EVALUATION OF THE EFFECT OF SOME INDIGENOUS MICROORGANISMS ON THE GROWTH AND YIELD OF WATER SPINACH UNDER GREENHOUSE AND FIELD CONDITIONS

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ABSTRACT

Indigenous microorganisms (IMO) have great potential in agricultural uses since they have high ability in biodegradation, nitrogen fixation, phosphate solubilization, plant growth hormone production. However, deep scientific knowledge about IMO's growth-promoting effect and vegetable yield has been limited and should be elucidated. This study aimed to evaluate the effect of some selected IMOs on the growth, yield of water spinach under greenhouse and field conditions. The greenhouse experiment was carried out with water spinach seeds that were inoculated with IMO solution at a dilution of 1.000-fold for 4 h and grown for 26-30 days. The level of chemical nitrogen fertilizer dose application for pot experiments were 25% recommended N and 50% recommended N. The field experiments were arranged in the condition that water spinach seeds were soaked in 500 times diluted IMO solution and reduced by 25% the amount of nitrogen fertilizer recommended for water spinach. The result showed that IMO helped to reduce 25% chemical N fertilizer, increase water spinach yield up to 18% as well as reduce from 42 to 59% of the total nitrate in fresh vegetables as compared to the control fertilizer recommended treatment. It is possible to exploit indigenous microorganisms as a source of beneficial microorganisms for stimulating plant growth and increasing vegetable yield.

ĐÁNH GIÁ ẢNH HƯỞNG CỦA HỆ VI SINH VẬT BẢN ĐỊA LÊN SINH TRƯỞNG VÀ NĂNG SUẤT CỦA RAU MUỐNG Ở ĐIỀU KIỆN NHÀ LƯỚI VÀ NGOÀI ĐỒNG

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TỪ KHÓA

Vi sinh vật bản địa
Nitrate trong rau
Kích thích sinh trưởng
Đạm tổng số trong rau
Rau muống

TÓM TẮT

Vi sinh vật bản địa (IMO) có tiềm năng lớn trong ứng dụng nông nghiệp vì chúng có khả năng phân hủy sinh học, cố định nitơ, hòa tan lân, tổng hợp hormone thực vật. Tuy nhiên, kiến thức khoa học về tác dụng thúc đẩy tăng trưởng và gia tăng năng suất rau của IMO còn giới hạn và cần được nghiên cứu. Mục đích của nghiên cứu này là đánh giá ảnh hưởng của các IMO lên sinh trưởng, năng suất của rau muống trong điều kiện nhà lưới và ngoài đồng. Thí nghiệm trong nhà lưới được thực hiện với hạt rau muống được ngâm trong dung dịch IMO ở độ pha loãng 1.000 lần trong 4 giờ và trồng trong 26-30 ngày. Mức phân đạm hóa học được giảm là 25% và 50% so với khuyến cáo. Thí nghiệm đồng ruộng được bố trí trong điều kiện ngâm hạt rau muống trong dung dịch IMO pha loãng 500 lần và giảm 25% lượng phân phân đạm khuyến cáo cho rau muống. Kết quả cho thấy IMO giúp giảm 25% lượng phân đạm hóa học, giúp kích thích sinh trưởng và làm tăng năng suất rau muống lên đến 20% đồng thời giảm từ 42% - 59% lượng nitrat trong rau tươi so với bón phân theo khuyến cáo. Có thể khai thác vi sinh vật bản địa làm nguồn vi sinh vật có ích để kích thích sinh trưởng cây trồng và tăng năng suất rau.

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1. Introduction

Soc Trang is a province with favorable climate and soil conditions, which is suitable for growing vegetables all year round. For a long time, many areas specializing in vegetable cultivation have been formed, including leafy vegetables. In the cultivation of vegetables, especially water spinach, Ceylon spinach, mustard green... to achieve maximum yield, beautiful vegetables, easy to sell and get high profit, farmers in many places have abused nitrogenous fertilizers and chemical growth stimulants, which increases the cost of vegetable cultivation. In addition, leafy vegetables have a short growth time, if the implementation of safe isolation is not guaranteed, it will lead to the residue of nitrate and growth stimulants in vegetables. This has a bad effect on consumers' health [1]. Therefore, at present, it is interested in developing safe and sustainable vegetable farming, which focus on the use of organic biological products and plant growth-promoting microorganisms. Plant growth-promoting microorganisms, especially indigenous microorganisms play an important role in stimulating plant growth as well as improving soil fertility, heath and quality because of the innate microbial consortium that is highly adaptable, survived and well developed under adverse soil, weather, climatic and environmental conditions [2].

The concept of indigenous microorganism (IMO) was developed by Cho Han Kyu and it contains consortia of beneficial microorganisms by trapping soil microorganisms when using cooked rice as an attractive source of nutrients, including fungi, bacteria, actinomyces... [2], [3]. IMO has a great potential in bio-fertilizers, bio-composting, biodegradation, bioleaching, bio-composting, phosphate solubilization, nitrogen fixation, soil fertility improvement and in the production of plant growth hormones as well as bio-control [4]. The positive effects of IMOs on soil physical, chemical and biological properties and soil enzyme activity, soil health and crop yield have been demonstrated by numerous previous studies [5]-[12]. In addition, Chiemela et al. [13] indicated that the application of IMO in agriculture was a friendly environmental method because IMO helped to enhance organic matter decomposition, plant nutrition, crop yields and soil fertility. Thus, the utilization of IMO resources in agricultural production brings many benefits to crops, soil, and humans. However, evaluation of the efficacy of IMOs on the growth and yield of vegetables under greenhouse and field conditions has been limited on both national and international scales. Therefore, in this study we evaluated the effect of some IMO on the growth, yield of water spinach under greenhouse and field conditions.

2. Materials and methods

2.1. Materials

Five IMOs were collected from 5 different cropping systems which had long-term cultivation with different crops such as guinea arrowroot, Ceylon spinach, chili, bamboo, and mixing of IMO within Soc Trang province. Refer to Le and Nguyen [14], [15] for more information about the content of microorganism. The origin and functions of the five IMOs are presented in Table 1 [14].

Table 1. *The origin and function of 5 collected IMO samples in Soc Trang province*

No.	Code	Origin of IMO (Farming system)	Nitrogen Fixing (mg/L)	IAA synthesizing (mg/L)
1	IMO guinea arrowroot	guinea arrowroot	5.33	35.4
2	IMO Ceylon spinach	Ceylon spinach	8.13	42.2
3	IMO chili	chili	8.09	12.0
4	IMO bamboo	bamboo	6.26	28.8
5	Mix IMO	mixed IMO	0.03	42.6

2.2. Evaluation of the efficiency of 5 IMOs on growth and yield of water spinach in the greenhouse condition

The study was carried out under the greenhouse of the Soil Science Department, College of Agriculture, Can Tho University. The soil was collected from vegetable cultivation land with topsoil (0-30 cm) in Tai Van commune, My Xuyen district, Soc Trang province. The collecting soil was mixed into a large sample. Then put 6 kg of soil (dry weight) into plastic pots (30 cm (height) x 30 cm (diameter). Soil samples were analyzed for some physical, chemical, and biological parameters including pH, EC, organic matter (CHC), nitrate (NO_3^-), ammonium (NH_4^+), total nitrogen (N), total phosphorus (P), total potassium (K), and the number of bacteria, fungi and actinomycetes in the soil. The soil characteristics were suitable for setting up experiments to evaluate the effect of fertilizers and bacteria on the growth and yield of water spinach in greenhouse condition. The soil was mixed well before sowing. The experiment was carried out continuously through 3 cropping seasons on the same experimental soil.

The experiment was arranged in a completely randomized design with a total of 12 treatments and 4 replicates for each treatment. The recommended chemical fertilizer formula for water spinach plants was 100N- $48P_2O_5$-$24K_2O$ [15], [16], the level of chemical nitrogen fertilizer dose application for both experiments were 25% recommended N and 50% recommended N. Information about pot experiments is presented in Table 2. Collected parameters included growth, yield, and total nitrogen and nitrate content in vegetables.

Table 2. *The treatments in field experiments for water spinach in greenhouse condition*

No.	Treatment	Content of treatment
1	T1	Control 0N-$0P_2O_5$-$0K_2O$
2	T2	Control 100N-$48P_2O_5$-$24K_2O$
3	T3	75N-$48P_2O_5$-$24K_2O$ + IMO guinea arrowroot
4	T4	75N-$48P_2O_5$-$24K_2O$ + IMO Ceylon spinach
5	T5	75N-$48P_2O_5$-$24K_2O$ + IMO chili
6	T6	75N-$48P_2O_5$-$24K_2O$ + IMO bamboo
7	T7	75N-$48P_2O_5$-$24K_2O$ + Mix IMO
8	T8	50N-$48P_2O_5$-$24K_2O$ + IMO guinea arrowroot
9	T9	50N-$48P_2O_5$-$24K_2O$ + IMO Ceylon spinach
10	T10	50N-$48P_2O_5$-$24K_2O$ + IMO chili
11	T11	50N-$48P_2O_5$-$24K_2O$ + IMO bamboo
12	T12	50N-$48P_2O_5$-$24K_2O$ + Mixed IMO

Water spinach seeds were inoculated with IMO solution at a dilution of 1.000-fold for 4 hours before sowing into the pot and growing for 27-28 days. The fertilizer application schedule and composition of each specific application for water spinach are presented in Table 3.

Table 3. *Fertilizer schedule for experimental water spinach in the greenhouse condition*

Fertilizer kinds	The amount of fertilizer (kg/ha)	1st time (3 DBS) (%)	2nd time (10 DAS) (%)	3rd time (20 DAS) (%)	4th time (25 DAS) (%)
N	100	20	30	30	20
P_2O_5	48	100	0	0	0
K_2O	24	50	20	20	10

**Note: DBS: day before sowing; DAS: day after sowing*

Collecting agronomic parameters: plant height, diameter of plant, number of leaves per plant, total chlorophyll content in leaf, fresh biomass of water spinach per pot. Agronomic parameters will be taken at harvest time. Methods for agronomic parameter collection are presented in Table 4.

Table 4. *Methods for agronomic parameter collection of water spinach*

Item	Survey Standard	Unit
Plant height	Measured from the ground to the top of leaf	cm
Plant diameter	Measured the maximal diameter of the stem	mm
Number of leaves/ plant	Count the number of leaves per each plant	leaf
Chlorophyll content in leaf	Using Chlorophyll Content Meter 200 Plus	CCI
Fresh biomass	Measure total fresh biomass of water spinach per pot	cm

**Note: CCI is Chlorophyll Content Index*

2.3. Evaluation on the efficiency of 2 selected IMOs on growth and yield of water spinach in the field condition

Field experiments were carried out at Truong An hamlet, Truong Khanh commune, Long Phu district, Soc Trang province. Information about the time of greenhouse experiments is presented in Table 5.

Table 5. *Time schedule of two field experiments*

	Seeding time	Harvesting time	Day of growth
Crop1	Apr. 27th 2020	May 27th 2020	30 days
Crop 2	June 23rd 2020	July 18th 2020	26 days

The experiment was arranged in a randomized complete block design with 5 treatments and 3 replicates for each treatment. Information about treatment in field experiments is presented in Table 6. The area of each treatment was 15 m^2 (1.5 m width x 5 m length) and 30 cm height. The distance between the 2 experimental plots was 0.5 m and the distance between the 2 blocks was 0.5 m. Before sowing, the soil was levelled, and the weed was removed.

Table 6. *The treatments in field experiments for water spinach in Truong Khanh commune, Long Phu district, Soc Trang province*

No.	Treatment	Details
1	T1	100N- 48P_2O_5-24K_2O (control)
2	T2	75N-48P_2O_5-24K_2O (control)
3	T3	75N-48P_2O_5-24K_2O + CMP EMpb
4	T8	75N-48P_2O_5-24K_2O + IMO spinach
5	T9	75N-48P_2O_5-24K_2O + IMO bamboo

**Note: CMP is commercial microbial products of Center for Commercial Microbial Products Ho Chi Minh City Limited Liability Company*

It was prepared with the 1000-fold dilution of IMO solution, water spinach seeds were soaked in 500 times diluted IMO solution, then sowing these soaked seeds into. Chemical fertilizers for water spinach plants were applied according to the formula 100N-48P_2O_5-24K_2O [16], [17]. The fertilizer application schedule and composition of each specific application for water spinach in the field are presented in Table 7.

Table 7. *Fertilizer schedule for experimental water spinach in the field condition*

Elements	The amount of fertilizer kg/ha	1st time (3 DBS) (%)	2nd time (10 DAS) (%)	3rd time (15 DAS) (%)	4th time (20 DAS) (%)
N	100	0	50	30	20
P_2O_5	48	100	0	0	0
K_2O	24	50	20	20	10

**Note: DBS: day before sowing; DAS: day after sowing*

Agronomic parameters including growth, yield, and total nitrogen and nitrate content in vegetables were determined at the harvest time. The yield was measured the weight of fresh biomass within a fixed area of 1 m^2. Total nitrogen content (N_t) in fresh vegetable biomass was determined by sample inorganic method and Kjeldahl distillation followed by titration with 2% H_3BO_3 with color indicator. The nitrate (NO_3^-) content in vegetables was analyzed by extraction with distilled water and coloration with vanadium chloride reagent solution.

2.4. Data analysis

The data were ANOVA analyzed by using MINITAB 16.2 software.

3. Results and discussions

3.1. The effect of 5 selected IMOs on the growth, yield of water spinach under greenhouse condition

3.1.1. Height of water spinach plant

Results of evaluating the effect of IMO on the height of water spinach plants in pot conditions are presented in Table 8. Generally, over 3 consecutive vegetable crops, the treatments with reduction of nitrogen fertilizer at 25% and 50% reduced the height of water spinach compared to the treatment with 100% N fertilization as recommended, however, in the treatments of IMO inoculation, the height of water spinach was not a significant difference at the level of 5% together. IMO bamboo with an average height of 43 cm was higher than the other treatments but not statistically significant ($p>0.05$) when compared together and this value was still lower than the positive control treatment received with the recommended formula as 100N-48P_2O_5-24K_2O.

Table 8. *Average of some agronomic parameters of water spinach over 3 crops in pot conditions*

No.	Treatment	Height (cm)	NBL (leaf)	Diameter (mm)	Chlorophyll (CCI)	Fresh biomass (g/pot)
1	Control 0N-0P_2O_5-0K_2O	27.5^{b}	7.77^{d}	3.44^{b}	8.4^{b}	12.4^{b}
2	Control 100N-48P_2O_5-24K_2O	46.7^{a}	10.1^{ab}	5.84^{a}	13.9^{a}	41.3^{a}
3	75N-48P_2O_5-24K_2O + IMO guinea arrowroot	39.2^{ab}	10.4^{a}	5.46^{a}	12.3^{a}	31.5^{ab}
4	75N-48P_2O_5-24K_2O + IMO Ceylon spinach	40.9^{ab}	10.0^{abc}	5.45^{a}	12.8^{a}	31.4^{ab}
5	75N-48P_2O_5-24K_2O + IMO chili	40.8^{ab}	10.1^{ab}	5.37^{a}	12.7^{a}	29.5^{ab}
6	75N-48P_2O_5-24K_2O + IMO bamboo	43.0^{ab}	10.1^{ab}	5.65^{a}	13.1^{a}	32.7^{ab}
7	75N-48P_2O_5-24K_2O + Mix IMO	40.6^{ab}	9.88^{abc}	5.58^{a}	13.3^{a}	30.8^{ab}
8	50N-48P_2O_5-24K_2O + IMO guinea arrowroot	38.1^{ab}	8.54^{bcd}	5.23^{a}	11.8^{ab}	23.7^{ab}
9	50N-48P_2O_5-24K_2O + IMO Ceylon spinach	41.0^{ab}	9.22^{abcd}	5.19^{a}	11.6^{ab}	28.5^{ab}
10	50N-48P_2O_5-24K_2O + IMO chili	40.5^{ab}	9.12^{abcd}	5.04^{a}	11.8^{ab}	27.4^{ab}
11	50N-48P_2O_5-24K_2O + IMO bamboo	40.5^{ab}	8.60^{bcd}	5.21^{a}	12.1^{ab}	28.1^{ab}
12	50N-48P_2O_5-24K_2O+ Mixed IMO	40.7^{ab}	8.44^{cd}	5.12^{a}	11.4^{ab}	28.8^{ab}
	F	*	*	*	*	*
	CV (%)	15.4	9.97	13.8	5,67	26.0

**Note: Values in the same column having the same letters are not significant difference at 5% level of Tukey test; CCI: chlorophyll content index*

3.1.2. Number of water spinach leaf

Table 8 presents the results of evaluating the effect of IMO on the number of water spinach leaves. In general treatments with IMO inoculation in a combination with N fertilizer reduction (25-50% N as recommended) had a good effect on the number of water spinach leaves with leaves ranging from 8 to 10 leaves and a significant difference from each other was found when compared with each other. The numbers of leaves of the water spinach plants of the treatments received with 25%N fertilizer reduction were significantly higher ($p<0.05$) than the treatment

with 50%N fertilizer reduction. Besides, the number of leaves was maintained at the same level as those of the positive control treatment received with the recommended formula as 100N-$48P_2O_5$-$24K_2O$.

3.1.3. Diameter of stem

The diameter of the stem of the water spinach plant is presented in Table 8. Overall, in all 3 experiments of water spinach, the treatments fertilizing with 75N-$48P_2O_5$-$24K_2O$ and inoculation of IMO had no statistically significant difference in diameter of the stem as compared to the control recommended treatment (100N-$48P_2O_5$-$24K_2O$).

3.1.4. Chlorophyll content in leaf

The results from 3 consecutive crops showed that the chlorophyll content of water spinach leaves was significantly different among treatments ($p < 0.05$). The treatments with reduction of nitrogen fertilizer decreased the chlorophyll content of water spinach leaf, but they were not significantly different at the level of 5% when compared with each other.

3.1.5. Fresh biomass per pot

In general, treatments that received 75N and 50N-$48P_2O_5$-$24K_2O$ in a combination with indigenous microorganisms had a lower fresh weight than the treatment received only recommended formula (100N-$48P_2O_5$-$24K_2O$), but not statistically significantly different when compared together. In general, treatments with IMO inoculation in a combination with 2 levels of N fertilizer reduction (25-50% N as recommended) were not significantly different when compared together. Among studied IMOs, IMOs from bamboo, guinea arrowroot and Ceylon spinach soils showed the greatest stimulating effect in the growth and yield of water spinach in three crops. This means that these IMOs had a great effect on the growth of water spinach. This can be explained by the fact that the nitrogen fixation ability by microbial communities in these IMOs was much better than other remaining IMOs, however, because of the 1000 times dilution, the population of nitrogen-fixing bacteria was low, leading to the amount of nitrogen fixed not enough to replace 25%N in the fertilizer. Therefore, in field experiments 500 times dilution from the microbial stock of IMOs was chosen.

This result is consistent with the results from the previous studies about nitrogen fixation capacity of IMOs [14] that indicated that IMO from bamboo and IMO Ceylon spinach had the best ability in nitrogen fixation. Especially when comparing the effectiveness of IMOs with each other in promoting the growth of canola (*Brassica juncea*), Keli'ikuli [18] reported that IMO collected from bamboo was effective as a plant growth promoter and gave the highest yield of the studied crop as compared with other IMOs collected from others and the control without IMO inoculation. Therefore, the two IMOs which collected from bamboo and Ceylon spinach soil were chosen for further study under the field condition.

3.2. Effect of 2 selected IMOs on growth, yield of water spinach in field condition

3.2.1. Water spinach growth and yield

The results of evaluating the effect of IMO on the growth and fresh weight of water spinach which are presented in Table 9 showed that the agronomic parameters and fresh weight of water spinach had big variation between the two crops, but the pattern of treatments was the same. Treatments inoculated with IMO showed better efficacy on stimulating the growth of water spinach than the treatments received only chemical fertilizers and even better than the commercial microbial products EMpb.

Especially, in the first crop, when the weather conditions were severely bad with drought, and salinity, the treatment applied with bamboo and Ceylon spinach IMOs showed the best efficiency in an increase of the agronomic parameters of water spinach including height, number of leaves,

the diameter of the stem, chlorophyll content in leaf and consequently increasing the yield of water spinach. Particularly, these two treatments helped to increase by 18.5-20.7% of water spinach's yield as compared to the positive control treatment. This could be explained by the fact that the nitrogen-fixing bacteria strains in IMO fixed nitrogen in the air to replace enough of the amount reduced nitrogen from fertilizers, as well as stimulating water spinach to grow well in the severely dry season. This suggests that the IMO strain may have supported water spinach plants in the process of nitrogen fixation and better nitrogen uptake by water spinach in the dry season.

Similarly, in the second crop, the treatment with IMO inoculation under the condition of 25% reduction in nitrogen from fertilizer had significantly higher growth and yield of water spinach as compared to the control treatment which only received recommended 100%NPK fertilizer. However, the actual yield of water spinach of these two treatments was not significantly different ($p>0.05$) when compared together.

Table 9. *Agronomic parameters and yield of water spinach in two crops under field conditions*

No.	Treatment	Agronomic parameters				
		Height (cm)	NBL (leaf)	Diameter (cm)	Chlorophyll (CCI)	Yield (kg/m^2)
Crop 1 from April 27 to May 27, 2020						
1	Control 100N-48P_2O_5-24K_2O	42.7^c	12.7^b	8.28^a	18.8^b	3.52^{ab}
2	Control 75N-48P_2O_5-24K_2O	39.2^d	12.1^c	8.12^a	18.3^b	3.16^b
3	75N-48P_2O_5-24K_2O + CMP EMpb	47.1^b	12.2^{bc}	6.41^b	19.7^b	3.84^{ab}
4	75N-48P_2O_5-24K_2O + IMO Ceylon spinach	47.3^b	14.0^a	7.11^{ab}	21.8^a	4.17^a
5	75N-48P_2O_5-24K_2O + IMO bamboo	49.4^a	13.7^a	7.09^{ab}	19.8^b	4.25^a
	F	*	*	*	*	ns
	CV (%)	8.49	6.37	9.50	6.89	15.63
Crop 2 from June 23 to July 18. 2020						
1	Control 100N-48P_2O_5-24K_2O	60.4^b	10.8	7.60^b	16.8^a	5.21^{ab}
2	Control 75N-48P_2O_5-24K_2O	52.4^d	10.4	6.88^c	14.7^c	3.39^c
3	75N-48P_2O_5-24K_2O + CMP EMpb	58.4^c	11.0	6.89^c	15.7^b	4.87^b
8	75N-48P_2O_5-24K_2O + IMO Ceylon spinach	61.7^a	11.4	8.40^a	15.4^{bc}	5.59^a
9	75N-48P_2O_5-24K_2O + IMO bamboo	60.2^b	11.1	7.43^{bc}	15.4^{bc}	5.31^{ab}
	F	*	ns	*	*	*
	CV (%)	5.80	4.75	8.31	9.63	22.89

**Note: Values in the same column having the same letters are not significant difference at 5% level of Tukey test*

This study result also showed that weather conditions strongly influenced the growth and yield of water spinach, especially in the dry season the growth and yield of water spinach were reduced strongly. However, treatment applied with IMO helped to resist vegetable yield losses for farmers under unfavorable weather conditions. The effectiveness of IMO in this study was similar to previous studies about IMO. The study of Sanchez et al. [11] showed that the addition of 1L of IMO solution at a frequency of 4 weeks would significantly increase plant height and yield components of basmati rice and lead to an increase in the actual yield of basmati rice. Similar results were found in other studies by Koon-Hui et al., Sakimin et al., Desiré et al., Keli'ikuli and Sekhar et al. [7], [9], [10], [12], [19], [20].

3.2.2. Chemical content in water spinach biomass

Similarly, the results of the evaluation of the effectiveness of IMO on some chemical parameters of water spinach presented in Table 10 revealed that there was also a big difference

between the two crops in terms of chemical parameters and there was a statistically significant difference between treatments in each crop when compared with each other.

Table 10. *Some chemical parameters in water spinach biomass in 2 crops under the field condition*

No.	Treatment	N_t (g/kg)	P (g/kg)	K (g/kg)	NO_3^- (mg/kg)
Crop 1 from April 27 to May 27, 2020					
1	Control 100N-48P_2O_5-24K_2O	32.2	6.89^{ab}	85.4^{a}	1.232^{a}
2	Control 75N-48P_2O_5-24K_2O	31.8	9.99^{b}	74.5^{b}	1.097^{a}
3	75N-48P_2O_5-24K_2O + CMP EMpb	32.7	6.90^{ab}	73.1^{b}	384^{b}
8	75N-48P_2O_5-24K_2O + IMO Ceylon spinach	31.9	7.23^{a}	70.6^{b}	348^{b}
9	75N-48P_2O_5-24K_2O + IMO bamboo	31.0	6.95^{ab}	71.6^{b}	406^{b}
	F	ns	*	*	*
	CV (%)	3.58	8.12	8.51	59.0
Crop 2 from June 23 to July 18. 2020					
1	Control 100N-48P2O5-24K2O	41.7^{a}	4.30^{ab}	71.3^{c}	2.186^{a}
2	Control 75N-48P2O5-24K2O	32.6^{b}	3.82^{b}	72.7^{bc}	348^{f}
3	75N-48P2O5-24K2O + CMP EMpb	37.5^{ab}	5.25^{a}	83.4^{ab}	1.348^{c}
8	75N-48P2O5-24K2O + IMO Ceylon spinach	41.0^{a}	4.28^{ab}	86.8^{a}	1.636^{b}
9	75N-48P2O5-24K2O + IMO bamboo	38.6^{ab}	5.09^{a}	74.5^{bc}	987^{d}
	F	*	*	*	*
	CV (%)	10.4	13.9	9.47	53.3

**Note: Values in the same column having the same letters are not significant difference at 5% level of Tukey test*

For total nitrogen content, in the first crop, there was not significantly different among treatments and the amount of nitrogen content in treatment was lower than that in crop 2. In addition, the second crop also showed the total nitrogen content in water spinach that was significantly different among treatments. In which the IMO inoculation showed their good efficacy in increasing the total nitrogen content compared to the control treatment with only NPK fertilizer application. Thus, reducing fertilizer may reduce total nitrogen taken up by vegetables, but the treatment applied with IMO enhanced the total nitrogen.

Concerning phosphate (P) and potassium (K), it was found that P and K were less affected by the IMO inoculation. But there was a big variation between the two crops. In which in the dry season higher concentrations of P and K were found as compared to the rainy season. In addition, for potassium, it was found that in crop 2, the treatment with IMO or EMpb gave higher potassium content. This means that the microorganisms in IMO or EMpb helped plants to uptake potassium better and prevent water spinach from falling [20]-[22].

Water spinach is leafy vegetable and commonly used in daily meals in our country [23]. Studies show that this vegetable has a high ability to accumulate nitrate when applied with chemical fertilizers [24], [25]. Therefore, in vegetable farming, it is necessary to reduce both the recommended nitrogen fertilizer and nitrate content. Reducing 25% of recommended nitrogen fertilizer in a combination with inoculation of IMO helped to significantly enhance the fresh biomass of water spinach by 18% and up to 20.7% as compared with the control treatments without microbial inoculation. Moreover, inoculation with IMO during vegetable cultivation helped to reduce up to 59% of nitrate content accumulated in water spinach biomass but still maintained the same amount of total nitrogen content in water spinach biomass. This result is similar to studies on the effectiveness of bacterial strains [26]-[28].

4. Conclusions

Application of IMO from different cropping systems not only helped to reduce the amount of nitrogen fertilizer recommended by 25%, but also increase vegetable yield up to 20% and reduce the amount of nitrate accumulation in vegetables by 42-59%, while maintaining the total nitrogen

in vegetables when compared with the recommended NPK fertilizer treatment. It is recommended to continue to research more about IMO for vegetable cultivation in Soc Trang as well as other provinces in the Mekong Delta.

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