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Growth, Yield and Microbial Contamination of Lettuce with Organic and Inorganic Fertilizer Treatments under Pipe-borne Water Irrigation Systems in Ghana

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ABSTRACT

The effect of organic (T_1- poultry manure at 2kg/m^2, T_2 - cattle manure at 2kg/m^2, T_3 - sheep manure at 2kg/m^2) and inorganic fertilizer (T_4 -DI grow green at 0.00024 l/m^2 and T_5 - NPK (15:15:15) and sulphate of ammonia at 25.2 g/m^2) on growth, yield and microbial contamination of lettuce was elucidated in Ghana using Randomized Complete Block Design (RCBD) with three replications. The results showed that application of organic and inorganic fertilizer had not significant effect on plant establishment, number of leaves, plant height, chlorophyll accumulation leaf area, fresh weight and dry matter of the shoots and the roots (P<0.05). Poultry manure recorded the highest mean leaf counts (12.47 and 13.87 in 26 days and 33 days but was overtaken by DI grow green with 28.73 mean leaf counts in 40 days), respectively. The total coliform numbers on lettuce grown with poultry, cattle and sheep manure, D1 Grow Green, NPK/Sulphate of Ammonia and control or zero fertilization were [(7.0 x 10^5), (9.0 x 10^{17}), (1.0 x 10^{17}), (1.0 x 10^{18}), (1.0 x 10^{17}) and (2.0 x10^{18})], respectively. Feacal coliform numbers in the same sequence treatments were low, [(3.11 x 10^5), (2.18 x 10^6), (9.36 x 10^6), (4.57 x 10^6), (1.39 x 10^6) and (7.84 x 10^6)], respectively. Similarly, *Enterococci* numbers on lettuce were lower, (6.52 x 10^2), (90), (57), (1.03 x10^2), (58) and (1.23 10^2) in the same treatment order. *Salmonella* numbers on lettuce were much lower [(1.0 x 10^4), (2.66 x 10^4), (1.33 x 10^4), (4.0 x 10^4), (3.0 x 10^4) and (3.33 x 10^5)] in the order of treatments for the total coliforms. *Escherichia coli* numbers on lettuce were also lower than the total and faecal coliforms [(3.05 x 10^6), (2.25 x 10^5), (1.42 x 10^6), (2.92 x 10^5), (3.05 x 10^4) and (2.91 x 10^5)] in the same treatment order. Generally, guano yielded the highest bacteria count on the lettuce far exceeding the recommendation of the World Health organization (WHO) and International Commission on Microbial Specifications for Foods (ICMSF) standard of 103. High dependency on the guano to produce lettuce may be a contributing factor to contamination and a health threat to lettuce consumers. Intensification of innovative education on the production, processing and consumption of lettuce in and around the College of Agriculture Campus of the University of Education, Winneba at Asante Mampong is thus, a good food hygiene and safety improvement strategy.

Key words: Organic and inorganic fertilizers, Lettuce, Yield and Microbial contamination.

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INTRODUCTION

Lettuce (*Lactuca sativa*) is a temperate annual or biennial in plant daisy family *Astereceae*. It is often grown as a leafy vegetable and usually eaten raw, especially in salads, sandwiches, hamburgers, tacos and many other dishes or cooked Chinese cuisine in which the stem is as important as the leaf. It is one of the earliest crops to be domesticated by man and has been in cultivation since ancient times (Tweneboah, 1997). Lettuce probably was

first cultivated by the ancient Egyptians who turned it from weed, whose seeds were used to produce oil, into a plant grown for its leaves. Lettuce spread to Greeks and Romans the latter of whom gave it the name, lactuca from which the English “lettuce” ultimately derived (Addo-Quaye et al., 1993). Lettuce is an important vegetable for human consumption, mainly due to the minerals and vitamins they contain and are essential to the diet. It provides biologically active substances that prevent some diseases whose incidence worldwide has been increasing (Sinnudurai, 1992). The demand for lettuce is in the increase due to the springing up of restaurants, hotels, supermarkets and wayside fast food joints in Ghana (Sinnadurai, 1992). Although demand for lettuce is high, production constraints including high cost of Agro-chemicals used in lettuce production have scared many people about the consumption of lettuce since the crop is usually eaten in its raw state. Moreover about 1700 cases of typhoid was reported by the hospitals in Ashanti, Brong Ahafo and Central regions of Ghana in 2012 (Kwabena et al., 2014). The avoidable alarming rate of typhoid and other associated bacterial related diseases in Ashanti-Mampong Municipality and beyond is rather unfortunate (Kwabena et al., 2014). Given the increased demand for ready-to-eat foods, particularly those containing uncooked fresh vegetables, there is great concern regarding the safety of these foods in the presence of unhygienic and improper management of the raw produce. Successful intervention strategies are therefore reliant on identifying not only the practices that are important for consumer protection, but also barriers that prevent consumers from responding to these interventions from the market chain (Fischer et al., 2006). Purposively, the study compared the effect of organic fertilizer (chicken, cattle, sheep manures, and D’ grow green made from Dynapharm Company Limited) and composite inorganic fertilizer (NPK 15:15:15 and sulphate of ammonia) on the growth, yield and microbial contamination levels of lettuce by specifically determining local indicative performance effects of the treatments on: vegetative growth, yield, and quality (microbial contamination level) of the lettuce leaves.

The rationale behind this study was to discover the best fertilizer application method for lettuce farmers in Mampong-Ashanti and beyond. The results could improve the knowledge of farmers on the agronomic practices to adopt for higher yield and safer ways of production to meet the targets of increasing number of lettuce consumers in the Municipality. Scientists proof that chemical fertilizers are usually salts of various elements and do nothing to feed the soil. Thus, a long term application of such fertilizers depletes soils and disrupts biosynthesis; even, the mostly used fertilizers in these fields are not broad spectrum most supplying 3 to 4 nutrients, the basis being nitrogen, phosphorus and potassium (Dick et al., 1993). Lettuce requires more than these four (4) elements. Major nutrient deficiencies may negatively affect the growth and yield of lettuce. Chemical fertilizers applied to crops often leach into the soil and carried by rain to rivers (Tiimub et al., 2012). Organic fertilizers are broad spectrum containing at least 20 nutrients; they add organic matter to the soil and feed the life that lives within the soil (Vimala, 2000). They actually feed and build the soil which in turn means the plants growing in that soil will be healthier and more able to fend off pest and diseases (Dick et al., 1993). Organic fertilizer from various sources contains both major and trace elements improve soil texture and ability to hold moisture and nutrients (Agyeman, 2005). There is an urgent need to determine the most suitable type of fertilizers that will not only increase yield but support the production of safe and nutritious lettuce. It was expected that the experiment would identify the most suitable fertilizer for increasing yield and quality of lettuce and safer ways of processing it since the crop is usually eaten raw and introduce innovations to increase lettuce production to meet huge nutritional demands for growing population farmers in at large around Mampong Municipality. Students studying agriculture at the university could utilize the information for lettuce production research and practical work.

MATERIALS AND METHODS

Site Description

The project was carried out at the University of Education Winneba research farm at Mampong Campus in Ashanti region of Ghana from May to October, 2012. Mampong is between latitude 07^{o} and 08^{o}North of equator and longitude 01^{o} and 02^{o} West of equator and has an altitude 490.2 m (Mampong Meteological Station, 2007). The major rain season usually starts from March and ends in July whiles the minor season starts from September and ends in November. It is between 800 and 1500 mm it is bimodal and fairly distributed. The mean daily temperatures and monthly rainfall are 30^{o}C and 91.2 mm, respectively (Mampong meteorological station, 2007). Mampong and its surrounding environment experience Relative humidity as high as 90% in June and as low as 55% in February (Mofa.gov.com District Ash. 01/04/2013). The vegetation of the area is semi-decidous type with thick grass such as; *Pennisetum purpureum*, *Panicum maximum*, *Cyperus spp* and *Chromolaena odorata*, with the *Cyperus spp*. The municipality is part of the Savannah transitional zone of Ghana, characterized by savannah woodland with patches of dry forest and grassland to the south; 80% of land area is used for small scale farming. There are 4 forest reserves covering the total area of 111.59km^{2}. These are Abrimsu, Aboma, Offin Hardwater and Onwnani reserves (Mofa.gov.com District Ash. 01/04/2013). The experimental soil belongs to Savannah Ochrosol class and the Bediesi Series which is well drained, friable, red in colour and permeable and has moderate organic matter content with good water holding

capacity. The soil was derived from voltarian sandstone and is classified as the chromic Luvisol in the FAO-UNESCO system (Asiamah, 1998). The soil is good for cultivation of vegetables, including lettuce sweet pepper and cash crops such as cocoa, cowpea and maize.

Field Preparation and Planting

An area of 14 m long and 8 m wide was measured and marked out. The total field area was 112m^2. Lining and pegging were done and plots were prepared into beds. Each bed measured 2 m long and 1.2 m wide given an area of 2.4 m^2 with distance of 0.5 m created as pathways between the beds. There were three (3) block and each had six (6) plot (bed). Lettuce seeds (Great lake brand) were purchased from reputable Agro-chemical shop (Kyeiwaa Agro-chemical shop) in Mampong-Ashanti planted on nursing beds at dimensions of 2 m long, 1 m wide and about 0.25 m high. The seeds were sown in drills and slightly covered with soil treated with carbofuran at the rate of 40 g per plot. The beds were shaded with palm frond and watered. The palm fronds were used to raise shade over the seedlings to avoid direct sunlight and direct rain. Three days after seedlings emergence, regular watering was done with watering can twine daily in the mornings and evenings at 12 h intervals.

Experimental Design and Crop Establishment

The RCBD was used in assigning the five treatments (T_1-T_5) which were replicated three (3) times and alongside with the control (T_6) detailed as follows: T_1 - poultry manure at 2 kg/m^2, T_2 -Cattle manure at 2 kg/m^2, T_3 -Sheep manure at 2kg/m^2, T_4 -DI grow green at 0.00024 l/m^2, T_5 - NPK (15:15:15) and sulphate of ammonia at 25.2 g/m^2, respectively at different times and T_6 -Control (no fertilizer or manure). The treatments were assigned to the plots. The poultry cattle and sheep manure and N.P.K and were obtained from the University's farm management section. D_1 grow green was bought from a branch of Dynapharm company in Kumasi-Ghana. Organic manure apart from D_1 grow green were applied a week before the transplanting. Transplanting was done when the seedlings were 29 days old at spacing of 30 × 25 cm, with 21 seedlings per bed. A total of three hundred and seventy eight (378) seedlings were transplanted on the experimental field. The seedlings were transplanted at four weeks old. Dead transplanted seedlings were replaced as a result of death. When the plants were 19 days old crop establishment count took place. Results of the actual plant population from establishment beds were as follows: T_1 – 61, T_2 – 63, T_3 – 62, T_4 – 62, T_5 – 58 and T_6 – 50.

Cultural Practices and Harvesting Of Lettuce

Manual weed control was done using cutlass and hoe. Weeding was done alongside stirring to aerate the beds. Immediately after transplanting the first watering was done. In ensuring adequate soil moisture for plants, watering was done every day except the rainy days. Watering was done using tap irrigation scheme. Butterfly species of Hemoptera (order) were later found on the lettuce. The pupae stage of the butterfly produced some foam like fluid on inner leaves. The insect were destroyed physically without using chemicals. The final harvesting and yield determination took place when the crops were 44 days old. It was done by uprooting and weighing on a scale.

Data Collection on Plant Growth Effects

Five selected plants per each treatment bed were tagged after labeling each treatment when the crops were 11 days old after transplanting.

Plant Height

The heights of five plants per treatment were taken every week. The measurement was done from the ground level of the stem to the tip of the highest spinal bud using a 30 cm rule. The mean height of all experimental plants on each plot was determined. The numbers of leaves were counted alongside the height of the tagged plants and recorded.

Chlorophyll Level

When the plants were 32 days old, chlorophyll level in mmol/L/g was tested with chlorophyll meter at the crop physiological maturity. The tagged plants were tested on lower, middle and upper leaves.

Dry Matter Determination

When the lettuce were 34 to 44 days old, samples were harvested, weighed and preserved in the oven for 48 h to determine the crop's dry matter at physiological maturity and harvesting stage, respectively.

Leaf Area Index

Leaf Area Index (LAI) = leaf area / ground area of the plants was computed using graph sheet as done for an area of an irregular object.

Yield Parameters

Mature plants were harvested when they were 44 days old after transplanting. The yield was determined by weighing the plants per treatment bed.

Data Analysis

The statistical analysis of the raw data was run with SASS

Table 1. Effects of Inorganic and organic fertilizer on the vegetative parameters of lettuce.

Descriptive Variable	Mean Height of Crops under Establishment 19 days after planting (cm)	Chloropyll (mmol/L/g)	Leaf Area (m^2 /m^2)	Number of Leaves/ Days After Transplanting (Dap)			
				19	26	33	40
10 tonnes/ha of poultry manure (T_1)	20.33	756	350	8.67	12.47	17.33	24.40
10 tonnes/ha of cattle manure (T_2)	21.00	506	234	7.00	10.00	14.80	20.27
10 tonnes/ha of sheep manure (T_3)	20.67	624	410	8.93	11.67	17.07	27.07
1 litre/ha of D1 Grow (T_4)	20.67	580	314	7.53	11.47	15.87	28.73
104.2kg/ha of NPK/SOA (T_5)	19.33	607	281	7.00	10.00	13.87	28.53
Control T6	16.67	444	311	7.27	10.47	14.27	24.27
L. S. D	2.433	138.1	100.7	0.78	0.88	1.47	3.82
CV (%)	6.8	12.9	17.5	5.6	4.4	5.2	8.2

(Version 18.0 Pack) using two way analysis of variance (ANOVAS) to test the least significant differences between the separate treatment means.

Microbial Analysis of Total, Faecal and Thermo-tolerant Coliforms

The Most Probable Number method was used to determine total and faecal coloforms in the samples. Serial dilutions of 10^{-1} to10^{-4} were prepared by picking 1ml of the sample into 1g and sterile distilled water. One milliliter aliquots from each of the dilutions were inoculated into 5 ml of MacConckey Broth incubated at 35°C for total coliforms and 44°C for faecal coliforms within a maximum duration of 18 to 24 h. Tubes showing colour change from purple to yellow and gas collected in the Durham tubes after 24 h were identified as positive for both total and faecal coliforms. Counts per 100 ml were calculated from the Most Probable Number (MPN) tables.

E. coli

From each of the positive tubes identified a drop was transferred into a 5 ml test tube of buffered tryptophan water and incubated at 44°C for 24 h. A drop of Kovacs' reagent was then added to the tube of tryptophan water. All tubes showing a red ring colour development after gentle agitation denoted the presence of indole and recorded as presumptive for thermo tolerant coliforms (*E.coli*). Counts 100 $^{-}$ ml were calculated from the MPN tables.

Faecal *enteroccoci*

Serial dilutions of 10^{-1} to 10^{-2} were prepared by picking 1ml of the sample into 9 ml sterile distilled water. One millilitre aliquots from each of the dilutions were inoculated on a Slanetz, and Bartley Agar prepared on sterile Petri dishes. The Petri dishes were pre-incubated at a temperature of 37°C for 4 h to aid bacterial resuscitation. The plates are then incubated at 44°C for a further 44 h. After incubation all red, maroon and pink colonies that were smooth and convex were counted and recorded as faecal *enteroccoci*.

Salmonella

Prepared 10 ml of manufactured formula of Buffered peptone water (BPW) was in a universal bottle and serial dilution of samples added to it. It is incubated at 37°C for 24 h. Then 0.1 ml of the sample from the BPW is placed in a 10 ml of selenite broth in universal bottle and incubated at 44°C for 48 h. Swaps from the bottle onto SS agar and incubated at 48hrs at 37**°**C. C. Black colonies on the SS agar indicate the presence of *salmonella*.

Total Viable Counts

About 9 ml of distilled water was sterilized at a temperature of 121^{o}C for 15 min and weighed; 10 gm of lettuce and manure samples were added and pulsified for 15 sec. Serial dilutions of 10^{-1} to 10^{-4} were prepared by dilution of 1ml of the sample. One millilitre aliquots from each of the dilutions were inoculated into petri dishes were already prepared P.C.A plate count Agar. The plates were then incubated at 35^{o}C for 24 h. After the incubation all white spots or spread were counted and recorded at the total viable count using the colony counter

RESULTS

Effect of Organic and Inorganic Fertilizers on Lettuce Performance

The results from Table 1 of crop establishment indicate that, cattle manure (T_2) recorded the highest height with the mean value of 21.00 cm, 19 days after transplanting. There was no significant difference between T_2, T_3 and T_5. However, T_6 recorded the lowest crop establishment with the mean value of 16.67 cm, 19 days after transplanting. The data on level of chlorophyll pigmentation with the

Table 2. Effects of organic and inorganic fertilizers on yield parameters of lettuce.

Descriptive Variable	Fresh Weight (mg/kg)		Dry Matter (mg/kg)		Total Harvest
Days After Transplanting (DAP)	34	44	34	44	44
10 tonnes/ha of poultry manure (T_1)	126.7	367	9.33	18.7	57.1
10 tonnes/ha of cattle manure (T_2)	66.7	180	7.00	10.0	37.7
10 tonnes/ha of sheep manure (T_3)	226.7	349	7.00	23.3	57.3
1 litre/ha of D1 Grow (T_4)	126.7	460	8.33	21.0	50.7
104.2kg/ha of NPK/SOA (T_5)	156.7	333	10.00	16.3	48.9
control T_6	126.7	373	7.00	20.3	41.9
L. S. D	73.00	279.2	2.86	7.91	13.19
CV (%)	29.00	44.6	19.2	23.8	14.9

various treatments revealed that, poultry manure (T_1) recorded the highest value of 756.3 mmol/L/g. There was significant difference between the treatments with the exception of T_4 and T_2. However, T_6 (controlled) recorded the lowest mean value of 443.9 mmol/L/g. The sheep manure (T_3) recorded the highest mean leaf area index value of 410.4m^2/m^2. There was significant difference between the various treatments except T_4 and T_6. T_2 (cattle manure) recorded the lowest mean value of 234.0m^2/m^2. Looking at T_6 (control) with mean value of 310.8m^2/m^2 recording the fourth highest. This may be due to the fact that the soil was already fertile for vigorous plant growth. Although the highest leave number (8.93) was obtained from the application of sheep manure, there was no significant difference between T3 and T1 (Table 1). The lowest number of leaves recorded was7.00 for both T_2 and T_5 treatments in 19 days after transplanting, respectively. However, there was significant difference between T_1 and T_4.

The leaf counts checks after 26 days of vigorous plant growth showed that poultry manure (T_1) recorded the highest mean value of 12.47, whereas T_2 and T_5 both recorded the lowest mean value of 10.00. Apart from T_2, T_6 and T_5 there was significant differences in lead counts between the rests of the treatments. The number of leaves in 33 days after planting with the poultry manure (T_1) recorded the highest mean value of 17.33 whilst NPK/SOA (T_5) recorded the lowest mean value of 13.87. There was no significant difference between T_6 and T_5 but there was significant difference among the rest of the treatments. The number of leaves in 40 days after planting with DI grow green (T_4) recorded the highest mean value of 28.73. Cattle manure (T_2) recorded the lowest mean value of 20.07. There were no significant differences between T_4 and T_5 and between T_3 and T_1.

Fresh Matter Determination of Successive Days after Plating of Lettuce

The data from Table 2 on the fresh matter determination of the lettuce samples after 34 days reveal that, sheep manure (T_3) treatment produced the highest mean value of 226.7 mg/kg whiles cattle manure (T_2) produced the lowest (66.7 mg/kg). There was significant difference between T_3 and T_5. There was no significant difference among T_4, T_1 and T_6. The data analysis from Table 2 of fresh matter determination after 44 days after transplanting indicates that, DI grow green (T_4) recorded the highest mean value of 460.0mg/kg and T_2 recorded the lowest mean value of 66.7mg/kg. There was no significant difference among T_1, T_6, T_3 and T_5.

Dry Matter Determination on Successive Days after Plating

The highest mean value of 10.00 mg/kg dry matter weight of the lettuce in 34 days after was recorded from NPK/SOA (T_5) treatment. The least mean value of 7.00 was recorded from T_3 and T_2 treatments, respectively. There was no significant difference among the mean dry matter weights of T_1, T_4 and T_6. The dry matter weights in 44 days after transplanting with sheep manure (T_3) treatment was the highest (23.33 mg/kg) whereas cattle manure (T_2) yielded the lowest mean value of 10.00 mg/kg apart from T_5 and T_2, and there was no significant difference among the rest of the treatments.

Variations in Plant Heights in Successive Days After Transplanting

The data on Figure1 shows that poultry manure (T_1) treatment recorded the highest mean value (14.11 cm) 19 days after transplanting whiles cattle manure (T_2) recorded the lowest mean value (11.4 cm) 19 days after transplanting. There was significant difference between T_1 and T_6, T_1 and T_5 and T_1 and T_2. Also, there was significant difference among the T_3, T_4, T_5 and T_6 mean heights. The results further indicates that poultry manure (T_1) recorded the highest mean value of 20.73 cm growth height within 26 days after transplanting whiles cattle manure (T_2) recorded the lowest mean height of 16.36 cm. There was significant difference among the T_1, T_5, T_2, T_4 and T_6 treatment heights and among the T_3 and T_6, T_5 and T_2, respectively. The (Figure 1) data sets indicate that in 33 days after transplanting with poultry manure (T_1) treatment the highest mean plant height recorded was 25.33 cm whiles the control (T_6) treatment gave the least response of 19.70 cm. There was significant difference among the

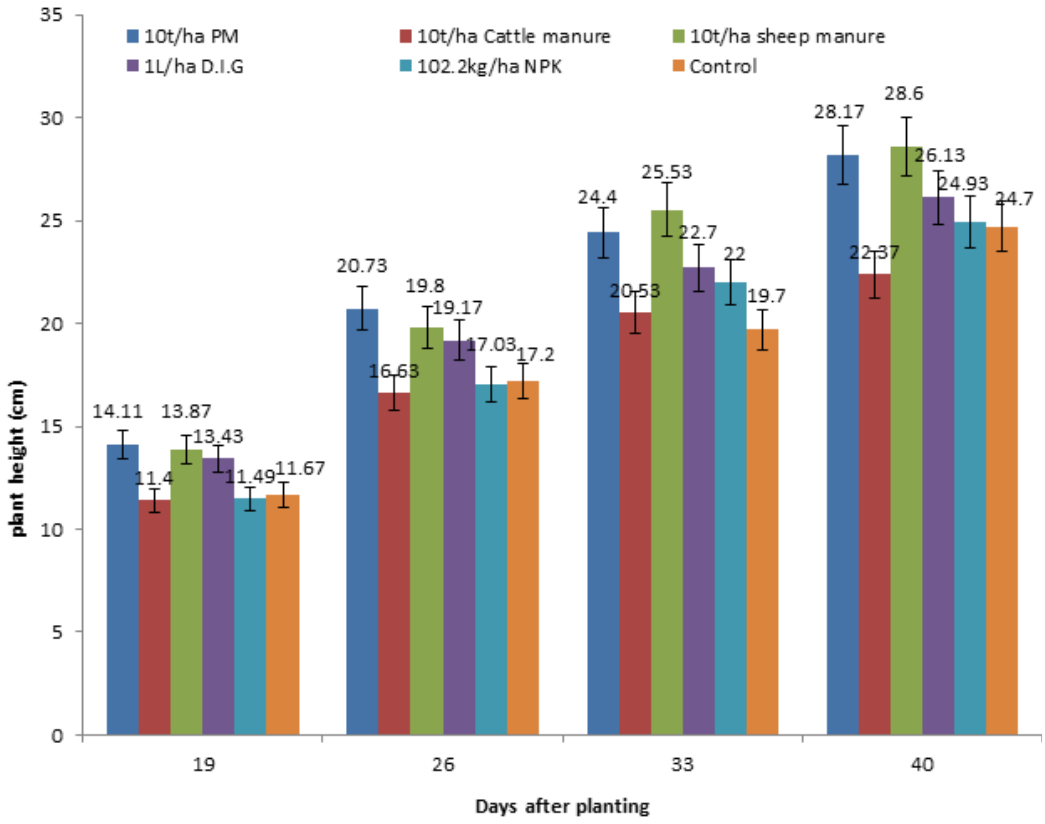

Figure 1. Effect of different soil amendments on the height of lettuce.

T_3 and T_5, T_3 and T_6, T_3 and T_4 heights, respectively. Significant differences among the T_1 and T_2, T_1 and T_6, T_4 and T_6 treatments growth heights of lettuce were further observed. The results of plant growth heights (figure1) with the sheep manure recorded the highest observed mean of 28.60 cm, followed by poultry manure treatment (28.17 cm). Cattle manure (T_2) recorded the least mean height of 22.37 cm. There were significant differences between the T_3 and T_5, T_3 and T_2 treatment responses. Also, there was significant difference in mean lettuce heights between T_1 and T_6, T_1 and T_2, T_1 and T_5; T_4 and T_2 treatments.

Effect of Organic and Inorganic Fertilizers on Lettuce Contamination with Bacteria

The results from Table 3 for estimated number of spotted *E coli* on the lettuce growths shows that, poultry manure (T_1) recorded the highest mean value of 3. 05 x 10^6 whilst NPK (15:15:15) and sulphate of ammonia (T_5) treatment recorded the lowest mean value of 3.05 x 10^4. D1 Grow green (T_4) recorded the highest mean value of 4.0 x 10^4 *Salmonella* contamination whilst poultry manure recorded the lowest mean value of 1.0 x 10^4 without significant difference among the treatments in Table 3. The results from Table 3 show the level of faecal *enteroccocus* contamination on the lettuce. The poultry manure (T_1) treatment recorded the highest *enterococcus* number 6.52 x 10^2 whiles sheep manure (T_3) recorded the lowest mean value of 56.7x10^1. Generally there was significant difference among the treatments. The control (T_6) treatment recorded the highest mean value (2.0 x 10^{18}) of total coliform whilst T_5 recorded the lowest (1.0 x 10^{17}). There was significant difference between T_3 and T_5 treatments.

The results from Table 3 on feacal coliform counts show that there was no significant difference among T_4, T_1, T_2 and T_3. However, there was significant difference between T_6 and T_5 and T_6 and T_3. Also, T6 recorded the highest mean value of 7. 84 x 10^6 of feacal coliform whiles T_3 recorded the least value of 3.96 x 10^5. The data sets on Table 3 on total bacterial count shows that, T_3 recorded the highest mean value of 1.72 x 10^5 whiles T_5 recorded the lowest mean value of 5.78 x 10^4. The results from Table 3 on total viable counts show that T_2 recorded the highest mean value of 9.24 x 10^2. T_5 recorded the least value of 67.6 and there was no significant difference among the various treatments.

E. coli in the Manure

The results from Table 4 shows that *E. coli* numbers dominated poultry manure treatment 9.0 x 10^{15} sources but

Table 3. Bacterial contamination on lettuce (*E coli*, Salmonella, feacal *Enteroccoci,* feacal coliform, total coliform, total count and total viable count).

Type Of Soil Treatment	*E. coli*	*Salmonella* 100 ml	Feacal *Enteroccoci*	Feacal coliform 100 ml	Total coliform 100 ml	Total count	Total viable count
10 tonnes/ha of poultry manure (T_1)	3.05×10^6	1.0×10^4	6.52×10^2	3.11×10^5	7.0×10^5	1.47×10^5	7.40×10^2
10 tonnes/ha of cattle manure (T_2)	2.25×10^5	2.66×10^4	90	2.18×10^6	9.0×10^{17}	1.55×10^5	9.24×10^2
10 tonnes/ha of sheep manure (T_3)	1.42×10^6	1.33×10^4	57	9.36×10^6	1.0×10^{17}	1.72×10^5	1.22×10^2
1 litre/ha of D1 Grow Green (T_4)	2.92×10^5	4.0×10^4	1.03×10^2	4.57×10^6	1.0×10^{18}	1.59×10^5	1.23×10^2
104.2kg/ha of NPK/SOA (T_5)	3.05×10^4	3.0×10^4	58	1.39×10^6	1.0×10^{17}	5.78×10^4	68
control T_6	2.91×10^5	3.33×10^5	$1.23\ 10^2$	7.84×10^6	2.0×10^{18}	1.22×10^5	1.57×10^2
L. S. D	$4.44\ 10^{-6}$	5.26×10^{-4}	7.48×10^{-2}	5.49×10^{-6}	1.41×10^{-8}	1.76×10^{-5}	1.21×10^{-3}
CV (%)	275.8	113.3	227.8	92.2	95.9	71.7	186.9

was lower in cattle manure and sheep manure (2.0 x 1017). There was significant difference among the treatments means.

Feacal *Enteroccoci in Manure*

The results from Table 4 shows that faecal *enterococci* was higher in the poultry manure (1.80×10^4) but lower in the sheep manure (1.0×10^2) although the differences between the means were not significant.

Salmonella in the Manure

Salmonella was mainly encountered from poultry manure and sheep manure treatments sources *(*4.0×10^{14}*and (*4.2×10^{15}), respectively with no significant difference in mean contamination levels from these sources (Table 4).

Feacal Coliform Contamination Level of the Manure

The faecal coliform numbers were higher in the cattle manure (9.0×10^{15}) than in sheep manure (4.8×10^{16}) and poultry manure (2.0×10^{17}). There were significant differences among the treatments (Table 4).

DISCUSSION

Crop Establishment

The result in Table1 reveals that, the beds treated with fertilizers had the highest crop establishment as compared to the control. The probable reason may be that, the crops needed fertilizer for good establishment (Crew and Peoples, 2004). Since those plots were treated with fertilizer the crop establishment was better than the control plot.

Test for Chlorophyll

The treatments had effect on the chlorophyll level of lettuce. All the treatments performed better than the control. Looking at the performance, there was no significant differences among T_1, T_3 and T_5. These three treatments probably released enough nitrogen and other valuable nutrients for the plants than the cattle manure and the DI grow green. This observation may be consistent with the earlier remarks of Dick et al. (1993) that organic fertilizers are broad spectrum containing at least 20 nutrients; they add organic matter to the soil and feed the life that lives within the soil. They actually feed and build the soil which in turn means the plants growing in that soil will be healthier and more able to fend off pest and diseases (Organic fertilizer from various sources contains both major and trace elements improve soil texture and ability to hold moisture and nutrients (Addo - Quaye et al., 2003). This is probably because the cattle manure takes a longer time to decompose due to its lumpy nature. The high percentage of chlorophyll indicates the presence of nitrogen in the plant.

Leaf Area Index

The results in Table1 indicated that treatments with organic manure (T_1, T_3) performed better than the rest. The poor performance of the cattle manure may be due to its inability to decompose fast enough. The increase in area of leaf by poultry manure (T_1) might be as a results of addition of high population of beneficial soil microbes that play a role in N-mineralization and non-symbiotic fixation as earlier on confirmed by Ademiyan et al. (2005).

Number of Leaves, Plant Height, Marketable Yield and Mean Leaf Dry Mass

The results showed that the type of treatment applied significantly affected the number of leaves, plant height marketable yield and dry mass ($P<0.05$). Poultry manure (T_1) recorded the highest mean value 19 to 40 days after transplanting followed by sheep manure (T_3) and the

Table 4. Bacterial load in the manure.

Type of soil treatment	*E. coli*	Feacal *enteroccoci*	*Salmonella*	Total coliform	Feacal coliform
10 tonnes/ha of poultry manure (T_1)	9.0×10^{15}	1.80×10^{4}	4.0×10^{14}	2.4×10^{14}	2.0×10^{17}
10 tonnes/ha of cattle manure (T_2)	2.0×10^{17}	1.16×10^{4}	4.2×10^{15}	9.2×10^{20}	9.0×10^{15}
10 tonnes/ha of sheep manure (T_3)	2.0×10^{17}	1.0×10^{2}	0.0×10^{10}	9.2×10^{17}	4.8×10^{16}
L. S. D	1.85×10^{-5}	0.000	0.000	0.000	1.67×10^{-5}
CV (%)	0.5	0.0	0.0	0.0	0.4

control (T_6) which recorded the lowest mean value. Generally treatments with organic manure perform better than inorganic manure apart from cattle manure. Crew and Peoples (2004) reported that the type of fertilizer applied significantly affected the growth, yield and nutritional values of lettuce. Trends in superiority of different types of organic fertilizer were observed as the chicken manure observed exhibited relatively higher values of the number of leaves, plant height and dry mass. The low mean value recorded by cattle manure may be as a result of low decomposition. This may be due to inorganic fertilizer showing less-broad spectrum, supplying about 3 to 4 essential nutrients while organic manure supplies about 20 nutrients out of which 17 are essential to the growth of plants as reported by Sanders (2001).

Bacterial Contamination Levels of Lettuce (*E. coli, Salmonella*, Feacal *Enteroccoci*)

The microbial analysis proved that the organic manure (poultry, sheep and cattle) shows significant contamination levels of bacteria such as *E. coli*, which recorded a mean value of 3.0×10^5 in poultry manure (T_1) and the lowest mean value of 3.05×10^6 in NPK/SOA (T_5). In the test for salmonella on lettuce, poultry manure recorded the lowest mean value of 1.0×10^4 whiles DI Grow green recorded the highest mean value. The analysis on feacal enteroccoci shows poultry recorded the highest mean value 6.52×10^2 whiles sheep manure recorded the lowest mean value of 56.7×10^1.When laboratory test was conducted on the samples of organic manure (poultry, sheep and cattle) the results indicated the presence of high concentration of *E. coli*, *Salmonella* and feacal *Enterococci*. This goes to confirm the high concentration of these bacteria on the leaves of lettuce due to the splashing of water during watering (Grubben et al., 2004). Furthermore, the treatment of T_1, T_2 and T_3 generally recorded the highest mean values. Though, control (T_6) bed was not treated with any fertilizer it recorded a substantial concentration of the bacterial (*E. coli*, salmonella, and feacal enteroccoci). This may be due to the use of the contaminated soil for previous experiments. Bacterial numbers recorded in this study were still above the International commission on microbial specification for foods. (ICMSF) (1998) limit of 10^3 to 10^5 coliforms $100g^{-1}$ weight of vegetables usually eaten raw when compared with previous research findings of Tiimub et al. in the Kumasi Metropolitan Area in 2011 where by, three major lettuce production centres were monitored for its bacterial contamination levels in Ghana.

CONCLUSION

The results reveal that, generally the organic manure performed better than the inorganic fertilizer in the production of lettuce. The results further specified that a treatment with inorganic fertilizer is less contaminated with bacteria as compared to the organic manure. It is clearly established from this study that bacteria could transfer from the organic manure to the lettuce through splashing of water during watering. This confirms the recent comparative analysis of organic produce versus conventional produce from university of Minnesota as far back as 2001 which reported that organically grown produce have 9.7% positive samples of the presence or generic *E. coli* bacteria versus only 1.6% for the conventional produce on farms. However, the bacteria numbers in all the treatment exceeded both the World Health Organization (WHO, 1989) and (ICMSF, 1998) recommended levels, making the lettuce risky for consumption in the raw state without proper processing. Analysis of the results obtained from the data indicates clearly that although the various soil enrichment treatments exhibit different levels of positive effects on yield of lettuce, the organic manure proved to be outstanding from this study.

RECOMMENDATIONS

The research has yielded empirical evidence through the field measurements and experimental trials that organic manure is the best soil amendment remedy to be recommended for adoption by farmers in and around Mampong-Ashanti for cultivating lettuce and could work effectively for other leafy vegetables. Furthermore, organic manure should be converted into compost due to the lumpy nature of cattle manure making them to delay in decomposition and this affects the growth, yield and quality of lettuce. Policy makers, Agriculture Extension Officers, NGO's and other stakeholders in the agriculture sector should advise farmers on the proper way of applying organic manure to the soil since it improves biological

activity and soil structure. It is also recommended that the experiment be repeated on infertile soils to ascertain clearly the effects of organic and inorganic fertilizers on the growth and yield of lettuce. Moreover, public education on the consumption of lettuce should be mounted in market and health centres. The general public should be made aware to use vinegar or salt solution to wash any lettuce before consumption.

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