Growth, nodulation and nutrients uptakes of cowpea (*Vigna unguiculata* L. Walp) following Zinc fertilizer applications in the semi-deciduous forest zone of Ghana

Abstract

Cowpea can fix atmospheric nitrogen through symbiotic association with indigenous rhizobia but unfortunately, the amount of N_2-fixed is usually not enough due to the presence of ineffective or low numbers of indigenous rhizobia in the soil. The effect of Zinc applications on growth, nodulation and nutrient uptakes of cowpea was investigated during the major and minor cropping seasons (2016). Cowpea seed varieties were treated to foliar spray with three different rates of Zinc sulfate (0, 5 and 10 kg Zn ha^{-1}) at 3 and 5 weeks after sowing. The split plot design was used for both cropping seasons. This study shows that the supply of Zn fertilizer applications did not affect growth and nodulation in 2016 major and minor cropping seasons. The application of the 5 kg Zn ha^{-1} led to better cowpea production and greatly improve the quantity (haulm and grain yield) and quality (NPK content and crude protein) of cowpea in both haulm and grain. The Zinc fertilizer significantly enhanced N_2-Fixed in both cropping season's trial investigating. These findings suggest that cowpea responds differently to Zinc Sulphate application depending on its rates and the application of the 5 kg Zn ha^{-1} is the optimum rate that will enhance the yield and nutrient quality of cowpea in the Semi-Deciduous Forest Zone of Ghana.

Keywords: N_2-fixed, Nodulation, Zn fertilizer, NPK uptake and yield

INTRODUCTION

Proper nutrition of plants with micronutrients depends on various factors, such as the rate of absorption of nutrients by the plants, distribution of nutrients to functional sites and nutrient mobility within the plant. Interactions occur between the micronutrients and some nutrients

[1, 2, 3]. The amount of nitrogen fixed is usually high in soils with low mineral N but with sufficient water and enough of other nutrients capable of supporting plant growth [4]. Another factor is the differential response of plants to one nutrient in combination with varying levels of a second element applied simultaneously i.e. the two elements combine to produce an added effect not due to each of them acting alone [1, 2]. Such interactions may take place in the soil and within the plant [3]. However, the amount of nutrients uptake is strongly dependent on nutritional and environmental factors.

Cowpea is especially important for dry savannah of West Africa between latitudes 7 and 14°N [5] and second after groundnut as the most important legume of Ghana in terms of space under cultivation (156,000 ha) and quantity produced and consumed annually (143,000 Mg) making Ghana among the largest cowpea producer in Africa [6]. Cowpea is a protein-rich component of an otherwise protein-poor diet [7]. Many researchers have observed that Zn have a positive relationship with the nitrogen metabolism pathway of plants, its deficiency cause a reduction in protein synthesis into the plants. Epstein and Bloom [8] identified the positive relationship between the flowering and fruiting process and Zn. As micronutrient, Zinc has received much recent attention [9] because it is present in all body tissues and fluids [10].

The native rhizobia are often low in numbers or ineffective and are therefore not able to fix enough nitrogen to meet the nitrogen demand of plants. Including the Zn effect in this study will help to determine the optimal rate that can maximize the dual mineral contents and nodulation for better cowpea production.

MATERIALS AND METHODS

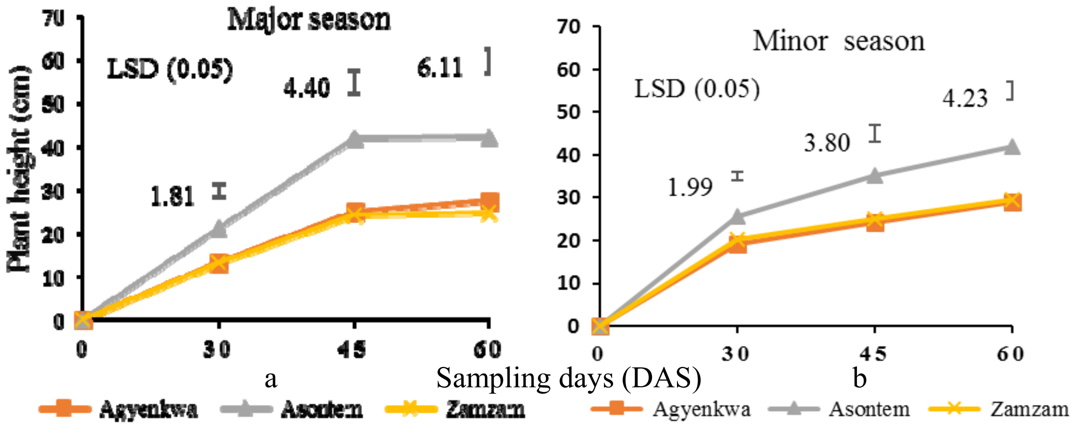
The study was conducted at the Plantation Section of the Department of Crop and Soil Sciences, Faculty of Agriculture, KNUST, in the cropping seasons of 2016. The site is located at 06° 45’ N and 01° 31’ W in the rainforest belt of Ghana. The total nitrogen content was low with a mean value of 0.06%, available P content was low with value of 6.4 mg kg^{-1}, soil Zn content was moderately low, found to be 1.290 mg kg^{-1}. The experiment design was split plot, with treatments arranged in Randomized Completely Block Design (RCBD). The factors assessed were cowpea varieties (main-plot factor) and Zn fertilizer (sub-plot factor). The treatment combinations were replicated four times in 2016 major and minor cropping seasons. Cowpea varieties were obtained from the Council for Scientific and Industrial Research (CSIR) at Fumesua, Kumasi, Ghana. Three early maturing cowpea varieties (Asontem, Agyenkwa and Zamzam) were grown in 2016 major and minor experiments and selected according to their yield, number of days to physiological maturity (62-70 days) and availability in the study area. The Zn fertilizer was obtained from “Chinese woman company”, one of fertilizer shops in Kumasi, Ghana. Zinc sulfate heptahydrate ($ZnSO_4.7H_2O$) was applied at a rate of 44.86 kg $ZnSO_4$ ha^{-1} (equivalent to 10 kg Zn ha^{-1}) and 22.43 kg $ZnSO_4$ ha^{-1} (equivalent to 5 kg Zn ha^{-1}). Foliar application to cowpea was done by dissolving 1.0 kg of the zinc sulphate salt into 278 litres of distilled water [11] and was sprayed on plant leaves at 3 weeks (40%) and 5 weeks (60%) after sowing when canopy/leaves had established. The application was done early morning before 9:00 am, using a sprayer. Urea and Triple Super Phosphate (TSP) fertilizers were applied as band placement by making a furrow of 5-7 cm deep and covering with 2 cm of soil. As starter nitrogen, Urea at the rate of 20 kg N ha^{-1} and Triple Super Phosphate (TSP) at the rate of 40 kg P_20_5 ha^{-1} were applied uniformly to all plots at two weeks after sowing (WAS). The plot (3 x 2 cm) was demarcated three days after harrowing and seeds were sown by hand using manual labour. Seeds were sown at a spacing of 60 cm x 20 cm with a rate of two seeds per hill at the depth of 3-5 cm. The first (3 weeks after sowing) and the second (7 weeks after sowing) weeding were done manually using hand hoe. Standard agronomic and plant protection treatments were used uniformly across the plots for the duration of the experiment. Grass hoppers *(Empoasca kerri Pruth),* Thrips *(Caliothrips indicus Bagnall)* and Aphids (*Aphis craccivora Koch*) were pests, respectively at vegetative stage and flowering to the end of pod filling. Lambda master 2.5 % E.C. [Active ingredients (Lambda-Cyhalothrin, 9.8 %)] was the pesticide used for pests' control.

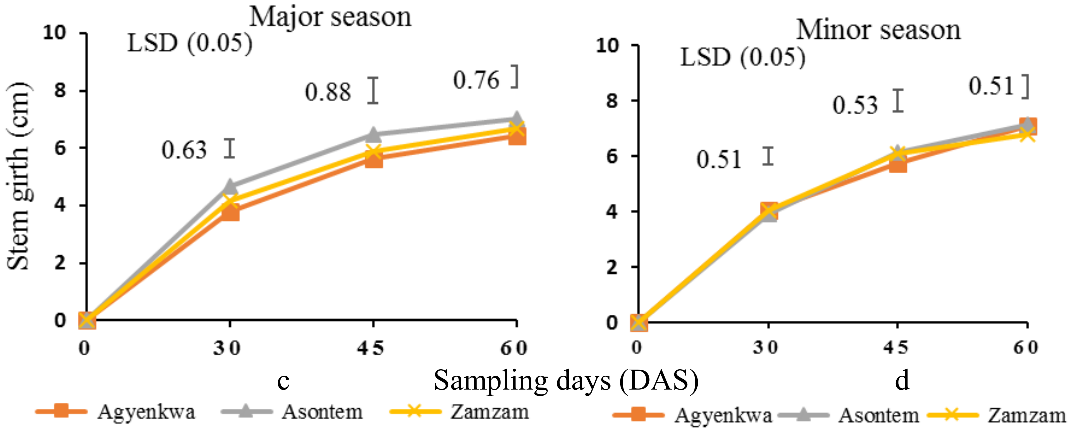
A random sample of five plants from each plot were selected and tagged to measure. Plant height, stem girth and number of leaves were measured at 30, 45 and 60 days after sowing (DAS) and mean for each plot was calculated. Nodules were sampled at 30 and 45 days after sowing. Plant samples were uprooted gently washed with water and the total nodules counted and the mean calculated for each plot. The ground was sufficiently soaked with water 48 hours before sampling to each uprooting of plants. To determine nodule effectiveness, nodules were cut open using a razor blade and hand lens. Nodules with pink or reddish colour were considered effective and fixing nitrogen, while those with green or colourless appearance were recorded as ineffective nodules. Nodules per plot were kept in labelled envelops and sent to the laboratory to oven-dry at 70°C for 48 hours. Average dry weight of nodules per plant was computed and expressed in grams. For mineral content analysis, random samples of five plants were uprooted gently from each plot at harvest and the root system was removed. The above ground parts were put in labelled envelops and oven dry at 70° C for 72 hours and milled and one hundred gram samples of each of the plant part (seeds and haulms) were taken to determine nitrogen, phosphorus and potassium content. The nitrogen content was determined using the Kjeldahl method [12]. The protein content of seed was determined on the basis of total nitrogen content [13]. Phosphorus (P), the content was measured on the Spectronic 20 spectrophotometer to give absorbance measurements at a wavelength of 420 nm. The observed absorbance was used to determine the P content from the standard curve [15, 16] and Potassium (K) was obtained using the flame photometer. From the standard curve, the concentration of K was calculated using the particular absorbance observed for the sample. NPK uptake were done by multiplying the grain and haulm yield in kilograms per hectare by each analysed parameter separately, nitrogen, phosphorus and potassium, in the grain and haulm then divided by 100 percent. This was done by multiplying the haulm yield in kilograms per hectare by concentrations of Nitrogen, Phosphorus and Potassium. From total N in both grain and haulm of cowpea and reference crop (Omankwa maize variety), N-uptake by was obtained using the N-difference method [14]. The reference crop was planted at the same time with cowpea varieties during the major and minor seasons (2016). The total nitrogen content of the maize was 1.27 % in the grain and 0.62 % in the haulm. The yield of the reference crop was 1949 kg ha^{-1} (grain yield) and 2285 kg ha^{-1} (haulm yield). Zn content was determined using Perkins model 403 atomic absorption spectrophotometer after digestion. The file for the type of analysis and hollow cathode lamps were selected with appropriate wavelengths of 213.9 nm [17]. The grain and straw yields were recorded separately. Total Zn uptakes by grain and tissue were computed by multiplying Zn content and their respective dry weights ha^{-1}. Data collected were subjected to analysis of variance (ANOVA) according to steel and Torrie [18] using GenStat statistical software [19]. The Least Significant Difference (LSD's test) was used to compare mean data when the probability level was significant.

RESULTS

1. Effects of cowpea varieties on growth

Fig. 1 illustrates the effect of different cowpea varieties on plant height (cm), stem girth (cm) and number of leaves/plant over the period of the experiment. The significant effect at 5% level of probability of cowpea varieties used was recorded over all sampling period of the study. The tallest plant was obtained by Asontem variety and the lowest by Zamzam. However, cowpea varieties did not show any significant ($P > 0.05$) effect on stem girth and number of leaves. Additionally, variety by Zinc rates was not significantly different on all days of sampling.





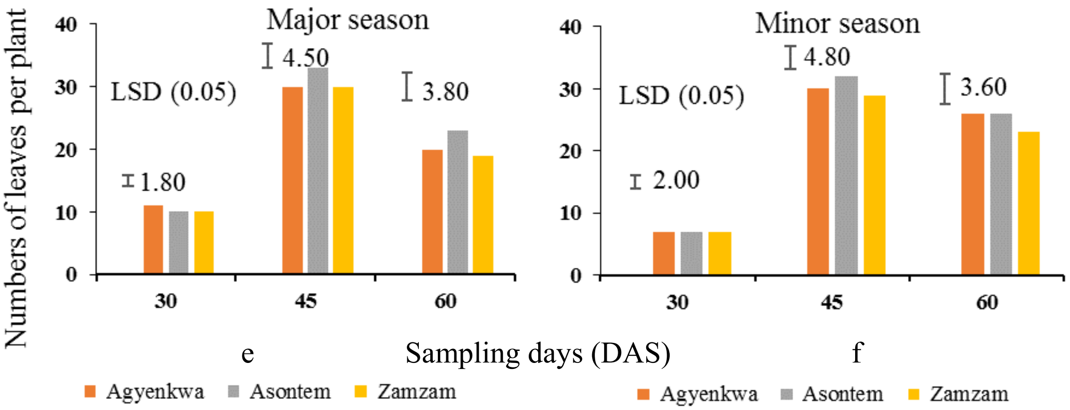


Figure 1. Effects of cowpea varieties on Plant height (a and b), stem girth (c and d) and number of leaves (e and f) under Zinc foliar application in 2016 cropping seasons

2. Effects of Zinc rates on growth

Fig. 2 illustrates the effect of different rates of Zn fertilizer application on plant height (cm), stem girth (cm) and number of leaves/plant over the period of the experiment. Analysis of variance showed no significant effect of Zn fertilizer on plant height and leaf production. Branch production was significantly affected by Zn rates in major and minor cropping seasons. At 30 DAS and 45 DAS, the 5 kg/ha treatment effect was significantly higher than other treatment effects. At 60 DAS, the control treatment effect was significantly lower ($P < 0.05$) than all Zinc treatments. Variety by zinc rates interaction was not significant at 5% level of probability on all sampling days.

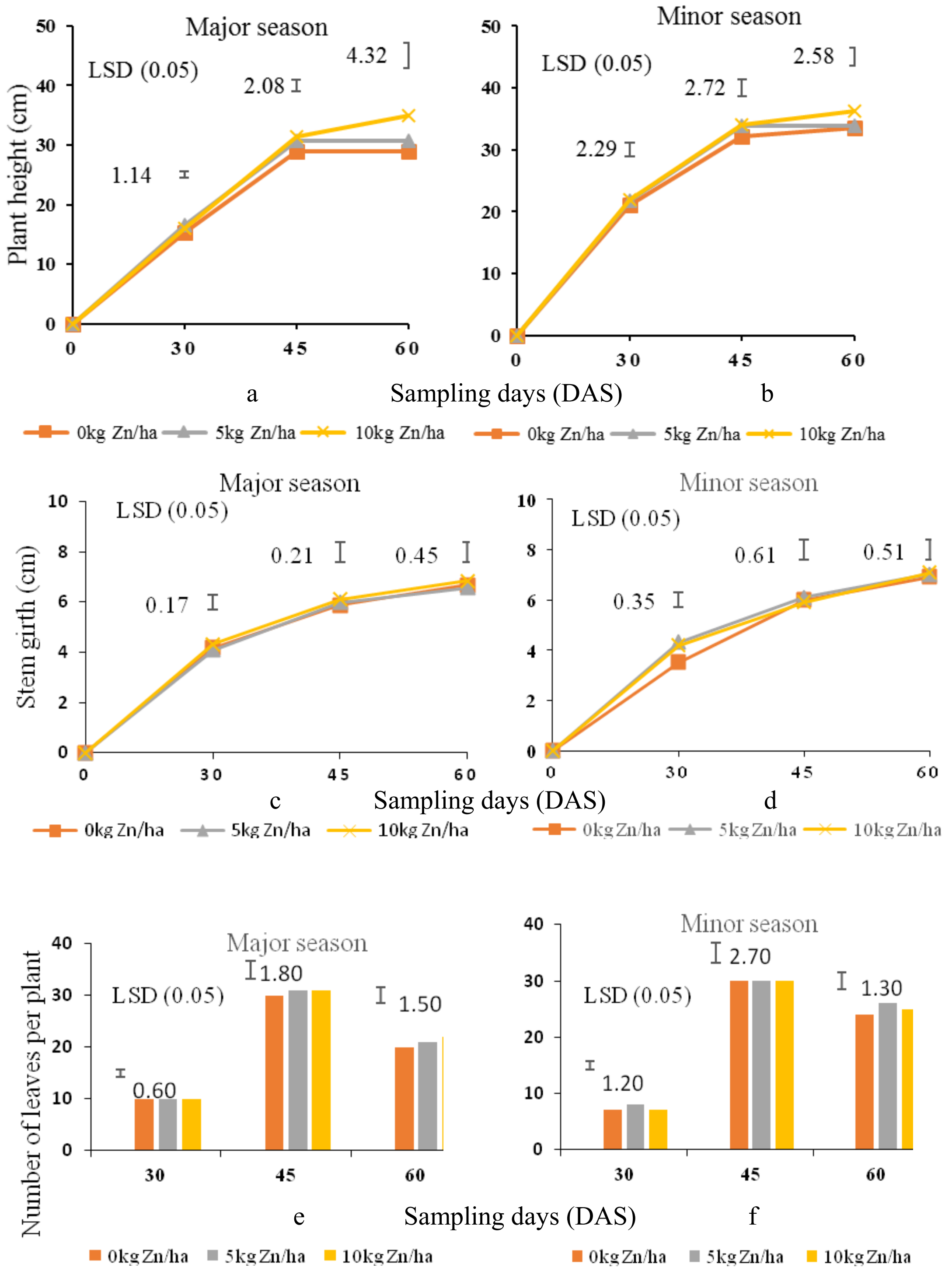


Figure 2. Effects of Zinc rates on plant height (a and b), stem girth (c and d) and number of leaves (e and f) in 2016 cropping seasons

3. Nodulation parameters

Results on number of nodules per plant, effective nodules per plant and nodule dry weight per plant as influenced by cowpea varieties and Zinc fertilizer application in the two sampling periods in both experiments are presented in Tables 1 and 2. Treatment differences for all parameters on all days at both seasons were not significant (P > 0.05). The interaction effect was also not significant at 5% probability. Nodule number was nearly successively decreased over time at all treatments and is not correlated with the Zinc fertilizer applied. No interaction effect was significant for all parameters at all sampling periods.

Table 1. Effect of cowpea varieties in changes of nodule number (nodules/ plant), nodule dry weight (g/ plant) and effective nodules (%)

	Time (Days after sowing)			
	Major season		**Minor season**	
	30	**45**	**30**	**45**
Varieties	**Nodule number (nodules/ plant)**			
Agyenkwa	5	3	7	6
Asontem	8	5	10	6
Zamzam	5	4	8	6
LSD (0.05)	NS	NS	NS	NS
CV (%)	21.8	31.8	14.2	22.4
	Nodule dry weight (g/ plant)			
Agyenkwa	0.06	0.09	0.22	0.16
Asontem	0.04	0.08	0.19	0.14
Zamzam	0.05	0.12	0.18	0.13
LSD (0.05)	NS	NS	NS	NS
CV (%)	18.8	21.0	10.5	20.3
	Effective nodules (%)			
Agyenkwa	82.54	37.82	84.48	47.55
Asontem	74.44	32.64	76.56	42.11
Zamzam	76.94	45.13	79.72	68.27
LSD (0.05)	NS	NS	NS	NS
CV (%)	4.9	5.5	2.4	13.3

Variety x Zn rate	NS	NS	NS	NS

Table 2. Changes in nodule number (nodules per plant), nodule dry weight (g per plant) and effective nodules (%) of cowpea growing under Zinc foliar application

	Time (Days after sowing)			
	Major season		Minor season	
	30	45	30	45
Rates	**Nodule number (nodules/ plant)**			
0	6	4	8	6
5	5	4	8	5
10	7	4	9	6
LSD (0.05)	NS	NS	NS	NS
CV (%)	11.3	13.8	5.0	11.9
	Nodule dry weight (g/ plant)			
0	0.05	0.11	0.18	0.15
5	0.05	0.09	0.20	0.15
10	0.06	0.08	0.21	0.14
LSD (0.05)	NS	NS	NS	NS
CV (%)	9.8	22.8	7.1	28.5
	Effective nodules (%)			
0	78.56	43.33	81,97	49.25
5	74.26	53.11	76,48	48.88
10	81.11	39.16	82,31	59.80
LSD (0.05)	NS	NS	NS	NS
CV (%)	3.6	8.8	4.4	8.4
Variety x Zn rate	NS	NS	NS	NS

4. Yield

The cowpea grain yield was significant ($P < 0.05$) under Zinc fertilizer application in all the two seasons (Table 3). Cowpea grain yield recorded on the application of Zinc fertilizer at 5 kg/ha increased at 28 % for Agyenkwa > Zamzam (20 %) > Asontem (19%) compare to the control in major season and the minor follows the same trend. The Zinc levels are increased the cowpea grain yield in the order: 5 kg Zn ha^{-1} > 10 kg Zn ha^{-1} > 0 kg Zn ha^{-1} during all cropping seasons (2016 major and minor seasons). The cowpea grain yield decline over increasing the Zinc rate beyond 5 kg ha^{-1}. There was about 6 % and 10 % yield reduction in the major season obtained with Agyenkwa and Asontem respectively. For one hundred seed weights, there were different results at 5% level of probability. One hundred seeds weight was higher with Zamzam following by Agyenkwa and at the end Asontem with the lowest one. The shoot dry weight was significant ($p < 0.05$) affect by Zinc fertilizer application. Similarly, cowpea varieties did significant ($P < 0.05$) affect the cowpea biomass yield in all sampling periods and the interaction follows the same trend.

Table 3. Effects of Zn rates on harvest index, haulm and grain yield of cowpea

Treatments	**Major season**			**Minor season**		
	100 seeds weight	**Haulm**	**Grain yield**	**100 seeds weight**	**Haulm**	**Grain yield**
	← g →	**← kg/ha →**		**← g →**	**← kg/ha →**	
Varieties						
Agyenkwa	16.25^{a}	1352.74^{b}	1142.23^{ab}	16.20^{a}	1311.58^{b}	1620.10^{a}
Asontem	13.63^{b}	1596.68^{a}	1082.15^{b}	13.26^{b}	1650.58^{ab}	1326.17^{b}
Zamzam	17.18^{a}	1470.64^{ab}	1423.62^{a}	17.07^{a}	1630.33^{a}	1707.63^{a}
LSD (0.05)	1.29	242.94	268.69	1.25	317.90	125.21
CV (%)	4.7	13.40	12.80	4.71	6.40	6.10
Zn levels (kg ha^{-1})						
0	15.34	1351.48^{b}	1087.45^{b}	14.93^{b}	1340.58^{b}	1493.22^{b}
5	15.58	1440.24^{ab}	1283.94^{a}	15.61^{a}	1587.67^{a}	1600.13^{a}
10	16.14	1558.34^{a}	1276.60^{a}	16.00^{a}	1564.25^{a}	1560.27^{a}
LSD (0.05)	NS	205.13	120.87	0.79	218.49	79.41
CV (%)	5.5	17.20	16.00	4.72	10.40	4.70
Variety x Zn rate	*	NS	*	*	NS	*

5. N-uptake and crude protein

The results of cowpea N-uptake and crude protein at all sampling periods are presented in Table 4. In this study, Zinc fertilizer application interacted to significantly ($p < 0.05$) affect cowpea N-uptake and crude protein. Zinc levels increased the cowpea N-uptake and crude protein in the order: 5 kg Zn ha^{-1} > 10 kg Zn ha^{-1} > 0 kg Zn ha^{-1}. N-uptake and crude protein interaction differed significantly ($p < 0.05$) among some the treatment interactions. Zamzam variety interacted markedly to produce the highest value of N-uptake in cowpea haulm and grain and Agyenkwa presented the lowest one. For the cowpea crude protein, the result is presented in the following order: Asontem > Agyenkwa > Zamzam.

Table 4. Effects of Zn rates on cowpea N-uptake and crude protein

Treatment	Major season			Minor season		
	Grain N-uptake	Tissue N-uptake	Crude protein	Grain N-uptake	Tissue N-uptake	Crude protein
	← kg ha^{-1}	→	(%)	← kg ha^{-1}	→	(%)
Varieties						
Agyenkwa	22.40	16.72^{b}	25.80^{b}	39.61^{a}	15.70	24.84^{b}
Asontem	26.40	27.64^{a}	29.44^{a}	32.23^{b}	16.59	26.86^{a}
Zamzam	31.80	30.82^{a}	24.87^{c}	39.14^{a}	20.95	23.44^{c}
LSD (0.05)	NS	12.32	0.91	4.81	NS	1.46
CV (%)	16.60	13.50	2.00	10.40	3.20	3.40
Zn levels (kg ha^{-1})						
0	21.80^{b}	24.12^{b}	25.88^{b}	33.59^{b}	14.12^{b}	24.27^{b}
5	30.90^{a}	27.26^{ab}	27.28^{a}	38.39^{a}	20.09^{a}	25.56^{a}
10	27.90^{a}	31.79^{a}	26.95^{a}	39.00^{a}	19.03^{a}	25.01^{ab}
LSD (0.05)	5.26	7.11	0.59	4.81	5.58	1.15
CV (%)	17.30	14.30	1.40	4.00	2.30	4.50
Variety x Zn rate	*	NS	*	*	NS	*

6. Effects of Zinc rates on NPK content

The results of grain nutrients analysis showed no significant varietal effects for content of nitrogen and phosphorus (Table 5). Additionally, potassium content in the haulms was not different among varieties. However, for potassium content in seed, the Zn treatments effects were similar, but greater either effect was greater than the control treatment effect in both cropping seasons (2016 major and minor seasons).

Table 5. NPK content of cowpea as affected by varieties

	Nutrient uptakes (kg ha^{-1})			
	Major season		Minor season	
	Haulm	Grain	Haulm	Grain
Rates		**N**		
Agyenkwa	40.92	47.21	29.90	64.41
Asontem	41.84	51.18	30.80	57.03
Zamzam	45.46	56.61	35.20	63.94
LSD (0.05)	NS	NS	NS	NS
CV (%)	8.4	6.9	9.40	6.2
		P		
Agyenkwa	3.23	5.46	3.53	12.19
Asontem	3.39	5.09	3.80	14.33
Zamzam	2.75	5.68	3.77	12.33
LSD (0.05)	NS	NS	NS	NS
CV (%)	7.5	6.5	7.1	7.70
		K		
Agyenkwa	14.32	12.19^{b}	13.95	20.92^{a}
Asontem	13.17	14.33^{a}	13.58	16.84^{b}
Zamzam	10.79	13.33^{a}	11.72	20.43^{a}
LSD (0.05)	NS	1.27	NS	2.89
CV (%)	12.1	7.70	7.70	4.30

For the Zn treatments, N content of grain was affected by Zn application in major and minor cropping seasons. Haulm N content was also significantly affected by Zn fertilizer application. In all these cases, treatments differences between the Zinc treatments were similar, but either effect was greater than the control treatment (Table 6). Haulm P contents were significantly affected by Zn fertilizer application (Table 6), with the exception haulm Zinc content in the major season, where the control treatment effect was similar to the 5-kg ha^{-1} Zn treatment. In all cases, the Zn treatment effects were similar, and either effect was significantly higher than the control treatment effect. Haulm K content was not affected by Zn fertilizer in the major and minor seasons (Table 6). However, grain K content significantly affected by the Zn fertilizer in the two cropping seasons. In both cropping seasons, grain K content in the control treatment was lower than the Zn treatment effects.

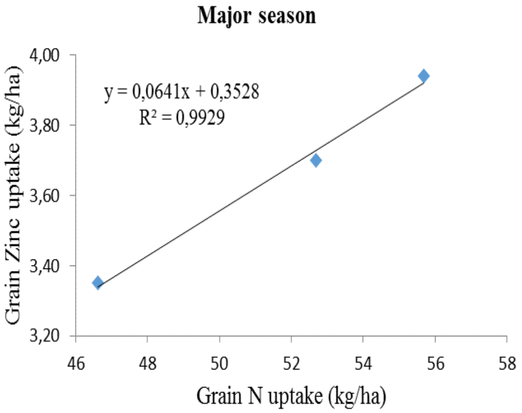
Table 6. NPK content of cowpea as affected by Zinc fertilizer

	Nutrient uptakes (kg ha^{-1})			
	Major season		Minor season	
	Haulm	Grain	Haulm	Grain
Rates		**N**		
0	40.32	46.63^{b}	28.30^{b}	58.39^{b}
5	41.46	55.69^{a}	34.30^{a}	63.19^{a}
10	45.99	52.68^{a}	33.20^{ab}	63.80^{a}
LSD (0.05)	NS	5.26	5.59	2.29
CV (%)	8.90	8.20	9.40	2.40
		P		
0	2.94^{b}	4.70^{c}	3.22^{b}	12.01^{b}
5	2.89^{b}	6.09^{b}	3.88^{ab}	13.61^{a}
10	3.54^{a}	5.44^{a}	4.01^{a}	13.83^{a}
LSD (0.05)	0.55	0.70	0.65	1.27
CV (%)	9.20	13.50	9.60	7.20
		K		
0	12.45	12.11^{b}	12.42	17.84^{b}
5	13.38	13.81^{a}	14.38	20.27^{a}
10	12.45	13.93^{a}	12.45	20.08^{a}
LSD (0.05)	NS	1.26	NS	1.67
CV (%)	14.30	8.30	17.30	8.10

7. Interrelationship between Zinc and NPK uptake in plant grain

The linear regression showed the positive relationship between grain Zn uptake and NPK content for the sampling period during the experiment in the major and minor seasons (Fig. 3). The argument on the enhanced NPK uptake by Zn content was ably supported by the significant positive relationship observed in the present study between NK and Zn uptake (0.9929*** with N and 0.9096** with K) in the major cropping season. The minor cropping season also follows the same trend with 0.9942** and 0.9389** with N and K respectively.

And with P the relationship was weak but positive (0.3839 in major season and 0.7289 in minor season).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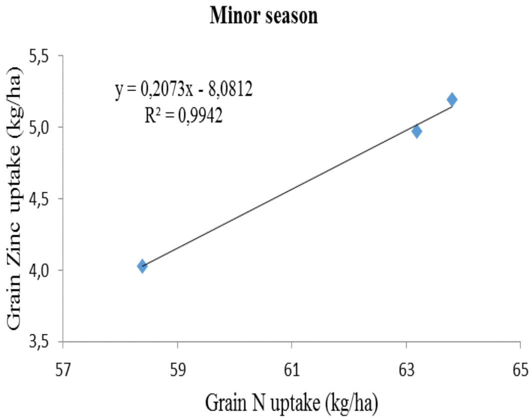
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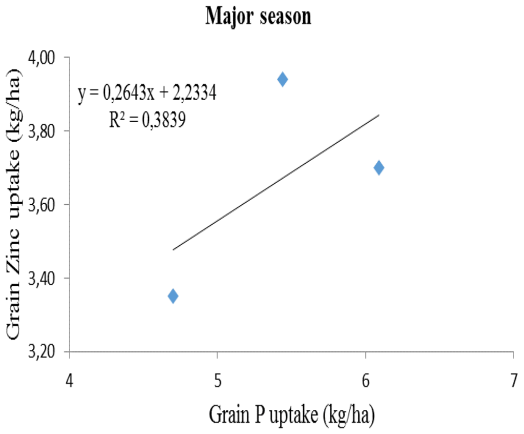
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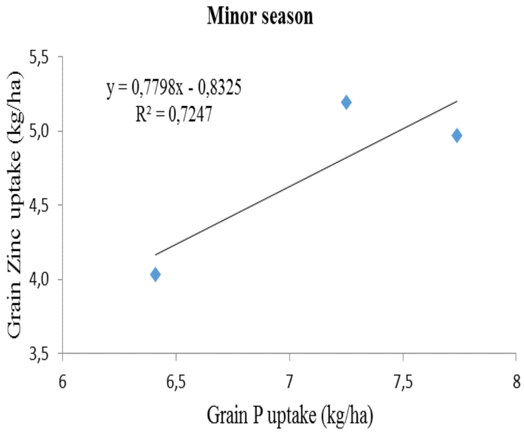
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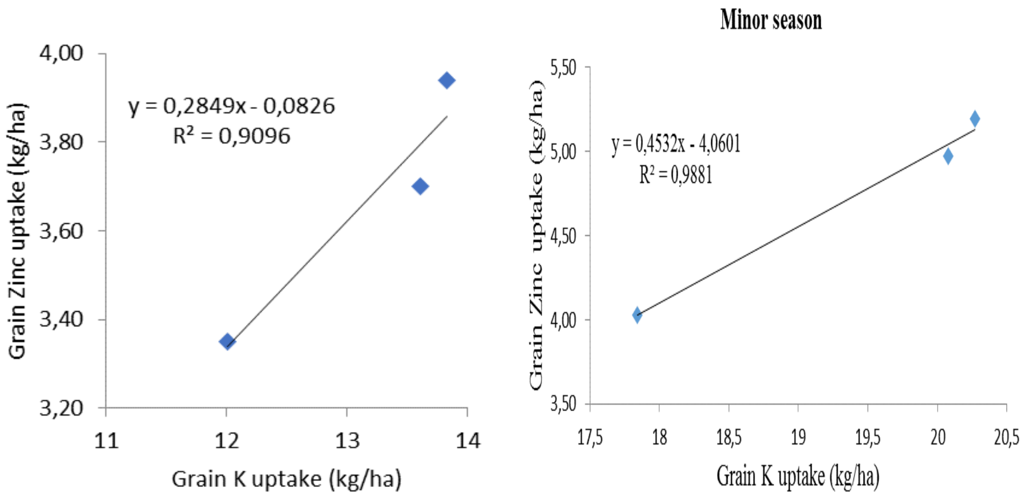

Figure 3. Relationship between Zn uptake and the macronutrients (NPK) in 2016 cropping seasons

DISCUSSION

Plant height was affected by both variety and Zn rates. Among the Zn treatments, Plant height was greatest in the Zn plots applied in both cropping seasons. Malakooti *et al.* [20] reported that added Zn significantly increased plant height by increasing internodes distances. Kaya *et al.* [21] stated that grain yield was positively correlated with leaf weight, stem weight, plant height and number of branching per plant. Zinc fertilizer application did not, however, have any significant effect on the number of leaves and stem girth in all the days examined. It was reported that application of zinc had positive effects on growth parameters [22]. Contrarily, foliar application of micronutrients increased the diameter of plant over the control treatment [23]. So, these findings conclude that the entire cowpea varieties gave equal stem diameter at all treatments of zinc application.

The application of the Zn fertilizer did not affect nodulation, indicating that some of inoculation factors were limiting such as soil pH, initial phosphorus and others micronutrients. And also, Gourion *et al.* [24] reported that the nodule initiation may depend on the relative concentrations of plant-specific signals and host species appears to be a significant factor determining the maximum number of nodules generated. Effective nodule is essential for a functioning Legumes-Rhizobium symbiosis and Zinc, chloride and cobalt have no effect on nodulation but are required for the growth of the host legume [25]. Two hosts may have the same sensitivity to bacterial signal molecules, but might differ in their ability to elicit synthesis of required nodulation signals in the bacteria [24, 26]. Cowpea root exudates have also been reported to contain substances that enhance nodule initiation [27, 28, 29]. However, lower efficiency of cowpea cannot be readily explained in terms of reduced numbers of bacteria in contact with the root [24]. Varieties most susceptible to infection and capable of producing effective nodules should have greater potential to fix more atmospheric N. However, this assumption often depends on other factors such as the environment and crop management [30]. Indeed Giller [31] reported that the ability to form nodules is not enough to obtain an effective nitrogen fixation symbiosis. Nodule number was nearly successively decreased over time at all treatments and is not correlated with the Zinc fertilizer applied. The amount of nodule dry biomass was drastically reduced with the mineral Zinc fertilizer, whereas the amount of nodule biomass was not affected in the control group, probably because the soil had satisfactory levels of available N and P. Nodule number correlated negatively with nodule dry weight [32]. The interaction effect was also not significant at 5% probability.

The present results were supported by Arif *et al.* [33] who reported that foliar application of micronutrients help in improving yield. In the two sampling seasons, foliar spray of Zn fertilizer had effect on hundred grain weights. In all these parameters, the control treatment effect was lower than Zn treatments, whereas among the Zn treatments. Pandey *et al.* [34]

reported that following Zn fertilization increased hundred seed weight. Also, Zeidan *et al.* [35] reported that yield and its components in lentil are improved by foliar application of micronutrients. Crop yields and quality are reduced by Zn inadequate in soil; therefore, Zn utilization is essential to obtain high yield and quality in crops as showed the results (Table 3). These results are in close conformity with those of Sharma and Jat [36], Yadav [37] and Tripathi *et al.* [38]. This was because of the fact that better and higher availability of Zinc, resulting better nutritional environment, higher dry matter accumulation and its associated effect on growth attributes increased haulm and grain yield. It is also evident from table 3 that all the Zinc treated plots increased the grain yield over the control, as there was a consistent increase in cowpea grain yield up to 10 kg Zn ha^{-1}. This suggests that, the application of Zn significantly affect cowpea yield. Similar results were reported as in Moswatsi [39] and Oseni [40] studies. In this connection, Banks [41] reported that the foliar application of Zn affected yield and its components of soybean. Also, Seifi *et al.* [42] reported that the highest yield of common bean was obtained by Zinc foliar application. Abdoli *et al.* [43] reported that more production of chlorophyll and IAA can cause delay in plant oldness and prolong the period of photosynthesis. This incident improves the production of carbohydrates and their transportation to the growing seeds.

The Zn deficiency symptoms can be prevented by the application of Zn fertilizers. The actual causal relationship and mechanisms are still not fully understood [3]. As shown in Tables 5 and 6 the mean percentage total nitrogen, phosphorus and potassium uptakes in the harvested leaves were quantitatively higher under zinc fertilizer application and increased with incremental zinc rates. These results corroborate the findings of Fagaria [44] and Sunitha et *al.* [45] who reported that zinc is an essential micronutrient for plant growth and plays an important role in the catalytic part of several enzymes its deficiency will result in stunted growth and nutrient uptakes. And also, Potarzycki and Grzebisz [46] reported that zinc exerts a great influence on basic plant life processes, such as (i) nitrogen metabolism – uptake of nitrogen and protein quality; (ii) photosynthesis - chlorophyll synthesis and carbon anhydrase activity. Also, many researchers have observed that Zn is closely related to the nitrogen metabolism pathway of plants, thus causing a reduction in protein synthesis for Zn deficient plants. Zinc deficiency significantly affects the root system including root development [47].

CONCLUSION

Zn fertilizer significantly affected NPK content and grain yield of cowpea varieties used. The increment of Zn content in the grain had a positive relationship with NK, which will definitely enhance nutrition of both human and animals. At all sampling periods, nodule number per plant was not affected by Zinc rates and nodule number was nearly successively decreased over time at all treatments and is not correlated with the Zinc fertilizer applied. The Zinc fertilizer significantly enhanced N_2-Fixed and Crude protein in both cropping season's trial investigating effect of Zinc rates on growth, nodulation and mineral content of cowpea in the semi-deciduous forest zone of Ghana. This implies the 5 kg Zn ha^{-1} is the optimum rate that will enhance the yield and nutrient quality of cowpea in the Semi-Deciduous Forest Zone of Ghana.

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