Sains Malaysiana 53(1)(2024): 23-37
http://doi.org/10.17576/jsm-2024-5301-03

Tripartite Introductions of PGPR, Humic Acid, and N-Fertilizer Improve the Growth and Yield of Sweet Potato under Glasshouse Conditions

(Pengenalan Tripihak PGPR, Asid Humik dan Baja N Meningkatkan Pertumbuhan dan Hasil Ubi Keledek di bawah Keadaan Rumah Kaca)

Buraq Musa Sadeq[1], Ali Tan Kee Zuan[1,*], Susilawati Kasim[1], Jawadyn Talib Alkooranee[2], Wong Mui Yun[3], Nur Maizatul Idayu Othman[4], Amaily Akter[1], Sayma Serine Chompa[1], Abba Nabayi[5] & Md Ekhlasur Rahman[1,6]

[1]*Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia*
[2]*Department of Plant Protection, Faculty of Agriculture, University of Wasit, Wasit, Iraq*
[3]*Department of Plant Protection, Faculty of Agriculture, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia*
[4]*Faculty of Plantation and Agrotechnology, Universiti Teknologi MARA, Melaka, Kampus Jasin 77300 Merlimau, Melaka, Malaysia*
[5]*Department of Soil Science, Faculty of Agriculture, Federal University Dutse, Nigeria. PMB 7156, Ibrahim Aliyu bye-pass Jigawa state, 720101, Nigeria*
[6]*Divisional Laboratory, Soil Resource Development Institute, Krishi Khamar Sharak, Farmgate, Dhaka-1215, Bangladesh*

Received: 17 April 2023/Accepted: 4 December 2023

ABSTRACT

This study was undertaken to investigate the effects of plant growth-promoting rhizobacteria (PGPR) with humic acid (HA) as amendments on the morphological and physiological growth characteristics and yield of Sepang Oren sweet potato (*Ipomoea batatas* [L.] Lam). The experiment was conducted under glasshouse conditions at the Faculty of Agriculture, Universiti Putra Malaysia, for 110 days. Two factors were used in this experiment: The first factor was PGPR-HA inoculations (UPMB10, UPMRB9, and mixed strains) and non-inoculation-HA, and the second factor was the Nitrogen fertilizer levels (50, 75, and 100%). The treatments were replicated three times and arranged factorially in a randomized complete block design. The results showed that inoculations with PGPRs-HA (UPMRB9 and UPMB10 strains) positively affect the plant growth significantly (SPAD measurements, number of leaves, vine length, root length, leaf area index, root dry weight, shoot dry weight, and root: shoot ratio) of sweet potato upon addition of 50% and 75% of N-fertilizer, respectively. The nutrient content of soil and plant leaf significantly increased by 12-15% and 14-18%, respectively, compared to the uninoculated, when applied with the same inoculation. After 30 days, the population of soil bacteria increased, reaching a value of 8.65 $\log_{10}$ CFU/g soil. The use of PGPR-HA inoculations with N-fertilization resulted in a considerable rise in the majority of plant and soil parameters compared to the treatments without PGPR inoculation. Therefore, PGPR supplemented with humic acid (HA) may be considered a viable and sustainable strategy for enhancing sweet potatoes' morphological and physiological attributes. This technique can result in increased crop productivity and serve as a substitute for nitrogen-based fertilizers.

Keywords: Bacterial population; humic acid (HA); PGPR; soil nutrients; sweet potato yield

ABSTRAK

Penyelidikan ini dijalankan untuk mengkaji kesan rizobakteria penggalak pertumbuhan tumbuhan (PGPR) dengan asid humik (HA) sebagai pindaan terhadap ciri pertumbuhan morfologi dan fisiologi serta hasil ubi keledek Sepang Oren (*Ipomoea batatas* [L.] Lam). Uji kaji dijalankan di bawah keadaan rumah kaca di Fakulti Pertanian, Universiti Putra Malaysia, selama 110 hari. Dua faktor telah digunakan dalam uji kaji ini: Faktor pertama ialah inokulasi PGPR-HA (UPMB10, UPMRB9 dan strain campuran) dan bukan inokulasi-HA serta faktor kedua ialah kepekatan baja Nitrogen (50, 75 dan 100%). Setiap rawatan mengandungi tiga replikasi dan disusun secara faktorial dalam reka

bentuk blok lengkap rawak. Hasil menunjukkan bahawa inokulasi dengan PGPRs-HA (strain UPMRB9 dan UPMB10) memberi kesan positif kepada pertumbuhan tumbuhan dengan ketara (ukuran SPAD, bilangan daun, panjang pokok, panjang akar, indeks luas daun, berat kering akar, berat kering pucuk dan nisbah akar: pucuk) ubi keledek selepas penambahan 50% dan 75% baja N. Kandungan nutrien tanah dan daun tumbuhan masing-masing meningkat dengan ketara sebanyak 12-15% dan 14-18% berbanding dengan tumbuhan yang tidak diinokulasi. Selepas 30 hari, populasi bakteria tanah meningkat, mencapai nilai 8.65 $\log_{10}$ CFU/g tanah. Penggunaan inokulasi PGPR-HA dengan pembajaan N menghasilkan peningkatan yang ketara dalam majoriti parameter tumbuhan dan tanah berbanding dengan rawatan tanpa inokulasi PGPR. Oleh itu, PGPR Bersama-sama asid humik (HA) boleh dianggap sebagai strategi yang mampan untuk meningkatkan sifat morfologi dan fisiologi ubi keledek. Teknik ini boleh menghasilkan peningkatan produktiviti tanaman dan berfungsi sebagai pengganti baja berasaskan nitrogen.

Kata kunci: Asid humik (HA); hasil ubi keledek; nutrien tanah; PGPR; populasi bakteria

Introduction

Sweet potato (*Ipomoea batatas* [L.] Lam) is a tuber crop native to Central America and the western coast of South America. In Malaysia, after cassava, sweet potato is the second-most significant root crop (Karim, Devarajan & Ahmad 2022; Shankar & Kaushik 2022). Sweet potato is one of the healthy foods recommended by the United Nations' Food and Agriculture Organization (FAO) due to the presence of anthocyanins, β-carotene, minerals (Ca, Zn, K, and Mg), and vitamins B1, B2, E, and C (Muhammad et al. 2022). Several nations, like China, Vietnam, and India, prioritize the cultivation of sweet potatoes as their primary agricultural product. In Malaysia, sweet potatoes hold significant economic value and may be classified as cash crops, holding around 24.93% of the country's total planted area (Zhu & Sun 2019). The production rate of sweet potatoes has declined in recent years, and this could be attributed primarily to the poor soil nutrient status, which consequently leads to lower yield (Agbede & Oyewumi 2022).

Nitrogen (N) is a vital element in the production of sweet potatoes, and N fertilizers are widely utilized in modern agriculture to nourish plants, enhance production, and improve end-use quality. Ensuring enough nutrition is crucial for cereal crops' optimal growth and productivity. Therefore, agricultural practitioners throughout various global areas prefer utilizing nitrogen-based fertilizers to augment and optimize crop yield (Folina et al. 2021). Excessive use of nitrogen fertilizer incurs higher production expenses and diminishes economic profits. It also engenders substantial environmental deterioration, encompassing the occurrence of acid rain, soil acidification, and groundwater acidification. These adverse consequences adversely impact soil fertility and the depletion of beneficial soil microorganisms. One technique for sustaining crop production is to use beneficial microbes such as plant growth-promoting rhizobacteria (Zeng et al. 2022; Zhao et al. 2022).

In sustainable farming, biofertilizer has been identified as an alternative for enhancing soil fertility and crop yield. In soil-integrated nutrient management, biofertilizers can play an essential role. N-fixers, potassium and phosphorus solubilizers, plant growth-promoting rhizobacteria (PGPR), endo- and ectomycorrhizal fungi, and other beneficial microscopic creatures are examples of microorganisms that are often utilized as biofertilizer inoculum (Itelima et al. 2018; Manna et al. 2021). In addition, biofertilizers benefit plants by converting and mobilizing unavailable elements to available forms via various biological processes (Meena et al. 2017). PGPR species such as *Bacillus tequilensis* and *Bacillus subtilis* have been found colonizing plant roots (Mahapatra, Yadav & Ramakrishna 2022). When comparing *B. subtilis* and *B. tequilensis* to other bacteria and how they affect plants, it is important to remember that different strains of bacteria have different traits and effects. Nevertheless, extensive research has demonstrated that these strains have several advantageous impacts on plants' development and overall well-being (Goswami, Thakker & Dhandhukia 2016). For instance, research has demonstrated that it facilitates the development of plants and their ability to withstand stress through mechanisms such as improved absorption of nutrients, the synthesis of phytohormones including indole-3-acetic acid (IAA), gibberellin, and cytokines, as well as the activation of systemic resistance against infections (Etesami et al. 2015). Furthermore, apart from

the strains mentioned, a plethora of different bacterial varieties have the potential to provide advantageous effects on plant development. Rhizobacteria are renowned for their capacity to engage in nitrogen fixation within the soil, facilitating plant development and production enhancements. In a similar vein, it has been demonstrated that Pseudomonas can enhance plant development through the production of siderophores, which facilitate the absorption of iron by plants from the surrounding soil (Mabrouk et al. 2018; Pandita 2022). Many PGPR inoculants have increased nutrient uptake in various crops, including cotton, pea, tomato, peanut, and maize (Egamberdiyeva & Höflich 2004). The PGPR has been reported to stimulate root growth and thereby increase root surface area for nutrient uptake and is crucial for promoting plant growth (Zilaie et al. 2022).

Humic acids (HA) are crucial as a medium for transferring nutrients from the soil to the plant because they can store ionized nutrients and prevent them from being flushed away. Humic acids keep and supply the plant's water and nutrients (Wu, Li & Chen 2020). Direct impacts of HA on plant development have been thoroughly reported, including increased macro- and micronutrient uptake and root expansion, improved microbial growth by providing a carbon source that serves as food for microbes, and increased soil water retention (Tang et al. 2022). Developing a good strategy for combining HA and biofertilizer for agricultural application under open field conditions is critical for improving soil fertility and productivity and decreasing the use of inorganic fertilizer for higher crop production (Al-Taey et al. 2019). Initially, humic chemicals have primarily been examined as carbon or micronutrient sources or for their overall impact on microbial development (Benz, Schink & Brune 1998). Using organic elements such as HA and beneficial bacteria as fertilizer has improved soil health and increased broccoli crop output (Al-Taey et al. 2019). Adding humic acid and urea fertilizer (HA-N) considerably increased the yield of sweet potato plants by 29.6%, the average fresh weight per storage root, and the number of storage roots per plant (Chen et al. 2017). In a study conducted by Sadeq et al. (2023), it was shown that a concentration of 0.1% HA resulted in a notable increase in the bacterial population of *B. tequilensis* (UPMRB9) and *B. subtilis* (UPMB10) strains. Additionally, this concentration of HA demonstrated efficacy in extending the shelf-life of both bacterial strains.

Sweet potato is one of the most important root crops worldwide, providing numerous raw materials for the agricultural industry, as well as food and feed for people and animals (Neela & Fanta 2019). In this study, we hypothesized that a combination of beneficial bacteria with humic acid (HA-PGPR) and optimal N-fertilizer would boost the growth and yield of sweet potatoes while enhancing the soil nutrient status in an economically and environmentally favorable manner. Thus, a pot trial was conducted with two locally isolated bacteria (UPMRB9 and UPMB10) with humic acid and three different rates of N-fertilizer (50%, 75%, and 100%) under glasshouse conditions. The study aimed to evaluate the effect of formulated biofertilizer inoculation with different N-fertilizer rates on the growth and yield of sweet potatoes. The study also determined the effect of applying PGPR-HA formulation on the physiological and morphological measurements of the sweet potato leaf, as well as the relationships between the soil and plant growth parameters.

Materials and Methods

PGPR strains and humic acid collection, preparation and characterization

Locally isolated PGPRs, *B. tequilensis* (UPMRB9), and *B. subtilis* (UPMB10) were collected from the Soil Microbiology Laboratory, Department of Land Management, Faculty of Agriculture, Universiti Putra Malaysia. These PGPRs were selected due to their beneficial biochemical and morphological characteristics, including N_2 fixation, phosphate and potassium solubilization, siderophore production, and pectinase production. These PGPR strains were also reported to increase the concentrations of N, P, and K in shoots and storage roots (Kapadia et al. 2021). Pure humic acid was purchased from Sigma-Aldrich (102098564, 53680-50G Swiss). Humic acid is black, has a pH of less than 6, and has low solubility in water (Table 1). HA is insoluble in acid but is soluble in alkali and in tryptic soy broth media (TSB) due to its higher pH (7.23).

Collection and characterization of initial soil sample

Mineral soil was collected from the University Agriculture Park, Universiti Putra Malaysia, Selangor, Malaysia. The location coordinates were 30 02′ N latitude, 1010 42′ E longitude, and 31 m above sea level. The soil was classified as sandy clay-textured, according to the United States Department of Agriculture (USDA) soil

classification system. Air-dried soil samples weighing 10 g were grounded to pass through a 2 mm sieve. The soil sample was characterized by its physical and chemical properties (Table 2). The soil pH in a 1:2.5 (weight/volume) ratio of soil and water, and the electrical conductivity (EC) for soil samples were determined by a pH meter and conductivity meter, respectively. The total carbon (C), nitrogen (N), and sulfur (S) were analyzed by dry combustion method using a CNS auto-analyzer (LECO Corporation, St. Joseph, MI, USA). The cation exchange capacity (CEC) was assessed by titration after being extracted by the leaching method using neutral 1 M ammonium acetate (NH_4OAc) solution at pH 7 (Thomas 1983). The total soil nutrients were extracted using the Aqua Regia method in a 0.5:1:3 (weight/volume/volume) ratio of soil, nitric, and hydrochloric acid, respectively. Later, the nutrients were determined by inductively coupled plasma optical emission spectroscopy (ICP-OES, Perkin Elmer, Boston, MA, USA). All the analyses were conducted in triplicate.

THE GLASSHOUSE TRIAL SETUP

The pot experiment was conducted in a glasshouse at the Faculty of Agriculture, Universiti Putra Malaysia, Selangor (298036.60 N latitude: 10173081.90 E longitude). Seven kg of mineral soil was filled in plastic pots measuring 23 cm height by 26 cm inner diameter. Before planting, Sepang Oren cuttings of the sweet potato variety (30 cm in length with 8 nodes) were cleaned carefully using sterilized distilled water and soaked in 48 h-old PGPRs-HA formulations for 6 h (Figure 1). About 0.1 g of humic acid was mixed with 100 mL of TSB and the solutions were autoclaved for 15 min at 121 °C. One full loop (approximately 1×10^6 CFU mL^{-1}) was taken from *B. tequilensis* and *B. subtilis* cultures and dipped into broth media for single and mixed strain, before incubated under a constant shaker at 150 rpm for 48 h at 30 °C. Each sample was replicated thrice. During the planting process, 20 mL (about 10^9 CFU mL^{-1}) of PGPRs-HA formulations was introduced onto each plant, with 2 week intervals of inoculation. Meanwhile, the uninoculated plants received the same volume of sterile media but without bacterial inoculation. To supply the recommended rate of the P and K as suggested by Kashiani (2012), triple superphosphate (TSP) and muriate of potash (MOP) was used at 80 kg ha^{-1} P_2O_5 and 120 kg ha^{-1} K_2O, respectively. Urea was used as the source of N fertilizer and was applied at three levels 50%, 75%, and 100% N. Furthermore, N, P, and K were added at 7 and 45 days after planting. Based on the recommended rates, urea was applied at 2.16 g for 50% N, 3.24 g for 75% N, and 4.38 g for 100% N, for each pot, respectively. About 13.14 g of P_2O_5 and 14.22 g of K_2O. The plants were irrigated regularly twice daily at 8 am and 5 pm. Soil samples were collected from the vicinity of plant roots to determine bacterial counts using the total plate count technique (TPC). The initial count of the bacteria before planting was 6.42 $\log_{10}$ CFU/g soil. Then, the bacterial count was assessed at three different time points: 30, 60, and 90 days after planting (DAP). Plants were harvested three months after they were planted.

TABLE 1. Physical and chemical properties of humic acid

Properties	Humic acid
Color	Black
Solubility	low
Molecular formula	$C_9H_9NO_6$
Molecular weight	227.17
Organic C (%)	30-50
Hydrogen (%)	5
Nitrogen (%)	3
pH	3.5- 6
CEC (cmolc/kg)	70-166
Moisture (%)	15

EXPERIMENTAL DESIGN AND TREATMENT

The experiment was arranged in a Randomized Complete Block Design (RCBD) with 12 treatment combinations. The treatment combination consists of two factors: The first factor was the bacterial strains, mixed strains and uninoculated control, all amended with 0.1% humic acid (HA), and the second factor was the three levels of N fertilizer; 50, 75, and 100 %. Consequently, 12 treatment combinations with three replications were obtained (Table 3).

TABLE 2. Selected physical and chemical properties of initial soil

Properties	Soil
Textural class	Sandy clay loam
% Sand	67.70
% Silt	5.50
% Clay	27.60
pH	5.13
EC (mS/cm)	17.78
CEC (cmolc/kg)	5.82
Total C (%)	1.75
Total N (%)	0.05
Total S (%)	0.04
Available P (mg /kg)	2.13
Exchangeable K (cmolc/kg)	0.49
Exchangeable Mg (cmolc/kg)	0.47
Exchangeable Ca (cmolc/kg)	1.68

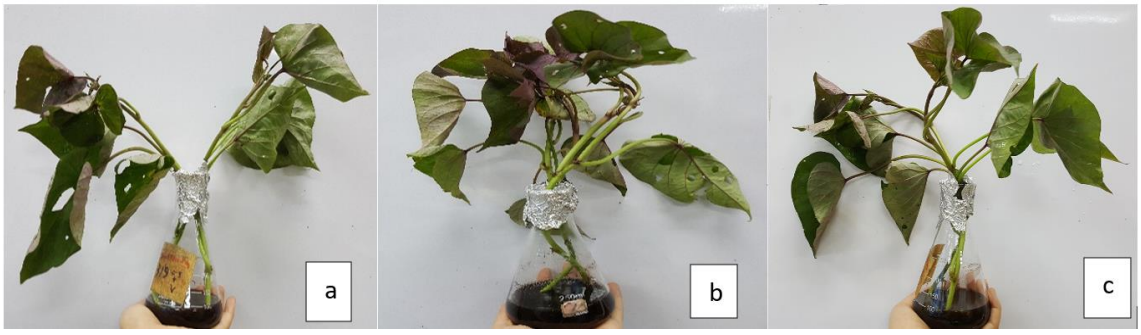

FIGURE 1. Sweet potato cuttings soaked in PGPR-HA formulation for 6 h in (a) UPMB10, (b) UPMRB9, and (c) mixed strain

TABLE 3. Treatments for the pot experiment

Treatment code	Description of the treatments
T1	50% Nitrogen fertilizer + 0.1% HA (uninoculated)
T2	50% Nitrogen fertilizer + 0.1% HA with UPMB10
T3	50% Nitrogen fertilizer + 0.1% HA with UPMRB9
T4	50% Nitrogen fertilizer + 0.1% HA with mixed strain
T5	75% Nitrogen fertilizer + 0.1% HA (uninoculated)
T6	75% Nitrogen fertilizer + 0.1% HA with UPMB10
T7	75% Nitrogen fertilizer + 0.1% HA with UPMRB9
T8	75% Nitrogen fertilizer + 0.1% HA with mixed strain
T9	100% Nitrogen fertilizer + 0.1% HA (uninoculated)
T10	100% Nitrogen fertilizer + 0.1% HA with UPMB10
T11	100% Nitrogen fertilizer + 0.1% HA with UPMRB9
T12	100% Nitrogen fertilizer + 0.1% HA with mixed strain

SOIL NUTRIENT ANALYSES AFTER HARVEST AND SWEET POTATO LEAF NUTRIENT CONCENTRATION

After harvest, a soil sample was taken, air-dried, and grounded to pass through a 0.2 mm sieve for the analyses of soil pH, base cations K, Ca, Mg, and available P. The Aqua Regia method (Kamali et al. 2021) was used to determine soil nutrients by using nitric and hydrochloric acids at a 1:3 ratio. The oven-dried sweet potato leaf was pulverized with an electric grinder and then sieved through a 0.5 mm sieve. The dry-ashing method was used to extract K, P, Ca, and Mg from sweet potato plant shoot using HCl and 20% HNO_3, and the filtrates were determined using inductively coupled plasma optical emission spectroscopy (ICP-OES, Perkin Elmer, Boston, MA, USA). The Micro-Kjeldahl method was used to determine the total N concentration of soil and sweet potato leaf. This was followed by adding concentrated H_2SO_4, K_2SO_4, and catalyst, followed by 1 h of digestion at 360 °C, using block digestion (Bowman, Paul & Carlson 1988).

PLANT GROWTH AND BIOMASS PRODUCTION MEASUREMENTS

Plant-growth measurements were taken during the plant's growth period. The Soil Plant Analysis Development (SPAD) chlorophyll meter was used to estimate the leaf greenness and chlorophyll concentrations through the dynamic chlorophyll distribution (Yuan et al. 2016). The number of leaves was counted based on taking the count of the leaves 10 cm from the tip of the vine. Vine and root length were measured by using a tape measure. After harvest, the roots were cleaned using running tap water. The shoot and storage root specimens were dried at 70 °C until a stable weight was achieved. The resulting dry weight was then measured using a digital weighing device, and the ratio of shoot to storage root (R/S ratio) was then calculated. Leaf Area Index (LAI) was measured using the LAI-2000 Plant Canopy Analyzer (Li-CoR Inc; Lincoln, NE, USA).

PERCENT RELATIVE DATA

The relative data of the values were expressed as percentages relative to the control value for each element following Mosharrof et al. (2021).

$$\text{Relative data (\%)} = \frac{(\text{Treatments value} - \text{control value})}{\text{control value}} \times 100$$

where the treatment value was the PGPR-HA with N-fertilizer, and the control value was without PGPR inoculation.

STATISTICAL ANALYSIS

All the data collected were analyzed using the Statistical Analysis System (SAS) 9.4. Following the analysis of variance procedure (ANOVA), the Least Significant Difference (LSD) comparison method was used to compare differences between treatment means at $p = 0.05$.

RESULTS AND DISCUSSION

EFFECT OF TREATMENTS ON SWEET POTATO GROWTH AND YIELD-CONTRIBUTING PARAMETERS

The treatments differ significantly ($p \leq 0.05$) from one another in terms of SPAD readings, number of leaves, vine length, root length, and LAI of sweet potato with the higher values in treatment combinations of PGPRs-HA with nitrogen fertilization rates (Table 4). The treatments UPMRB9+50%N (T3) and mixed strain+100%N (T12) exhibited the highest SPAD values of 43.32 and 43.24, respectively, which were found to be statistically significant at ($p<0.05$). Conversely, the treatments UPMB10+50%N (T2) and UPMRB9+75%N (T7) had the lowest SPAD values of 28.24 and 28.20, respectively. Table 4 demonstrates that the use of PGPRs-HA inoculation and N-fertilizer in soil amendments resulted in a notable increase in the number of leaves, vine length, root length, and LAI of sweet potatoes, as compared to the soil that was not inoculated. Both treatments of UPMRB9+50%N (T3) and UPMB10+75%N (T6) exhibited a significantly greater leaf count, surpassing the uninoculated treatments by 100%. Treatments T3 and T6 had the most extensive vine length, measuring 156.2 and 154.3 cm, respectively, demonstrating a notable divergence from the remaining treatments. Nonetheless, there was no statistically significant difference between the UPMRB9+75%N treatment (T7) and the mixed strain+100%N treatment (T12) concerning the length of sweet potato vines, which ranged from 90.4 to 89.2 cm. However, it is worth noting that the uninoculated treatment (T1) exhibited the shortest vine length among all treatments.

The treatment UPMRB9+50%N (T3) exhibited the largest root length (15.4 cm) and leaf area index (408.24 cm), which were statistically different from the other treatments. Conversely, treatment T1 (uninoculated) had a considerably shorter root length, measuring 46.73% lower than the other treatments. The UPMB10+50%N (T2) and mixed strain+100%N (T12) treatments had considerably lower LAI values, measuring 225.39 cm and 234.08 cm, respectively. Treatments T3 and T6 exhibited statistically significant increases in root dry weight, shoot dry weight, and root-to-shoot ratio (Table 4), indicating notable distinctions from the remaining treatments. The plants subjected to UPMRB9+50%N (T3) and UPMB10+75%N (T6) exhibited notably elevated dry weights in both root (43.6 g) and shoot (29.38 g) components, as well as higher values for the root-to-shoot ratio (2.7) when compared to the remaining treatments.

Leaf number, vine length, root length, and LAI increased with PGPR-HA inoculation and N-fertilization compared to the uninoculated (Table 4). The observed enhancement in plant development in treatments infected with PGPR-HA may be attributed to the capacity of *B. tequilensis* and *B. subtilis* to synthesize indole acetic acid (IAA), siderophores, and protease activity, which acts as a growth-promoting hormone (Baard et al. 2023; Bahadur et al. 2017). According to Ekin (2019), applying a mixed culture of PGPR combined with humic acid resulted in a significant increase of around 140% in potato tuber production. According to Meng et al. (2021), humic acid as a soil amendment can accelerate nutrient absorption, reduce toxins, increase water retention, improve microbial growth by providing shelter and carbon sources as food for bacteria, and improve soil structure. Chen et al. (2017), El-Sawah et al. (2021), and Khan et al. (2021) conducted studies that demonstrated the positive effects of combining nitrogen application with PGPR and humic acid on plant development and yield. These studies found that this combination outperformed fertilizer alone and significantly improved the efficiency of nitrogen fertilizer production. According to Wang et al. (2022), combining humic acid (HA) and N-fertilizer improves nutrient utilization and crop yield.

TOTAL BACTERIAL POPULATION OF SOIL AFTER HARVEST

The total bacterial population in the soil was affected by PGPRs-HA inoculation and nitrogen fertilization rates (Figure 2). There was a significant effect of PGPR-HA inoculation and nitrogen fertilization on the bacterial population in the soil. There were apparent changes in the population of the bacteria during various stages of crop development. Generally, the population of bacteria was at its highest level 30 days after planting and started decreasing at 60 and 60 DAP. Compared to the uninoculated control (7.5 $\log_{10}$ CFU/g soil), the inoculation treatments had a larger bacterial population. The highest population (8.65 $\log_{10}$ CFU/g soil) was obtained with the inoculation of UPMB9 with 50% N-fertilizer at 30 DAP and reduced with the increase of the plant age.

The application of PGPR-HA inoculation and N-fertilization substantially impacted the microbial population density in the soil surrounding the plant roots throughout the plant growth stages. The overall bacterial population in the soil was shown to be considerably greater in treatments where *B. tequilensis* (UPMRB9) inoculations were applied, compared to other treatments across different phases of N-fertilizer application. The inoculation of PGPR-HA may stimulate the sweet potato plant to produce metabolites and hormones that promoted the production of the other native bacterial strains (Ahmad et al. 2016). The inoculation with *B. tequilensis* may accelerate root development and increased root exudate output (Kapadia et al. 2021). Moreira et al. (2022) reported that adding N fertilizer to the soil provides bacteria with a nitrogen supply, thus, enhancing their growth.

PGPR inoculation and nitrogen fertilization significantly affected microbial populations in plants and soil (Kaur et al. 2022). The microbial population density in the soil around plant roots during plant growth was shown to be strongly impacted by the inoculation of PGPR-HA and the administration of N fertilizer. The overall population of bacteria exhibited high levels at 30 days post-planting but subsequently decreased as the plants aged, as shown in Figure 2. The rhizobacteria implanted likely stimulated the production of plant growth hormones and other metabolites, hence promoting the proliferation of native bacteria. According to Huang et al. (2014), it has been proposed that root exudates consist of many components such as sugars, amino acids, vitamins, tannins, alkaloids, phosphatides, and additional chemicals that have not yet been found. The sugars released by root exudates serve as easily accessible carbon and energy sources for the bacterial population in the soil.

The use of a minimal amount of nitrogen (N) fertilizer supplies nitrogen for bacteria, hence enhancing their growth and development. In their study, Bashan et al. (2014) reported a notable impact of nitrogen fertilization and PGPR inoculation on microbial communities inside the rhizosphere soil of barley plants. The observed decline in population size after 60 and 90 days of planting may be attributed to competitive interactions for limited resources, such as nutrients and space, between the introduced inoculants and the native bacterial community inside the soil. The rhizobacteria are likely engaged in competition for carbon and energy resources and vying for colonization opportunities inside the rhizosphere (Yasmin, Othman & Maziz 2020).

EFFECT OF TREATMENTS ON SOIL NUTRIENTS AFTER HARVEST

After harvest, the soil pH was found to increase significantly ($p \leq 0.05$) in the different treatments as compared to the uninoculated (Table 5). The pH values that exhibited the most significant magnitude were recorded in the UPMRB9+50%N treatment (T3) and the UPMB10+75%N treatment (T6). The pH values of 6.59 were recorded in treatments with inoculation, whereas the lowest was observed in treatments without inoculation. Significant variations in the total nitrogen content resulting from the treatment combinations of PGPR-HA with varying N-fertilizer rates were observed (Table 5). The soil nutrients were found to be considerably higher ($p \leq 0.05$) in the treatments UPMRB9+50%N (T3) and UPMB10+75%N (T6). The lowest values were seen in the treatments without inoculation, specifically T1, T5, and T9. The T3 treatment exhibited notable increases in soil N, exchangeable K, and Ca levels, with corresponding increments of 36.84%, 44%, and 30% compared to the uninoculated treatment. The T6 treatment exhibited a notable increase in soil-accessible phosphorus (P) and magnesium (Mg), respectively, 148.98% and 76.40% compared to the uninoculated treatment. There was no statistically significant difference between the UPMRB9+50%N (T3) and UPMB10+75%N (T6) treatments in relation to the availability of soil phosphorus. Both treatments exhibited the maximum soil phosphorus levels of 14.79 mg/kg and 14.49 mg/kg, respectively. Nevertheless, there were no significant differences seen among the treatments, including mixed strain+50%N (T4), mixed strain+75%N (T8), UPMB10+100%N (T10), and UPMRB9+100%N (T11) with regards to the availability of phosphorus. In terms of soil K, no statistically significant changes were seen among the treatments UPMB10+50%N (T2), mixed strain +50%N (T4), UPMRB9 +75%N (T7), UPMB10 +100%N (T10), and UPMRB9+100%N (T11). The UPMB10+75%N treatment (T6) exhibited the greatest level of exchangeable Mg, which was statistically significant. The value of 1.57 cmolc/kg did not exhibit a statistically significant difference compared to the other treatments, except for the uninoculated control, which had the lowest value of 0.89 cmolc/kg.

TABLE 4. Means (±SE) of growth and yield- contributing parameters of sweet potato as affected by different treatments

Treatments	SPAD meter	Leaf number	Vine length (cm)	Root length (cm)	Leave area index (cm^2)	Root dry weight (g)	Shoot dry weight (g)	Root: Shoot ratio
T1	38.92 ± 1.04 abc	3 ± 0.7 d	77.8 ± 8.9 b	9.2 ± 0.6 de	344.15 ± 11.8 ab	22.4 ± 4.8 d	21.57 ± 1.4 b	1.0 ± 0.3 c
T2	28.24 ± 5.3 c	5 ± 0.2 abc	91.8 ± 19.7 ab	11.6 ± 0.3 bcd	225.39 ± 113 b	26.9 ± 0.6 cd	24.57 ± 2.1 ab	1.1 ± 0.08 c
T3	43.32 ± 0.5 a	6 ± 0.2 a	156.2 ± 10.5 a	15.4 ± 0.4 a	408.24 ± 34.3 a	43.6 ± 5.7 a	29.38 ± 0.4 a	2.2 ± 0.3 ab
T4	40.72 ± 0 .9 abc	5 ± 0.2 abcd	124.6 ± 8.4 ab	11.1 ± 0.4 cde	362.03 ± 3.6 ab	33.7 ± 8.5 bcd	22.47 ± 1.2 ab	1.5 ± 0.5 bc
T5	42.41 ± 1.5 ab	3 ± 0.7 cd	95.9 ± 9.6 ab	11.1 ± 0.8 cde	330.34 ± 17.5 ab	29.8 ± 0.6 bc	21.76 ± 0.4 b	0.9 ± 0.04 c
T6	40.14 ± 0.7 abc	6 ± 0.2 a	154.3 ± 17.2 a	13.5 ± 0.2 ab	340.45 ± 17.9 ab	41.8 ± 1.1 b	27.52 ± 0.3 ab	2.7 ± 0.1 a
T7	28.20 ± 5.3 c	5 ± 0.2 ab	90.4 ± 22.2 ab	10.1 ± 0.5 de	316.34 ± 1.4 ab	26.8 ± 6.6 cd	23.85 ± 2.2 ab	1.1 ± 0.3 bc
T8	40.64 ± 1.1 abc	5 ± 0.3 ab	117.5 ± 13.5 ab	10.6 ± 0.8 cde	339.74 ± 30.2 ab	39.7 ± 8.2 ab	23.09 ± 1.3 ab	1.7 ± 0.2 abc
T9	41.80 ± 1.18 ab	4 ± 0.8 bcd	105.5 ± 10.3 ab	10.7 ± 0.5 cde	314.05 ± 1.5 ab	32.6 ± 4.7 bcd	21.14 ± 0.4 b	1.5 ± 0.3 bc
T10	29.44 ± 5.6 bc	5 ± 0.3 ab	145.2 ± 13.9 ab	9.8 ± 1.09 de	337.83 ± 13.2 ab	39.3 ± 0.4 ab	22.14 ± 1.8 ab	0.8 ± 0.05 c
T11	36.34 ± 1.8 abc	5 ± 0.2 ab	136.5 ± 12.1 ab	10.6 ± 0.3 cde	366.40 ± 53.9 ab	35.4 ± 4.1 bc	12.42 ± 1.3 ab	1.1 ± 0.2 c
T12	43.24 ± 1.1 a	5 ± 0.1 ab	89.2 ± 21.9 ab	12.5 ± 0.8 bc	234.08 ± 117 b	35.8 ± 9.5 bc	23.19 ± 1.7 ab	1.5 ± 0.3 bc

Means within the same column followed by the same letter are not significantly different at $p \leq 0.05$ using Least Significant Difference (LSD test). The columns represent the mean values ± standard error

The higher soil pH in T3 and T6, as shown in Table 5, indicated that PGPR-HA inoculations were more efficient in increasing the soil pH than uninoculated treatments. This could be attributed to the higher basic cations of the treatments because of the PGPR-HA inoculations, which were primarily responsible for the neutralization of the soil acidity (Ashwini et al. 2022; *Çığ* et al. 2021). The pH of the soil and plant exudation may affect the ability of PGPR to colonize plant roots in the rhizosphere. Humic acid has a favorable impact on the soil and crop by neutralizing the pH of acidic soil, which increases the soil's capacity as a buffer, reducing the impact of acidic precipitation on soil reactions. In addition, humic acids promote soil life and root growth and accelerate the pace of nitrification (Yang et al. 2021). Based on a scholarly investigation, it has been shown that the utilization PGPR may effectively enhance the accessibility of essential nutrients, including P, inside the soil. Certain PGPRs possess the capacity to solubilize phosphate within the soil matrix, leading to an augmented abundance of phosphate ions in the soil that may be readily accessed by plants (Prasad et al. 2015; Sharma et al. 2017). One potential explanation for the observed increase in accessible soil P in the soils treated with PGPR-HA compared to the uninoculated treatments might be attributed to this factor. Applying humic acid amendments with PGPR strains positively influenced the soil K contents (Table 5). The elevated K concentration in the soil may be attributed to the same underlying factor that has caused the increase in P levels in the soil.

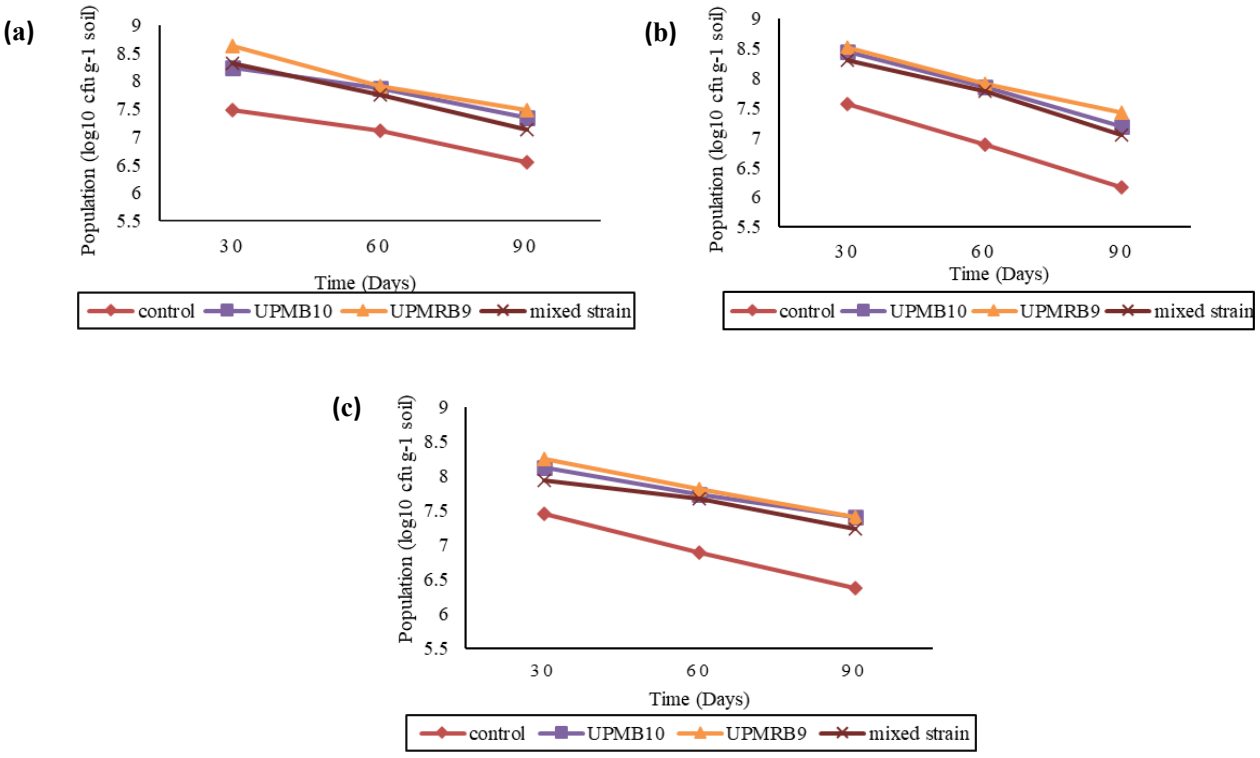

FIGURE 2. Means (±SE) of soil bacterial population at different growth stages of sweet potato as affected by PGPR-HA inoculation with (a) 50% N, (b) 75% N, and (c) 100% N recommended rate

Our result aligned with Abdelrahman et al. (2021), who found that soil inoculation with PGPR strains combined with NPK and humic acid spraying gave the highest records of all nutrient activity. The addition of PGPR-HA treatments had increased Ca and Mg concentrations in soils, particularly in the T3 treatments, due to both the PGPR-HA and the 50% N fertilizer rate. This result was similar to the findings of Ekin (2020), who found that sunflower plant inoculation with PGPR strain (*B. subtilis*) at different humic acid doses significantly increased Ca and Mg accumulation in seed by 62.9% and 81.1%, respectively, compared with untreated plants. Ashwini et al. (2022) reported that humic acid is the principal component of soil organic matter, and it affects soil factors such as nutrient solubility by creating complex forms with humic materials' chemical components. The potential positive effects of adding HA to bacteria can be better understood by examining its role as a catalyst for enhancing microbial growth and activity. Incorporating PGPR-HA formulations with nitrogen fertilizer resulted in an augmentation of mineral nutrient levels within the soil and a subsequent elevation in pH values.

EFFECT OF TREATMENTS ON THE NUTRIENT CONTENTS OF SWEET POTATO LEAF

The nutrient contents (N, P, K, Ca, and Mg) in the leaf of sweet potato plants were also significantly increased ($p \leq 0.05$) by the application of PGPR-HA inoculation and the addition of different N-fertilizer rates (Table 6). The shoot's highest N, P, and K values of 2.88, 1.40, and 1.40%, respectively, were observed in UPMRB9+50% N (T3). However, the highest Ca was found in UPMB10+50% (T2)-treated plants with 0.67%. T3 did not differ significantly from UPMB10+75% (T6) and mixed strain +75% (T8) in terms of leaf K contents, while the uninoculated treatments had the lowest leaf nutrient contents as compared with the PGPR-HA-inoculated plants.

Higher plant nutrient contents in T3 and T6 were due to the presence of the beneficial microbes that are known to fix N, solubilize P and K, and produce IAA. Incorporating reduced nitrogen fertilizer rates in the experimental treatments contributes to the augmentation of the potential of the PGPR inoculations, thereby improving the nutritional composition of the plants. Savarese et al. (2022) have reported that the augmentation of PGPR with humic acid as an amendment can enhance plant physiological processes by enhancing the accessibility of macro- and micronutrients. Furthermore, the incorporation of humic acid amendment in conjunction with bio-inoculants, namely PGPR, was shown to result in a significant increase in the levels of N, P and K in the leaves (Ashwini et al. 2022). PGPR is an environmentally acceptable alternative technology to promote crop growth and agricultural productivity. PGPR application improved onion mineral nutrition, which led to the maximum mineral content in the leaves and bulb (Gupta et al. 2021; Laftah & Alabdulla, 2022).

TABLE 5. Means (±SE) of soil nutrients after harvest as affected by different treatments

Treatments	PH	N (%)	P Mg/ kg	K cmol /kg	Ca cmol/kg	Mg cmol/kg
T1	5.33 ± 0.02 c	0.19 ± 0.01 ab	5.94 ± 0.3 d	0.50 ± 0.01 d	2.87 ± 0.009 ab	0.89 ± 0.09 b
T2	6.48 ± 0.04 ab	0.21 ± 0.00 ab	10.01 ± 0.5 b	0.64 ± 0.02 abc	3.22 ± 0.1 ab	1.33 ± 0.3 ab
T3	6.59 ± 0.02 a	0.26 ± 0.004 a	14.49 ± 0.2 a	0.72 ± 0.02 a	3.78 ± 0.2 a	1.52 ± 0.2 ab
T4	6.52 ± 0.00 ab	0.21 ± 0.004 ab	8.84 ± 0.5 bc	0.62 ± 0.01 abc	2.77 ± 0.05 ab	1.02 ± 0.01 ab
T5	5.40 ± 0.02 c	0.21 ± 0.004 ab	6.12 ± 0.2 d	0.56 ± 0.02 dc	2.87 ± 0.009 ab	1.02 ± 0.04 ab
T6	6.59 ± 0.01 a	0.24 ± 0.004 ab	14.79 ± 0.3 a	0.68 ± 0.01 ab	3.38 ± 0.08 ab	1.57 ± 0.3 a
T7	6.54 ± 0.01 ab	0.20 ± 0.004 ab	10.12 ± 0.8 b	0.65 ± 0.01 abc	2.80 ± 0.1 ab	1.40 ± 0.2 ab
T8	6.35 ± 0.08 ab	0.18 ± 0.02 b	9.54 ± 0.1 bc	0.60 ± 0.02 bc	2.81 ± 0.2 ab	1.03 ± 0.08 ab
T9	5.38 ± 0.01 c	0.22 ± 0.004 ab	5.36 ± 0.2 d	0.59 ± 0.02 dcb	2.60 ± 0.3 b	1.22 ± 0.3 ab
T10	6.42 ± 0.09 ab	0.21 ± 0.03 ab	9.14 ± 0.4 bc	0.63 ± 0.007 abc	2.79 ± 0.2 ab	1.07 ± 0.1 ab
T11	6.50 ± 0.02 ab	0.22 ± 0.008 ab	9.14 ± 0.3 bc	0.63 ± 0.009 abc	2.80 ± 0.1 ab	1.35 ± 0.3 ab
T12	6.46 ± 0.01 ab	0.23 ± 0.008 ab	8.66 ± 0.5 c	0.62 ± 0.01 bc	2.56 ± 0.1 b	1.11 ± 0.008 ab
p-value	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001

Means within the same column followed by the same letter are not significantly different at $p \leq 0.05$ using Least Significant Difference (LSD test). The columns represent the mean values ± standard error

TABLE 6. Means (±SE) of leaf nutrient contents as affected by different treatments

Treatments	N (%)	P (%)	K (%)	Ca (%)	Mg (%)
T1	0.98 ± 0.1 c	0.88 ± 0.01 d	1.68 ± 0.02 d	0.50 ± 0.05 bc	0.88 ± 0.1 d
T2	1.67 ± 0.08 bc	1.27 ± 0.1 ab	1.93 ± 0.01 bcd	0.67 ± 0.08 a	1.27 ± 0.1 ab
T3	2.88 ± 0.09 a	1.40 ± 0.06 a	2.34 ± 0.1 a	0.58 ± 0.008 ab	1.40 ± 0.06 a
T4	1.28 ± 0.1 c	1.07 ± 0.03 bcd	1.98 ± 0.003 bc	0.40 ± 0.004 c	1.07 ± 0.03 bcd
T5	1.23 ± 0.1 c	1.0 ± 0.1 cd	1.76 ± 0.01 cd	0.48 ± 0.06 bc	1.00 ± 0.1 cd
T6	2.20 ± 0.06 ab	1.21 ± 0.1 abc	2.29 ± 0.01 a	0.58 ± 0.02 ab	1.21 ± 0.1 abc
T7	1.75 ± 0.07 bc	1.17 ± 0.02 abc	2.00 ± 0.01 bc	0.52 ± 0.04 abc	1.17 ± 0.02 abc
T8	1.10 ± 0.04 c	1.28 ± 0.1 ab	2.40 ± 0.2 a	0.55 ± 0.05 abc	1.28 ± 0.1 ab
T9	1.04 ± 0.2 c	1.21 ± 0.1 abc	1.88 ± 0.01 cd	0.50 ± 0.09 bc	1.21 ± 0.1 abc
T10	1.62 ± 0.3 bc	1.24 ± 0.06 abc	2.01 ± 0.01 bc	0.55 ± 0.01 abc	1.24 ± 0.06 abc
T11	1.41 ± 0.1 bc	1.27 ± 0.1 ab	2.15 ± 0.01 ab	0.60 ± 0.02 ab	1.27 ± 0.1 ab
T12	1.30 ± 0.1 c	1.12 ± 0.1 bcd	2.02 ± 0.06 bc	0.62 ± 0.04 ab	1.12 ± 0.1 bcd
p-value	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001

Means within the same column followed by the same letter are not significantly different at $p \leq 0.05$ using Least Significant Difference (LSD test). The columns represent the mean values ± standard error

Conclusions

Based on this study, the inoculations of PGPR-HA (*B. tequilensis* and *B. subtilis* strains) and the optimum nitrogen fertilization (50 and 75%) significantly enhanced the availability of N, P, and K in the soil and plant tissue, higher plant physiological parameters, and higher soil microbial population which ultimately translated into the higher growth and yield of the sweet potato plant. The higher measured parameters were achieved by using the amendment of humic acid along with the inoculations, which is attributed to the synergetic effect of their combined application rather than using either alone as indicated in their sole application. Hence, these PGPR and humic acid combinations have the potential to be used as a biofertilizer for sweet potato plant production and to sustain soil health in sustainable agriculture practices.

Acknowledgements

The authors sincerely acknowledge the research grant (6300406) by a private Malaysian company (Hj Mat Hj Jantan Pvt. Ltd).

References

Abdelrahman, H.M., Zaghloul, R.A., Hassan, E.A., El-Zehery, H.R.A. & Salem, A.A. 2021. New strains of plant growth-promoting rhizobacteria in combination with humic acid to enhance squash growth under saline stress. *Egyptian Journal of Soil Science* 61(1): 129-146.

Agbede, T.M. & Oyewumi, A. 2022. Benefits of biochar, poultry manure and biochar–poultry manure for improvement of soil properties and sweet potato productivity in degraded tropical agricultural soils. *Resources, Environment and Sustainability* **7:** 100051. https://doi.org/10.1016/j.resenv.2022.100051

Ahmad, S., Daur, I., Al-Solaimani, S.G., Mahmood, S., Bakhashwain, A.A., Madkour, M.H. & Yasir, M. 2016. Effect of rhizobacteria inoculation and humic acid application on canola *(Brassica napus L.*) crop. *Pakistan Journal of Botany* 48: 2109-2120.

Al-Taey, D.K., Al-Shareefi, M.J., Mijwel, A.K., Al-Tawaha, A.R. & Al-Tawaha, A.R. 2019. The beneficial effects of biofertilizers combinations and humic acid on growth, yield parameters and nitrogen content of broccoli grown under drip irrigation system. *Bulgarian Journal of Agricultural Science* 25(5): 959-966.

Ashwini, N., Kumar, P., Joshi, A.K., Sharma, N.C., Sharma, N. & Sharma, N. 2022. Synergistic action of humic acid substances and bio-inoculants in guava (*Psidium guajava* L.): Impact on growth traits, fruiting, nutrient profiling and rhizosphere stochiometry in meadow rainy season plant-soil interface. *Journal of Plant Nutrition* 46(4): 574-588.

Baard, V., Bakare, O.O., Daniel, A.I., Nkomo, M., Gokul, A., Keyster, M. & Klein, A. 2023. Biocontrol potential of *Bacillus subtilis* and *Bacillus tequilensis* against four fusarium species. *Pathogens* 12(2): 254.

Bashan, Y., de-Bashan, L.E., Prabhu, S.R. & Hernandez, J.P. 2014. Advances in plant growth-promoting bacterial inoculant technology: Formulations and practical perspectives (1998-2013). *Plant and Soil* 378: 1-33.

Bahadur, I., Maurya, B.R., Meena, V.S., Saha, M., Kumar, A. & Aeron, A. 2017. Mineral release dynamics of tricalcium phosphate and waste muscovite by mineral-solubilizing rhizobacteria isolated from indo-gangetic plain of India. *Geomicrobiology Journal* 34(5): 454-466.

Benz, M., Schink, B. & Brune, A. 1998. Humic acid reduction by *Propionibacterium freudenreichii* and other fermenting bacteria. *Applied and Environmental Microbiology* 64: 4507-4512.

Bowman, D.C., Paul, J.L. & Carlson, R.M. 1988. A method to exclude nitrate from Kjeldahl digestion of plant tissues. *Communications in Soil Science and Plant Analysis* 19: 205-213.

Chen, X., Kou, M., Tang, Z., Zhang, A. & Li, H. 2017. The use of humic acid urea fertilizer for increasing yield and utilization of nitrogen in sweet potato. *Plant, Soil and Environment* 63(5): 201-206.

Çığ, F., Sönmez, F., Nadeem, M.A. & Sabagh, A.E. 2021. Effect of biochar and PGPR on the growth and nutrients content of einkorn wheat (*Triticum monococcum* L.) and post-harvest soil properties. *Agronomy* 11: 2418.

Egamberdiyeva, D. & Höflich, G. 2004. Effect of plant growth-promoting bacteria on growth and nutrient uptake of cotton and pea in a semi-arid region of Uzbekistan. *Journal of Arid Environments* 56(2): 293-301.

Ekin, Z. 2020. Co-application of humic acid and bacillus strains enhances seed and oil yields by mediating nutrient acquisition of safflower (*Carthamus tinctorius* L.) plants in a semi-arid region. *Applied Ecology and Environmental Research* 18: 1883-1900.

Ekin, Z. 2019. Integrated use of humic acid and plant growth promoting rhizobacteria to ensure higher potato productivity in sustainable agriculture. *Sustainability* 11(12): 3417.

El-Sawah, A.M., El-Keblawy, A., Ali, D.F.I., Ibrahim, H.M., El-Sheikh, M.A., Sharma, A. & Sheteiwy, M.S. 2021. Arbuscular *mycorrhizal* fungi and plant growth-promoting rhizobacteria enhance soil key enzymes, plant growth, seed yield, and qualitative attributes of guar. *Agriculture* 11: 194.

Etesami, H., Alikhani, H.A. & Mirseyed Hosseini, H. 2015. Indole-3-acetic acid and 1-aminocyclopropane-1-carboxylate deaminase: Bacterial traits required in rhizosphere, rhizoplane and/or endophytic competence by beneficial bacteria. *Bacterial Metabolites in Sustainable Agroecosystem*. *Sustainable Development and Biodiversity, vol 12*, edited by Maheshwari, D. Springer, Cham. pp. 183-258.

Folina, A., Tataridas, A., Mavroeidis, A., Kousta, A., Katsenios, N. & Efthimiadou, A. 2021. Evaluation of various nitrogen indices in N-Fertilizers with inhibitors in field crops: A review. *Agronomy* 11: 418.

Goswami, D., Thakker, J.N. & Dhandhukia, P.C. 2016. Portraying mechanics of plant growth promoting rhizobacteria (PGPR): A review. *Cogent Food & Agriculture* 2(1): 1127500.

Gupta, S., Stirk, W.A., Plačková, L., Kulkarni, M.G., Doležal, K. & Van Staden, J. 2021. Interactive effects of plant growth-promoting rhizobacteria and a seaweed extract on the growth and physiology of *Allium cepa* L. (onion). *Journal of Plant Physiology* 262: 153437.

Huang, X.F., Chaparro, J.M., Reardon, K.F., Zhang, R., Shen, Q. & Vivanco, J.M. 2014. Rhizosphere interactions: Root exudates, microbes, and microbial communities. *Botany* 92(4): 267-275.

Itelima, J.U., Bang, W.J., Onyimba, I.A., Sila, M.D. & Egbere, O.J. 2018. Bio-fertilizers as key player in enhancing soil fertility and crop productivity: A review. *Direct Research Journal of Agriculture and Food Science* 6(3): 73-83.

Kapadia, C., Sayyed, R.Z., El Enshasy, H.A., Vaidya, H., Sharma, D., Patel, N. & Zuan, A.T.K. 2021. Halotolerant microbial consortia for sustainable mitigation of salinity stress, growth promotion, and mineral uptake in tomato plants and soil nutrient enrichment. *Sustainability* 13: 8369.

Karim, N.A., Devarajan, T. & Ahmad, A. 2022. Principal component analysis for phenotypic characterization of sweet potato (*Ipomoea batatas* (L.) Lam.) genotypes in Malaysia. *Trends in Sciences* 19: 4612.

Kashiani, P. 2012. Genetic potential of selected sweet corn inbred lines and analysis of their combining ability assisted by microsatellite DNA markers. Doctoral dissertation. Universiti Putra Malaysia (Unpublished).

Kamali, S.R., Tsai, C.H. & Chen, C.N. 2021. Comparison of three digestion methods for determination of selenium in green tea samples using fluorescence spectrometry. *IOP Conference Series: Earth and Environmental Science* 712(1): 012016. doi:10.1088/1755-1315/712/1/012016

Kaur, T., Devi, R., Kumar, S., Sheikh, I., Kour, D. & Yadav, A.N. 2022. Microbial consortium with nitrogen fixing and mineral solubilizing attributes for growth of barley (*Hordeum vulgare* L.). *Heliyon* **8**(4): e09326.

Khan, Z., Rahman, M.H.U., Haider, G., Amir, R., Ikram, R.M., Ahmad, S., Schofield, H.K., Riaz, B., Iqbal, R., Fahad, S., Datta, R., Baazeem, A., El Sabagh, A. & Danish, S. 2021. Chemical and biological enhancement effects of biochar on wheat growth and yield under arid field conditions. *Sustainability* 13(11): 5890.

Laftah, S.K. & Alabdulla, S.A. 2022. Response of some growth traits of safflower (*Carthamus tinctorius* L.) to spray with humic acid under levels of phosphorus fertilizer. *IOP Conference Series: Earth and Environmental Science* 1060(1): 012115.

Mabrouk, Y., Hemissi, I., Salem, I.B., Mejri, S., Saidi, M. & Belhadj, O. 2018. Potential of rhizobia in improving nitrogen fixation and yields of legumes. *Symbiosis* 107: 73495.

Mahapatra, S., Yadav, R. & Ramakrishna, W. 2022. *Bacillus subtilis* impact on plant growth, soil health and environment: Dr. Jekyll and Mr. Hyde. *Journal of Applied Microbiology* 132(5): 3543-3562.

Manna, M.C., Rahman, M.M., Naidu, R., Bari, A.F., Singh, A.B., Thakur, J.K. & Subbarao, A. 2021. Organic farming: A prospect for food, environment and livelihood security in Indian agriculture. *Advances in Agronomy* 170: 101-153.

Meena, K., Ram, R.B., Meena, M.L., Meena, J.K. & Meena, D.C. 2017. Effect of organic manures and bio-fertilizers on growth, yield and quality of broccoli (*Brassica oleracea* var.) cv. KTS-1. *Chemical Science Review and Letters* 6: 2153-2158.

Meng, F., Huang, Q., Yuan, G., Cai, Y. & Han, F.X. 2021. The beneficial applications of humic substances in agriculture and soil environments. In *New Trends in Removal of Heavy Metals from Industrial Wastewater*, edited by Shah, M.P., Couto, S.R. & Kumar, V. Elsevier. pp. 131-160.

Moreira, B.C., Prates Júnior, P., Dell, B. & Kasuya, M.C.M. 2022. Roots and beneficial interactions with soil microbes. In *Subsoil Constraints for Crop Production*, edited by Oliveira, T.S.D. & Bell, R.W. Springer, Cham. pp. 263-287.

Mosharrof, M., Uddin, M., Sulaiman, M.F., Mia, S., Shamsuzzaman, S.M. & Haque, A.N.A. 2021. Combined application of biochar and lime increases maize yield and accelerates carbon loss from an acidic soil. *Agronomy* 11(7): 1313.

Muhammad, R., Nor, N.M., Ikram, E.H.K. & Sharif, M.S.M. 2022. Sustainability of underutilized local crop through product development of Malaysia purple sweet potato cracker. *Malaysian Journal of Medicine & Health Science* 18: 305-314.

Neela, S. & Fanta, S.W. 2019. Review on nutritional composition of orange-fleshed sweet potato and its role in management of vitamin A deficiency. *Food Science & Nutrition* **7**: 1920-1945.

Prasad, R., Kumar, M. & Varma, A. 2015. Role of PGPR in soil fertility and plant health. In *Plant-Growth-Promoting Rhizobacteria (PGPR) and Medicinal Plants*, edited by Egamberdieva, D., Shrivastava, S. & Varma, A. Soil Biology, vol. 42. Springer, Cham. pp. 247-260.

Pandita, D. 2022. Plant growth promoting bacteria (PGPB): Applications and challenges in bioremediation of metal and metalloid contaminated soils. In *Metals Metalloids Soil Plant Water Systems,* edited by Aftab, T. & Hakeem, K. Massachusetts: Academic Press. pp. 485-500.

Sadeq, B.M., Zuan, K., Tan, A., Kasim, S., Yun, W.M., Othman, N.M.I., Alkooranee, J.T., Chompa, S.S., Akter, A. & Rahman, M.E. 2023. Humic acid-amended formulation improves shelf-life of plant growth-promoting rhizobacteria (PGPR) under laboratory conditions. *Pertanika Journal of Science & Technology* 31(3): 1137-1155.

Sharma, I.P., Chandra, S., Kumar, N. & Chandra, D. 2017. PGPR: Heart of soil and their role in soil fertility. In *Agriculturally Important Microbes for Sustainable Agriculture,* edited by Meena, V., Mishra, P., Bisht, J. & Pattanayak, A. Singapore: Springer. pp. 51-67.

Savarese, C., Cozzolino, V., Verrillo, M., Vinci, G., De Martino, A., Scopa, A. & Piccolo, A. 2022. Combination of humic biostimulants with a microbial inoculum improves lettuce productivity, nutrient uptake, and primary and secondary metabolism. *Plant and Soil* 481: 285-314.

Shankar, A.B. & Kaushik, P. 2022. Visiting sweet potato from a breeding perspective: An update. *PrePrints* 2020: 2022040149.

Tang, C., Cheng, K., Liu, B., Antonietti, M. & Yang, F. 2022. Artificial humic acid facilitates biological carbon sequestration under freezing-thawing conditions. *Science of The Total Environment* 849: 157841.

Thomas, G.W. 1983. Chapter 9. Exchangeable cations. In *Methods of Soil Analysis: Part 2 Chemical and Microbiological Properties*, edited by Page, A.L. Madison: American Society of Agronomy, Soil Science Society of America. pp. 159-165.

Wang, D., Chen, X., Tang, Z., Liu, M., Jin, R., Zhang, A. & Zhao, P. 2022. Application of humic acid compound fertilizer for increasing sweet potato yield and improving the soil fertility. *Journal of Plant Nutrition* 45(13): 1933-1941.

Wu, Y., Li, S. & Chen, G. 2020. Impact of humic acids on phosphorus retention and transport. *Journal of Soil Science and Plant Nutrition* 20: 2431-2439.

Yang, F., Tang, C. & Antonietti, M. 2021. Natural and artificial humic substances to manage minerals, ions, water, and soil microorganisms. *Chemical Society Reviews* 50(10): 6221-6239.

Yasmin, F., Othman, R. & Maziz, M.N.H. 2020. Yield and nutrient content of sweet potato in response of plant growth-promoting rhizobacteria (PGPR) inoculation and N fertilization. *Jordan Journal of Biological Science Jordan* 13: 117-122.

Yuan, Z., Cao, Q., Zhang, K., Ata-Ul-Karim, S.T., Tian, Y., Zhu, Y. & Liu, X. 2016. Optimal leaf positions for SPAD meter measurement in rice. *Frontiers in Plant Science* 7: 719.

Zeng, Q., Ding, X., Wang, J., Han, X., Iqbal, H. & Bilal, M. 2022. Insight into soil nitrogen and phosphorus availability and agricultural sustainability by plant growth-promoting rhizobacteria. *Environmental Science and Pollution Research* 29: 45089-45106.

Zhao, C., Liu, G., Chen, Y., Jiang, Y., Shi, Y., Zhao, L., Liao, P., Wang, W., Xu, K., Dai, Q. & Huo, Z. 2022. Excessive nitrogen application leads to lower rice yield and grain quality by inhibiting the grain filling of inferior grains. *Agriculture* 12(7): 962.

Zhu, F. & Sun, J. 2019. Physicochemical and sensory properties of steamed bread fortified with purple sweet potato flour. *Food Bioscience* 30: 100411.

Zilaie, M.N., Arani, A.M., Etesami, H., Dinarvand, M. & Dolati, A. 2022. Halotolerant plant growth-promoting rhizobacteria-mediated alleviation of salinity and dust stress and improvement of forage yield in the desert halophyte *Seidlitzia rosmarinus*. *Environmental and Experimental Botany* 201: 104952.

*Corresponding author; email: tkz@upm.edu.my