LABORATORY CULTIVATION OF THE EARTHWORM
Eudrilus eugeniae,(Kinberg)

BY

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MARCH, 1995

DECLARATION

I hereby declare that this thesis has been written by me and that it is a complete record of my own research work. It has not been presented in any previous application for a higher degree.

All sources of information are specifically acknowledged by means of references.

Signed: _____

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27/04/95

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CERTIFICATION

This thesis titled "LABORATORY CULTIVATION OF THE EARTHWORM Eudrilus eugeniae,(Kingerg)" by Ibrahim,Baba Usman meets the regulations governing the award of degree of Master of Science of Ahmadu Bello University and is approved for its contribution to scientific knowledge.

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To

The Glory of Almighty God

AND

In honour and everlasting memory of my dear, courageous and beloved friend Late Alhaji Mamman Musa(Anaruwa Bako Kontagora).

May his soul rest in eternal peace.

Amen.

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May the Glory and Blèssing of Almighty God be upon all of you Amen.

ABSTRACT

Earthworms, Eudrilus eugeniae (Kinberg), were cultured using Loamy, sandy and clayey soils in earthenware pots for ten weeks under laboratory conditions. Two experiments were set up, the experimental containing organic matter, in form of cowdung, and the control without organic matter. Watering was done every three days to keep the culture moistened, but not saturated.

Results of these treatments showed that the number of earthworms harvested from the experimental soil samples, were significantly higher (P 0.05), than those of the control. The loamy soil, had the lighest number of earthworms harvested, fallowed by sand and lastly the clayey soil. The differences in the number of earthworms harvested from each soil type were highly significant (P 0.05).

There was positive correlation between the weight of earthworms and their lengths.

Soil moisture, organic matter, temperature, pH and porosity of the soil were found to affect the breeding of the earthworms. Loamy soil had the most favourable soil factors that enhanced the growth and survival of the earthworms.

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CHAPTER ONE

1.0. INTRODUCTION

1.1. HISTORY OF THE RESEARCH PROBLEM

Whenever laboratory animals are mentioned, it is usually the vertebrates that readily come to mind. Therefore this research would like to draw attention towards invertebrater, such as earthworm, as a laboratory animals. The earthworms have been successfully bred in captivity under laboratory conditions by scientists interestedin using them as research models (Frank et al.,1937. Bullough, 1958. Luker and Luker, 1971. Archenhold et al., 1978). Earthworms have been used in the laboratory for phylogenic studies and demonstration of behaviour. Most of the time, preserved earthworms are employed in biological studies, because it is often difficult to procure fresh live specimens from the wild, when required in large quantities. Freshly killed ones are better than the preserved specimens for the study of certain organ systems. Therefore there is a need to maintain a culture of earthworms in the laboratory to meet these demands, especially in the dry season when they are not easily available . The only documented work on Nigerian earthworms was that of Professor Segun of Obafemi Awolowo University, Ile-Ife, who succeeded in identifying many new species of earthworms. These species include, Hyperiodrilus Oshogbonensis and Hyperiodrilus malakai from Oshogbo and Ughelli respectively. Other new species were Iridodrilus tonyii and Iridodrilus vomiensis and Vomia Prima from Vom and Jos, a new genus Nsukkadrilus mbae from Nsukka.

Fourteen different species of Eudrillid earthworms were identified from Obafemi Awolowo University, Ile-Ife campus, which included four new ones Eminoscolex nigeriensis, Eminoscolex ifensis, Hippopera ajokei and Hippopera ifensis (Segun, 1992).

1.2. JUSTIFICATION

There has been a lot of work on earthworms, but for the most part, this has been concentrated on their biology, especially morphology, growth, population reproductive potential and taxonomic studies. However, in Nigeria not much work has been done on the possibility of breeding and maintaining them in the laboratory, on a continuous basis.

Earthworms are of great importance as they serve as food for other laboratory animals, such as toads and frogs, as bait for fishing, and for creating and improving soil fertility. They also serve as a source of revenue to many people in some parts of Sokoto and Borno States, where they have been sold to fishermen for fish baiting, and as food for man too, a delicacy in Japan. Earthworm have been used as a common example of the Phylum Annelida during teaching, their external features, behaviour, locomotion, burrowing and feeding also make it interesting to study them.

Despite the importance of earthworms they are not always available throughout the year, due to unfavourable seasonal changes in the environment.

1.3. OBJECTIVE

The objective of this research is to determine which of the three soil samples namely, loamy , clayey and sandy is most suitable for breeding and maintaining earthworms, under laboratory conditions.

CHAPTER TWO

2.0. REVIEW OF LITERATURE

2.1. CLASSIFICATION

The earthworm used in this work according to Edwards and Lofty(1972) can be classified as follows:-

Phylum - Annelida

Class - Chaetopoda

Order - Oligochaetae

Family - Eudrilidae

Genus - Eudrilus

Species - Eugeniae(Kinberg)

2.2. CHARACTERISTICS

Earthworms have well defined segmentation with setae, and a coelom usually divided into compactments by intersegmental septa.

Bullough(1958) recognised two living orders of chaetopoda , order polychaetae which are exclusively marine including swimming, crawling, burrowing and tube dwelling worms. Order oligochaetae , are land or fresh water animals. According to Bullough (1958), the body has a cylinderical pointed anterior end, a flattened blunt posterior end , the mouth and the anus are terminal. There are about 150 well marked segments , of which number 32-37 are swollen to form a clitellum, which secrete a cocoon. The head lacks sensory appendages such as eyes, tentacles and palps(fig.1) Earthworms vary in size , from a fraction of a centimeter to exceptional individuals of " Megascolides australis, measuring upto 2.75m in length, and 3cm in diameter(Edward and Lofty, 1972).

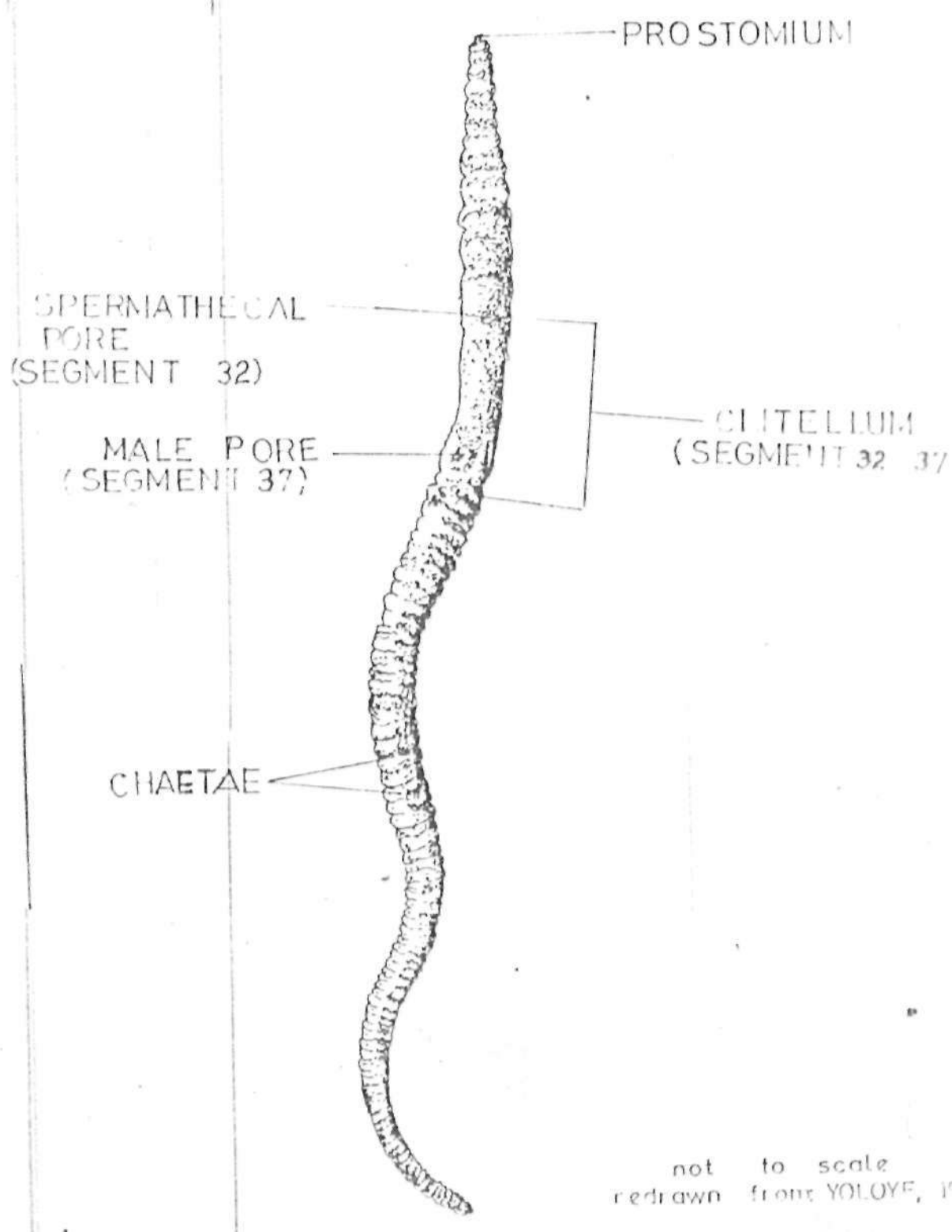


FIG. 1 EXTERNAL FEATURES OF EARTHWORM

2.3. HABITAT

Earthworms are found to inhabit almost any place where soil and green plant exist. They can be found everywhere in the world except in the desert which is too hot and too dry, and in the polar region, which is too cold (white, 1975)

2.4. DISTRIBUTION OF EARTHWORMS

Guild(1952) and Murchie(1958) mentioned possible factors that are responsible for the variability in the distribution of earthworms as follows:

(a) Physicochemical factors , such as soil, temperature, moisture, pH, inorganic salt, aeration, and texture of soil.

(b) Availability of food such as herbage, leaf litter, dung and consolidated organic matter.

(c) Reproductive potential and dispersive power of the species.

Murchie(1958) concluded that no single factor was solely responsible for the distribution, but rather the interaction of several factors.

2.4.1. HYDROGEN ION CONCENTRATION (pH)

Nielsen(1951) reported that the distribution of earthworm was mainly due to the exchangeable calcium in the soil , while Satchell(1955) suggested that the difference in the number of earthworms between plots could be explained more by pH, than by the exchangeable calcium. Madge and Sharma (1969) also reported that higher acidity was more important in limiting the distribution of some earthworms, compared with available calcium, and that some species of earthworms were acid tolerant , and occurred in organic soil with a pH of less than five. According to Satchell(1955), Reynold(1955) , and Wallwork(1970), earthworms were also found in slightly acidic and slightly basic soils, and that their tolerance to pH varied from species to species. There were few earthworms in soil with a pH below 4.3. except for one species Distichopus octaedra, which seemed to be acid tolerant (Bornesbusch, 1930). Eisenia foetida had been reported to prefer soils with a pH between 7.0 and 8.0, while certain tropical species of Megascolex thrive in acid soils from pH 4.5-4.7 (Bachelier, 1963). Madge(1969) reported that optinum pH for Hyperiodrilus africanus lay between 5.6.-9.2 indicating that the species is acid tolerant as well as basic tolerant. Duweini and Ghabour(1965) found that an increase of pH from 7.25 to 8.25 was associated with a decrease in number of earthworm in fourteen Egyptian soils investigated , thus showing

that soil can be too alkaline to favour earthworms.

2.4.2. LAND CULTIVATION AND VEGETATION

The differences in the number of earthworms between a cultivated soil and an uncultivated soil had been studied by many workers.

Evans and Guild(1948a), Jackson and Raw(1966), observed that less number of earthworms occur in cultivated soil, then uncultivated soil. They based their conclusion on the fact that the worms were killed. by cultivation and that cultivation replaces many of the earthworms activities. Edward (1975), reported that more earthworms were found in cultivated soil, then in uncultivated one. He concluded that the effects of cultivation on the population of earthworms depend on the frequency of cultivation i.e the high number of cultivation is deleterious , while lower frequency of cultivation increases the number of earthworms. The type of soil has a great influence on the distribution of earthworms. Guild(1948), studied the influence of soil types on earthworm populations , and reported that there were differences in total number and relative number of each species. He concluded that light and medium loams had higher total population of worms than heavier clay or more open gravellly sends and alluvial soils. Several workers including Ghilarov(1956, 1965), showed that the species of earthworms found in a soil can be an indicator of type of soil and its properties. Wallwork(1970) reported that maximmum density of

earthworms generally occurs in grassland and mullwood land, and minimum occurs in moorland and raw humus.

2.4.3. MOISTURE

Satchell(1971) reported a 38% soil moisture for the maximum activities of earthworms, and decreases in activities at 44% . Water constitutes 75-90% of the body weight of earthworms (Grant,1955). Therefore, prevention of mater loss is major problem of earthworm survival. Earthworms have considerable ability to survive adverse moisture conditions, either by moving to a more suitable area or by aestivating. If they can not avoid dry soil, they can survive the loss of a large part of the total water of their bodies. Lumbricus terrestris can lose 70% and Allolobophora Chlorotica 75% of their total body water and still survive(Roots, 1956). Gerard (1960), showed that some species can withstand dry conditions better than others, for instance Lumbricus. terrestris can survive in non irrigated plots as well as in irrigated ones, whereas Allolobophora chlorotica, Allolobophora caliginosa and Allolobophora rosea did not survive in the non irrigated plots. In the tropics the most important factor influencing seasonal variation in the number of earthworms is rain. Madge (1969), reported that the principal activity of Hyperiodrilus africanus in Nigeria was during May and June, at the beginning of the wet season, and thereafter the number decreases-gradually until in November when few were found. According to

Gerrard(1960), Lack of moisture can cause earthworms to become quiescent or go into diapause, for instance when individuals of Allolobophora caliginosa were kept in soil that was dried slowly in the Laboratory, they went into diapause but when kept in moist soil, they remained active for eighteen months. Madge (1969) placed earthworms Hyperiodrilus africanus in moisture gradients, and reported that they prefered soils with 12.5% and 17.2% moisture, while soil with a moisture content of about 23.3% was the optimum for them to produce cast. Thus earthworms are much more active in moist soil than dry ones. The distinct climatic seasons, such as winter and summer has been found to have influence on the number of earthworms. There is a high number in the summer than in the winter. This has been found by Evans and Guild (1947) , Edward and Lofty(1972) and VanRhee(1973), The general consensus is that , this fluctuation in number is brought about by changes in moisture and seasonal variation in the atmospheric temperature.

2.4.4. TEMPERATURE

Kollmannsperger (1956) found that the number of earthworms on the soil surface at night was positively correlated with temperature, and estimated 10.5°C as the optimum temperature for the activities of the earthworm. Satchell (1967) also concluded that the most suitable condition for activities of earthworms on the surface were lights , when the soil temperature did not exceed 1[illegible] .

10.5°C. Duweini and Ghabbour (1965) determined the prefer and lethal temperatures for Pheretima californica and Alma sp, and found the prefer temperature for these species to be 26°C-35°C and 24°C-26°C respectively, and the upper lethal temperatures 37°C and 38°C respectively. Madge (1969), tested Hyperiodrilus africanus in a temperature gradient, and found that they aggregated between 23.9°C and 31.5°C, while temperature above 34°C were avoided. According to Hopp(1947), the most important factor affecting seasonal variation in the number of earthworms in the arable soil of United States was the winter frost, which killed the worms in the unprotected surface.

2.4.5 ORGANIC MATTERS

The distribution of organic matter in the soil greatly influenced the distribution of earthworms. Soils that are poor in organic matter do not usually support large number of earthworms(Edward andLofty,1972) Edward and Lofty, 1972 also stated that large amount of organic matter in pasture usually coincide with large number of earthworms, and it is probably the gradual decrease in soil organic matter. If there are few number of earthworms, the decaying organic matter usually lie in a thick mat on the soil surface. Such mats of undisturbed organic matter occur in both woodlands(Richardson, 1938). and grassland(Raw,1962).

2.5. THE ROLE OF EARTHWORMS IN SOIL FERTILITY

There are certain activities of earthworms that have influence on the soil structure, and these eventually improve the fertility of the soil. These activities includes the ingestion of soil, partial breakdown of organic matter, intimate mixing of these fractions, ejection of these materials as surface cast, and burrowing through the soil and bringing subsoil to the surface (Edwards and Lofty, 1972). During these processes, they thoroughly mix the soil, and improve its water holding capacity. Thus improving fertility of the soil.

2.6. BREAKDOWN OF SOIL PARTICLES

The presence of small amount of sand and other large soil fractions in worm casts than in the surrounding soil, has been taken as evidence that worms can break down mineral particles to smaller units(Te,tia et al., 1950). Evans (1948) reported that the amounts of coarse sand relative to silt and clay in two pasture lands with large number of earthworms increased with depth, and he suggested that earthworms might be breaking down the coarse sand in the surface soil.

2.7. FORMATION OF AGGREGATES

Aggregates are mineral granules joined together in such a manner that they can resist wetting, erosion and remain loose when the soil is either dry or wet Edward and Lofty, 1972). A soil that is rich in

aggregates remain well drained and aerated, thus the formation of aggregates is of great importance to soil fertility. Guild (1955) reported that earthworm casts contain more water-stable aggregates than the surrounding soil. Hopp and Hopkins(1946) found out that the percentage of aggregates in soil to which earthworms, were added was greater than those in soil without earthworms.

2.8. AERATION,POROSITY AND DRAINAGE

Earthworms improve soil aeration by their burrowing activity and also influence the porosity of soils by their effect on soil structure. According to Edward and Lofty(1972), earthworm burrows increase the soil-air volume from 8% to 30% of the total soil volume. Teotia et al (1950), reported that earthworm activity increased the porosity of two soils from 27.5% to 31.6% , and 58.8% to 61.8% respectively. Soils with earthworms have been found to drain four to ten times. faster than soils without earthworms(Guild,1952).

In order to prove that earthworms play a significant role in soil fertility, many attempts have been made to add earthworms to poor soil, or to encourage the build up of earthworm population by addition of organic matter. According to Rhee Van (1969), such addition of earthworms to soil is of great importance in reclaiming flooded areas, that are subsequently drained, and put into cultivation, as in the Dutch polders.

The addition of earthworm casts to soil can also improve its structure and fertility. Casts usually have a higher pH, nitrate 'nitrogen organic matter, exchangeable magnesium, Phosphorus and moisture equivalent (Lunt and Jacobson, 1944).

2.9. FOOD AND FEEDING HABIT

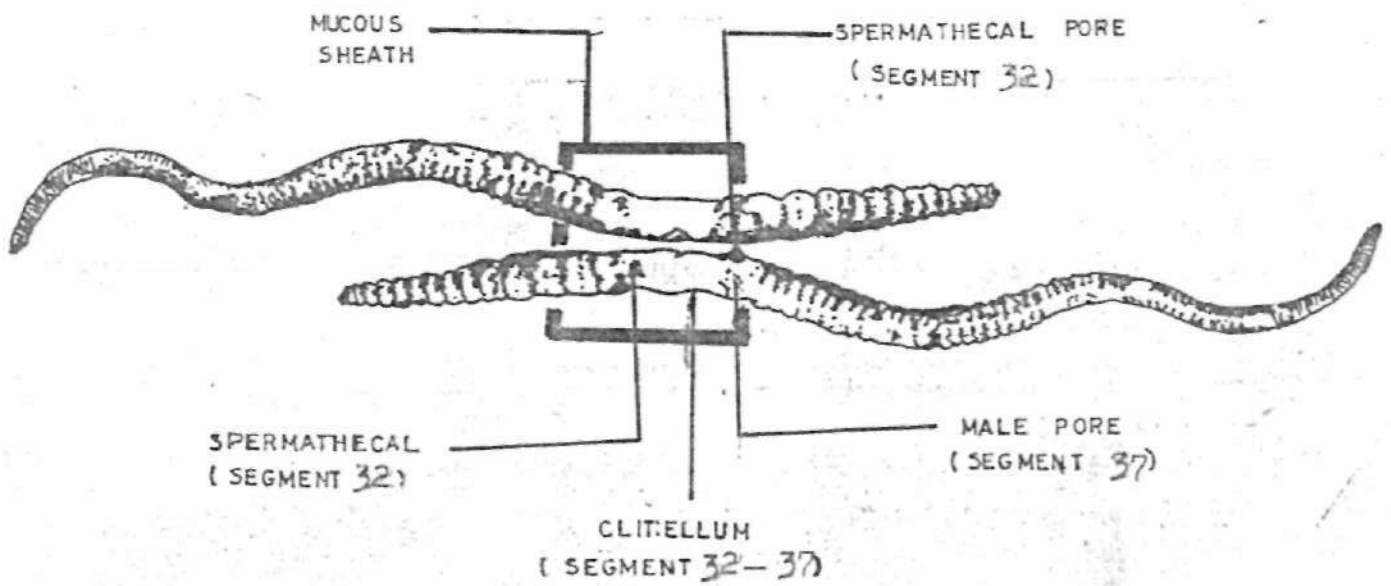
Whatever earthworm eats depends on the presence or absence of food. White(1975) stated that in the absence of leaves and tiny animals, the earthworm bulk-feed by taking in earth through the mouth land then take out of it, tiny bits of animals and vegetable materials. After the useful materials have been eaten up, the sand grains and gritly clay are passed on through the digestive system, and out through the anus into the soil, as worm casts. Guild (1955) reported that all species of earthworms prefer dungs and succulent herbage to the tree leaves. Barley (1959) fed individuals of Allolobophora caliginosa on different diets and reported that they grew at very different rates on these diets, growing fastest when fed on dung. Lindquist (1941) also reported that worms of species Allolobophora rosea and Allolobophora caliginosa were not attracted to litter but readily eat dung. Worms consume large amounts of food, for instance, Guild (1955) calculated that worms of 0.1g body weight ate as much as 80mg of food per day per gram of body weight of worm.

2.10. REPRODUCTIVE BEHAVIOUR

Earthworms are hermaphrodites , the male system consists of two pairs of testis enclosed in vesicular seminalis. The female system consists of a pair of ovaries, two oviducts and the spermotheca in which sperms received during corpulation are stored , until when needed to fertilized the eggs (Yoloye, 1988). Edward and Lofty (1972) also reported that earthworms, although hermaphradites, are not self fertilising , and that individuals usually mate (fig.2) and fertilize each other before fertile cocoons are produced. Cocoons contain eggs which hatch into immature worms, similar to the adults. A mature worm can be recognised by the presence of the clitellum. The clitellum is the glandular portion of the epidermis associated with cocoon production (Edward and Lofty, 1976). It usually appears swollen, and sometimes can only be differentiated externally by colour. The clitellum are recognisable throughout the year, or only at breeding season (Dale, 1963)

2.11. GROWTH

Growth in earthworms varies from species to species. According to Hyman(1940) , earthworms continue to grow through out their lives by adding segments proliferating from a growing zone, just infront of the anus. Angbashim (1991) reported that earthworm emerged from the cocoon possessing the full adult number of segments, and grew only by enlargement of segment.



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FIG. 2 - TWO EARTHWORMS IN COPULATION

Another study by moment (1963), on Eisenia foetida culture showed that newly emerged worms posseed average the same number of segments as adult, and the only worms that grew by adding segments were those dissected from cocoons. Vail (1974) reported that young of Eisenia foetida possessed between 28 and 122 distinct segments, and those of Bimastos tumidus, 69-128 distinct segments. Edwards and Lofty (1976) observed that Lumbricus sp. had fewer segments as they get older even though they continue to increase in size.

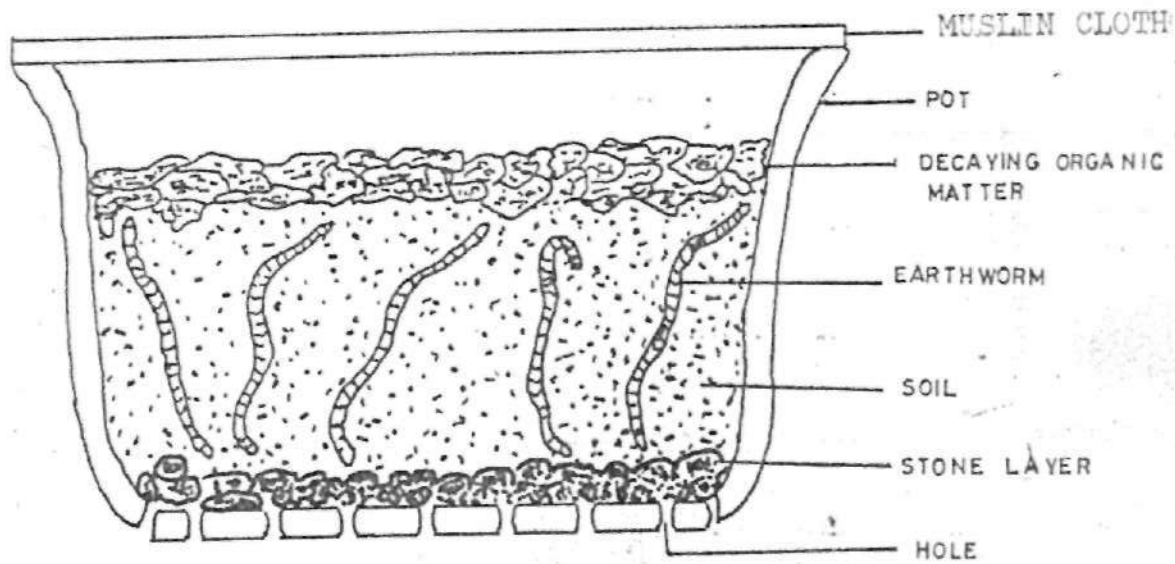
2.12. COLLECTION OF EARTHWORM

Earthworms can be collected for culture purpose by digging up soil samples and sorting this by hand. Workers who had used the above method included Hopp (1947), Barley(1959) and Rhee Van and Nathans(1961). Earthworms can also be collected by using the formalin extraction method (Raw, 1959). Several workers have compared the relative efficiency of extracting earthworms from soil by two or more of these methods. Svendsen(1955) reported that handsorting was much more efficient than using potassium permanganate. Raw(1959), compared formalin with potassium permanganate, and obtained more earthworm with formalin, then with no potassium permanganate.

2.13. EARTHWORM CULTURE

Several types of containers used with success for culturing earthworms included jam jars, specimen

tubes, luncheon boxes, oil drums, biscuit tin and wooden boxes (Ashby, 1976). He also recommended the size of boxes to be used in earthworm culture to be base 46x38cm height 20cm. Although not much attention has been given to the use of locally available products , such as earthenware pot, which are easily accessible and also at a lesser cost, Edward and Lofty(1972) recommended that stocks of earthworms can be kept in large earthenware pots, about 12cm in diameter and 30cm deep, with the top cover with muslin (Fig.3). He also stated that earthworm culture should be kept in a cool building or cellar at a temperature between 10°C and 15°C, or buried in the soil, and that the ideal culture medium for most of the common pasture and garden species is friable loamy soil with 25-30% mositure content. According to Ashby(1976) when considering the type oi container to be used in culture, it is important to bear in mind the necessity of providing sufficient aeration, soggy and waterlogged areas must not be allowed to develop in the medium, and if glass jar or tubes without adequate drainage are to be used , the medium must be carefully turned over at intervals . Guild(1957) reported that incubation of eggs took from 7 to 14 days in summer, and upto a month in winter , while in nature it took upto several weeks in summer and several months in winter. He also reported that smaller species of earthworms which take up to a year to mature in nature will mature in



REDRAWN FROM:- MADGE & SHARMA 1969

FIG.3 CULTURE METHOD FOR EARTHWORMS

6-8 weeks in Laboratory. The brandling, Eisenia foetida, a fast growing worm takes 4-6 weeks only to mature , while the larger species will mature in the Laboratory in 4-5 months. The cocoons are roughly spherical in shape and vary in size according to species. In smaller species such as Allolobophora to caliginosa, Allolobophora chlorotica, Lumbricus rubellus and Eisenia foetida , the cocoons vary in length from 2.5mm-3mm, where as those of Lumbricus terrestris are 6-7mm Long (Guild, 1957).

2.14. KILLING AND PRESERVATION OF EARTHWORMS

Earthworms can be killed by exposure to the fumes of chloroform , carbontetrachloride, and ethylacetate, then they can be preserved in formalin. For immediate dissection 60% ethyl alcohol as preservative allows the worm to remain pliable for several months. If they are wanted for dissection after a longer period of time, they should be preserved in 70% alcohol(Guild, 1957) . According to Edwards and Lofty(1972), earthworms for taxonomic study can be killed by placing them either in a 5% formalin solution, or in a fairly hot ($50^{o}C$) water, but formalin is more convenient and is also the most useful permanent preservative.

CHAPTER THREE

3.0. MATERIALS AND METHOD

3.1. COST SURVEY OF EARTHENWARE POTS, PLASTIC LUNCHEON BOXES, PLASTIC BUCKETS AND KLINER GLASS JAR.

A cost survey of the earthenware pots, plastic luncheon boxes, and plastic buckets were conducted at Samaru market, three different sizes of the earthenware pots, plastic luncheon boxes and plastic buckets were priced and photographed respectively. Three different sizes of kilner glass jars were also photographed and their prices obtained from Carolina Biological supply company catalogue.

3.2. COLLECTION OF SOIL SAMPLES

Loamy and clayey soil samples were collected from the Botanical Garden of the Department of Biological Sciences, Ahmadu Bello University, Zaria. While the sandy soil was collected at the University Dam in the main campus (Fig.4.)

3.3. COLLECTION OF EARTHWORMS

The earthworms were collected from the muddy and marshy areas along the University Dam, in the main campus, by digging and hand sorting (fig.4).

3.4. PERIOD OF STUDY

The experiment was conducted for a period of ten weeks, from April 16th to June, 24th, 1993.

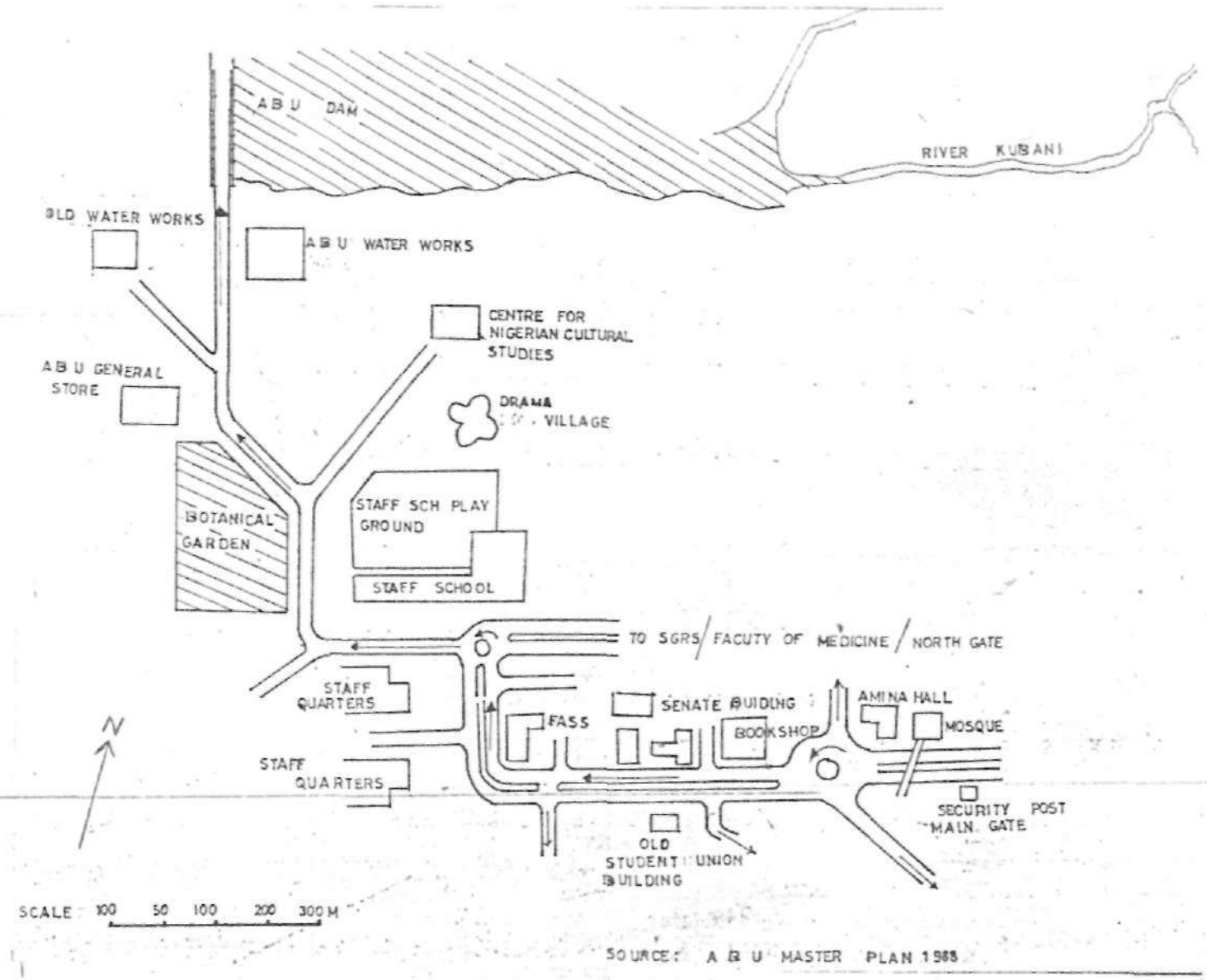


FIG. 4 MAP SHOWING THE BOTANICAL GARDEN AND A B U DAM

3.5.0. PROPERTIES OF SOIL SAMPLES DETERMINED

3.5.1 DETERMINATION OF SOIL pH(HYDROGEN ION CONCENTRATION)

The pH of the three types of soil samples were determined using the pH meter(Pye Model 292) . Fifty grams of each soil sample(Sandy, Loamy and Clayey soils) were weighed into 250cm^3 beaker respectively . 100 cm^3 of deionised water was added to the soil in each beaker. The content of each beaker was stirred with a glass rod intermittently, every five minutes, for about thirty minutes. This enabled the air to escape more rapidly. The soil solution was allowed to stand for another thirty minutes, while waiting the pH meter was standardised with buffer solution of pH4, pH7,and pH9. After thirty minutes, the electrode was washed with distilled water, and immersed in the solution so that it touches the soil. The pH of the soil sample was then read from the pH meter. The procedure was repeated for the second time and the average recorded.

3.5.2. DETERMINATION OF PERCENTAGE SOIL MOISTURE CONTENT

The percentage moisture content of the three types of soil samples was determined, by weighing thirty grams each soil sample in accrucible, and left in an electric oven, set at 105^{o}c, for twenty four hours, after which they were removed, cooled and reweighed. The difference in weight was the amount of water in the soil sample. The percentage soil moisture was calculated as follows:

% Percentage moisture content=

$$= \frac{\text{Initial weight - Final weight}}{\text{Initial weight of the soil.}} \times \frac{100}{1}$$

$$= \frac{\text{Loss of weight at } 105^{\circ}C}{\text{weight of oven dry soil}} \times \frac{100}{1}$$

This procedure was repeated for the second time, and the average recorded.

3.5.3. DETERMINATION OF SOIL ORGANIC CONTENT

The organic content of the three types of soil samples was determined. Thirty grams of the soil sample was put in a crucible, and dried for 24 hours in an oven set at $105^{\circ}C$. It was removed after 24hours, then cooled and re-weighed. The remains of the soil sample was kept in a furnace, and burnt overnight to ashes. The furnace was then switched off, and the soil sample, remained until it cooled down and re-weighed. The differences in weight between the oven dry weight and that of furnace gave the organic matter content of the soil sample. This was repeated for the second time. The percentage organic matter was calculated as follows:

$$\frac{\text{Initial weight- Final weight of the soil}}{\text{Initial weight of the soil}} \times \frac{100}{1}$$

Comparison were then made between the three soil samples.

3.5.4. DETERMINATION OF SOIL TEMPERATURE

The soil temperature of the three types of soil samples was determined by inserting a thermometer five

centimeters (5cm) deep below the soil surface, and the reading recorded. The procedure was repeated twice for each soil sample and the average recorded.

3.5.5. DETERMINATION OF POROSITY OF SOIL BEFORE AND AFTER THE EXPERIMENTS.

The porosity of the three types of soil samples was determined before and after the experiments. The bulk density of the soil sample was first determined by using a core sampler to obtain soil, the soil was then transferred in a crucible, and dried in an oven. The bulk density was obtained by dividing mass of dry soil, by volume of dry soil.

Db = Bulk Density $g\ Cm^{-3}$ ----------(1)

$$= \frac{\text{Mass of oven dry soil } g}{\text{volume of dry soil } cm^3}$$

Volume of dry soil = Volume of the core sampler

$$= \pi r^2 \times L$$

r = radius of the core sampler

L = Length of the core sampler

$$\pi = \frac{22}{7}$$

After obtaining the bulk density, the porosity of the soil was then determined as follows:

F = Porosity of the soil----------(2)

$$= 100 - \left(\frac{Db}{Dp} \times 100\right)$$

$$= 1 - \frac{Db}{Dp}$$

$$= 1 - \frac{\text{Bulk Density}}{\text{Particle Density.}}$$

Dp = Particle density is a constant, the international accepted particle density of soil ranges between 2.6- 2.7, but the mean is taken = 2.65(Hillel,1980)

Porosity of soil is normally in percentages

:. 1= 100 %.

3.6.0. PHYSICAL PARAMETERS OF THE EARTHWORM:

3.6.1. BODY WEIGHT OF THE EARTHWORM

The fresh body weight of the earthworm was measured in grams using an electric top loading balance (Metler balance). The weight was measured twice, to be certain of the measurement. Body weight of each earthworm was measured, and the initial stocking weight of adult earthworms in each experimental and control group, as well as the final body weight of fifty randomly selected earthworms during harvesting was recorded for each soil sample.

3.6.2. CONTRACTED LENGTH

The length of the earthworms were measured in centimeter using a measuring board, and a thirty centimeter ruler. The contracted length of adult earthworms were recorded when they were not moving. This was repeated, three times for each earthworm, and the average recorded.

3.6.3. EXTENDED LENGTH

The extended length of the adult earthworm was taken, when it was moving. Few drops of sodium chloride solution was applied to touch its posterior

end, thus inducing quick movement of the earthworm. This was repeated three times, and the mean extended length for each worm was calculated.

3.7.0. CULTURE METHOD

3.7.1. CULTIVATION OF EARTHWORM USING THREE TYPES OF SOIL SAMPLE MEDIA

Two experiments were set up . Experiment group A is the control group and experiment group B is the experimental group. Three types of soils were used as culture media, namely loamy, sandy and clayey soils. The soil was seived using 2mm mesh size seive, to remove the debris materials. In the control group, twelve pots were used (Plate 1). Four pots for each soil sample media designated A_1 A_1^1, C_1 C_1^1 Loamy soil , A_2 $A2^1$. C2 C_2^1 sandy soil and A_3 A_3^1 C_3 C_3^1 clayey soil. Each pot was filled with 9500g of soil samples, to a depth of 25cm. Ten earthworms were introduce to top of each pot, and allow to burrow down. The bottom of each pot with a hole, was lined with a layer of stones to allow free drainage of water, and the content sprinkled with the same amount of water (750 cm^3) every three days, so as to keep the culture medium moistened, but not saturated (plate 2) . They were kept in a cool place.

The second experimentel group B, was set up as in group A. Twelve pots were used. Four pots for each soil, sample medium designated B_1 B_1^1 D_1 D_1^1 loamy soil,



PLATE 1: 24 EARTHENWARE POTS USED FOR EARTHWORM CULTURE.

KEY TO THE PLATE 1

CONTROL GROUP			EXPERIMENTAL GROUP		
A1	=	LOAMY SOIL	B1	=	LOAMY SOIL
A_1^1	=	LOAMY SOIL	B_1^1	=	LOAMY SOIL
A2	=	SANDY SOIL	B2	=	SANDY SOIL
A_2^1	=	SANDY SOIL	B_2^1	=	SANDY SOIL
A3	=	CLAYEY SOIL	B3	=	CLAYEY SOIL
A_3^1	=	CLAYEY SOIL	B_3^1	=	CLAYEY SOIL
C1	=	LOAMY SOIL	D1	=	LOAMY SOIL
C_1^1	=	LOAMY SOIL	D_1^1	=	LOAMY SOIL
C2	=	SANDY SOIL	D2	=	SANDY SOIL
C_2^1	=	SANDY SOIL	D_2^1	=	SANDY SOIL
C3	=	CLAYEY SOIL	D3	=	CLAYEY SOIL
C_3^1	=	CLAYEY SOIL	D_3^1	=	CLAYEY SOIL.

12 POTS ON THE LEFT CONTAINED THE CONTROL SOIL SAMPLES WITHOUT ORGANIC MATTER, AND THE OTHER 12 ON THE RIGHT CONTAINED THE EXPERIMENTAL SOIL SAMPLES TREATED WITH ORGANIC MATTER (COW DUNG).



PLATE 2: THE RESEARCHER WATERING THE CULTURE WITH WATER SPRINKLER.

B2 B_2^1 D2 D_2^1 Sandy soil and B3 B_3^1 D3 D_3^1 clayey soil (plate 1). However decayed leaves and cowdung were introduced into the experimental group as food. The decayed leaves were mixed with the soil at the beginning of the culture while three hundred grams of cowdung were spread on top of the culture medium every week, and covered with about one centimeter of the soil sample. Thus the experimental group contained organic matter. The culturing lasted for a period of ten weeks, after which the earthworms were harvested from each pot. Their numbers, length and weight were taken. Comparison were then made between loamy soil treated with organic matter. And clayey soil treated with organic matter, and clayey soil not treated with organic matter. Comparison were also made between different soil samples to which organic matters were added. The earthworms harvested were bottled and preserved in 5% Formalin

3.8.0. STATISTICAL ANALYSIS

The mean length, number and weight of the earthworms in each pot were calculated. The standard deviation(SD), standard error(SE), analysis of variance(ANOVA) and coefficient of correlation of the length and weight of the earthworms were also determined for each soil sample.

3.8.1. ANALYSIS OF VARIANCE(ANOVA)

The analysis of variance (F-ratio test) was used to test the differences in growth in the three soil samples. The significant level was set at 95% confidence level (0.05).

3.8.2. COEFFICIENT OF CORRELATION(r)

The coefficient of correlation(r) was used to compare the two means of the weight and length of the earthworms (Appendix 9,10,11).

CHAPTER FOUR

4.0. RESULTS

4.1. COST SURVEY OF EARTHENWARE POTS, PLASTIC LUNCHEON BOXES , PLASTIC BUCKETS, AND KILNER GLASS JAR.

The result (Table 1) of the cost survey of three different sizes of earthenware pots, plastic luncheon boxes, plastic buckets and kilner glass jars, shows that the earthenware pots were the least costly of all, and were easily obtained. It was also easier to request for a particular size of earthenware pot from the manufacturer than the plastic luncheon boxes, plastic buckets and kilner glass jars. Plate 3,4,5 and 6 show the three different sizes of each container. And the earthenware pots were eventually chosen for the present study.

4.2. SIZE AND DIMENSION OF THE EARTHENWARE POTS USED FOR THE EXPERIMENTS.

The size and dimensions of the pots were indicated in Fig 5,, which shows the elevation and plans of the culturing pot at various positions.

4.4.0. PHYSICAL PARAMETERS

4.4.1. SOIL pH

The result obtained for pH , for both experimental and control soil samples were all basic (Table 2). There was no significant difference in the pH of each

TABLE 1

COST SURVEY OF EARTHENWARE POTS, PLASTIC LUNCHEON BOXES PLASTIC BUCKETS AND KILNER GLASS JAR.

NUMBER	COST											
	EARTHENWARE POTS			PLASTIC LUNCHEON BOXES				PLASTIC BUCKETS.			KILNER GLASS JARS		
	SIZE(cm)		PRICE	SIZE(cm)			PRICE	SIZE(cm)		PRICE	SIZE (cm)		PRICE
	L	D		L	B	H		L	D		L	D	
1	16	19	₦5:00	14	9	6	₦15:00	24	22	₦25:00	9.5	7.5	₦104: 75
2.	18	22	₦7:00	22	16	14	₦22:00	26	24	₦35:00	15	7.5	₦201: 34
3.	22	26	₦10:00	35	20	16	₦35:00	28	26	₦50:00	18.5	7.5	₦880: 88



PLATE 3: THREE DIFFERENT SIZES OF EARTHENWARE POTS PRICED AT SAMARU MARKET.

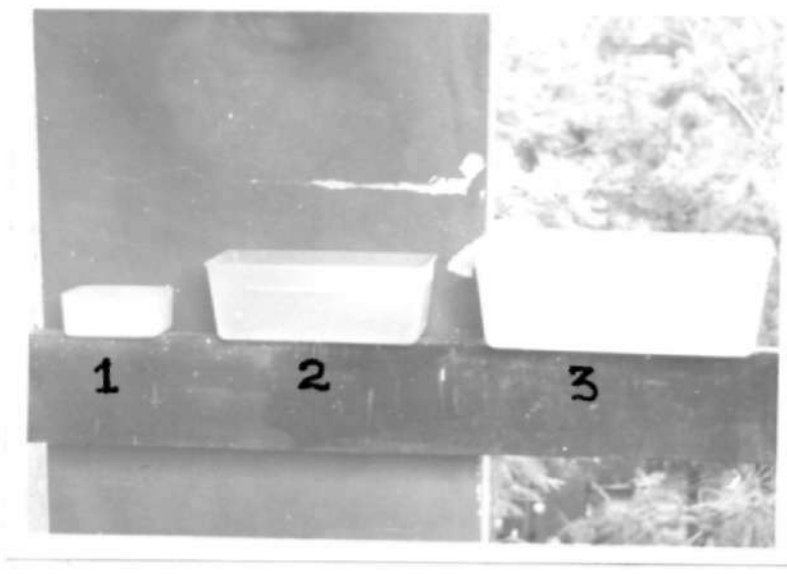


PLATE 4: THREE DIFFERENT SIZES OF PLASTIC LUNCHEON BOXES PRICED AT SAMARU MARKET.



PLATE 5: THREE DIFFERENT SIZES OF PLASTIC BUCKETS PRICED AT SAMARU MARKET.

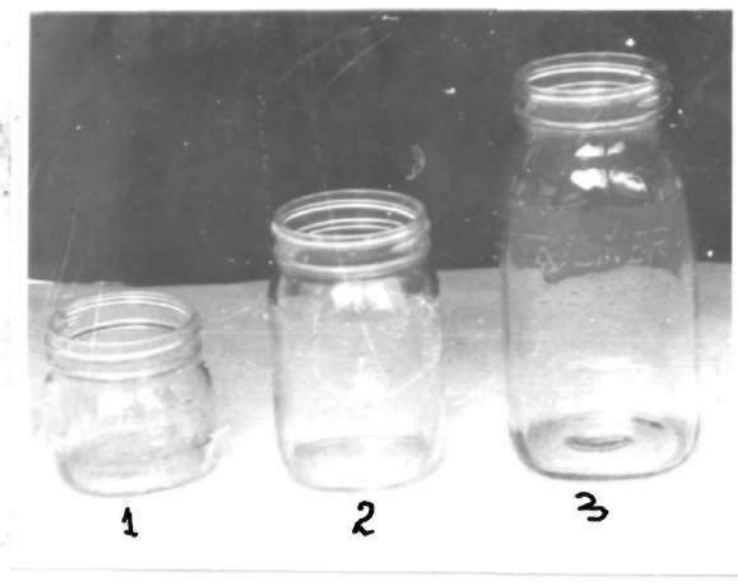


PLATE 6: THREE DIFFERENT SIZES OF KLINER GLASS JAR

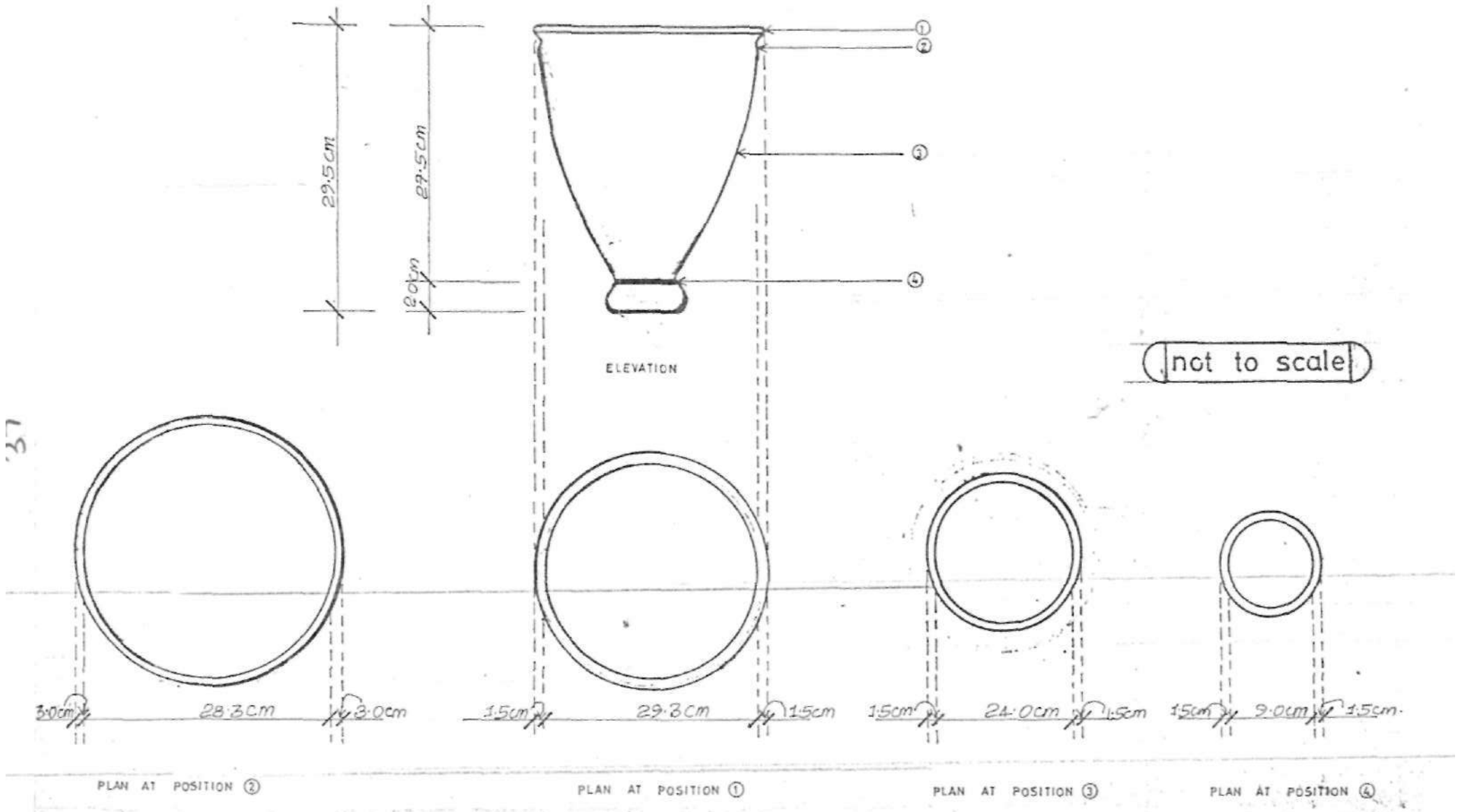


FIG.5 ELEVATION AND PLANS OF THE CULTURING POT

TABLE 2

MEAN SOIL PARAMETERS FOR THE THREE SOIL SAMPLES DURING THE CULTURES PERIOD

SOIL PARAMETER.	LOAMY SOIL		SANDY SOIL		CLAYEY SOIL	
	CONTROL .	EXPERIMENTAL.	CONTROL	EXPERIMENTAL.	CONTROL	EXPERIMENTAL
MEAN SOIL pH	8.3 ± 0.19	8.8 ± 0.21	8.3 ± 0.18	8.9 ± 0.21	7.6 ± 0.35	8.6 ± 0.25
Mean % Soil Moisture content	36.39± 1.64	47.59± 4.36	7.56± 0.54	34.29± 13.05	16.53± 1.08	40.30± 9.44
Mean % Orga Organic Content	13.52± 1.02	19.80± 2.71	0.86± 0.39	12.48± 3.89	3/14± 0.25	15.08± 3.08
Mean Soil Temperature	26.6± 0.69	26.7± 0.82	26.5 ± 0.53	27 ± 0.82	26.6 ± 0.52	27± 0.67
Mean % Soil Porosity after experiment.	71± 0.58	78± 2.38	51 ± 0.50	67± 3.09	45 ± 4.43	62± 2.16
Mean % Soil Porosity Before experiment	37± 1.41			47		43

soil sample in both experimental and control soil samples during the culture period (Appendix 1)

4.4.2. PERCENTAGE SOIL MOISTURE CONTENT

The percentage moisture content of the soil samples varied considerably in the experimental soil sample, during the study period , while in the control soil samples , the variation in moisture is not significant The mean value is highest for the loamy soil, followed by clayey and lastly sandy soil respectively. The result shows a significant difference(P 0.05), in the percentage moisture content between the experimental and the control soil sample with the experimental soil sampler having the highest values(Table 2,fig.6 and Appendix 2).

4.4.3. PERCENTAGE ORGANIC CONTENT

The percentage organic content varied with the soil types in both experimental and control soil samples Table 2, fig.7 and Appendix 3). The mean value was highest for the Loamy, followed by clayey and lastly a sandy soil types in both experimental and control soil samples.

4.4.4. SOIL TEMPERATURE

The mean soil temperature recorded for the soil types in both experimental and control soil samples showed no significant difference(Table 2) Also the temperature recorded for each soil type in both experimental and

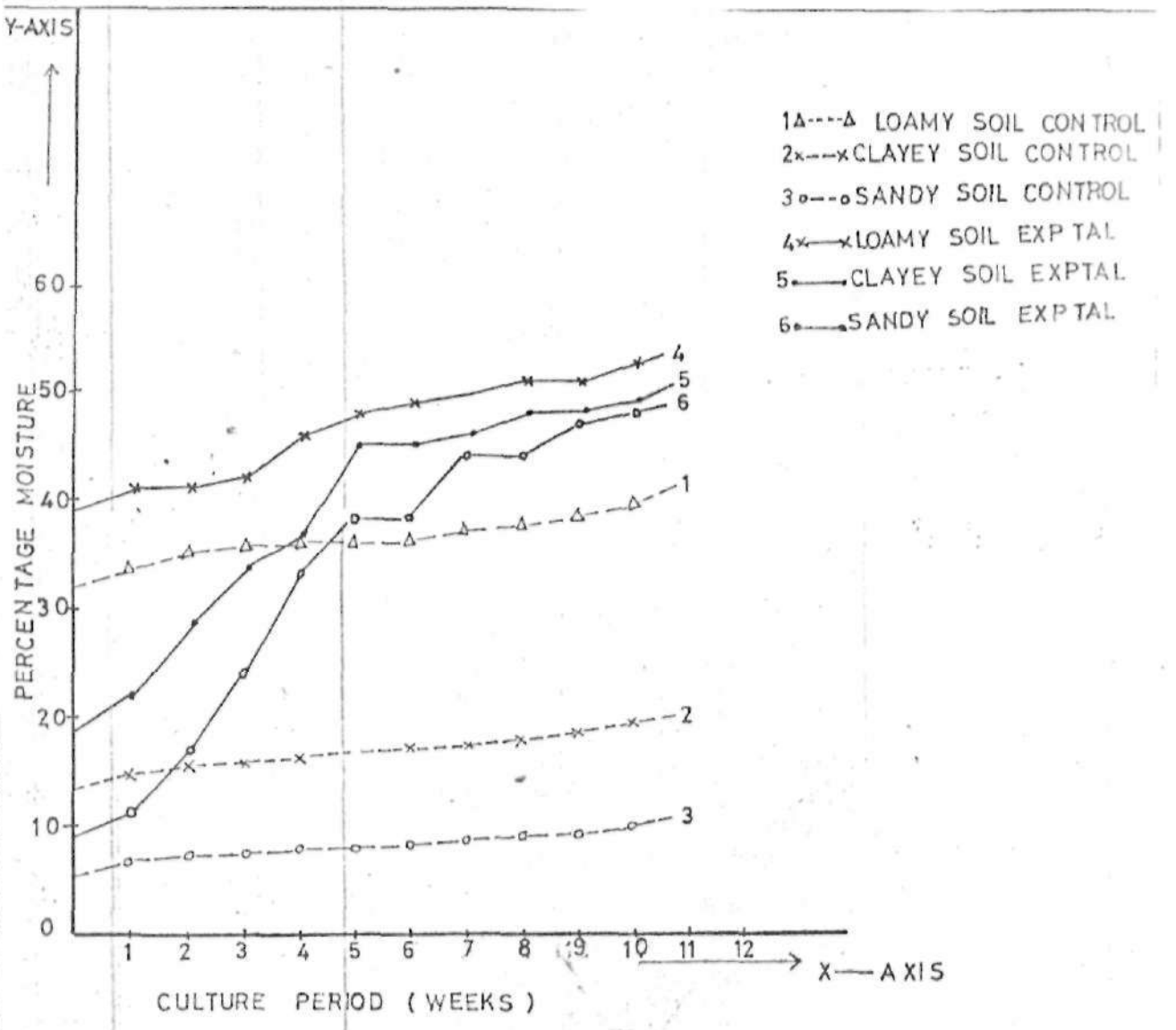


FIG. 6 VARIATION IN MOISTURE CONTENT FOR LOAMY CLAYEY AND SANDY SOILS

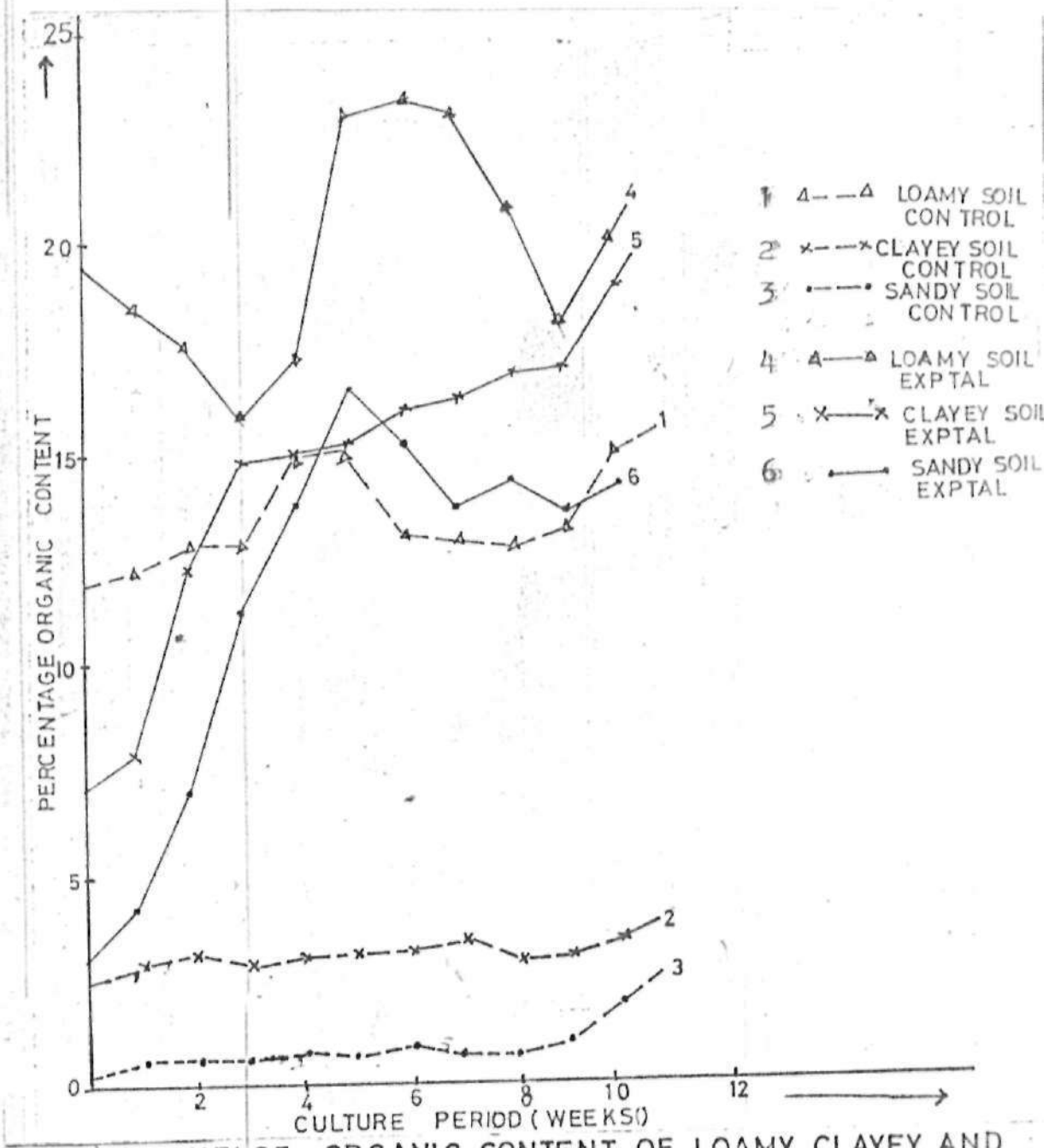


FIG.7 PERCENTAGE ORGANIC CONTENT OF LOAMY, CLAYEY AND SANDY SOIL

control soil samples were relatively constant, throughout the period (Appendix 6)

4.4.5. PERCENTAGE SOIL POROSITY

The percentage soil porosity varied with the soil types in both experimental and control soil samples, and within the period of the experiment i.e. before and after the culture period. The mean percentage porosity after the experiment was highest for the loamy soil, followed by sandy and lastly clayey (Table 2 and appendix 7.

4.5.0 CULTURING EXPERIMENT

4.5.1. EARTHWORMS HARVESTED FROM THE THREE SOIL SAMPLE

Table 4 showed a significant difference (P 0.05), between the population of earthworms harvested from the the three soil types, with the Loamy soil having the highest number of earthworms, followed by sand and lastly the clayey soil. There was also a significant difference(P 0.05) between the number of earthworms harvested in the control and experimental group for the soil types(Table 3, and appendix 8). The length of the earthworms harvested varied with the soil types, with the clayey soil having the longest length of the earthworms, followed by loamy and lastly the sandy soil. However, earthworms harvested from the experimental soil, were longer than those of the control(Table 3 Appendix 10 and 11).

TABLE 3

MEAN LENGTH, WEIGHT AND TOTAL NUMBER OF EARTHWORMS HARVESTED IN THE THREE SOIL SAMPLES

SOIL SAMPLE.	INITIAL NUMBER OF EARTHWORMS.	FINAL NUMBER OF EARTHWORMS.	INITIAL MEAN WEIGHT. (g)	FINAL MEAN WEIGHT. (g)	MEAN CONTRACTED LENGTH. (cm)	MEAN EXTENDED. LENGTH. (cm)	INCREASED IN NUMBER OF EARTHWORMS.	% INCREASED IN NUMBER OF EARTHWORMS.	INCREASED IN NUMBER WEIGHT.	% INCREASED IN MEAN WEIGHT.
LOAMY SOIL CONTROL	40	108	0.21	0.33	6.51	7.49	68	170%	0.12	57%
LOAMY SOIL EXPERIMENTAL	40	1398	0.28	0.54	9.12	10.54	1358	3395%	0.26	93%
SANDY SOIL CONTROL	40	77	0.16	0.25	5.18	6.02	37	93%	0.09	56%
SANDY SOIL EXPERIMENTAL	40	720	0.21	0.36	8.39	9.52	680	1700%	0.15	71%
CLAYEY SOIL CONTROL	40	69	0.21	0.41	5.89	6.93	29	73%	0.20	95%
CLAYEY SOIL EXPERIMENTAL	40	264	0.32	0.79	9.73	10.79	224	560 %	0.47	146%

There was a significant difference (P 0.05), between the mean weight of the earthworms harvested from the soil samples,with the clayey soil having the largest weight, followed by loamy and sandy soils respectively(Table 3).

4.5.2. EARTHWORM CASTS

It was observed that earthworms burrowed into the soil immediately they were introduced onto the culture surface. They started producing casts the second day of the experiment . More earthworm casts were noticed on the experimental soils treated with cowdung, compare with those of the control

CHAPTER FIVE

5.0. DISCUSSION

5.1. HYDROGEN ION CONCENTRATION EFFECTS

The pH of the soil samples obrained in this study showed that the basic pH served as a good condition for the survival of the earthworms. This result is in agreement with similar observations made by Fitchell (1955), Reynold (1955) and Wallwork (1970), who reported that earthworms were found in slightly basic soils. They further concluded that although earthworm tolerance to pH varied from species to species Madge (1969) reported that optimum pH for Hyperi lrilus africanus was between 5.2. and 9.2, indicating that this species can survive in acidic and basic soils. However, the basic pH recorded in the present study did not agree with the findings of Bachelier (1963) and Angbashim (1991), who reported acidic pH condition of 4.5-4.7 and 6.3 for Megascolex sp and Lumbricus rubellus respectively. This differences might be due to environmental condition during the period of the study.

5.2. COWDUNG EFFECT

The organic matter content of the experimental soil samples were greater than those of the control soil samples, probably this was because cowdung was introduced into the experimental soils. This could explain why there was a significant difference (P 0.05) between the population of earthworms in the experimental, and control soil samples.

Also, the loamy soil sample, had the highest population of earthworms , than the clayey and sandy, and this could be attributed to the fact that the loamy soil contained more organic matter, than the clayey and sandy soils. These results were in agreement with Edward and Lofty(1972), who reported that soils that were poor in organic matter did not usually support large number of earthworms. Duweini and Ghabbour (1965) observed that increase in the organic content of the soil were associated with increase in numbers of earthworms. Angbashim (1991), obtained a similar result using decayed leaves as a source of organic matter. However, the result of the present study, revealed that earthworms grow well when cowdung was introduced as a source of organic matter, than when decayed leaves were introduced by Angbashim (1991). 2636 earthworms of Hyperiodrilus eugeniae were harvested from the present study, using cowdung as a source of organic matter, while 67 earthworms of Lumbricus rubellus were obtained by Angbashim (1991) , using decayed leaves as organic matter content. The differences might be due to two different species of earthworms used and also differences in the conditions during the period of experiments. This is in agreement with earlier report of Guild(1955), that all species of earthworms prefer dungs to leaves. Barley (1959)

feeding individuals of Allolobophora caliginosa on different diets, reported that they grew at different rates on these diets , growing fastest on dung.

5.3. SOIL MOISTURE EFFECT

The result indicated that the loamy soil had more moisture than the sandy and clayey soil samples. Loamy soil supported the highest population of earthworms, because it provided more favourable condition for their growth and survival. This result does not agree with that of Angbashim (1991), who reported that clayey soil samples contained more mositure than loamy and sandy soils, but supported less population of earthworms than the loamy soil. This was attributed to the fact that the clayey soil sample had few air spaces filled with water, and also the kilner glass jar container used by Angbashim (1991) did not have holes to allow free drainage of water, thus the clayey soil became water logged, and this condition did not favour earthworm activities, thus resulting in fewer populations observed.

5.4. TEMPERATURE EFFECT

The temperature recorded for the activities of the earthworms in the soil samples during the culture period was between 26.5-27^{o}C , as opposed to 10.5^{o}C reported by Kollmann sperger(1955), Satchell (1967) and 21.9^{o}C reported by Angbashim (1991). However the result of the present study , agreed with that of Madge(1969), who recorded 23.9^{o}C- 31.5^{o}C as prefered temperature for

Hyperiodrilus africanus, Duweini and Ghabbour (1965) recorded 26-35°C and 24-26°C as prefered temperature for Pheretima californica and Alma sp respectively.

5.5. POROSITY EFFECT

The porosity of the soils after the experiment was greater than the porosity before the experiment, due to the increase in number and activities of the earthworms, during the culture period. This result is in line with those obtained by Teotia et al. (1950), who reported that earthworm activities increased the porosity of two soils from 27.5% to 31.6% and 58.8% to 61.8% respectively. Guild (1952) reported that soils with earthworms drained four to ten times faster than soils without earthworms.

5.6. EARTHWORM POPULATION HARVESTED FROM SOIL SAMPLES

The highest population of earthworms were harvested from the loamy soil, followed by the sandy and lastly the clayey soil samples. The result of Guild(1948) agreed with the present study, when he reported that light and medium loamy soils had higher total population of earthworms, than heavier clayey and more open gravelly sand. These results can be attributed to the soil structure, which influenced ability of the earthworms to penetrate into the soil successfully. Thus too compact soils like clay hinder earthworm penetration, while too loose soils as sand, make the burrows unable to retain their shape, and the earthworms may suffocate due to inadequate aeration (Madge and Sharma, 1969) Therefore they prefer the loamy soil to either sandy or clayey soil. The difference in the population of

earthworms obtained in the present study and that of Angbashim (1991) may be due to the fact that earthenware pots used in the present study may be better for culturing earthworms than the kilner glass jar used by Angbashim (1991). Also the differences may be due to differences in species of earthworms used and the number that can be housed by the containers used. The larger sized earthenware pot had more space to accommodate more earthworms and their activities. Also, there were holes at the bottom of the earthenware pot, which provided free drainage, preventing waterloging condition in the culture compared to kilner glass jars that had no outlet., and would therefore require periodic turning over to avoid waterloging. Thus Ashby (1976) stated that when considering the type of container to be used in earthworm culture, it is important to bear in mind the necessity of providing sufficient aeration. Edward and Lofty (1972), recommended that a stock of earthworms could be kept in large earthenware pots, about 12cm in diameter and 30cm deep. The pots used in this study met this recommendation for they were easily accesible , and can be obtained at less cost than kilner glass jar and plastic luncheon boxes.

5.7. BODY WEIGHT AND LENGTH OF THE EARTHWORM

The difference in weight and length of the earthworms could be attributed to the available organic matter in form of cowdung. The organic matter promoted their growth and multiplication. The results were in line with observation of Angbashim (1991), who used leaves as organic

matter. Barley(1959) also reported fast growth of earthworms fed with dungs. From the available literature, earthworms bulk-feed by taking in earth in the absence of leaves and dungs (White, 1975). This might be responsible for the survival of the earthworms in the control soil samples without organic matter content, and therefore they possessed smaller weight due to lack of adequate nutrition to promote proper growth. The earthworms from the clayey soil samples recorded the highest weight compared to sandy and loamy soil samples, probably because of the few population of worms in clayey soil sample there was more space and less competition for the available food.

5.8. EARTHWORM CASTS

Earthworm casts were more in the experimental soil treated with cowdung than in the control, probably because of the increase in number and activities of earthworms in the experimental soil sampler, than the control soil samples. This result was in line with the observation of Evans (1948), who reported that the presence of casts was a good indication of the amount of earthworms activity.

5.9. CONCLUSION

The species of earthworm Eudrilus eugeniae was found to prefer loamy to the clayey and sandy soil. The loamy soil with a pH of 8.8± 0.21 seemed to offer the most favourable conditions also included an organic matter content of 19.80± 2.71%, a soil moisture of

47.59± 4.36% and soil temperature of 26.7°C± 0.82. Apart from using Loamy soil, cowdungs could be used as a source of organic matter while earthenware pots had the advantage of being spacious and could be constructed with drainage holes at the bottom, thereby making them more suitable than either the smaller plastic luncheon boxes or kilner jars which are very expensive and more difficult to obtain.

Earthworms could thus be cultivated in the laboratory to provide ready live specimens for demonstration, teaching and research and for introduction into infertile lands. Thus reducing the dependency on preserved specimens for these purposes. This means that constant supply of the worms could be maintained throughout the year even during the cold and dry seasons when they may not be easily procured from the wild. Also, the earthworms could be bred for feeding to fish in ponds, hence making the upkeep of fish ponds easier especially during the dry seasons, when there is usually general food scarcity. Cultivated earthworms could also be exported to countries such as Japan, that relish them as special items of human food.

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APPENDIX 1

WEEKLY MEAN SOIL pH

WEEKLY	LOAMY SOIL		SANDY SOIL		CLAYEY SOIL.	
	CONTROL.	EXPERIMENTAL.	CONTROL.	EXPERIMENTAL.	CONTROL.	EXPERIMENTAL
1	8.4.	8.4.	8.4	8.6	7.4	8.2
2.	8.4	8.7	8.5	9.0	7.8	8.4
3.	8.1	8.6	8.5	8.6	7.7	8.7
4	8.2	8.7	8.4	8.7	7.5	8.7
5.	3.3	9.0	7.9	8.9	6.8	9.0
6	3.6	8.9.	8.4	8.9	8.2	8.9
7	3,3	8.7	8.5	9.1	7.6	8.8
8	8.5	9.0	8.3	9.2	7.6	8.8
9	8.1	9.1	8.3	9.0	7.6	8.5
10	8.C	8.7	8.2	9.1	7.7	8.4
$\bar{X}$	8.3	8.8	8.3	8.9	7.6	8.6
S^2	0.04	0.05	0.03	0.05	0.12	0.06
S.D	0.19	0.21	0.18	0.21	0.35	0.25
S.E.	0.06	0.07	0.06	0.07	0.11	0.08

APPENDIX 2

PERCENTAGE MOISTURE CONTENT

WEEK.	LOAMY SOIL		SAND SOIL		CLAYEY SOIL	
	CONTROL .	EXPERIMENTAL.	CONTROL.	EXPERIMENTAL.	CONTROL .	EXPERIMENT
1.	33.52	41.70	6.41	10.82	14.64	21.81
2.	35.25	41.76	7.09	16.90	15.13	28.68
3	35.53	42.48	7.29	23.63	15.80	33.91
4	35.75	46.13	7.41	32.84	16.31	37.43
5	35.95	48.04	7.55	37.59	16.57	44.86
6	36.11	49.19	7.76	38.20	16.93	44.86
7	36.80	50.83	7.88	43.94	16.93	45.98
8	37.48	51.10	7.99	44.02	17.30	48.28
9	38.40	51.15	7.99	47.01	17.72	48.39
10	39.20	53.56	8.27	47.96	17.98	49.04
$\bar{X}$	36.39	47.59	7.56	34.29	16.53	40.30
S^2	2.69	18.98	0.29	170.18	1.17	89.20
S.D.	1.64	4.36	0.54	13.05	1.08	9.44
S.E.	0.52	1.38	0.17	4.13	0.34	2.99

APPENDIX 3

PERCENTAGE SOIL ORGANIC CONTENT

WEEK	LOAMY SOIL		SANDY SOIL		CLAYEY SOIL	
	CONTROL	EXPERIMENTAL.	CONTROL.	EXPERIMENTAL.	CONTROL	EXPERIMENTAL
1.	12.25	18.41	0.60	4.30	2.99	7.81
2	12.83	17.68	0.72	7.04	3.02	12.40
3	12.74	15.89	0.58	11.34	2.78	14.92
4	14.90	17.30	0.82	13.73	3.11	15.02
5	14.97	23.0	0.69	16.67	3.14	15.22
6	13.22	23.41	0.81	15.39	3.17	16.14
7	13.15	23.10	0.72	13.72	3.40	16.36
8	12.98	20.79	0.70	14.43	2.94	16.94
9	13.22	18.14	1.09	13.67	3.15	16.97
10	14.98	20.29	1.89	14.46	3.69	18.99
$\bar{X}$	13.52	19.80	0.85	12.48	3.14	15.08
S^2	1.05	7.34	0.15	15.12	0.06	9.46
S.D.	1.02	2.71.	0.39	3.89	0.25	3.08
S.E.	0.32	0.86	0.12	1.23	0.08	0.97

APPENDIX 4

DAILY ATMOSPHERIC TEMPERATURE READING DURING THE CULTURE PERIOD

DAY	APRIL	MAY	JUNE
1	-	32°C	28°C
2	-	32°C	28°C
3	-	30	26
4	-	30	30
5.	-	29	29
6.	-	30	28
7	-	30	28
8	-	32	28
9	-	30	28
10	-	30	28
11	-	31	29
12	-	30	29
13	-	30	28
14	-	32	28
15	-	32	28
16	32°C	33	28
17	33°C	31	28
18	32	31	28
19	32	27	28
20	32	31	29
21	32	30	29
22	32	30	28
23	29	28	28
24	33	29	29
25	31	32	-
26	31	30	-
27	31	29	-
28	32	29	-
29	32	29	-
30	32	28	-
31	-	28	-
$\bar{X}_2$	32	30	28
S^2	0.92	1.96	0.54
S.D	0.96	1.39	0.74
S.E	0.25	0.25	0.15

APPENDIX 5

WEEKLY MEAN ATMOSPHERIC TEMPERATURE READING DURING THE CULTURE PERIOD

WEEK	ATMOSPHERIC TEMPERATURE
1.	32°C
2.	31°C
3.	30
4.	30
5.	31
6.	30
7	28
8	28
9	28
10	28
$\bar{X}$	29.60
S^2	2.27
S.D.	1.51
S.E.	0.48

APPENDIX 6

WEEKLY MEAN SOIL TEMPERATURE READING DURING THE CULTURE PERIOD

	LOAMY SOIL		SANDY SOIL		CLAYEY SOIL	
WEEK	CONTROL	EXPERIMENTAL	CONTROL	EXPERIMENTAL	CONTROL	EXPERIMENTAL
1	28	28	27	28	27	27
2	27	27	27	27	27	27
3	27	27	27	27	27	27
4	27	28	27	28	27	28
5	27	28	27	28	27	27
6	25	27	26	27	27	27
7	26	27	26	26	26	26
8	26	26	26	26	26	26
9	26	26	26	27	26	26
10	26	26	26	26	26	26
$\bar{X}$	26.6	27	26.5	27	26.6	26.7
S^2	0.40	0.67	0.28	0.67	0.27	0.46
S.D.	0.69	0.82	0.53	0.82	0.52	0.67
S.E.	0.22	0.26	0.17	0.26	0.16	0.21

APPENDIX 7

POROSITY OF SOIL BEFORE AND AFTER EXPERIMENT

POROSITY BEFORE THE EXPERIMENT.			POROSITY AFTER THE EXPERIMENT						
			EXPERIMENTAL GROUP.			CONTROL GROUP			
Loamy Soil.	Sandy Soil.	CLAYEY Soil.	Loamy Soil	Sandy Soil.	Clayey Soil.	Loamy Soil	Sandy Soil	Clayey Soil	
68%	47%	43%	B_1 = 80%	B_2=64%	B_3=63%	A_1=70%	A_2=52%	A_3=47%	
66%	47%	43%	B_1^1 = 75%	B_2^1 =71%	B_3^1=62%	A_1^1=71%	A_2^1=51%	A_3^1=49%	
-	-	-	D1 =76%	D_2 =67%	D_3=59%	C_1=70%	C_2=51%	C_3 =39%	
-		-	D_1^1 =79%	D_2^1 =65%	D_3^1=64%	C_1^1=71%	C_2^1=51%	C_3^1=43%	
$\bar{X}$=67%	47%	43%	78%	67%	52%	71%	51%	45%	
S^2=2.0		-	5.7	9.58	1.67	0.33	0.25	19.67	
S.E=1.41	-	-	2.38	3.09	2.16	0.58	0.50	4.43	
S.E=1.0	-	-	1.19	1.55	1.08	0.29	0.25	2.22	

APPENDIX 8

ANALYSIS OF VARIANCE (ANOVA) OF MEAN POPULATION OF EARTHWORMS HARVESTED IN THE THREE SOIL SAMPLES

MEAN POPULATION OF EARTHWORMS IN SOIL SAMPLES COMPARED.		ANOVA RESULT	REMARK
Sample 1 Vs Sandy experimental '180.0)	Sample 2 Sand Control (17.75)	F-147.98 df(1,6) P 0.05	Significantly different
Sample 3 Vs Loamy experimental 348.25	Sample 4 Loamy Control 27.0	F=106.44 df (1,6) P 0.05	Significantly different.
Sample 5 Vs Clayey experimental (65.50)	Sample 6 Clayey Control (17.25)	F= 55.67 df=(1,6) P 0.05	Significantly different
Sample 1 Vs Sandy experimental (180.0)	Sample 3 Loamy experimental (348.25)	F=24.77 df=(1,6) P 0.05	Significantly different
Sample 1 VS Sandy experimental (180.0)	Sample 4 Loamy Control (27.0	F=128.97 df (1,6) P 0.05	Significantly different.
Sample I Vs Loamy experimental (180.0))	Sample 5 Clay experimental. (65.50)	F= 60.11 df (1,6) P 0.05	Significantly different.
Sample 3 VS Loamy experimental (348.25	Sample 5 Clay experimental. (65.50)	F= 79.45 df (1,6) P 0.05	Significantly different.
Sample 3 Vs Loamy experimental (348.25)	Sample 4 Loamy control (27.0)	F= 106.44 df (1,6) P 0.05	Significantly different.
Sample 1 Vs Sandy experimental (180.0)	Sample 6 Clay control (17.25	F= 148.75 df(1,6) P 0.05	Significantly different
Sample 2 Vs Sandy control (17.75)	Sample3 Loamy experimental (348.25)	F= 113.07 df(1,6) P 0.05	Significantly different
Sample 2 Vs Sandy control (17.75	Sample 4 Loamy control (27.0)	F=16.90 df(1,6) P 0.05	Significantly different

APPENDIX 8 CONT'D

MEAN POPULATION OF EARTHWORMS IN SOIL SAMPLES COMPARED	ANOVA RESULT	REMARK
Sample 2 Vs Sample 5 Sandy Control Clay experimental. (17.75 (27.0)	F= 54.74 df(1,6) P 0.05	Significantly different
Sample 4 Vs Sample 5 Loamy control Clay experimental. (27.0) (65.50)	F= 32.75 df(1,6) P 0.05	Significantly different
Sample 4 Vs Sample 6 Loamy Control Clay Control (27.0) (17.25)	F= 18.17 df(1,6) P 0.05	Significantly different
Sample 2 Vs Sample 6 Sandy Control Clay Control (17.75) (17.25)	F= 0.15 df(1,6) P 0.05	Not Significantly difference

APPENDIX 9

CORRELATION COEFFICIENT(r)

	Weight		Length		
No	X	X^2	Y	Y^2	XY
1					
2					
3					
4					
5					

(i) Variance of $X = \dfrac{X^2 - \frac{(x)^2}{N}}{N-1}$

(ii) Variance of $Y = \dfrac{Y^2 - \frac{(Y)^2}{N}}{N-1}$

(iii) Variance of $XY = \dfrac{xY - \frac{X.\ Y}{N}}{N-1}$

However, $XY - \dfrac{(X\ Y)}{N}$

$$r = \frac{\dfrac{}{N-1}}{\sqrt{\dfrac{X^2 - \frac{(X)^2}{N}}{N-1} \cdot \dfrac{Y^2 - \frac{(Y)^2}{N}}{N-1}}}$$

$$r = \frac{X.Y}{X.\ Y}$$

The calculate 'r' value is then compared with the table value to determine the kind of relationship.

(To further confirm the result of the association t-test was used)

That is $t = \dfrac{r}{\sqrt{\dfrac{1 - r^2}{N-2}}}$

APPENDIX 10

THE CORRELATION COEFFICIENT ANALYSIS BETWEEN BODY WEIGHT AND THE CONTRACTED LENGTH OF THE EARTHWORMS.

POT NO.	BODY WEIGHT X	X^2	CONTRACTED LENGTH (Y)	Y^2	XX
1.	0.39	0.15	7.81	60.99	3.05
2	0.48	0.23	8.27	68.39	3.97
3.	0.46	0.21	9.68	93.70	4.45
4.	0.50	0.25	10.76	115.78	5.38
5	1.09	1.19	10.46	109.41	11.40
6.	0.57	0.32	10.90	118.81	6.21
7	0.47	0.22	8.64	74.65	4.06
8	0.42	0.18	8.86	78.49	3.72
9	0.38	0.14	7.99	63.84	3.04
10	0.43	0.18	8.05	64.80	3.46
11	0.48	0.23	8.19	67.08	3.93
12	1.02	1.04	9.37	87.79	9.56
13	0.28	0.08	7.34	53.88	2.06
14.	0.35	0.12	6.84	46.79	2.39
15.	0.31	0.09	6.22	38.69	1.93
16.	0.19	0.04	5.15	26.52	0.98
17	0.34	0.12	6.17	38.07	2.09
18	0.35	0.12	6.27	39.31	2.19
19	0.28	0.08	5.41	29.27	1.51
20	0.27	0.07	6.46	41.73	1.74
21	0.25	0.06	4.42	19.54	1.11
22	0.24	0.06	4.93	24.30	1.18
23	0.31	0.09	5.56	30 .91	1.72
24	0.32	0.10	5.57	31.02	1.78
	10.18	5.3/	179.32	1423.76	82.91

r= + 0.73. The calculated 'r' value (+0.73) is greater than the table value 0.396 at 5% level of significance and thus the correlation is significant. Similar result was obtained when t-test was conducted (t=5.10) compare to table value 2.069 at P= 0.5% level of significant.

APPENDIX 11

THE CORRELATION COEFFICIENT ANALYSIS BETWEEN BODY WEIGHT AND THE EXTENDED LENGTH OF THE EARTHWORMS.

POT NO	BODY WEIGHT. X	X^2	EXTENDED LENGTH (Y)	Y^2	XY
1	0.39	0.15	8.84	78.15	3.45
2.	0.48	0.23	9.48	89.87	4.55
3.	0.46	0.21	11.10	123.21	5.11
4.	0.50	0.25	12.18	148.35	6.09
5	1.09	1.19	11.59	134.33	12.63
6.	0.57	0.32	11.96	143.04	6.82
7	0.47	0.22	9.68	93.70	4.55
8	0.42	0.18	10.07	101.40	4.23
9	0.38	0.14	9.48	89.87	3.60
10	0.43	0.18	9.39	88.17	4.04
11	0.48	0.23	9.16	83.91	4.39
12	1.02	1.04	10.45	109.20	10.66
13	0.28	0.08	8.38	70.22	2.35
14	0.35	0.12	7.82	61.15	2.74
15	0.31	0.09	7.02	49.28	2.18
16	0.91	0.83	5.95	35.40	5.41
17	0.34	0.12	6.93	48.02	2.36
18	0.35	0.12	7.04	49.56	2.46
19	0.28	0.08	6.46	41.73	1.81
20	0.27	0.07	7.32	53.58	1.98
21	0.25	0.06	5.29	27.98	1.32
22	0.24	0.06	5.80	33.64	1.39
23	0.31	0.09	6.71	45.02	2.08
24	0.32	0.10	7.05	49.70	2.26
	10.90	6.16	205.15	1848.48	98.46

r= +0.49. The calculated 'r' value(+0.49) is greater than the table value (0.396) at 0.5% level of significance, and thus the correlation is significant. Similar result was obtained with t-test (t=2.69 greater than table value 2.069 df 23) at 0.5% level of significance, thus the correlation is significant.

APPENDIX 12

IDENTIFICATION FEATURES

Eudrilus eugeniae

1. Clittellum commencing in front of the 15 segment.
2. Setae arrangement is lumbricus
3. Dorsal pore lacking
4. Inter segmental furrows visible on clittelum
5. Length 90 - 185 mm
6. Colour- Dorsal surface only red