doi:10.14720/aas.2024.120.2.13447 Original research article / izvirni znanstveni članek

Screening and identification of IAA-capable and cellulose-degrading bacteria with the potential for plant growth-promoting traits

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Received April 19, 2023; accepted May 06, 2024.
Delo je prispelo 19. aprila 2023, sprejeto 6. maja 2024.

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Abstract: Strains with both straw degradation and plant growth promotion ability were selected from the cultivated soil in Bac Kan, Vietnam to solve the problems of poor soil microbial diversity status, weak corrosion promotion effect, and poor crop growth caused by fungal rot diseases. Among seventeen bacteria isolated, strain NR1 presented the highest value for cellulase enzyme activity (Hydrolysis index = 24.8 mm), and IAA production (20.15 mg l^{-1}), and was identified as *Bacillus amyloliquefaciens* Priest et al., 1987. Inoculation with NR1 significantly increased the rot promotion rate of straw under liquid fermentation by 54.71 % compared with the control and increased the root length and average diameter, and SPAD value of maize under soil culture by 18.3 %, 22.0 %, and 5.24 % respectively ($p < 0.05$). In addition, fertilizing 8 or 9 tons of NR1-degraded compost fertilizer per hectare had the best effect on the growth, development, and productivity of the L14 peanut variety. These results suggest strain NR1 could be used to produce multi-functional humus, accelerate the decomposition of straw in the cultivated soil, and promote crop growth.

Key words: *Bacillus* sp., cellulose, organic matter, peanut plant, PGPR

Iskanje in določanje bakterij, ki so sposobne razgraditi celulozo s pomočjo IAA kot potencialnih pospeševalcev rasti rastlin

Izvleček: Sevi bakterij sposobni razgradnje slame in sposobnostjo pospeševanja rasti rastlin so bili izolirani iz kmetijskih tal na območju Bac Kan v Vietnamu z namenom razrešiti problem majhne mikrobne raznolikosti tal, šibke sposobnosti razgradnje in slabe rasti poljščin, ki jo povzročajo glive, povzročiteljice gnilobe korenin. Med sedemnajstimi izoliranimi sevi je sev NR1 pokazal največjo vrednost aktivnosti celulaze (Indeks hidrolize = 24,8 mm), in tvorbe IAA (20,15 mg l^{-1}). Sev je bil določen kot vrsta *Bacillus amyloliquefaciens* Priest et al., 1987. Inokulacija s sevom NR1 je značilno povečala rast korenin v slami pri tekočinski fermentaciji za 54,71 % v primerjavi s kontrolo in povečala dolžino, poprečni premer korenin in SPAD vrednost pri koruzi pri gojenju v tleh za 18,3 %, 22,0 % in 5,24 % ($p < 0.05$). Dodatno je imelo gnojenje z 8 ali 9 t ha^{-1} od NR1-razgrajenega komposta najboljši učinek na rast, razvoj in produktivnost L14 sorte graha. Rezultati nakazujejo, da bi sev NR1 lahko uporabili za pripravo multifunkcionalnega humusa pri pospeševanju razgradnje slame v obdelovalnih tleh in s tem pospešili rast gojenih rastlin.

Ključne besede: *Bacillus* sp., celuloza, organska snov v tleh, arašidi, PGPR

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1 INTRODUCTION

Soil organic matter (SOM) decomposed from plant or animal biomass is returned to the soil. In soil, the SOM is an important source of nutrients for plants, especially in sloping soils. It was reported that the turnover of SOM was strongly affected by several factors such as moisture, temperature, clay content, soil porosity, soil cover, and the structure of the soil microbial communities (Don et al., 2017). So, the SOM accumulation in the soil depends on the biomass of the crop when it is returned to the soil and the soil microbial communities responsible for the organic matter decomposition. In addition, the shift of structures and activities of the microbial community in soil has been shown important roles in SOM accumulation due to their ability to secrete different types of enzymes in soil, which are involved in C cycling in soil (Sardans et al., 2008). Hence, the reaction of microbial communities to plant inputs plays significant implications for nutrient cycling and ecosystem functioning.

Moreover, the significant role of plant inputs to the soil microbial processes relies on C availability, the main factor inhibiting microbial growth and activity (Fierer et al., 2009). Notably, the main component of plant biomass is cellulose, which was the dominant waste material from the agricultural industry in the form of stalks, stems, and husks (Shankar et al., 2011). Using these agricultural byproducts as a source to produce biofertilizers is attracting scientific interest because it both reduces waste and utilizes it to make compost to provide nutrition for crops. Furthermore, the application of microbiological technology, especially applying cellulase-producing bacteria, to compost agricultural byproducts is an emerging solution for sustainable agriculture. For example, previous studies have demonstrated that soil added cellulose presented a strong stimulation of cellulose-degrading enzymes (Fontaine et al. 2004) and lignin-degrading enzymes (Talbot & Treseder 2012). These microorganisms are all available in the wild and belong to the group of mycelial fungi, bacteria, actinobacteria, and yeast (Fontaine et al., 2004). Therefore, there has been great interest in screening microorganisms with strong cellulose-degrading capabilities from soil that could be applied in compositing agroforestry byproducts into compost, which reclaimed fertility for the soil.

It was reported that plant-stimulating bacteria produce plant hormones that directly stimulate seedling growth (Do et al., 2023). Therefore, if the straw-degrading bacteria screened from the soil have both plant-promoting functions, then their potential application will be broader. To meet the multifaceted needs faced in agricultural production, more and more researchers are trying to breed strains with multiple functions. For example, Luo et al. (2018) screened multi-functional strains that degraded cellulose, starch, protein, and oil from forest soil, which played a significant role in improving soil fertility and improving crop quality on agricultural farmland. From the above, when the straw is returned to the soil field, the straw-degrading bacteria with the ability to produce IAA is applied as the core of the saprophytic agent or can solve the two major problems faced by the direct return of straw to the field.

It was reported that perennial cropping systems present extensive root networks and high allocation of belowground C, which may enhance the plant–microbial linkages. For example, a significant source of C inputs to soils from perennial root systems and a change in microbial community composition were observed during grassland restoration (Bach et al., 2010) or during crop cultivation (Dodor & Tabatabai, 2003). Especially, a similar observation was reported in annual agroecosystems that applied organic residues, cover crops (Bandick & Dick, 1999), or rotated diverse crops (Dodor & Tabatabai, 2003). These suggest the root rhizosphere is a source of microorganisms that could be exploited to support the development of sustainable agriculture.

Thus, the study was conducted to screen cellulose-degrading bacteria from soil samples grown in perennial and annual crops; and also to investigate their abilities in composting agricultural byproducts, and finally study their effects on the growth of maize.

2 MATERIAL AND METHODS

2.1 ISOLATION AND SCREENING OF CELLULOSE-DEGRADING BACTERIA FROM RHIZOSPHERE

A total of 15 soil samples were collected in cultivated fields in Na Ri district, Bac Kan, Vietnam in June 2021. In each sample plot, three sub-plots were randomly selected to collect five soil cores (2.5 cm diameter × 15 cm in length) at depths of 0–15 cm. Then these five soil cores were homogenized and bulked into one composite sample and kept in a ziplock bag. The soil samples were stored on ice until they could be transferred to the laboratory refrigerator.

Mass 1g of rice soil sample and dilute with 100 ml of sterile distilled water, shake for 15 minutes, then pipetted 20 µl and spread it on a carboxymethyl cellulose (CMC) medium (Ulrich et al., 2008). The inoculated plates were incubated at 30 °C for 2-3 days. After incubation, each colony was transferred to a new plate. Then the medium plate was stained with Congo Red solution (1 g l^{-1}) for 15 min and finally washed with 1M NaCl saline. Bacterial

isolates that hydrolyzed CMC would produce a colorless zone around the colony (halo). The hydrolysis index (HI) was calculated as the following formula:

Hydrolysis index (HI) = D – d

In which D: Halo Diameter (mm); d: Colony Diameter (mm).

CMC medium included 1 g $(NH_4)_2SO_4$, 1 g K_2HPO_4, 0.5 g $MgSO_4.7H_2O$, 0.001 g NaCl, 10 g CMC, 15 g agar, water was added to a volume of 1 liter, and adjusted pH 7.

2.2 CHARACTERIZE THE ISOLATED STRAINS

2.2.1 Cellulase enzyme activity assay

Inoculate the cultures in a liquid medium with corn stover powder as the only carbon source. The culture was incubated in a liquid flask at 37°C for 60 h, and the fermentation broth was centrifugated at 4 °C, 5000 rpm for 10 min, and the collected supernatant is crude enzyme solution. Cellulase enzyme activity was measured by determining the reducing sugar content in enzyme solutions by DNS (3,5-dinitrosalicylic acid) method (Chen et al., 2014). One unit (U) of enzyme activity was defined as the amount of enzyme equivalent to 1 μmol of reducing sugars released per minute in a 1 ml enzyme solution at a temperature of 50 °C and a pH of 4.8.

2.2.2 IAA production

Inoculation of cellulose-degrading bacteria in Luria-Bertani (LB; 10 g l^{-1} Peptone, 5 g l^{-1} Yeast Extract, 5 g l^{-1} NaCl) liquid medium containing L-tryptophan (100 mg l^{-1}). The inoculated media were incubated at 30 °C for 1 day on the shaker at 180 rpm, then centrifuged for 10 min at 5000 rpm. Then 2 ml supernatant was mixed with an equal volume of Salkowski colorimetric solution, and kept at room temperature for 30 min, and the IAA content was calculated based on spectral absorbance measurements of the standard curve at 530 nm (Liu et al., 2017).

2.2.3 Molecular identification of selected cellulose-degrading bacteria

The total DNA of selected microorganisms was extracted using a Rapid Bacteria Genomic DNA Isolation Kit (Biobasic, Canada) as per the kit instructions. The PCR amplification of 16S rDNA was done with the extracted DNA by using the universal primers 27 F (5′-AGA GTT TGA TCC TGG CTC AG-3′), and 1492 R (5′-TAC GGT TAC CTT GTT ACG ACT T-3′). The amplification was done in a GeneAmp PCR System 2700 thermocycler (Applied Biosystems, CA, USA) using the following program: 95 ˚C for 5 min; 30 cycles at 95 ˚C for 30 s, 55 ˚C for 30 s, and 72 ˚C for 90 s; and 72 ˚C for 7 min. The fragment of 16S rDNA sequences (1.5 kb) was obtained and purified by using the QIAquick PCR Purification Kit (Qiagen, USA). The purified 16S rDNA fragment was sequenced by First Base Company (Singapore). The obtained sequence was blasted on NCBI to identify the species.

2.3 EVALUATION OF THE MAIZE GROWTH PROMOTION OF BACTERIA UNDER GREENHOUSE CONDITION

The experiment was carried out in a greenhouse belonging to the Central Institute for Natural Resources and Environmental Studies, Vietnam National University Hanoi, Vietnam.

The soil of the tillage layer in the field at Na Ri, Bac Kan, Vietnam was collected. Soil samples have removed the gravel and weed dead branches, and mixed well through a 5 mm pore size sieve. The soil properties were organic matter 13.01 g kg^{-1}, total nitrogen 0.126 g kg^{-1}, alkaline nitrogen 81.03 mg kg^{-1}, available phosphorus 16.1 mg kg^{-1}, and available potassium 101.2 mg kg^{-1}. The soil was used to fill the pots.

The maize seeds (VN595 hybrid variety) were surface sterilized with 0.1 % $HgCl_2$ for 10 min and rinsed 5 times with sterile distilled water. The sterilized seeds were coated with bacteria by soaking in the bacterial solution (1×10^8 CFU ml^{-1}) for 1 hour. The bacterized seeds were sown 5 seeds per pot, and the soil moisture content was adjusted to 60 % of the maximum water-holding capacity in the field. Two treatments have been set up, each treatment was repeated 5 times. Every two weeks, 10 ml of bacterial solution (1×10^8 CFU ml^{-1}) was applied at the base of corn seedlings. For control treatment (CK), sterile water was used instead of the bacterial solution. Soil fertilization was performed as the recommendation for maize crops according to QCVN 01-56:2011/BNNPTNT.

The plant growth parameters (the root length, surface area, root tip number, plant height, SPAD value, and plant fresh mass) were measured after 49 days.

Steel tape measure and TYS- were selected for maize plant height and SPAD value, respectively-Type A chlorophyll analyzer determination; Aboveground fresh mass of the plant was measured on a one-percent scale (Lu et al., 2019). Maize root length, diameter, and surface area

with a root scanner (LA1600 + scanner, Canada) obtained images of the roots of individual plants for assays (Liu et al., 2017).

2.4 EVALUATION OF THE CELLULOSE DEGRADATION PROMOTION OF BACTERIA UNDER *IN VITRO* CONDITION

Weighted and crushed 5 g of sieved wheat straw powder in a 250 ml Erlenmeyer flask. Then added 30 ml of water, 2 g of sodium nitrate, and 2 ml of bacterial solution (1×10^8 CFU ml^{-1}) into the flash. After that, the inoculated mixtures were incubated on a constant temperature shaker at 28 °C, 120 rpm. After 15 days of incubation, the culture was centrifugated (5 000 rpm for 10 min) to remove the supernatant and washed the pellets with distilled water three times. The pellet was dried to constant mass at 80 °C. For the control experiment, replaced the bacterial solution with sterile water, other steps are consistent. Each treatment was done in triplicates. The decomposition rate of straw was determined by mass loss (%), which was calculated by the formula:

$$\text{Mass loss (\%)} = [(M - M_1)/M] \times 100$$

Where M and M1 are the initial and final mass, respectively.

2.5 EVALUATION OF SELECTED CELLULOSE-DEGRADING BACTERIA IN COMPOSTING AGRICULTURAL BYPRODUCTS UNDER GREENHOUSE CONDITIONS

By-product waste materials include 200 kg of straw, waste after mushroom cultivation; 120 kg of water hyacinth; and 80 kg of corn stalks, beans, and peanuts.

The selected bacteria were cultured in a mixture of rice bran and cornstarch (3 : 1 ratio) supplemented with 50 ml of sterile distilled water for 1 kg. The mixtures were incubated at laboratory temperature (28 ± 2 °C) and after 7 days counted the number of microbial cells. The results were: 5.21×10^8 CFU g^{-1}, in accordance with the standard of microbial production (> 10^8 CFU g^{-1}).

The composting experiment consisted of 2 formulas (Table 1) and was carried out on a cement base. A mixture of 200 kg of annealing material + 0.4 kg of lime and 0.5 kg of phosphate compounds was prepared and mixed well. The mixture was incubated for 7 days, then mixed well with the bacterial seed, stacked in 70 cm high piles, and covered with plastic. The incubation period was 30 days.

After 30 days of incubation assessed the total protein content (N %) according to TCVN6498:1999, the total P content (P_2O_5 %) according to TCVN 8940:2011; total potassium (K_2O %) according to TCVN 8660:2011 and cellulose content of the composting formula with the bacterial inoculation compared to controls (no bacterial inoculation) to assess the effectiveness of the microbial mixture.

2.6 EVALUATION OF THE PEANUT GROWTH PROMOTION OF NR1 STRAIN-PRODUCED COMPOST UNDER FIELD CONDITION

The experiments were carried out in Na Ri, Bac Kan, Vietnam. Different combinations were designed to evaluate the impact of compost on the growth and productivity of the L14 peanut variety, as in Table 2. The L14 peanut variety was cultivated on the field in spring 2021 (12/1–20/5/2021) with a planting density was 33 seedlings m^{-2}. The experiments were designed in completely randomized blocks (10 m^2) with 3 repetitions.

At harvest, the growth, development, and productivity of peanuts were collected according to the national technical regulation on testing for the value of cultivation and use of groundnut varieties (QCVN 01-57:2011/BNNPTNT).

Soil samples taken on the 0–20 cm depth before and after the experiment were dried in the air and analyzed the following indicators: pH_{KCl} by pH meter method, organic carbon content (OC) according to TCVN 8941:2011; total nitrogen by TCVN 6498:1999; total phosphorus (P_2O_5) according to TCVN 8940:2011; total potassium (K_2O) according to TCVN 8660:2011.

2.7 DATA ANALYSIS

All experiments were repeated three times the re-

Table 1: The formula for experimenting with composting

Formula	Amount of byproduct mixture (kg)	Bacterial addition	Seed inoculation rate (%)
I (Control)	200	No	0
II	200	Yes	5

Table 2: The formula for experimenting with composting

Experimental formulas	Amount of fertilizer for 1 hectare
CT1 (control) = Background	30 kg N + 60 kg P_2O_5 + 60 kg K_2O + 400 kg lime
CT2	7 tons of composting byproduct + Background
CT3	8 tons of composting byproducts + Background
CT4	9 tons of composting byproducts + Background

sults were presented as mean values ± SD. Data were statistically analyzed using Excel 2010 and SPSS 13.0 software, and the least significant difference (LSD) test was used for multiple comparisons ($p < 0.05$).

3 RESULTS AND DISCUSSION

3.1 ISOLATION AND IDENTIFICATION OF CELLULOSE-DEGRADING BACTERIA

From 12 soil samples, 17 strains of bacteria capable of degrading cellulose compounds were isolated with different shapes, sizes, colors, and cellulose degradation (Table 3).

As can be seen, bacterial strains have a diversity of colors: pale yellow, milky white, light yellow, light pink, and different cellulose-degrading abilities (Table 3). Among those, strains NR1 and NR10 presented strong cellulose degradation ability with a hydrolysis index (HI) larger than 20 mm; while 3 strains (NR7, NR9, and NR12) had a weak ability (HI < 10 mm) and other 12 isolates showed a medium ability with HI ranged from 10 to 20 cm.

3.2 SCREENING THE BACTERIAL ISOLATES FOR CELLULASE ENZYME AND IAA PRODUCTION CAPABILITIES

Seventeen strains of cellulose-degrading bacteria were screened for cellulase activity and IAA production (Figure 1). Strain NR1 produces the highest activity of CMC enzymes, up to 20.60 U ml^{-1}, which was significantly higher than that of other strains. This was followed by NR4, with a CMC enzyme activity of 17.98 U ml^{-1}, and

Table 3: Characterization of cellulose-degrading bacteria strains

Bacterial strain	Colony characteristics	Hydrolysis index (mm)
NR1	Milky white, round shape, serrated edge	24.8 ± 0.00 ^{a}
NR2	Pale yellow, viscous, irregular edge, flat	17.3 ± 0.33 ^{bc}
NR3	Pale yellow, viscous, flat, irregular edge	16.1 ± 037 ^{bc}
NR4	Milky white, flat, irregular edge	14.2 ± 0.32 ^{c}
NR5	Milky white, irregular edge, wrinkled	10.8 ± 0.23 ^{d}
NR6	Milky white, wrinkled, irregular edge	12.3 ± 0.22 ^{cd}
NR7	Milky white, irregular round, viscous	8.3 ± 0.26 ^{e}
NR8	Milky white, irregular round, viscous	18.7 ± 0.45 ^{bc}
NR9	Filamentous, uniform round, light pink	9.6 ± 0.01 ^{de}
NR10	Milky white, irregular round, viscous	20.2 ± 0.13 ^{b}
NR11	Milky white, rough, wrinkled	10.7 ± 0.23 ^{d}
NR12	Milky white, rough, wrinkled	7.6 ± 0.25 ^{e}
NR13	Milky white, rough, wrinkled	10.2 ± 0.12 ^{d}
NR14	Pale yellow, flat, wrinkled, with concentric rings	10.9 ± 0.04 ^{d}
NR15	Light yellow, slightly viscous, round edges	10.1 ± 0.33 ^{d}
NR16	Transparent, viscous, round, concentric ring	13.4 ± 0.43 ^{cd}
NR17	Milky white, uniformly round, concentric ring, viscous	12.2 ± 0.26 ^{cd}

Data are means ± SD (n = 3). Values in the same column with the same letter(s) are not significantly different as determined by the least significant difference (LSD) test ($p < 0.05$)

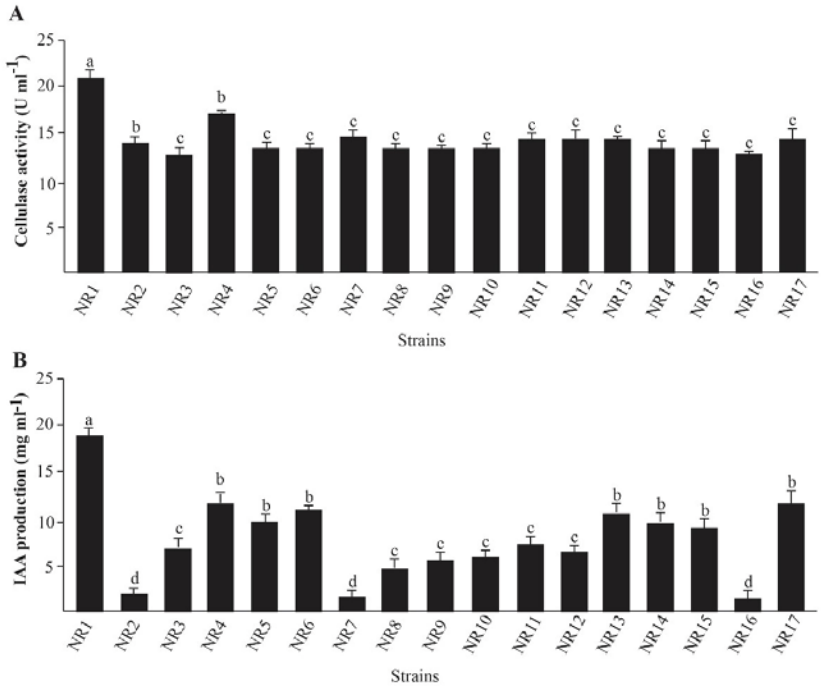

Figure 1: The ability of different strains to produce CMC enzymes (A) and IAA (B). Plotted data are means ± SD (n = 3). The same letter(s) are not significantly different as determined by the least significant difference (LSD) test ($p < 0.05$)

there was no significant difference in CMC enzyme activity among other strains (Figure 1A). The IAA-producing capacity of the NR1 strain is also the strongest, with concentrations of up to 19.83 mg l^{-1}, significantly higher than other strains ($p < 0.05$), and none of the NR2, NR7, and NR16 strains produced IAA (Figure 1B). Therefore, we selected strain NR1 as a multifunctional strain capable of degrading cellulose and synthesizing IAA and carried out straw degradation experiments and pot experiments to verify its decay-promoting and growth-promoting effects.

The 16S rDNA sequence of the NR1 strain was compared to the NCBI database for homology using the blast function. The results showed that the NR1 strain has the highest homology (identity percentage of 99.47 %) and is most closely related to *Bacillus amyloliquefaciens* Priest et al., 1987. The 16S rDNA sequence was deposited on Genbank with the accession number MZ484519.

3.3 EVALUATION OF PLANT GROWTH PROMOTION OF NR1 STRAIN

The greenhouse experiment showed that maize plant and root traits were significantly improved after inoculating with the NR1 strain. The root length, the mean diameter of the root, and the SPAD value increased significantly by 18.3 %, 22.0 %, and 5.24 % respectively ($p < 0.05$, Table 4), compared with the control.

It was reported that IAA participates in many physiological and biochemical regulations in plants, such as cell elongation, cell division, etc., and can promote plant growth (Yue et al., 2005). In this study, the IAA production of the strain NR1 reached 20.15 mg l^{-1}, which is significantly higher than the average amount of the screened strains in the previous reports (Sun et al., 2020). As the plant growth showed a low-promotion and high-inhibition effect with the increase in IAA concentration

Table 4: Effect of strain NR1 on straw degradation promotion and maize growth

Formula	Strawdegradation rate (%)	SPAD value	Average diameter (mm)	Root length (cm)	Root surface area (cm^2)	Plant height (cm)	Aboveground fresh plant mass (g)
Control	9.76 ± 0.90 [a]	40.2 ± 1.62 [a]	0.41 ± 0.04 [a]	23.5 ± 2.29 [a]	111.5 ± 23.0 [a]	51.8 ± 1.25 [a]	3.91 ± 0.42 [a]
NR1	15.1 ± 0.12 [b]	42.3 ± 0.82 [b]	0.50 ± 0.05 [b]	27.8 ± 5.02 [b]	114.3 ± 43.9 [a]	54.4 ± 1.71 [b]	4.12 ± 0.25 [a]

Data are means ± SD (n = 3). Values in the same column with the same letter(s) are not significantly different as determined by the least significant difference (LSD) test ($p < 0.05$)

(Jiang et al., 2000). Therefore, the growth-promoting effect of IAA-producing strains must be comprehensively analyzed with pot experiments. The experiments showed that the SPAD values of maize plants inoculated with strain NR1 were significantly increased compared with the control. This may be because the SPAD value represents the chlorophyll content of the plant, and the higher the value, the stronger the plant's photosynthetic ability. Strain NR1 could belong to nitrogen-fixing bacteria, which can promote the absorption and accumulation of nitrate nitrogen in maize after inoculation (Wu et al., 2011). Nitrogen is an important component of chlorophyll. Therefore, the SPAD value of maize plants significantly increased (Wu et al., 2011). The root length and average diameter of the corn inoculated strain NR1 also increased, indicating that the inoculated maize formed a more developed root system. This confirmed the previous research that IAA produced by microorganisms can promote cell division and differentiation changing the root morphology of plants (Xi et al., 2005); The root surface area is not significantly increased compared with the control, which may be related to external pressure and soil type (Zhang et al., 2018). The condition of the root system directly affects plant growth and nutrient supply, and a well-developed root system can fully interact with nutrients in the soil, thereby improving nutrient utilization and promoting growth (Liu et al., 2017; Nguyen & Nguyen, 2018). However, in this study, the plant height and aboveground fresh mass of inoculated corn increased by 5.0 % and 5.4 % respectively compared with the control, which did not reach a significant correlation level, which may be attributed to the influence of plant species and cultivation conditions (Yu et al., 2015).

3.4 EVALUATION OF STRAW DEGRADATION-PROMOTING ABILITY OF STRAIN NR1 UNDER *IN VITRO* CONDITION

The liquid shake flask test showed that the degradation rate of wheat straw inoculated with strain NR1 reached 15.1 %, which was 54.71 % higher than that of the control ($p < 0.01$, Table 4). These results suggest strain NR1 could produce external cellulase that degrades cellulose into monosaccharides.

The result of this study showed that the CMC enzyme-producing ability of strain NR1 was as high as 20.60 U ml^{-1}. A variety of cellulose-degrading strains have been found. For example, the decomposing bacteria ZJA-6 isolated by Wei et al. (2015) exhibited a CMC enzyme activity of 13.20 U ml^{-1}; Li et al. (2019) reported the enzyme activity of Actinomycetes C31 reached 4.8 U ml^{-1}; the enzyme activity of *Burkholderia* ME27-1 reported by Liang et al. (2014) was only 2.08 U ml^{-1} under optimized conditions. The CMC enzyme activity of strain NR1 was 1.56 times, 4.29 times, and 9.90 times that of strains ZJA-6, C31, and ME27-1, respectively, indicating that strain NR1 had relatively high CMC enzyme activity. It should be noted that the application of the strain NR1 needs to be comprehensively judged in combination with CMC enzyme activity and straw degradation test (Wang et al., 2016) because straw is composed of cellulose, hemicellulose, and lignin through covalent bonds, hydrogen bonds and it is a water-insoluble polymer compound composed of a variety of molecular forces such as wax bonds. The outside of cellulose is tightly wrapped by lignin and hemicellulose, which is difficult to be decomposed by cellulase (Yu & Guo, 2019). The straw degradation test showed that the straw degradation rate of strain NR1 reached 15.1 % in 15 days, which was 54.71 % higher than that of natural degradation straw. These results indicated that the addition of strain NR1 can significantly improve the straw degradation yield. Therefore, the strain NR1 isolated in this study not only exhibits high enzyme activity but also can accelerate the process of straw degradation, which is expected to improve the comprehensive utilization rate of straw in practical applications.

3.5 EVALUATION OF THE ABILITY TO DEGRADE AGRICULTURAL BYPRODUCTS OF NR1 STRAIN

The strain NR1 was investigated for its potential ability to decompose cellulose-rich agricultural byproducts under natural conditions. The results are presented in Table 5.

The data in Table 5 showed that the cellulose content was dramatically reduced at the formula inoculated with NR1 strain (66.7 %) while a slight decrease of it was observed for the control. These results suggest strain NR1 still kept its strong ability to degrade indigestible organic compounds very well by secreting the external cellulase under natural conditions.

Protein, phosphorus, and potassium are the necessary nutritional elements that determine crop yield. Determination of the total protein, phosphate, and potassium amount in compost plays an important role in considering the possibility of supplying N, P, and K from the manure. The results in Table 5 showed that the total amount of protein, phosphorus, and potassium content in the formula for supplementing strain NR1 all increased and were higher than the formula without supplements of strain NR1. Compared to research reports on the quality of microbial compost composted from agricultural byproducts, the total protein, phosphate, and potassium

Table 5: Effect of strain NR1 on cellulose degradation and compost quality

	Cellulose content (%)		Compost quality (%)		
Formula	Before incubation	After incubation	Total N	Total P_2O_5	Total K_2O
I (Control)	14,5 ± 0.3 [a]	11,2 ± 0.2[a]	0,83 ± 0.2 [a]	0,36 ± 0.3 [a]	0,63 ± 0.4 [a]
II	14,4 ± 0.4 [a]	4,8 ± 0.3 [b]	1,22 ± 0.1 [b]	0,45 ± 0.2 [a]	0,76 ± 0.2 [a]

Data are means ± SD (n = 3). Values in the same column with the same letter(s) are not significantly different as determined by the least significant difference (LSD) test ($p < 0.05$)

content in the compost of this study is equivalent to or higher than some other studies (Tran et al., 2011).

3.6 EVALUATION OF THE ABILITY TO STIMULATE PEANUT GROWTH UNDER FIELD CONDITIONS OF NR1-PRODUCED COMPOST

The results showed that the height of the peanut plant in the formulas added to the compost (CT2, CT3, and CT4) is higher than the one in the control (CT1, Table 6). Especially, the increase of compost (7, 8, and 9 tons ha^{-1}) enhanced the peanut plant height (33.34, 36.43, and 37.45 cm, respectively).

In addition, the addition of compost produced by the NR1 strain also enhanced the yield of the peanut plant (Table 6). As can be seen, all treatments added compost fertilizer did not affect the total number of fruits on the plant and the total number of fertilized fruits on the plant, with no statistically significant discrepancies. However, the mass of 100 fruits and the practical yield differed between the experimental formulas and the control and among the experimental formulas.

The mass of 100 fruits is the indicator that determines the productivity of the experimental formulas. The results showed that the highest mass of 100 fruits was observed in CT4 (160.34 g), followed by the one in CT3 (154.12 g), and the lowest values were in CT1 (142.85 g) and CT2 (147.31 g). Moreover, practical yield is an important indicator for assessing the effectiveness of the compost in the growth, development, and productivity of peanut plants. The practical yield of peanut plants in CT2, CT3, and CT4 was 3.01, 3.35, and 3.48 t ha^{-1}, respectively, and was significantly different from the one of the control (2.83 t ha^{-1}). These results indicated that NR1 strain in the compost produced IAA compounds and the composition of nutrients in composted organic fertilizers has added timely nutrition for peanut growth and hence improved productivity.

The analysis results of some chemical properties of the soil before and after the experiment are presented in Table 7.

The data presented in Table 6 aligns with previous research, demonstrating a positive correlation between rice straw compost application and practical yield. Iqbal (2008) observed similar trends, reporting that a combination of rice straw compost and 75 % recommended nitrogen fertilizer yielded the highest protein content in grains. This finding can be attributed to the compost's contribution of readily available phosphorus (P) and potassium (K) nutrients, essential for protein and carbohydrate synthesis within the plant (Iqbal, 2008). Furthermore, Madejon et al. (2001) documented improvements in plant nutritional status, growth response, and overall productivity following compost amendments.

Our study reinforces the potential of composted agricultural by-products as organic fertilizers for soil enhancement. This aligns with the established role of organic matter in improving soil properties, as demonstrated by Giller et al. (1998) and Nguyen & Nguyen (2018). These prior investigations highlight the potential for or-

Table 6: Effect of NR1strain-produced compost on the plant development and yield of peanuts

Formula	Plant height (cm)	Total number of fruit/plant (fruit)	Number of fertilized fruits/plant (fruit)	Mass of 100 fruits (g)	Theoretical yield (tons ha^{-1})	Practical Yield (tons ha^{-1})
CT1	31.21 [a]	21.24 [a]	12.91 [a]	142.85 [a]	4.89 [a]	2.83 [a]
CT2	33.34 [ab]	22.58 [ab]	13.41 [ab]	147.31 [ab]	5.13 [ab]	3.01 [b]
CT3	36.43 [b]	23.17 [b]	14.53 [b]	154.12 [b]	5.49 [b]	3.35 [ab]
CT4	37.45 [b]	23.75 [b]	15.15 [c]	160.34 [c]	5.91 [c]	3.48 [c]
LSD0.05	3.56	5.34	3.64	9.11	8.94	4.17

Data are means (n = 3). Values in the same column with the same letter(s) are not significantly different as determined by the least significant difference (LSD) test ($p < 0.05$)

Table 7: Effect of compost on some chemical properties of groundnut soil

Formula	pH_{KCl}	OC (%)	Total N (%)	Total K_2O (%)	Total P_2O_5 (%)
Before experiment	4,97	1,37	0,09	0,30	0,05
CT1	4,99	1,55	0,13	0,35	0,05
CT2	5,80	1,64	0,13	0,28	0,05
CT3	5,81	1,73	0,11	0,30	0,06
CT4	5,50	1,69	0,11	0,36	0,07

The data showed that fertilization not only increases the yield of peanuts but also improves soil fertility. After the experiment, the pH_{KCl} and organic content (OC) increased fluctuated in the experimental formulas from 4.99–5.81 and 1.55-1.73, respectively. Other factors (total N, K_2O, and P_2O_5) showed no difference between the treatment and the control. These results suggest the NR1-produced compost could be applied to increase the soil's OC and balance pH

ganic fertilizers to replace chemical fertilizers partially or fully in agricultural practices.

4 CONCLUSIONS

In this study, *Bacillus amyloliquefaciens* NR1 screened from the root rhizosphere has both straw degradation and crop growth-promoting abilities. The above conclusions have a certain positive significance for guiding the creation and application of multifunctional straw-degrading bacteria in composting agricultural waste that could improve plant growth, plant yield, and soil fertility.

5 ACKNOWLEDGEMENTS

This research has been supported by the VNU-Central Institute for Natural Resources and Environmental Studies (VNU-CRES). The VNU-CRES provided funds for collecting samples and buying chemicals for experiments and supported experimental facilities to carry out experiments.

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