

Using *Streptomyces* spp. as Plant Growth-Promoting Inoculants for Growth of Napier Grass under Low Water System

Waraporn Chouychai¹, Aphidech Sangdee², Areeya Phunee², Phakamas Senarit² and Khanitta Somtrakoon^{2,3*}

¹Biology Program, Department of Science, Faculty of Science and Technology, Nakhonsawan Rajabhat University, Nakhon Sawan 60000, Thailand

²Department of Biology, Faculty of Science, Mahasarakham University, Kantharawichai, Maha Sarakham 44150, Thailand

³Digital Innovation Research Cluster for Integrated Disaster Management in the Watershed, Mahasarakham University, Kantharawichai, Maha Sarakham 44150, Thailand

ABSTRACT

Napier grass can be used as feed for livestock and possibly for bioenergy production. However, the stimulation of the growth of Napier grass by plant growth-promoting bacteria (PGPB) has been rarely found. Thus, this study was performed to investigate the ability of *Streptomyces* spp. PB5, SRF1, St8, STRM104, and STRM302 to support the growth of Napier grass (*Pennisetum purpureum* × *Pennisetum americanum* cultivar Pak Chong 1) under a low water system. Among the five bacterial isolates, *Streptomyces* sp. St8 was the most suitable bacterial inoculant to stimulate the growth of plants grown under a low water system. Napier grass grew under a low water system and inoculated with *Streptomyces* sp. St8 had the highest shoot and root weight compared to the other inoculated isolates. The shoot and root fresh weights of plants grown under a low water system were 21.3 ± 1.53 g and 4.29 ± 0.77 g when inoculated with *Streptomyces* sp. St8. Moreover, *Streptomyces* sp. St8 also stimulated the growth of plants grown under a normal water system: the highest shoot

length (61.3 ± 5.67 cm), shoot fresh weight (26.9 ± 4.07 g), and root fresh weight (4.84 ± 0.54 g) were found in plants inoculated with this bacterial isolate. Furthermore, the plant's root-to-shoot ratios grown under a low water system were inoculated with each isolate of *Streptomyces* sp. (PB5, SRF1, St8, STRM104, and STRM302) were lower than for plants grown in the control pots. It means that bacterial inoculation under a low water

ARTICLE INFO

Article history:

Received: 05 January 2022

Accepted: 15 March 2022

Published: 11 April 2022

DOI: <https://doi.org/10.47836/pjtas.45.2.10>

E-mail addresses:

waraporn.c@nsru.ac.th (Waraporn Chouychai)

aphidech.s@msu.ac.th (Aphidech Sangdee)

61010210030@msu.ac.th (Areeya Phunee)

61010210018@msu.ac.th (Phakamas Senarit)

khanitta.s@msu.ac.th (Khanitta Somtrakoon)

* Corresponding author

system could protect the efficiency of roots from producing shoot biomass in the plants. Based on the results found in this study, *Streptomyces* sp. St8, a microbial inoculant, can be used with Napier grass cropping to produce feed for livestock or bioenergy production.

Keywords: Low water, Napier grass, plant growth-promoting bacteria, *Streptomyces*

INTRODUCTION

Napier grass is a fast-growing perennial grass usually found in humid soils in areas with over 1,000 mm of rainfall per year. Napier grass is a stress-tolerant forage crop, including plant disease and short drought stresses, and it can grow under low fertility (Negawo et al., 2017; Odiyi & Oludare, 2015). In Thailand, it is mainly used to feed livestock, and it is expected to be used for other purposes, including as a substrate for bioenergy production and biomass for electricity generation (Nantasaksiri et al., 2021; Osman et al., 2020; Waramit & Chaugool, 2014). Some genotypes of Napier grass can generate large biomass and accumulate nitrogen derived from biological nitrogen fixation when grown under low levels of nitrogen in the soil (Videira et al., 2012). Information about the possibility of using Napier grass as a resource for bioenergy production in Thailand is required in numerous areas for plantations. Moreover, biomass production from Napier grass for bioenergy production cannot compete with food or forage crop production for arable land. Thus, bioenergy

crops should be grown on non-fertile soils, which are not appropriate for other economic crops (Mei et al., 2021). Using plant growth-promoting bacteria (PGPB) is one way to improve plant growth and yield under unfavorable conditions. The application of PGPB to stimulate the growth of Napier grass has been rarely found, even though several PGPB have been isolated from Napier grass, including diazotrophic nitrogen-fixing bacteria belonging to the genera *Azospirillum* and *Gluconacetobacter* (Videira et al., 2012). PGPB from the genera *Bacillus*, *Enterobacter*, and *Sphingomonas* can solubilize insoluble phosphate, fix nitrogen, produce indole-3-acetic acid, ammonia, and siderophores have also been isolated from Napier grass, which could enhance salt tolerance in hybrid *Pennisetum* (Li et al., 2016).

The objective of this study was to investigate the ability of five isolates of *Streptomyces* spp. (PB5, SRF1, St8, STRM104, and STRM302) to stimulate the growth of Napier grass under low water conditions. The reason for using *Streptomyces* spp. as a model PGPB in this study was that many species had been shown to alleviate undesirable effects from drought stress on the plants in Gramineae. For example, *Streptomyces coelicolor* DE7, *Streptomyces olivaceus* DE10, and *Streptomyces geysiriensis* DE27 have been previously isolated from arid and drought-affected areas, and they could promote the growth of wheat cultivar WR-544 when grown in water-stress soil using the combined effects from phytohormone

production and water stress tolerance ability (Yandigeri et al., 2012). In addition, *Streptomyces pseudovenezuelae* MG547870 could produce indole-3-acetic acid and ACC deaminase, and it could increase the growth and alleviate severe effects from drought on maize (Chukwuneme et al., 2020). Moreover, *Streptomyces albidoflavus* OsiLf-2 increased the osmotic modification ability of rice grown under drought and salt stresses by increasing proline and sugar content in the plant (Niu et al., 2022). Even though the five isolates of *Streptomyces* spp. used in this study have never been tested to promote the growth of Napier grass previously, all isolates have plant growth-promoting activities. For example, *Streptomyces* sp. St8, STRM104, and STRM302 can solubilize phosphate and produce indole-3-acetic acid (Somtrakoon et al., 2019a, 2021). *Streptomyces* sp. SRF1 has only indole-3-acetic acid production activity (Somtrakoon et al., 2019a) during *Streptomyces* sp. PB5 has never been tested for plant growth-promoting activity, but it was tested in this study. Moreover, these five bacterial isolates have not been isolated from Napier grass. However, if they can stimulate the growth of Napier grass under low water, a biofertilizer from bacteria in this genus may be developed for Napier grass planting in the future.

MATERIALS AND METHODS

Plant Growth-Promoting Activity

Five isolates of *Streptomyces* spp., PB5, SRF1, St8, STRM104, and STRM302, were kindly provided by the Microbiology

and Applied Microbiology Research Unit, Faculty of Science, Mahasarakham University. Each *Streptomyces* sp. isolate was isolated from different agricultural areas in Thailand. *Streptomyces* sp. SRF1 (Sangdee et al., 2016) and PB5 were isolated from paddy field and integrated agricultural area in Lopburi and Buriram Provinces, respectively. *Streptomyces* sp. St8 was isolated from soil planted with a mango tree in Kalasin Province. *Streptomyces* sp. STRM104 and STRM302 were isolated from soil planted with tomatoes in Maha Sarakham Province. Each isolate of *Streptomyces* sp. was sub-cultured in half-strength potato dextrose agar (PDA) [potato dextrose broth powder (Himedia™, India) 12 g, agar powder (Difco, USA) 20 g, distilled water 1,000 ml, and the pH was adjusted to 7.0]. Then, the plant growth-promoting activities of *Streptomyces* sp. PB5 to solubilize phosphate, produce indole-3-acetic acid and ammonia were tested using the methods described in Ahmad et al. (2008), while the exopolysaccharide producing activity was tested using the methods described in Lakshminarayanan et al. (2015). Only the exopolysaccharide and ammonia-producing activities of *Streptomyces* sp. SRF1, St8, STRM104 and STRM302 were tested using the methods described in Lakshminarayanan et al. (2015) and Ahmad et al. (2018).

Preparation of Bacterial Culture

To prepare the bacterial inoculum used in the pot experiment, *Streptomyces* spp. PB5, SRF1, St8, STRM104, and STRM302

were grown in half-strength PDA, pH 7.0, and incubated at 37 °C for 14 days. Approximately 2–3 ml of 0.85% sodium chloride (NaCl) + 0.1% Tween 80 solution were poured onto the agar surface, and the cells and spores of each isolate of *Streptomyces* sp. were scraped with a loop and re-suspended in 0.85% NaCl + 0.1% Tween 80 solution (adapted from Somtrakoon et al., 2019b). A suspension of cells and spores was transferred into the culture tube, and the optical density was adjusted to be 0.5 at an optical wavelength at 600 nm. The initial cell number of each bacterial isolate of *Streptomyces* sp. from the culture suspension was serially diluted and counted on half-strength PDA by the drop plate method before use as an inoculum. The initial cell numbers of each isolate of *Streptomyces* sp. used to prepare the bacteria suspension in the pot experiment for the first and the second inoculations were recorded (Table 1).

Preparation of Soil

The soil used in this study was collected from wasteland in Khamriang Sub-district, Khantharawichai District, Maha Sarakham Province, Thailand. The soil was air-dried

for two weeks before use. After serial dilution, the total heterotrophic bacteria in the soil used in this study were counted on nutrient agar using the spread plate method. At the beginning of the experiment, the number of total heterotrophic bacteria was 5.3×10^4 CFU/g dry soil. Then, these soils were sub-divided into the experimental pots, with each experimental pot containing 4 kg of dry soil. There were 120 pots for the experiment.

Experimental Design

The ability of each isolate of *Streptomyces* sp. to stimulate the growth of Napier grass was determined in a 2 x 6 factorial, completely randomized design with ten replicates. Two factors were two levels of the water system (normal water and low water irrigation) x six levels of bacterial inoculation (non-inoculation and inoculation with PB5, SRF1, St8, STRM104, and STRM302). The details of each treatment are given in Table 2.

Pot Experiment

Stems of Napier grass cultivar ‘Pak Chong 1’ were cut into 13-14 cm pieces, with each piece having only one node and then soaked

Table 1
Initial cell numbers of *Streptomyces* spp. used in pot experiments

Bacterial isolates	1 st inoculation (CFU/ml) (14 days after transplantation)	2 nd inoculation (CFU/ml) (31 days after transplantation)
<i>Streptomyces</i> sp. PB5	8.7×10^{10}	8.7×10^{10}
<i>Streptomyces</i> sp. SRF1	2.5×10^{10}	1.9×10^{10}
<i>Streptomyces</i> sp. St8	3.5×10^8	3.3×10^8
<i>Streptomyces</i> sp. STRM104	1.0×10^{10}	9.3×10^9
<i>Streptomyces</i> sp. STRM302	4.3×10^9	4.3×10^9

Table 2
Details of each treatment

Treatment	Water system	<i>Streptomyces</i> isolate
1	Normal water	Non-inoculation
2	Normal water	PB5
3	Normal water	SRF1
4	Normal water	St8
5	Normal water	STRM104
6	Normal water	STRM302
7	Low water	Non-inoculation
8	Low water	PB5
9	Low water	SRF1
10	Low water	St8
11	Low water	STRM104
12	Low water	STRM302

in water for 72 hours. One cutting of Napier grass was planted in each experimental pot until the young plant was 14 days old. At this age, 2 ml of each bacterial inoculum (Table 1) was mixed with 250 ml of water and poured into the experimental pot. Pots that did not receive the bacterial inoculum had distilled water added as a non-inoculated control. The water system was set into two patterns; 250 ml of water was added to the experimental pot once every three days for the normal water system and once every six days for the low water system. The second bacterial inoculation was performed one month after planting. Again 2 ml of each bacterial inoculum (Table 1) was mixed with 250 ml of water and poured into the planted soil. Napier plants were grown until they were 49 days old when the experiment was terminated. Then, the physical and chemical characteristics of the soil in a low water system at the end of the experiment were analyzed at the Soil-Fertilizer-Environment Academic

Development Project, Department of Soil Science, Kasetsart University, Bangkok, Thailand.

Plant Growth Measurement

Plant growth parameters were determined at the end of the experiment, including shoot length, root length, shoot fresh weight, shoot dry weight, root fresh weight, root dry weight, and the number of leaves. Total chlorophyll, chlorophyll *a*, and chlorophyll *b* contents in leaves of Napier plants were determined according to the methods described in Huang et al. (2004). The relative water content (RWC) in the leaves of the Napier plants was analyzed according to the methods described in Sade et al. (2015). The specific root length was calculated from the root length/root dry weight (Calvelo Pereira et al., 2010). The root to shoot ratio was calculated from the root dry weight/shoot dry weight (Xu et al., 2018).

Statistical Analysis

A two-way analysis of variance (ANOVA) and least square difference (LSD) tests were used for variance analysis and pairwise comparison for plant growth. Microsoft Excel was used for statistical analysis.

RESULTS AND DISCUSSION

Relative Water Content and Chlorophyll Content in Leaves

The growth levels of Napier grass planted under normal and low water systems in this study were similar. This study did not change

Napier grass's growth under the low water system. The RWC confirmed it in Napier grass leaves that were not significantly different between normal and low water systems for the same bacterial isolate (Table 3). However, RWC in leaves differed between some inoculations within the same water system, for example, *Streptomyces* sp. St8 and STRM302 under the normal water system, and non-inoculation and *Streptomyces* sp. STRM104 under the low water system. Normally, the RWC in leaves

of plants decreases when encountering drought conditions (Machado & Paulsen, 2001). It may be due to Napier grass being tolerant to short droughts. It has been reported that Napier grass could survive under non-irrigated conditions and could generate higher biomass during the dry season than in the rainy season (Haegele et al., 2017).

Under the normal water system, inoculation of Napier grass with *Streptomyces* sp. isolates PB5, SRF1, St8, and STRM104

Table 3

Chlorophyll content and relative water content of Napier grass leave grown under normal system and low water condition for 49 days [mean ± standard error (SE)]

Treatment	Chlorophyll <i>a</i> (mg/ml)	Chlorophyll <i>b</i> (mg/ml)	Total chlorophyll (mg/ml)	RWC (%)
<u>Normal water system</u>				
Control	5.09 ± 1.02cA	6.81 ± 0.39cB	11.90 ± 1.42dB	78.2 ± 21.6abA
PB5	12.32 ± 1.13abA	9.84 ± 0.63bA	22.15 ± 0.50bA	58.5 ± 8.5abA
SRF1	10.00 ± 0.29bA	6.85 ± 0.07cA	16.85 ± 0.32cA	51.2 ± 13.6abA
St8	16.14 ± 2.08aA	16.35 ± 1.12aA	32.48 ± 1.14aA	85.9 ± 9.8aA
STRM104	11.80 ± 1.29bA	10.19 ± 0.38bB	21.98 ± 1.06bB	49.7 ± 19.2abA
STRM302	4.09 ± 1.12cA	6.20 ± 0.04cA	10.29 ± 1.09dA	20.7 ± 13.1bA
<u>Low water system</u>				
Control	10.85 ± 1.57abA	16.99 ± 0.97aA	27.83 ± 2.36aA	96.9 ± 11.5aA
PB5	14.87 ± 0.29aA	8.17 ± 0.21cA	23.03 ± 0.50bA	57.5 ± 28.8abA
SRF1	8.00 ± 0.53bA	6.81 ± 0.35cdA	14.81 ± 0.24cA	64.8 ± 14.1abA
St8	14.86 ± 3.23aA	13.19 ± 2.33bB	28.04 ± 1.43aB	76.5 ± 9.8abA
STRM104	14.52 ± 0.74aA	14.77 ± 0.20abA	29.29 ± 0.70aA	48.2 ± 22.5bA
STRM302	4.62 ± 0.48bA	5.04 ± 0.15dA	9.66 ± 0.55dA	59.1 ± 6.4abA
Water system	ns	**	**	ns
Bacteria	**	**	**	*
Water system x bacteria	ns	**	**	ns

Note. Different lower-case letters show significant differences between inoculations of bacterial isolates under the same water system ($P < 0.05$), and different capital letters show significant differences between normal system and low water system with the same bacterial isolate inoculations ($P < 0.05$). Abbreviations: ns, *, ** denote non-significance ($P > 0.05$), statistical significance ($P < 0.05$), and high statistical significance ($P < 0.01$) for each factor, respectively

increased the total chlorophyll content in the leaves of the plant when compared to the control pots (Table 5). The highest total chlorophyll content in the plant's leaves was observed in soil inoculated with St8. Under the low water system, inoculation of St8 and STRM104 could maintain the chlorophyll content in the leaves of Napier grass because the total chlorophyll content in the leaves of plant inoculation with *Streptomyces* sp. isolates St8 (28.04 ± 1.43 mg/ml) and STRM104 (29.29 ± 0.70 mg/ml) were not significantly different from the control pots (27.83 ± 2.36 mg/ml). However, the total chlorophyll content in the leaves of plants inoculated with *Streptomyces* sp. SRF1, STRM302, and PB5 were lower than the total chlorophyll content in the plant's leaves in the control pots (Table 3). Normally, drought stress decreases the chlorophyll content in plants (Chandra et al., 2018), but a decrease in the chlorophyll content in the low water system was only found in the leaves of plants inoculated with *Streptomyces* sp. St8. On the other hand, the chlorophyll content in the leaves of plants inoculated with *Streptomyces* sp. STRM104 and non-inoculated plants were increased in the low water system.

Shoot and Root Growth of Napier Grass

The leaf numbers of Napier grass grown under the normal water system were similar between the control pots and pots inoculated with each bacterial isolate. However, decreased leaf numbers were found in plants grown in the control pots under the low water system (Table 4). This phenomenon

is prominently found in plants grown under drought stress because decreasing the leaf number is one of the adaptation mechanisms in plants. In general, the plant responds to drought via many adaptations in the leaves to limit water loss, such as thickening the palisade parenchyma in the leaf, decreasing the leaf area, stomatal size, and leaf number (Deblonde & Ledent, 2001; Zhang et al., 2018). Surprisingly, using *Streptomyces* sp. PB5, St8, and STRM104 could increase the leaf number of plants grown under the low water system to be comparable to plants grown under the normal water system. It corresponds to the results of shoot growth because increasing shoot growth was also observed in the experimental pot inoculation with *Streptomyces* sp. PB5, St8, STRM104, and STRM302 under normal and low water systems (Table 4). Application of *Streptomyces* sp. St8 under both normal and low water systems tended to give the highest shoot fresh weight (26.9 ± 4.07 g and 21.3 ± 1.53 g) and shoot dry weight (3.60 ± 0.540 g and 2.84 ± 0.190 g) compared to the inoculation with the other bacterial isolates (Table 4 and Figure 1). Moreover, the highest root growth in fresh and dry weight was also observed in the experimental pots inoculated with *Streptomyces* sp. St8 under both normal and low water systems (Table 4). The root's fresh and dry weights were 4.29 ± 0.77 g and 0.62 ± 0.099 g when the soil was inoculated with *Streptomyces* sp. St8 under the low water system. However, *Streptomyces* sp. SRF1 was unsuitable as a microbial inoculant for Napier grass cultivation. This bacterial isolate stimulated the growth of

Table 4
Shoot and root growth of Napier grass grown under normal water and low water systems for 49 days [mean ± standard error (SE)]

	Leaf number	Shoot length (cm)	Shoot fresh weight (g)	Shoot dry weight (g)	Root length (cm)	Root fresh weight (g)	Root dry weight (g)	Root to shoot ratio	Specific root length (m/g)
<u>Normal water</u>									
Control	8.4 ± 0.40	41.0 ± 1.57bA	11.5 ± 1.24cA	1.25 ± 0.150bA	48.6 ± 5.60aA	2.49 ± 0.42bA	0.23 ± 0.042cA	0.19	2.09
PB5	8.5 ± 0.55	61.1 ± 6.39aA	20.2 ± 2.19bA	1.92 ± 0.328bA	45.2 ± 4.51aA	3.35 ± 0.60bA	0.48 ± 0.087bA	0.25	0.93
SRF1	8.0 ± 0.24	53.0 ± 2.34abA	15.7 ± 1.53bcA	1.99 ± 0.252bA	50.8 ± 3.70aA	3.62 ± 0.70abA	0.50 ± 0.100bA	0.21	1.02
St8	9.3 ± 0.42	61.3 ± 5.67aA	26.9 ± 4.07aA	3.60 ± 0.540aA	43.5 ± 4.35aA	4.84 ± 0.54aA	0.76 ± 0.119aA	0.24	0.57
STRM104	9.3 ± 0.37	56.8 ± 5.62aA	20.3 ± 2.09bA	2.63 ± 0.428abA	47.9 ± 3.64aA	2.79 ± 0.26bA	0.63 ± 0.150abA	0.20	0.76
STRM302	8.6 ± 0.50	57.3 ± 5.38aA	19.6 ± 1.63bA	2.59 ± 0.357abA	50.6 ± 2.11aA	3.80 ± 0.48abA	0.53 ± 0.070abA	0.25	0.95
<u>Low water</u>									
Control	6.9 ± 0.43	40.8 ± 3.57bcA	9.7 ± 1.44cA	1.17 ± 0.194bA	39.3 ± 3.12cA	2.67 ± 0.41bA	0.13 ± 0.049bA	0.27	1.24
PB5	9.0 ± 0.30	58.4 ± 4.09abA	16.0 ± 1.08bA	2.24 ± 0.165aA	45.2 ± 2.58bcA	2.29 ± 0.26bA	0.25 ± 0.036bA	0.11	1.78
SRF1	6.5 ± 0.58	36.8 ± 4.11cA	8.9 ± 1.34cB	1.17 ± 0.227bA	57.8 ± 6.44aA	2.00 ± 0.25bB	0.25 ± 0.036bB	0.22	2.27
St8	8.9 ± 0.23	59.0 ± 4.22abA	21.3 ± 1.53aB	2.84 ± 0.190aA	48.4 ± 1.71abA	4.29 ± 0.77aA	0.62 ± 0.099aA	0.24	0.71
STRM104	9.3 ± 0.17	66.5 ± 4.92aA	18.2 ± 1.13abA	2.23 ± 0.196aA	36.6 ± 3.53cA	2.45 ± 0.22bA	0.46 ± 0.065abA	0.21	0.79
STRM302	7.3 ± 0.59	52.2 ± 3.58bA	16.6 ± 1.36abA	2.16 ± 0.254aA	48.9 ± 2.90abA	1.84 ± 0.31bB	0.38 ± 0.056bA	0.12	1.28
Water		ns	**	ns	ns	*	*		
Bacteria		**	**	**	*	**	**		
Water x bacteria		ns	ns	ns	ns	ns	ns		

Note. Different lower-case letters show significant differences between inoculations of bacterial isolates under the same water system ($P < 0.05$), and different capital letters show significant differences between normal system and low water system with the same bacterial isolate inoculations ($P < 0.05$). The data were not normally distributed for leaf number, and the statistical calculation was not performed. Abbreviations: ns, *, **, ** denote non-significance ($P > 0.05$), statistical significance ($P < 0.05$), and high statistical significance ($P < 0.01$) of each factor, respectively

plants grown under both normal and low water systems to a lesser extent than the other isolates (Table 4 and Figure 1). It may be due to no phosphate solubilization activity detected in *Streptomyces* sp. SRF1 and only a slight level of indole-3-acetic acid were produced by this bacterial isolate (Somtrakoon et al., 2019a).

The stimulation of the growth of Napier grass in this study may be due to the plant growth-promoting activities of *Streptomyces*. Our previous work (Somtrakoon et al., 2019a, 2021), and

recent tests on plant growth-promoting activity, revealed that *Streptomyces* sp. St8, STRM104, STRM302, and PB5 can produce indole-3-acetic acid, exopolysaccharide, ammonia, and solubilize phosphate (Table 5). These activities assist in promoting the growth of plants by several mechanisms. For example, IAA production supports plant growth by increasing root growth, which permits the plant to uptake more soil nutrients (Goswami et al., 2013). In addition, increasing the soil water holding capacity by bacterial exopolysaccharides promotes plant

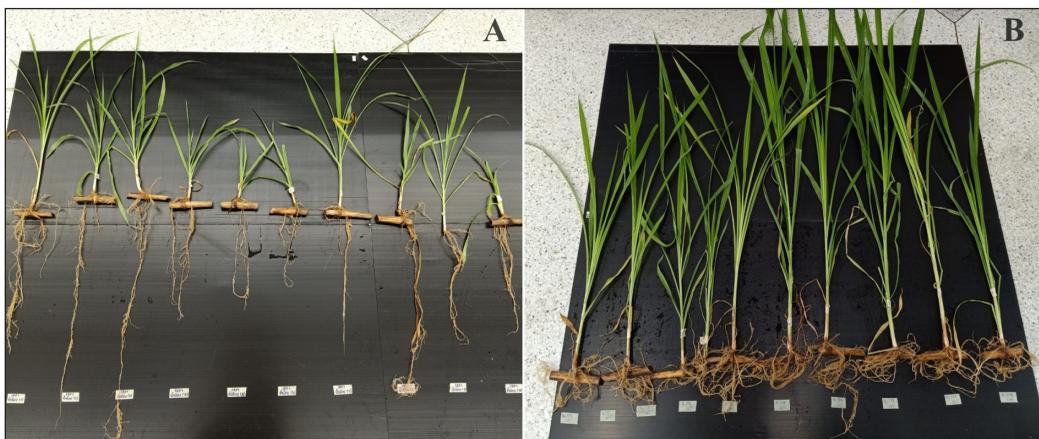


Figure 1. The 49-day-old Napier grass grown under a low water system when inoculated with *Streptomyces* sp. SRF1 (A) and St8 (B), respectively

Table 5
Plant growth-promoting activity of *Streptomyces* sp. PB5, SRF1, St8, STRM104, and STRM302

Bacteria	IAA production	Phosphate solubilization	Exopolysaccharide production	Ammonia Production
PB5	+	+	+	+
SRF1	ND ^A	ND ^A	+	+
St8	ND ^A	ND ^A	+	+
STRM104	ND ^B	ND ^B	+	+
STRM302	ND ^B	ND ^B	+	+

Note. ND^A mean not determined in this study. Plant growth-promoting activity was determined in Somtrakoon et al. (2019a); ND^B mean not determined in this study. Plant growth-promoting activity was determined in Somtrakoon et al. (2021); Symbols + and - indicate positive and negative activities, respectively

growth via increasing the nutrient uptake and aiding the colonization of PGPB to the plant root zone (A. Kumar et al., 2020; Khan et al., 2017). Bacterial colonization of plant roots is a significant procedure for PGPB to survive, grow, and function in the soil (de Souza et al., 2015). In addition, increasing phosphorus mobilization by PGPB could promote phosphorus uptake by plants and support plants grown in soil (Pereira et al., 2020). The ammonia-producing ability of PGPB also provides a nitrogen source for plants (Goswami et al., 2013), and it can act to protect the plants from phytopathogens (Fahsi et al., 2021).

In general, indigenous bacteria have been proposed to be used as microbial inoculants because of their adaptation capacity to the environment after inoculation into the environment again (B. L. Kumar & Gopal, 2015). However, the results of this study confirmed that the *Streptomyces* sp., which has not previously been isolated from soil planted with Napier grass, could promote the growth of plants to an obvious extent compared to the control. *Streptomyces* sp. St8 was the most suitable microbial inoculant for Napier grass planting based on the root to shoot ratio. It is confirmed by a similar root to shoot ratio of plant inoculation with *Streptomyces* sp. St8, which was similar between the normal and low water system conditions. It means that growing under a low water system did not affect the integrity of the root of Napier grass. The root to shoot ratio of Napier grass inoculation with *Streptomyces* sp. STRM104 was also constant between the normal water

and low water systems, but the ability to stimulate the growth of Napier grass by this bacterial isolate was poor. Meanwhile, the root to shoot ratio of the plants in the control pots was increased under the low water system. It means that the roots of Napier grass grown under a low water system were not healthy. Therefore, using *Streptomyces* sp. St8 is the best to protect the root integrity of the plant in this study. However, the nutrient elements in all soils planted with Napier grass and inoculated with each isolate of *Streptomyces* sp. were lower than those in soil planted with Napier grass only (Table 6). The soil organic matter, available phosphorus, exchangeable potassium, exchangeable calcium, exchangeable magnesium, and total nitrogen in planted soil inoculated with *Streptomyces* sp. PB5, SRF1, St8, STRM104, and STRM302 were not increased compared to the control pots (Table 6). Available phosphorus, exchangeable potassium, and exchangeable calcium in the control pots were higher than those inoculated with *Streptomyces* sp. PB5, SRF1, St8, STRM104, and STRM302.

CONCLUSION

Inoculation with *Streptomyces* could increase Napier grass growth, and it is possible to use it as a biofertilizer for Napier grass planting. The different bacterial isolates had important factors that affect the Napier grass's growth and *Streptomyces* sp. St8 was the best isolate. The different systems in this study did not decrease the Napier grass's growth. For Napier grass inoculated with *Streptomyces* sp. St8, only

Table 6
Physical and chemical characteristics of soil under low water condition after Napier grass planting for 49 days

Treatment	pH	Calcium carbonate requirement (CaCO ₃ /rai)	Organic matter (g/kg)	% sand	% silt	% clay	Soil texture	Available phosphorus (mg/kg)	Exchangeable potassium (mg/kg)	Exchangeable calcium (mg/kg)	Exchangeable magnesium (mg/kg)	Total nitrogen (g/kg)
Control	3.88	403	2.2	66	21	13	Sandy loam	6.1	72	813	39	0.26
PB5	3.99	403	2.0	71	18	11	Sandy loam	4.6	30	354	30	0.26
SRF1	4.01	403	2.8	70	18	12	Sandy loam	5.3	57	475	35	0.26
St8	3.96	403	2.1	70	19	11	Sandy loam	4.6	26	587	33	0.17
STRM104	3.98	269	2.1	70	19	11	Sandy loam	5.3	37	399	30	0.22
STRM302	4.07	403	1.9	69	19	12	Sandy loam	4.2	29	378	33	0.22

the shoot fresh weight was decreased in the low system condition. Even though inoculation of soil with *Streptomyces* sp. did not increase the planted soil's fertility in this study, the nutrient accumulation in Napier grass inoculated with *Streptomyces* should be analyzed in further experiments.

ACKNOWLEDGEMENTS

This research was financially supported by Mahasarakham University.

REFERENCES

- Ahmad, F., Ahmad, I., & Khan, M. S. (2008). Screening of free-living rhizospheric bacteria for their multiple plant growth promoting activities. *Microbiological Research*, 163(2), 178-181. <https://doi.org/10.1016/j.micres.2006.04.001>
- Calvelo Pereira, R., Monterroso, C., & Macías, F. (2010). Phytotoxicity of hexachlorocyclohexane: Effect on germination and early growth of different plant species. *Chemosphere*, 79(3), 326–333. <https://doi.org/10.1016/j.chemosphere.2010.01.035>
- Chandra, D., Srivastava, R., Glick, B. R., & Sharma, A. K. (2018). Drought-tolerant *Pseudomonas* spp. improve the growth performance of finger millet (*Eleusine coracana* (L.) Gaertn.) under non-stressed and drought-stressed conditions. *Pedosphere*, 28(2), 227-240. [https://doi.org/10.1016/S1002-0160\(18\)60013-X](https://doi.org/10.1016/S1002-0160(18)60013-X)
- Chukwuneme, C. F., Babalola, O. O., Kutu, F. R., & Ojuederie, O. B. (2020). Characterization of actinomycetes isolates for plant growth promoting traits and their effects on drought tolerance in maize. *Journal of Plant Interaction*, 15(1), 93-105. <https://doi.org/10.1080/17429145.2020.1752833>
- de Souza, R., Ambrosini, A., & Passaglia, L. M. P. (2015). Plant growth-promoting bacteria as inoculants in agricultural soils. *Genetics and Molecular Biology*, 38(4), 401-419. <https://doi.org/10.1590/S1415-475738420150053>
- Deblonde, P. M. K., & Ledent, J. F. (2001). Effects of moderate drought conditions on green leaf number, stem height, leaf length and tuber yield of potato cultivars. *European Journal of Agronomy*, 14(1), 31-41. [https://doi.org/10.1016/S1161-0301\(00\)00081-2](https://doi.org/10.1016/S1161-0301(00)00081-2)
- Fahsi, N., Mahdi, I., Mesfioui, A., Biskri, L., & Allaoui, A. (2021). Phosphate solubilizing rhizobacteria isolated from jujube *Ziziphus lotus* plant stimulate wheat germination rate and seedlings growth. *PeerJ*, 9, e11583 <https://doi.org/10.7717/peerj.11583>
- Goswami, D., Vaghela, H., Parmar, S., Dhandhukia, P., & Thakker, J. N. (2013). Plant growth promoting potentials of *Pseudomonas* spp. strain OG isolated from marine water. *Journal of Plant Interactions*, 8(4), 281-290. <https://doi.org/10.1080/17429145.2013.768360>
- Haeghele, T., Bunnom, T., Khumhom, S., Braeuchler, C., Liplap, P., & Arjhan, W. (2017). Expanding the farming potential of Napier grass (*Pennisetum purpureum* SCHUMACH.) under low-fertile conditions. *Suranaree Journal of Science and Technology*, 24(2), 137-151.
- Huang, X., El-Alawi, Y., Penrose, D. M., Glick, B. R., & Greenberg, B. M. (2004). Response of three grass species to creosote during phytoremediation. *Environmental Pollution*, 130(3), 453-363. <https://doi.org/10.1016/j.envpol.2003.12.018>
- Khan, N., Bano, A., & Babar, M. A. (2017). The root growth of wheat plants, the water conservation and fertility status of sandy soils influenced by plant growth promoting rhizobacteria. *Symbiosis*, 72(3), 195-205. <https://doi.org/10.1007/s13199-016-0457-0>
- Kumar, A., Singh, S., Gaurav, A. K., Srivastava, S., & Verma, J. P. (2020). Plant growth-

- promoting bacteria: Biological tools for the mitigation of salinity stress in plants. *Frontiers in Microbiology*, *11*, 1216. <https://doi.org/10.3389/fmicb.2020.01216>
- Kumar, B. L., & Gopal, D. V. R. S. (2015). Effective role of indigenous microorganisms for sustainable environment. *3 Biotech*, *5*, 867-876. <https://doi.org/10.1007/s13205-015-0293-6>
- Lakshminarayanan, V., Ponnuswamy, R., & Rengaraju, B. (2015). Screening, purification and characterization of β -glucan from a novel strain *Bacillus cereus* LVK13 (KC 898956). *International Journal of ChemTech Research*, *8*(3), 1156-1162.
- Li, X., Geng, X., Xie, R., Fu, L., Jiang, J., Gao, L., & Sun, J. (2016). The endophytic bacteria isolated from elephant grass (*Pennisetum purpureum* Schumach) promote plant growth and enhance salt tolerance of hybrid *Pennisetum*. *Biotechnology for Biofuels*, *9*, 190. <https://doi.org/10.1186/s13068-016-0592-0>
- Machado, S., & Paulsen, G. M. (2001). Combined effects of drought and high temperature on water relations of wheat and sorghum. *Plant and Soil*, *233*(2), 179-187. <https://doi.org/10.1023/A:1010346601643>
- Mei, C., Amaradasa, S., Sikaroodi, M., Zhang, X., Gillevet, P., Nowak, J., & Lowman, S. (2021). Chapter 7 - Potential application of plant growth promoting bacteria in bioenergy crop production. In J. White, A. Kumar, & S. Droby (Eds.), *Microbiome stimulants for crops* (pp. 109-123). Woodhead Publishing. <https://doi.org/10.1016/B978-0-12-822122-8.00014-5>
- Nantasaksiri, K., Charoen-Amornkitt, P., & Machimura, T. (2021). Land potential assessment of Napier grass plantation for power generation in Thailand using SWAT model. Model validation and parameter calibration. *Energies*, *14*(5), 1326. <https://doi.org/10.3390/en14051326>
- Negawo, A. T., Teshome, A., Kumar, A., Hanson, J., & Jones, C. S. (2017). Opportunities for Napier grass (*Pennisetum purpureum*) improvement using molecular genetics. *Agronomy*, *7*(2), 28. <https://doi.org/10.3390/agronomy7020028>
- Niu, S., Gao, Y., Zi, Z., Liu, Y., Liu, X., Xiong, X., Yao, Q., Qin, Z., Chen, N., Guo, L., Yang, Y., Qin, P., Lin, J., & Zhu, Y. (2022). The osmolyte-producing endophyte *Streptomyces albidoflavus* OsiLf-2 induces drought and salt tolerance in rice via a multi-level mechanism. *The Crop Journal*, *10*(2), 375-386. <https://doi.org/10.1016/j.cj.2021.06.008>
- Odiyi, B. O., & Oludare, P. A. (2015). Impact of simulated salinity gradient on growth indices of *Pennisetum purpureum* Schumach. *Jordan Journal of Agricultural Sciences*, *11*(3), 661-667. <https://journals.ju.edu.jo/JJAS/article/view/10315/4651>
- Osman, N. A., Roslana, A. M., Ibrahima, M. F., & Hassana M. A. (2020). Potential use of *Pennisetum purpureum* for phytoremediation and bioenergy production: A mini review. *Asia Pacific Journal of Molecular Biology and Biotechnology*, *28*(1), 14-26. <https://doi.org/10.35118/apjmbb.2020.028.1.02>
- Pereira, N. C. M., Galindo, F. S., Gazola, R. P. D., Dupas, E., Rosa, P. A. L., Mortinho, E. S., & Teixeira Filho, M. C. M. (2020). Corn yield and phosphorus use efficiency response to phosphorus rates associated with plant growth promoting bacteria. *Frontiers in Environmental Science*, *8*, 40. <https://doi.org/10.3389/fenvs.2020.00040>
- Sade, N., Galkin, E., & Moshelion, M. (2015). Measuring *Arabidopsis*, tomato and barley leaf relative water content (RWC). *Bio-Protocol*, *5*(8), e1451. <https://doi.org/10.21769/BioProtoc.1451>
- Sangdee, A., Kornphachara, S., & Srisawat, N. (2016). *In vitro* screening of antagonistic activity of soil *Streptomyces* against plant pathogenic fungi and assessment of its characters. *International*

- Journal of Agricultural Technology*, 12(1), 173-185.
- Somtrakoon, K., Sabutong, B., Srinoi, P., Chaiyasit, R., Sangdee A., & Chouychai W. (2021). Pattern of *Streptomyces* sp. culture filtrate application on seedling growth of rice cv. RD6 cultivated under fluorene or phenanthrene contamination. *Journal of Agricultural Research and Extension*, 38(3), 15-27.
- Somtrakoon, K., Sangdee, A., Chouychai, W. (2019a). Roles of plant growth promoting bacteria on growth of ornamental plants grown in anthracene-spiked soil. *Journal of Agricultural Research and Extension*, 36(2), 11-22.
- Somtrakoon, K., Sripasa, N., Ladsena, S., Sangdee, A., & Chouychai, W. (2019b). Optimum conditions for indole-3-acetic acid production by *Streptomyces* and its stimulation on seed germination of rice cv. KDML105. *Journal of Agricultural Research and Extension*, 36(3), 12-22.
- Videira, S. S., de Oliveira, D. M., de Moraes, R. F., Borges, W. L., Baldani, V. L. D., & Baldani, J. I. (2012). Genetic diversity and plant growth promoting traits of diazotrophic bacteria isolated from two *Pennisetum purpureum* Schum. genotypes grown in the field. *Plant Soil*, 356, 51-66. <https://doi.org/10.1007/s11104-011-1082-6>
- Waramit, N., & Chaugool, J. (2014). Napier grass: A novel energy crop development and the current status in Thailand. *Journal of the International Society for Southeast Asian Agricultural Sciences*, 20(1), 139-150.
- Xu, Z., Mei, X., Tan, L., Li, Q., Wang, L., He, B., Guo, S., Zhou, C., & Ye, H. (2018). Low root/shoot (R/S) biomass ratio can be an indicator of low cadmium accumulation in the shoot of Chinese flowering cabbage (*Brassica campestris* L. ssp. *chinensis* var. *utilis* Tsen et Lee) cultivars. *Environmental Science and Pollution Research*, 25, 36328–36340. <https://doi.org/10.1007/s11356-018-3566-x>
- Yandigeri, M. S., Meena, K. K., Singh, D., Malviya, N., Singh, D. P., Solanki, M. K., Yadav, A. K., & Arora, D. K. (2012). Drought-tolerant endophytic actinobacteria promote growth of wheat (*Triticum aestivum*) under water stress conditions. *Plant Growth Regulation*, 68, 411-420. <https://doi.org/10.1007/s10725-012-9730-2>
- Zhang, S., Xu, X., Sun, Y., Zhang, J., & Li, C. (2018). Influence of drought hardening on the resistance physiology of potato seedlings under drought stress. *Journal of Integrative Agriculture*, 17(2), 336–347. [https://doi.org/10.1016/S2095-3119\(17\)61758-1](https://doi.org/10.1016/S2095-3119(17)61758-1)