

Rice straw degradation by *Pseudomonas aeruginosa* AMB-CD-1, isolated from fresh cow dung and its impact on rice plants

Kishan Kumar Raj, Anup Kumar Singh, Tapas Chowdhury, S B Gupta, Ravindra Soni*

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Abstract

Purpose Rice straw degradation in the soil is slow because of the high percentage of complex carbohydrates. Hence, farmers prefer to burn it in the agricultural field as a quicker alternative to the open burning of rice straw. Therefore, this study aims to characterize the bacteria from cow dung that can degrade rice straw.

Method In the present study, four bacterial isolates were isolated from the fresh cow-dung sample. After initial screening, one isolate, i.e. AMB-CD-1, was used to degrade rice straw for 25 days. The degraded straw samples were then analyzed through FTIR and SEM analysis. Furthermore, the degraded sample was used for growing paddy plants.

Results Out of four isolates, only the AMB-CD-1 isolate performed the best in all the screening analyses. The results concluded that the AMB-CD-1 culture inoculated straw had a low level of cellulose and hemicellulose, indicating these particles' degradation. The isolated AMB-CD-1 was identified as *Pseudomonas aeruginosa* through the Sanger sequencing technique of the 16S rRNA gene. Results also revealed that the highest germination percentage, shoot weight and shoot length were found to be significantly superior in T4 (50% Sand + 50% Compost) whereas the highest chlorophyll content was found in T7 (25% Sand + 25% Soil + 50% Compost), and T1 (100% Sand) was effective in terms of root length.

Conclusion Overall, the study concluded that *Pseudomonas aeruginosa* AMB-CD-1 might be a fast decomposer of paddy straw.

Keywords *Pseudomonas aeruginosa*, Paddy straw, Degraded Straw, Biodegradation

Introduction

Rice straw is used for various purposes like animal feed, biogas production, mushroom media and non-wood-

based paper preparation, material for mulching and packing, etc. Chemically, paddy straw is composed of cellulose (32–47%), hemicelluloses (19–27%), and lignin (5–24%) (Yang et al. 2020). The increased production, productivity, and area under the paddy cultivation led to high production of rice straw. It is produced in large quantities, reaching about 731 million tons per

✉ Ravindra Soni rs31693@gmail.com

Department of Agricultural Microbiology, College of Agriculture, Indira Gandhi Krishi Vishwavidyalaya, Raipur, India

year, with contributions from Africa, Asia, Europe, and America (Hegazy 2016). Straw is the most important by-product of agriculture, consisting primarily of dry stalks of crops. Potential microorganisms must degrade it to make it a better product for further use. Its assimilation into soils by microbial translocation is a sustainable method of increasing the soil fertility. As a result, microbial fermentation techniques can be used to manufacture various value-added products using these resources, either directly or after pretreatment (Sonwani et al. 2020; Tse et al. 2021). It decomposes slowly in the soil because it is poorly digestible and contains a lot of disease-causing lignin and silica. Therefore, farmers favor burning it in the agricultural field as a speedier alternative; unfortunately, open burning of rice straw releases harmful greenhouse gasses and creates air pollution, which is a severe hazard to human health; annually, tons of straw are burned without being employed (Sharma et al. 2020). Massive amounts of plant nutrients and organic materials are also lost as a result of the burning. In addition, the abundance of rice straw as an organic waste can be transformed into manure during the degraded strawing process. Degraded strawing is a cost-effective biological treatment to treat different types of organic waste (Tiquia and Tam 2002). It is one of the beneficial methods for immediately consuming a substantial proportion of these wastes for the production of degraded straw, which can be utilized as a source of nutrients to improve soil structure, increase organic matter, and promote plant growth. Further, being herbivores, exploration of microbial flora from cow dung for cellulase-producing bacteria (Bai et al. 2012; Islam and Roy 2018; Hong-li et al. 2015) and enzymatic activities (Vijayaraghavan et al. 2016) is well reported. Therefore, the aim and objective of this study were to isolate and characterize the bacteria from cow dung that can degrade rice straw.

Material and methods

Collection of cow dung sample

A morning fresh dung sample was collected directly from a lactating indigenous cow from the dairy farm located at Dairy Farm College of Agriculture, IGKV Raipur (India). Soon after collecting the cow dung samples, they were stored in sterilized polythene bags. A 1mL aliquot of a one-gram cow dung sample was plated on a cellulose activity agar plate (10^{-5} to 10^{-7}). All plates were incubated at 37 °C for 24–48 hrs.

Selection of cellulose degrading bacteria

For the selection of cellulose-degrading bacteria, twenty grams of cow dung were inoculated in 150 mL of nutrient broth for 48 h. One milliliter of each sample was taken for serial dilution. The diluted sample was plated in CMC media and incubated at 37 °C for 48 h. Morphologically different isolates were selected and re-streaked to obtain a pure culture. Screening isolates also did biochemical characterizations for urease, amylase, catalase, citrate utilization, and triple sugar iron (TSI) Agar test.

Antibiotic susceptibility test

Further, overnight grown bacterial cultures (100 µL) were evenly spread over nutrient agar plates having HiPer antibiotic sensitivity discs (Himedia Laboratories Pvt. Ltd., India) consisting of eight antibiotics (Streptomycin, Tetracycline, Chloramphenicol, Rifamycin, Ampicillin, Kanamycin, Gentamicin, and Nalidixic acid). These plates were kept at 37 °C in the BOD incubator for 24 h. On the next day, the inhibition zone of antibiotics obtained was measured using the Himedia scale.

Estimation of reducing sugar from rice straw

A volume of 100 mL of nutrient broth medium with 1 gram of dry rice straw was prepared into separate conical flasks and distributed into separate 200 mL conical flasks, followed by autoclaving at 121°C. The bacterial suspensions were inoculated into the conical flasks and incubated at 37 °C for 30 days, and the degraded culture filtrate was collected. The reduced sugar was analyzed using the dinitrosalicylic acid (DNS) method (Sonwani et al. 2020).

Screening for straw utilization

A 15-day long experiment was set up in a 150 mL conical flask. Carboxymethyl cellulose (CMC) and/or rice straw were used with selected isolates as a sole carbon source. All treatments were performed in triplicate. The details of the treatments are given in Table S1. All the flasks were kept at 37 °C with shaking for 15 days. A 3 mL sample was taken at a regular time interval from each treatment to record the O.D. at 600 nm.

Rice straw degraded strawing by cow dung bacteria

Degradation of rice straw was observed for 25 days with bacteria (AMB-CD-1) in a wooden box (70 x 70 x 50 cm). The bottom of the box is filled with a sand layer (5 cm) before adding a presoaked paddy straw layer (10 cm). Bacterial broth AMB-CD-1 was sprayed at regular intervals and the straw was used for degradation for 25 days in the open environment.

Nitrogen (N) content in degraded rice straw

The nitrogen (N) content of straw samples was determined by placing 0.25 g of uniformly prepared sample in a digestion tube, adding 1 g of salt mixture (K_2SO_4

and $CuSO_4 \cdot 5H_2O$ mixed in a 10:1 ratio), 10 mL of concentrated sulphuric acid (H_2SO_4), and digesting the mixture at 350°C in the digestion unit until the material became colorless. Then the nitrogen in the digested material was distilled by the automatic KEL plus system (Jackson 1973).

Phosphorus and Potassium content

In a digestion tube, one gram of straw samples was placed, and 10 mL of the tri-acid mixture (concentrated HNO_3 , $HClO_4$, and H_2SO_4 in the ratio of 9:4:1) was added. This mixture was digested at 150 °C in a digestion unit till the digested material became colorless, then allowed to cool. The digested clear material was transferred and filtered into a 100 mL volumetric flask by repeated washing with distilled water and making the volume up to the mark. This digested material is then used to estimate P and K content analysis.

Phosphorus content was determined by the vanadomolybdo-phosphoric acid yellow color complex method as described by Jackson (1973). An aliquot of 10 mL was taken and 10 mL of vanadomolybdate yellow reagent was added in a 50 mL volumetric flask and the volume was made up to the mark. After half an hour, color intensity was measured by a Spectrophotometer at 540 nm. As Chapman and Pratt (1961) described, the potassium content was determined by a flame photometer. An aliquot of 10 mL was taken in a 25 mL volumetric flask and the volume was made up to the mark. A flame photometer was then used to determine the potassium content.

Fourier Transform Infrared Spectroscopy (FTIR) analysis

FTIR was used to analyze functional groups of the untreated and treated rice straw samples (after zero-day, 10

days, and 25 days of degradation). Rice straw samples were sent for FTIR analysis at NCNR, PRSU Raipur (India). The FTIR analysis parameter was set as per our previous study by Sonwani et al. 2020.

Scanning electron microscopy (SEM)

SEM observed the change in surface structures of 25-day degraded paddy straw. The SEM procedure was done at the Nano and Electron Microscopy Laboratory at the National Institute of Technology, Raipur, Chhattisgarh, India. SEM was performed at 1000, 2000, 5000, 8000, and 10,000x magnification.

Molecular characterization

The culture was sent to Microbial Type Culture Collection, IMTECH, Chandigarh, India for its 16S rRNA gene segment sequence was submitted to NCBI GenBank for accession number phylogenetic tree is prepared with nearby genera.

Evaluation of rice straw degraded straw on rice plant growth

A 15-day long polybag experiment was conducted to evaluate the rice straw's degraded straw performance on rice plants. All treatments were performed in triplicate. The medium used for growing the rice crop was sand, soil, and straw degraded straw. The polybag is filled with the desired medium. The details of the treatments are summarised in Table 1. The experimental data (germination percentage, shoot weight, root and shoot length and chlorophyll content) obtained at 15 days after sowing (DAS) on various selected variables was analysed by the standard statistical analysis of CRD. Chlorophyll content (by ethanol method) was measured by using the ethanol extraction method as described by Lichtenthaler

(1987) and Arnon's (1949) methods. Statistical analysis took the mean value of ten plants from each treatment in each replication. A Student's t-test was performed for the parameters like nitrogen, phosphorus, and potassium content for both untreated and treated rice straw using paired samples for means, which would give a clear view for this comparison study.

Table 1 The details of the treatments used to evaluate the performance of degraded rice straw compost

S. No	Treatment	Details
1	T1	100% Sand
2	T2	100% Soil
3	T3	100% Compost
4	T4	50% Sand + 50% Compost (1:1)
5	T5	50% Soil + 50% Compost (1:1)
6	T6	25% Sand + 75% Compost (1:3)
7	T7	25% Sand + 25% Soil + 50% Compost (1:1:2)

Results and discussion

Isolation, purification and characterization of bacterial culture

The total viable count in the 1 g dung sample was 4.9×10^{10} , showing a good amount of bacterial population inhabiting the fresh cow dung sample previous reports also suggested that there was around 10^7 to 10^9 cfu/g of cow dung (Munshi et al. 2018). Four morphologically different isolates (AMB-CD1 to AMB-CD14) were selected for further study. The bacteria of interest were isolated in the laboratory on CMC agar plates from cow dung using the serial dilution method. The single discrete pure colonies of isolates were taken and again streaked on fresh CMC plates. Out of four bacteria, selected for further study, two isolates are gram-negative bacteria, and the rest of the two bacteria that appear purple are referred to as gram-positive. The biochemical properties of all the selected isolates are summarized in Table 2.

Table 2 Biochemical characterization of isolates selected from fresh cow dung

S.N	Name of isolated bacteria	Catalase test	Citrate utilization test	Urease test	Starch hydrolysis	Glucose	Lactose	Sucrose	Gas production	H2S production
1	AMB-CD-1	-	-	-	-	-	-	-	-	-
2	AMB-CD-2	-	-	-	-	-	-	-	-	-
3	AMB-CD-3	-	-	-	-	-	-	-	-	-
4	AMB-CD-4	-	-	-	-	-	-	-	-	-

Antibiotic susceptibility test

All four bacterial isolates were spread on nutrient agar medium with 9 antibiotic discs such as Streptomycin, Penicillin, Tetracycline Chloramphenicol, Rifamycin, Ampicillin, Kanamycin, Gentamycin, and Nalidixic acid. Afterwards, the plates were incubated at 37 °C for 24 hours to ensure an even zone of bacterial growth. The isolate AMB-CD-1 was shown to be resistant to penicillin, tetracycline chloramphenicol, rifamycin, ampicillin, Kanamycin, and nalidixic acid but sensitive to streptomycin and gentamycin. AMB-CD-2, AMB-CD-3, and AMB-CD-4 were resistant to Kanamycin and susceptible to Streptomycin, Penicillin, Tetracycline, Chloramphenicol, Rifamycin, Ampicillin, Gentamycin, and Nalidixic acid.

Reducing sugar estimation

Enzymatic hydrolysis of cellulose and hemicelluloses in rice straw was vital for reducing sugar production. The glucose concentration was observed for various samples using a UV double beam spectrophotometer at 540 nm. The sample with strain AMB-CD-1 had the highest concentration of 7.1 mg/mL followed by AMB-CD-2 (6.6 mg/mL). The results showed that the straw sample treated with the bacterial isolate AMB-CD-1 had more cellulose-degrading activity when compared to others.

Screening for straw degradation

Four fast-growing cultures, AMB-CD-1, AMB-CD-2, AMB-CD-3, and AMB-CD-4 were chosen for further study based on their growth on CMC agar plates. Their growth in the presence of rice straw and CMC was examined at various time intervals after incubation at 37°C. In the case of minimal broth containing rice straw,

the maximum growth is observed in AMB-CD-1 on the 3rd day of inoculation compared to all other treatments, including control. Similarly, these isolates also had maximum growth where CMC was the only carbon

source. However, an extended growth till 7 days was observed where CMC was the only carbon source (Fig 1a and b).

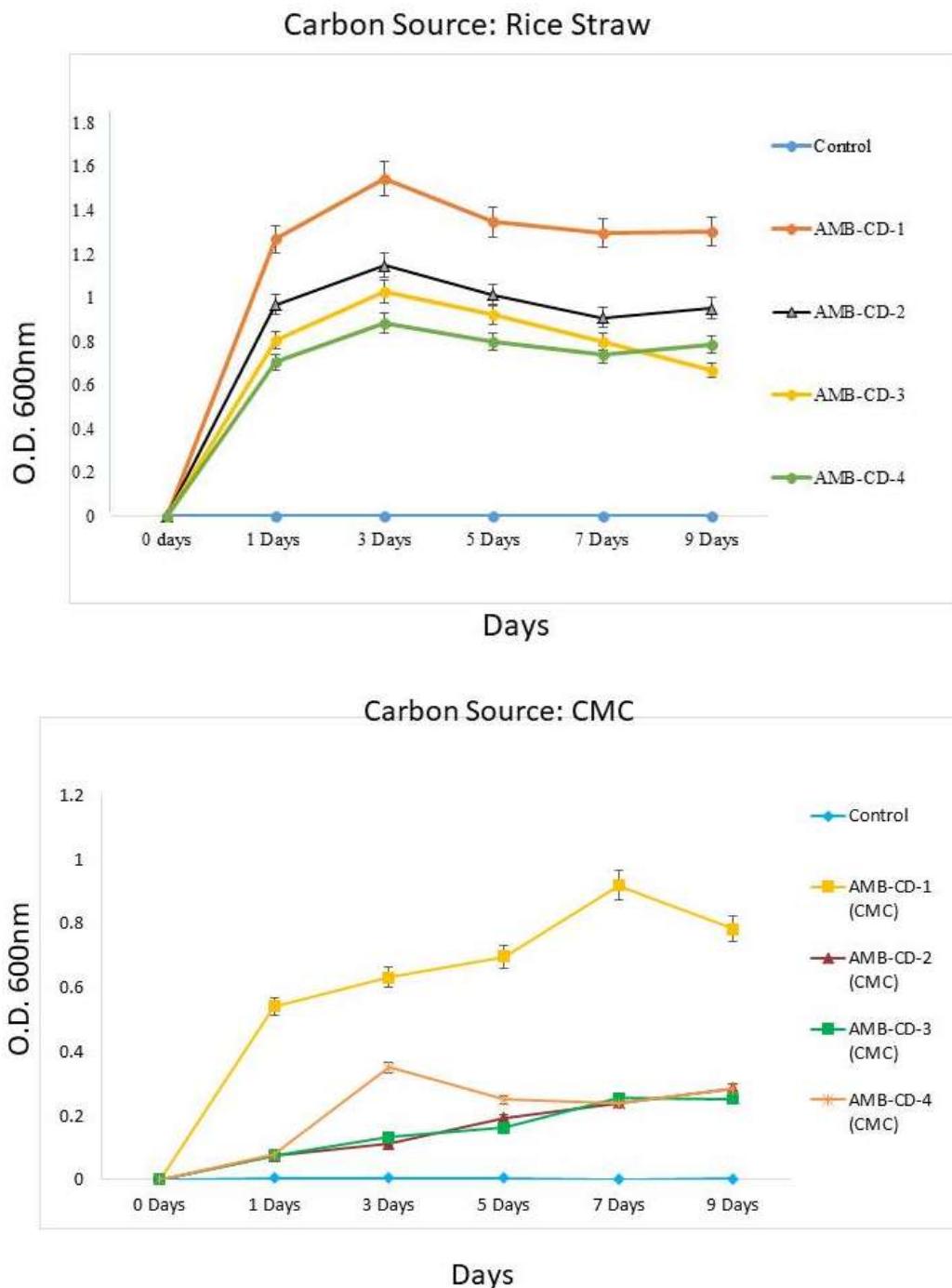


Fig. 1 Growth of cow dung isolated bacteria in presence of straw and Carboxy Methyl Cellulose

Each data represents the mean \pm SE obtained from independent experiments with 3 replicates ($p < 0.05$). Control (only broth) compared with the rest of the treatments.

Molecular characterization and phylogenetic study

The results obtained after Sanger sequencing of the isolate AMB-CD-1 revealed that the strain belonged to *Pseudomonas aeruginosa*. This species is well known for its biodegradation properties. After phylogenetic analysis with the nearest genera, the 16S rRNA gene sequence was submitted to NCBI GenBank with accession number MZ819967.

Rice straw degradation by *Pseudomonas aeruginosa* AMB-CD-1

Rice straw was degraded with the help of the isolated AMB-CD-1 culture by spraying over the rice straw at 2 days for a time of 25 days for further testing. The color of the straw changed from yellow to brown and then to a dark brown/ black color (Fig. 2). For analyzing structural changes during degraded strawing, three samples (control, 10 DAI, and 25 DAI) were sent for FTIR analysis. However, the sample after 25 DAI was also analyzed by scanning electron microscopy (SEM).

Analysis of N, P and K content in the degraded straw

Nitrogen content was estimated for the control (untreated straw) and *Pseudomonas aeruginosa* AMB-CD-1 treated paddy straw. It was observed that the nitrogen content in treated rice straw was higher (0.7%) compared to control (0.504%). Further, the phosphorus content in treated rice straw was also higher (0.22%) compared to control (0.14%). Similarly, the higher potassium content in treated rice straw was 0.244% compared to control (untreated rice straw) of 0.176% (Fig. 3).

The FTIR result of degraded rice straw

The FTIR results obtained for the rice straw without the culture (control) had the highest cellulose and lignin levels. The assignment of FTIR peaks corresponding to the functional groups of lignocellulose components revealed the characteristics of cellulose bonds. Compared to the control (3275.80 cm^{-1}), the distinctness in the crystal structure of cellulose was disrupted with a peak of 3340 cm^{-1} for 10 days of degraded straw and 3250 cm^{-1} for 25 days of composing straw were all exhibited O-H stretching. Similarly, for cellulose with C-H stretching, CH_2 bending, and out-of-phase ring stretching at different wavelengths, the peak values of 2930 , 1438 , 720 , & 650 cm^{-1} for 10-day degraded straw and 3250 , 2933 , & 720 cm^{-1} for 25 days degraded straw respectively when compared to control. Apart from cellulose, hemicellulose and lignin were also observed in this FTIR study (Table 3, Fig. 4).

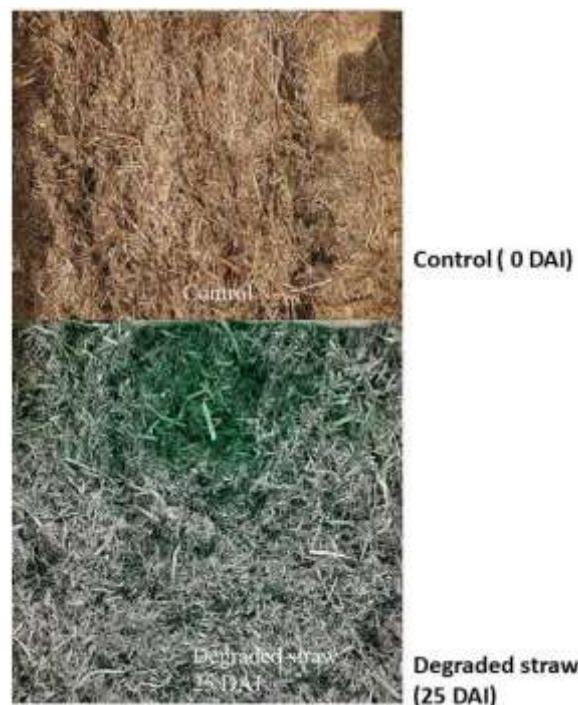


Fig. 2 Degraded strawing of rice straw for 25 days. Control rice straw (a); Straw after 25 days (b)

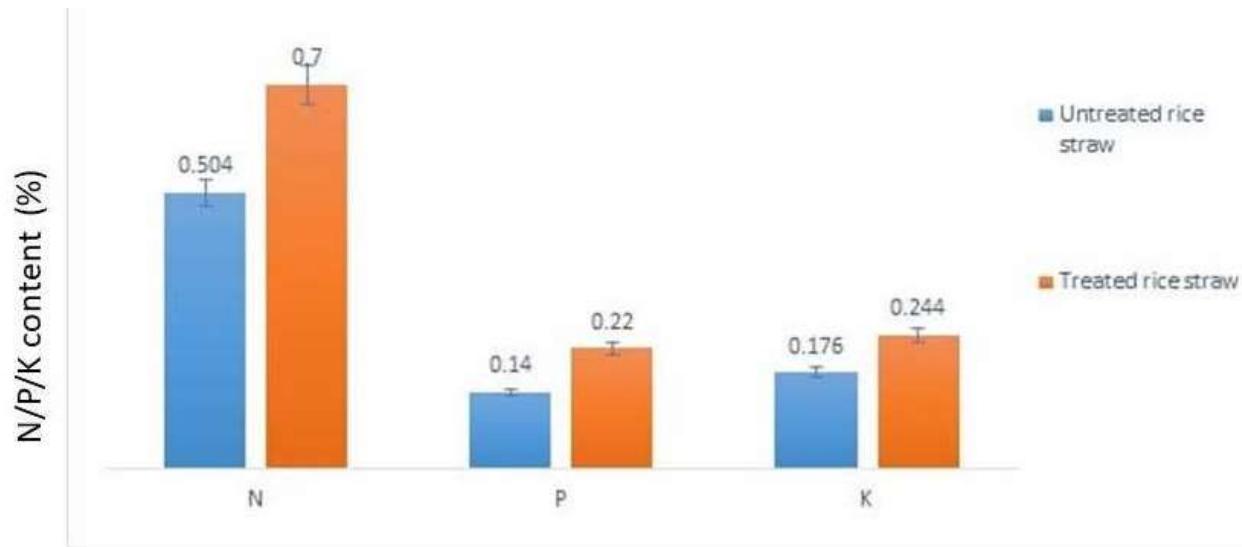


Fig. 3 Analysis of N, P and K content in the degraded straw

Each data represents the mean \pm SE obtained from independent experiments with 3 replicates. Control compared with the rest of the treatments. Student's t test was used to analyze the data with $p < 0.05$.

Scanning Electron Microscopy (SEM) of 25 days of degraded rice straw

The degraded straw kept for 25 days was observed under a scanning electron microscope under different magnifications. The distinct changes in the surface structure were visible in the tissue of the paddy straw. In general, paddy straw exhibits a rigid and highly compact structure, whereas the straw treated with *Pseudomonas aeruginosa* AMB-CD-1 bacterial culture shows the opening of the holo-cellulose fibrils due to the creation of pores of different sizes. Microfibrils were separated from the initial connected structure and were fully exposed, thus increasing paddy straw's external surface area and porosity (Fig. 5). Bacterial cells were also observed on the degraded rice straw surfaces.

Evaluation of rice straw degraded straw on rice plant

The treatments significantly affected the germination percent of rice seeds and summarized in Fig. 6 among

all the treatments, the maximum average germination percentage (100%) was observed in the treatment T₄ (50% Sand + 50% Degraded straw), which was at par by treatment T₃ (87.5%). Maximum shoot weight was observed in the treatment T₄ (0.200 g) contained 50% Sand + 50% Degraded straw followed by treatments T₇ i.e. 25% Sand + 25% Soil + 50% Degraded straw (0.150 g). For the chlorophyll content, the maximum chlorophyll was observed for the treatment T₇ (0.9 mg/g) followed by T₄ where sand and degraded straw are in equal proportion. The minimum chlorophyll content was observed in the treatment T₃ (0.3 mg/g) in which neither soil nor sand was mixed with degraded straw. Among all the treatments, the maximum average shoot length was observed in treatment T₇ (13.7 cm) consisting of 25% Sand + 25% Soil + 50% Degraded straw. The minimum shoot length was observed in treatment T₃ (7.9 cm), consisting of 100% degraded straw. The maximum root length for the treatment T₁ (9.0 cm) consisted of 100% sand which was at par with treatment T₇ (8.0 cm). The minimum root length was observed in the treatment T₃ (5.5 cm) containing 100% degraded straw (Fig. 6).

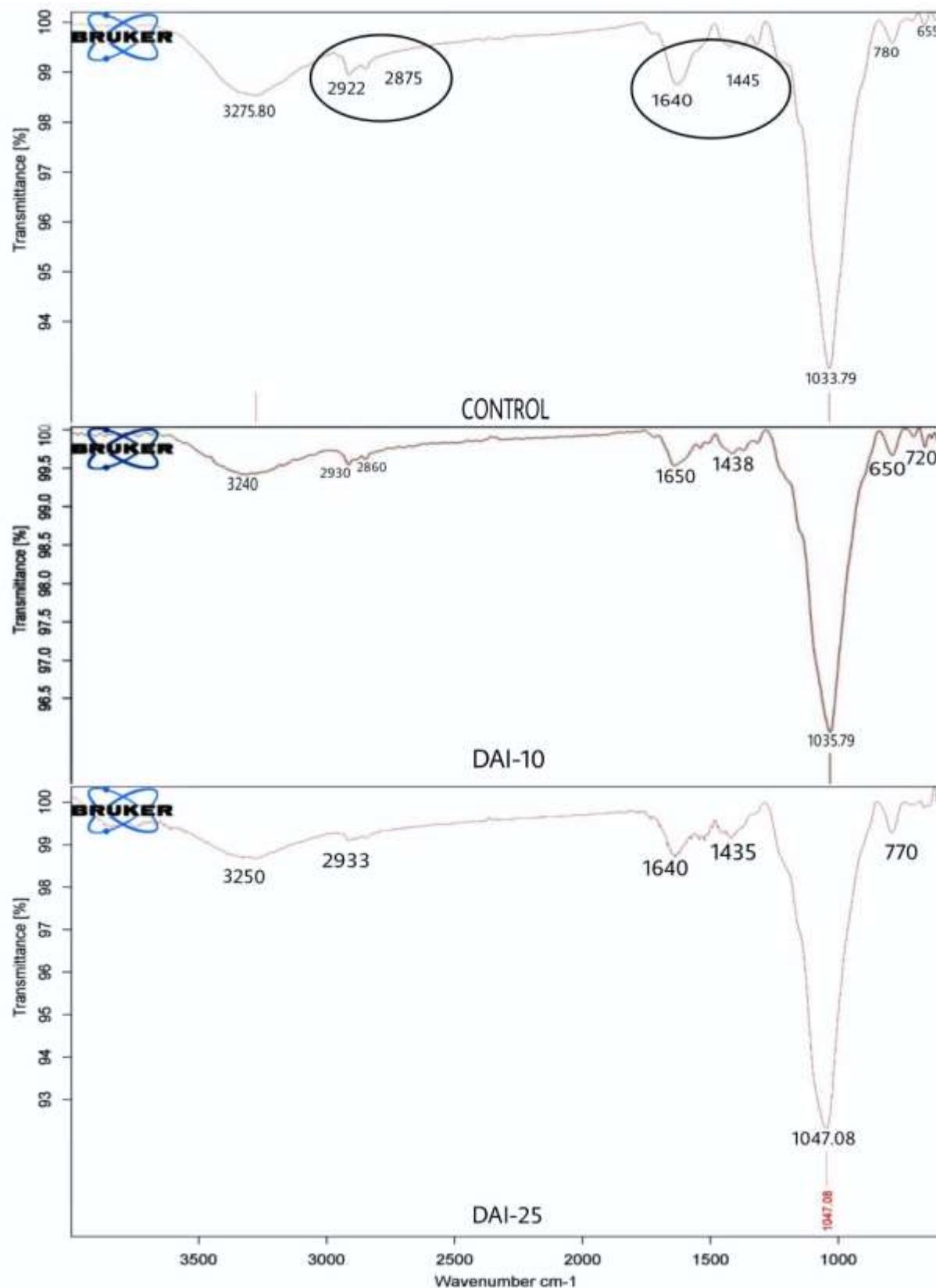


Fig. 4 The FTIR spectrums of biodegraded rice straw samples collected at different time intervals

DAI: Days after first inoculation

Table 3 Assignment and description of peaks corresponding to the functional groups of rice straw biomass components by FTIR analysis

Related biomass component	Functional group assignment	Wavelength (cm ⁻¹)		
		0 DAI	10 DAI	25 DAI
Cellulose	O-H stretching (hydrogen bond)	3275.80	3240	3250
Cellulose	C-H stretching	2922	2930	2933
Hemicellulose/lignin	C=O stretching	2875	2860	-
Lignin	C=C aromatic skeletal vibration	1640	1650	1640
Cellulose	CH ₂ bending	1445	1438	1435
deformation in	C-O stretching, aromatic C-H in plane	1033.79	1035.79	1047.08
Cellulose, Lignin				
Cellulose	Out of phase ring stretching	780	720	770
Cellulose	Out of phase ring stretching	655	650	-

DAI: days after first inoculation

Dung is a biological waste of herbivorous animals who consume food consisting of lignin, cellulose, and hemicelluloses. Dung can be considered a rich reservoir of microorganisms as it possesses a wide variety of microbes comprising over 60 different bacterial species (Bhatt and Maheshwari 2019; Gupta and Rana 2021). The fresh dung sample collected in the present study was 4.9×10^{10} , which is good enough and similar to the previously reported values. Reports suggest that there is around 10^7 to 10^9 cfu/g of cow dung (Munshi et al. 2018). Previous studies on the microbiological analysis of cow dung also reported that the population and stability of gram-negative bacteria is higher than gram-positive ones (Quraishi et al. 2018). *Pseudomonas aeruginosa* is a well-known gram-negative bacterium with brilliant biodegradation properties (Wu et al. 2018; Muriel-Millán et al. 2019). *Pseudomonas aeruginosa* However, it is, also, an opportunistic animal, and human pathogens generally infect some patients with compromised immune systems (He et al. 2004; Diggle and

Whiteley 2020). It also observed that the strain *Pseudomonas aeruginosa* AMB-CD-1 is resistant to several antibiotics like Penicillin, Tetracycline Chloramphenicol, Rifamycin, Ampicillin, Kanamycin, and Nalidixic acid, but sensitive to Streptomycin and Gentamycin. Furthermore, it is considered that the antibiotics administered for veterinary use can be excreted in dung, either unchanged or as an active metabolite (Spielmeyer 2018; Filippitzi et al. 2019; Huygens et al. 2021). However, *P. aeruginosa* presents a large genome that can develop several factors associated with antibiotic resistance involving almost all classes of antibiotics (Bassetti et al. 2018).

In the presence of rice straw as a sole carbon source, the maximum growth is observed in AMB-CD-1 indicating the use of rice cellulosic material by *P. aeruginosa* AMB-CD-1. It was also reported that *Pseudomonas* sp. can degrade cellulosic and lignocellulose materials (Yang et al. 2018; Zhang et al. 2016).

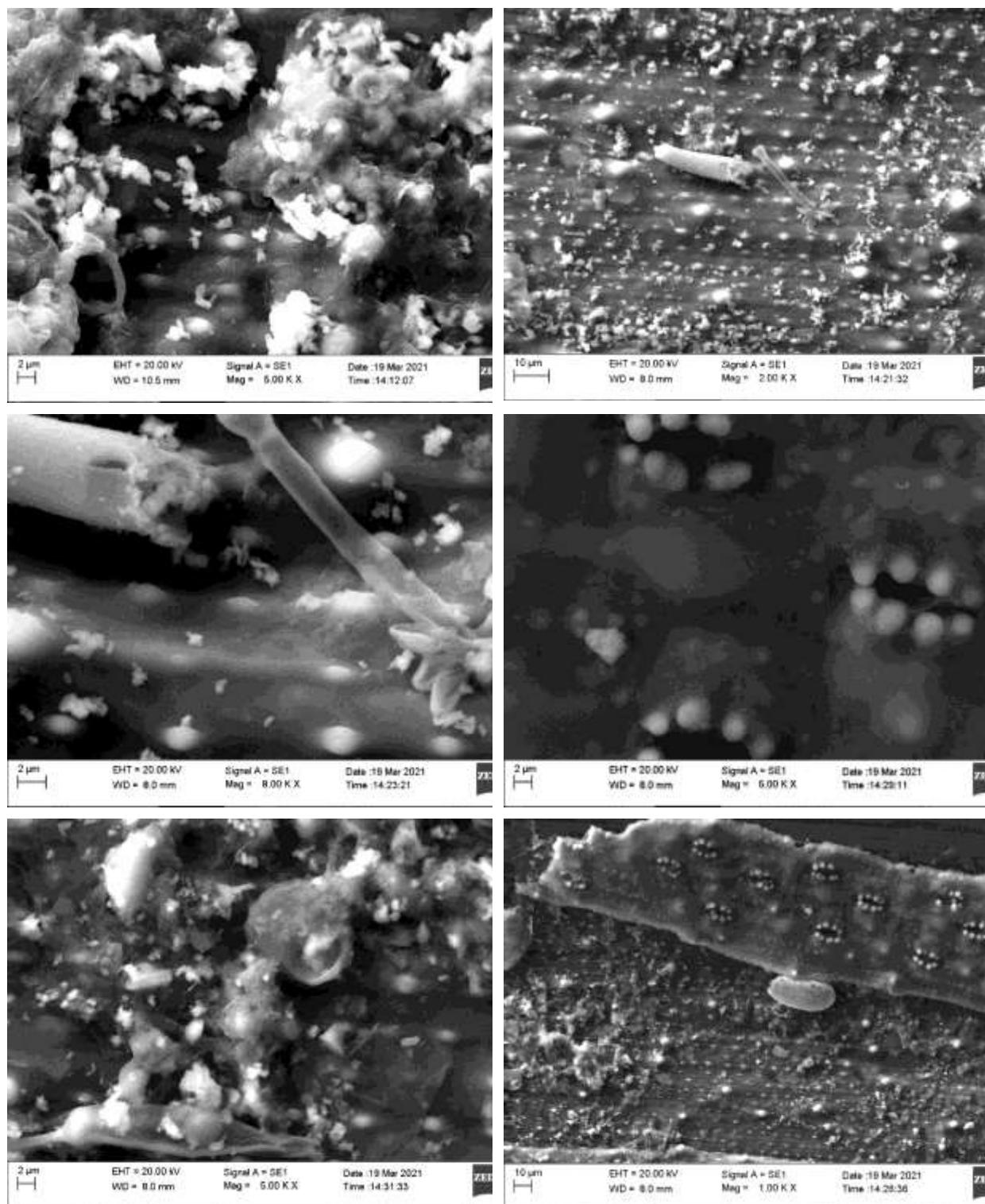


Fig. 5 Scanning Electron Microscopy (SEM) of 25 days degraded rice straw. SEM performed 1000, 2000, 5000, 8000 and 10000x magnification

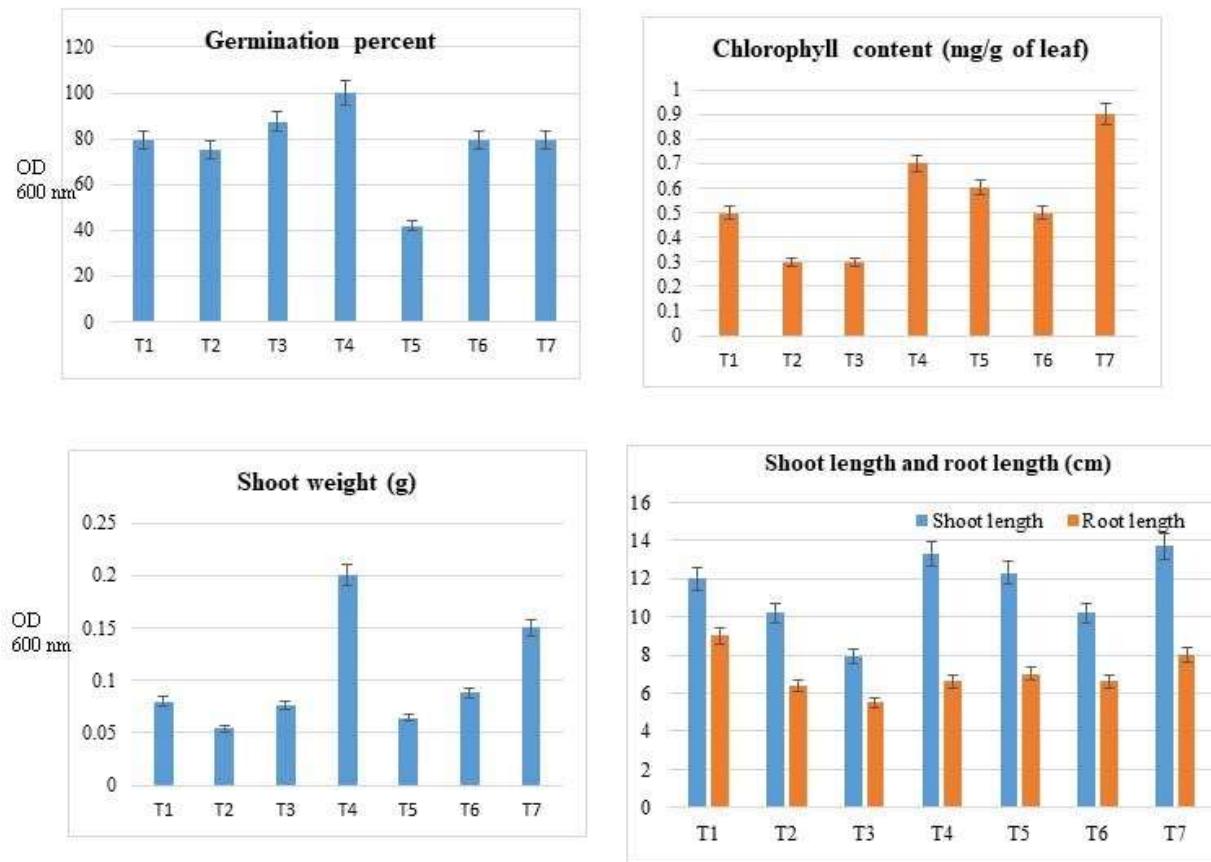


Fig. 6 Evaluation of rice straw degraded straw effect on 15 days grown rice plant

The degradation of straw was confirmed by FTIR analysis of AMB-CD-1 inoculated straw. Results showed specific peaks of cellulose around 3200, 2900, 1400, and 1000 cm^{-1} . In IR spectra, the broad band at 3000-3500 cm^{-1} is associated with O-H stretching of cellulose or hemicelluloses (Bhattacharyya et al. 2020). The peaks near 2,930 cm^{-1} are associated with the C-H stretching of methyl or methylene groups (Chen et al. 2015; Kumar et al. 2009; Tandy et al. 2010; Wang and Ren 2008). Their intensity weakened during the biodegradation process in every group, implying the rupture of the methyl or methylene portions in the lignocellulose. The peak at 1030-1050 cm^{-1} is assigned to the-C-O- group of secondary alcohols and ether functions existing in the cellulose chain backbone (Dinh Vu et al. 2017). The SEM

analysis (Fig. 5) showed that there was modification in the surface structure of rice straw after biodegradation of *P. aeruginosa* AMB-CD-1. The surface of rice straw got hollow, chapped and cracked, and this changed the structure and surface area of the degraded straw favor of enzymatic hydrolysis (Xu et al. 2007; Phutela and Sahni 2013). Furthermore, the nutrient content for degraded straw rice straw showed that N, P, and K experienced an increase in the percentage. Paddy straw contains 0.65% nitrogen, 0.20% phosphorus, and 0.30% potassium (Goyal and Sindhu 2011). Our results observed that the nitrogen content in treated straw (inoculated with AMB-CD-1) was 0.7% compared to control (untreated rice straw) of 0.504%. The phosphorus content in treated rice straw (AMB-CD-1) was 0.22% as compared to the

control (0.14%). However, Saludes et al. (2008) reported that the cattle manure degraded straw's total N, P, and K did not change significantly after 28 days of degraded strawing. Rice straw utilization by degraded strawing would provide a means to avoid air pollution from residual burning while also preventing the loss of nutrients in organic materials (Sidhu and Beri 2008). Barus (2011) reported that the application of degraded straw increased N and K uptake more than dry biomass and consequently increased the yield. Our results also revealed that the growth of 15-days-old rice plants was affected by using rice degraded straw along with sand or soil. A maximum germination percentage (100%) was observed in the treatment where 50% Sand + 50% Degraded straw were used. Maximum chlorophyll (0.9 mg/g) was observed in the treatment containing 25% Sand + 25% Soil + 50% Degraded straw, followed by treatment containing 50% Sand + 50% Degraded straw (0.7 mg/g). Badar and Qureshi (2014) also reported that use of degraded strawed rice application increases the chlorophyll content in leaves of sunflower plants. The highest germination percentage, shoot weight and shoot length were significantly superior in T4 (50% Sand + 50% Degraded straw). Similar observation was also reported by Ashrafi et al. (2019), where straw compost increases the plant growth in strawberries. The treatment consisting of 75% straw, on the other hand, was not nearly as successful; this could be because of an imbalance in the ratio of sand to compost.

Conclusion

Rice straw is a farm waste that comes in large amounts during harvest. Most farmers are still unaware of the degraded strawing of rice straw and its application for crop growth improvement. All the parameters analyzed for degraded straw indicate that it has all the properties to improve plant growth. Our results also revealed that *P. aeruginosa* AMB-CD-1 might be a potential fast degrading bio inoculant for degraded strawing of rice straw. This species of *Pseudomonas* is well documented for its degradation ability.

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Compliance with ethical standards

Conflict of interest The authors declare that there are no conflicts of interest associated with this study.

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Appendix

Table S1 Treatments of experiments set for screening of isolates for straw utilization

S. No	Treatment	Details
1.	Control	
2.	T ₁	AMB-CD 1 + Rice straw
3.	T ₂	AMB-CD 2 + Rice straw
4.	T ₃	AMB-CD 3 + Rice straw
5.	T ₄	AMB-CD 4 + Rice straw
6.	T ₅	AMB-CD 1 + CMC Broth
7.	T ₆	AMB-CD 2 + CMC Broth
8.	T ₇	AMB-CD 3 + CMC Broth
9.	T ₈	AMB-CD 4 + CMC Broth

Bacteria strain AMB-CD-1, AMB-CD-2, AMB-CD-3 and AMB-CD-4

References

- Arnon D (1949) Estimation of total chlorophyll. Plant Physiology 24:1-15
- Ashrafi R, Saem RM, Kamruzzaman M, Mamun MSAA, Begum HA (2019) Utilization of rice straw compost for strawberry and tomato production. Progress Agric 30(4):335-343. <https://doi.org/10.3329/pa.v30i4.46887>
- Badar R, Qureshi SA (2014) Degraded strawed rice husk improves the growth and biochemical parameters of sunflower plants. J Bot 427648:6. <https://doi.org/10.1155/2014/427648>
- Bai S, Kumar M, Balashanmugam P, Kumaran M, Kalaichelvan P, Mani R (2012) Cellulase production by *Bacillus subtilis* isolated from cow dung. Arch Appl Sci Res 4(1):269-279
- Barus Y (2011) Application of rice straw degraded straw with different bioactivators on the growth and yield of rice plant. J Trop Soils 17(1):25-29. <https://doi.org/10.5400/jts.15.2.95>
- Bassetti M, Vena A, Croatto A, Righi E, Guery B (2018) How to manage *Pseudomonas aeruginosa* infections. Drugs Context 7:212527. <https://doi.org/10.7573/dic.212527>
- Bhatt K, Maheshwari DK (2019) Decoding multifarious role of cow dung bacteria in mobilization of zinc fractions along with growth promotion of *C. annuum* L. Sci Rep 9: 14232. <https://doi.org/10.1038/s41598-019-50788-8>
- Bhattacharyya P, Bhaduri D, Adak T, Munda S, Satapathy BS, Dash PK, Padhy SR, Pattanayak A, Routray S, Chakraborti M, Baig MJ, Mukherjee AK, Nayak AK, Pathak H (2020) Characterization of rice straw from major cultivars for best alternative industrial uses to cutoff the menace of straw burning. Ind Crops Prod 143:111919. <https://doi.org/10.1016/j.indcrop.2019.111919>
- Chapman HD, Pratt PF (1962) Soil water and plant analysis. Univ. California Agri Div 93 (1): 68
- Chen Y, Huang J, Li Y, et al (2015) Study of the rice straw biodegradation in mixed culture of *Trichoderma viride* and *Aspergillus niger* by GC-MS and FTIR. Environ Sci Pollut Res 22: 9807-9815. <https://doi.org/10.1007/s11356-015-4149-8>
- Diggle SP, Whiteley M (2020) Microbe Profile: *Pseudomonas aeruginosa*: Opportunistic pathogen and lab rat. Microbiol 166(1):30-33. <https://doi.org/10.1099/mic.0.000860>
- Dinh Vu N, Thi Tran H, Bui ND, Duc Vu C, Viet Nguyen H (2017) Lignin and cellulose extraction from vietnam's rice straw using ultrasound-assisted alkaline treatment method. Int J Polym Sci 1-8. <https://doi.org/10.1155/2017/10636952017>
- Filippitti ME, Devreese M, Broekaert K, Rasschaert G, Daeseleire E, Meirlaen J, Dewulf J (2019) Quantitative risk model to estimate the level of antimicrobial residues that can be transferred to soil via manure, due to oral treatments of pigs. Prev Vet Med 167:90–100. <https://doi.org/10.1016/j.prevetmed.2019.03.022>
- Goyal S, SS Sindhu (2011) Degraded strawing of rice straw using different inocula and analysis of degraded straw quality. Microbiol J 1:126-138. <https://doi.org/10.3923/mj.2011.126.138>
- Gupta KK, Rana D (2021) Spectroscopic and chromatographic identification of bioprospecting bioactive compounds from cow feces: Antimicrobial and antioxidant activities evaluation of gut bacterium *Pseudomonas aeruginosa* KD155. Biotechnol Rep 29: e005772. <https://doi.org/10.1016/j.btre.2020.e00577>
- He J, Baldini RL, Déziel E, Saucier M, Zhang Q, Liberati NT, Lee D, Urbach J, Goodman HM, Rahme LG (2004) The broad host range pathogen *Pseudomonas aeruginosa* strain PA14 carries two pathogenicity islands harboring plant and animal virulence genes. Proc Natl Acad Sci 24;101(8):2530-5. <https://doi.org/10.1073/pnas.0304622101>
- Hegazy R (2016) Report: Rice straw collection. Figshare J Contribution. <https://doi.org/10.6084/m9.figshare.3206839.v1>
- Hong-li Z, Xiao Y, Dong-mei X, Lu Z, Kai-zhong T, Xing-yao X, Yun L, Xiao-jun S (2015) Isolation, identification and characterization of cellulosedegradation bacteria from fresh cow dung and fermentation biogas slurry. Res Rev J Microbiol Biotechnol 4(3):30-37
- Huygens J, Daeseleire E, Mahillon J, Van Elst D, Decrop J, Meirlaen J, Dewulf J, Heyndrickx M, Rasschaert G (2021) Presence of antibiotic residues and antibiotic resistant bacteria in cattle manure intended for fertilization of agricultural fields: A one health perspective. Antibiotics. 10:410. <https://doi.org/10.3390/antibiotics10040410>
- Islam F, Roy N (2018) Screening, purification and characterization of cellulase from cellulase producing bacteria in molasses. BMC Res Notes 11:445. <https://doi.org/10.1186/s13104-018-3558-4>
- Jackson ML (1973) Soil chemical analysis. Pub. Prentice Hall of Indian Pvt. Ltd, New Delhi
- Kumar R, Mago G, Balan V, Wyman CE (2009) Physical and chemical characterizations of corn stover and poplar solids resulting from leading pretreatment technologies. Bioresour Technol 100:3948–3962. <https://doi.org/10.1016/j.biortech.2009.01.075>
- Lichtenthaler HK (1987) Chlorophylls and carotenoids: Pigments of photosynthetic membranes. Methods in Enzymology 148:350-382
- Munshi SK, Roy J, Noor R (2018) Microbiological investigation and determination of the antimicrobial potential of cow dung samples. Stamford J Microbiol 8(1):34-37. <https://doi.org/10.3329/sjm.v8i1.42437>
- Muriel-Millán LF, Rodríguez-Mejía JL, Godoy-Lozano EE, Rivera-Gómez N, Gutierrez-Rios R-M, Morales-Guzmán D, Trejo-Hernández MR, Estradas-Romero A, Pardo-López L (2019) Functional and genomic characterization of a *Pseudomonas aeruginosa* Strain isolated from the southwestern Gulf of Mexico reveals an enhanced adaptation for long-chain alkane degradation. Front Mar Sci 6:572. <https://doi.org/10.3389/fmars.2019.00572>
- Phutela UG, Sahni N (2013) Microscopic structural changes in paddy straw pretreated with trichoderma reesei MTCC 164

- and Coriolus versicolor MTCC 138. I J Microbiol 53(2):227-31. <https://doi.org/10.1007/s12088-012-0321-8>
- Quraishi T, Kenekar A, Ranadive P, Kamath G (2018) Evaluation of performance of cow dung as microbial inoculum in industrial wastewater treatment and its environmental implications. Indian J Sci Technol 11(20):1-7. <https://doi.org/10.17485/ijst/2018/v11i20/122616>
- Saludes RB, Iwabuchi K, Miyatake F, Abe Y, Honda Y (2008) Characterization of dairy cattle manure/wallboard paper degraded straw mixture. Bioresour Technol 99:7285-7290. <https://doi.org/10.1016/j.biortech.2007.12.080>
- Sharma S, Zhang M, Anshika Gao J, Zhang H, Kota SH (2020) Effect of restricted emissions during COVID-19 on air quality in India. Sci Total Environ 728: 138878
- Sidhu BS, Beri V (2008) Rice residue management: farmer's perspective. Indian J Air Pollut Control 8:61-7
- Sonwani R, Gupta SB, Soni R (2020) Production of bioethanol from biodegraded alkali pretreated rice straw. Vegetos 33:128-134. <https://doi.org/10.1007/s42535-019-00089-2>
- Spielmeyer A (2018) Occurrence and fate of antibiotics in manure during manure treatments: A short review. Sustain Chem Pharm 9:76-86. <https://doi.org/10.1016/j.scp.2018.06.004>
- Tandy S, Healey JR, Nason MA, Williamson JC, Jones DL, Thain SC (2010) FT-IR as an alternative method for measuring chemical properties during degraded strawing. Bioresour Technol 101:5431-5436. <https://doi.org/10.1016/j.biortech.2010.02.033>
- Tiquia SM, Tam NFY (2002) Characterization and composting of poultry litter in forced-aeration piles. Process Biochem 37: 869-880
- Tse TJ, Wiens DJ, Chicilo F, Purdy SK, Reaney MJT (2021) Value-added products from ethanol fermentation-A review. Fermentation 7:267. <https://doi.org/10.3390/fermentation7040267>
- Vijayaraghavan P, Arun A, Vincent SGP, Arasu MV, Al-Dhabi NA (2016) Cow dung is a novel feedstock for fibrinolytic enzyme production from newly isolated *Bacillus* sp. IND7 and its application in *in vitro* clot lysis. Front Microbiol 7: 361. <https://doi.org/10.3389/fmicb.2016.00361>
- Wang X, Ren H (2008) Comparative study of the photo-discoloration of moso bamboo (*Phyllostachys pubescens* Mazel) and two wood species. Appl Surf Sci 254:7029-7034. <https://doi.org/10.1016/j.apsusc.2008.05.121>
- Wu T, Xu J, Xie W, Yao Z, Yang H, Sun C, Li X (2018) *Pseudomonas aeruginosa* L10: A hydrocarbon-degrading, biosurfactant-producing, and plant-growth-promoting endophytic bacterium isolated from a reed (*Phragmites australis*). Front Microbiol 9:1087. <https://doi.org/10.3389/fmicb.2018.01087>
- Xu Z, Wang Q, Jiang Z, Yang X, Ji Y (2007) Enzymatic hydrolysis of pretreated soybean straw. Biomass Bioenergy 31:162-167. <https://doi.org/10.1016/j.biombioe.06.015>
- Yang C, Yue F, Cui Y, Xu Y, Shan Y, Liu B, Zhou Y, Lü X (2018) Biodegradation of lignin by *Pseudomonas* sp. Q18 and the characterization of a novel bacterial DyP-type peroxidase. J Ind Microbiol Biotechnol 45(10):913-927. <https://doi.org/10.1007/s10295-018-2064-y>
- Yang X, Zhao J, Liang J, Zhu J (2020) Efficient and selective catalytic conversion of hemicellulose in rice straw by metal catalyst under mild conditions. Sustainability. 12(24):10601. <https://doi.org/10.3390/su122410601>
- Zhang J, Hou H, Chen G, Wang S, Zhang J (2016) The isolation and functional identification on producing cellulase of *Pseudomonas mendocina*. Bioengineered 7(5):382-391. <https://doi.org/10.1080/21655979.2016.1227143>