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THE EFFECT OF STREPTOMYCIN ON SOME MORPHOLOGICAL PARAMETERS OF THREE ACCESSIONS OF BAMBARA NUT (*Vigna subterrenea* (L) Verde)

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ABSTRACT
Bambara nut (*Vigna subterrenea* (L) Verde) is a major source of vegetable protein in sub - Saharan Africa. It is well adapted to harsher conditions and constitutes an important part of the local diet, culture and economy. The effects of streptomycin on some morphological parameters of three accessions of Bambara nuts was investigated in this study. Three healthy and viable bambara nut *(Vigna subterranean)* accessions were collected from IITA, Ibadan, Nigeria. They include: TVSU-1, TVSU-9 AND TVSU-10. Streptomycin was obtained from a reputable pharmaceutical store located along Abuja Road, Lafia, Nasarawa state. Garden soil was collected from the research garden and filled into polyethene pots after which they were perforated at the bottom. Randomized Complete Block Design (RCBD) was adopted with each treatment consisting of 3 replicates. Polyethylene bags were properly labeled according to a given treatment name and replicate number. Five (5) different concentrations of Bambara Nut were measured with the aid of weigh balance. This concentration is; 0.1g/L, 0.2g/L, 0.3g/L, 0.4g/L and lastly 0.5g/L. Different Accessions of Bambara Nut were primed into this different concentration. From the results obtained, the number of leave was highest at the lowest concentration in TVSU-1. In TVSU-9 0.3g/L had the highest number of leaves. However, control had the highest number of leaves in TVSU-10. In Plant height, the lower concentration had more plant height compared to other treatments. In the Stem Girth, the mid-concentration had more stem girth size (0.3g/L) across accessions. Same observation was recorded in the Leaf Area of all Accessions where treatment 0.3g/L had the highest Leaf Area. In the number of flowers, treatment 0.3g/L recorded the highest Number of Flowers. However, in Number of days to germination, the least treatment recorded the fastest days to germination.

Keywords: Streptomycin, accessions, Bambara nuts, yield

INTRODUCTION

Bambara nut (*Vigna subterrenea* (L) Verde) is a major source of vegetable protein in sub - Saharan Africa. It is well adapted to harsher conditions and constitutes an important part of the local diet, culture and economy (Goli, 1997; Heller *et al*., 1997). Bambara nut fixes atmospheric nitrogen in symbiosis with Brady rhizobium strains through a nodulation process (Gueye *et al*., I998). The crop requires relatively low inputs and contributes to the sustainability of the cropping systems in West Africa). The seed is regarded as a completely balanced food because it is rich in iron 4.948mg/100g, compared to a range of 2.0 - 10.0 mg/100g for most food Legumes, Protein 18.0 - 24.0% with high Lysine and Methionine contents (Rowland, 1993), Ash 3.0 - 50%, Fat 5.0 - 7.0%, Fibre 5.0 - 12.0%, Potassium 1144 -1935mg/100g, Sodium 2.9 - 12.0 mg/l00g, Calcium 95.899mg/100g, Carbohydrate 51 - 70%, oil 6 - 12%, and energy 367 - 414kal/100mg (Rowland, I993).

Bambara seed and haulm have been used to feed livestock and poultry (Anchirina *et al*., 2001). Bambara nut fixes atmospheric nitrogen in symbiosis with Bradyrhizobium strains through a nodulation process (Gueye *et al*., I998). The crop requires relatively low inputs and contributes to the sustainability of the cropping systems in West Africa. It is grown mostly by women who intercrop with maize, millet, sorghum, yam, and groundnut (Goli, 1997, Anchirina, *et al*., 2001).

Aminoglycoside are a group of highly potent, broad spectrum antibiotic which have amino sugars that linked by glycosidic bonds (Madigen *et al*., 2009). Antibiotic has been gainfully exploited in the manufacture of substances that promote the growth of transformed tissue, called positive selectable marker genes (Miki and McHugh, 2004) in transgenic and genetically modified plants. These antibiotics, which are basically called aminoglycosides, have a basic chemical structure necessary for both potency and the wide range of antimicrobial activity of aminoglycosides is that of one or several amino sugars joined in glycosidic linkages to a dibasic cyclitol. In most clinically used aminoglycosides, the latter is 2-deoxystreptamine and it is streptidime in streptomycin and derivatives; and fortamine in the fortmicin series. This is achieved by its strong interaction with the ribosome thus disrupting translation. These reactions are quite specific and bind to the rRNA. Also, they inhibit transcription by inhibiting RNA synthesis (Madigan *et al*., 2009). Antibiotic has also been gainfully exploited in the manufacture of substances that promote the growth of

transformed tissue, called positive selectable marker genes (Miki and McHugh, 2004) in transgenic and genetically modified plants. The aim of the research therefore was to determine the effect of streptomycin on some morphological and yield parameters of Bambara nuts while the objective is to identify the effect of streptomycin in growth and yield of Bambara nut. Bambara nut (Vigna subterranean) is a major source of vegetable protein in sub - saharan Africa. It is well adapted to harsher condition and constitutes an important part of local diet culture and economy (Goli, l997; Heller, *et al*., 1997). According to Linnemann (1990), Bambara nut flour is used to make Bread. Bambara milk is preferred to that prepared from other pulses because of its flavor and colour (Goli, 1997). Bambara seed and Haulm have been used to feed Livestock and poultry (Anchirina, *et al*., 2001). Introduction of streptomycin into Bambara nut might help improve its morphological and yield characteristics, hence this study.

MATERIAL AND METHODS

Study Area

The study was carried out at the Botanical Garden, Federal University of Lafia, Nasarawa state, Nigeria.

Collection of Planting Materials

Three healthy and viable bambara nut *(Vigna subterranean)* accessions were gotten from International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria (TVSU 1, TVSU 9 AND TVSU 10).

Streptomycin was obtained from a Namples pharmaceutical store located along Abuja Road, Lafia, Nasarawa state.

Garden soil was collected from the research garden and filled into polyethene pots after which they were perforated at the bottom to allow excess water to flow out.

Reparation of Antibiotic Solution

Streptomycin was weighed, 0.1g, 0. 2g, 0.3g, 0.4g and 0.5g and each gram of streptomycin was dissolved into 1 liter of distilled water. The control used was water without any additive (Mshelmbula *et al*., 2012).

Pre – Treatment of Seeds with Streptomycin

Three accessions of various Bambara nut were primed in the varying concentrations of streptomycin for three hours. The treated seeds were washed in running water to remove excess chemicals and exudates from seeds and ready to be sown (Mshelmbula *et al*., 2019).

Sowing

The pre – soaked seeds were sown directly into the soil. Planting was done in the evening, just beyond sunset Ikhajiagbe, *et al*. (2014). Seeds were sown at the rate of three seeds per hole. The polyethylene bags filled with soil was placed at the research garden, thereafter, constant irrigation was carried out every morning and evening until full maturity was attained.

Experimental Design

The experimental design selected was Randomized Complete Block Design (RCBD) for field work. There were 5 treatments with each treatment consisting of 3 replicates. Polyethylene bags were properly labeled according to a given treatment name and replicate number. Polyethylene bags were randomized over the whole plot, each bearing an identification tag.

Parameters Measured

Number of Leaves per plant, Plant Height, stem girth, Leaf area, Number of Flowers and Number of Days to germination were considered.

Statistical Analysis

Statistical analysis was done using Analysis of Variance (ANOVA).

RESULTS

The number of leaves and plant height of the three accessions under different treatments

The number of leaves and plant heights of the three accessions under different treatments and control are presented in Table 1. The results showed that highest number of leaves (22.10) was observed in treatment 0.1g/L of TVSU-1 and it was significantly different from the control which had the lowest number of leaves (13.05). In TVSU-9, the number of leaves of control and treatment 0.3g/L were not significantly different and they had the highest number of leaves compared with others. The lowest number of leaves (13.76) was observed in the treatment 0.5g/L. Also, it was observed that the control, treatments 0.1g/L, 0.3g/L, and 0.4g/L had the highest number of leaves in TVSU-10 but were not significantly different from each other. Treatment 0.5 also recorded the lowest number of leaves (12.56) in TVSU-10. The plant heights of the control and all the treatments in TVSU-1 were not significantly different from each other. In TVSU9, the highest plant height (10.69 cm) was observed in the control and is significantly different from all the treatments except treatment 0.3g/L. However, in TVSU-10, treatment 0.3g/L had the highest plant height (12.04 cm) and it was significantly different from the control. Treatment 0.5g/L still had the lowest plant height (5.68 cm).

Table 1: The number of leaves and plant height of the three accessions under different treatments

TREATMENT	NUMBER OF LEAVES			PLANT HEIGHT (CM)		
	TVSU-1	TVSU-9	TVSU-10	TVSU-1	TVSU-9	TVSU-10
CONTROL	$13.05^{a} \pm 1.85$	$27.21^{e} \pm 2.08$	$24.41^{n} \pm 1.90$	$7.95^{a} \pm 1.08$	$10.69^{h} \pm 0.67$	$9.44^{g} \pm 0.52$
0.1	$22.10^{b} \pm 3.16$	$20.53^{d} \pm 2.89$	$20.36^{n} \pm 1.86$	$20.36^{n} \pm 1.86$	$7.96^{g} \pm 1.08$	$11.04^{gh} \pm 0.71$
0.2	$19.72^{ab} \pm 2.69$	$15.46^{d} \pm 2.11$	$13.23^{g} \pm 1.85$	$7.28^{a} \pm 0.95$	$6.37^{g} \pm 0.86$	$7.40^{a} \pm 0.89$
0.3	$13.64^{a} \pm 1.78$	$30.46^{e} \pm 2.12$	$18.79^{n} \pm 1.89$	$8.67^{a} \pm 1.08$	$10.65^{h} \pm 0.61$	$12.04^{h} \pm 0.68$
0.4	$21.12^{b} \pm 1.56$	$19.10^{d} \pm 2.19$	$22.21^{n} \pm 1.83$	$10.16^{a} \pm 0.78$	$7.44^{g} \pm 0.87$	$9.77^{g} \pm 0.49$
0.5	$20.13^{ab} \pm 2.93$	$13.76^{d} \pm 1.19$	$12.56^{g} \pm 1.97$	$8.26^{a} \pm 1.18$	$6.13^{g} \pm 0.85$	$5.68^{a} \pm 0.91$

Values represent mean ± standard error

Values with the same superscript across same column are not significantly different ($P \leq 0.05$)

The Girth and Leaf Area (Cm^2) of the three accessions under different treatments

The Girth and Leaf Area of the three accessions under different treatments and control are presented in Table 2. The results showed that there was no significant difference across all treatment of Streptomycin concentration on TVSU-1 accession. TVSU-9 was significantly different across all treatments, however, treatment 0.3g/L had the highest Girth (0.13). On TVSU-10, there was significant difference across all treatments, however treatment 0.4g/L had the highest Girth alongside Control (0.11).

On the leaf area, there was no significant difference across all treatment in TVSU-1. TVSU-9 had significant difference across all treatment, however Control had the highest Leaf area (9.11). TVSU-10 had significant difference across all treatment, 0.4g/L had the highest leaf area.

Table 2: The Girth and Leaf Area (Cm^2) of the three accessions under different treatments

TREATMENTS	GIRTH			LEAF AREA (cm^2)		
	TVSU-1	TVSU-9	TVSU-10	TVSU-1	TVSU-9	TVSU-10
CONTROL	0.10a ± 0.20	0.12ab ± 0.21	0.11wj ± 0.02	5.72d ± 0.98	9.11d ± 0.70	6.83g ± 0.60
0.1	0.11a ± 0.22	0.10ab ± 0.21	0.12j ± 0.02	8.68d ± 1.11	5.80a ± 0.70	7.91g ± 0.60
0.2	0.10 a± 0.21	0.70a ± 0.20	0.10wj ± 0.02	9.46d ± 1.50	5.40a ± 1.30	4.20dy ± 0.32
0.3	0.12a ± 0.21	0.13b ± 0.21	0.13j ± 0.02	7.00d ± 0.94	6.91ad ± 0.80	0.53d ± 0.33
0.4	0.11a ± 0.21	0.10ab ± 0.20	0.11wj ± 0.02	7.91d ± 0.84	6.31a ± 0.80	8.02g ± 0.50
0.5	0.10a ± 0.22	0.10a ± 0.02	0.10w ± 0.02	9.48d ± 1.40	5.60a ± 0.90	3.20y ± 0.5

Values represent mean ± standard error
Values with the same superscript across same column are not significantly different (P≤0.05)

The Number of Flowers and Number of Days to Germination of the three accessions under different treatments

The Number of Flowers and Number of Days to germination of the three accessions under different treatments and control are shown in Table 3. On the Number of Flowers, there was no significant difference across all treatments in TVSU-1. In TVSU-9 there was significant difference between Control compared all other treatments. There was no significant difference across all treatments in TVSU-10 accession.

On the number of days to germination, there was no significant difference across all treatments in TVSU-1. Similarly, there was no significant difference across all treatments in TVSU-9 and TVSU-10.

Table 3: The Number of Flowers and Number of Days to Germination of the three accessions under different treatments

TREATMENTS	NO. OF FLOWERS			NO. OF DAYS TO GERMINATION		
	TVSU-1	TVSU-9	TVSU-10	TVSU-1	TVSU-9	TVSU-10
CONTROL	0.23b ± 0.12	0.80dg ± 0.30	0.50a ± 0.30	12.00s ± 2.10	12.00g ± 1.20	10.00p ± 1.20
0.1	0.53b ± 0.23	0.33d ± 0.14	0.90a ± 0.31	8.00s ± 0.00	6.33g ± 3.20	9.00p ± 0.00
0.2	0.50b ± 0.20	0.21d ± 0.12	0.50a ± 1.19	5.33s ± 2.70	11.00g ± 0.60	11.33p ± 1.33
0.3	0.80b ± 0.43	1.21g ± 0.40	0.60a ± 0.24	10.00s ± 1.53	9.00g ± 0.60	9.33p ± 1.20
0.4	0.21b ± 0.83	0.50d ± 0.19	0.50a ± 1.20	13.33s ± 2.20	10.33g ± 0.33	12.33p ± 0.90
0.5	0.53b ± 0.30	0.41d ± 0.17	0.51a ± 0.21	10.33s ± 5.20	14.33g ± 3.33	10.70p ± 0.40

Values represent mean ± standard error
Values with the same superscript across same column are not significantly different (P≤0.05)

DISCUSSION

From the results, it was observed that there was significant difference in the Number of Leaves of TVSU-1 compared to the control at the least antibiotic treated seeds. The least antibiotic treatment seeds (0.1g/L) had a greater number of leaves compared to other treatments. This agrees with work done by Ikhajiagbe *et al*. (2014) who reported that there was increase in leaf primordial number among aminoglycosides treated Soybean seeds compared to the control. Mshelmbula *et al*., 2019 also reported that lower concentrations of antibiotics treated seeds of two maize varieties increased leaf number. This observation however differed among the various accessions considered; TVSU-9 had similar observation, however, higher antibiotics treatment seeds (0.3g/L) had the highest number of leaves (30.46). Also, it was observed that there was significant difference recorded among all the antibiotics treated seeds in terms of number of leaves. But then, the control had the highest number of leaves (24.40) compared to all other treatments which of course is in variant with the aforementioned results from the other accession under consideration. For the Plant Height, there was significant difference across all treatment in TVSU-1. The least concentration of Streptomycin had the highest Plant Height in this accession. This quite agrees with work done by Hills *et al*., 2011 who opined that tetracycline treated seeds showed increase in shoot height at all concentrations. Similar observation was recorded in TVSU-9 and TVSU-10 where concentration 0.3g/L had the highest plant height (10.65cm) at the end of week 14. This result however does not correlate with Mensah *et al.,* 2012 who reported that streptomycin treated seeds significantly reduced plant height of Bambara Nut. Further observations showed that the girth in TVSU-1 and TVSU-9 had no significant difference across all treatments considered; though treatment 0.3g/L had the highest girth size (0.12cm) in TVSU-9. This correlates with Mshelmbula *et al.* (2018) who reported that there were no significant differences in the stem girth of two varieties of Pepper after the application of Carboxylic acid. This suggests that there was no biochemical reaction that took place in the girth of the plant as a result of the presence of streptomycin concentrations. From the results obtained, the number of flowers obtained from TVSU-1 were all not significantly different; however, treatment 0.3mg/L recorded the highest number of flowers at the 14 weeks after planting. Similar finding was seen from TVSU-10 where the least treatment (0.1g/L) recorded the highest number of flowers at the end of week 14 (0.90). This result is in agreement with Mshelmbula *et al*. (2019) who reported that there was increase in the number of flowers of white and yellow maize antibiotic treated varieties after the application of ampiclox. This suggests that there was biochemical reaction in the number of flowers as a result of the presence of streptomycin concentrations in Bambara nut accessions. The number of days to germination showed that significant difference was not recorded across all

treatments and accessions, however, treatment 0.3g/L, 0.2g/L and 0.1g/L had the earliest days to germination across the three accessions respectively. Marguiles (1962, 1964) reported that chloramphenicol hinders some of the light dependent responses of bean seedlings, including the synthesis of chlorophyll and the development of photosynthetic activity. As both factors are key to total development of the seedling, this may account for the significantly low dry weight of the sprouted seedling as well as the low number of root branches. These light dependent responses inhibited by Chloramphenicol have been reported to be associated with the maturation of the chloroplast (Marguiles, 1962, 1964). This therefore strongly suggests the presence of Streptomycin aided hastening of days to germination among all the treated accessions. Also, Ikhajiagbe *et al*., 2014 said streptomycin had the least germination effects on Bambara nuts compared to Tetracycline.

CONCLUSION

In conclusion, from the results obtained, Streptomycin at high concentration increased the Number of leaves in TVSU-1 and TVSU-10, increased Girth in TVSU-1 and TVSU-9, Leaf Area also increased in TVSU-1 and TVSU-10. Also, there was increase in Number of flowers in accession TVSU-1 and TVSU-10. However, at lower concentration, plant height increased in TVSU-1 and TVSU-10. At those concentrations, agronomical characters were triggered. This enhanced treated Bambara nut compared to the control. This indicates that Streptomycin could be used at different concentrations for crop improvement.

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