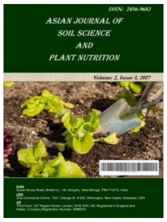

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Effect of Biofertilizer on Growth and Yield Characteristics of *Zea mays* L. in Different Ecological Zones in Kenya

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Authors' contributions

This work was carried out in collaboration among all authors. Author WNW designed the study, wrote the protocol and wrote the first draft of the manuscript. Authors ASK and NKK reviewed the study design and all drafts of the manuscript. Author WNW managed the analyses of the study and performed the statistical analysis. Author ASK managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

In order to reduce the dependence on chemical fertilizers, alternative methods should be developed which will provide nutrients to plants. The increased cost of inorganic fertilizers, including their inability to condition the soil and their polluting effect on the environment, has directed attention towards other sources of soil fertilization to enhance maize production. Hence, this study was carried out to determine the effect of Effective Micro-organisms (biofertilizer) comprising *Pseudomonas spp*, *Saccharomyces spp*, *Bacillus subtilis* and *Lactobacillus spp*. on the growth and yield components of *Zea mays* L. The trial plots measured 4 m×3 m and the experimental design was randomized complete block design (RCBD) with 4 treatments namely; Biogrovit (biofertilizer) alone; conventional fertilizer alone, Biogrovit plus conventional fertilizer

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combined and the control. It was replicated three times. Biogrovit was soil drenched in plants at an interval of 14 days in crops established at two sites in Kirinyaga and Machakos County. Significant differences were observed in the leaf area where application of the biofertilizer had the largest (995 cm^2) while the least was under the control (529 cm^2). The grain yield was notably influenced by application of treatments at both sites, where the highest was recorded under the biofertilizer in Kirinyaga (8.6 t/ha) and Machakos (7.77 t/ha) which was not significantly different from that of the conventional fertilizers in Kirinyaga and Machakos at 7.55 t/ha and 6.87 t/ha respectively. The control had the lowest grain yield in both sites. The 1000-grain mass, ear length, cob weight, number of cobs per plant and the number of kernels per cob directly influenced the actual grain yield as they were higher for both biofertilizer and chemical treatments. The application of Biofertilizer and conventional fertilizer combined at full rates were antagonistic as most of the parameters tested had lower counts than when independently applied. Therefore, the findings of this study suggest that biofertilizers enhance the growth of maize and as such its use should be encouraged because it is eco-friendly.

Keywords: Biofertilizer; Pseudomonas spp; Saccharomyces spp; Bacillus subtilis; Lactobacillus spp.; Zea mays L.

1. INTRODUCTION

Maize is a valuable cereal that is highly cultivated in Kenya because of its domestic and industrial use. According to Tollenaar and Dwyer, [1], maize is the third most important cereal crop after wheat and rice in the world based on area and production. The productivity of maize is dependent on its nutrient requirement and management, particularly that of nitrogen, phosphorus and potassium [2]. The increased cost of conventional fertilizers including their inability to condition the soil and their polluting effect on the environment has directed attention towards other sources of soil fertilization to enhance maize production. Micro-organisms are involved in a range of processes that affect the transformation of soil phosphorus and atmospheric nitrogen into usable forms for plant growth. Microbial inoculants of bacteria, algae and fungi, either separately or in combination enhance the availability of nutrients to plants by nitrogen fixation and solubilizing phosphorus for the benefit of plants. Microorganisms are therefore critical in the conversion of atmospheric nitrogen and transfer of phosphorus from accessible soil pools to plants in available forms.

Biofertilizers are preparations containing live or latent cells of efficient nitrogen fixing, phosphate solubilizing algae, bacteria or fungi. The application of biofertilizers can be either to the seed or soil to speed up microbial processes in the soil thereby augmenting the availability of nutrients which can be assimilated by crop plants. Indeed, certain soil micro-organisms have the inherent capacity to dissolve part of the bound phosphorus and make it available to crops by secreting organic acids such as acetic acid, succinic acid, lactic acids, etc. [3]. These attributes make the micro-organisms important as biofertilizers. The plant promoting rhizobacteria can influence plant growth directly through the production of phytohormones and indirectly through nitrogen fixation and production of biocontrol agents against the soil-borne pathogens [4].

The use of microbial inoculants as biofertilizers has become a hope for most countries as far as economic and environmental viewpoints are concerned. Biologically fixed nitrogen is such a source that can supply an adequate amount of nitrogen to plants and other nutrients to some extent [5]. It is a non-hazardous way of fertilization of the field. Moreover, biologically fixed nitrogen consumes about 25% to 30% less energy than the chemical fertilizers. The application of biofertilizers provides effective implementation of biological mechanisms of plant nutrition, growth promotion and protection [6]. In order to reduce the dependence on chemical fertilizers, an alternative method is to be developed which will provide nutrients to plants. Through effective research and technology, Biogrovit combines useful micro-organisms primarily; Lactic acid bacteria, photosynthetic bacteria, yeast, pseudomonas and actinomycetes leading to a soil friendly, organic solution for improving soil fertility and restoring pH balance in the soil. Each of these micro-organisms has an important role complementing each other and are mutually beneficial. They collectively work towards the betterment of the soil, environment and plants. However, there is limited information on the advantages of biofertilizers prompting this study on maize in Kirinyaga and Machakos Counties in Kenya.

2. MATERIALS AND METHODS

2.1 Study Sites

There were three crops grown (Maize, French beans and Kale) in two locations of two counties of Kenya namely Mwea in Kirinyaga County and Kitengela in Machakos County. The maize variety used was DKC-9089 from Monsanto Seed Company.

2.2 Experimental Design and Application of Treatments

The trial was laid out in a randomized complete block design (RCBD). The treatments included; Biogrovit (biofertilizer) alone, conventional (chemical) fertilizer alone, Biogrovit plus conventional fertilizer, and no application (control). The experimental plots measured 4 m long and 3 m wide with a 0.5 m pathway between plots and 1 m pathway between blocks. The treatments were replicated three times.

The land was ploughed and re-ploughed at a two-week interval then harrowed to a fine tilth where the experimental units were demarcated. Furrows were opened at a spacing of 75 cm by 30 cm for maize. Biogrovit was soil drench-applied near the plants at one week after emergence and after every two weeks for five times to physiological maturity. A knapsack sprayer at full pressure at a rate of 2 litres per acre was used after mixing with water in ratio of 1 litre of Biogrovit in 100 litres of water. The conventional treatment had DAP fertilizer applied at planting and top-dressed with CAN at the 5-leaf stage of the crop. All other agronomic practices were carried out uniformly as recommended for the respective crops.

2.3 Data Collection and Statistical Analysis

Plant height was measured vertically based on the distance from the stem base to the highest growing point of leaf segregation while the leaf number included standard counting of leaves per plant including discoloured ones for cases of senescence. The leaf area was measured using a leaf area meter Model. The cob weight and the number of rows per cob were determined. To determine grain yield, biomass yield and harvest index, we removed and cleaned all the seeds produced within 1 m^2 central rows in the field. Then grain yield and biomass yield were recorded on a dry weight basis. Yield was defined in terms of grams per square meter and quintals per hectare. Replicated samples of clean seed (broken grain with foreign material removed) were sampled randomly and 1000-grain were counted and weighed.

The data collected was statistically analyzed by using the computer statistical program SAS package. Analysis of variance technique was employed to test the overall significance of the data, while the least significance difference (LSD) test at P= 0.05 was used to compare the differences among treatment means [7].

3. RESULTS AND DISCUSSION

3.1 Growth Components

In both sites, a conclusive trend observed among the test treatments showed differences, with the absolute control having the lowest growth rate of maize in both sites. The leaf area, number of leaves per plant and plant height significantly (P= 0.05) differed between the treatments where the highest were under the biofertilizer and conventional treatments while the lowest were under the control in both sites (Tables 1 & 2). The tallest plants were recorded under the biofertilizer treatments at 160 cm and 158 cm for Kirinyaga and Machakos respectively while the shortest were under the control at 102.5 cm and 93.8 cm for Kirinyaga and Machakos respectively. Also, the leaf area was highest under the biofertilizer and conventional fertilizer treatments in both sites with the control exhibiting the lowest in both sites. The average number of leaves per plant was signficantly different between the treatments where the highest in Kirinyaga was recorded under the biofertilizer and conventional fertilizer and the highest number of leaves per plant in Machakos was under the biofertilizer treatment. There were no significant differences between the treatments on the stand count in both sites.

The increase in the growth components compared to the control might be due to the effect of micro-organisms in the biofertilizer which colonized the plant and soil thus directly releasing nutrients or by increasing availability of nutrients in the soils to plants. This is in accordance to Vessey [8] who reported that biofertilizers are defined substances which contain living micro-organisms and when applied to seed, plant surfaces or soil, colonize the plant and promote its growth by increasing the nutrient availability. Also, Ali et al. [9] reported that application of plant growth promoting rhizobacteria increased plant height and biological yield. This agrees with Zahir et al. [10] that Azotobacter and Azospirillum are the most important plant growth promoting rhizobacteria which affect the growth and development of crops. Vessey, [8] posited that Azotobacter and Azospirillum enhance crop growth conditions through several mechanisms especially through growth hormone production and improving the efficiency of roots. Such growth promoting effect was maximal in response to inoculation with mixture of *Azotobacter, Azospirillum* and phosphate-solubilizing microorganisms (PSM) for all the growth parameters when compared with the control. Improved plant growth by *Azospirillum sp* has been attributed to both production of plant hormones, especially growth promoters, and by supplying combined nitrogen [11].

3.2 Yield Components

The application of treatments positively and notably influenced the grain yield and yield components of maize in both sites (Tables 3 and 4). The highest grain yield in Kirinyaga was recorded under the biofertilizer treatment (8.60 t/ha) which however, did not differ significantly with that under the conventional fertilizer treatment (7.55 t/ha). The lowest grain yield was observed under the control (4.45 t/ha). The 1000-grain mass, number of cobs per plant and the ear height were highest under the biofertilizer treatment, thus directly influencing the total grain yield in the end.

In Machakos, there were no significant differences between the biofertilizer and conventional fertilizer treatments which were however signficantly higher than those under the combination of the biofertilizer and the conventional fertilizer, and the untreated control. A maximum of 7.77 t/ha was recorded under the biofertilizer treatment and 6.87 t/ha under the conventional fertilizer. The ear height, 1000-grain mass and number of cobs per plant are important traits that significantly differed due to the treatments with the highest recorded under the biofertilizer and conventional fertilizer treatments individually.

The positive effect of biofertilizer may have resulted from its ability to increase the availability of phosphorus and other nutrients especially under the influence of the calcareous nature of the soil, which cause decrease in the nutrients availability. These results agree with [12]. Some researchers have also determined that enhanced phosphorus release increases evaluations for the trait of grain yield, biomass yield and 100-seed weight [13]. The 1000 grain weight increases due to better transfer of photosynthetic substances under the biofertilizer treatments. It may be concluded that the photosynthetic capacity of plants treated with phosphorus-solubilizing micro-organisms increases due to increased supply of phosphorus nutrition. Cob weight increase may have been under the effect of the phosphorus biofertilizer which induced the nutrient uptake ability of the roots and positively increased the yield parameters because of improving the root system as a source-sink relationship to the reproductive part (shoot), this agrees with [14]. Grain yield and biomass yield increase were reported with the biofertilizer application which accounts important benefits to the maize producers and maize production, causing a decrease in the inputs of production because of economizing money compared to chemical fertilizers in order to increase gram yield and biological yield. Biomass yield increased under application of biofertilizers, because there was a significant increase in the dry weight of shoots at the prestilking stage, that

Table 1. Influence of test treatments on growth parameters of maize at Kirinyaga

Treatment	Leaf area (cm^2)	Leaf number	Height (cm)-80 DAP	Stand count
Biogrovit	995a	16.3a	160.0a	26a
Biogrovit+Conventional	643c	15.3ab	132.3b	24a
Control	529d	14.4b	102.5c	17a
Full Conventional	752b	15.5ab	155.0a	20a
SE	74.6	0.369	2.826	2.612
CV%	12.9	9	6.4	15.1
F pr.	<.001	<.001	<.001	0.173

Means followed by the same letter in each column are not significantly different at P≤0.05. DAP-Days after planting

Table 2. Influence of test treatments on growth parameters of maize at Machakos

Treatment	Leaf area (cm^2)	Stand count	Height (cm)-80 DAP	Leaf no
Biogrovit	881a	15a	158.0a	17.5a
Biogrovit+Conventional	774b	16a	125.3b	11.0c
Control	436c	15a	93.8c	9.8c
Full conventional	858a	16a	128.0b	13.8b
SE	66.7	0.453	2.911	0.632
CV%	14.6	4.2	3	6.2
F pr.	<.001	0.287	<.001	<.001

Means followed by the same letter in each column are not significantly different at P≤0.05. DAP-Days after planting

Table 3. Influence of test treatments on 1000-grain mass, number of cobs per plant, ear length and grain yield of maize at Kirinyaga

Treatment	1000-Grain weight (g)	Cobs/plant	Ear Length (cm)	Grain Yield (t/ha)
Biogrovit	351.5a	2.0a	15.9a	8.60a
Biogrovit+Conventional	260.8c	1.0c	11.4b	6.15b
Control	209.8d	1.0c	6.5c	4.45d
Full Conventional	299.5b	1.3b	13.2a	7.55a
SE	12.75	0.288	0.407	0.431
CV%	5.9	24.1	4.4	10.4
F pr.	<.001	<.001	<.001	<.001

Means followed by the same letter in each column are not significantly different at P≤0.05

Table 4. Influence of test treatments on 1000-grain mass, number of cobs per plant, ear length and grain yield of maize at Machakos

Treatment	1000-Grain weight (g)	Cobs/Plant	Ear Height (cm)	Grain Yield (t/ha)
Biogrovit	335.6a	1.78a	23.8a	7.77a
Biogrovit+Conventional	278.1b	1.13b	17.7b	5.95b
Control	198.7c	1.01b	13.0c	5.38d
Full Convectional	329.5a	1.13b	18.5b	6.87a
Standard error	12.75	0.0537	0.805	0.287
CV%	5.9	5.5	5.7	7.9
F pr.	<.001	<.001	<.001	<.001

Means followed by the same letter in each column are not significantly different at P≤0.05

may be related to the favouritism of some environmental factors which directly affected the bio fertilizer and its impact on the nutrient availability and growth, which positively influenced maize photosynthesis and dry matter accumulation more actively that agree with [15,16]. Azimi et al. [17] found that application of Supernitroplus biofertilizer with Phosphate (Barvar 2) treatment had the highest seed yield (7.6 ton/ha) and non-application of biofertilizers treatment that had Pishtaz cultivar had the lowest seed yield (6.3 ton/ha) [18]. Also, Azimi et al. [18] found that the application nitrogen and phosphate biofertilizers increased yield and yield components of barley under Boroujerd environmental condition. They suggested that grain yield and biomass yield increase was reported with the biofertilizer application which is seen to be beneficial, causing a decrease in the production costs because of spending less money compared to chemical fertilizers as mentioned earlier [19].

The reduction in yield in the combined treatment of biofertilizer and conventional fertilizer might be due to an antagonistic effect on plants and soils. The symbiotic association of micro-organisms with plant roots is one of the most enhanced biological activities in the soil. The neglectful interference of human activities such as over-application of fungicides and frequent chemical phosphorus and nitrogenous fertilizer application has seriously threatened this advantageous symbiosis as is in this case.

4. CONCLUSION

Application of biofertilizers is essential in the production of maize and therefore recommended for its proper use is an environmentally friendly way of strengthening plant growth and improvement.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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