Microbial-induced mobilization of micronutrients in soil for increased yield and improved nutritional quality of sweet potato and purple yam

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Outline

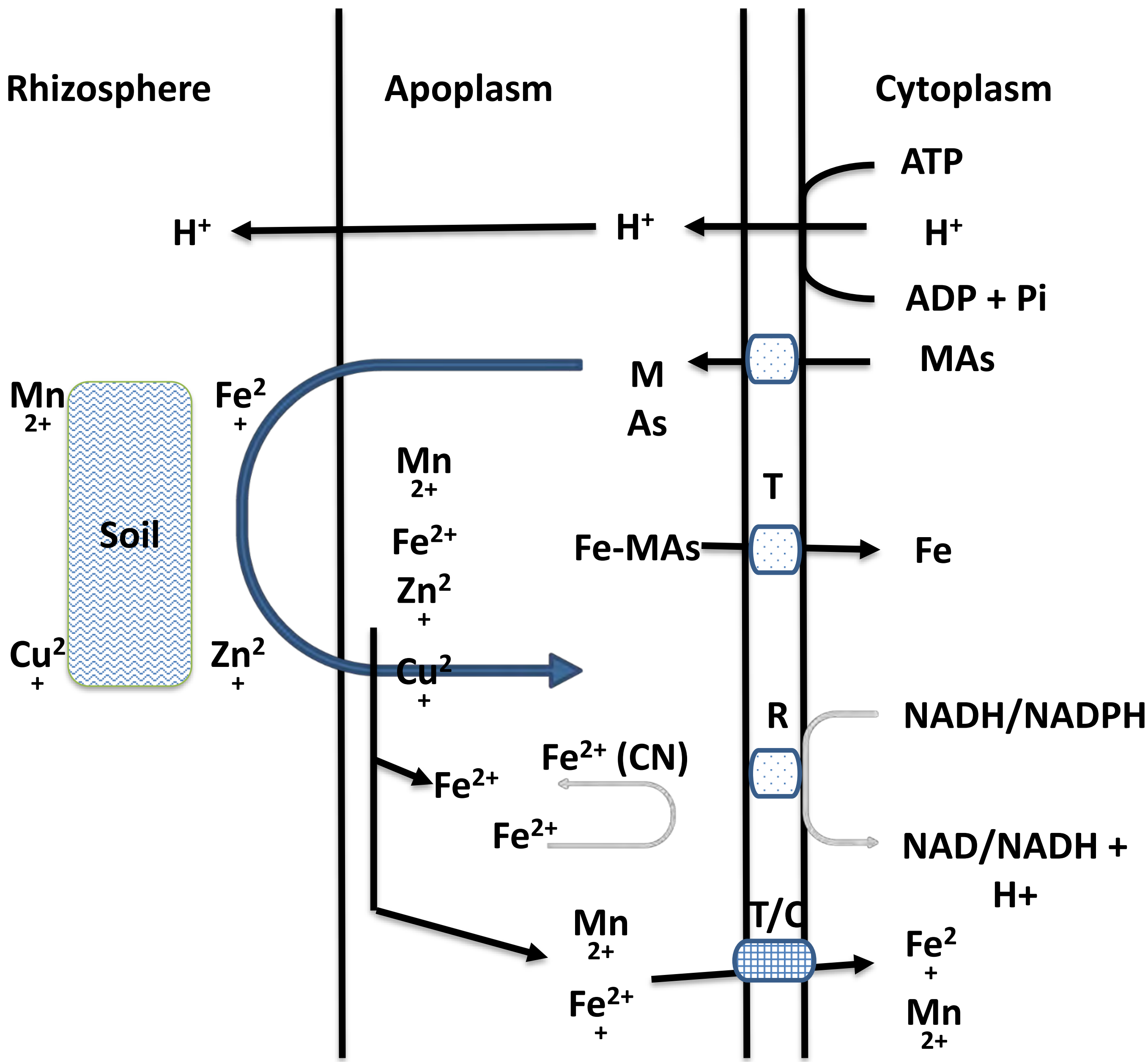
- Introduction
- Objectives
- Methodology
- Physico-chemical characteristics of soil samples
- Isolation, characterization, functional evaluation and identification of microbial isolates
- Development of organic-based cultivation medium
- Process flowsheet for mass production of the bacterial inoculant
- Shelf-life experiment of inoculant
- Evaluation of effectivity of microbial inoculant (pot experiments)
- Summary and Next Steps

Introduction

- The success of food production depends greatly on the fertility status of the soil
- Continuous planting leads to fast depletion of available nutrients causing tremendous problem in crop production
- The limited availability of agricultural lands makes farmers utilize marginal areas and highly degraded lands

Introduction

- In the Philippines, there are many types of soils that are classified as marginal areas
- Notable among these are the highly weathered acid soils and volcanic ash derived soils which abound in mountainous and hilly areas
- Soil analysis revealed that these marginal soils are rich in micronutrients such as Fe, Zn and Cu which is insoluble thus cannot be utilized by plants.
- Knowing the critical roles of micronutrients in the growth and development of crops, it is necessary to find ways to facilitate the availability of these micronutrients to plants.



T or C – Transporter or Channel

Rs – Standard Reductase

Possible Mechanism

1. Bacteria produces/secretes enzymes that helps solubilize micronutrients in soil
2. Bacteria produce pytohormones such as cytokinins, IAA and other unknown metabolites that facilitate transfer and stabilize growth
3. Bacteria produce reactive ligand groups that bind to cationic micro-nutrients for translocation

Ref. Yang and Romheld, 1999

Introduction

- Micronutrient deficiency is very common in the country. Such deficiencies in plant affect the human health through food chain resulting to loss of productivity.
- Increasing attention has been given to them in recent years not only by nutritionists and medical scientists but also by economists and social scientists because of their serious threat to human health especially to child health.

Introduction

- Experiencing the deteriorative effects of synthetic and chemical fertilizers, there is a need to employ useful microbial population responsible for continuous availability of micronutrients from natural sources to revive soil health and improve soil quality in order to support sustainable crop production system. They also improve the plant immune system thereby increasing its resistance against pest and diseases.

Objectives

General

To isolate and evaluate microbial inoculants for the release of micronutrient from soil minerals naturally present in various soil environments for increased yield and improved nutritional quality of sweetpotato and purple yam

Objectives

Specific

1. To isolate naturally occurring soil microorganisms capable of oxidation and dissolution of soil minerals specifically oxides of Zn, Fe and Cu from different soil environments.
2. To characterize and identify the above-isolated microorganisms.
3. To develop an organic-based microbial cultivation method to mass produce the above microbial inoculants.
4. To evaluate the effectivity of the isolated microbial inoculants in increasing yield and improving the nutritional quality of sweetpotato and purple yam.

Methodology

Soil collection and analysis

- Collection of soil samples from six different soil environments
- Preparation of soil samples
- Soil chemical analyses (pH, % organic matter, total nitrogen, extractable Fe, Mn, Zn and Cu. Available phosphorous and exchangeable bases of K, Na, Ca and Mg)

Microbial assay and characterization

- Isolation of soil-borne microorganisms
- Preparation of microbial inoculum
- Dissolution experiments of Fe, Cu and Zn Stefanescu et al. (2009) and Kalinowski et al.(2000).
- Characterization and identification of isolates

Microbial production and evaluation

- Development of culture conditions of isolates
- Mass production of the isolates
- Pot experiments to evaluate microbial inoculants

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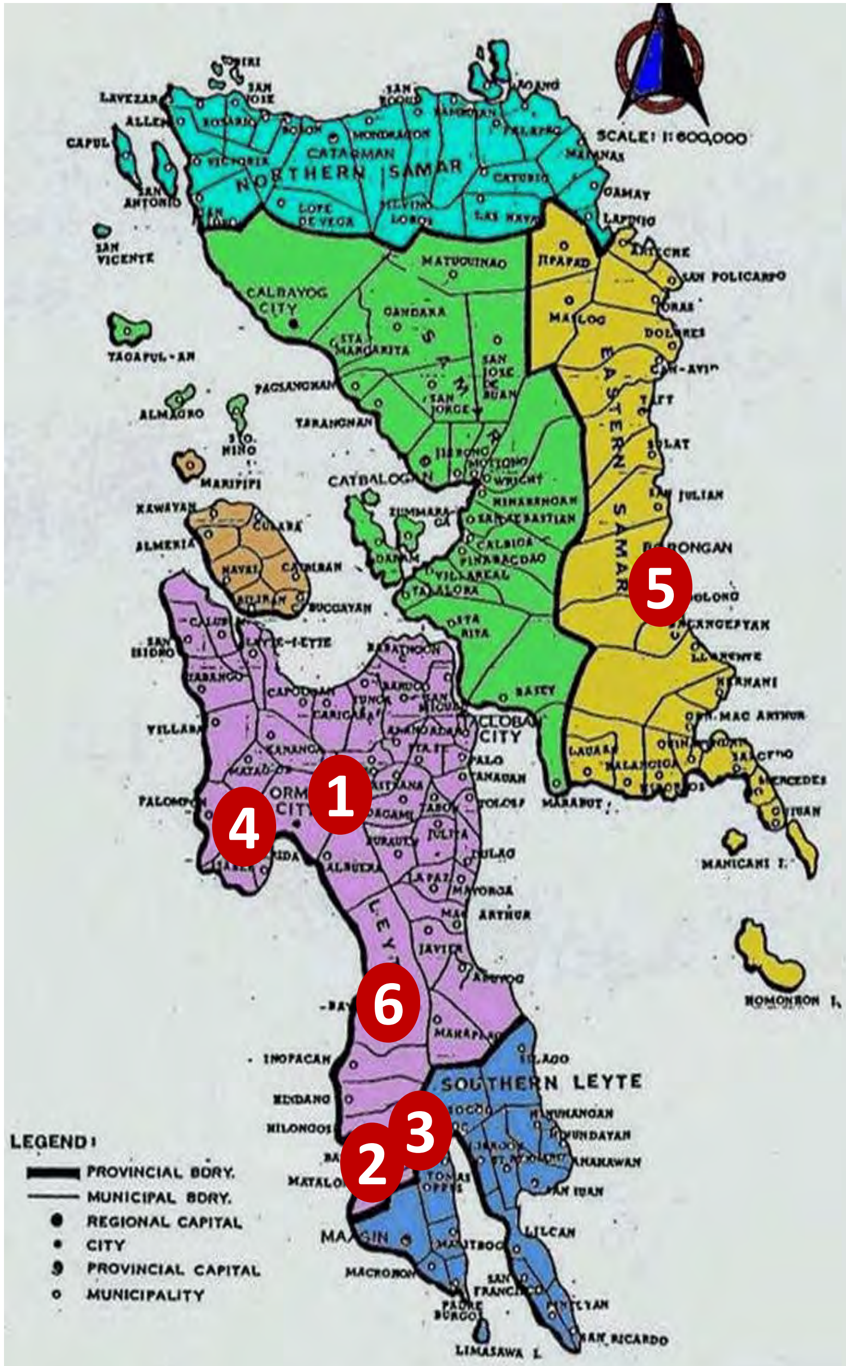
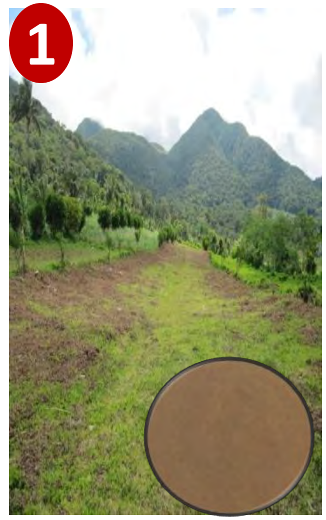
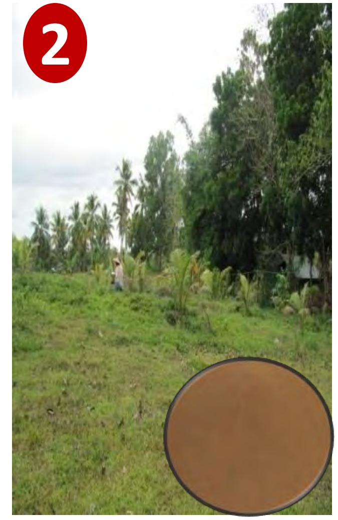




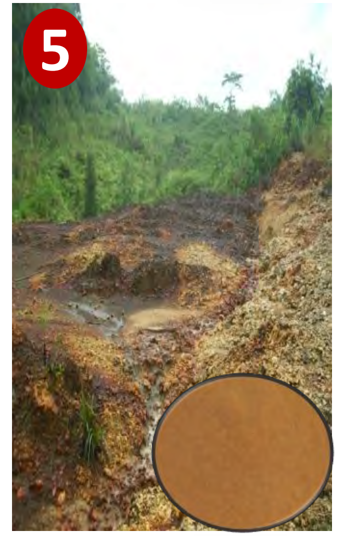
Cabintan, Ormoc City



San Salvador, Matalom Leyte

Alta Vista, Matalom Leyte

PASAR, Isabel, Leyte



Bagacay, Western Samar

Pangasugan, Baybay Leyte

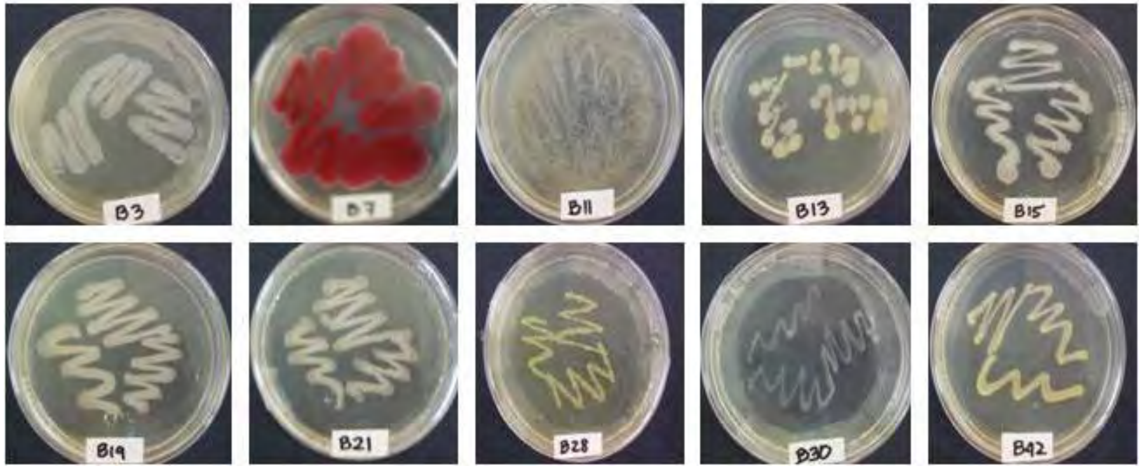
Locations of sampling sites

Characteristics of soils from various collection sites.

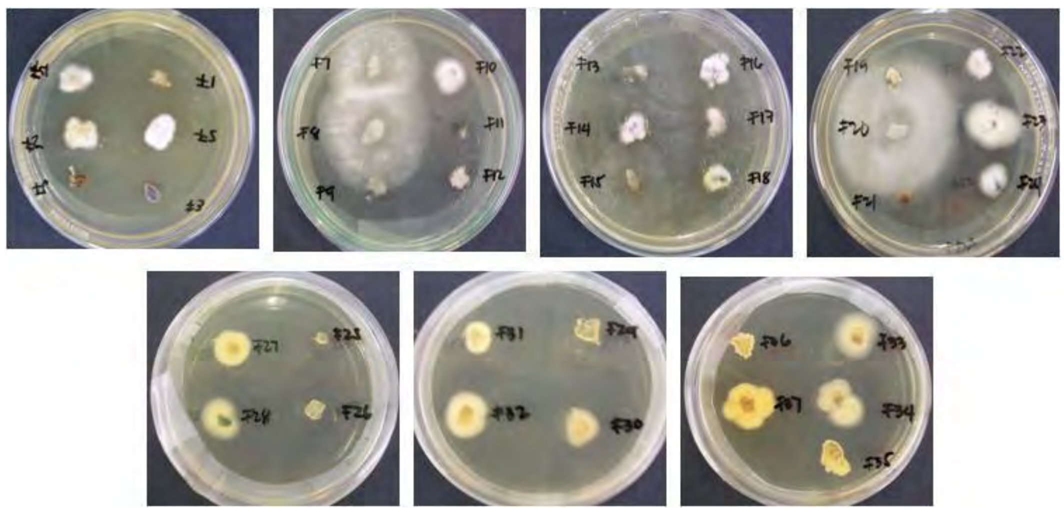
Collection area	Soil description
1. Cabintan, Ormoc City	Volcanic soil
2. San Salvador, Matalom, Leyte	acidic soil
3. Alta Vista, Matalom, Leyte	calcareous (basic)
4. PASAR, Isabel, Leyte	copper smelter area with calcareous soil
5. Pangasugan, Baybay, Leyte	Neutral soil
6. Bagacay, Western Samar	previous mining area, acidic soil

Results of the pH, organic matter and micronutrient analysis of the soil samples collected from six locations in Leyte and Samar, Philippines.

Sample Source	pH	OM (%)	Total N, %	Fe (ppm)	Zn (ppm)	Cu (ppm)	Mn (ppm)	K cmol(+)/ kg soil	Na cmol(+)/ kg soil	Ca cmol(+)/ kg soil	Mg cmol(+)/ kg soil
Cabintan, Ormoc (disturbed)	5.29	1.25	0.26	55.70	1.91	4.20	17.40	0.12	0.14	0.22	0.06
Cabintan, Ormoc (undisturbed)	5.80	7.58	0.66	58.45	1.93	4.20	21.25	0.13	0.15	0.36	0.18
San Salvador, Matalom (D)	5.72	1.16	0.11	18.50	0.30	1.40	42.65	0.03	0.11	0.65	0.37
San Salvador, Matalom (UD)	5.83	2.42	0.14	12.80	1.08	1.70	30.75	0.03	0.12	1.19	0.83
Alta Vista, Matalom (D)	8.71	4.23	0.21	4.34	0.31	1.41	5.41	0.07	0.13	36.1	0.59
Alta Vista, Matalom (UD)	8.75	1.76	0.20	14.10	0.56	3.32	11.69	0.09	0.15	33.9	1.20
Pasar, Isabel (D)	8.62	0.32	0.06	3.28	3.88	9.63	2.56	0.06	0.07	30.3	0.42
Pasar, Isabel (UD)	7.81	5.48	0.43	2.28	4.27	41.21	0.07	0.04	0.04	21.1	0.32
Pasar, Isabel (Slag)	7.56	6.77	0.20	18.44	8.02	38.57	0.13	0.07	0.02	1.38	0.03
Bagacay, W. Samar (D)	5.96	7.23	0.05	402.7	91.91	398.5	21.65	0.04	0.03	45.1	2.05
Bagacay, W. Samar (UD)	5.50	1.42	0.10	132.3	0.67	2.00	trace	0.03	0.05	1.04	0.22
VSU, Baybay (D)	6.59	1.42	0.13	311.5	3.43	6.80	55.50	0.55	0.18	12.1	4.18
VSU, Baybay (UD)	6.20	1.45	0.23	159.9	2.57	3.95	38.70	0.93	0.13	15.4	5.12



The 10 fast growing bacteria isolated from different sites.



Comparative growth of fungal isolates in solid medium after 48 h of cultivation.

Criteria for selection of isolates

- Fast growers
- Spore-formers
- High dissolution capacity of Fe, Cu and Zn

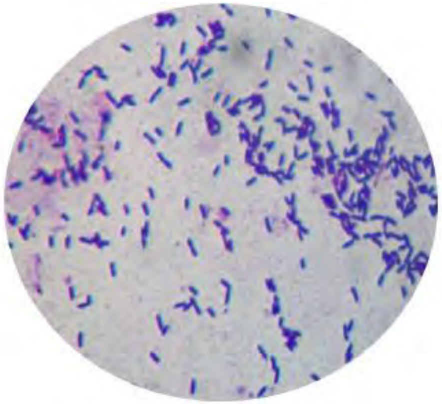
Top three bacterial and fungal isolates

Bacterial isolates

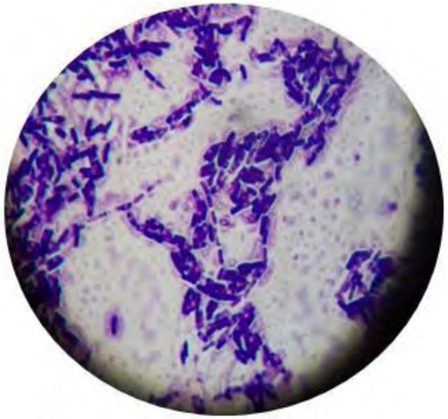
1. B21 – *Bacillus subtilis*
2. B28 – *Bacillus sp.*
3. B42 – *Bacillus megaterium*

Fungal isolates

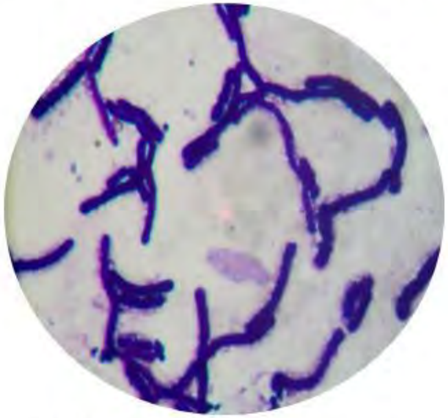
1. F2 – *Mucor sp.*
2. F5 – *Aspergillus sp.*
3. F8 – *Mucor sp.*



B21 *Bacillus subtilis*

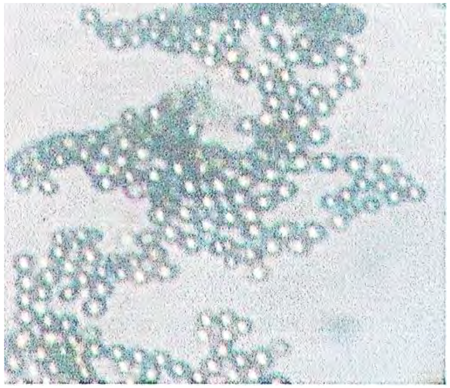


B-28 *Bacillus subtilis*

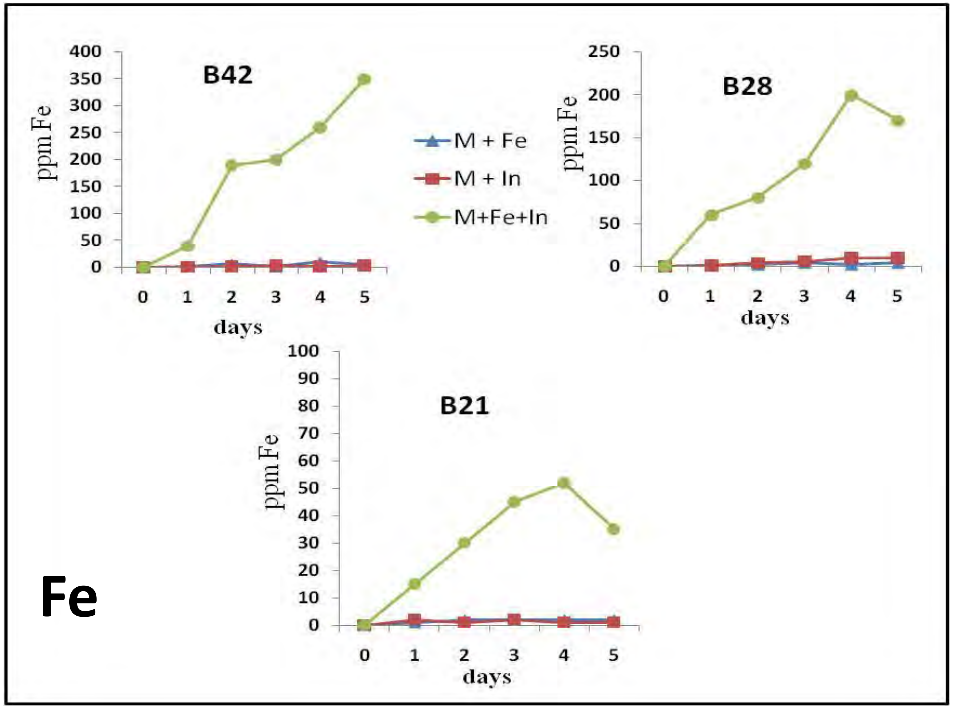


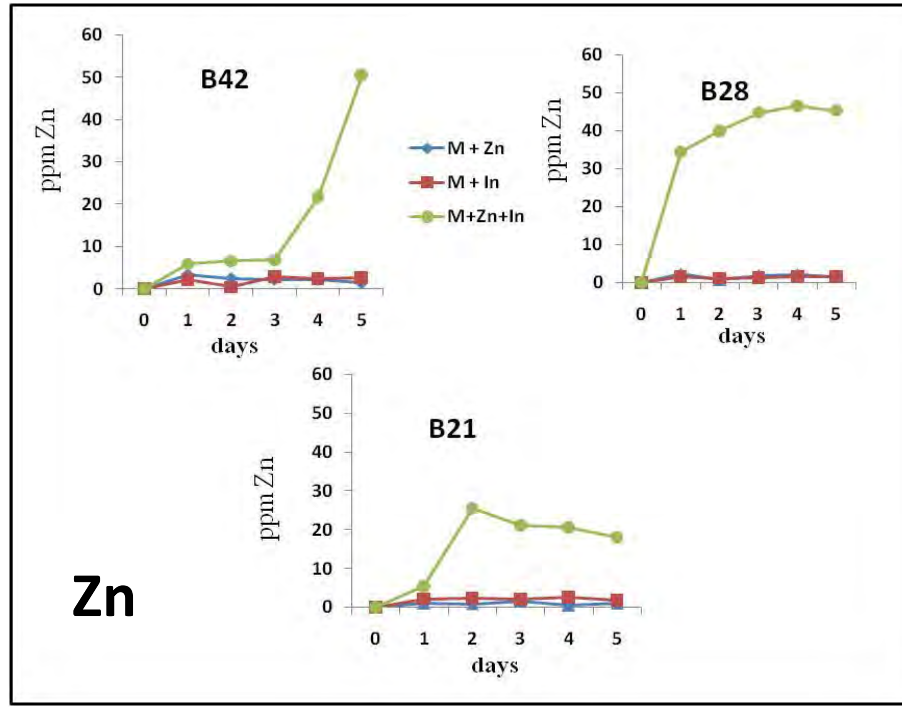
B-42 *Bacillus megaterium*

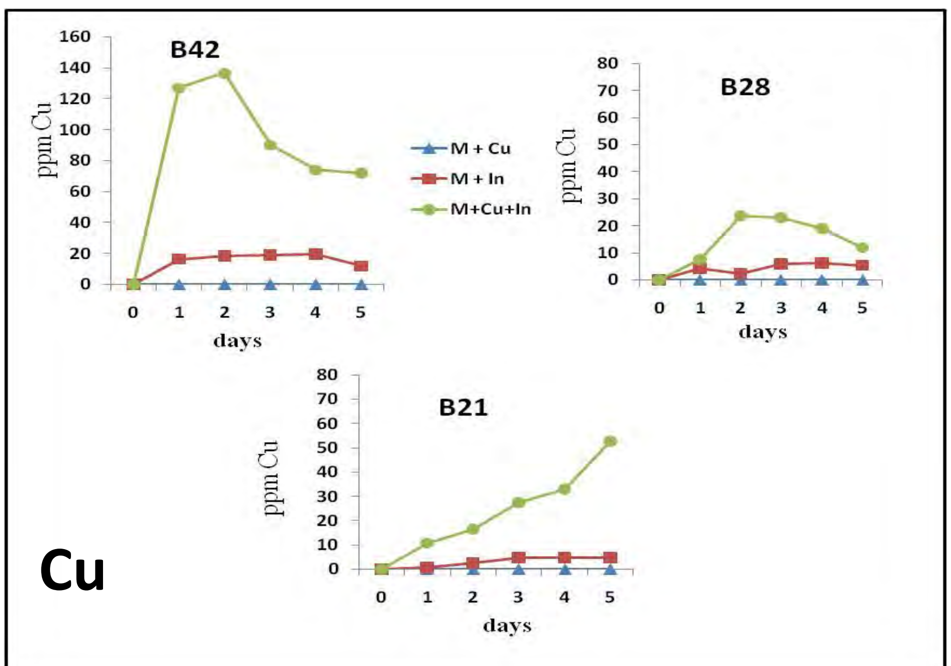
F8 *Mucor spp*.



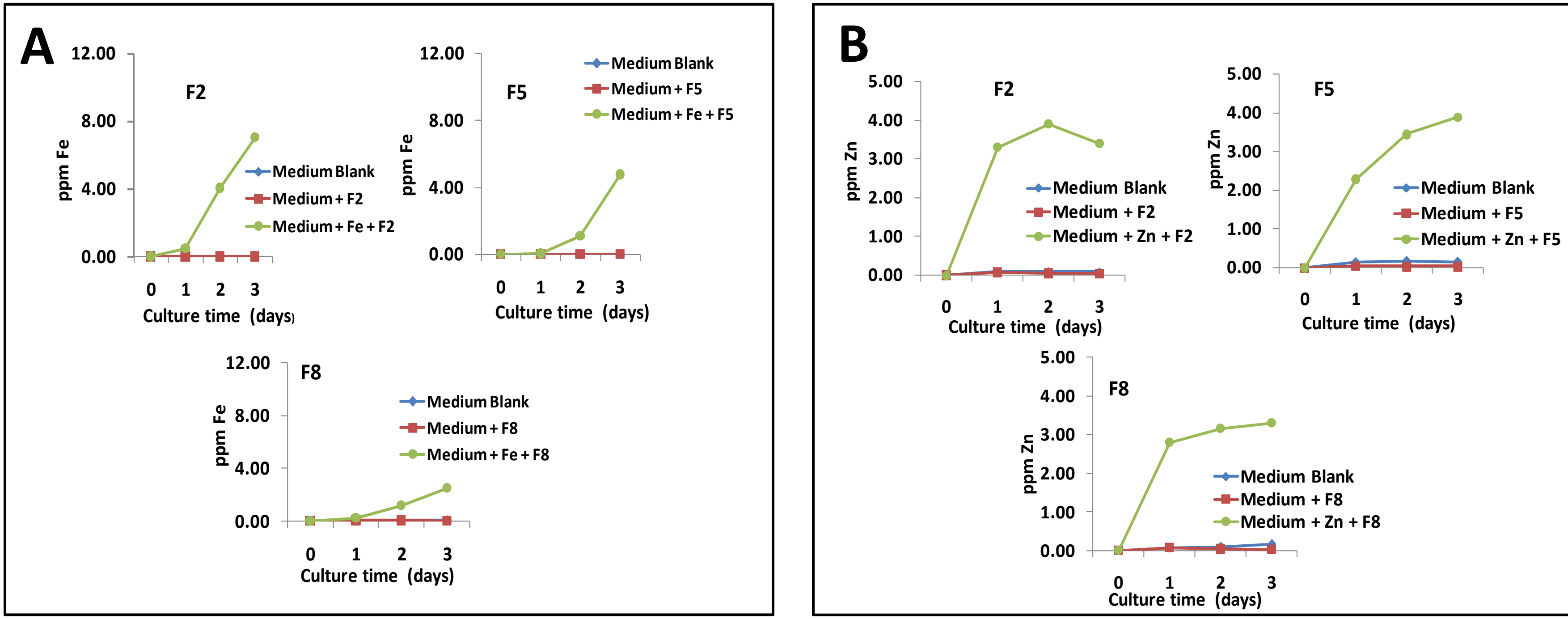
F5 *Aspergillus spp*.







Dissolution of micronutrients in top three bacterial isolates cultured for 5 days



Dissolution of Fe (A) and Zn (B) in top three fungal isolates cultured for 3 days.

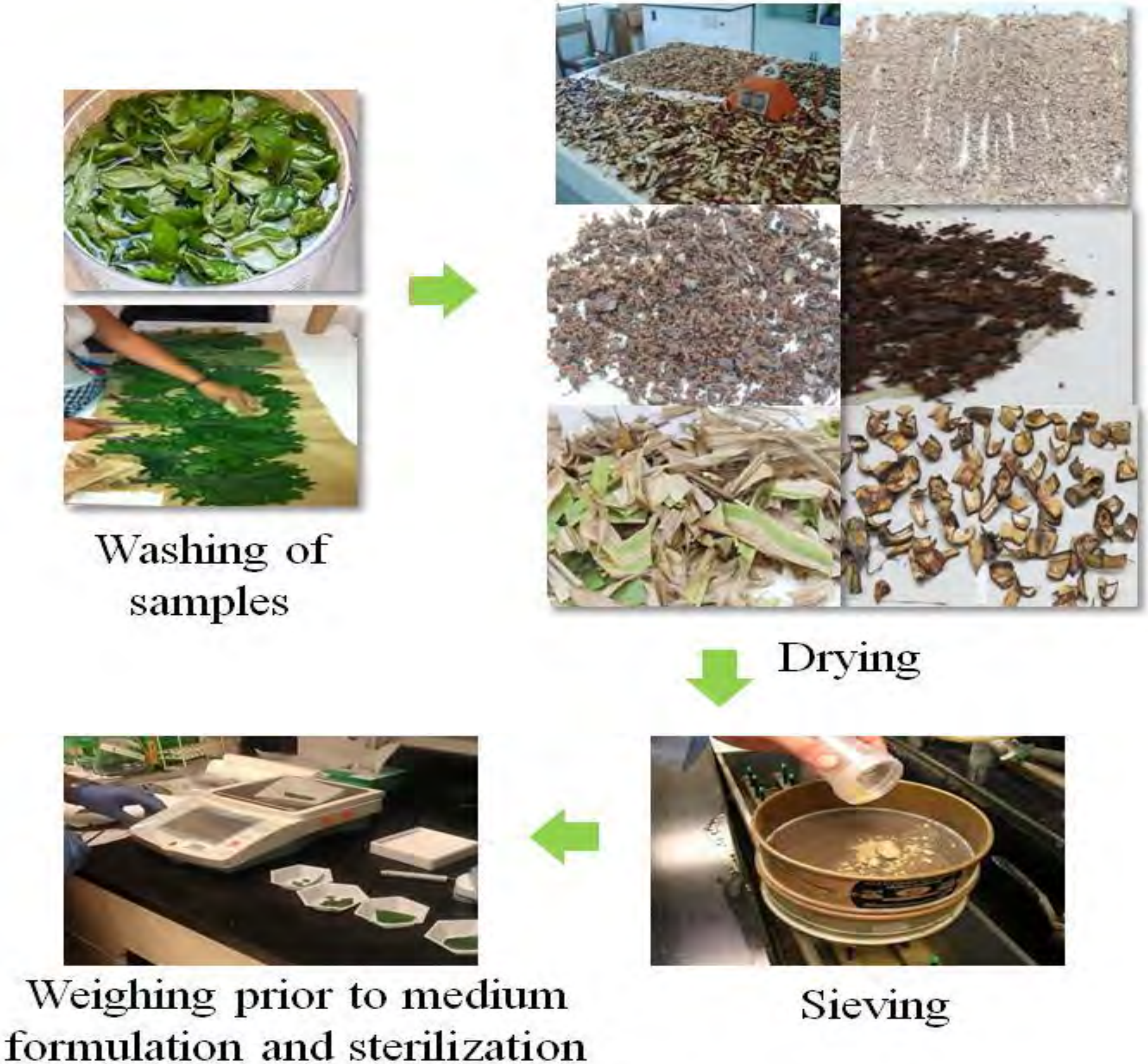
Morphological and biochemical characterization of the top three bacterial isolates.

Characteristics	**B21**	**B28**	**B42**
Morphological characterization			
Oxygen requirement	aerobic	aerobic	aerobic
Temperature	37°C	37°C	37°C
Colonies	dirty white	light yellow	grayish yellow
Shape and arrangement	short bacilli	short bacilli	large bacilli
Gram staining	+	+	+
Motility	+	+	+
Culture characterization on agar plates			
MRS (gas pak)	-	-	-
Differential and selective medium			
BSA	-	-	-
BGA	-	-	-
EMB	-	-	-
MacConkey Agar	-	-	-
SSA	-	-	-
XLD	-	-	-
Staph 110	-	-	-
Biochemical tests			
Indole	-	-	-
MR			
VP	-	-	-
SC			
Catalase	-	-	-
Nitrate	-	+	-
Litmus milk	alkaline	stormy clot	alkaline
Gelatin	+	-	+
H2S gas	-	-	-
Urease	-	-	-
Carbohydrates fermentation			
Arabinose	+	+	+
Cellobiose	+	+	+
Lactose	-	-	-
Mannitol	+	+	+
Melibiose	+	+	+
Mannose	+	+	+
Sucrose	+	+	+
Glucose	+	+	+
Maltose	+	+	+
Raffinose	+	+	+
Rhamnose	+	+	+
Salicin	+	+	+
Sorbitol	+	+	+
Xylose	+	+	+
Trehalose	+	+	+
Possible Microorganism	***Bacillus subtilis***	***Bacillus subtilis***	***Bacillus megaterium***

Morphological and biochemical characterization of the top three fungal isolates.

Growth Characteristics	Fungal isolate		
	F2	**F5**	**F8**
Sabaraud Dextrose Agar	+	+	+
growth	abundant	abundant	abundant
temperature °C	30°C	30°C	30°C
colony color :			
2-3 days	fluffy cottony white	powdery velvety green yellow	white
5-7 days	grayish white	greenish brown	grayish white
Morphological Characteristics :			
sporangiophores	branched		branched
Apophyses	absent		absent
Sporangia	spherical		spherical
Rhizoid	absent		absent
Stylophores	absent		absent
Conidiophores		smooth/ greenish	
Phialides		uniseriate	
vesicle		round,columnar head	
Possible identity	***Mucor spp.***	***Aspergillus spp.***	***Mucor spp.***

Development of organic-based cultivation medium for microbial production



Substrates used for mass production of isolate B42 (*Bacillus megaterium*) and cultivation of the same. A – dried cassava peelings, B – dried ipil-ipil leaves, C – seed culture of isolate B42, D –half-liter culture of isolate B42.

Root Crops Biotech

Microbial cultivation set-up for monitoring microbial growth.

Substrate selection: Growth ($x10^8$ cfu/ml) of bacterial isolate B42 in different C and N combinations. Number of cells was counted after 5 days of culture.

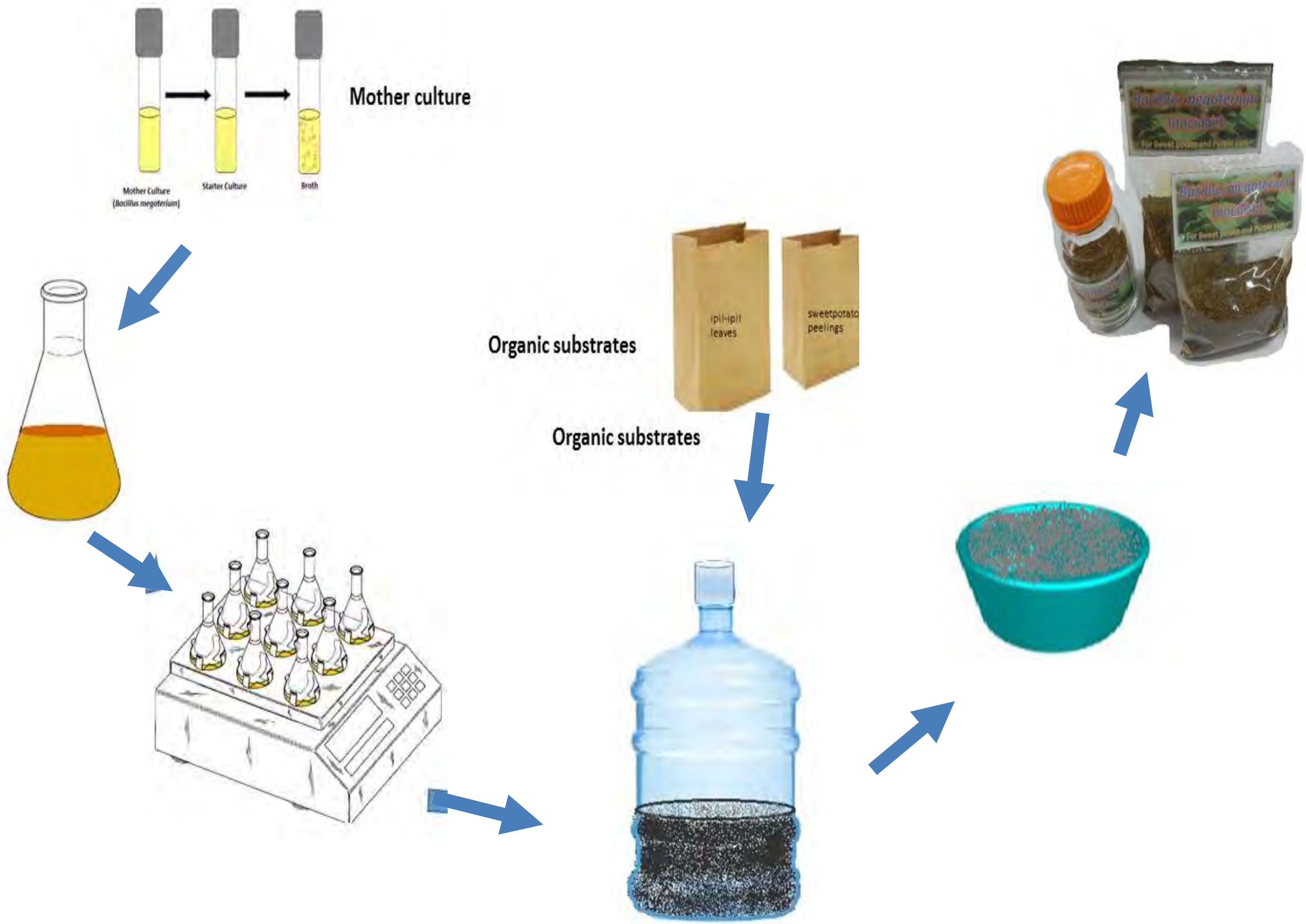
N-source	C-source					
	None	Rice straw	Cassava peel	Sweet potato peel	Banana peel	Molasses
None		21	14	17	26	31
Ipil-ipil leaves	tmtc	tmtc	60	24	36	37
Kakawate leaves	65	23	tmtc	26	22	tmtc
Chicken dung	12	13	13	15	11	21
Corn leaves	34	26	27	21	39	36

tmtc – too many to count

Substrate optimization: Growth (x10^8 cfu/ml) of bacterial isolate B42 in different substrate combinations. Number of cells was counted after 5 days of culture.

Particle Size (mm)	Moisture Content		
	30%	50%	70%
>19.05	5.1x10^9	3.2 x10^9	4.1 x10^9
19.05	1.8 x10^9	5.1 x10^9	8.1 x10^9
8.64	2.0 x10^9	3.7 x10^9	8.7 x10^9
2.34	1.9 x10^9	TMTC	TMTC
0.66	TMTC	9.3 x10^9	9 x10^9

TMTC – too many to count



PROCESS FLOWSHEET FOR MASS PRODUCTION OF THE BACTERIAL INOCULANT

Inoculant from *Bacillus megaterium* (dried product)

Shelf-life Study

Viability of stored dried *B. megaterium* inoculant.

Storage Period (months)	Storage Container	
	Plastic Polyethylene	Pyrex Bottles
1	TMTC	TMTC
2	TMTC	TMTC
4	TMTC	$3.4 X10^9$
6	$1.4 X10^9$	$1.1 X10^9$
7	$6.7 X10^8$	$7.7 X10^8$
8	$6 X10^8$	$6.3 X10^8$
9	$5.3 X10^8$	$5.7 X10^8$
10	$4.0 X10^8$	$4.3 X10^8$

TMTC – too many to count

Performance evaluation of the microbial experiment

Collection of soil samples for use in pot experiments.

A – Bagacay Samar (previous mining area)

B – Cabintan, Ormoc (volcanic soil)

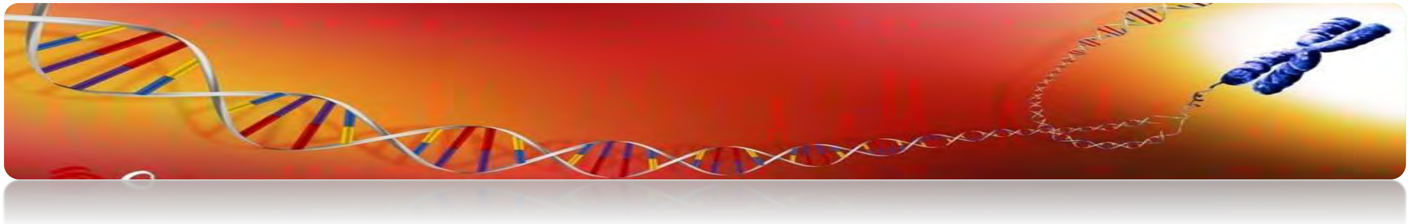
C – Matalom, Leyte (acidic soil)

EXPERIMENTAL TREATMENTS

Purple Yam VU-2 variety	**Sweetpotato** LSU-25 variety
T1 – control	T1 – control
T2 – 150-50-150 kg/ha (N-P2O5-K2O) recommended rate for yam	T2 – 60-60-60 kg/ha (N-P2O5-K2O) recommended rate for sweetpotato
T3 – T2 + 8 kg/ha Biozome-200	T3 – T2 + 8 kg/ha Biozome-200
T4 – T2 + Mycovam-1 (5 g/plant, manufacturer's recommended rate)	T4 – T2 + Mycovam-1 (5 g/plant, manufacturer's recommended rate)
T5 – T2 + 4 kg/ha microbial inoculant	T5 – T2 + 4 kg/ha microbial inoculant
T6 – T2 + 8 kg/ha microbial inoculant	T6 – T2 + 8 kg/ha microbial inoculant
T7 – T2 + 12 kg/ha microbial inoculant	T7 – T2 + 12 kg/ha microbial inoculant

* Applied in split , at 1 month after planting and the other half at one month after 1st application.

* Applied 2 weeks after planting.



Composition of Biozome S-200

S – 16%

Mg – 0.45

Mn – 3.0

Zn – 12.0

Cu – 1.0

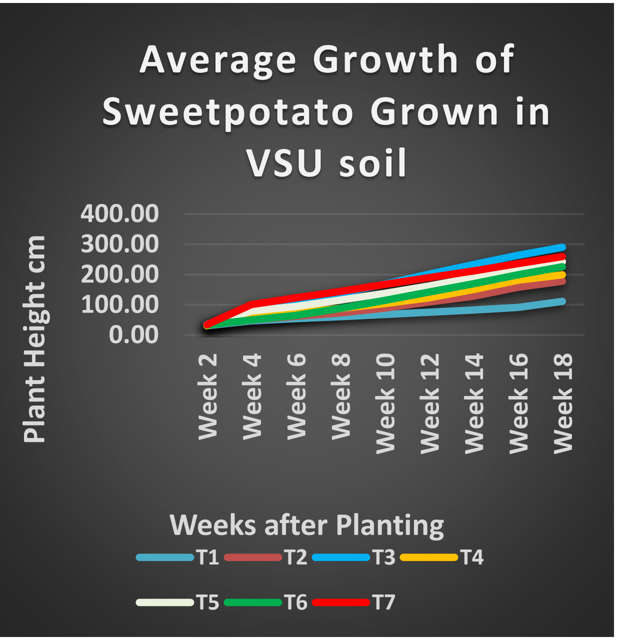
B – 0.50

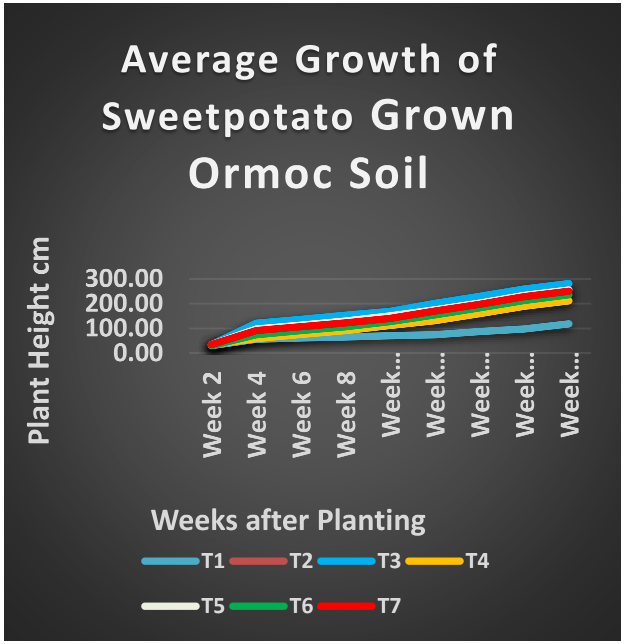
Fe – 1.0

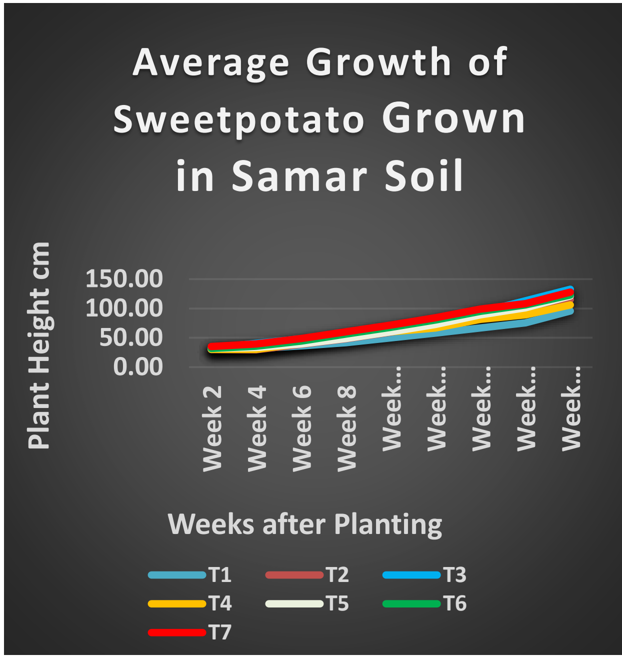
Mo – 0.05

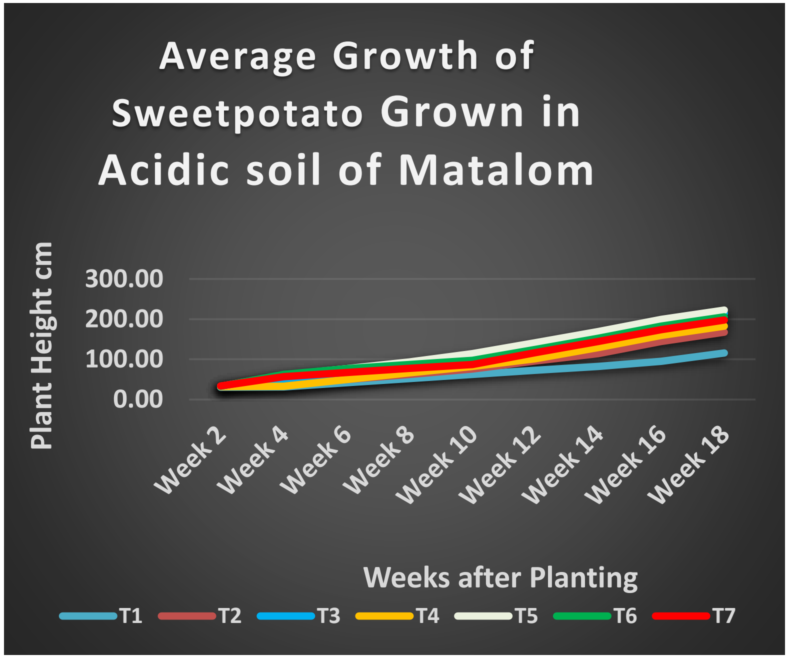
Co – 0.03

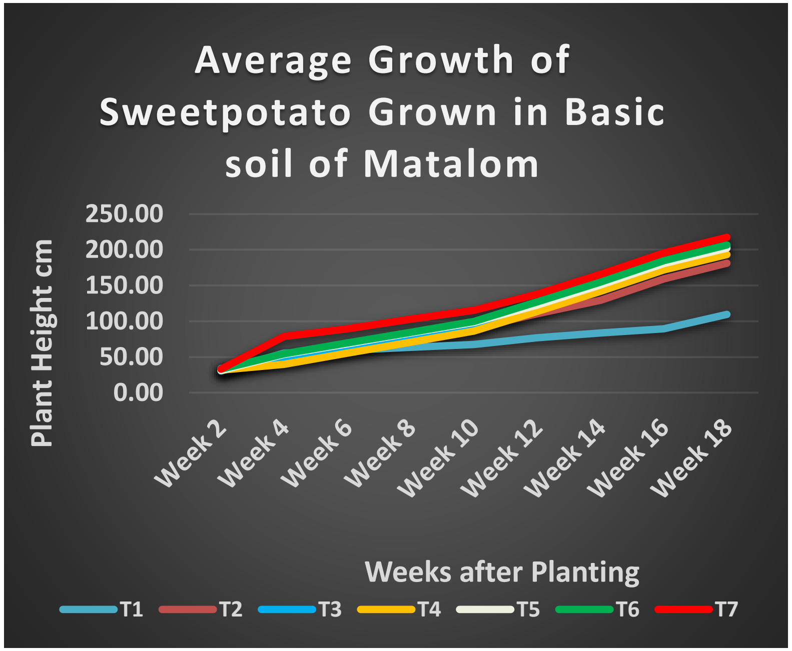
POT EXPERIMENT OF SWEETPOTATO LSU-25 VARIETY


Average Growth of Sweetpotato Grown in VSU soil
Plant Height cm
400.00
300.00
200.00
100.00
0.00
Week 2
Week 4
Week 6
Week 8
Week 10
Week 12
Week 14
Week 16
Week 18
Weeks after Planting
T1
T2
T3
T4
T5
T6
T7


Average Growth of Sweetpotato Grown Ormoc Soil
Plant Height cm
300.00
200.00
100.00
0.00
Week 2
Week 4
Week 6
Week 8
Week...
Week...
Week...
Week...
Week...
Weeks after Planting
T1
T2
T3
T4
T5
T6
T7


Average Growth of Sweetpotato Grown in Samar Soil
Plant Height cm
150.00
100.00
50.00
0.00
Week 2
Week 4
Week 6
Week 8
Week...
Week...
Week...
Week...
Week...
Weeks after Planting
T1
T2
T3
T4
T5
T6
T7


Average Growth of Sweetpotato Grown in Acidic soil of Matalom
Plant Height cm
300.00
200.00
100.00
0.00
Week 2
Week 4
Week 6
Week 8
Week 10
Week 12
Week 14
Week 16
Week 18
Weeks after Planting
T1
T2
T3
T4
T5
T6
T7


Average Growth of Sweetpotato Grown in Basic soil of Matalom
Plant Height cm
250.00
200.00
150.00
100.00
50.00
0.00
Week 2
Week 4
Week 6
Week 8
Week 10
Week 12
Week 14
Week 16
Week 18
Weeks after Planting
T1
T2
T3
T4
T5
T6
T7

Average main vine length (cm) of sweet potato (LSU-25 variety) before harvest and grown under different soil types as affected by the varying sources and rates of micro nutrient fertilizers.

	Soil Type				
Treatments	VSU Soil	Ormoc Soil	Matalom Soil-Acidic	Matalom Soil-Basic	Samar Soil
T1 = control	111.6 D	118.1 B	115.3 B	109.5 C	95.5 B
T2 = 60-60-60 kg/ha NPK	178.1 CD	252.6 A	167.7AB	181.0 B	115.33 AB
T3 = T2 + 8 Kg/h Biozome 200	290.5 A	281.2 A	214.9 A	205.3 AB	133.0 A
T4 = T2 + Mycovam-1 (5 g/plant)	199.1 BC	211.2 AB	182.8 AB	193.3 AB	106.35 AB
T5 = T2 + 4 Kg/ha microbial inoculant	242.7 ABC	254.5 A	222.8 A	202.2 AB	121.4 A
T6 = T2 + 8 Kg/ha microbial inoculant	225.8 ABC	235.0 AB	205.9 A	206.5 AB	123.0 A
T7 = T2 + 12 Kg/ha microbial inoculant	259.3AB	249.3 A	197.5 A	217.3 A	128.0 A
Average	349.66	228.86	186.69	187.90	117.50

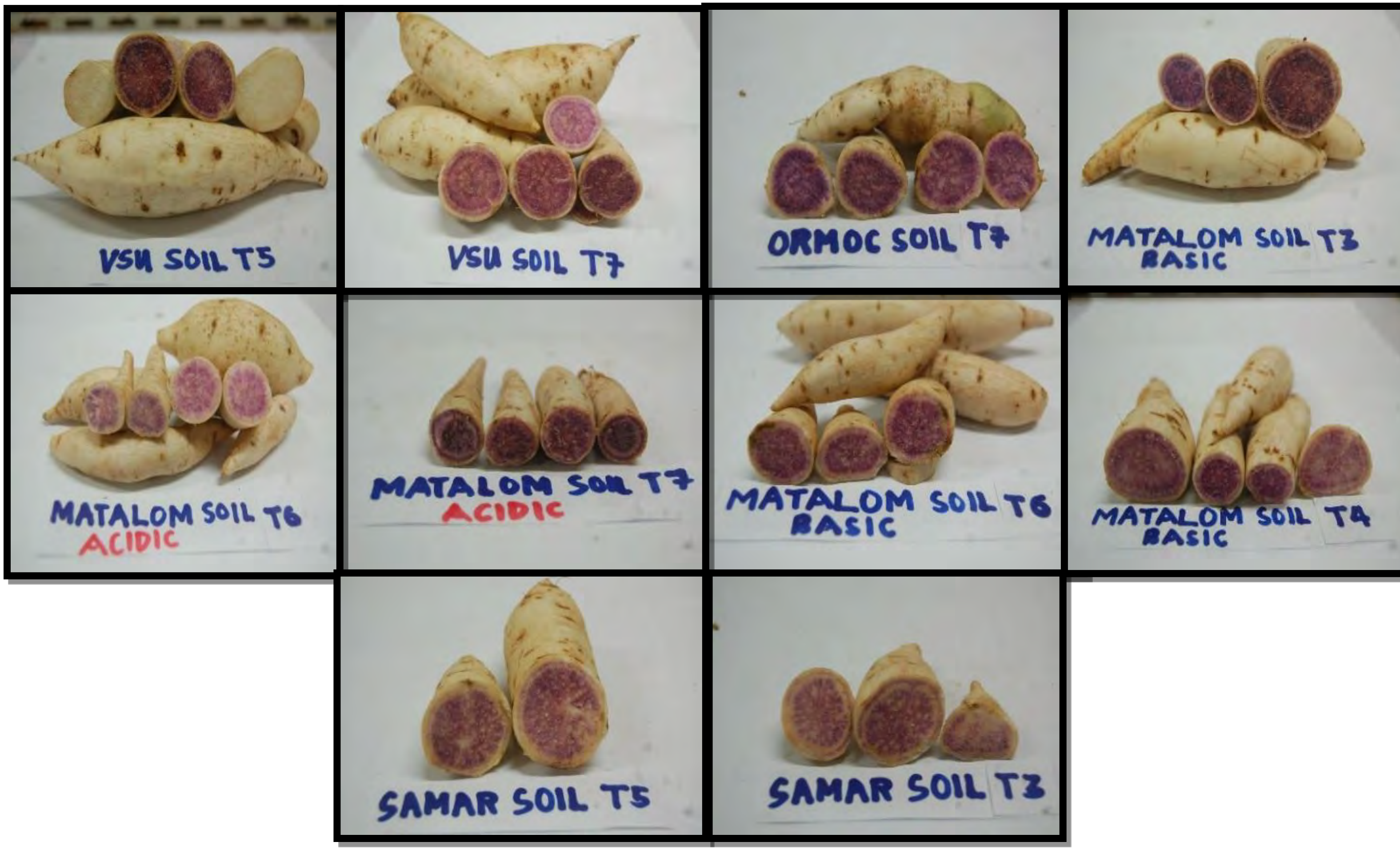
Note: Means followed by the same letter are not significantly different from each other at 1% α.

Yield (g/pot) of marketable sweet potato tubers (LSU-variety) grown under different soil types supplemented by the varying sources and rates of micro nutrient fertilizers.

	Soil Type				
Treatments	VSU Soil	Ormoc Soil	Matalom Soil-Acidic	Matalom Soil-Basic	Samar Soil
T1 = control	193.19 C	181.03 B	161.71 B	192.16 B	88.70 B
T2 = 60-60-60 kg/ha NPK	237.68 C	228.81 B	219.33 AB	232.02 AB	116.64 AB
T3 = T2 + 8 Kg/h Biozome 200	548.21 A	443.91 A	280.35 A	260.05 A	132.02 A
T4 = T2 + Mycovam-1 (5 g/plant)	380.37 B	400.05 A	211.02 AB	246.70 AB	103.40 AB
T5 = T2 + 4 Kg/ha microbial inoculant	446.39 AB	402.53 A	231.41 AB	249.62 AB	121.28 AB
T6 = T2 + 8 Kg/ha microbial inoculant	465.51 AB	426.18 A	255.32 AB	287.72 A	102.39 AB
T7 = T2 + 12 Kg/ha microbial inoculant	503.69 AB	419.65 A	241.45 AB	260.14 A	123.56 AB
Average	396.43	357.45	228.66	246.92	112.57

Note: Means followed by the same letter are not significantly different from each other at 1% α.

Treatments with the highest yield (g/pot) of marketable sweet potato tubers grown in different locations.



Treatments with the highest yield (g/pot) of marketable sweet potato tubers grown in different locations. T3 (60-60-60 kg/ha N-P-K + 8 kg/ha Biozome-200), T4 (60-60-60 kg/ha N-P-K +Mycovam 5g/plant inoculant), T5 (60-60-60 kg/ha N-P-K + 4 kg/ha microbial inoculant) T6 (60-60-60 kg/ha N-P-K + 8 kg/ha microbial inoculant) and T7 (60-60-60 kg/ha N-P-K + 12 kg/ha microbial inoculant).

Nutrient Uptake Analysis

Initial and final soil analysis of VSU (neutral) soil of Baybay City, Leyte.

Nutrients Applied	Initial Nutrient Content - Start of Study							
Soil type - VSU Soil			Exchangeable mg/kg			Extractable mg/kg		
Treatment	Total N %	Available P*	K*	Ca*	Mg*	Fe*	Cu	Zn
T1 = control	0.16	17.56	125.33	1882.63	984.25	50.82	2.66	4.81
T2 = 60-60-60 kg/ha NPK	0.18	21.96	146.96	2001.25	889.65	61.71	2.57	4.06
T3 = T2 + 8 Kg/h Biozome 200	0.27	26.58	161.11	2093.50	1095.18	66.22	2.64	5.26
T4 = T2 + Mycovam-1 (5 g/plant)	0.22	19.34	231.65	1972.75	909.08	58.83	2.70	4.10
T5 = T2 + 4 Kg/ha microbial inoculant	0.23	20.16	187.71	2084.50	1013.43	65.53	3.01	5.81
T6 = T2 + 8 Kg/ha microbial inoculant	0.26	20.62	156.31	2004.75	1008.18	66.86	2.64	5.24
T7 = T2 + 12 Kg/ha microbial inoculant	0.21	26.46	160.84	1990.50	952.40	65.97	2.89	5.60
	Nutrient Content at the End of study							
			Exchangeable mg/kg			Extractable mg/kg		
	Total N %	Available P*	K**	Ca	Mg*	Fe	Cu	Zn
T1 = control	0.19	90.32	68.59	2479.00	563.98	54.20	2.43	4.38
T2 = 60-60-60 kg/ha NPK	0.20	126.96	75.89	2550.43	885.48	54.57	2.60	5.59
T3 = T2 + 8 Kg/h Biozome 200	0.24	139.28	182.89	2795.20	1179.83	57.71	2.69	8.15
T4 = T2 + Mycovam-1 (5 g/plant)	0.20	120.16	148.16	2552.45	971.91	62.36	2.66	5.98
T5 = T2 + 4 Kg/ha microbial inoculant	0.24	114.32	135.07	2801.25	1117.83	60.54	2.83	5.88
T6 = T2 + 8 Kg/ha microbial inoculant	0.21	161.92	195.19	2748.50	1240.80	60.53	2.71	6.54
T7 = T2 + 12 Kg/ha microbial inoculant	0.22	96.00	132.04	2561.50	1000.05	61.19	2.93	7.51

Nutrient Uptake Analysis

Initial and final soil analysis of volcanic soil of Ormoc City, Leyte.

Table 9. Initial soil analysis of disturbed volcanic soil of Ormoc city, Leyte.

Nutrients Applied	Initial Nutrient Content - Start of Study							
Soil type -Ormoc Soil	Total	Available	Exchangeable mg/kg			Extractable mg/kg		
Treatment	**N (%)**	**P**	**K***	**Ca***	**Mg***	**Fe**	**Cu**	**Zn**
T1 = control	0.35	9.56	187.20	311.00	80.58	70.29	1.35	0.83
T2 = 60-60-60 kg/ha NPK	0.50	15.22	192.70	416.25	89.20	72.35	1.47	1.18
T3 = T2 + 8 Kg/h Biozome 200	0.40	12.98	330.19	717.75	181.05	79.54	1.50	2.05
T4 = T2 + Mycovam-1 (5 g/plant)	0.47	12.36	272.74	431.25	102.90	73.13	1.54	1.65
T5 = T2 + 4 Kg/ha microbial inoculant	0.45	12.00	206.41	802.00	145.58	74.84	1.71	1.11
T6 = T2 + 8 Kg/ha microbial inoculant	0.50	11.28	217.84	1021.25	96.35	75.56	1.65	1.33
T7 = T2 + 12 Kg/ha microbial inoculant	0.47	11.18	219.39	574.50	140.03	77.17	1.79	1.10
	Nutrient Content at the End of study							
	Total	Available	Exchangeable mg/kg			Extractable mg/kg		
	N (%)	**P***	**K***	**Ca***	**Mg***	**Fe***	**Cu**	**Zn**
T1 = control	0.10	12.40	10.28	358.75	94.98	35.57	1.15	3.89
T2 = 60-60-60 kg/ha NPK	0.10	45.04	12.18	385.50	101.05	40.12	1.30	4.08
T3 = T2 + 8 Kg/h Biozome 200	0.10	58.40	61.61	517.75	135.88	44.72	1.36	5.45
T4 = T2 + Mycovam-1 (5 g/plant)	0.11	85.12	44.27	389.00	122.40	57.42	1.25	4.47
T5 = T2 + 4 Kg/ha microbial inoculant	0.19	120.32	45.61	421.75	111.05	54.36	1.53	7.21
T6 = T2 + 8 Kg/ha microbial inoculant	0.11	140.00	52.26	396.00	116.83	59.66	1.41	5.42
T7 = T2 + 12 Kg/ha microbial inoculant	0.12	84.88	50.03	464.00	142.70	47.84	1.27	5.51

Nutrient Uptake Analysis

Initial and final soil analysis of acidic soil of Matalom, Leyte.

Table 10. Initial soil analysis of the acidic soil of Matalom, Leyte.

Nutrients Applied	Initial Nutrient Content - Start of Study							
Soil type -Matalom Acidic Soil	Total	Available	Exchangeable mg/kg			Extractable mg/kg		
Treatment	**N (%)**	**P***	**K***	**Ca**	**Mg**	**Fe***	**Cu**	**Zn**
T1 = control	0.14	7.78	71.94	<0.000	46.88	38.04	1.57	1.66
T2 = 60-60-60 kg/ha NPK	0.13	10.90	79.79	<0.001	35.63	49.89	1.72	2.36
T3 = T2 + 8 Kg/h Biozome 200	0.23	14.18	96.01	<0.001	36.95	51.48	1.70	3.05
T4 = T2 + Mycovam-1 (5 g/plant)	0.20	10.06	86.40	<0.001	21.98	48.04	1.78	5.13
T5 = T2 + 4 Kg/ha microbial inoculant	0.13	13.20	94.48	<0.001	30.08	56.76	1.54	3.08
T6 = T2 + 8 Kg/ha microbial inoculant	0.21	13.90	127.10	<0.001	23.90	55.98	1.75	1.67
T7 = T2 + 12 Kg/ha microbial inoculant	0.16	25.42	106.31	<0.001	23.18	59.49	1.58	3.51

	Nutrient Content at the End of study							
	Total	Available	Exchangeable mg/kg			Extractable mg/kg		
	N (%)	**P***	**K***	**Ca**	**Mg**	**Fe***	**Cu**	**Zn**
T1 = control	0.16	64.96	56.25	<.003	22.55	36.69	2.22	2.93
T2 = 60-60-60 kg/ha NPK	0.17	85.52	63.75	<.003	22.75	53.15	2.27	4.06
T3 = T2 + 8 Kg/h Biozome 200	0.20	156.24	61.00	<.003	24.88	52.91	2.34	5.55
T4 = T2 + Mycovam-1 (5 g/plant)	0.20	108.88	63.75	<.003	37.45	55.54	2.74	4.04
T5 = T2 + 4 Kg/ha microbial inoculant	0.18	123.36	64.50	<.003	30.65	52.05	2.96	3.57
T6 = T2 + 8 Kg/ha microbial inoculant	0.20	92.16	68.00	<.003	35.63	58.54	3.13	3.10
T7 = T2 + 12 Kg/ha microbial inoculant	0.17	132.72	81.25	<.003	44.75	46.60	2.67	5.35

Nutrient Uptake Analysis

Initial and final soil analysis of basic soil of Matalom, Leyte.

Table 11. Initial soil analysis of the basic soil of Matalom, Leyte.

Nutrients Applied	Initial Nutrient Content - Start of Study							
Soil type -Matalom Basic Soil	Total	Available	Exchangeable mg/kg			Extractable mg/kg		
Treatment	**N (%)**	**P***	**K***	**Ca***	**Mg****	**Fe***	**Cu**	**Zn**
T1 = control	0.13	14.72	93.73	1252.00	40.99	38.99	1.57	1.65
T2 = 60-60-60 kg/ha NPK	0.18	16.16	135.55	1872.88	42.15	40.32	1.94	2.00
T3 = T2 + 8 Kg/h Biozome 200	0.26	30.52	443.00	1931.50	264.85	69.89	2.15	2.61
T4 = T2 + Mycovam-1 (5 g/plant)	0.21	23.36	283.90	1796.50	165.10	40.50	1.95	2.01
T5 = T2 + 4 Kg/ha microbial inoculant	0.21	36.02	361.91	1782.00	195.13	51.68	1.91	2.22
T6 = T2 + 8 Kg/ha microbial inoculant	0.28	38.70	174.38	1576.50	52.10	55.15	1.86	5.72
T7 = T2 + 12 Kg/ha microbial inoculant	0.21	29.24	168.96	1360.25	36.45	44.97	1.59	2.07
	Nutrient Content at the End of study							
	Total	Available	Exchangeable mg/kg			Extractable mg/kg		
	N (%)	**P***	**K***	**Ca***	**Mg***	**Fe**	**Cu**	**Zn**
T1 = control	0.15	50.72	96.44	848.50	43.18	27.63	1.76	1.84
T2 = 60-60-60 kg/ha NPK	0.20	64.48	116.83	1192.80	63.23	41.07	1.79	2.60
T3 = T2 + 8 Kg/h Biozome 200	0.25	206.80	350.27	1964.85	227.65	54.38	2.10	2.95
T4 = T2 + Mycovam-1 (5 g/plant)	0.20	126.48	154.65	1222.25	74.10	42.22	2.55	2.89
T5 = T2 + 4 Kg/ha microbial inoculant	0.19	279.92	217.43	1749.58	209.20	53.21	2.11	2.77
T6 = T2 + 8 Kg/ha microbial inoculant	0.21	139.68	145.80	1711.75	154.35	46.56	1.80	3.24
T7 = T2 + 12 Kg/ha microbial inoculant	0.18	131.20	131.66	1659.73	160.58	46.78	2.44	3.88

Table 12. Initial tissue analysis of sweetpotato (leaves) before planting.

Initial Tissue Analysis	Total	Available	Exchangeable mg/kg			Extractable mg/kg		
	N (%)	**P**	**K**	**Ca**	**Mg**	**Fe**	**Cu**	**Zn**
	1.19	**1.06**	**2.13**	**0.94**	**0.16**	**885.95**	**23.5**	**75.48**

Table 13. Tissue analysis of sweetpotato grown under VSU (neutral) soil of Baybay city, Leyte.

Nutrients Applied	**Tissue Analysis after harvest**							
Soil type - VSU Soil			Exchangeable mg/kg			Extractable mg/kg		
Treatment	**Total N %**	**Available P**	**K**	**Ca**	**Mg**	**Fe***	**Cu**	**Zn**
T1 = control	1.27	1.17	2.76	1.23	0.29	1027.20	22.00	57.12
T2 = 60-60-60 kg/ha NPK	1.37	1.28	2.75	1.05	0.21	970.85	26.15	57.19
T3 = T2 + 8 Kg/h Biozome 200	1.56	1.73	2.69	1.88	0.23	1330.35	28.65	62.80
T4 = T2 + Mycovam-1 (5 g/plant)	1.51	1.37	2.58	1.13	0.26	1175.50	24.55	73.07
T5 = T2 + 4 Kg/ha microbial inoculant	1.64	1.48	2.76	1.50	0.39	1299.15	26.30	61.93
T6 = T2 + 8 Kg/ha microbial inoculant	1.56	1.69	2.88	1.07	0.29	1238.70	42.50	113.52
T7 = T2 + 12 Kg/ha microbial inoculant	1.65	1.49	2.91	1.94	0.45	1182.05	24.70	100.39

Table 14. Tissue analysis of sweetpotato grown under the disturbed volcanic soil of Ormoc City, Leyte.

Nutrients Applied	**Tissue Analysis after harvest**							
Soil type - Ormoc Soil			Exchangeable mg/kg			Extractable mg/kg		
Treatment	**Total N**	**Available P**	**K**	**Ca**	**Mg***	**Fe***	**Cu**	**Zn**
T1 = control	1.34	1.19	2.57	1.02	0.18	696.70	20.55	51.25
T2 = 60-60-60 kg/ha NPK	1.48	1.24	2.81	1.20	0.14	1018.05	23.45	65.40
T3 = T2 + 8 Kg/h Biozome 200	1.65	1.29	2.66	1.95	0.18	1089.55	25.80	98.88
T4 = T2 + Mycovam-1 (5 g/plant)	1.52	1.16	2.58	1.02	0.19	886.00	20.10	68.54
T5 = T2 + 4 Kg/ha microbial inoculant	1.51	1.46	2.89	1.02	0.27	1050.95	22.05	81.46
T6 = T2 + 8 Kg/ha microbial inoculant	1.84	1.41	2.96	1.05	0.34	1360.70	23.85	100.56
T7 = T2 + 12 Kg/ha microbial inoculant	1.48	2.54	2.84	2.54	0.52	1591.00	36.05	166.87

Table 15. Tissue analysis of sweetpotato grown under the acidic soil of Matalom, Leyte.

Nutrients Applied	**Tissue Analysis after harvest**							
Soil type - Matalom Acidic Soil			Exchangeable mg/kg			Extractable mg/kg		
Treatment	**Total N %**	**Available P**	**K**	**Ca**	**Mg**	**Fe**	**Cu**	**Zn**
T1 = control	0.92	1.04	2.47	0.41	0.14	1303.50	13.20	57.21
T2 = 60-60-60 kg/ha NPK	1.04	1.47	2.66	0.76	0.14	1368.90	15.85	53.84
T3 = T2 + 8 Kg/h Biozome 200	1.07	1.56	2.96	0.68	0.12	1293.50	23.35	65.39
T4 = T2 + Mycovam-1 (5 g/plant)	1.26	1.72	2.77	0.53	0.17	1709.95	26.20	52.77
T5 = T2 + 4 Kg/ha microbial inoculant	1.21	1.52	2.72	0.69	0.17	1774.65	17.65	62.05
T6 = T2 + 8 Kg/ha microbial inoculant	1.15	1.38	2.77	0.97	0.15	1640.55	27.85	53.74
T7 = T2 + 12 Kg/ha microbial inoculant	1.34	1.49	2.66	0.62	0.14	1842.65	15.95	88.17

Table 16. Tissue analysis of sweetpotato grown under the basic soil of Matalom, Leyte.

Nutrients Applied	Tissue Analysis after harvest							
Soil type - Matalom Basic Soil			Exchangeable mg/kg			Extractable mg/kg		
Treatment	Total N	Available P	K	Ca	Mg	Fe	Cu	Zn
T1 = control	0.89	1.33	2.56	1.17	0.16	781.65	14.90	37.08
T2 = 60-60-60 kg/ha NPK	1.20	1.51	2.66	0.98	0.20	869.55	19.35	50.55
T3 = T2 + 8 Kg/h Biozome 200	1.76	1.77	2.74	1.24	0.14	935.70	28.35	62.23
T4 = T2 + Mycovam-1 (5 g/plant)	1.51	1.64	2.78	1.07	0.19	891.25	21.50	52.20
T5 = T2 + 4 Kg/ha microbial inoculant	1.34	1.49	2.76	1.55	0.12	1228.55	20.05	60.96
T6 = T2 + 8 Kg/ha microbial inoculant	1.34	1.44	2.82	1.42	0.20	1057.30	23.60	74.26
T7 = T2 + 12 Kg/ha microbial inoculant	1.17	1.63	2.86	1.31	0.14	1222.80	25.19	71.27

Table 17. Tissue analysis of sweetpotato grown under Samar (abandoned mine site) soil.

Nutrients Applied	Tissue Analysis after harvest							
Soil type - Samar Soil			Exchangeable mg/kg			Extractable mg/kg		
Treatment	Total N %	Available P	K	Ca	Mg	Fe	Cu	Zn
T1 = control	1.06	1.16	2.58	0.79	0.15	1181.05	19.90	117.16
T2 = 60-60-60 kg/ha NPK	1.62	1.25	2.79	0.79	0.22	1309.10	20.35	275.79
T3 = T2 + 8 Kg/h Biozome 200	1.86	1.30	2.90	1.82	0.47	1393.75	33.00	125.78
T4 = T2 + Mycovam-1 (5 g/plant)	1.67	1.49	2.84	1.02	0.27	1636.05	25.75	158.74
T5 = T2 + 4 Kg/ha microbial inoculant	1.76	1.26	2.91	1.03	0.22	1412.55	25.20	162.93
T6 = T2 + 8 Kg/ha microbial inoculant	1.65	1.63	2.83	0.69	0.28	1777.25	30.65	227.11
T7 = T2 + 12 Kg/ha microbial inoculant	1.62	1.87	2.88	0.97	0.30	1551.65	28.55	151.85

Table 7. Anthocyanin content of sweetpotato LSU-25 variety as influence by varying rates micronutrient supplement and microbial inoculants grown under five different soil types.

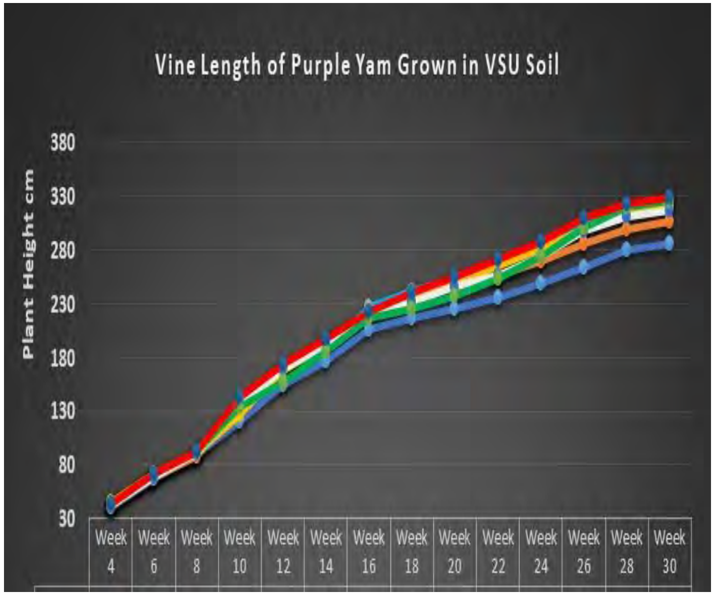
Treatment	Anthocyanin Content (ml/L)				
	VSU soil	Ormoc Soil	Matalom Acidic Soil	Matalom Basic Soil	Samar Soil
T1	0.18	2.50	2.63	2.31	0.81
T2	0.72	2.79	2.19	1.23	0.62
T3	0.06	2.66	2.35	3.10	2.54
T4	2.46	2.46	0.09	2.28	0.50
T5	2.48	2.80	3.27	2.42	2.67
T6	0.09	2.37	1.05	2.38	2.82
T7	2.51	2.60	3.07	2.71	2.53

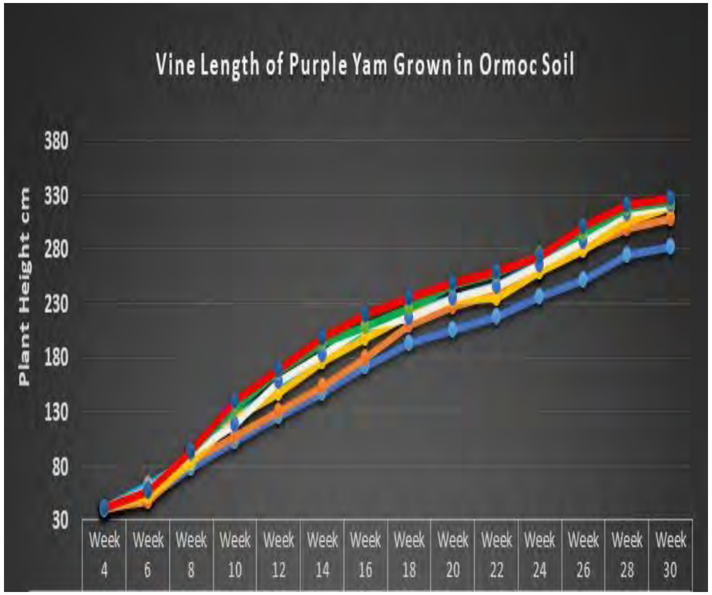
Table 21. Anthocyanin content of sweetpotato LSU-25 variety as influence by varying rates of micronutrient supplement and microbial inoculant grown under five different soil types.

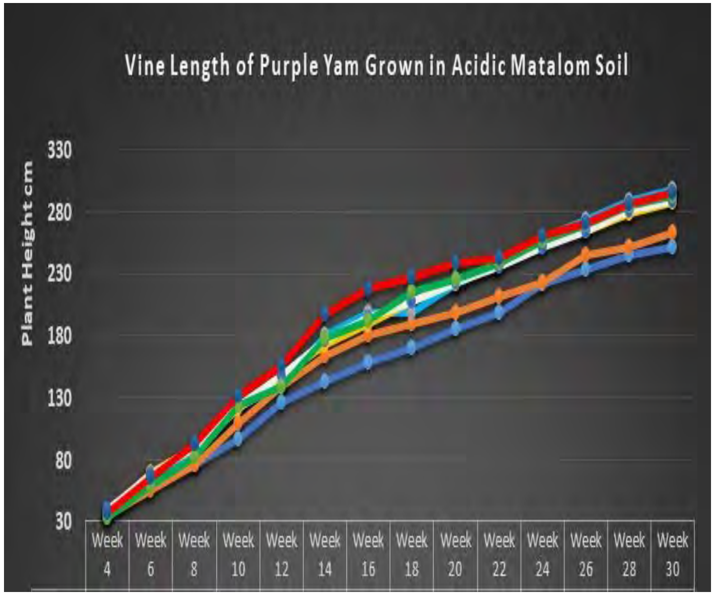
Anthocyanin Content

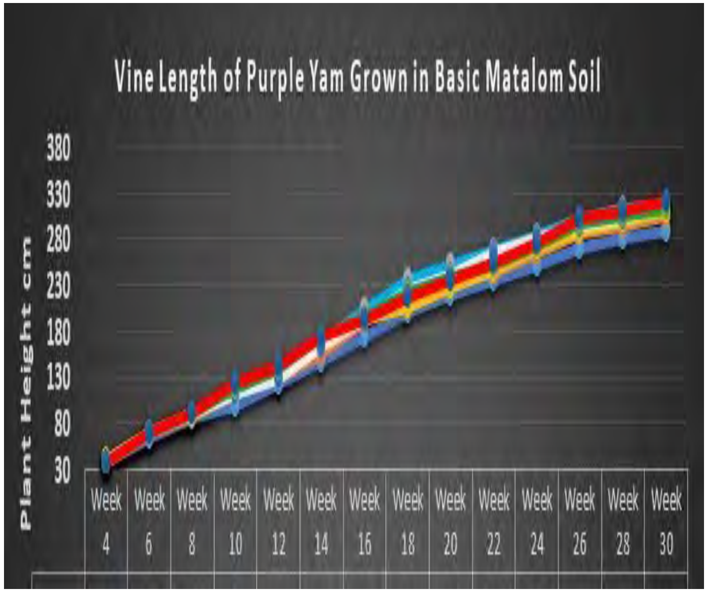
(ml/L)

POT EXPERIMENT OF PURPLE YAM VU-2 VARIETY.


Vine Length of Purple Yam Grown in VSU Soil
Plant Height cm
380
330
280
230
180
130
80
30
Week 4
Week 6
Week 8
Week 10
Week 12
Week 14
Week 16
Week 18
Week 20
Week 22
Week 24
Week 26
Week 28
Week 30


Vine Length of Purple Yam Grown in Ormoc Soil
Plant Height cm
380
330
280
230
180
130
80
30
Week 4
Week 6
Week 8
Week 10
Week 12
Week 14
Week 16
Week 18
Week 20
Week 22
Week 24
Week 26
Week 28
Week 30


Vine Length of Purple Yam Grown in Acidic Matalom Soil
Plant Height cm
330
280
230
180
130
80
30
Week 4
Week 6
Week 8
Week 10
Week 12
Week 14
Week 16
Week 18
Week 20
Week 22
Week 24
Week 26
Week 28
Week 30


Vine Length of Purple Yam Grown in Basic Matalom Soil
Plant Height cm
380
330
280
230
180
130
80
30
Week 4
Week 6
Week 8
Week 10
Week 12
Week 14
Week 16
Week 18
Week 20
Week 22
Week 24
Week 26
Week 28
Week 30

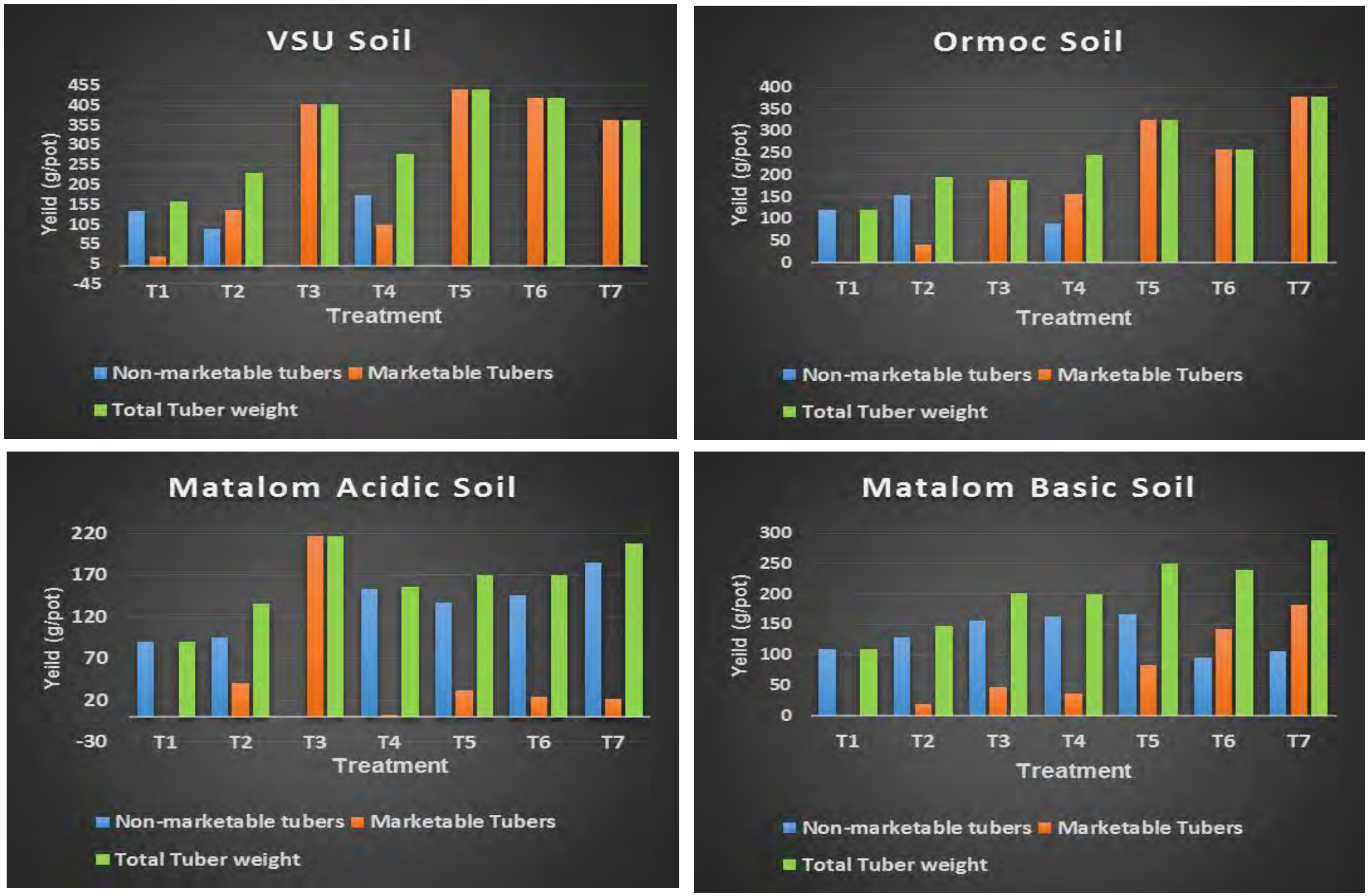


Fig. 19. Marketable and non-marketable yields of purple yam (VU2 var.) grown in different locations and treatments.

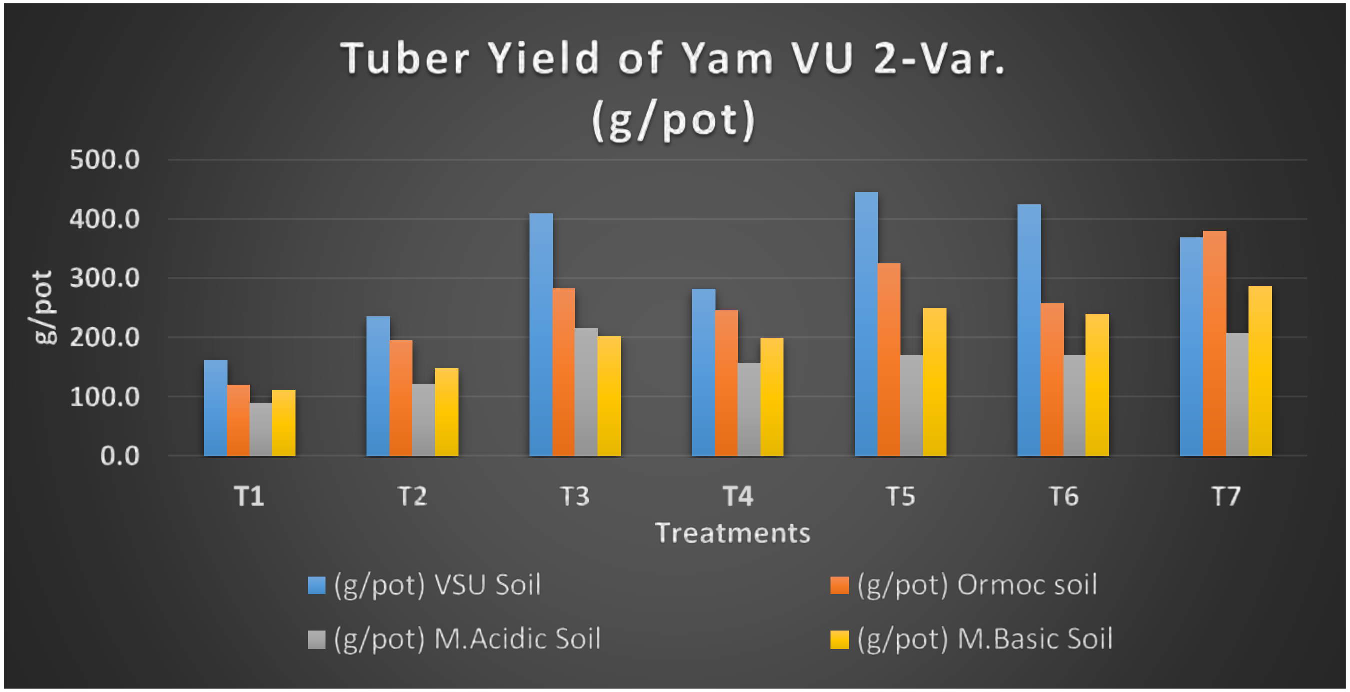
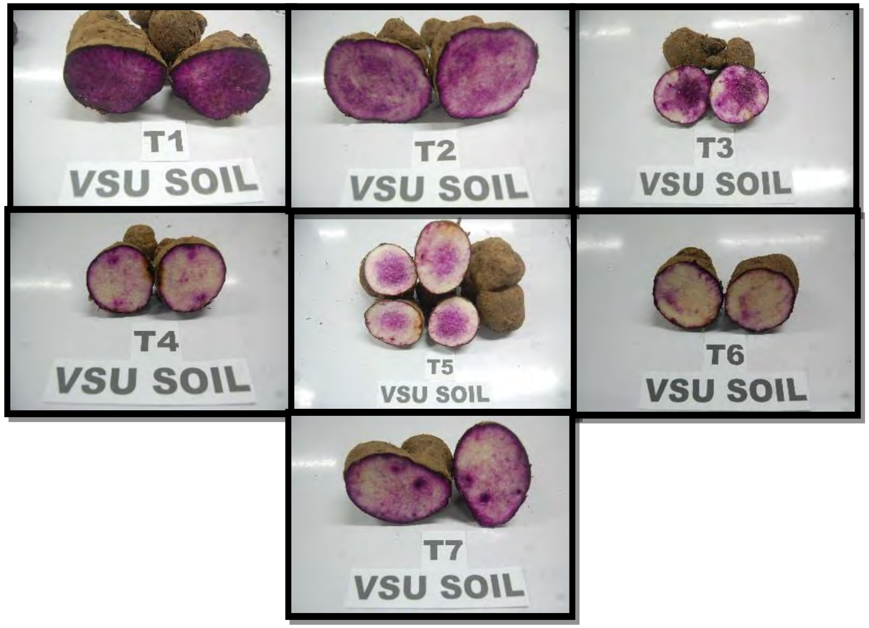
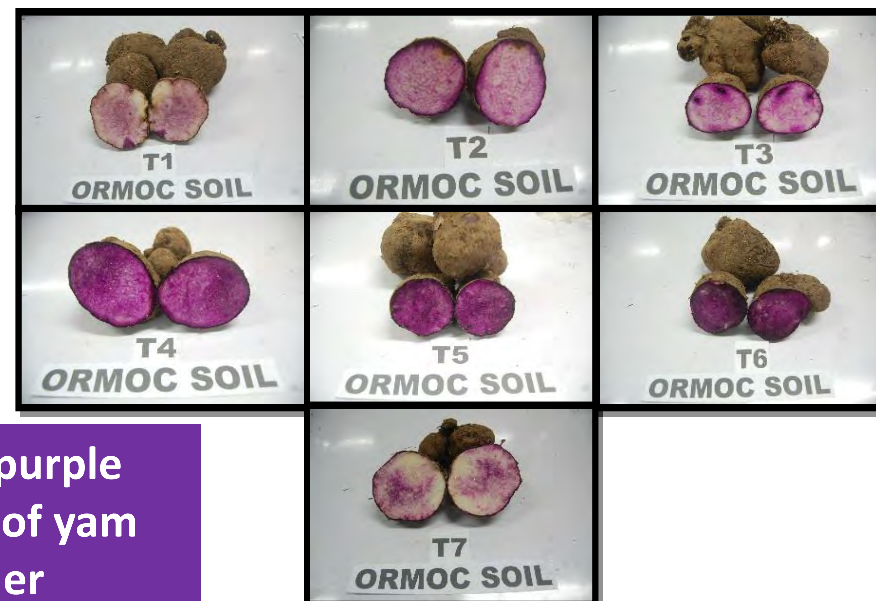


Fig. 18. Tuber yield of purple yam VU-2 grown in different locations as supplemented by varying rates of commercial and organic-based microbial inoculant.

Anthocyanin contents of purple yam VU-2 variety affected by soil types and fertilizer application.

Anthocyanin Content (ml/l)				
Treatment	VSU soil	Ormoc Soil	Matalom Acidic Soil	Matalom Basic Soil
T1	4.65	1.09	3.74	1.12
T2	2.40	1.16	4.83	1.08
T3	1.96	1.18	2.91	4.59
T4	0.94	4.77	4.99	4.41
T5	1.09	4.90	4.75	5.08
T6	0.87	4.97	4.62	4.78
T7	2.95	1.11	5.17	4.53





Images of purple coloration of yam grown under various soils.

Summary

- The microbial inoculant developed in this work was able to increase tuber yield of sweetpotato and purple yam which is comparable or even better than the performance of the crops applied with commercially available micronutrient fertilizers Biozome-200 and another microbial inoculant, Mycovam.
- Higher micronutrient contents of sweetpotato was also obtained from plants applied with the microbial inoculant than the control plants which was not applied with any fertilizer or with complete fertilizer only.

Next Steps

- Conduct field trial to assess and validate the effectivity of the developed microbial inoculants in actual field conditions particularly in areas with marginal soil characteristics as tested in this study.

- Conduct pot and field trials using other short-term crops such as vegetables (cabbage, lettuce, sweet pepper) to evaluate wider applicability and suitability of the microbial inoculants.

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- **Philrootcrops**
- **LGU’s of Samar, Matalom and Ormoc City**

Thank you for your attention.

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