

EFFECT OF BIOFERTILIZER (WITH MYCORRHIZA INOCULUM), COWDUNG, NITROGEN AND PHOSPHOROUS ON GROWTH AND YIELD OF THREE VARIETIES OF AMARANTH (*Amaranthus tricolor* L.)

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ABSTRACT: *The experiment was conducted to study the effect of biofertilizer (with soil based mycorrhiza inoculum, AM), cowdung (CD), two inorganic fertilizers (Urea, N and Phosphorous, P) on the growth and yield of three different varieties of amaranth (*Amaranthus tricolor*). The experiment was carried out with nine treatments in a Completely Randomized Design (CRD) with three replications and 81 treatment combinations. Plant height, plant diameter, total number of leaves per plant, fresh weight of whole plant (shoot and root), root length, dry weight of whole plant (shoot and root) of three varieties of amaranth were recorded after 40 DAS of sowing. Two way analysis of variance (ANOVA) was carried out and the mean differences of the treatments were adjusted by Turkey pairwise comparisons at $P \leq 0.05$ of significant level.*

Application of AM, CD and N significantly influenced the shoot height of amaranth. The highest (49.07 cm) and lowest (31.78 cm) shoot height were recorded in T9 treatment (AM + CD + N) and T1 (basal dose; control) respectively and both are significantly different at $P \leq 0.05$. The result shown that root length of amaranth was significantly similar ($P \leq 0.05$) for all treatments which was either by individual or combined of test medium. The highest root length (18.85 cm) was noted from T2 treatment (AM) on variety 1 (green).and the lowest length (12.94 cm) was recorded in T1 (control) on variety 3 (mixed).

The combined application improved stem diameter of amaranth in T9 (AM+CD+N). The plant obtained adequate nutrition from T9 and the stem growth was thicker when compare to others. Among the varieties tested, variety 1 (green) showed the highest average diameter (1.12 cm) of stem. Significant effect of AM, CD and N was found on number of leaves per plant in amaranth. The highest number of leaves per plant was observed in T9 on variety 1 (green) and treatments T1, T3, T4, T5, T6 and T7 had statistically identical effect. Significant variation in fresh and dry weight of amaranth was observed due to the effect of AM, CD and N. It was observed that the maximum fresh and dry weight per plant (59.7 g and 7.35 g respectively) was observed in T9 (AM + CD + N) on variety 3 (mixed) and that was significantly higher than all other treatments.

The experimental results indicated that there were significant differences in growth and yield characters such as plant height, plant diameter, number of leaves per plant, shoot fresh and dry weights, root fresh and dry weights of amaranth., Since a remarkable higher response in growth and yield of variety 1 (green) and variety 3 (mixed) were observed by the influence of T9 and hence the cost of adding of phosphorous to the soil can be reduced.

Keywords: Biofertilizer, Cowdung, Nitrogen, Phosphorous, growth and yield, *Amaranthus tricolor*

1. INTRODUCTION:

The Department of Agriculture has introduced a biofertiliser with the adequate quantity of soil based Mycorrhiza inoculum in order to enhance the yield of rice cultivation. It is believed that mycorrhiza helps most of the plant species in the uptake of moisture and nutrients from the soil by forming symbiotic association

with roots (Marschner and Dell 1994). Asghari and Cavagnaro (2014) studied on mycorrhizal colonization using mycorrhizal and non mycorrhizal plants, arbuscular mycorrhizal external hyphae development, plant growth, nutrient uptake and NO₃, NH₄ and available P in soil and leachate. They reported that mycorrhizal fungi highly colonized roots of arbuscular mycorrhizal plant (exotic grass *Phalaris aquatica*) and significantly increased plant growth and nutrient uptake. Since Mycorrhizal fungi colonize the roots of more than 90% of plant species having mutual plant and fungus benefit (Smith and Read 2008), and this symbiosis can improve the nutritional status and growth of plants under both optimal and restricted water levels.

Loss of soil organic matter can only be replenished in the short term by application of organic matter such as manure (Glaser et al. 2001). Cowdung is one of the most important organic materials and good source of organic matter. Cowdung releases nutrient slowly so that nutrient loss is less followed by more plant uptake. It is quite promising to use of organic manure and chemical fertilizers in integrated manner for soil fertility. The present day concern about global environmental pollution can be reduced to a considered extent by the judicious use of chemical fertilizers and increasing the use of organic manure.

Phosphorus is one of the major limiting plant nutrients in soils world over. The presence of phosphorus in the soil encourages plant growth because phosphorus is essential mineral macronutrients. Most of the essential plant nutrients, including phosphorus, remain in insoluble form in soil (Yadav and Verma 2007). It was reported that applications of phosphorus solubilizing microbe significantly improved yield of maize, but had no real effect towards potential-P, available-P, phosphatase and P uptake of plants (Fitriatin et al. 2014). Because of such benefits, the Mycorrhizae fungi can be used as bio-fertilizer for enhancing growth and yield of most crop plants.

Of the necessary nutrient elements, N is required in the largest quantities, and its availability and internal concentration affect the partitioning biomass between roots and shoots (Glaser et al, 2001). Costa et al (2002) reported that root length and root surface area were increased under intermediate N levels and that root growth was reduced under both higher and lower fertilizer levels. Abbasi (2007) and Saeidi (2008) announced that application of nitrogen fertilizer increases the average weight, the number and the operation of the tuber in potato, and these traits decrease will be decreased if the optimum nitrogen rate exceeds.

Amaranth is a multipurpose crop whose leaves and grains are tasty and of high nutritional value, additionally it can be cultivated as an ornamental plant (Venskutonis and Kraujalis, 2013).

The present study was conducted to study the effect of biofertilizer (with soil based mycorrhiza inoculum, AM) mainly along with cowdung (CD), two inorganic fertilizers (Urea: N and Phosphorous: P) on the growth and yield of three different varieties of amaranth (*Amaranthus tricolor*).

2. MATERIALS AND METHODS:

2.1 Collection and preparation of soil sample

The soil used in this experiment was collected from area surrounding of the Agricultural Research Centre, Sammanthurai, Ampara. The soil was dried and the clods were broken and sieved to remove weeds, stubbles and hard clods.

2.2 Test crops

Three varieties of *Amaranthus tricolor*; Pure green colour (V1), Pure purple colour (V2) and mixed colours of green and purple (V3) were used as plant materials for the experiment. Commercially available seeds registered under the seed act and approved by the Department of Agriculture were obtained from Department of Agriculture.

2.3 Treatment combinations Experimental design

The experiment was laid out in Completely Randomized Design (CRD) with three replications and total treatment combinations were 81 (3 varieties X 9 treatments X 3 replicates) and for each test variety the number of combination was 27.

2.4 Fertilizer dose

Biofertilizer with Arbuscular Mycorrhizae (AM) =150g/polybag,
(Obtained from Agricultural Research Centre, Sammanthurai, Ampara)
Cowdung (CD) = 37.5g/polybag,
Phosphorous (P) = 0.56 g Tri Super Phosphate (TSP)/polybag
Nitrogen (N) = 1.95g Urea/ polybag

Basal dose:

Urea = (1.78 g/polybag),
Muriatic of potash (MOP) = (0.375 g/polybag)
Gypsum = (0.166g/polybag)

Per polybag 15 kg soil was used. Basal doses were added according to the fertilizer recommendation guide of Department of Agriculture (2005).

2.5 Treatment combinations

T1: Control (only basal dose)	T4: P	T7: AM + N
T2: AM	T5: N	T8: AM + CD + P
T3: CD	T6: AM + P	T9: AM + CD + N

2.6 Experimental plot

Table 1: The plot of completely randomized design. (CRD)

Right					Left			
T ₁ V ₁ R ₁	T ₈ V ₁ R ₁	T ₉ V ₁ R ₁	T ₇ V ₁ R ₁	T ₃ V ₁ R ₁	T ₅ V ₁ R ₁	T ₄ V ₁ R ₁	T ₂ V ₁ R ₁	T ₆ V ₁ R ₁
T ₆ V ₂ R ₁	T ₉ V ₂ R ₁	T ₄ V ₂ R ₁	T ₇ V ₂ R ₁	T ₂ V ₂ R ₁	T ₃ V ₂ R ₁	T ₈ V ₂ R ₁	T ₅ V ₂ R ₁	T ₁ V ₂ R ₁
T ₁ V ₃ R ₁	T ₂ V ₃ R ₁	T ₃ V ₃ R ₁	T ₈ V ₃ R ₁	T ₅ V ₃ R ₁	T ₉ V ₃ R ₁	T ₇ V ₃ R ₁	T ₄ V ₃ R ₁	T ₆ V ₃ R ₁
T ₉ V ₁ R ₂	T ₃ V ₁ R ₂	T ₅ V ₁ R ₂	T ₂ V ₁ R ₂	T ₈ V ₁ R ₂	T ₆ V ₁ R ₂	T ₇ V ₁ R ₂	T ₄ V ₁ R ₂	T ₁ V ₁ R ₂
T ₃ V ₂ R ₂	T ₆ V ₂ R ₂	T ₂ V ₂ R ₂	T ₁ V ₂ R ₂	T ₇ V ₂ R ₂	T ₄ V ₂ R ₂	T ₈ V ₂ R ₂	T ₅ V ₂ R ₂	T ₉ V ₂ R ₂
T ₈ V ₃ R ₂	T ₉ V ₃ R ₂	T ₃ V ₃ R ₂	T ₅ V ₃ R ₂	T ₇ V ₃ R ₂	T ₆ V ₃ R ₂	T ₂ V ₃ R ₂	T ₄ V ₃ R ₂	T ₁ V ₃ R ₂
T ₄ V ₁ R ₃	T ₁ V ₁ R ₃	T ₈ V ₁ R ₃	T ₆ V ₁ R ₃	T ₅ V ₁ R ₃	T ₇ V ₁ R ₃	T ₉ V ₁ R ₃	T ₃ V ₁ R ₃	T ₂ V ₁ R ₃
T ₂ V ₂ R ₃	T ₄ V ₂ R ₃	T ₉ V ₂ R ₃	T ₁ V ₂ R ₃	T ₈ V ₂ R ₃	T ₅ V ₂ R ₃	T ₃ V ₂ R ₃	T ₆ V ₂ R ₃	T ₇ V ₂ R ₃
T ₇ V ₃ R ₃	T ₉ V ₃ R ₃	T ₈ V ₃ R ₃	T ₃ V ₃ R ₃	T ₅ V ₃ R ₃	T ₁ V ₃ R ₃	T ₄ V ₃ R ₃	T ₂ V ₃ R ₃	T ₆ V ₃ R ₃



Figure 1: *Amaranthus tricolor* at the experimental plot

2.7 Application of fertilizer in soil and sowing of seed

Basal doses were carefully weighed and mixed with each treatment substances separately added into reusable polythene bags (> 15 Kg capacity) except the Organic fertilizer with Arbuscular Mycorrhizae (AM) which was surface mixed with basal dose. Seeds of each variety was sown in each bag with three replicates. After sowing the seed, the soil was saturated with water and bags were covered (50% shadow) up to germination period to give a shady condition. Plants were regularly irrigated every day. During the growth stage a pesticide ('Marshal', 30 ml of chemical was mixed with 10 L of water) was applied to avoid the pest attack.

2.8 Collection of Data

Plant height, plant diameter, total number of leaves per plant, fresh weight of whole plant (shoot and root), root length, dry weight of whole plant (shoot and root) of three varieties of amaranth were recorded after 40 DAS of sowing.

2.9 Statistical analyses

Two way analysis of variance (ANOVA) was done following the completely randomized design (CRD). Data were analysed in Minitab 16.1 (2007) at $P \leq 0.05$ of significant level. The mean differences of the treatments were adjusted by Turkey pairwise comparisons.

3. RESULTS AND DISCUSSION:

3.1 Shoot height

Different growth parameters such as plant height, plant diameter and number of leaves per plant are given best performance in the yield of amaranth. Application of AM, CD and N significantly influenced the shoot height of amaranth (Table 2). The highest (49.07 cm) and lowest (31.78 cm) shoot height were recorded in T9 treatment (AM + CD + N) and T1 (basal dose; control) respectively and both are significantly different at $P \leq 0.05$.

Table2. Effect of AM, CD, N, P on growth and yield of Amaranth (*Amaranthus tricolor*)

Treatment	SH/cm	RL/cm	SD/cm	NL	FW/g	DW/g
T1: control	31.78b	12.94a	0.95b	10b	42.4abc	4.80b
T2: AM	43.81a	18.85a	1.08ab	13ab	45.9abc	4.39b
T3: CD	36.53b	16.3a	0.98b	12b	48.8abc	3.79b
T4: P	39.46ab	17.0a	1.00b	11b	42.43bc	4.38b
T5: N	40.46ab	16.2a	1.02b	12b	43.50bc	3.97b
T6: AM+P	36.11b	17.3a	0.94b	11b	42.91bc	4.45b
T7:AM+N	38.31ab	16.0a	1.07ab	12b	40.00c	3.57b
T8:AM+CD+P	42.51ab	16.0a	1.11ab	13ab	56.03ab	5.42ab
T9: AM+CD+N	49.07a	17.7a	1.12a	14a	59.74a	7.35a

SH – Shoot height, RL – Root length, SD – Stem diameter, NL – Number of leaf, FW – Fresh weight, DW – Dry weight. In a column, the figure(s) having same letter are not significantly different at $P \leq 0.05$ by Turkey pairwise mean comparisons.

The combined application of AM biofertilizer along with nitrogen and CD application employ a positive effect on all growth and yield parameters of amaranth. Treatments T4, T5, T7 and T8 were statistically identical in their effect on growth and yield. Individual application of AM, CD, P and N (T2, T3, T4 and T5) indicated significantly higher shoot height over control plant. Shoot height showed best performance on variety 1 (green) and low performance on variety 2 (purple) (Fig. 2).

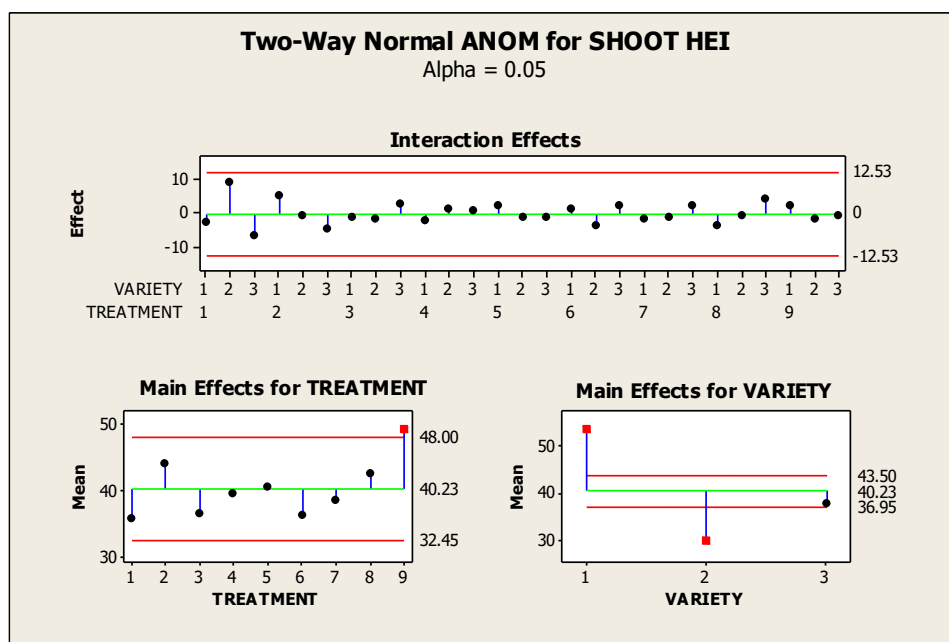


Figure 2. Interaction and main effects on shoot height of *Amaranthus tricolor*

The present study revealed that AM inoculants had significant effect on plant growth of amaranth. Similarly it was found in three varieties of same species, Such as by (Karagiannidis *et al.* 2002) in tomato and egg plants, (Ergin and Gülser 2016) in lettuce and (Caglar and Akgun 2006), who said that the treatment of mycorrhiza increased plant growth compared to control. The results specified that the combined application of AM, cowdung with nitrogen fertilizer increased the plant height, diameter and number of leaves and so on. This result was supported by Wang *et al.* (2006) also. The symbiotic association between mycorrhizal fungi and the roots of plants is widespread in the natural environment. AM benefit their host principally by increasing uptake of macronutrients other than P, including nitrogen (N) potassium (K) and magnesium (Mg) has also been measured as well as increased uptake of some micronutrients. (Hodge *et al.* 2001).

3.2 Root length

The result shown that root length of amaranth was significantly similar ($P \leq 0.05$) for all treatments which was either by individual or combined test medium (Table 2). The highest root length (18.85 cm) was noted from T2 treatment (AM) on variety 1 (green).and the lowest length (12.94 cm) was recorded in T1 (control) on variety 3 (mixed) (Table 2 and Fig. 3). There is improved interest in proper and effective use of organic manure to maintain soil fertility (Olatunji and Oboh, 2012). Organic manure helps to increase the population of soil micro-organisms which have some influence in protecting plant against pathogens like nematodes and soil born insects and also provides plant growth hormones like auxins (Agbede and Ojeniyi, 2009). Organic manure also supports to improve the physical condition of the soil and offers the required plant nutrients. It enhances cation exchange capacity and performances as a buffering agent against undesirable soil pH fluctuations (Ojeniyi *et al.*, 2007; Akanni and Ojeniyi, 2008). The application of organic manure has been found to have higher comparative economic advantage over the use of inorganic fertilizer.

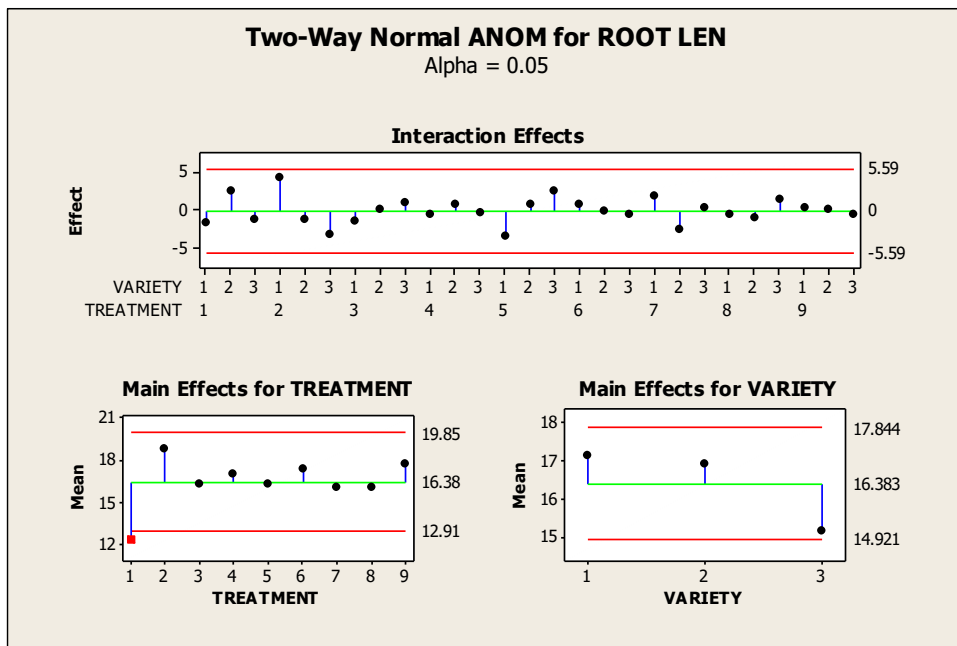


Figure 3. Interaction and main effects on root length of *Amaranthus tricolor*

3.3 Stem diameter

The combined application of fertilizers affected diameter of amaranth in T9 (AM+CD+N). The plant obtained adequate nutrition from T9 and the stem growth was thicker when compare to others. Among the varieties tested, variety 1 (green) showed the highest average diameter (1.12 cm) of stem. The least average diameter (0.95 cm) was produced by T1 (control) on variety 2 (purple) (Table 2 and fig. 4).

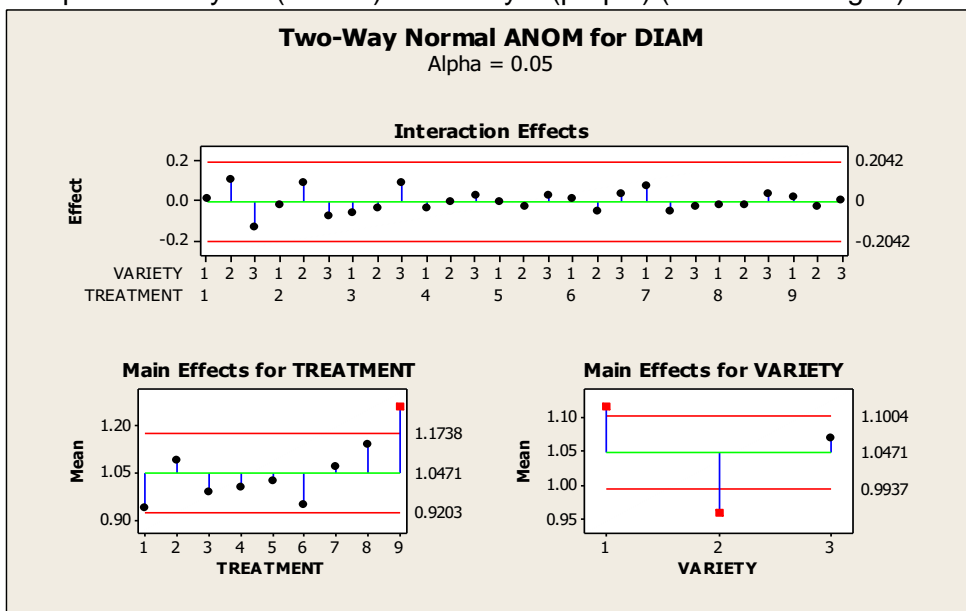


Figure 4. Interaction and main effects on stem diameter of *Amaranthus tricolor*

Mycorrhizal inoculation improved higher shoot diameter in AM inoculated seedlings than in non-inoculated seedlings (Giri et al 2005). Subhan (1988) has reported that application of organic manure increased the head diameter of cabbage. The result

showed that the plant diameter of amaranth was significantly influenced by the effect of AM biofertilizer, cowdung and phosphorus (Guo et al 2006).

3.4 Number of leaves per plant

Significant effect of AM, CD and N was found on number of leaves per plant in amaranth. The highest number of leaves per plant was observed in T9 on variety 1 (green) where AM, CD and N were applied. The treatments T1, T3, T4, T5, T6 and T7 had statistically identical effect (Table 2). The variety 2 (purple) showed least performance of having number of leaves (fig. 5).

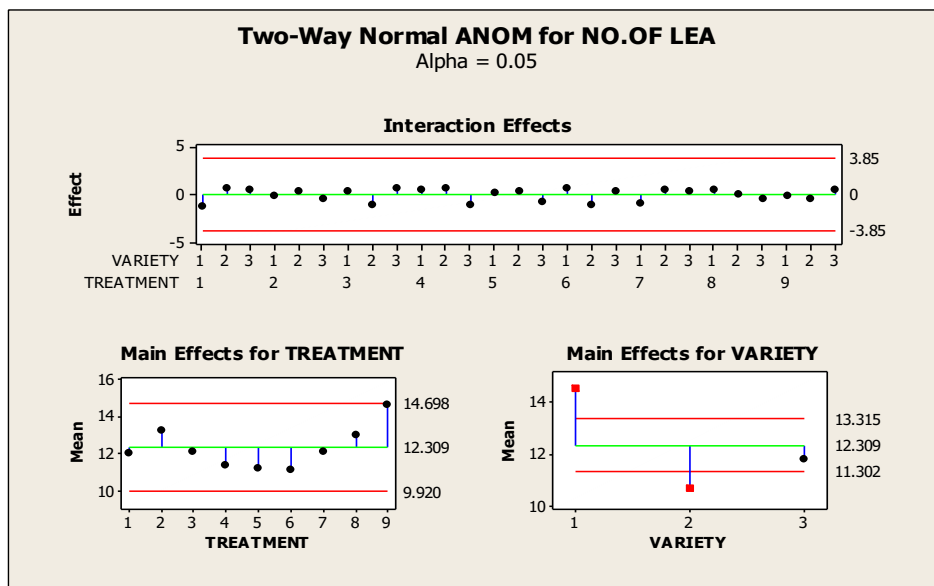


Figure 5. Interaction and main effects on number of leaf of *Amaranthus tricolor*

It was noted that the number of leaves per plant increased successively with the combined applications as in T8 and T9.

3.5 Fresh and dry weight per plant

Significant variation in fresh and dry weight of amaranth was observed due to the effect of AM, CD and N (Table 2). It was observed that the maximum fresh and dry weight per plant (59.7 g and 7.35 g respectively) and that was observed in T9 (AM + CD + N) in variety 3 (mixed). It was significantly higher than all other treatments. The lowest fresh and dry weight were obtained in treatment T7 (AM + N) on variety 1 (green), (fig. 6). Shoot fresh weight and dry weight of amaranth was also influenced significantly by the application of AM, phosphorus and CD. Shoot fresh and dry weight was increased by AM colonization (Tawaraya et al. 2006).

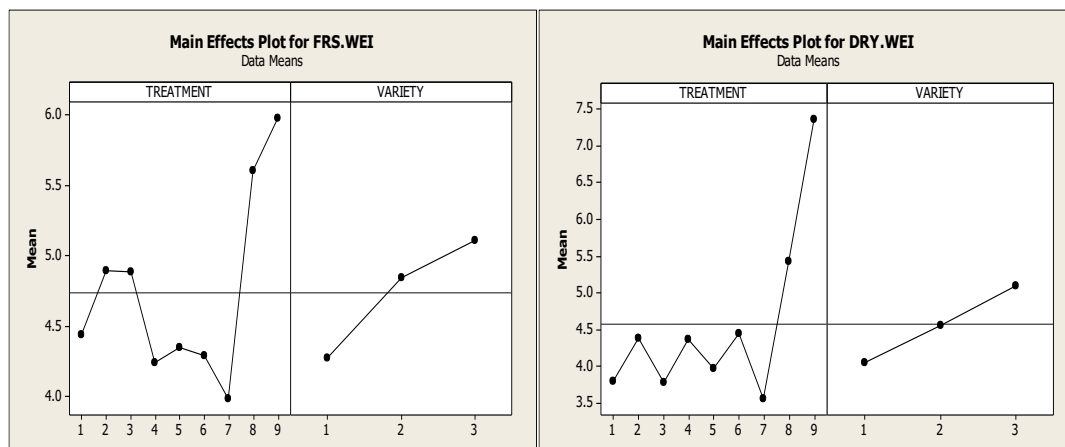


Figure 6. Main effects on fresh and dry weight of leaf of *Amaranthus tricolor*

The shoot dry weight was increased with AM inoculation (Andersen and Andersen 2006) and the increasing rate of manure (Asghari et al. 2015). Under nursery conditions mycorrhizal inoculation improved growth of *Glomus fasciculatum*, *G. macrocarpum* and *Cassia siamea* seedlings. Shoot dry weight was higher in mycorrhizal than non-mycorrhizal plants (Giri et al. 2005).

4. Conclusions

Application of AM, CD and N significantly influenced the shoot height of amaranth. The highest and lowest shoot height were recorded in T9 treatment (AM + CD + N) and T1 (basal dose; control) respectively and both are significantly different at $P \leq 0.05$. The result shown that root length of amaranth was significantly similar ($P \leq 0.05$) for all treatments which was either by individual or combined of test medium. The highest root length was noted from T2 treatment (AM) on variety 1 (green).and the lowest length was recorded in T1 (control) on variety 3 (mixed).

The combined application improved stem diameter of amaranth in T9 (AM+CD+N). Among the varieties tested, variety 1 (green) showed the highest average diameter (1.12 cm) of stem. Significant effect of AM, CD and N was found on number of leaves per plant in amaranth. The highest number of leaves per plant was observed in T9 on variety 1 (green) and treatments T1, T3, T4, T5, T6 and T7 had statistically identical effect. Significant variation in fresh and dry weight of amaranth was observed due to the effect of AM, CD and N. It was observed that the maximum fresh and dry weight per plant was observed in T9 (AM + CD + N) on variety 3 (mixed) and that was significantly higher than all other treatments.

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