Effect of organic manure on growth, yield and nitrates content of *spinach oleracea* in Gashua Yobe State, Nigeria

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Abstract- This study investigated the effect of cow dung manure application rates on spinach (Spinacia oleracea) growth, yield, and nitrate content. Conducted during the 2019/2020 dry season at two irrigated locations in Nigeria (CIDS-FUGA and YOSADP farms), the experiment used a Randomized Complete Block Design (RCBD) with seven treatments: five cow dung rates (0, 1.0, 1.5, 2.5, 3.0 t/ha), a control (no manure), and a check (100 kg/ha NPK 15:15:15). Data on growth (plant height, leaf count, stem diameter, leaf area) and yield (fresh/dry weight per plot/hectare) were analyzed using SAS and DMRT. Results showed no significant effect of cow dung on growth or yield due to low soil fertility. However, nitrate content increased with higher manure rates, peaking at 3.0 t/ha (lowest at 1.0 t/ha, still above control). A positive correlation linked yield to growth parameters, but a negative correlation existed between yield and nitrate (higher yield = lower nitrate). The study concluded that while cow dung did not significantly improve spinach growth or yield, it enhanced nitrate accumulation, with 3.0 t/ha being optimal for nitrate content.

Indexed Terms- Spinach oleracea, Cow dung, Nitrate, Nitrite and Gashua

I. INTRODUCTION

Spinach is an important vegetable crop consumed for its nutritional, medicinal and health benefits. It also serves as source of income for small holder farmers, particularly the rural women who produce and sell the crop. Nitrogen fertilizers have been identified as one of the major factors that influence optimum crop yield in vegetables. It has been reported that Green leafy vegetables such as spinach, generally contain higher levels of nitrate than other foods. Therefore, excessive application of N may result in higher nitrate concentration in spinach. Nitric oxide (NO) is oxidized in the body naturally to form nitrite (NO_2^-) and nitrate (NO_3^-) [1]. The nitrate taken in through diet is typically found in functional foods such as dark leafy vegetables for like lettuce, broccoli, cabbage, spinach amaranths and fruits like strawberries. About 85% of dietary nitrate comes from vegetables, while the remaining amount can be obtained from drinking water.

Vegetables in diets are functional foods because they aid in lowering blood pressure due to the dietary nitrates that are found in them, which have a beneficial effect on the circulatory system by decreasing blood pressure [2]. Nitrate can function as an antioxidant when it is converted to nitric oxide [3]. This quality allows for the possibility to be beneficial and be able to prevent diseases and promote efficiency in the regional blood flow in patients with sickle cell [4]. Conversely, it has also been established that excessive intake of nitrate due to increasing consumption of vegetable, especially leafy crops may increase the risks of nitrate and nitrosamine exposure. Therefore, it is important to increase crop yield using N without leading to excessive accumulation of nitrate beyond the 2500 ppm. Maximum limit for nitrate in fresh vegetables. The amount of nitrate found in vegetables depends on its genotype, and a variety of other conditions such as soil and growth, conditions. Season, fertilizer, the type of cooking procedure, and crop types [5, 6].

The study was carried out to determine the effect of different concentration of cow dung manure application on the growth, yield and nitrate content of spinach with a view to increase its productivity, thereby improving the health and nutritional benefit of smallholder farmers in northern Nigeria.

II. MATERIALS AND METHOD

The experiment was conducted at two locations (Centre for Irrigation and Desertification Studies {CIDS} Research Farm of Federal University, Gashua {FUGA} and Yobe State Agricultural Development Project {YOSADP} Farm, Gashua) during the 2020 dry season under irrigated conditions. The treatments consist of five levels of cow dung manure (*0.t/ha, 1.0t/ha, 1.5t/ha, 2.0t/ha,2.5t/ha, 3.0t/ha of cow dung manure and the recommended NPK (100kg/ha)* for spinach in the study area and the check, which is non application of any type of fertilizer. This was factorial combination and arranged in a Randomized Complete Block Design (RCBD) and replicated three (3) times. There were 7 treatments in each replication and 21 treatments in the trial.
Soil samples were taken randomly at 0 – 30 cm soil depths from the experimental sites before establishing the trial. A tubular auger was used to take the samples. The composite sample was analyzed in the laboratory to determine their physical and chemical properties. Samples of cow dung manure that was used for the experiment was analyzed in the laboratory to determine their chemical properties. The samples were mixed thoroughly in a container after which a representative sample was scooped out from the bulk and analyzed to determine the initial soil fertility levels. The pH was measured in water (1:2.5 soil: water) and in 0.01M $Cacl_2$ using a cyber scan 20 pH meter. The soil organic carbon was determined through the wet oxidation method [7]. The total nitrogen was determined by micro kjeldahl digestion distillation method [8]. While available phosphorous was determined by Bray 1 method [9]. The exchangeable cations (Ca, Mg, K and Na) and the cation exchange capacity (CEC) were determined using standard procedures as described by [10].

The land was ploughed, harrowed before planting, and marked into plots of 3 m by 2 m (6 m^2) from which sunken beds was made manually to accommodate for the manure. The blocks were separated by an alley of 1 m and the plots were demarcated with high bunds of 20 cm to minimize lateral movement of fertilizer from one plot to another, in addition to the conservation of water within the plot. The cow dung manure was applied as per treatments at 2 weeks before planting which was uniformly spread on the beds and lightly worked into soils with hoe. The land was harrowed into a fine tilt and beds of 3 x 2m^2 (6m^2) were made where the seeds were sown by broadcasting and lightly covered with soils. The seeds were planted at a spacing of 20cm x20cm. The beds were mulched with straw grasses. The mulch was removed after seed emergence to facilitate seedlings establishment. The cow dung manure was applied at planting. It was uniformly spread on the ridge and lightly worked into soils with hoe. Samples of cow dung manure that was used for the experiment was analyzed in the laboratory to determine their chemical and physical properties. No Content of the vegetable at harvest was analyzed using standard procedure to determine the effect of the different treatments on the vegetable.

Plant height was measured from ground level of the tip of height growing point at 3, 4, and 5 weeks after sowing (WAS) using metre rule. Later the average height was determined. Plant stem diameter of the two plants was measured using vernier caliper at 3, 4, 5 WAS, the average was calculated and recorded. Number of leaves per plant was determined by counting the total number of leaves on each tagged plant at 3, 4 and 5 WAS, and later the average was taken. Leaf area was determined using a Ll-3100c leaf area meter at 3, 4 and 5 WAS and the average was determined. Fresh vegetables were harvested at 6 WAS and weighted. The weight determined was later expressed in tons/ha. The samples were weighted using Metter weighing balance. Fresh weight per plant was obtained by up-rooting two plants per plot at 6 WAS, the roots were washed to remove the soil, samples were later put into a small, marked envelope labeled with the plot number and oven dried at temperature of 70º to a constant weighed. Harvest was done at physiological maturity, all plants in the net plot was harvested to determine yield, while plants outside the net plot will be used for analysis.

Weeds were controlled by hand pulling as at when necessary. Harvest was made at 6 weeks after planting, all plants in the net plot were harvested to determine yield and NO content. Nitrate Content of the spinach at harvest was analyzed using the standard procedure to determine the effect of the different treatments on the crop. Data collected was subjected to Analysis of Variance (ANOVA) using general linear model GLM of the Statistical Analysis System package and the means were separated using Duncan's Multiple Range Test. All statistical procedures were be done using SAS.

III. RESULTS

The physical and chemical property of the soil of the experimental site during the 2019 dry season is shown in Table 1. The textural class of the soil was loam. The pH in water was 6.53 the organic carbon and total nitrogen were 14.60 and 1.21 respectively while the available phosphorus was 3.42. The exchangeable cation values for Ca and K were 3.61 (molkg^{-1}) and0.22 (molkg^{-1}) respectively. The concentration of H+AL was 0.21 (cmol kg^{-1}) and the CEC was 4.87(cmol kg^{-1}).

Table 1: Properties of the soil of the experimental sites in 2020/2021 dry seasons at Gashua

Soil Parameters	Location I	Location II
Particle size distribution (g kg^{-1})		
Clay	16	12
Silt	28	34
Sand	56	54
Textural class	sandy loam	sandy loam
Chemical properties		
pH in water 1: 2.5	5.28	6.83
pH in $Cacl_2$ 1: 2.5	4.78	5.99
EC (dsm^{-1})	0.014	0.05
Organic Carbon (g kg^{-1})	1.344	0.58
Total N (g kg^{-1})	0.21	0.35
Available P (mg kg^{-1})	9.81	13.95
Exchangeable Cations		
K (cmol kg^{-1})	1.71	0.73
Mg (cmol kg^{-1})	0.92	0.92
Ca (cmol kg^{-1})	8.85	7.70
Na (cmol kg^{-1})	0.87	0.77
H + Al (cmol kg^{-1})	1.20	0.80
ECEC (cmol kg^{-1})	13.55	10.92

The chemical composition of cow dung manure used for the study is shown in Table 2 and it was observed that nitrogen content was 13.11 (gkg^{-1}) followed by phosphorus 3.96 (gkg^{-1}) then potassium 1.17gkg^{-1}.

Table 2: Analysis of the cow dung manure used.

Total nitrogen (g kg^{-1})	13.11
Available Phosphorous (g kg^{-1})	3.96
Potassium (g kg^{-1})	1.17

Application of 3.0t/ha of cow dung manure produced the highest content of nitrate, this was followed by plots that received 2.5t/ha, 2.0t/ha and NPK 100kg/ha, and then 1.5t/ha and 1.0t/ha of cow dung manure while the control plot produced the least content of nitrate as shown in Table 3.

Table 3: Effect of cow dung manure on nitrate content of spinach during the 2020 dry seasons at Gashua

	CIDS	YOSADP
Treatment	Nitrate	
0.1t/ha	8.45c	7.12d
1.0t/ha	10.44b	9.23c
1.5t/ha	10.44b	10.27c
2.0t/ha	11.54b	13.08b
2.5t/ha	12.06b	13.78b
3.0t/ha	13.99 a	15.67a
NPK 100kg/ha	12.06b	15.45a
SE±	0.432	0.544

The response of plant height of spinach in relation to application of organic fertilizers is shown in table 4. The result revealed that there was no significant difference in plant height at 3, 4 and 5 weeks after sowing.

The response of stem diameter of spinach in relation to application of organic fertilizer is shown in Table 4. The result shows that there was no significant difference in stem diameter at 3, 4 and 5 weeks after sowing.

Table 4: Effect of Organic Fertilizer on Plant height and diameter of Spinach at 5WAS during the 2020 dry Season in Gashua

Treatment	Plant Height (cm)		Plant Diameter (cm)	
	CIDS	YOSADP	CIDS	YOSADP
0t/ha	8.92	7.32	0.22	0.22
1.0t/ha	8.33	6.57	0.23	0.28
1.5t/ha	10.8	9.05	0.25	0.28
2.0t/ha	11.95	9.78	0.23	0.33
2.5t/ha	10.67	9.23	0.25	0.30
3.0t/ha	12.95	11.00	0.30	0.35
NPK100kg/ha	10.18	8.58	0.17	0.20

SE±	0.698	0.668	0.088	0.103

The response of number of leaves of spinach in relation to application of organic fertilizer is shown in Table 5. The result shows that there was no significant difference in the number of leaves at 3, 4, and 5 weeks after sowing. The effect of organic fertilizer on Leaf Area of Spinach is shown in Table 5. The result shows that there was no significant difference in Leaf Area at 3, 4 and 5 weeks after sowing.

Table 5. Effect of organic fertilizer on number of leaves and leaf area of spinach during the 2020 dry season at Gashua

Treatment	Number of leaves per plant		Leaf area (cm^2)	
	CID	YOSAD	CID	YOSAD
0.1t/ha	8.17	9.83	15.0	15.33
1.0t/ha	9.50	11.67	15.4	15.90
1.5t/ha	9.83	11.50	15.3	15.93
2.0t/ha	8.67	11.33	19.7	21.53
2.5t/ha	9.50	11.67	9.70	9.57
3.0t/ha	10.1	12.17	14.3	15.03
NPK100kg/ha	7.83	9.17	10.5	12.17
SE±	0.50	0.504	1.00	1.049

The effect of organic fertilizer on fresh weight of spinach is shown in table 6. The result shows that there was no significant difference in fresh weight at 3, 4, and 5 weeks after sowing. The effect of organic fertilizer on dry weight of spinach during the 2020 dry season at Gashua shown in Table 6. The result shows there was no significant difference in dry weight at 3, 4 and 5 weeks after sowing.

Table 6: Effect of Organic Fertilizer on Fresh Weight of Spinach during 2020 Dry Season in Gashua

Treatment	Fresh Weight (g)		Dry Weight (g)	
	CIDS	YOSADP	CIDS	YOSADP
0.1t/ha	0.57	0.80	0.30	0.47
1.0t/ha	0.80	1.07	0.50	0.67
1.5t/ha	1.07	1.23	0.63	0.70
2.0t/ha	1.57	1.80	0.97	1.23
2.5t/ha	0.80	0.83	0.47	0.50
3.0t/ha	1.47	1.67	1.07	1.00
NPK100kg/ha	0.90	0.97	0.57	0.53
SE±	0.319	0.335	0.275	0.306

The effect of organic fertilizer on yield of fresh weight per plant of spinach during the 2020 dry season at Gashua is shown in Table 7.

Table 7: Effect of Organic Fertilizer on Yield of Fresh Weight Per Plot During the 2020 Dry Season at Gashua.

Treatment	CIDS	YOSADP
0.1t/ha	2.20	1.20
1.0t/ha	3.47	1.63
1.5t/ha	2.97	1.50
2.0t/ha	6.03	2.70
2.5t/ha	3.23	0.80
3.0t/ha	4.63	1.77
NPK 100kg/ha	3.10	0.80
SE±	0.607	0.411

IV. DISCUSSION

The result of the soil analysis shows that the soil textural class is loam. The soil is slightly acidic, very low in organic carbon, total nitrogen, available phosphorus and potassium as described by [11]. The calcium, magnesium, CEC and Sodium content is medium while Aluminum is low as described by [11].

A. Effects of Organic Fertilizer on Growth and Yield of Spinach

As observed from the result, there was no significant difference on growth and yield of spinach throughout the period of experimental. This was because the soil was low in total nitrogen, organic carbon, phosphorus and potassium. Among inorganic fertilizer, NPK fertilizers are designed to give plants the three major nutrients: nitrogen (N), phosphorus (P) and Potassium (K) that the plants need in appropriate Proportion and amount. [12, 13]. Many workers have tried to access the importance of organic manures in crop production. [14] reported that the use of organic manures improves all the growth parameters of the leaf vegetable they worked with. Poultry, cow and pig dung are the basic sources organic manure [15]. There is the need to comparatively study the effect of different levels of NPK and animal dung on the growth performance of common vegetables in order to quantify their applications in the production of such vegetable in Nigeria [16].

This study investigated the effects of different levels of inorganic fertilizer and cow dung manure on the growth and yield of Indian Spinach (Basella alba). There were significant differences in the numbers of leaves between control each of the plant with the other treatment. There was no significant difference in the number of leaves among treatment PM 5t/ha, PM 2t/ha. The number of leaves in treatment NPK 200kg/ha (6.5g 1pot) was significantly higher than that of every other treatment. The control had the lowest shoot length followed by NPK 100 kg/ha and PM 3t/ha. NPK 200kg/ha has the highest significant difference in shoot length compared to very other treatment. But this is contrary, because of the low availably in soil; it could not support growth and yield. The application of manure was not enough to satisfy the fertility requirement.

B. Effect of Organic Fertilizer on Nitrate Content of Spinach

As observed from the result, there was significant difference on nitrate content increased with increasing application of cow dung manure analysis. Manure is both a natural by-product of livestock production and an excellent source of plant nutrients. The use of manure as a fertilizer for crop production is a very beneficial way to recycle manure nutrients within an agricultural system. Manure not only acts as a source of plant nutrients but through the addition of organic matter, it also helps to improve soil tilt, structure, aeration and water holding capacity. Manure application rates should be based on crop nutrient requirements or removal. Nitrate (NO_3) constitutes the most important form of nitrogen, when nitrate uptake for exceeds assimilation by the plant, accumulation of nitrate in the plant tissues can occur. For this reason, leafy vegetable, (i.e spinach, lettuce, celery, rocket, Swiss chard) are considered as prominent nitrate accumulating species [17].

Nitrogen is an essential element for plant growth and development; however, due to environmental pollution, high nitrate concentrations accumulate in the edible parts of these leafy vegetables, particularly if excessive nitrogen fertilizer has been applied. Consuming these crops can harm human health, thus developing a suitable strategy for the agricultural application of nitrogen fertilizer is important [18].

Organic, inorganic and liquid fertilizers were utilized in this study to investigate their effect on nitrate concentration and lettuce growth. The results of the pot experiment shows that the total nitrogen concentration in soil and the nitrate concentration in lettuce increased as the amount of nitrogen fertilizer increased. If the recommended amount of inorganic fertilizer (200kg Nha^{-1}) is used as a standard of comparison, lettuce augmented with organic fertilizer (200kg Nha^{-1}) have significantly longer and wide leaves, higher shoot and lower concentration of nitrate.

Once nitrogen fertilizers are applied to agricultural systems, the fertilizers are absorbed directly by plants or converted into various other forms through the oxidation process. Excess nitrogen is lost in ionic or gaseous form through leaching, volatilization and de-nitrification. If nitrate is not absorbed by plant not, it is carried away by run off or leaches into the soil along with water. The Phyto-availability of the nitrogen fertilizers and environmental problems such as eutrophication, the greenhouse effect, and acid rain.

Consuming contaminated groundwater or crops with a high concentration of nitrate has negative effects on human health. In a study by Donner and Kucharik, when the application rate of nitrogen. Fertilizer was increased by 30% on the corn yield increased 4% but the amount of nitrate loss through leaching increased by 53%. Although the yield decreased by 10% when the application rate of nitrogen fertilizer was reduced by 30%, the leaching loss was 37% less [19]. Applying Manure (150kg –N-ha^{-1} $year^{-1}$) raises the yield of phylum pretense L. champ; however, the excess nitrogen that accumulates in the soil if double the amount of manure is applied may result in a decrease in yield.

According to the results of previous studies, the accumulation of nitrate in the edible parts of crops is directly related to the type of nitrogen fertilizer used as well as the soil properties. Regarding lettuce, the light intensity, timing of fertilizer N release and lettuce type have been shown to affect the accumulation of nitrate in this crop.

A pot experiment was conducted using various types of nitrogen fertilizers, application rates and combinations of rates and fertilizers. The objective of the present study is to develop an agricultural system that produces healthy lettuce that has low nitrate content. Green leafy vegetable contains the highest nitrate levels and lettuce is classified as having very high nitrate content. Because consuming high levels of nitrate may further lead to severe pathologies in humans, cultivating edible crops with low nitrate content very important. The joint expert committee of the food and agriculture (JECFA) Organization of the United Nations World Health Organization and the European Commission (EC) Scientific Committee on Food have also set an acceptable daily intake for nitrate of 0-3.7mg.kg^{-1} body weight. The U.S Environmental Protection Agency (EPA) reference does for nitrate is equivalent to about 7.0mg.kg^{-1} body weight per day. Previous studies indicate that vegetables are the major source of nitrate intake by humans and constitute approximately 40%-92% of the average daily intake. Assuming lettuce grown in OM200 and OM200 + LF was the only vegetable consumed, a person eating 400g of vegetables, as recommended by the World Health Organization, would have dietary exposure to approximately 1,670-1,746mg of nitrate. Therefore, consuming a balanced diet of vegetables could be an effective method for decreasing the amount of nitrate exposure in humans.

CONCLUSION

Based on the result of the trial, there was no significant differences between all cow dung manure rates on growth and yield of spinach production. However, nitrate content increased up to application of 3t/ha of cow dung manure.

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