INTEGRATED APPLICATION OF NAPHTHALENE ACETIC ACID, STAKING AND INORGANIC NITROGEN AND PHOSPHORUS ON NUTRIENT USE EFFICIENCY IN CLIMBING BEANS (*Phaseolus vulgaris L.*)

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Abstract

In Kenya climbing beans (*Phaseoulus vulgaris* L.) are mainly grown by small-scale farmers under limited resources, such as limited land and high cost of inorganic fertiliser inputs. These have had led to declining trends and low productivity of climbing beans as well as increasing environmental concerns in cases of excess fertilizer inputs. To mitigate these problems there is need examine the factors that can lead to increased nutrient use efficiency (NUE) by climbing beans. A study was conducted at Kaguru Farmers Training Centre, Kenya, to determine the effect of integrating naphthalene acetic acid (NAA), staking, Diammonium phosphate (DAP) fertiliser on NUE in climbing beans. A 3x3x4 factorial experiment laid out in Randomised Complete Block Design was used. Treatments included NAA concentrations (0, 200 and 400 ppm), staking (no staking, staking with wooden stakes and maize plants as support), and DAP (0, 200, 250 and 300 Kg DAP ha^{-1}). The DAP was the source of both nitrogen and phosphorus. The yield data was used to calculate NUE. The obtained data was then subjected to analysis of variance using SAS version 9.4 and significant means separated using LSD at $\alpha = 0.05$. There was significant difference ($p < 0.05$) in NUE amongst the different treatments. The integration of 200 ppm NAA, staking with wooden stakes and application of 250 Kg DAP ha^{-1} resulted in the highest NUE (18.84 kg/kg). Though increase in NAA showed a general tread of increase in NUE, application of DAP fertiliser seems to have reached an optimum at around 250 Kg DAP ha^{-1} The study demonstrate that integration of NAA, staking with wooden stakes and application of appropriate amount of DAP can lead to increased NUE by climbing beans. This study recommended use of appropriate agronomic practices that integrate 200 ppm of NAA, 250 Kg of DAP ha^{-1} and staking with wooden stakes for sustainable climbing bean production.

Key words: Climbing beans, Naphthalene acetic acid, Nutrient use efficiency, Staking

INTRODUCTION

Common beans (*Phaseolus vulgaris L.*) are an important grain legume for food and cash to the smallholder farmers worldwide (Nassary *et al.,* 2020) and are produced for direct consumption with commercial value exceeding all the other legumes (Broughton *et al*., 2003; Graham and Vance, 2003; Porch *et al*., 2013). The annual global production is approximately 26.5 million tonnes (FAOSTAT, 2018) and grain legumes have been used for improving food and nutrition security, generation of income, soil fertility improvement, providing livestock feed, soil erosion control, water conservation and as a source of fuel (Kebede, 2020). However, despite their importance the production per unit area in Kenya has remained constant or is declining (FAOSTAT 2018). This decline has been attributed to factors including unreliable weather, low soil fertility, especially nitrogen (N) and phosphorus (P) and poor agronomic practices (Namugwanya *et al*., 2014).

There are two types of bean growth habits, determinate (bush beans) and indeterminate (climbing), which require different agronomic practices. Climbing beans have a better yield potential of up to 4 to 5 tonnes ha^{-1}, produce more biomass and fix more nitrogen than bush beans (Bliss, 1993; Wortmann, 2001; Ramaekers *et al.,* 2013), with average yield of 1.45 tonnes ha^{-1} in Kenya. However, climbing beans have great potential for agricultural intensification especially in areas of high population pressure and small farm sizes (Katungi *et al*., 2018). The indeterminate growth habit requires support for optimal productivity. But most of the farmers either grow beans without staking or intercrop climbing beans with maize, thus using maize as their supporting material, hence reduced productivity of beans (Muraya *et al*, 2006). Moreover, intercropping beans and maize has been found to substantially reduce the maize yield. In addition, most farmers apply suboptimal levels of N and P due to insufficient information on optimal requirement of the inorganic N and P for climbing beans for different environments (Franke and de Adolf, 2011; Ruganzu *et al.,* 2014). The suboptimal application of these plant nutrients can mainly be attributed to inadequate agronomic information available on rates of fertiliser application for climbing beans, as many farmers apply same practices ment for bush beans in climbing beans (Wondimu and Tana, 2017). In Kenya, the recommended rates of fertiliser application for bush bean is 50 Kg/ha DAP fertiliser (KARI, 2008). This may not be appropriate for climbing beans since they have larger biomass production, therefore, require more nutrient input (Sperling and Muyaneza, 1995).

Despite the need to increase the fertiliser input for climbing beans, excess application of fertilizers not only increases production cost, but also cause severe environmental problems (Zhang *et al.,* 2020). Therefore, there is need to determine integration of various inputs, such as optimum application of inorganic fertilisers, NAA, and staking that will enhance nutrient use efficiency (NUE), while maintaining sustainable climbing beans production. Nutrient use efficiency is a measure of how well plants use the available mineral nutrients (Hawkesford *et al*., 2014; Salim and Raza, 2020). Fertilizers are considered to be utilized efficiently when maximum yield is obtained with minimum possible amount of fertiliser application (Duarah *et al.,* 2011). Improvement in NUE depends upon the management practices (McDonald *et al.* 2015) and it is important to derive strategies like application of NAA and staking that may increase efficiency of utilization of any applied N and P in order to enhance climbing bean yields per unit of applied N and P.

Application of phytohormones has become a useful practice to improve the yields of many crops (Alleon *et al*., 2000). Plant hormones regulate important processes in plants such as mineral nutrition, water relations, resistance to pathogens, and antioxidant functions (Kudoyarova *et al.,* 2019). The exogenous application of phytohormones affects the endogenous hormonal pattern of a plant either by supplementation of sub-optimal levels or by interaction with their synthesis, translocation or inactivation of existing hormone levels (Adam and Jahan 2011 and Basuchaudhuri, 2016). Naphthalene acetic acid (NAA) as a hormone, plays an important role in physiological processes that include synthesis of growth regulators (Abd *et al.,* 2018; Ismail and Fayed, 2020) and it plays an important role in growth and development of plants by promoting vegetative growth by active cell division, cell enlargement and cell elongation. The greater vegetative growth is crucial towards supporting the reproductive growth phase eventually increasing yield. The excessive vegetative growth due to NAA may require extra inorganic N and P. However, there is limited information on the role of NAA on growth and consequent yields of climbing beans. Naphthalene acetic acid also increases the osmotic uptake of water and nutrients (Bairwa and Mishra, 2017). It had been shown to have positive effect on plant height and leaf chlorophyll content (Khandaker *et al*., 2017).

Although climbing beans are highly accepted by farmers, their major shortcoming is that they require stakes to support their growth and or they should be grown in association with other crops on which they climb (Takusewanya *et al.,* 2018). Staking is a very valuable agronomic practice in climbing bean production (Lwakuba *et al.,* 2003) because it support the weak, long and twisted stems and branches that enables reproductive adaptation (which results in pods distributed from the base to the top of the plant or production of pods for a long time), that increases the yield per unit area (Takusewanya *et al.,* 2018). Stakes up to 3.5 m high have been reported to be economical (Mcharo and Katafiire, 2014). Staking has a positive effect on the crop in that it allows for the vegetative adaptation that results in the crop growing faster. Staking of a climbing bean plant as it extends in length may provide it with the ability to grow without bending allowing it to obtain the sunlight required for growth and it also prevent the pods from contacting the ground and start rotting and the overall growth occurs optimally. Consequently, probably leading to improved nutrient use efficiency. This study determined the effect of integrating NAA, staking, and DAP fertilizer on nitrogen use efficiency and phosphorus use efficiency of the climbing beans.

MATERIALS AND METHODS

Study Site

The study was carried out at Kaguru Agricultural Training Centre (ATC), located in Imenti South Sub county of Meru County, Kenya, at latitude, 0^0 05’ south and longitude 37^0 40’ east. The average temperature range is 16 – 23 ^{0}C and lies at an altitude of 1289 m above sea level. The site has well drained, deep dark reddish-brown friable clay i.e. humic nitosols. The rainfall is bimodal with long rains occurring between March and May and short rains between October and December of every year. The site soil characteristics are presented on table 1.

Table 1: Experimental site soil characteristics

Soil characteristics	Unit	Value	Rating/Remarks
pH	-	5.83	Low
P	ppm	40	very low
K	ppm	111	very low
Ca	ppm	1030	Low
Mg	ppm	198	Medium
Na	ppm	39.7	Medium
Organic matter	%	4.2	Medium
N	%	0.32	Medium
CEC	Meq/100g	14.3	Low
Ca %	%	52.64	Low
Mg %	%	15.9	Medium
K %	%	2.39	Medium
Na % (ESP)	%	1.28	Medium

Experimental Design

In this study a 3x3x4 factorial experiment laid out on Randomised Complete Block Design was used. There were three factors, i.e, Naphthalene acetic acid (NAA) concentrations at three levels (0, 200 and 400 ppm), staking at three levels (no staking, staking with wooden stakes and maize plants as support) and Diammonium phosphate (DAP) fertiliser at four levels (0, 200, 250 and 300 Kg DAP ha^{-1}), resulting in a total of 36 treatments. The DAP was the source of N (equivalent of 0, 36, 45 and 58 kg N ha^{-1}) and P (equivalent of 0, 92, 115 and 138 kg P ha^{-1}), respectively. Each treatment had 135 bean plants arranged into three blocks. The experimental field was divided into three blocks and each block was then divided into 36 experimental plots. Each experimental plot measured 2.5 by 2.7 m and consisted of 5 rows of climbing beans and each row had 9 plants therefore making a total of 45 plants per each experimental plot.

Preparation of Naphthalene Acetic Acid

A stock solution of 1000 ppm was prepared by taking 1 ml of active ingredient in 1000 ml volumetric flask and diluted to mark. The lower concentrations of 200 ppm and 400 ppm was prepared by further dilution of stock solution using distilled water.

Field Trials

The MAC 34 (Kenya Mavuno) climbing bean variety, which was sourced from Kenya Agricultural and Livestock Research organisation (KALRO – Embu) was used in this study. MAC 34 is a high yielding climber which does well in all areas that have adequate rainfall of above 1000 mm. It matures in approximately 90 - 105 days and has a yield potential of 4 -5 tonnes/ha (KARI, 2008). The optimal production altitude lies between 1000 m and 2100 m above sea level. Maize hybrid H513 sourced from Kenya Seed Company was used in this experiment as a one of the staking treatment. The hybrid is well suited in the area and has relatively high yields.

The field experiments were carried out over three growing seasons, i.e., season 1 (March – July 2017), season 2 (October 2017 to February 2018) and season 3 (October 2018 to February 2019). The DAP (18-46-0) fertiliser supplying inorganic nitrogen and phosphorus was applied at planting time. Naphthalene acetic acid (NAA) was applied twice during the growing period. The first application was done fourteen days after crop emergence while the second application was done at the onset of flowering. A knapsack sprayer was used for spraying the NAA. In order to avoid the NAA spray to drift to unintended area, big tarpaulins mats were used for covering the area that was being sprayed. Stakes were applied at 21 days after planting. Wooden stakes measuring 3.5 m and diameter of approximately 2 cm were pegged next to the bean plants this ensured that every shoot touched the stake in order to enhance early coiling hence offering support. Each stake supported two climbing bean plants. The maize for supporting bean plants were planted seven days before planting of beans at a spacing of 75 by 25 cm.

Determination of the Grain Yield

At physiological maturity the grain yield per experimental plot was determined for all harvestable rows of the net plot. All pods were harvested, sundried for 14 days, threshed and then cleaned. The grains were then weighed. These grains from the net plot (experimental plot) were converted to Kgha^{-1}after adjustment to 12% moisture content using a John Deere SW moisture tester.

Determination of Nitrogen Use Efficiency and Phosphorus Use Efficiency

The nitrogen use efficiency was determined by dividing the total yield per hectare with the actual amount of nitrogen applied per hectare as per the rate according to Sabine *et al.*, (2014). The nitrogen use efficiency was computed as follows; NUE = Total grain yield / N supply. The phosphorus use efficiency was determined by dividing the total yield per hectare of the climbing beans in every treatment with the actual amount of phosphorus applied per hectare as per the rate according to Hawkesford *et al.*, (2014). The phosphorus use efficiency (PUE) was computed as follows; PUE = Total grain yield / P applied.

Data Analysis

The data collected were subjected to analysis of variance using Statistical Analysis Software (SAS) version 9.3.The significant means were separated using LSD at $\alpha = 0.05$.

RESULTS

The results showed that there was significant effect ($p < 0.05$) of treatments and the seasons on the nutrient use efficiency (Table 2). However, there was no significant interaction effect ($p > 0.05$) of treatments and seasons on the nutrient use efficiency of the climbing beans. Indicating homogeneity of variance and hence the data for the three season was pooled in further analysis.

The study revealed that there was significant ($p < 0.05$) effect of each factor and their interaction on nutrient use efficiency (Table 3). However, the blocking of treatments was found to be effective ($p = 0.2518$).

An increase in amount of foliar application of NAA, resulted to an increase in NUE by climbing beans, with the application of 200 ppm giving the highest NUE, while no application of NAA giving the lowest (Table 4). Staking with wooden stakes gave the highest NUE by climbing beans compared to other types of staking. Application of 250 Kg DAPha^{-1}and 350 Kg DAPha^{-1} did not result in a significant increase in NUE by the climbing beans, perhaps suggesting that an optimum DAP application had been achieved.

Analysis of combined effect of factors (treatments) revealed significant ($p < 0.05$) difference in NUE by climbing beans, ranging from 0 to 18.84 kg/kg (Table 5). The treatment integrating 200 ppm NAA, staking with wooden stakes and 250 Kg of DAPha^{-1} fertiliser (A2S2F3) resulted in the highest NUE by the climbing beans. Generally, treatments which integrated 200 ppm NAA had higher NUE by the climbing beans. Moreover, treatments integrating staking with wooden stakes also gave higher NUE by the climbing beans compared to those integrating other types of staking.

Table 2: Analysis of variance for effect of naphthalene acetic acid, staking, and application of Diammonium phosphate fertiliser on nutrient use efficiency of climbing beans

Source of variation	df	SS	MS	F-value	p-value
Block	2	14.897141	7.448571	1.54	0.2157
Treatment	35	8842.292525	252.636929	52.40	<0.0001
Season	2	89.090204	44.545102	9.24	0.0001
Treatment*Season	70	416.513307	5.950190	1.23	0.1293

Table 3: Analysis of variance for each factor and their combined effect for nutrient use efficiency

Source of variation	df	SS	MS	F-value	p-value
Block	2	14.897141	7.448571	1.39	0.2518
NAA	2	180.440845	90.220423	16.78	<0.0001
Staking	2	1533.257440	766.628720	142.62	<0.0001
DAP	3	6299.946737	2099.982246	390.66	<0.0001
NAA*Staking	4	74.271273	18.567818	3.45	0.0089
NAA*DAP	6	116.282261	19.380377	3.61	0.0018
Staking*DAP	6	532.258556	88.709759	16.50	<0.0001
NAA*Staking*DAP	12	105.835413	8.819618	1.64	0.0800

Where, NAA = naphthalene acetic acid and DAP = Diammonium phosphate fertiliser

Table 4: Mean of nutrient use efficiency of climbing beans under different factor levels.

Factor levels	NUE-NAA	Factor levels	NUE-Staking	Factor levels	NUE-DAP
N2	8.99a	S2	10.66a	F3	10.67a
N3	7.76b	S1	7.44b	F4	10.45ab
N1	6.98c	S3	5.22c	F2	9.70b
LSD	0.621	LSD	0.621	F1	0c
				LSD	0.717

Where, NUE-NAA = Nutrient use efficiency due to naphthalene acetic acid levels (N1 = 0 ppm, N2 = 200 ppm and N3 = 400 ppm); NUE-Staking = Nutrient use efficiency due to staking (S1 = No staking, S2 = Staking using wooden stakes, S3 = Supporting beans with maize as stakes) and NUE-DAP = nutrient use efficiency due to DAP application (F1 = 0 Kg DAPha^{-1}, F2 = 200 Kg DAPha^{-1}, F3 = 250 Kg DAPha^{-1} and F4 = 300 Kg DAPha^{-1}). [a]Means followed by the same letters are not significantly different at 5% probability level.

DISCUSSION

The foliar application of NAA, staking and application of DAP fertiliser produced significant effect on NUE by the climbing beans. Naphthalene acetic acid belongs to synthetic forms of auxins, which play a key role in cell elongation, cell division, vascular tissue, differentiation, root initiation, apical dominance, leaf senescence, leaf and fruit abscission, fruit setting and flowering (Davies, 1987). In this study, an increase in concentration of foliar application of NAA results to an increase in NUE by the climbing beans. Therefore, its application can be a good crop management practice towards sustainable climbing beans productivity. Application of NAA has been shown to improve plant growth under different growth stresses (Abou El-ghit, 2015). Application of NAA has been reported to promote plant growth, yield and yield attributes (Jahan *et al.,* 2019), thus potentially increasing NUE.

The staking with wooden stakes recorded higher NUE than the unstaked beans as well as beans supported by maize plants. Ronner *et al*., (2018) reported that climbing beans need stakes in order to realize their climbing potential. Staking has been reported to improve leaf arrangement and thus possibly reducing the occurrence of mutual leaf shedding, which might depress plant photosynthetic capacity (Onwueme & Johnston, 2000). In this study staking with maize is expected to have resulted to maize-climbing bean competition for growth resources and shading by maize plants that contributed to lower yields of climbing beans that reduced the NUE. Increasing solar radiation interception is an important practice associated with increases in crop yield, and where maize was used as a stake could have resulted the shading of climbing bean plants making them suffer from poor light conditions. Where the climbing beans were staked using wooden stakes, it is possible that there was better exposure of the leaves to sunlight enabling them to capture more solar radiation that resulted to optimum photosynthesis.

Plant productivity is limited by source activity (photosynthesis) and the source to sink ratio can be easily reduced by partial shading of leaves (Glanz-Idan *et al.,* 2020). A possible strategy to improve photosynthetic efficiency is to optimize light collection and use (Cardona *et al.,* 2018). Staking the climbing beans causes an increase in vegetative growth, thus increasing photosynthetic surface and eventually contributes to increase in yield (Takusewanya, 2018). The stakes that are placed in vertical position make the crop grow upwards producing fresh healthy leaves and pods. The production of fresh healthy leaves, the upward growth and greater exposure of leaves resulting from staking with wooden stakes created a better plant architecture, photosynthetic capacity and production of pods, which cumulatively increased climbing bean grain yield and thus NUE.

Table 5: Means of nutrient use efficiency of climbing beans under different treatment

Treatment	Nutrient use efficiency
A2S2F3	18.84 a
A2S2F4	17.12 b
A2S2F2	14.50 c
A3S2F2	14.20 c
A1S2F3	13.33 cd
A3S2F4	12.78 cd
A3S2F3	12.75 cd
A2S1F3	11.78 de
A1S2F4	11.67 de
A1S2F2	1f1.51 d
A3S1F4	11.10 defg
A2S1F4	10.26 efgh
A3S1F2	9.50 fghi
A1S1F4	9.17 ghij
A2S1F2	9.07 ghij
A3S1F3	8.94 hijk
A1S1F2	8.77 hijk
A1S1F3	8.62 hijk
A2S3F4	7.73 ijklm
A2S3F3	7.50 ijklm
A2S3F2	9.49 ijklm
A3S3F3	7.41 jklm
A3S3F4	6.91klm
A1S3F2	6.71 lm
A1S3F3	6.23 m
A1S3F4	5.98 m
A3S3F2	5.80 m
A1S1F1	0 n
A1S2F1	0 n
A1S3F1	0 n
A2S1F1	0 n
A2S2F1	0 n
A2S3F1	0 n
A3S1F1	0 n
A3S2F1	0 n
A3S3F1	0 n
LSD	0.901
CV%	28.82
R^2	2.04

Where, A1= 0 ppm, A2 = 200 ppm, A3 = 400 ppm, S1 = No staking, S2 = Staking using wooden stakes, S3 = Supporting beans with maize as stakes, F1 = 0 Kg DAPha^{-1}, F2 = 200 Kg DAPha^{-1}, F3 = 250 Kg DAPha^{-1} and F4 = 300 Kg DAPha^{-1}. ^{a}Means followed by the same letters are not significantly different at 5% probability level.

The unstaked climbing bean plants had limited vegetative growth and the stems of the beans coiled around each other for support. This led to clustering of the leaves together which might have resulted to less exposure to the solar radiation, which may have led to lesser photosynthesis that resulted to lower yields. Musoni *et al*. (2014) reported that the use of stakes was the best option to maximise potential yield benefits from the climbing beans. The climbing bean plants supported by maize plants had an advantage over the unstaked plants, in that the maize plants offered support to climbing beans. The climbing beans coiled around maize plants, however, bean leaves were overshadowed by the maize plants, which resulted to low solar radiation interception. This causes differential light interception, which can be attributed to inefficiency in nutrient use. Studies have shown that crop development causes differences in light interception and dry matter production, with the latter being always positively correlated with light interception (Zhang *et al*. 2016; Zhi-qiang *et al.,* 2018). The less solar radiation interception resulted to less photosynthesis and decreased supply of photoassimilates leading to low yields in case of unstaked and supported by maize plant treatments. Consequently, lower NUE compared to the climbing beans supported by wooden stakes. Moreover, intercropping maize and beans is a major contributing factor to low climbing bean production (Muraya *et al*, 2006). Staking with wooden stakes or supporting bean plant with maize was also found to increase the quality of harvested bean seeds, since the pods were off the ground that minimized diseases like rots, thereby increasing marketable yield.

Application of DAP fertilizer significantly influence NUE by climbing beans. Plant nutrition is one of the most important factors that increase plant productivity (Barzegar *et al.,* 2020). Climbing bean require more nutrients uptake because of their larger biomass (Sperling and Muyaneza, 1995) and the extra vegetative growth due to the influence of staking demanded for more inorganic N and P. Nitrogen promote production, partitioning and accumulation of dry matter in crop plant (Akanbi *et al*. 2007). Nitrogen has been reported to influence root development, high nutrient uptake and long duration of photosynthesis (Ning *et al*., 2013; Dordas, 2009) while phosphorus improves early root formation which facilitate early nodulation hence enhanced productivity (Chekanai *et al*., 2018). However, in this study application of very high DAP fertiliser rates (as source of N and P) showed non-significant increase in NUE. This demonstrated the importance of optimising inorganic fertiliser, to avoid nutrient use inefficiency, which may lead to environmental pollution due to excess fertilizer inputs.

CONCLUSION

The results of this study showed that integration of optimum foliar application of NAA, DAP fertilizer and staking increases NUE by the climbing beans. This can be attributed to change in plant canopy arrangement, improved vegetative growth, increased photosynthetic capacity, increased radiation interception, increased number of pods and eventually increased marketable grain yield. The study recommends foliar application of 200 ppm of NAA, 250 Kg DAP/ha and staking with wooden stakes for optimum NUE by the climbing bean and thereby ensuring sustainable climbing beans production.

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COMPETING INTERESTS

Authors have no competing interests.

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