (U mL}^{-1}\text{)} = \frac{\Delta A_{665}/\text{min} \times V_t \times 10^6}{\epsilon \times V_e \times l}$$

Where:

ΔA₆₆₅/min = Change in absorbance per minute at 665 nm

V_t = Total reaction volume (3.0 mL)

V_e = Volume of enzyme extract (0.5 mL)

ε = Extinction coefficient of Methylene Blue (74,000 M⁻¹ cm⁻¹)

l = Cuvette path length (1 cm)

10⁶ = Conversion factor from moles to micromoles

Pectinase assay

Pectinase activity was quantified using the DNS method with citrus pectin as substrate. Briefly, 0.5mL crude enzyme extract was incubated with 0.5mL 1% (w/v) pectin in 0.1 M potassium phosphate buffer (pH 7.0) at 40°C for 30 min. Reaction termination was achieved by adding 2mL DNS reagent and boiling for 10 min. Absorbance was read at 540nm after cooling. D-galacturonic acid served as the standard. One Unit (U) of enzyme activity was defined as 1 μmol of galacturonic acid liberated per minute per milliliter U/mL.

$$\text{Enzyme Activity (U/mL)} = \frac{\Delta M \times DF}{MW \times T \times V}$$

where:

ΔM = amount of reducing sugar released (μg)

DF = dilution factor of enzyme extract

MW = molecular weight of glucose (180.16 μg/μmol)

T = incubation time (30 min)

V = volume of crude enzyme extract used in the assay (0.5 mL)

Molecular characterization

Genomic DNA was extracted from the bacterial isolate PBN6 using a microbial genomic DNA isolation kit according to the manufacturer's instructions. The 16S rRNA gene was amplified by PCR using the universal bacterial primers 27F (5'-AGAGTTTGTACCTGGCTCAG-3') and 1492R (5'-GGTTACCTTGTACGACTT-3'). PCR products were purified and sequenced commercially.

The obtained sequence was compared with sequences available in the NCBI GenBank database using the BLASTn algorithm to identify closely related taxa. Homologous sequences were retrieved from GenBank and aligned using ClustalW implemented in MEGA software (version 12). A phylogenetic tree was constructed using the Neighbor-Joining (NJ) method based on the Kimura two-parameter model. The robustness of the tree topology was evaluated with 1,000 bootstrap replicates. *Stenotrophomonas maltophilia* strain IAM 12423 (NR_041577) was included as the outgroup.

Statistical Analysis

All experiments were conducted using a completely randomized design (CRD) with three independent replications. Quantitative enzyme activity data were subjected to analysis of variance (ANOVA) using the OPSTAT statistical software package and treatment means were compared at the 5% level of significance according to Gomez and Gomez (1984). When significant differences

among bacterial isolates were observed, treatment means were separated using the Critical Difference (CD) test at the 5% level of significance. The results are presented as mean $\pm$ standard error (SE) and differences among treatment means were considered statistically significant when they exceeded the corresponding CD value.

Result and discussion

Collection of samples

To check for the presence of potential bacteria that degrade lignocellulose, a total of 10 soil samples were collected from various ecological niches such as rhizosphere soils of crop, decomposing plant residue, soil near decaying wood and other organic matter rich areas of Parbhani district. Collected samples had their physical properties varied according to their origin source and were meticulously labelled and then packed in sterile bags and sent to the lab. The samples were stored at 4 °C for further processing to ensure the integrity of the microbial population and maintain the microbial viability. The microorganisms were sampled from a wide range of environments associated with organic matter, which raised the likelihood of isolating microorganisms with different types of ligninolytic, cellulolytic and pectinolytic capabilities, because they are constantly exposed to plant polymers which help to maintain the enrichment of the substrate-degrading microbial community. Likewise, Aljo James *et al.* (2024) took soil sample from forest litter and compost pits and agricultural wastes.

Isolation and preservation of samples

A total of 25 bacterial isolates were isolated from the soil samples collected by serial dilution plate method. The soil suspensions were serially diluted and plated on nutrient agar medium under aseptic conditions and then incubated to allow the growth of the bacteria. The morphological variations in terms of colour, shape, size, margin and elevation of the bacterial colonies were selected and purified by repeated streaking to obtain pure cultures. The purified isolates were then kept on nutrient agar slants and stored at 4 °C for short term storage. The isolated bacterial population was then qualitatively and quantitatively screened for ligninolytic, cellulolytic and pectinolytic enzyme activity to find efficient bacteria that can degrade the lignocellulosic wastes. In the same way, U. Jagadeesh and R. Muthuraju (2022) ^[10] isolated 173 microbial cultures (bacteria, fungi and actinobacteria) from various substrates like paddy straw, decayed wood, termite midgut, cow dung, forest soil, paddy field soil, tree bark, compost and vermicompost.

Assessment of screening

A total of 35 bacterial isolates were quantitatively screened for their lignocellulose-degrading potential using five different selective media, namely carboxymethyl cellulose (CMC), methylene blue, ABTS, tannic acid and pectin agar media. The isolates were tested for their clear zone or colour change activity in relation to the degradation of particular substrates. 8 bacterial isolates were able to produce cellulase enzyme, which was identified by their ability to breakdown CMC medium, out of the tested isolates. In the same way, 12 isolates showed a ligninolytic activity on medium containing methylene blue, indicating the ability to degrade lignin. On pectin agar, 5 bacterial isolates were found to be pectinolytic while 2 were found to produce phenolics

compound-degrading enzymes on the tannic acid medium. No activity was noted for the other isolates on each screening culture medium. The positive isolates were subjected to further evaluation and identification based on its overall enzyme producing potential. During qualitative screening, all isolates showed good cellulolytic activity which was evident by the large halo zone formed on CMC agar after Congo red staining as observed by Yadav *et al.* (2025) ^[26]. Congo red staining for determination of cellulolytic activity has been widely used in bacteria as well as fungi (Shinde *et al.* 2017, Jagtap *et al.* 2026) ^[21, 11]. Another recent study by Supreet Kaur *et al.* (2025) ^[12] aimed to isolate ligninolytic bacteria from the soil of Western Ghats and determined their potential to grow on the lignin containing substrates and decolourize dyes mimicking lignin revealed significant differences in functional diversity among the isolated bacteria. Likewise, T. Murugan *et al.* (2021) ^[17] reported the identification of pectinase producing bacteria in fruit-waste soil and that 12 of 36 isolates were pectinolytic and one isolate of *Bacillus* species was chosen as the most efficient isolate for further study.

Quantitative estimation

CMC assay

There was overall significant difference in endoglucanase activity among the bacterial isolates, which was determined quantitatively. The enzyme activity ranged from 0.020 to 0.616 U mL⁻¹. Among the isolates, PBNB 6 recorded the highest endoglucanase activity (0.616 ± 0.009 U mL⁻¹), followed by PBNB 18 (0.305 ± 0.012 U mL⁻¹). In contrast, PBNB 22, PBNB 3, PBNB 1 and PBNB 17 exhibited substantially lower activities of 0.035 ± 0.001 , 0.023 ± 0.000 , 0.021 ± 0.000 and 0.020 ± 0.000 U mL⁻¹, respectively. The critical difference (CD) value obtained at 5% probability level was 0.019 U mL⁻¹, which suggested that the differences among the isolates were significant enough for them to be differentiated as high and low active isolates. The higher activity of PBNB 6 indicates that it is possible to hydrolyze more cellulose extracellularly. The biological conversion of the lignocellulosic biomass has a significant impact by bacterial cellulases and the capacity of efficient endoglucanase production is important in depolymerization of cellulose. Therefore, PBNB 6 is a potentially valuable crop-residue degradation research model that can be further optimized and tested. Table 1 and Fig.1 shows the endoglucanase activity of six isolates. In the same way, Luo *et al.* (2023) ^[15] evaluated cellulose-degrading bacterial strains isolated from industrial waste, reporting endoglucanase (CMCase) titers between 0.531 U mL⁻¹ and 1.874 U mL⁻¹. Shrestha B. G *et al.* (2017) ^[23] also performed CMC assay where endoglucanase activity recorded was about 0.035 U mL⁻¹.

Lignin peroxidase assay

The production of lignin peroxidase was quantitatively estimated and significant differences in the amount of peroxidase produced were observed among the bacterial isolates. The activity of the lignin peroxidase was between 0.009 and 0.103 U U mL⁻¹. The highest activity was recorded in PBNB 22 (0.103 ± 0.004 U mL⁻¹), followed by PBNB 18 (0.033 ± 0.001 U mL⁻¹), PBNB 6 (0.031 ± 0.001 U mL⁻¹) and PBNB 1 (0.021 ± 0.000 U mL⁻¹). PBNB 19 and PBNB 20 exhibited comparatively low activities of 0.010 ± 0.000 and 0.009 ± 0.000 U mL⁻¹, respectively. The

differences between the isolates were large with the CD at 5% being 0.006 U mL⁻¹. Remarkably, the activity of PBNB 22 was around 3.1 times higher than that of PBNB 18, which is the second highest isolate, indicating its comparatively higher potential to produce the ligninolytic enzyme. The data shown in Table 2 and Fig.2 shows the lignin peroxidase activity of the six isolates. This was also observed by Satwika *et al.* (2024) ^[25] who observed that measurable lignin peroxidase activity was detected in only a few of the 41 isolates of bacteria they tested from soil, kitchen waste, leaf litter or cow dung.

Pectinase assay

Quantitative estimation of pectinase activity revealed differences among the three selected bacterial isolates. The enzyme activity ranged from 0.022 to 0.030 U mL⁻¹. The highest pectinase activity was recorded in PBNB 20 (0.030 ± 0.001 U mL⁻¹), followed by PBNB 22 (0.023 ± 0.000 U mL⁻¹) and PBNB 6 (0.022 ± 0.000 U mL⁻¹). Based on the CD at 5% probability level (0.002 U mL⁻¹), PBNB 20 was significantly superior to PBNB 22 and PBNB 6, whereas the difference between PBNB 22 and PBNB 6 was non-significant. Thus, PBNB 20 was identified as the most promising pectinase-producing isolate among the isolates evaluated. The data presented in Table 3 and Fig.3 reveals the pectinase activity of six isolates. Anju Mohandas *et al* (2018) ^[16] reported that, seven isolates from spoiled fruits and vegetables were screened for pectinase production using pectin agar plates and the maximum obtained enzyme activity was 2.43 (U/mL)/min by *Bacillus sonorensis*.

Molecular identification and phylogeny

The 16S rRNA gene sequence of bacterial isolate PBNB 6 showed highest sequence similarity with members of the genus *Pseudomonas* based on BLASTn analysis. Phylogenetic analysis placed isolate PBNB 6 in a well-supported clade with *Pseudomonas juntendi* strain BML3 (NR_180497), confirming its affiliation with this species (Fig. 4). The clustering was supported by a bootstrap value of 97%, indicating a close evolutionary relationship between PBN6 and *P. juntendi*. The *P. juntendi* clade was further

grouped with *Pseudomonas entomophila*, whereas *P. laurylsulfatovorans* and *P. baetica* formed a separate sister clade. *Stenotrophomonas maltophilia* strain IAM 12423 was used as an out-group to root the phylogenetic tree. Bacterial identification using 16srRNA molecular approach has been already reported in earlier studies. Cellulose degrading *Pseudomonas putida*, *Pseudomonas plecoglossicida*, *Psuedomonas* species were isolated from termite guts and identified by 16sRNA gene amplification approach (Shinde *et al.* 2017) ^[21].

Table 1: Endoglucanase activity U mL⁻¹ of bacterial isolates

Isolates	Endoglucanase activity (Mean ± SE)
PBNB 1	0.021 ± 0.000 ^d
PBNB 3	0.023 ± 0.000 ^d
PBNB 6	0.616 ± 0.009 ^a
PBNB 18	0.305 ± 0.012 ^b
PBNB 22	0.035 ± 0.001 ^c
PBNB 17	0.020 ± 0.000 ^d
SE(m)	0.006
CD at 5%	0.019

Table 2: Lignin peroxidase activity U mL⁻¹ of bacterial isolates

Isolates	Lignin peroxidase activity (Mean ± SE)
PBNB 1	0.021 ± 0.000 ^c
PBNB 6	0.031 ± 0.001 ^b
PBNB 18	0.033 ± 0.001 ^b
PBNB 20	0.009 ± 0.000 ^d
PBNB 22	0.103 ± 0.004 ^a
PBNB 19	0.010 ± 0.000 ^d
SE(m)	0.002
CD at 5%	0.006

Table 3: Pectinase activity U mL⁻¹of bacterial isolates

Isolates	Pectinase activity (Mean ± SE)
PBNB 6	0.022 ± 0.00 ^b
PBNB 22	0.023 ± 0.00 ^b
PBNB 20	0.030 ± 0.001 ^a
SE(m)	0.001
CD at 5%	0.002

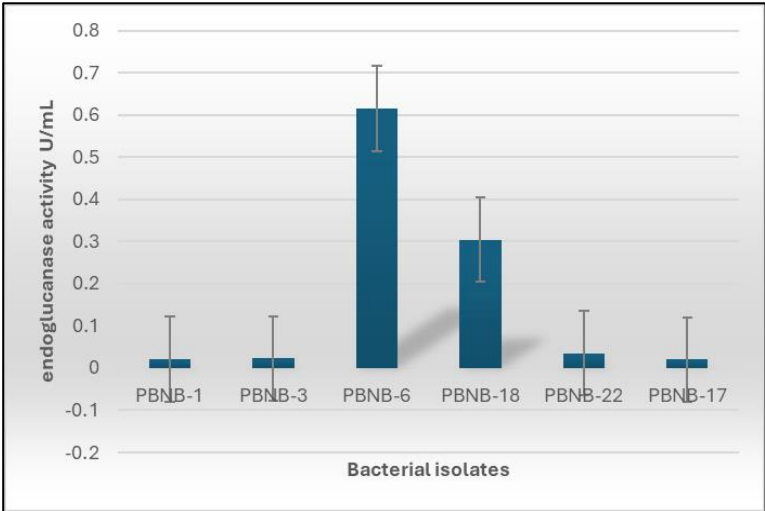


Fig 1: Endoglucanase activity of bacterial isolates

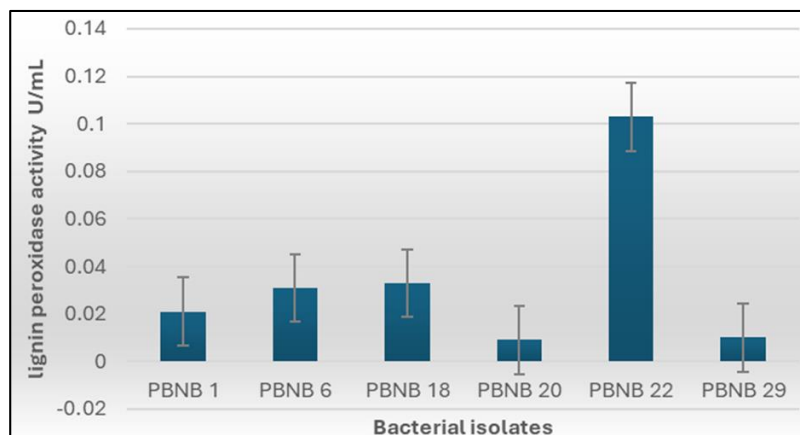


Fig 2: Lignin peroxidase activity of bacterial isolates

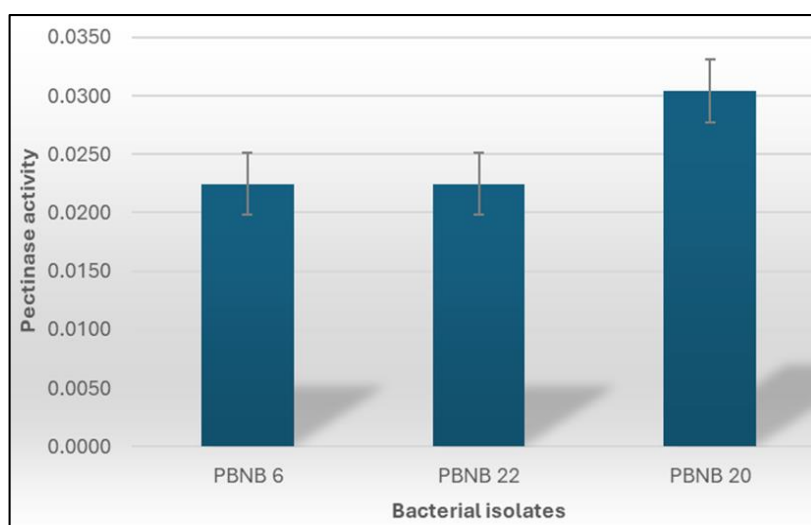


Fig 3: Pectinase activity of bacterial isolates

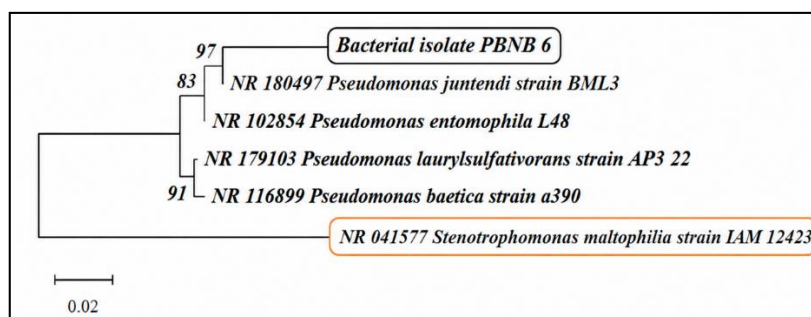


Fig 4: Neighbour-Joining phylogenetic tree of PBNB 6

Conclusion

The present investigation successfully isolated and characterized the lignocellulolytic bacteria from various soil habitats of Parbhani district successfully which showed that organic matter rich soil is a rich source of microorganisms which are efficient in degradation of crop residues. The 25 bacterial isolates were isolated and tested for their ability to produce ligninolytic, cellulolytic and pectinolytic enzymes on selective media and enzyme assays. In addition to the selection of the best possible strains, qualitative screening was used to choose several promising isolates with substrate degradation potential and quantitative enzyme assays were used to determine the most efficient strains. The superior cellulolytic potential was observed in the isolate PBNB 6 with an activity value of 0.616 ± 0.009 U mL⁻¹, while the isolate PBNB 22 showed the highest lignin peroxidase

activity (0.103 ± 0.004 U mL⁻¹) which indicated its strong lignin-degrading capability. The most efficient pectinase producer among them was the PBNB 20 which has an activity of 0.030 ± 0.001 U mL⁻¹. There were significant differences between isolates, indicating that there is a great degree of functional diversity present in bacteria that degrade lignocellulose. The study shows that these efficient bacterial isolates have a good potential in terms of their enzymatic capacity for biological degradation of crop residues namely lignocellulosic crop residues. The results obtained serve as a good basis for further screening, consortia development and optimization of the selected bacteria and evaluation for sustainable management of crop residues and environmentally friendly bioconversion of agricultural wastes.

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