Isolation of Mycorrhiza and Its Effect on Yield of Zea mays (L.) var. rugosa Bonaf. (Sweet corn)

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Abstract:- In this experiments were carried out to isolation of mycorrhiza from roots and spore population from rihzosphere in ten species of angiosperms and to investigate the effect of mycorrhiza fertilizer on the yield of Zea mays (L.) var. rugosa Bonaf. (sweet corn) in Magway University during February to December 2018. These experiments included laboratory, pot were conducted at Magway University. In laboratory experiment mycorrhiza infection of roots were calculated by grid-line intersection method and spore population were collected by spore decanting method. The highest infection was observed in roots of Dactylotenium aegyptium (L.)Willd. (72.1%) and the Lowest infection was occurred in *Phyllanthus urinaria* L. (19.5 %). Highest number of spore was observed in rhizosphere of Chloris barbata SW. (55). and lowest number of spore was observed in Leucas aspera (Willd.) Spreng (13). In pot experiment, the effect of mycorrhiza infected grass roots on yield of sweet corn were also investigated. It was observed that yield of treated plants were significantly higher than the control plants at 5% level. These results also indicated that yield parameter of the earliest day to 50% tasseling and earing was found in treated with VAM , and the latest was observed in the control. The yield component of ear yield ,ear size, ear weight and 1000 grains weight were obviously increased the effect of treated with VAM, than control at harvested time According to result suggested that, the use of mycorrhiza had a higher positive effect of yield on Zea may s(L.) var rugosa Bonaf.(sweet corn).

I. INTRODUCTION

The term mycorrrhiza was first used to fungus-root and originates from Greek myco, meaning 'fungus' and rhiza, meaning 'root'. Mycorrhizal are symbiotic associations, formed between plants and soil fungi that play an essential role in plant growth, plant protection and soil quality(Frank 1885). General nature of VAM to plant are; (1) Increase plant nutrient supply by extending the volume of soil accessible to plants. (2) Increase plant nutrient supply by acquiring nutrient forms that would not normally be available to plants. (3) Root colonization by VAM fungi can provide protection from parasitic fungi and nematodes. (4) Non nutritional benefits to plants due to change in wate relation, phytohormone levels, carbon assimilation, etc. (5) Mycorrhizal benefit can include greater yield, nutrient accumulation, and/or reproductive success (Brundrett et al. 1990).

Present agricultural system in Myanmar follow the traditional methods which utilize the available natural resources combined with improved cultural practices. Microbiological fertilizer, more commonly known as biofertilizer, include phosphate solubilizing and plant growth promoting microorganism (Goel 1999). Among biofertilizer benefiting the crop production are phosphate solubilizing microorganism and mycorrhiza fungi (Hedge *et al.* 1999).

II. STUDY OF MYCORRHIZA INFECTION AND SPORE POPULATION OF TEN SPECIES IN ANGIOSPERMS FROM MAGWAY UNIVERSITY CAMPUS

➤ Collection of sample site

In this research, roots and rhizospheres soil sample were collected from the growing ten species of angiosperms in Magway University. These are *Chloris barbata* SW., *Dactylotenium aegyptium* (L.) Willd., *Eragrostis cilianensis* (All.) Vignolo-Lut, *Eragrostis theinlwinii* Bor *Setaria verticillata* (L.)P.Beauv., *Lycopersicum esculentum* Miller *Leucas aspera* (Willd.) Spreng , *Helianthus annus* L., *Wedelia calendulacea* Less. and *Phyllanthus urinaria* L..

In this collection procedure, the soil sample collected from the plant base weighing 200 gm was dug about 5 cm away from the stem of each plant at the depth of 10 cm. The collected species of plants were mounted in the herbarium sheets. The collected soil samples were packed in plastic bags. The root samples were used freshly for isolation of mycorrhiza.

> Plant Identification

According to the morphology and floral characteristics, the collected plants were identified by using Backer (1963-1968), Cronquist (1981), Dassanayake (1980-2001), Heywood (1978), Hooker (1897), Kress (2003), Lawrence (1951).

> Mycorrhiza colonization by staining method

The root preliminary surveying and observation of mycorrhizal colonization by staining INVAM (International culture collection of vascular Arbuscular Mycorrhizal Fungi) method. The collected root samples were washed in water to remove adhering soil and sands. After through washing the fresh roots samples were removed all particulates and roots were cut into (1 cm) of root. About 20 segments (1 cm) of root from each plant was cut. They were cleared and stained to determine the intensity of VAM infection using the staining method (Phillips and Hayman 1970). Then the roots were boiled into 10% KOH for 10-15 minutes and after that washed the roots fragments with water.

Acidified into 2% HCL for about 10 minutes. Afterward, the stain was poured off into another container and the roots were immersed in the water in small beaker. Wash again in water and stained with hot 0.05% acidfuchsine for about 20 minute, the stain was prepared by mixing with water, glycerin and lactic acid (1:1:1). These roots were placed on the slides and the slides were studied under microscope.

➢ Mycorrhizal colonization analysis

Mycorrhizal colonization was calculated by grid–line intersection method Newman (1996) 20 fragments of stained roots were randomly placed in petridish (2 cm diameter) which has gird lines of 0.5 cm square block line Counted the intersection of mycorrhizal mycelia on the grid - line under microscope. The percent of VAM infection was determined by the following formula:

Percent of infection = $\frac{\text{Intersection of infected roots}}{\text{Total number of intersection roots}} \times 100$

Extraction of spores from rhizosphere

Mycorrhizal spores extraction were carried out from rhizosphere soil of ten species of angiosperms, which was naturally grown in Magway University by wet sieving and spore decanting method (Gerdeman and Nicolson 1963).

Take about 50 g of freshly soil with cut root sample from rhizosphere were taken. These samples were placed in a blender with small amount of water and blended at high speed for just about five second to break up root fragments. The spores were released and liberated from hyphal aggregate attached of roots and from the soil. After that the blended materials immediately poured through three sieves. Most sand remains in the blender. The spore were isolated from the collected soil by passing a suspension of soil through a series of sieves (500µm, 200µm, 100µm, 45µm) respectively. The spore size of top sieve generally has 500µm. The capture root debris and large spores. The middle sieve was 100µm and the bottom sieve has about 45µm. The bottom sieve captures the majority of spores. All the spore were isolated from the extract with a fine forceps into a watch glass with small quantity of water. The extract with AM spores were observed under microscope and number of spores were counted.



Fig 1:- Fragment of roots



Fig 2:- staining roots

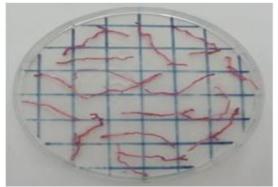


Fig 3:- Grid line intersection method



Fig 4:- Extraction of spore population procedure

> Preparation of materials

Firstly, soil were exactly weighed to get 5kg for each pot $(37.5 \text{cm} \times 50 \text{cm})$. Two kg of mycorrhiza were taken for each bag and then mixed thoroughly with the soils. In this experiment, totally 20 pots were used (10 for control and 10 for treatment). The seeds of sweet corn were loosely wrapped and soaked in water over night (at least 12 hours). Only three seeds were directly sown into the individual bags and regularly watered daily. In each pot, only one healthy seedling was left and the rest plants were thinned from the bags. During the planting sweet corn, the crop management and cultural practices such as spraying of water and weeds control were done when necessary.

Data collection and statistical analysis

Data were collected at weekly intervals as the follow: day of 50% tasselling and earing days, number of ear per plants, ear size (length and width), ear weight (husk and dehusked) and row per ear for yield. These data were statistical analysed by using Gomaz and Gomaz (1984) IRRI STAT(2000) software program and LSD (Least significant difference) test, Friedman test (F-test), and Value of Coefficient for variation percent (CV%) was used to analyze data for the variation.

Data of soil analysis

Soil samples were analyzed for their physical and chemical properties at Soil Analysis Laboratory of Myanmar Agriculture service (Land use), Insein Township, Yangon. The results of soil samples are described in Table 4.

III. RESULTS

In this research, roots and rhizospheres soil sample were collected from the growing ten species of angiosperms in Magway University during February to December 2018. The mycorrhiza infection in root of and the collected spore number were counted from rhizosphere were shown in Table 1.

No.	Family	Scientific name	Spore Number (50g Soil)	Mycorrhiza Infection (%)
1	Poaceae	Chloris barbata SW .	55	63.4
2	Poaceae	Dactyloctenium aegyptium (L.)Willd.	32	72.1
3	Poaceae	Eragrostis cilianensis(All.)Vignolo-Lut	28	41.2
4	Poaceae	Eragrostis theinlwinii Bor	42	30.2
5	Poaceae	Setaria verticellata (L.) P.Beauv.	33	54.3
6	Solanaceae	Lycopersicum esculentum Miller.	14	27
7	Lamiaceae	Leucas aspera (Willd.) Spreng	13	41.2
8	Asteraceae	Helianthus annus L.	16	17.2
9	Asteraceae	Wedelia calendulacea Less	28	37.2
10	Euphorbiaceae	Phyllanthus urinaria L.	17	19.5

Table 1:- Mycorrhiza infection in roots and spore population in ten species of angiosperms

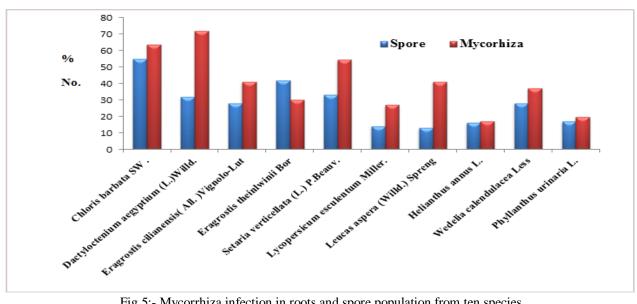


Fig 5:- Mycorrhiza infection in roots and spore population from ten species

✤ Pot experiment

Effect of mycorrhiza fertilizer on the yield parameters of sweet corn 50% tasseling, earing day and ear yield

The day to 50% tasseling in mycorrhiza supplemented plants were 50 DAS (Day After Sowing) and the control

plant were 58 DAS. The day to 50% earing in mycorrhiza supplemented plant were 70 DAS and the control plants were 80 DAS. The average number of ear yield in mycorrhiza supplemented plant is (1.2) and the control was (1) ear as shown in Table 2.

Treatment	No of Plants	Tasseling Days	Earing Days	EarYield (NO)
Control	10	58.5	79.5	1
VAM	10	50.2	70.0	1.2

Table 2:- The effect of mycorrhiza on the tasseling and earing day, ear yield in sweet corn VAM= Vasicular Arbuscular Mycorrhiza

Ear weight, length and width, number of row per ear

The average weight of ear with husk in mycorrhiza supplemented plant was 399 gm and the control is 256 gm. The average weight of ear without husk in mycorrhiza supplemented plant was 364 gm and the control is 228 gm. Average ear length ranged from 19.4 cm (mycorrhiza) to 13 cm (control) among the tested varieties. The highest of ear length was found in 19.4 cm in mycorrhiza and 13 cm in non mycorrhiza. The average number of row per ear in mycorrhiza supplemented plant was 13 rows and the control was 10 row as shown in Table 3.

Treatment	No of plant	Ear Weight		Ear	Size	Row per ear
		Husk	dehusk	Length	Width	
Control	10	256	228	13	5.3	13
VAM	10	399	364	19.4	8.34	10.3
F-Test		**	**	**	**	**
LSD		58.7	52.7	1.8	1.2	1.4
CV(%)		17.7	17.6	11.1	7.5	5.2

Table 3:- The effect of mycorrhiza on the ear weight (husk and dehusk), ear length, ear width and row per ear in sweet corn

VAM= Vasicular Arbuscular Mycorrhiza, Coefficient of variation, L.S.D = Least Significant Difference, **statistical significance at P=0.01(1%)

> Data of soil analysis

The result of soil sample were shown in (Table 4.)

рН	AvaliableN		Avilable P		Available K		Moisture	Texture
7.83	58	Low	19	Medium	199	Medium	4.12	Sandy
alkaline	ppm		ppm		ppm			Loam

Table 4:- Soil analysis data

IV. DISCUSSION AND CONCLUSION

In the present study emphasized on isolation of mycorrhiza from roots and spore number from rhizosphere of ten species were collected from. Mycorrhiza colonization was calculated by using grid-line intersection method (Newman 1996). Nearly, all about the selected plants are observed in mycorrhiza colonization. In this investigation, the range of mycorrhiza colonization in roots was 19.5% to 72.1%. as shown in Table 1. The results showed that, the highest colonization of mycorrhiza in roots was *Dactyloctenium aegyptium* (L.)Willd. is (72.1%) and the lowest mycorrhiza colonization was *Phyllanthus urinaria*

L.19.5 % . Powell (1997) indicated that, the different species might be considered as good hosts for arbuscular mycorrhiza fungi. In this study, the root colonization ten species of angiosperms were different from one another. The present results agree with the statement of Powell (1997).

The spores extraction from rhizosphere soil were collected by wet sieving and spore decanting method. the highest number of spore was observed in *Chloris barbata* SW.(55) rhizosphere and and the lowest number of spore was found in the *Leucas aspera* (Willd.) Spreng (13).Zaw Myo Tun (2009) reported that Variations in present of root colonization were not related to spore population numbers.In

this research, the spore number of ten species of angiospermae is different from one another, even at the same temperature and rainfall. The present results agree with the statement of Zaw Myo Tun(2009). In the present study, regarded soils and plants are mainly affected the mycorrhizal fungi and their development in conservation of soil health and soil fertility.

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