PRELIMINARY INVESTIGATION INTO SOME ASPECTS OF THE ECOLOGY OF COASTAL SAVANNAH FOREST SOILS IN GHANA: A Case Study of the University of Cape Coast Nature Reserve

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ABSTRACT
The study was conducted in the University of Cape Coast Nature Reserve to investigate the levels of urease in the soils and to relate these levels to soil organic matter (SOM) content, soil moisture (SM) content, pH, temperature, particle size distribution and bulk density. The stratified random sampling method was employed to collect data within three 10 × 200 m belt transects. Soil samples were collected from 0 to 15 cm depth in different zones of the nature reserve between January and April, 2005. The mean soil urease levels ranged from 80.91 ± 4.72 to 132.36 ± 10.80 NH_4^+-N mg kg^{-1}. Monthly variations in soil urease levels were highly significant ($p < 0.01$). The enzyme level varied significantly ($p < 0.001$) with topography. Multiple regression analysis showed that urease activity depended on SOM, SM, pH and soil temperature ($p < 0.05$). Soil texture and bulk density were similar in all the zones. SOM, SM, time and topography were the main factors which affected urease levels in the soils of the University of Cape Coast Nature Reserve.

INTRODUCTION
Forest soils are influenced by biotic and abiotic factors. Soil development depends on factors such as soil enzymes, organic matter content, moisture, pH, temperature, particle size distribution and bulk density. All enzyme activities in soils including that of urease, derive ultimately from micro-organisms, plants or soil animals and these are essential for soil development (Baligar *et al.*, 1991; Tabatabai, 1994).

Urease is a very important enzyme in soils. It converts urea to carbon dioxide and ammonia which is a source of nitrogen for organisms. Urease activity relates to soil properties such as organic matter content, moisture content, pH, temperature, particle size distribution and bulk density. Soil organic matter plays a crucial role in the maintenance and improvement of soil properties. Its distribution is influenced by topographical gradient whereby it increases from the summit to the foot of slopes (Chen and Chiu, 2000). Significantly, it is affected by climatic factors, particularly, rainfall and temperature (Brady, 1990). Soil organic matter content is influenced by urease activity. It has been reported that there is strong relationship between urease activity and soil organic matter content (Zantua *et al.*, 1977; Tejeda *et al.*, 2006). Soil moisture content plays vital role in the overall condition of soils. Variations in soil

moisture are influenced by topography, which includes elevation, slope and aspect (Qiu *et al.*, 2001). It influences availability of nutrients to organisms, including plants. By virtue of its high specific heat capacity, soil water affects soil organisms directly. It also serves as the medium through which enzymatic reactions can occur. Such reactions are strongly influenced by the pH of the medium. The pH of soils derives from natural geological and biological processes. Soil pH affects many soil processes including decomposition and mineralization of organic matter (Vepsalainen and Niemi, 2002). Decomposition of litter and organic matter and their mineralization are affected by temperature.

Temperature affects the physical, chemical and biological activities necessary for soil formation and development. Whilst low and high temperatures facilitate weathering of rocks, ample temperatures are required for biological and biochemical processes which are mediated by enzymes. The soil skeleton, namely; clay, silt and sand, influences the physical properties of the soil (Ricklefs, 1996). These components, together with bulk density, influence porosity, aeration and moisture content of soils. Soil air and moisture content are not only important as far as plant growth is concerned, but also, in determining the numbers and kinds of soil organisms (Enger and Smith, 2006). Bulk density decreases with increasing organic matter content. For a fine-textured soil, bulk density ranges between 1.1 and 1.3 Mg m^{-3} and ranges between 1.4 and 1.8 Mg m^{-3} for coarse soils (Biswas and Mukherjee, 1995).

Although information on some ecological aspects of the University of Cape Coast Nature Reserve exists (Yeboah, 1993; Pappoe *et al.*, 2008) knowledge on the assay of its soil enzyme is still lacking. The study was conducted to investigate the levels of urease in the soils of the Nature Reserve, and to relate these levels to soil organic matter content, moisture content, pH, temperature, particle size distribution and bulk density. The results would not only add to knowledge for better understanding of the soil ecology of similar forest types but would also serve as a base line data for monitoring the dynamics of these parameters in the University of Cape Coast Nature Reserve.

MATERIALS AND METHODS

Study area

The University of Cape Coast Nature Reserve lies on latitude 5^{o} 06' N and longitude 1^{o} 16' W (Etrex-Legend-Garmin Global Positioning System). The area (approximately 17.6 hectares) stretches from flat to hilly grounds. The climate follows the normal pattern of the coastal region of the country. The monthly mean temperatures fluctuate between 28 ^{o}C and 32 ^{o}C. The highest temperatures are recorded in February and March whilst the minimum temperatures are recorded between June and August. The relative humidity ranges between 68 % and 90 %. The annual rainfall is between 750 mm and 1000 mm (M. Sackey, personal communication). The reserve is a thicket vegetation and forms part of the southern marginal forest zone of Ghana (Hall and Swaine, 1976). The physiognomy presents a few scattered emergent trees, lianas and herbaceous climbers, shrubs and a ground layer. The forest floor is sparsely covered with litter.

Fieldwork

Soil samples were collected from each of three (10 x 200 m) belt transects, namely; Hill Bottom, Middle Slope and Hill Top from January to April, 2005. The stratified random sampling method was employed. Ten sampling areas were randomly located every month within each transect. At each sampling area, a sampling point was selected where a soil sample was collected to a depth of 15 cm by means of soil auger, after the surface litter has been removed. The soil collected from each zone was composited, put in a labelled black polythene bag and secured tightly. The soil samples were taken to the laboratory for analysis. Within each sampling area, a 5 cm diameter metal cylinder with both ends opened was driven into the soil till the upper rim was 5 mm below the soil surface to a depth of 3 cm, after the surface soil

had been cleared of litter, for determination of bulk density.

Laboratory analyses

In the determination of urease, organic matter content, pH, and particle size distribution soil analyses were replicated three times, whilst ten replicates were made for moisture content, temperature and bulk density determination.

Determination of urease activity

Urease activity was determined using the ammonium-nitrogen released method (Tabatabai and Bremner, 1972). Five grams of composted soil were placed in 50 ml volumetric flask, 0.2 ml toluene and 9 ml tris-hydroxymethyl aminoethane (THAM) buffer was added and swirled for a few seconds to mix. 1 ml aliquot of 0.2 M urea solution was added and swirled for a few seconds. The flask was stoppered and placed in a temperature-controlled water bath at 37 ^{o}C. After 2 hours, the stopper was removed, 35 ml KCl-Ag_2SO_4 solution was added and the flask was swirled for a few seconds. The flask was allowed to stand for the contents to cool to room temperature. The contents were topped to 50 ml with KCl-Ag_2SO_4 solution. The flask with its contents was stoppered and inverted several times to mix. NH_4^+-N in the resulting solution was determined by pipetting 20 ml of the suspension into 100 ml distillation flask. The NH_4^+-N was released by steam distillation. 0.2 g magnesium oxide (MgO) was added to the soil suspension in the distillation flask. On boiling, NH_4 that evolved was condensed and collected in boric acid indicator for 4 minutes. The distillate was titrated with 0.01 M HCl till the colour changed from green to permanent light pink colour at the endpoint. A control was set up as described above, except that the 1 ml 0.2 M urea was added after addition of 35 ml KCl-Ag_2SO_4 solution.

Determination of organic matter

The method described by the International Institute of Tropical Agriculture, IITA (1985) was followed. 0.5 g of soil sample was placed into 500 ml conical flask. A pipetted volume of 10ml normal $K_2Cr_2O_7$ solution was introduced into the flask and swirled gently to disperse the soil. A 20 ml volume of concentrated sulphuric acid (H_2SO_4) was added and swirled gently to mix, then vigorously for a minute and set on the laboratory bench. The contents were diluted with 200 ml of distilled water after 30 minutes. A 10 ml volume of concentrated orthophosphate acid (H_3PO_4) and 0.2 g sodium fluoride (NaF) were added followed by 1 ml of diphenylamine indicator and titrated with 0.5 N $FeSO_4$ (ferrous sulphate) solution till the colour changed from blue to green at the end-point. A blank titration was carried out in an identical manner as described above. The percentage organic carbon content was calculated from the following formula and used to estimate the percentage organic matter:

$$\text{organic carbon}(\%) = \frac{(b-s)\times 0.5\text{N}\times 0.3}{w\times 77}\times 100 \qquad (1)$$

where,

b = blank titre,

s = sample titre,

$0.5N$ = normality of ferrous sulphate solution and

w = weight of soil sample

Organic matter (%)

= organic carbon (%) x 1.724 (2)

Determination of soil moisture content

Soil moisture content was determined by following the process described by Allen *et al.* (1974). Ten grams of soil from each zone were measured into 10 separate crucibles of predetermined weights. The replicates were placed in an oven and dried at 105 ^{o}C for 24 hours, allowed to cool and transferred to a desiccator for 30 minutes to dry further. The moisture content was calculated using the formula below.

$$\text{Moisture content (\%)} = \frac{f-d}{d}\times 100 \qquad (3)$$

where,

f = fresh weight of soil (g)

d = dry weight of soil (g)

Determination of soil pH

Soil pH was determined on a 1:2.5 w/v suspension in distilled water (Rowell, 1994). Ten grams of air-dried soil from each zone were weighed separately into 3 plastic bottles with screw caps. A 25 ml volume of distilled water was added to the soil sample. The bottles were shaken for 15 minutes by means of a shaker (1KA HS 501 digital). The soil suspension was stirred and allowed to settle. The electrode of a pH meter (JENWAY 3330 research pH meter) was inserted and the pH was recorded after 30 seconds.

Measurement of soil temperature

Soil temperature was measured by inserting a soil thermometer to a depth of about 10 cm (Curtis and Cottam, 1962) at each sampling point for 3 minutes to allow the thermometer to adjust to the soil temperature. The temperatures were taken at 10:00 hours (Greenwich Mean Time) at monthly intervals.

Determination of soil particle size distribution and soil texture

The bouyoucos hydrometer method described by Anderson and Ingram (1989) was employed. Fifty grams of soil from each zone were weighed into separate 500 ml heat resistant (105 ^{o}C) screw lid bottle calibrated at 250 ml. A 125 ml volume of distilled water was added and swirled. A 20 ml volume of 30 % hydrogen peroxide was added and swirled gently. When the reaction subsided, the bottle was heated in a temperature controlled (105 ^{o}C) water bath to complete the reaction. Amyl alcohol was added drop-wise to contain the bubbles. When the reaction had subsided, the bottle was removed from the water bath and allowed to cool, 2.0 g sodium hexametaphosphate was added and the solution topped to the 250 ml mark with distilled water. The suspension was shaken overnight on a mechanical shaker. The contents were quantitatively transferred to a 1000 ml cylinder and topped up to the mark with distilled water. A blank was prepared in another cylinder by dissolving 2.0 g sodium hexaphosphate in 1000 ml of distilled water. The two cylinders were placed in the same tank and allowed to equilibrate for 30 minutes. The contents of the sample cylinder were mixed vigorously with a plunger. A stopwatch was started at the moment the plunger was removed. The bouyoucos hydrometer readings at 40 seconds and 5 hours as well as the temperature of the tank were recorded. The soil particle sizes were calculated from the formulae below:

40 seconds correction = 2(40 seconds reading – 40 seconds blank + T)

5 hours correction = 2(5 hours reading – 5 hours blank + T)

where

T = temperature correction [For every ^{o}C above 20 ^{o}C, T = 0.3; for every ^{o}C below 20 ^{o}C, T = – 0.3]

Sand (%) = 100 – 40 seconds correction.

Silt (%) = 40 seconds correction – 5 hours correction.

Clay (%) = 5 hours correction.

Particle sizes were defined as follows:

Sand = 2 mm – 0.06 mm.

Silt = 0.06 mm – 0.002 mm.

Clay = $\leq$ 0.002 mm.

The soil texture classification chart of the Food and Agriculture Organization, FAO (Sutherland, 1997) was used to determine the texture of the soils.

Determination of bulk density

The method used was adapted from Anderson and Ingram (1989). The litter was removed from the spot and top 2 cm surface soil was removed from the sampling points in each zone. A sampling metal cylinder (with open ends) of known weight and volume was driven into the soil to about 5 mm below the surface. The soil around the cylinder was excavated and the cylinder was undercut. Excess soil was trimmed off from both ends of the cylinder. The soil in the cylinder was dried at 105 ^{o}C for 48 hours, allowed to cool and weighed. The bulk density was calculated from the formula:

$$\text{Bulk density (g cm}^{-3}) = \frac{a - b}{v} \qquad (4)$$

where

a = weight of soil before drying (g)
b = weight of soil after drying (g)
v = volume of soil (cm^3)

Data analysis

The 2-way analysis of variance was carried out by using the Minitab 13 Statistical Software to analyse the data. Multiple regression was used to model the relationship between urease levels and some selected soil properties.

RESULTS

Tables 1 to 7 show variations in urease activity, organic matter content, moisture content, pH, temperature, particle size distribution and bulk density of soil samples taken from various zones of the University of Cape Coast Nature Reserve. The variations in the range of mean urease levels with respect to topography were considerable (96.58 ± 8.21 NH_4^+–N mg kg^{-1} at Hill Top – 127.51± 12.05 NH_4^+–N mg kg^{-1} at Hill Bottom). The lowest mean monthly urease activity (80.91 ± 4.71 NH_4^+–N mg kg^{-1}) was observed in March whilst the highest (132.36 ± 10.80 NH_4^+ – N mg kg^{-1}) activity occurred in February (Table 1). There was a very significant difference ($p<0.001$) in urease activity with respect to topography. Also, the monthly variations in the enzyme levels were very significant ($p<0.001$).

Generally, variations in soil organic matter content followed a topographical gradient. Monthly variations amongst the zones were slight for Hill Top (3.04 – 4.00 %) and Hill Bottom (0.91 – 2.38 %). Considerable variations were observed at Middle Slope (1.08 – 3.54 %). The variations were significant ($p = 0.044$ in terms of topography and $p = 0.005$ in the case of month). The mean organic matter content was highest in March (3.31%). The least values (approximately 2 %) were recorded for January and February. Soil moisture content was generally low, mostly for Hill Top and Middle Slope. Variations in moisture content were similar for Hill Top and Hill Bottom. The mean values ranged from 5.82 ± 0.45 % at Middle Slope to 9.14 ± 1.14 % at Hill Bottom. The monthly variations in soil moisture content were not significant ($p > 0.05$) with mean values ranging from 6.70 ± 0.45 % in March to 8.31 ± 1.65 % in February.

Table 1: Variations in urease activity with respect to topography and month

Topography	Monthly level (NH_4^+ - N mg kg^{-1})				
	January	**February**	**March**	**April**	**Mean ± s.e.**
Hill Top	102.11	116.22	70.85	97.14	96.58 ± 8.21
Middle Slope	118.41	122.32	81.01	114.21	108.98 ± 8.20
Hill Bottom	131.6	158.54	90.87	129.03	127.51± 12.05
Mean ± s.e.	117.37 ± 6.97	132.36 ± 10.80	80.91 ± 4.72	113.46 ± 7.53	

Table 2: Variations in organic matter content with respect to topography and month

	Monthly level (%)				
Topography	**January**	**February**	**March**	**April**	**Mean ± s.e.**
Hill Top	3.04	3.50	4.00	3.51	3.51 ± 0.20
Middle Slope	1.08	1.38	3.54	2.51	2.13 ± 0.56
Hill Bottom	1.77	0.91	2.38	1.67	1.68 ± 0.30
Mean ± s.e.	1.96 ± 0.47	1.93 ± 0.65	3.31 ± 0.39	2.56 ± 0.43	

Table 3: Variations in soil moisture content with respect to topography and month

	Monthly level (%)				
Topography	**January**	**February**	**March**	**April**	Mean ± s.e
Hill Top	9.50	7.65	7.52	7.61	8.07 ± 0.41
Middle Slope	5.28	5.19	7.38	5.42	5.82 ± 0.45
Hill Bottom	10.05	12.08	5.79	8.64	9.14 ± 1.14
Mean	8.28 ± 1.23	8.31 ± 1.65	6.70 ± 0.45	7.22 ± 0.78	

Soil pH varied slightly amongst the various zones (Table 4). The mean values ranged from 4.57 ± 0.21 at Middle Slope to 6.93 ± 0.47 at Hill Top. The variations in pH against topography were not significant (p = 0.44). Also, the monthly variations in soil pH were not significant (p = 0.419). The monthly mean values ranged from 5.39 ± 0.73 in February to 6.01 ± 0.61 in April. Variations in soil temperature within the zones and month were not much, particularly for Hill Top and Hill Bottom. However, temperatures were highest at Hill Top. Similar variations in temperature were observed for Middle Slope and Hill Bottom. The mean temperatures ranged from 27.73 ± 0.07 ^{o}C at Hill Bottom to 31.59 ± 0.46 ^{o}C at Hill Top. Whilst the temperature variations were not significant (p = 0.114) amongst the zones, variations amongst the various months were highly significant (p = 0.001). The highest mean monthly temperature (30.42 ± 1.29) was observed in February. The soil temperature was similar for January, March and April.

Considerable variations existed in particle size distribution with respect to clay and sand. Analysis of soil particle size distribution revealed that clay ranged from 24 % at Hill Top to 33 % at Middle Slope, silt ranged from 14 % at Hill Bottom to 19 % at Hill Top whilst sand ranged from 49 % at Middle Slope to 62 % at Hill Bottom (Table 6). Bulk density did not vary much with respect to topography and month. The values were close to one (Table 7). Data analysis involving multiple regression, showed that urease activity was significantly dependent on soil organic matter content (p = 0.002, R^2 = 81.0%) and moisture content (p = 0.015, R^2 = 81.0%). The activities of the enzyme with respect to soil pH were not significant (p = 0.257). Similarly, urease activity did

Table 4: Variations in pH with respect to topography and month

	Monthly level				
Topography	**January**	**February**	**March**	**April**	**Mean ± s.e.**
Hill Top	7.93	5.38	7.04	7.36	6.93 ± 0.47
Middle Slope	4.82	3.84	4.86	4.76	4.57 ± 0.21
Hill Bottom	4.29	6.94	5.80	5.91	5.74 ± 0.47
Mean ± s.e.	5.68 ± 0.93	5.39 ± 0.73	5.90 ± 0.52	6.01 ± 0.61	

Table 5: Variations in soil temperature with respect to topography and month

	Monthly level (^{o}C)				
Topography	**January**	**February**	**March**	**April**	**Mean ± s.e.**
Hill Top	30.60	33.10	31.30	31.36	31.59 ± 0.46
Middle Slope	28.45	30.50	27.75	28.71	28.85 ± 0.51
Hill Bottom	27.55	27.65	27.80	27.92	27.73 ± 0.07
Mean ± s.e.	28.87 ± 0.74	30.42 ± 1.29	28.95 ± 0.96	29.33± 0.85	

not vary significantly with variations in soil temperature ($p = 0.076$). The regression model of urease activity and the relevant soil factors is presented thus as;

$$UREASE = 5.0 - 19.6OMC + 6.02MC - 3.95pH + 4.45T \quad [R - Sq\ (adj) = 81.0\ \%] \qquad (5)$$

Where:

OMC	=	soil organic matter content (%),
MC	=	soil moisture content (%),
pH	=	soil pH, and
T	=	soil temperature (^oC).

DISCUSSION

Activity of urease was generally high at Hill Bottom compared to Middle Slope and Hill Top. Variations in urease activity at the micro-site levels can be ascribed to variations in chemical properties of these sites (Dowon *et al.*, 1998). Gupta and Bhardwaj (1990) have demonstrated that urease activity in forest and natural soils tends to be higher than those of other types of soil. Results obtained from the University of Cape Coast Nature Reserve confirm this observation. The mean soil urease levels of the Nature Reserve ranged from 80.91 ± 4.72 to 132.36 ± 10.80 NH_4^+–N mg kg^{-1} as compared to the levels, 5.3 – 79.2 NH_4^+–N mg kg^{-1} that was reported by Monreal *et al.* (1999) for agricultural soils.

It has been observed that increases in soil organic matter content enhance soil enzyme activities (Havlin *et al.*, 1995; Zaman *et al.*, 1999). However, this is in contrast to the results of the present study whereby the highest urease activity was recorded at Hill Bottom where the lowest organic matter content was recorded. The large differences in organic matter with respect to the three zones may be attributed to variations in microclimate that influence organic matter build-up. The soils at Hill Top recorded the highest levels of organic matter. This is contrary to the observation made by Chen and Chiu (2000) that total organic carbon increases from summit to the foot of a slope. Hill Top is relatively flat so that movement of materials by runoff water could be slow and this may have accounted for the build-up of organic matter there. Also, the highest mean soil organic matter recorded in March could be ascribed to the lowest soil moisture content realized in that month. The converse is true for February where the lowest mean organic matter and highest mean moisture content were obtained.

On the issue of soil moisture content, the highest moisture contents were recorded at Hill Bottom in January and February. This may be attributed to the drainage of the area as rainwater moves from the top of the hill to the bottom.

Table 6: Variations in soil particle size distribution with respect to topography and month

	Soil particle content (%)			
Topography	**Sand**	**Silt**	**Clay**	**Textural class**
Hill Top	28	19	53	sandy clay loam
Middle Slope	33	18	49	sandy clay loam
Hill Bottom	24	14	62	sandy clay loam

Table 7: Variations in soil bulk density with respect to topography and month

	Monthly value (Mg m^{-3})				
Topography	**January**	**February**	**March**	**April**	**Mean ± s.e.**
Hill Top	0.98	0.98	0.98	0.97	0.98 ± 0.002
Middle Slope	1.07	1.09	1.09	1.07	1.08 ± 0.005
Hill Bottom	0.97	0.97	0.96	0.95	0.96 ± 0.004
Mean ± s.e.	1.01 ± 0.03	1.01 ± 0.03	1.01 ± 0.03	1.00 ± 0.03	

Qiu *et al.* (2001) have observed that soil moisture shows significant correlation with relative elevation, slope and aspect. All these factors might have accounted for the highest moisture content at Hill Bottom. Urease activity varied significantly ($p = 0.015$) with soil moisture content. Following a decline in activity of the enzyme from its peak in February to its lowest level in March, urease activity rose in April to a level slightly lower than what was recorded in January. Similar trends were observed in soil moisture content and soil temperature. This shows that urease activity was related to these two parameters as shown by the multiple regression model. It must however, be noted that the highest urease activity recorded in February could be attributed to the fact that it rained heavily the previous two days before the soil samples were taken. Mayor *et al.* (1994) have also observed that water plays a critical role in microbial enzyme activity and attributed decline in soil enzyme activity to reduction in water content. Soil moisture content and the nature of the mineral grains also impact on the pH of the soil.

The mean soil pH was generally low at Middle Slope in contrast to that obtained at Hill Top. This can be attributed to the highest sand content of the soils at Middle Slope. Also, the clay content was lowest in this zone. Clay domains offer large surface area for adsorption of basic elements. Therefore, the lower the clay content, the lower the amount of basic elements that can be fixed on the lattices. It appears the interaction of sand and clay more than the silt content may have accounted for the variations in pH among the three zones. Soil acidity, which is a function of the nature of the parent material and soil inorganic and organic compounds; particularly acids from decaying organic matter (Bickelhaupt, 1998), is influenced by temperature which is known to affect soil formation.

The high soil temperatures recorded at Hill Top in relation to the other zones may be ascribed to the aspect of the slope. The aspect of the slope determines the amount of insolation that can impinge on it. It is also significant to note that the highest mean temperature was recorded in February where the highest mean urease and soil moisture levels and lowest mean organic matter and soil pH values were recorded. This shows that the mix of soil moisture, pH and temperature was crucial in decomposition of the organic matter resulting in increased urease activity.

Soil texture and bulk density did not vary much within and among the zones. As a result they could not have any meaningful effect on the activity of urease in the soils of the forest. Results of this study pose challenges to popular views about the influence of soil moisture and soil organic matter content on soil enzyme activities, particularly along topographic gradient that require further investigations. We hereby recommend extended investigations into interactions among soil biochemical, chemical and physical factors to span at least three years.

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