IDENTIFICATION OF NEMATODES AND MORPHOLOGICAL CHARACTERISATION OF *SCUTELLONEMA BRADYS* ASSOCIATED WITH YAM FROM GUINEA SAVANNA AND TRANSITIONAL RAIN FOREST ECOLOGICAL ZONES OF GHANA

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DECLARATION

I do hereby solemnly declare that, this thesis is the original research work of Saaka Abubakar J. Yahaya towards the award of the Master of Philosophy Crop Science (Plant Pathology) in the department of crop science, College of Basic and Applied Science (CBAS), University of Ghana, Legon except for other peoples' work which has been duly cited and acknowledged and thus, neither part nor whole of this thesis has been submitted elsewhere for another degree.

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ABSTRACT

Nematode parasitism is a major constraint and has resulted in major loses to both yam farmers and marketers in all yam producing areas in West Africa. Nematodes causes significant damage to yam tubers resulting in deformed, unsightly tubers or tubers with cracked and flaking skin that conceals the underlying rot. Farmers have over long periods been experiencing severe loses as result of nematode infestation both in the field and in storage. A study was therefore conducted from August, 2014 to May, 2015 to assess farmers' knowledge and perception about nematode occurrence and its' economic impact on their livelihoods; determine the different types, abundance and frequency of occurrence of plant-parasitic nematodes (PPNs) associated with yam; and determine morphological and morphometric variations among the yam nematode population. Assessment of farmers' knowledge and perception was ascertained through questionnaires and interviews of 60 yam farmers in the Guinea savanna and Transitional rain forest agro-ecological zones (AEZs). The questionnaire focused on farmers' demographic characteristics, their land use intensity, their knowledge, perception and experiences concerning occurrence, management and economic importance of nematodes associated with yam. The findings indicated that most farmers of yam aware of the physical damage that nematodes cause to yam tubers, but are completely unaware of what causes the damage, its spread and management. Nine genera of PPN belonging to six families of Tylenchida and Tryplonchida were identified from both the yam rhizosphere soils and yam tubers. They included; *Meloidogyne* spp., *Pratylenchus* spp., *Scutellonema* spp., *Tylenchus* spp., *Trichodorus* spp., *Tylenchorhynchus* spp., *Rotylenchus* spp., *Hoplolaimus* spp., and *Helicotylenchus* spp.The lesion nematode (*Pratylenchus* spp.), *Scutellonema bradys* and *Meloidogyne* spp. occurred with frequency of 100% in both yam rhizosphere soils and yam tubers in the field during the rainy season and yam tubers in storage during the dry season. *Pratylenchus* spp. was observed with the highest frequency of 22.9%, 46.3% and 22.3% for soil and yam tubers in the field during the rainy season and yam tubers in storage during the dry season respectively. *Scutellonema* spp. and *Meloidogyne* spp. had relative abundance of 18.8% and 18.2% for soils, 20.8% and 19.2% for yam tubers in storage respectively. The highest nematode population density in yam rhizosphere soils, yam tubers in the field and yam tubers in storage was recorded in Kanshegu in the Guinea savanna AEZ. Morphological descriptions and morphometric measurements are given for *S. bradys* female and male populations. Morphometric measurements from five populations in each AEZ using 14 female and male characters corresponded with previous descriptions of this species. Principal component analysis on eight variable that were identified as the most valuable characters in differentiating both male and female of *S. bradys*, could not distinctly separate the populations into groups.

DEDICATION

I dedicate this work wholeheartedly to my wife, Saaka Jamila, my sons; Saaka Biawuribi, Saaka Nunbrase and my daughter, Saaka Afeso for their endurance.

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LIST OF ABBREVIATIONS

AEA	Agricultural Extension Agent
AEZ	Agro Ecological Zone
ANOVA	Analysis of Variance
AS	Ashanti Region
BAR	Brong Ahafo Region
cm^3	Centimeter cube
F	Factor Analysis
FAO	Food and Agriculture Organization of the United Nations
FAOSTAT	Food and Agriculture Organization of the United Nations Statistics
FG	Formalin Glycerol
g	Gram
GPS	Global Positioning System
GS	Guinea Savanna
GTZ	German Technical Co-operation
IITA	International Institute of Tropical Agriculture
IPM	Integrated Pest Management
JHS	Junior High School
Kg	Kilogram
L	Liters
LSD	Least Significant Difference
MiDA	Millennium Development Authority
ml	Milliliters

MoFA	Ministry of Food and Agriculture
MT	Metric Tones
NPK	Nitrogen, Phosphorus and Potassium
NR	Northern Region
P	Probability
PC	Principal Component
PPN	Plant Parasitic Nematodes
SHS	Senior High School
SPSS	Statistical Package for Social Sciences
SRID	Statistics, Research and Information Directorate
TRF	Transitional Rain Forest
WA	West Africa
UG	University of Ghana
µm	Micrometers

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background

Edible yam (*Diosorea* spp.) is a major food security crop in the diet of over 70 million people in the world (FAO, 2012). Yam has a high calorie content (381 kCal per 100 g), protein (4.94 g per 100 g), vitamin (64 mg per 100 g), fibre (13.23 g per 100 g), and minerals (2.97 g per 100 g) (Jonathan *et al.*, 2011). It is the second most important tuber crop in West Africa after cassava (*Manihot utilisima*) in terms of tonnage of production and in food security terms (Robertson & Lupien, 2008). Yam is also a major staple food crop in Ghana and in other parts of the world especially West- and East-Africa, Caribbean, and Asia-Pacific regions where the crop is largely cultivated. Its' production occurs in all regions in Ghana except Greater Accra and Upper East regions. Brong-Ahafo region is the leading producer of yam (39%) in Ghana, Northern and Eastern regions produce 25% and 12% of yams in the coutry respectively, with the remaining 24% distributed between Upper West, Ashanti, Volta and Western regions (MoFA,SRID, 2011). The Guinea savanna and the transitional rain forest zones which consists of Northern, Brong Ahafo and some parts of Ashanti regions contributes about 72 % of the the total yam produced in Ghana (MoFA, SRID, 2011).

The major yam producing countries in West Africa include Nigeria, Ghana, Benin, Cote d'Ivore and Cameroon (FAO, 2013). Almost 80% of the world's yam production occurs in West Africa (Bridge *et al.*, 2005; Coyne *et al.*, 2006), Nigeria produces 38 million tonnes of yams to accounts for 69.3% of the total yam produced in West Africa whilst, Ghana, Cote d'Ivore and Benin accounts for 12.1%, 10.6% and 5.8% respectively (FAOSTAT, 2013). Ghana, however, is the leading exporter (20,841

MT) of yam accounting for 94% of total yam exports (MiDA, 2010). Yam contributes to 16% of Ghana's Agricultural domestic product (FAO, 2013).

Considering the impact of nematodes on crops, McCarter (2009) estimated a global total crop loss of $118 billion for 2001. It has also been reported that annual loses of over 25% of the yield of yam is due to diseases and pests globally (Ezeh, 1998; FAO, 2012). Kwoseh *et al.*, (2005) estimated loses of yam tubers in storage due to nematodes to be about 25%. Yield loses and poor tuber quality is the major effect of plant parasitic nematodes (Adegbite *et al.*, 2005; Bridge *et al.*, 2005; Coyne *et al.*, 2006; Humphreys *et al.*, 2014).

Although nematodes are the most numerous and diverse metazoans with a total number estimated around a million species (Lambshead, 2004), they are interetestingly the least studied with just about less than 3% of the different species described so far (Hugot *et al.*, 2001; Hallan, 2007).
Nematodes of utmost importance to yam include root knot (*Meloidgyne* spp.), lesion nematodes (*Pratylenchus* spp.), and the yam nematode (*Scutellonema brodys* steiner and Le Hew 1933, Andrassy 1958*)* (Coyne *et al.*, 2006). Yam production in Ghana is adversely affected by plant-parasitic nematodes, particularly the yam nematode *(Scutellonema bradys)*.

In the West African Sub region and in Ghana in particular, *Scutellonema bradys* has been confirmed as the causal agent of dry rot in yam tubers in storage and in the field (Kwoseh *et al.*, 2005). A study by Kwoseh *et al.*, (2005) revealed that stored yam tubers had 83% and 100% prevalence rate of *S. bradys* in the transitional forest zone and the Guinea savanna agro-ecological zones respectively whiles that of *Meloidogyne* spp. was about 72% and 28.6% in the Forest transitional zone and the Guinea savanna agro-ecological zones respectively. (Kwoseh *et al.*, 2005).

The genus *Scutellonema (*Andrassy *1958)* has 46 valid species (Siddiqi, 2000), but only three are recognized as agricultural pests; *Sentellonema bradys*, *Scutellenama cavenensis* and *scutellonema brachyunus*. *Scutallenama bradys* is a migratory endoparasite and has all of its life stages infective in the yam (*Dioscorea* sp) tubers and roots. This nematode causes significant loses of yam in sub-saharan Africa. The distribution of seed tubers harboring the yam nematode which may not show any symptoms inadvertently facilitates transmission of this nematode to new areas (Baimy *et al.*, *2006). Scutellonema bradys* also causes significant damage in sweet potato (*Ipormea batatas*) (Coyne *et al.*, 2006) and potato (*Solammun tuberosum* L) (Coyne *et al.*, 2011).

Nematodes are considered as some of the most difficult organisms to identify because of their microscopic size, morphological similarity and limited number of distinguishable taxonomic characters and overlapping morphometric measurements. This limits the ability of morphological and morphometric characters to delimit closely related species, but they are when facilities for genome analysis is limited. Accurate identification is a prerequisite to an understanding of the effects of nematodes as parasites of plants. It is also fundamental to understanding and communication of the ecological role of any organism. It is obvious that no significant study on plant pests can be initiated until the identity of the parasite has been established. Also, understanding nematodes occurrence, population densities and diversities is very fundamental for the success of any future management strategy for nematodes affecting yams in the Guinea savanna and Transitional rain forest agro ecological zones of Ghana.

In Ghana, although, the distribution and economic importance of soil nematodes have been studied long ago (Addoh, 1971) and also the presence of the yam nematode confirmed on stored yam (Kwoseh *et al*., 2005), there has been little or no documentation as to a comprehensive data on the

the occurrence and farmers perceptions across the major yam growing areas as well as the accurate identification of the yam nematode using morphological features and morphometric analysis.

This study therefore seek to generally establish the occurrence as well as accurately identify the yam nematode in Ghana using its' morphological features and morphometric analysis

The specific objectives were to:

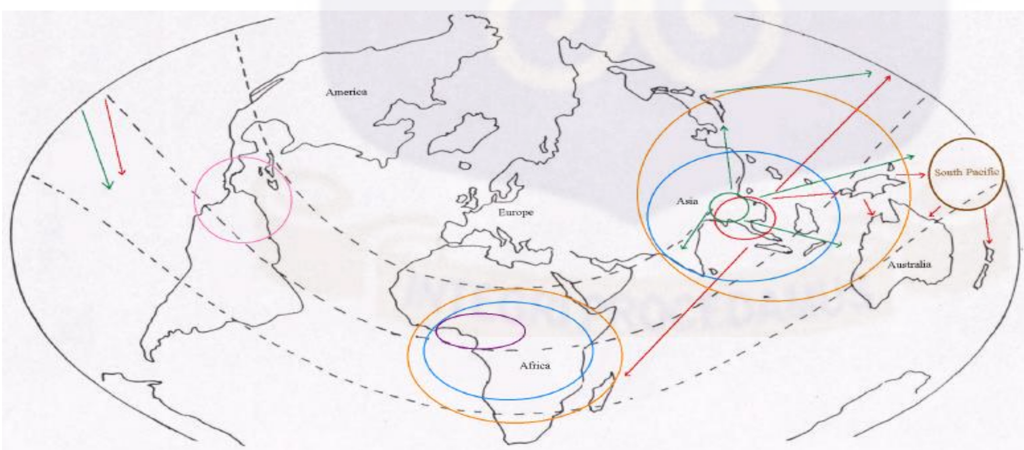
1. assess knowledge, perception and experiences of farmers in the Guinea savanna and Transitional agro ecological zones concerning the occurrence and management of yam nematode.
2. identify and assess populations of nematode genera affecting yam in the Brong- Ahafo and Northern regions of Ghana.
3. determine the variation within and among populations of *Scutellonema bradys* through morphometric analysis.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Origin of Yam

Yams (*Dioscorea* spp.) are annual climbing plants with edible underground tubers. It belongs to the genus *Dioscorea* (family *Dioscoreaceae*). There are an estimated 300-600 species available worldwide (Govaerts *et al.,* 2007), and of these only about six species are edible (Ofori and Hahn, 1991), while others are grown for medicinal purposes (Wu *et al.*, 2005; Lebot, 2009). Yams are believed to have originated from Far East and spread through the westwards (Coursey, 1975) (Fig 2). They have since evolved independently in the Eastern and Western Hemispheres and today yams are grown widely throughout the tropics (Asiedu *et al*., 2003). In the West African yam zone, which is the principal producer on a global basis, white yam (*D. rotundata), water yam (D. alata*), and yellow yam (*D. cayenensis)* are the most common species (Degras, 1993). White yam (*D. rotundata*) originated in Africa and is the most widely grown and preferred yam species (Aseidu *et al.*, 2003).



D. alata *D. dumetorum* and *D. hipsida* *D. trifida* *D. esculenta* *D. cayenensis* and *D. rotundata* *D. bulbifera*

Figure 1. Origin and distribution of *Dioscorea* species. Source: Degras (1993).

2.2 Economic importance and uses of yam

Yams are second to cassava as the most important tropical root crop (in terms of production or consumption) (Dadzie, 2004). They are staple crop in many parts of Africa and Southeast Asia (Amusa *et al*., 2003). In South Pacific, yam is a significant food crop, accounting for over 20%, 8.1% and 4.6% of the total dietary calorie intake in the Kingdom Tonga, Solomon Island, and Papua New Guinea, respectively (Coursey, 1975).

Besides their importance as food source, yam also play significant role in the socio-cultural lives of some producing regions like the celebrated New Yam Festival in West Africa, a practice that has also extended to overseas where significant population of tribes form yam growing areas observe it (Narula *et al*., 2007). In some parts of Southeast Nigeria, the meals offered to gods and ancestors consist principally of mashed yam. Yam store relatively longer in comparison with other tropical fresh produce, and therefore stored yam represents stored wealth which can be sold all-year-round by the farmer or marketer. In parts of Igbo land in Southeastern Nigeria, it is customary for the parents of a bride to offer her yam for planting as a resource to assist them in raising a family (Egesi *et al*., 2007).

The *Dioscorea* species have long been cultivated for their medicinal properties (sapogenin steroid, used in production of cortisone and synthetic hormones) in the following order of importance: *D. bulbifera*, *D. cayenesis*, *D. dumetorum*, *D. alata*, *D. trfida*, *D. laxiflora*, and *D. microbotrya* (Pedralli *et al*., 2002). The wide array of saponins and steroidal sapogenins from various cultivars provide the pharmaceutical industry with compounds for oral contraceptives and prevention of diseases such as malaria, yellow fever, skin rashes and dengue (Bhandari and Kawabata, 2004; Lebot, 2009). Wu *et al*. (2005) analyzed the consumption of *D. alata* by post-menopausal women and concluded that although the mechanisms are not yet fully understood, its consumption reduces the risk of breast cancer and cardiovascular disease. Nutritionists stress the importance of yam in treating anemic patients giving their wealth of nutrients, vitamins and minerals (Okwu and Ndu, 2006). Diosgenin can be extracted from *D. bulbifera* (Narula *et al.*, 2007) and can also be used as an anti-tumor agent (Gao *et al*., 2007).

Average daily consumption per capita of yams is highest in Benin (364 kcal), Cote d'Ivoire (342 kcal), Ghana (296 kcal), and Nigeria (258 kcal) (IITA, 2009). A typical analysis of the edible portion of the tubers is: water, 65-73 per cent; protein 1.12-2.78 per cent; fat 0.03-0.27 per cent; carbohydrate 22-29 per cent; fiber, 0.65-1.40 per cent; ash 0.67-2.06 per cent (IITA, 2000). The starch contains a high proportion of fairly large granules: size ranging from 5 to 50 microns have been reported. Unlike most other yam species, starch from *D. alata* has high gel strength. Starch form white-fleshed and purple-fleshed cultivars have similar typical composition averaging: moisture 13.6 per cent; protein 0.14 per cent; ash 0.22 per cent; amylose 21.1 per cent; reducing sugars 0.18 per cent; pH 7.1 (Tetteh and Saakwa, 1991). Ascorbic acid content ranging from 4.9 to 8.2 mg/100 g\ of dibble portion have been reported, while certain cultivars in the South Pacific have been found to contain 6 mg/100 g of carotene. Three anthocyanins have been isolated from *D. alata var*. *atropurpurea* and *rubella* and found to be cyaniding glycosides (Degras, 1993).

Yams are an important source of food security and access to income and thus adds to local food self-sufficiency (FAO, 2012). It serves a critical employment role with thousands of rural families making a living from yam cultivation. Farmers have constantly affirmed that yam cultivation is veriatable source of income.

2.3 World production and trade of yam

Most of the world production of yam is from Africa (about 96%) with Nigeria alone accounting for nearly 64.5% of the total world production; world annual production was estimated to be 25 million metric tons in 1974, and 24 million metric tons in 1992 (Moura-Costa *et al.*, 1993). In 1995, total world production increased from 32.7 million metric tons to 37.5 million metric tons 2000 (IITA, 2009) and 2012 production is estimated at 58.9 million metric tonnes (FAO, 2013). Also, in the period, export quantity declined slightly while export income remained fairly steady (FAOSTAT, 2010). The importance of yam in the economy of the main producing areas, however, appears to be declining due partly to competition with other crops like cassava in Nigeria and Ghana (Opara, 1999; Anaadumba, 2013). During the last four decades, the annual growth rate (%) of per capita production in the major yam zones in Africa has declined (Egesi *et al*., 2007).

2.4 Yam production in Ghana

In Ghana, yam is grown on free draining, sandy and fertile soil after clearing the first fallow. Land is prepared in the form of mound or ridge. The yams recommended for such soil conditions are the white yam or white guinea yam (*D. rotundata*) and water yam or yellow yam (*D. alata*). Planting is done by seed yam or cut setts from ware tubers (CCPGS, 2011). The setts are planted at an interval of 15-20 cm (5.9-7.9 inch) with the cut surface facing up. Mulching is essential during October-November with dry grass or plant debris weighed down with balls of mud. Dosage of fertilizer application, as essential, is decided after chemical analysis of the soil samples. Manual weeding by hoeing is done three or four times depending on rate of weed growth (Dadzie, 2004).

Ghana is the second largest producer of yams after Nigeria (FAOSTAT, 2013). In Ghana, during 1997 and 1998, the crop was ranked second in importance (in terms of tonnage) after cassava and was the most important crop in terms of value (Dadzie, 2004). A survey by GTZ in the Northern Region of Ghana identified yams as the most important cash and food crop in that region, followed by groundnuts, cassava and, maize (MoFA-SRID, 2011). Reasonable production, however, occurs in almost all regions except Upper East and Greater Accra region (Fig. 2).

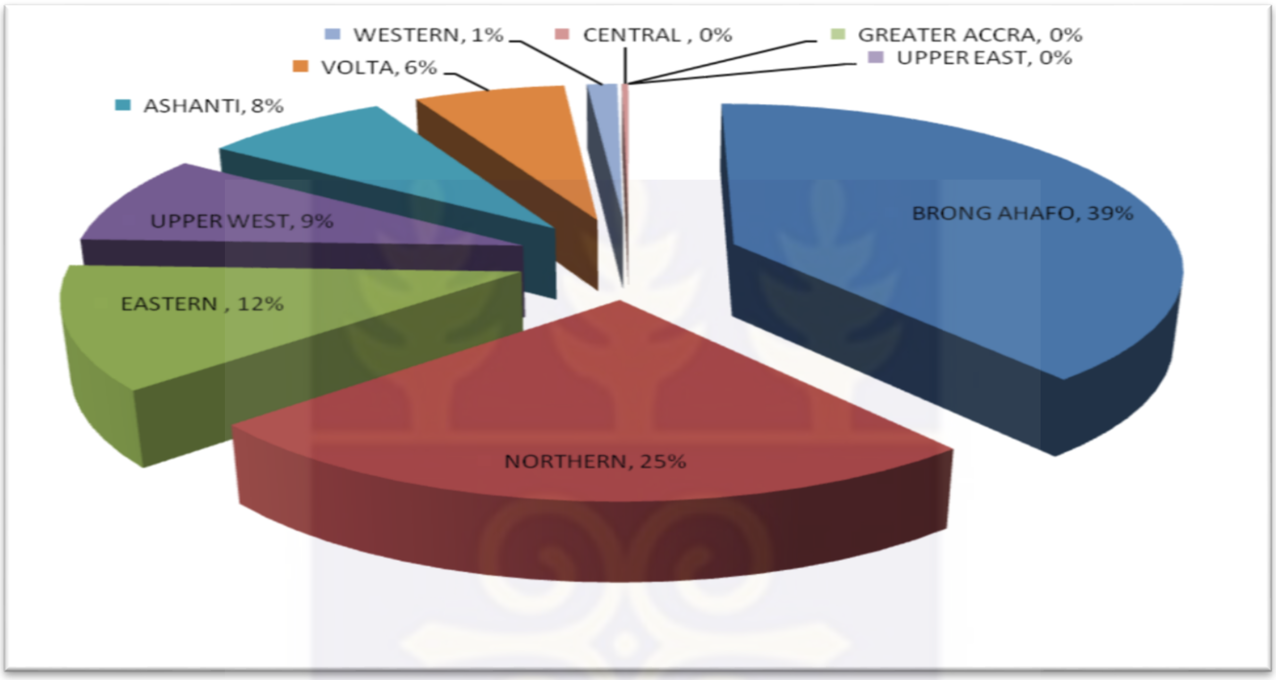


Figure 2. Distribution of Yam Production in Ghana by Region, 2010. Source: MOFA, SRID (2011)

Yams are almost entirely vegetatively propagated by planting pieces of tubers, or setts or whole small tubers. They can be planted 7 to 9 cm deep in 1.2 m rows with plants spaced 60 cm apart, or in hill plantings 1 m apart.

Traditionally in Ghana, farmers obtain their planting materials either from their own farms, or by buying the surplus from neighbouring farms (Kwoseh *et al.*, 2005). This means that the planting material is often of low quality, infected with nematode pathogens, virus and/or fungus and may be relatively expensive (Tetteh and Saakwa, 1991). In order to ensure the survival and growth of the planting materials, fairly large pieces of seed yam (220 g) are used which adds to the cost and results in a very low multiplication rate. Inter-cropping is the most common practice, with the distance between yam plants determined by the number and types of the other crops in the field. Where sole cropping is done, rows are 1 m apart, with intra-row spacing of 50-100 cm (Dadzie, 2004).

2.5 Constraints of yam production

Several factors hinder yam production. Some of these major factors include, limited availability and high cost of planting materials, diseases and pests, high labour costs, poor soil fertility and drought.

2. 5.1 Nematodes pests of yam

Many different nematode species have been found to be associated with yams. The nematodes of particular importance are endoparasites of roots and tubers. Those known to cause serious damage by mainly reducing tuber yield and quality are, the yam nematode (*Scutellonema bradys),* lesion nematode (*Pratylenchus* spp.*)*, and root knot nematode (*Meloidogyne* spp.) (Bridge, J and Page, S. L. J, 1984; Bridge *et al.*, 2005).

Scutellonema bradys the causal agent of dry rot is amongst the most important nematode pest of yam (Coyne *et al.*, 2006). Outside of West Africa, dry rot is however largely caused by the lesion nematode *Pratylenchus coffeae* (Bridge *et al.*, 2005). The yam nematode and *Meloidogyne* spp. were recorded in 2.84% and 2.94% of yam tubers respectively on marketed yam tubers in West African yam markets (Coyne *et al.*, 2006). Similar obsevations were recorded in Ghana by Kwoseh *et al.*, (2005).

The most severe symptoms of dry rot are seen in mature tubers, during storage, when it is associated with general decay. Symptoms begin with light yellow or brown lesions below the outer skin, which gradually progress deeper into the tuber as the nematodes feed and multiply causing infected tissues to turn from brown to black. The outer cortex may crack and flake exposing the necrotic tissue, but may also remain intact presenting the appearance of an otherwise healthy tuber. These symptoms are more severe as the length of storage increses (Coyne *et al.*, 2006; Baimey *et al.*, 2009). There is little doubt that the disease is highly debilitating to yam production (Bridge, 1982; Bridge *et al.*, 2005). *Scutellonema. bradys* has, in addition to other pests and pathogens, been related to the increasing scarcity of seed material, particularly healthy material (Green and Florini, 1996). As a vegetatively propagated crop, untreated material used for planting will perpetuate the disease cycle and a continued decline in quality.

In addition to *S. bradys*, root-knot nematodes (*Meloidogyne* spp.) also adversely affect yams (Hemeng, 1978; Bridge *et al.*, 2005). *Meloidogyne* spp. so far identified to be associated with yam include, *M. incognita, M. javanica, M. hapla* and *M. arenaria* and of all, *M. incognita* has been observed to be the most important (Bridge *et al.*, 2005). Root-knot nematodes have been reported on yam in Africa (Ghana, Côte d'Ivoire and Nigeria), the Caribbean (Jamaica, Martinique, Puerto Rico and Trinidad), Pacific (Fiji, Kiribati, Papua New Guinea and Western Samoa), Brazil, Guatemala and Japan (Osei *et al*., 2004; Kwoseh *et al.*, 2005 and Bridge *et al.*, 2005). Storage losses of up to 25% to 75% due to *Meloidogyne* spp infection on yam tubers stored within a period of 16 weeks have been reported in Nigeria (Nwauzor and Fawole, 1981). Infection of yams by root-knot nematodes results in an unsightly, warty appearance of the tubers, affecting marketability. In storage, affected tubers reputedly desiccate and lose weight much more quickly than unaffected tubers (Bridge *et al*., 2005). Light to heavy galling and knobbly tuber surfaces often covered with excessive rooting are some of the disease symptoms caused by *Meloidogyne* species. The life cycle of *M. incognita* in *D. rotundata* or *D. alata* tubers is 35 days (Nwauzor and Fawole, 1981). *Meloidogyne* spp. are found in relatively deep layers (2-6 mm) of *D. alata* and *D. rotundata* tubers

(Nwauzor and Fawole, 1981). The proportion of yam with galled tubers collected from yam barns and markets in Nigeria can be as high as 90 % for *D. alata* and 70 % for *D. rotundata* (Adesiyan and Odihirin, 1978). It is estimated that there is a reduction of 39-52 % in the price of galled tubers compared to healthy ones (Nwauzor and Fawole, 1981).

Pratylechus spp like *S. bradys* is migratory endoparasite usually infecting roots and underground stems and tubers. It has been reported as pest of *Dioscorea* species in all the major yam producing countries in West Africa (Ghana, Nigeria, Cote d'Ivore and Benin), Jamaica, Costa Rica, Puerto Rico and India (Osei *et al.*, 2004; Coyne *et al.*, 2006 Baimy *et al.*, 2006; Humphreys *et al.*, 2014; and Osei *et al*., 2013). *Pratylenchus* spp, has a life cycle of 3-4 weeks on yam tissues and also cause dry rot of surface and sub surface regions (Thompson *et al.*, 1973). In Jamaica, 67-100% of *D. rotundata* and *D. cayenensis* tubers were found to be infected with *Pratylenchus* spp (Thompson *et al.*, 1973).

Pratylenchus sudanensis has been reported on yam in Uganda (Coyne *et al.*, 2003) and was also confirmed as the most dorminant nematode on yam in that country (Mudiope *et al.*, 2004). *P. sudanensis* is morphologically similar to *P. pseudopratensis* (Coyne *et al*., 2003) and associated with symptoms such as cracked tubers (Mudiope *et al.*, 2001). Host range studies in Sudan reveals that it has a wide host range including, sorghum, cotton and pigeon pea (Saadabi, 1985).

2.5.2 Disease complexes of yam

Average yield losses of yams due to diseases and pest has been estimated to be over 25% annually (Ezeh, 1998; FAO, 2013). Lesions caused to yam tubers by nematodes facilitate invasion by fungi and other micro-organisms. The resulting complex often destroys the entire tuber in the ground and during storage (Bridge, 1982). The more extensive, internal decay of tubers known as "wet rot", "soft rot" or "watery rot" is associated with fungal and bacterial pathogens (Adeniji, 1970; Amusa *et al.*, 2003). Nematodes and fungi are found together in the transitional stage between dry rot and wet rot but nematodes do not occur in the "late stage" deep in the tubers (Adesiyan *et al.*, 1975). Another complex associated with serious losses in yam production is the occurrence of nematodes especially in soils of poor fertility. According to Adeniji (1970), in soils of low fertility, yam plants are weak, nematode attacks increase and yam yields decrease.

2.6 The yam nematode, *Scutellonema bradys* (Steiner and LeHew) Andrassy.

2.6.1 Taxonomy and nomenclature

The first description of the yam nematode was in 1933 from infected yam tubers in Jamaica. It was classified into the genus *hoplolaimus* (Steiner and Le Hew, 1933). *S. bradys* was redescribed from *hoplolaimus* to *Rotylenchus* by Goodey, (1952) and later redescribed to *Scutellonema* by Andrassy, (1958) (Morgan, 1971).

Scheme of classification:

Kingdom: Metazoa
Phylum: Nemata Cobb, 1919
Class: Secernentea von Loinstow, 1905
Subclass: Diplogasteria Maggenti, 1982
Order: Tylenchida Thorne, 1949
Suborder: Tylenchina Chitwood, 1950
Superfamily: Tylenchoidea Orley, 1880
Family: Hoplolaimidae (Filip'ev, 1934)
Subfamily: Hoplolaiminae Filip'ev, 1934
Genus: *Scutellonema* Andrássy, 1958
Species: *Scutellonema bradys* (Steiner and LeHew, 1933),Andrassy, 1958
Source: Morgan (1971).

2.6.2 Biology and ecology

The reproduction of *Scutellonema bradys* is amphimictic and eggs are laid in the soil or roots and tubers (Tchabi, 2008). Juveniles develop into mature male or female by moulting in stages within 21 days while all active stages are infective (Kwoseh and Krapa, 2008). Populations build up in the tubers with a maximum of 62000 nematodes/ 10 g of tuber peels has been recorded in Nigeria (Bridge, 1972). *S. bradys* population is affected by storage conditions with maximum populations of 6200 nematodes/ 10 g of yam tuber peels recorded in Nigeria (Bridge, 1982). The nematode invade primary roots through their growing tips and tuber and can enter developing tubers through their growing points. Invasion of tubers can also occur through cracks or damaged areas in the epidermis (Bridge, 1972, Coyne *et al.*, 2011). *S. bradys* feeds intracellularly in the tuber tissues resulting in rupturing of cell walls, loss of cell contents and formation of cavities (Bridge, 1982; 2005).

2.6.3 Importance of *Scutellonema bradys* in Yam Production

Scutellonema. bradys is a nematode of major economic importance in all yam growing regions in West Africa major economically important nematode pest of yam in West Africa, but not necessarily of other yam-growing regions of the world (Bridge *et al.*, 2005). *S. bradys* has been recorded on yam from West Africa (Ghana, Benin, Cameroon, Côte d'Ivoire, Nigeria, Senegal, Togo), the Caribbean (Cuba, Jamaica, Puerto Rico), Brazil and India (Bridge, 1982 and Bridge *et. al.*, 2005). Most edible yam species commonly grown in West Africa are susceptible to nematode infection (Coyne *et al.*, 2006). The primary importance of *Scutellonema bradys* is in the direct damage it causes to the tubers, but the relationships between this damage and loss in total yield is difficult to determine (Coyne *et al.*, 2006). Dry rot of yams alone causes a marked reduction in the quality, marketable value and edible portions of tubers, and these reductions are more severe in stored yams (Coyne *et al.*, 2006). When dry rot is followed by wet rot in stored yams, losses of whole tubers can be as high as 80–100% (Adesiyan and Odihirin, 1975, and Baimey *et al.*, 2009), but losses certainly increase with duration of storage. The degree of preharvest damage to tubers by *S. bradys* varied from 0 to 40% in Nigeria (Wood *et al*., 1980). About 46.6% of IITA yam germplasm screened were naturally infested with *S. bradys* (Kwoseh, 2000). Almost 47% of all tubers on sale in Nigerian markets were infested with *S. bradys* (Bridge, 1972), and both dry rot and wet rot diseases of tubers have been observed in all Nigerian yam barns and markets sampled (Adesiyan and Odihirin, 1977).

In Ghana, *Scutellonema bradys* was first reported in 1970 (Addoh, 1971). Following a survey of plant-parasitic nematodes associated with yam in the Guinea savanna and Transitional forest ecological zones of Ghana, Kwoseh *et al.,* (2005) found that 100% of the yam tubers in stores (all cultivar groups) were infected with *S. bradys*, whereas *M. incognita* (83.3%) for the Guinea savanna ecological zone whiles in the Transitional forest zone, *M. incognita* occurred in yam tubers in market stores was 72.2% and *S. bradys* was 28.6%.

2.6.4 Life Cycle of the Yam Nematode

The yam nematode (*Scutellonema bradys*) is a migratory endoparasite present in yam soils, roots and tubers. *S. bradys* is a vermiform nematode when mature. All active stages are infective (J2 – J4), thus invading the young developing tubers at the tuber growing points, alongside young developing roots and shoots and also through cracks or damaged areas in the tuber skin (Bridge, 1972; Coyne *et al*., 2011).

The yam nematode feed intracellularly in tuber tissues, resulting in rupture of cell walls, loss of cell contents and the formation of cavities (Bridge, 1972; Adesiyan *et al.*, 1975). They are mainly confined to the subdermal, peridermal and underlying parenchymatous tissues in the outer 1–2 cm of tuber. *S. bradys* continues to feed and reproduce in yams stored after harvesting. The yam nematode feed by sucking out ceels of the plant with their stylet and moving ahead leaving the lesions behind. They make no permanent feeding sites. The life cycle of *S. bradys* consist of hatching nematode eggs into second stage juveniles which immediately begins feeding on the plant cells. The nematode feed, molt and reproduce primarily within the plant tissue. All motile stages are capable of feeding from the plant and they are able to move into the soil in search of new roots to invade. Because these nematodes create extensive wounds in the plant root and tubers, secondary infection by bacteria and fungi can often occur, further damaging the root and tuber system. *S. bradys* may feed on external surfaces of roots but generally they burrow into the root to feed on internal root cortex. This tunneling results in considerable damage as the nematodes move from one feeding site to another. While some eggs are laid in the soil, most are laid inside roots and tubers. Eggs may hatch inside the root or remain until the root decays and the eggs are released into the soil. As the nematode develops in the egg, it molts to change from a first stage juvenile to a second stage juvenile (J2) which then hatches from the egg. The nematode grows and molts three more times to become a mature male or female. During periods of unfavourable weather conditions such as drought, the nematodes may remain quiescent (anhydrobiosis) until moisture increases and plants resume growth.

2.6.5 Symptoms and diagnostics

Scutellonema. bradys causes a major disease of yam *(Dioscorea* spp.) tubers known as 'dry rot disease' (Bridge *et al*., 2005; Coyne *et al*., 2006)). Dry rot of yams occurs in the outer 1 to 2 cm of tubers showing symptoms of *S. bradys* infection. The initial stage of dry rot consists of cream and light-yellow lesions below the outer skin of the tuber with no external symptoms at this stage

(Baimey, 2006; Humphreys *et al.,* 2014). As the disease progresses it spreads into the tuber, normally to a maximum depth of 2 cm but sometimes deeper. In these later stages of dry rot, infected tissues first become light brown and then turn dark brown to black. External cracks appear in the skin of the tubers and parts can flake off exposing patches of dark brown, dry rot tissues (Fig. 3A) and depending on the nematode species, galls and knots may be formed on the roots and tubers (Fig. 3B). The most severe symptoms of dry rot are seen in mature tubers especially during storage when it is often associated with general decay of tubers. No foliar symptoms have been observed on yams growing in soil infested with *S. bradys* (Bridge, 1994).

Figure 3: Yam tuber showing symptoms of *Meloidogyne* spp.infection (A); Yam tuber showing symptoms of *Scutellonema bradys* infection (B).

2.6.6 Host Range and distribution

The yam nematode has a wide range of distribution especially across the yam growing areas in West Africa (Coyne *et al.*, 2006). It has also been reported in Cuba, Jamaica, Brazil, India, Pueto Rico and Costa Rica (Bridge *et al.*, 2005; Humphreys *et al.*, 2010). All edible *Dioscorea* spp. are susceptible to to nematode infection. Two wild *Dioscorea* spp. growing in the forests of Nigeria have been reported as natural host of the yam nematode (Bridge *et al.*, 2005). Other crops such as yam bean (*Pachyrrhizus erosus*), green gram (*Phaseolus aureus*), pigeon pea (*Cajanus cajan*), kenaf (*Hibiscus cannabinus*), okra (*Hibiscus esculentus*), tomato (*S. lycopersicon*), sorghum (*Sorghum vulgare*) have been reported to support low population of *S. bradys* in the soil (Adesiyan, 1976; Kwoseh and Krappa, 2008). They also occurs in cassava (*Mannihot esculentan*). Other crops such as cowpea (*Vigna unguiculata*), melon (*Cucurbita melo*) and sesame (*Sesamum indicum*) in addition to yams have been found to actually increase populations of the nematode (Jatala and Bridge, 1990., Kwoseh and Krapa, 2008; Osei *et al*, 2011). These alternative hosts permit the yam nematode to survive in soil in the absence of yams.

2.7.1 Morphological Identification

The Phylum Nematoda is highly diverse in terms of species richness and one of the most abundant metazoan groups on earth. It is estimated that nematodes comprise nearly 90% of all multicellular organisms. Furthermore, Lambshead (1993) predicted the number of nematode species in marine habitats to be as high as one hundred million, although only 26,646 species have been described so far from all habitats (Hugot *et al.*, 2001).

In the taxonomy of *Scutellonema* species identification has been based primarily on light microscope observations (morphology) and measurements of morphological characters of mainly females and males (Fortuner, 1989; Siddiqi, 2000). Morphological resemblances also formed the basis for evolutionary systematics (Coomans, 2000). But the light microscope has its limitations and the use of scanning electron microscope has considerably improved the accuracy of these observations and descriptions of especially small morphological characters (Nguyen, 2010). The genus *Scutellonema* has about 46 valid species and the small number diagnostic morphological characters at species level and the overlapping of morphological characters among species is making it difficult to separate the species (Siddiqi, 2000; Van de Berg *et al.*, 2013). In order to aid accurate morphological identification (traditional identification aids), various dichotomous keys have been designed. Some of these include; Filipev, (1936); Chitwood, (1951); Siddiqi, (1972a, 1986), Andrassy, (1976); Fortuner, (1989) and Siddiqi, (2000).

The morphological and morphometric characters most commonly used to separate *Scutellonema* species are;

Total body length	Lip region height
Spermatheca width	Lip region diameter
Position of vulva to anterior	Lip annule shape
DGO	Pharynx length
Diameter at anus	Diameter at head region
Median bulb valve length	Diameter at mid body
Median bulb vave width	Diameter at tail region
Scutellum width	Tail length
Scutellum height	Stylet length
Diameter at anus	The de man's ratios (a, b, b’, c, c’, s)

Sources: Siddiqi, 2000; Van de Berg *et al*., 2013

2.8. Nematode disease management options

Most nematode species do not cause economically significant damage, except if they are present in the soil and tissues in high populations or the plant is highly stressed. Thus, any nematode management strategy should aim at maintaining nematode population density under threshold levels. Host plants may either suppress (i.e. resistance) or allow (i.e. susceptibility) nematode development and reproduction. However, they may suffer only little injury (i.e. tolerance) even when heavily infected with nematodes (Bos and Parlevliet, 1995). In order to prevent and control nematode infestation, the management of most plant parasitic nematodes can be achieved by one, or preferably, a combination of the following several measures;

2.8.1 Phytosanitary and clean planting materials

Phytosanitary is the first method used for nematode management on yam. This is done by separating infested tubers before storage and planting in order to prevent establishment of nematode infestation in the field. Healthy planting materials can also be obtained by using tissue-cultured planting material (Dropkin, 1980; Speijer *et al*., 2000). In yam, use of nematode-free planting material is a practical and economic means of preventing damage by *S. bradys* and their dissemination. A number of yam species, such as *D. alata*, *D. rotundata* and *D. dumetorum*, can also be produced from vine cuttings (Coursey, 1967). Although these methods of propagation are not practical for producing yam tubers, they can be used to produce nematode-free seed tubers (IITA, 2005). The use of 'microsetts' or 'minisetts', cut from mature tubers (IITA, 2007) can be used to provide clean planting material if the mother seed yam tubers selected is free of nematodes.

2.8.2 Agronomic practices in the field

Agronomic practices such as fallow, crop rotation with non-host or cover crops are an efficient for managing nematode infestation in the field. A fallow of 12 to 24 months as well as crop rotation with non-hosts, can reduce the nematode population in the soil (Adesiyan, 1976). However, high land pressure coupled with the intensification of yam and its component crops cultivation have rendered the fallow method inapplicable in most yam growing regions (Kwoseh and Krappa, 2008). When it is practical, rotation of yam with non-hosts or poor hosts such as groundnut (*Arachis hypogea*), tobacco (*Nicotiana tabacum*) or cotton will limit damage by *S. bradys*. However, crop rotation to effectively reduce *S. bradys* populations seems to be non-realistic because of its absence or low density in the soil (Tchabi, 2008).

Mucuna spp., *Crotalaria* spp. and *Tithonia diversolifia* cropped as cover crops have been reported to reduce soil nematode populations, particularly *Meloidogyne* spp. and *S. bradys* (Claudius-Cole *et al.,* 2004; Garrido *et al.,* 2008; Osei *et al.*, 2011).

Other cover crops such as *Tagetes* spp., *Stylosanthes* spp., *Centrosema* spp. and *Aspilia* spp. have been recommended to lower nematode populations and restore fertility for yam production in Nigeria (Atu *et al.,* 1984). Results from use of cover crops in a crop rotation have shown that they are not efficient managing nematodes especially, *Meloidogyne* spp. since these nematodes have a wide host range; moreover, as these crops are not edible, farmers are not enthused about using them as a rotation crop (Onalo *et al*., 2001). Superphosphate fertilizers were reported to reduce *S. bradys* populations in tubers of *D. alata* to a low level, but did not increase yam yield (Baimey *et al.*, 2006). In contrast, N (Nitrogen) alone was reported to increase both populations of *S. bradys* and also increase yields and the percentage of the infested tubers of *D. rotundata*, whereas P (Phosphorus) alone can decrease the percentage of infested tubers (Yahaya, 2011). In addition, the traditional practice of using wood ash on yam tubers before planting is reported to decrease nematode numbers (Adesiyan and Adeniji, 1976; Diby *et al.,* 2004).

2.8.3 Biological control

Biological control is considered to be an alternative to nematicides, especially concerning the environmental and health risks associated with the use of these chemicals (Viaenne *et al.,* 2003). Integrated crop pest control methods may benefit from studying plant-parasitic nematodes and natural antagonistic interactions in natural systems, which have co-evolved for longer than crop nematode-antagonist systems. Understanding how wild plants manage their association (plant parasitic nematodes) may ultimately result in improving the sustainability of crop protection against plant-parasitic nematodes. Current research has focused mainly on predacious and .parasitic micro-organisms. Nematophagous fungi such as *Pochonia chlamydosporium* and *Paecilomyces lilacinus* are nematode parasites (Gaspard *et al.,* 1990). Bacteria such as *Pasteuria penetrans* also appear promising biological control agents of *Meloidogyne* spp. (Pembroke *et al*., 1998). Recently, possible anti-nematode effects of the micro in the rhizosphere have been studied. Rhizobacteria such as *Rhizobium* spp. and *Pseudomonas* spp. not only have a positive effect on plants by promoting their growth, but in addition they show a repellent effect towards nematodes (Aalten *et al.,* 1998; Hallman *et al*., 2001).

2.8.4 Use of nematicides

Nematicides (carbofuran and organophosphates) can be used to successfully manage nematodes on yam, but these chemicals are very expensive and also toxic against non-target organisms, including users. They are poorly biodegradable and, therefore present an important ecological risk.

Application of furadan at rates of 100 kg/ha results in optimum yields and significant reduction in the incidence and severity of nematodes infection on yam in the field (Adegbite and Agbage, 2007) but information on the economics of this means of control is lacking for large-scale use. Nematicide treatment of planting material however, to generate healthy seed yam free of nematodes can have a major effect and proved to be highly economical (IITA, 2006).

2.8.5 Physical control or hot water treatment

Hot water treatment is done by heating water to a temperature of 50-55°C and dipping the tubers in the water for 45 min. This ensures better management of *S. bradys* without damaging tubers (IITA, 2005). Notwithstanding the effectiveness of the method, it is usually not pratical for farmers, due to the need for temperature control, labour and fuel, and more importantly due to the huge volume of yam material needed to be treated. Other factors, such as yam species, nematode densities, depth of infestation and age of tubers also have negative impacts on the efficiency of this method (Bridge *et al.,* 2005). It is however, feasible for small-scale operations and for establishing nematode-free planting material for research.

2.8.6 Resistance

There has not been any reported source of resistance to the yam nematode *S. bradys* in cultivated yam (IITA, 2004). Noncultivated yams are likely to present nematodes resistance sources but breeding this into cultivated yam lines will be a lengthy process and not necessarily successful due to the complex nature of yam botany (Coursey, 1967). Resistance could however prove to be the most economic and practical means of managing *S. bradys* as a way of reducing losses to such pests found in commercially acceptable varieties (Brandon, 2001). Crop resistance is not as common against the migratory endoparasitic nematodes however, such as *S. bradys*, as it is against the sedentary endoparasites with specialized feeding sites, such as *Meloidogyne* spp., *Globodera* spp., *Heterodera* spp., *Rotylenchulus* spp. and *Tylenchulus* spp. One cultivar of yellow yam, *D. Esculenta*, and one of *D. dumetorum* have shown some resistance to *S. bradys* (Bridge, 1987; Kwoseh, 2000, Tchabi, 2008).

2.8.7 Integrated pest management

A combination of several methods is the best way to control nematodes but is not usually applied by farmers for managing nematodes. For example, combinations of hot water treatment and phytosanitary measures as well as crop rotations were experimentally used in IITA-Ibadan, West Africa for successful yam nematode control (IITA, 2005).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Survey on nematodes associated with yam and assessment of farmers' knowledge and perceptions on occurrence, management and economic importance of nematodes associated with yam.

3.1.1 Selection of experimental sites within the Guinea savanna and transitional agro-ecological zones for survey on nematodes associated with yam

Ten communities from ten districts (Yendi, Karaga, East Gonja, Kumbungu, Savelugu/ Nanton, Kintampo North, Techiman, Wenchi, Atebubu/Amanten and Ejura/Sekyeredumase) were purposefully selected from the two leading yam producing ecological zones in Ghana. The communities were stepwisely selected by first obtaining a list of major yam producing communities in the districts from the Agricultural Extension Agents (AEAs) working in the ten districts. Initial focus group discussions were held with the AEAs in each district to determine the communities in their respective districts where the occurrence and prevalence of nematodes associated with yam was predominant. Six yam farmers per community were selected from the list of yam farmers provided by the AEAs. Survey sites, location, ecological zones and number of farmers selected for the survey are presented in Table 1.

3.1.2 Survey to assess farmers' knowledge, perception and experiences concerning occurrence, management and economic importance of nematodes associated with yam

A field survey was conducted between December and January, 2015 to obtain baseline data on farmers' knowledge, perceptions, experiences concerning prevalence, management and economic

Table 1. Regions, districts and communities selected for the questionaire survey, collection of soil and tuber sample and the number of respondents involved.

Locality	**District**	**Region**	**Location**	**Vegetation**	**No of respondents**
Choo	Yendi	Northern	N 09° 19’ 44.1’’ W 000° 00´ 09.7´´	Guinea Savannah	6
Shebo	Karaga	Northern	N 09° 55´ 12.6´´ W 000° 23´ 00.5´´	Guinea Savannah	6
Kpachi	Kumbungu	Northern	N 09° 25´ 44.1´´ W 000° 58´ 37.4´´	Guinea Savannah	6
Kaswurape	East Gonja	Northern	N 08° 41´ 39.0´´ W 000° 32´ 44.9´´	Guinea Savannah	6
Kanshegu	Savelugu/Nanton	Northern	N 09° 34´ 31.5´´ W 000° 49´ 17.0´´	Guinea Savannah	6
Promposo	Kintampo South	Brong Ahafo	N 07° 40´ 42.4´´ W 002° 39´ 52.0´´	Transitional Forest	6
Ayayo	Wenchi	Brong Ahafo	N 07° 36´ 45.1´´ W 002° 08´ 08.0´´	Transitional Forest	6
Atrensu	Techiman	Brong Ahafo	N 07° 37´ 55.5´´ W002° 03´ 04.0´´	Transitional Forest	6
Afrefreso	Atebubu/ Amanten	Brong Ahafo	N 07° 40´ 34.5´´ W 001° 02´ 01.5´´	Transitional Forest	6
Nokwarease	Ejura/Sekyeredumase	Ashanti	N 07^{0} 21` 24.4`` W 001$^{0}$ 18` 19.7``	Transitional Forest	6

importance of nematodes associated with yam in the Guinea savanna and the Transitional forest ecological zones of Ghana. Sixty (60) yam farmers (from both smallholders with ≤ 1 ha of yam and large scale growers with ≥ 10 ha of yam) were randomly chosen from the experimental sites in the two agro - ecological zones. Semi-structured questionnaires were designed, pretested and administered to farmers (read and interpreted to farmers) and their responses documented. The areas covered by the questionnaire included the demographical information of the farmers; their land use intensity, knowledge, perception and experiences concerning occurrence, control and the socioeconomic impact of nematodes on yam production in their localities (Appendix 1). Pictures (Appendix 2) were included to facilitate responses from farmers.

Data from questionnaire was anlysed using descriptive statistics (percentages, frequencies and tables) in statistical package for social scientists (SPSS) version 20.

3.1.3 Sampling of soil and yam tubers during the rainy season for nematode extraction

Sampling was undertaken in the months of August to September, 2014. These months corresponded with the yam maturity periods in the Agro ecological zones sampled. Five sites around the yam tubers were sampled from a farm randomly within each district. The soil samples were thoroughly mixed together and 200 cubic centimeter used for nematode extraction. Three yam tubers were also randomly selected from each field that exhibited symptoms of nematode parasitism. The tubers together with soil sub-samples were placed into plastic bags, sealed, and labelled accordingly, placed in a thermo cooler and transported to the Plant Pathology Laboratory of Crop Science Department, University of Ghana and stored at 4º C until processing. The GPS coordinates of location of farms sampled were recorded.

3.1.4 Sampling of yam tubers in storage

This was also done in the months February and March, 2015. The period also corresponded with the dry season agro ecological zones and harvested yam tubers were in storage. Yam tubers showing symptoms of *S. bradys* infection were sampled from all ten localities in the two AEZs. Five yam tubers were sampled from each locality. The tubers were packaged and well labelled before transporting them to Plant Pathology Laboratory of the Department of Crop Science, University of Ghana for nematode extraction.

3.2. Extraction, Identification and quantification of nematodes from soil and yam tubers

3.2.1 Extraction of nematodes from the soil

The various soil samples from each farm were thoroughly mixed and a 200 cubic centimeter (cc) was used in nematode extractions.

Extraction from the soil was done by sieving and sucrose centrifugation method (Jenkins, 1964). The sucrose solution was prepared by dissolving 454 g of sugar in 1 liter of distilled water. Soil sample was mixed thoroughly and passed through coarse sieve to remove rocks, roots and other debris. A 200 cc subsample of soil was taken into a beaker and 500 mL water added and stirred thoroughly and then given enough time (5 minutes) to settle. The supernatant was poured through a 90 µm mesh on a 36 µm sieve. A spray water bottle was used to gently wash the nematodes into centrifuge tubes. The centrifuge tubes were spun at 1700 rpm for five minutes in a thermo scientific Jouan MR 23 I centrifuge (made in Germany). After centrifugation and settling of the solution for a further five minutes, the supernatant was decanted, leaving approximately 1 cm above the pellet. The tubes were refilled with sucrose solution at room temperature and stirred with spatula to break the pellets. The samples were spun again in the centrifuge at 1000 rpm for one minute. The supernatant was poured through a 36 µm mesh – sieve and rinsed gently with water in a spray bottle and transferred into graduated labelled vials. The aliquots are concentrated to 10 mL in each labelled vial.

3.2.2 Nematode extraction from yam tubers.

Yam tubers with symptoms of dry rot were randomly collected to determine *S. bradys* population densities in these tubers in the field during the rainy season, and in storage during the dry season. The tubers were manually peeled using a kitchen peeler and the peel chopped (0.5 cm x 0.3 cm) and nematodes were extracted from 10 g sub-samples of peel per tuber using a modified Baermann method for 48 h (Hooper, 1990) (Fig. 4). The nematode suspension was collected in beakers, and settled for two hours, afterwards the supernatant was poured through a 90 µm mesh on a 36 µm sieve. A spray water bottle was used to gently wash the nematodes into centrifuge tubes to a final volume of 10 mL Nematodes densities were assessed in a counting dish using exacta 5BP light microscope (EXACTA OPTECH®, Munchen- Germany) and the mean nematode density calculated per unit peel (outer cortex) weight for each tuber sampled.

Figure 4. Setup of the Baerman funnel method of extracting nematodes from yam tuber peels

3.2.3 Handling and processing of nematodes for identification

To facilitate identification, nematode suspension was further concentrated to 10 mL. The nematode suspension was shaken and small batches of 2 ml nematodes were sucked using a shringe and transferred to a Hawksley counting dish (Fig. 5). Nematodes for identification were selected randomly from the counting dish using an eye lash attached to a needle. The process was repeated five times and the obtained mean was used to estimate the total plant parasitic nematodes per sample. The selected nematodes were mounted on a drop of water on a microscopic slide and placed on a hot plate at 60°C (Hooper, 1997) until nematode suddenly straightened out. The specimens were then examined under an illuminated stereomicroscope (Leica zoom 2000™. Buffalo – NY, USA) and identified to genera level using their morphological features.

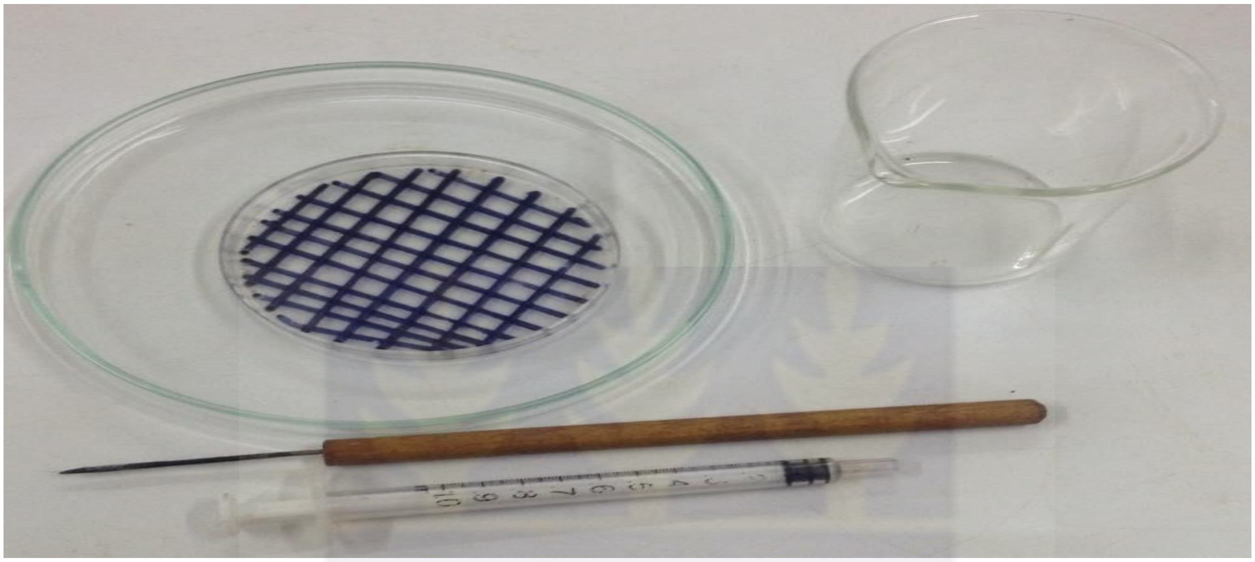


Figure 5. Shring, counting tray, beaker and a needle for handling nematodes

Extracted nematodes were examined directly under a compound microscope. The specimens were collected in a drop of water in the center of a clean glass slide. Fixative (4% Formalin with 1% glycerol -FG) was heated to about 70° C and an excess 2-3 ml, was quickly added to the specimens to fix and kill the nematodes in one process (Seinhorst, 1966). The fixed nematodes were immediately covered with coverslips and sealed with nail polish. They were viewed under the compound light microscope (Exacta – Optech® Biostar B5P; Munchen- Germany) at a magnification of 200x. Nematodes were concentrated in glass vial after settling and excess water pipette. Nematodes were then cleared with glycerol (Hooper, 1990) before being mounted on a slide and viewed under a compound microscope. Fixed specimens were then concentrated in glass vials and stored for further identification.

3.2.4 Nematode identification

Nematodes were identified to genera level through morphological features as described by Sher, (1964, 1965); Siddiqi (1972, 1989); Luc, *et al*., (1990); Siddiqi, (2000); Hallan, (2007) and the University of Nebraska Lincoln nematode identification website (http://nematode.unl.edu/konzlistbutt.htm).

3.2.5 Quantification of nematodes

The suspension of nematodes was shaken gently and 5 mL was sucked from the suspension with a shringe and poured into an open 50 mm plastic counting dish and place under a microscope (exacta optech® B5P, Munchen- Germany) magnification 100x. A hand tally counter was used for counting nematodes. Counting was repeated three times to obtain the mean which was used in calculating the total nematodes in a sample suspension. The nematodes enumerated were expressed as number of nematodes in 200 cc of dry soil, and 10 g of yam tuber peels.

3.3 Morphometric measurements

Ten female *S. bradys* and five male *S. bradys* were pooled from each population for morphometric measurement. Thirteen morphometric variables and four de Man's indeces were measured in total.The morphometric variables that were measured were; Total body length, Stylet length, Tail length, Stylet knob height, Stylet knob width, Diameter at head region, Diameter at tail region, Diameter at mid body, Postion of vulva to anterior, Spicule length, Scutellum height, Scutellum width, Lip region height and Lip region diameter. The four de Man's ratios that calculated include; ratio a = body length/diameter at mid body, ratio 'c' = body length/ tail length, ratio 'c^1' = tail length/ tail diameter and ratio 's' = Stylet length/diameter at head region (de Man, 1876).

Measurements were done with scope image 9.0 professional software installed in a computer and connected to a binocular compound light microscope (Fig. 6)

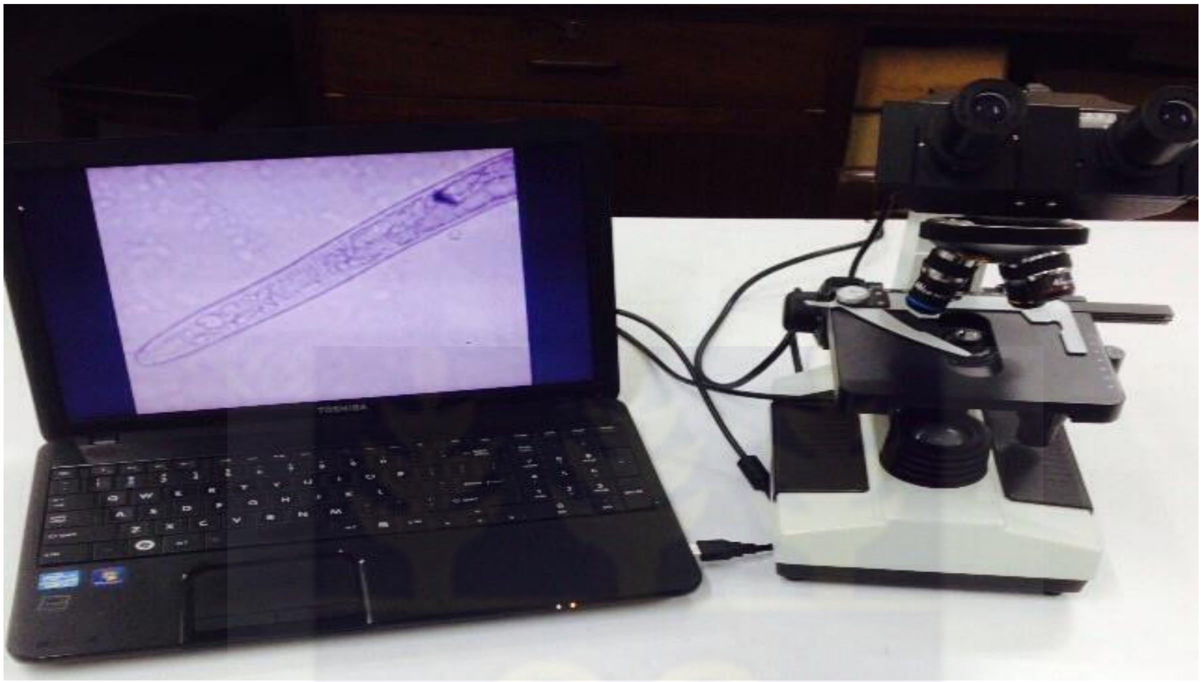


Figure 6. Binocular compound light microscope connected to a computer with scope image 9.0 professional imaging software

3.4 Multivariate principal component analysis of *Scutellonema bradys*

A multivariate principal component analysis was performed on both female and male *S. brabys* species from the nine populations to determine morphometric discrimination among the species. The analysis was based on the following characters: body length, stylet length, diameter at mid body, tail length, lip region diameter, lip region height, diameter at head region, diameter at tail region, stylet knob height, stylet knob width, scutellum height, scutellum width, position of vulva to anterior, spicule length, and the de Man ratios; a, c, c' and s.

3.5 Data analysis

3.5.1 Analysis of quantitative data on nematodes

Identified and enumerated PPN were subjected to various analyses. Relative abundance, population density, regression analysis, richness, diversity and evenness of various genera of PPNs from the soil and tuber samples were calculated based on methods of community analysis (Norton, 1978; Beals, 1960) where:

Relative Abundance = Ratio of a particular nematode genera over total nematode population x 100

2. Genera richness = number of genera in a given soil sample

3. Genera evenness was calculated using Simpson's index of diversity (D) to determine relative abundance with which each genus was represented in areas sampled. This was done using formula below;

$$D = 1 - \left(\frac{\Sigma n(n-1)}{N(N-1)} \right)$$ (Simpson, 1943),

Where D= Simpson's Diversity Index

n= the number of individuals of genera

N= the total number of genera in the sample

The D values obtained from different samples were compared using ANOVA ($p < 0.05$) and Least Significant Difference (5%).

3.5.3. Morphometric data analysis

Morphometric data was analysed using principal component analysis in xlstat version 2015.1. This analysis produced a set of variables (principal components) that were linear combinations of the original variables. The variables (principal components) were independent of each other and ranked according to the amount of variation accounted for.

The analysis also produced biplots showing the relationships between the morphometric characters and correlations between the variables and the principal component.

CHAPTER FOUR

4.0 RESULTS

4.1 Farmers' knowledge, perceptions' and experiences concerning occurrence and management of nematodes associated with yam production in the Guinea savanna and Transitional rain forest agro-ecological zones of Ghana

4.1.1 General background of Yam (*Dioscorea* spp) Farmers

The survey revealed that majority of *D.* species farmers in the Northern and Brong Ahafo region of Ghana were males (93.3%), females were (3.7%). Majority of the respondents were aged 41-50 years (59.3%). With the rest being 21-30 years (1.9%), 31-40 years (35.2%) or 51 years and above (3.7%) (Fig. 7).

Figure 7. Age distribution of yam farmers in the Guinea Savanna and Transitional rain forest agro-ecological zones.

Majority of *Dioscorea* species farmers (70.4%) in the two regions has no formal education (Table 2). The rest had formal education up to Junior High School (JHS) level (22.2%), Senior High School (3.7%) and a small proportion of the farmers (3.7%), however had tertiary education.

Table 2. Educational background of respondents

Educational level	Frequency	Percent	Cumulative Percent
Middle Sch /JHS	13	21.7	21.7
Secondary	2	3.3	25.0
Tertiary	2	3.3	28.3
None	43	71.7	100.0
Total	60	100.0	

Majority of the farmers (51.8%) had between 11 – 15 years farming experiences in *Dioscorea* species production (Fig. 8). The rest had between 6 – 10 years (27.8%), 1 – 5 years (3.7%) and greater than 15 years (16.7%) of experience.

Figure 8. Years of farming *Dioscorea* species by farmers in the Guinea savanna and Transitional rain forest agro ecological of Ghana.

A greater proportion of yam farmers (57.7%) in these two regions were smallholder farmers with holdings ranging between 2 - 3 acres and others between 4 -5 acres (39.5%) and 0 - 1 acre (2.8%) (Fig. 9)

Figure 9. Sizes of farmers' yam farms in the Guinea savanna and Transitional agro ecological zones in Ghana (2011 – 2014).

4.1.2 Sources of seed yam used by farmers

Farmers' sources of planting materials varied with a greater proportion (72.5%) sourcing their planting materials from their own farms. Others got their planting materials from family members (5.1%). There were also combinations of materials from various sources, these included materials from their own farms and from family members (19.6%), own farm and market (1.4%), family members and market (1 %) and a combination of both own farm, family members, and market (0.4 %) (Fig. 10). The major varieties grown by farmers in the two ecological zones include Pona, lareboko, afeshei, fuseinbilla, lillea, limo, matches, asobayere, lobere, gunguma and mankuma. Furthermore, a relatively greater proportion of yam farmers (26.7 %) in these 2 ecological zones grew combinations of 'Pona', 'Lareboko', 'Fusheinbilla' and 'Afashe' yam varieties. However most of the respondents grow either lareboko and pona or both (Table 3).

Figure 10. Farmers sources of seed yam between years 2011 and 2014.

Table 3. Major white yam varieties grown by farmers in the Guinea savannah and transitional ecological zones of Ghana.

Varieties	Frequency	Percent
Pona, larebroko,asobayere and afashe	6	10.0
Pona, asobayere, afashe and lilea	7	11.7
lobere, lilea and afashe	7	11.7
Pona, larebroko,fusheibila and afashe	16	26.7
Pona, larebroko matches, afia and limo	11	18.3
Pona, larebroko,seidubile and numa	2	3.3
Pona, larebroko,asobayere, gunguma, dedii and mankoma	10	16.7
Lobere	1	1.6
Total	**60**	**100**

4.1.3 Farmers Land Use Intensity

Majority of the yam farmers (46.3%) had continuously cultivated their fields for 7 – 10 years and a sizeable proportion (3.7%) had also cultivated their fields for the past 4 – 6 year. The rest were between 1 -3 years (7.4%), 11 – 15 years (7.4%) and more than fifteen years (1.9%) (Table. 4).

Table 4. Farmers land use intensity in yam production for Guinea savanna and transitional ecological zones of Ghana

No. of years	Percent of farmers
1- 3 years	7.4
4 - 6 years	37.0
7 - 10 years	46.3
11 - 15 years	7.4
> 15 years	1.9

The major crops that were intercropped with yam include cassava, pigeon pea, cowpea, maize, groundnut, millet, guinea corn, pepper, okra, and tomato. A greater proportion (46.3%) of farmers have been continuously grown yam on the same piece of land for the past 1 – 3 years, and others (42.6%) had for the past 4 – 6 years (Fig. 11).

Figure 11. Various periods of continuous cultivation of yam on same piece of land by farmers in the Guinea savanna and transitional agro ecological zones.

Almost all farmers (98.1%) had ever practiced crop rotation and intercropping in their yam production. The periods of rotation varied between 1 – 2 years (15.1%), 3 – 5 years (83.0%), and more than 5 years (1.9%) (Fig. 12).

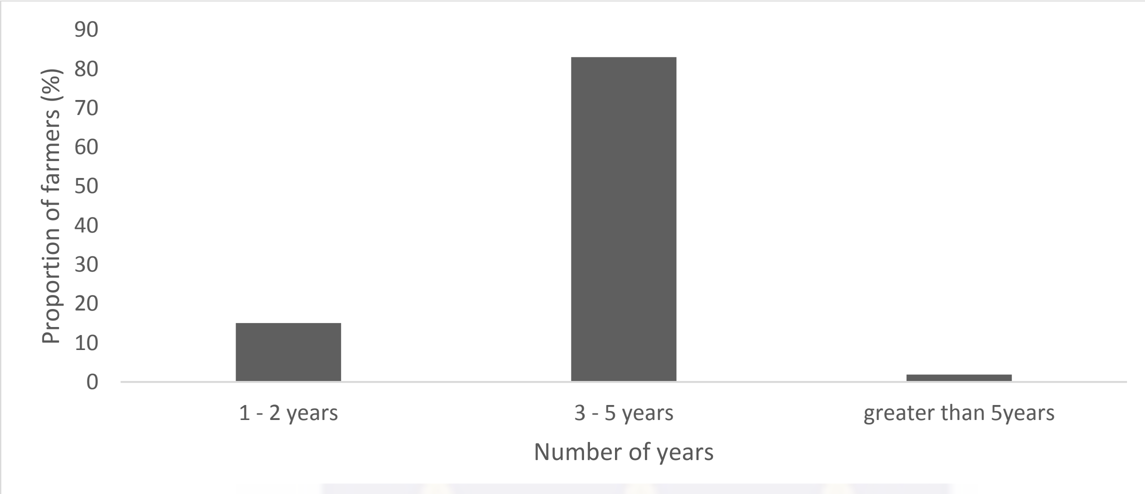


Figure 12. Periods of rotation practiced by yam farmers in the two agro ecological zones

The major crops involved in the rotational program and intercropping include maize, cassava, millet, sorghum, pigeon pea, cowpea, garden eggs, pepper, okra, groundnuts, and soybean (Table 5).

Table 5. Major crops rotated with yam in the ten districts

Types of crops	**Frequency**	**Percent**
Maize, cassava, and cowpea	29	49.2
maize and pigeon pea	2	3.4
maize, pepper, okro, and garden eggs	2	3.4
maize, groundnut, millet, and soya	18	30.5
maize, millet, and cassava	2	3.4
maize and garden eggs	4	6.8
maize and sorghum	2	3.4
Total	59	98.1
	60	**100**

A greater majority of the yam farmers (53.7 %) had ever practiced land fallow system, whiles (46.3 %) had never used this production practice in their yam farming. The periods of fallow however varied between 1 year (10.3 %), 2 – 3 years (75.9 %) and 5 -10 years (13.8 %) (Fig. 13).

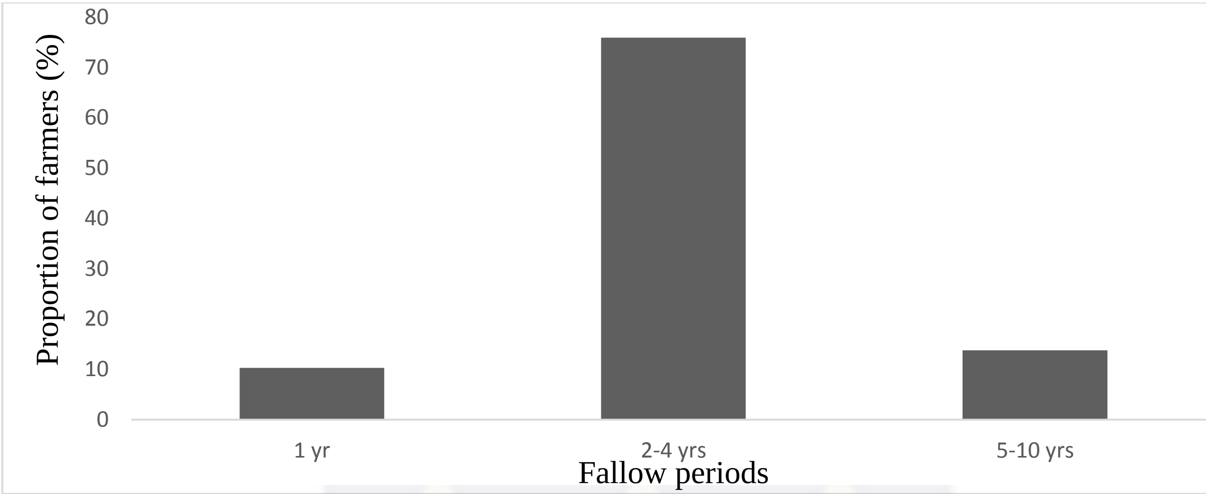


Figure 13. Duration of land fallow period allowed by yam farmers in the Guinea savanna and Transitional-agro ecological zones of Ghana

4.1.4 Farmers knowledge, perception and experiences concerning prevalence of nematodes on the farm and in stored yams

Majority of the yam farmers (96.3%) in both ecological zones mentioned they had experienced diseased conditions in their yam tubers both in the field and in storage. A greater proportion of the yam farmers (81.5%) could not confirm that the disease on the yams were caused by nematodes because, they did not have any knowledge about nematodes. However, the farmers (100%) believed that the yam varieties grown may be susceptible to nematode parasitism albeit different levels. Majority (26.7%) of the farmers perceived Lareboko to be highly susceptible to nematode parasitism followed by 'Pona' (20.0%), 'Afishei' (16.7%), 'Fusheinbilla' (11.7%), 'Asobayere' (12.9%), and 'Lillea' (7.4 %) (Table 6). However, 'limo' (5.5%), 'Gunguma' (1.9 %) and 'Lobere' (1.9 %) were perceived to be moderately susceptible whiles 'matches' and 'seidubille' were least susceptible.

Almost all the farmers (100%) agreed they observed the following symptoms: necrotic streaks or lesions on roots, reduced rooting system, internal rotting or discoloration of vegetative organs, andstunting and rosetting, typical nematode parasitism symptoms in their yam fields (Table 7).

Table 6. Farmers' knowledge on susceptibility of yam in the guinea savanna and transitional ecological zones of Ghana

Yam variety	Frequency	Percent of respondents
Lareboko	16	26.7
Pona	12	20.0
Afishei	10	16.7
Fuseinbilla	7	11.7
Asobayere	5	8.3
Lillea	5	8.3
Limo	3	5.0
Gunguma	1	1.7
Lobere	1	1.7
Total	**60**	**100**

Table 7. Farmers' knowledge and perception on symptoms of nematodes parasitisms in the field from the Guinea savanna and transitional agro ecological zones.

Symptoms	**Frequency**	**Percent of respondents**
Internal rotting and discoloration of vegetative organs	50	33.0
Necrotic streaks and lesions on roots	25	41.7
Stunting and resetting	8	13.3
Reduced rooting system	6	10.0

Generally, farmers were of the opinion that nematode parasitism occurred in almost all soil conditions. The farmers stated that nematode parasitism occurred mostly in sandy-clayey soils (59.3 %), waterlogging- clayey soils (24.1 %), well drained loamy soils (5.6 %), clayey-sandy and waterlogged soil conditions (3.7 %) (Fig. 14).

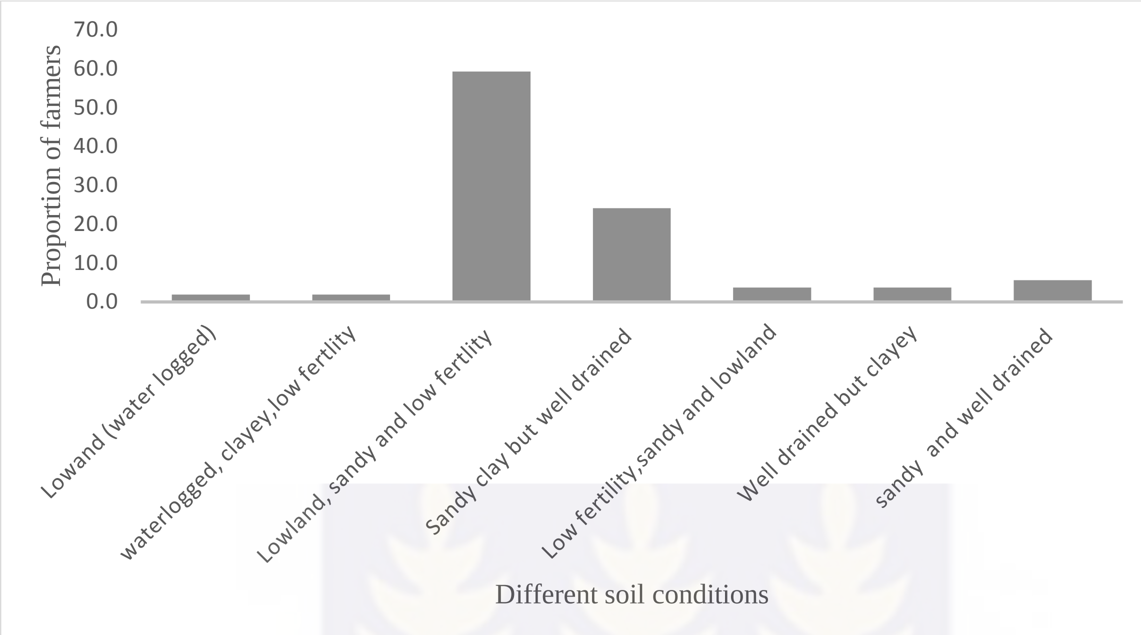


Figure 14. Farmers perception on soil conditions in which nematode parasitism occurred most in the study area

A greater proportion of the farmers (76.7 %) stored their yam tubers and setts in farmstead at their farms whiles some store their tubers in local yam barns at the farm or home (6.7 %), in both farmstead and in a store at home (6.7 %), in a farmstead and local yam barn at home or farm (5.0 %) and, under a shade in the farm (5.0 %) (Table 8).

All farmers (100 %) stated that they experienced storage loses in their yams due to nematode parasitism. Even though majority (81.5 %) of farmers did not have enough knowledge about nematodes, they indicated symptoms such as light yellow lesions below the outer skin of the tuber, external cracks on skin of tubers, rotting of the outer 1 – 2cm layer, hardened and dried tissues with varying discolorations, significant weight loses in tubers, flaking off- of external coverings exposing patches of dark brown and general decay of tubers which are symptomatic of nematode parasitism of yam in storage have been observed (Table 9).

Table 8. Place of storage of yam by farmers in the 10 districts

Type and place of storage	Frequency	Percent
Yam barn at home or farm	4	6.7
Farm hut	46	76.7
Under a shade in the farm	3	5.0
Farmstead and yam barn at home or farm	3	5.0
Farmstead and in a store at home	4	6.7
Total	**60**	**100**

Table 9. Farmers' knowledge on symptoms of nematode parasitism on yam tubers in storage in the two AEZs.

Symptoms	Frequency	Percent of respondents
Hardened and dried tissues with varying discolorations	32	53.3
External cracks on skin of tubers	45	75.0
Significant weight loses in tubers	42	70.0
Light yellow lesions below the outer skin of tubers	51	85.0
Rotting of the outer layer of tubers (1-2 cm)	36	60.0
Flaking-off of external coverings exposing patches of dark brown colorations	31	51.7
General decay of tubers	41	68.3

4.1.5 Farmers knowledge, perception and experiences concerning spread and management of nematodes affecting yam tubers in the field and in storage

Various modes of spread were perceived by yam farmers in the two agro-ecological zones of Ghana regarding nematode parasitism. Farmers perceived the nematodes were spread by infected seed yam (37%), infected soils (31.5%), insects (16.7%), and excessive heat (14.8%). Majority of the yam farmers (83.3%) in both ecological zones believe that nematodes could not be controlled. However, (16.7%) of the farmers believed nematodes could be managed through crop rotation, land rotation, and destruction of infected tubers.

4.1.6 Knowledge and experiences of farmers on yield and economic impact of nematodes associated with yam tubers on livelihood of yam farmers

Majority of the farmers (56.7%) in both ecological zones cultivated yam mainly for food security reasons. Another group (28.3%) stressed they cultivated yam exclusively for income generation, and (15.0 %) indicated they cultivated the crop in order to buy assets such as houses, land, tractors and bullocks (Table 10).

Table 10. Reasons for planting *Dioscorea* species by farmers in the Guinea savanna and transitional ecological zones

Reasons for planting yam	Frequency	percent of respondents	Cumulative percent
Food security	34	56.7	56.7
Income generation	17	28.3	83.3
Purchase of assets	9	15.0	100.0

The yields expected by farmers in both agro-ecological zones were between 4.0 tons in small farms (< 1 ha) and 21.0 tons in large farms (> 10), but the actual harvest of most farmers (27) was between 1 and 3 tons (Table 11). Among the 45.0% of farmers who cultivated 1 ha of farmland, only 14.8%

obtained their expected yields (4 – 6 tons/ 1-2 ha), whiles majority (23) had yields between 1 – 3 tons/2 ha farm size. None of the farmers who cultivated farmlands ranging from 5 to 10 ha achieved his/her expected yield; implying that 35.2% of the farmers could not attain their anticipated yield.

Table 11. Expected and actual yield of *Dioscorea* species of farmers in the Guinea savanna and Transitional rain forest agro ecological zones.

Farm size/ha	**Expected yield (tons/ha) of yam per annum**	**Actual yield (tons/ha) per annum**								
		1 - 3	4 - 6	7 - 9	10 - 12	13 - 15	16 – 18	19 – 21	**Freq**	**%**
1-2	4.0 – 6.0	23*	4	0	0	0	0	0	27	45.0
4	7.0 – 9.0	4	4	6	0	0	0	0	14	23.3
5	10.0– 12.0	0	3	5	0	0	0	0	8	13.3
7	13.0– 15.0	0	0	2	4	0	0	0	6	10.0
8	16.0– 18.0	0	2	1	0	0	0	0	3	5.0
10	19.0– 21.0	0	0	0	0	2	0	0	2	3.3
	Total	**27**	**13**	**14**	**4**	**2**	**0**	**0**	**60**	**100**

Majority of the respondents (40.0%) attributed their yield loses in the field to extensive drought (Table 12). Other farmers ascribed their yield loses in the field to field diseases and pests (23.3%), non – staking (18.5%), inadequate labour (10.0 %), poor land preparation (6.7%), and flooding (3.3%).

Table 12. Farmers' reasons for not achieving expected yield of *Dioscorea* species in the Guinea Savanna and Transitional agro-ecological zones.

Reasons for not achieving expected yield	**Frequency**	**Percent respondents**
Field diseases and pest	14	23.3
Extensive drought	24	40.0
Poor land preparation	4	6.7
Non-staking	10	16.7
Inadequate labour	6	10.0
Flooding	2	3.3

Farmers (100%) stated that, they experienced tuber losses in storage mainly because of nematode infestation and other disease causing organisms. Among the 27 farmers who stored about 1.5 tons of tubers after harvest, they all suffered from about 25% losses due to nematodes, those storing 2 tons, 3 tons, 4 tons, and 5 tons suffered 20%, 24%, 15%, and 18% losses respectively due to nematode parasitism (Table 13).

Table 13. Loses suffered by farmers due to nematode infestation of yam tubers in storage

Actual Yield/tons	**Quantity stored/tons**	**Percent loses due to nematodes**	**Freq**	**Percent response**
3	1.5	25	27	45.0
4	2.0	20	11	18.3
5	3.0	24	8	13.3
7	4.0	15	6	10.0
8	4.0	20	5	8.3
10	5.0	18	4	6.7

Ware yam, 1 tuber = > 1 kg. Source: Ogbonna *et al.*, 2011

Nematodes also impacted negatively on the livelihoods of farmers as majority were unable to pay: their wards school fees (88.9%), medical bills (83.3%), service loans (87%) and purchase of assets (72.2%) (Table 14). A greater proportion (77.8%) of the farmers reported that losses due to nematodes parasitism in stored tubers impacted negatively on their social relations with their spouses in some cases, with extended family members. Most farmers (57.4%) were reluctant or unwilling to contact extension agents when yields were low in the fields or when losses were experienced in storage. There were however some farmers who indicated that, poor yields and storage losses due to nematodes parasitism and other diseases, did not negatively influence their relationship with extension agents.

Table 14. Socio-economic impacts of nematodes associated with yam on the livelihoods of farmers in the Guinea Savanna and the Transitional ecological zones. n = 60

Economic and social responsibilities of farmers	**Level of significance**				
	1	2	3	4	5
Payment of school fees	88.9*	7.4	3.7	0	0
Payment of medical bills	83.3	12.9	3.7	0	0
Purchase of assets	72.2	18.5	9.3	0	0
Payment of service loans	87	9.3	3.7	0	0
Relationship with spouse and neighbours	77.8	14.8	7.4	0	0
Relationship with extension officer	57.4	20.4	11.1	7.4	3.7

1 = High negative influence; 2 = Negative influence; 3 = Neutral; 4 = Positive influence; 5 = High positive influence.
*Values represent proportion of farmers associated with various socio-economic responses, related to nematode damages on yam.

4.2 Genera of plant parasitic nematodes associated with yam rhizosphere and yam tubers in the Guinea savanna and Transitional agro ecological zones of Ghana

When nematodes were extracted from the soil, yam tubers in the field and in storage, nine genera belonging to six families of Order Tylenchida and Triplonchida were found associating with yams (Table 15).

Table 15. Plant parasitic nematodes extracted from yam rhizosphere soil and yam tubers in the Guinea savanna and Transitional rain forest agro ecological zones of Ghana

Order	Sub- Order	Family	Genus
Tylenchida	Tylenchina	Pratylenchidae	*Pratylenchus*
		Hoplolaimidae	*Helicotylenchus*
			Hoplolaimus
			Rotylenchulus
			Scutellonema
		Meloidogynidae	*Meloidogyne*
		Tylenchulidae	*Tylenchus*
		Tylenchorhynchudae	*Tylenchorhynchus*
Tryplonchida	Diphtherophorina	Trichodoridae	*Trichodorus*

The nine genera of nematodes were present in soils sampled, whiles 8 genera with the exception of *Rotylechulus* spp. (Reniform) occurred in yam tubers sampled from storage. Three out of the nine nematode genera were recovered from yam tubers in the field during the rainy season. Nematode genera extracted from yam tubers in storage include: *Pratylenchus* spp. *(*lesion nematode), *Scutellonema bradys* (yam nematode), *Meloidogyne* spp. (Root- knot nematode), *Tylenchus* spp., *Tylenchorhynchus* spp. (Stunt nematode), *Hoplolaimus* spp. (Lance nematode), *Trichodorus* spp. (Stubby nematode) and *Helicotylenchus* spp. (Spiral nematode). All these genera including *Rotylechulus* spp. (Reniform nematode) were also identified in soil sampled in the rainy season. Nematode genera extracted from yam tubers in the field during the rainy season included; *Pratylenchus* spp. (Lesion nematode), *Scutellonema bradys* (Yam nematode), and *Meloidogyne* spp. (Root-knot nematode).

Eight (8) out of the nine nematode genera identified belonged to the Order Tylenchida, while only one (1) belonged to Triplonchida. The family Hoplolaimidae had the highest representative with four genera out of the nine. The family Tylenchulidae had three genera of nematodes associated with it. The occurrence of most of these PPN genera cut across the two AEZs.

4.3 Morphological descriptions of some common nematode genera identified during the survey

4.3.1 *Helicotylenchus* spp.

The body is spiral when relaxed (Fig. 15). Lip region rounded and bearing five annules. Stylet long with cuped knobs. Dorsal oesophaegal gland outlet behind the stylet.

Figure 15: *Helicotylenchus* spp. showing the characteristic spiral morphology.

4.3.2 *Hoplolaimus* spp.

They are large sized nematodes (1 mm – 2 mm). Lip region high, offset with prominent transverse striae, It has huge cephalic framework. Big stylet, basal knobs bearing anterior tooth like projections. Excretory pore posterior or anterior for most species. It has well developed esophagus. Tail is short, bluntly rounded and annulated. Phasmids large and scutellum like.

4.3.3 *Meloidogyne* spp.

Mature female have pear shaped body. Stylet knobs are set off, rounded to transversely elongated. Juveniles has basal knobs offset, rounded to transversely elongated (Fig. 16). The male has long thin cylindrical shape but the lip region has a distinct head cap. The vulva and anus are terminal. The stylet is shorter.

Figure 16*: `Meloidogyne* spp., juvenile tail region

4.3.4 *Pratylechus* spp.

The body is slender in shape and width increases from the level of pharyngeal bulb towards the vulva and also decreases from the level of vulva towards the tail tip. Cuticular annulations very conspicuous (Fig, 17). The lip region bears two distinct annules. The tail tapering strongly with a narrow tip. Male tail pointed, bursal edges faintly crenated.

4.3.5 *Rotylenchus* spp.

Body shape after fixation form a spiral to an open 'C' shape. The posterior part is tightly coiled. Head region clearly separated from the rest of the body. Stylet knobs rounded and anteriorly flattened. Tail bluntly conical and tapered. Body is slender. Spicules slightly curved with rounded head and weakly enlarged central part. Bursa enveloping tail.

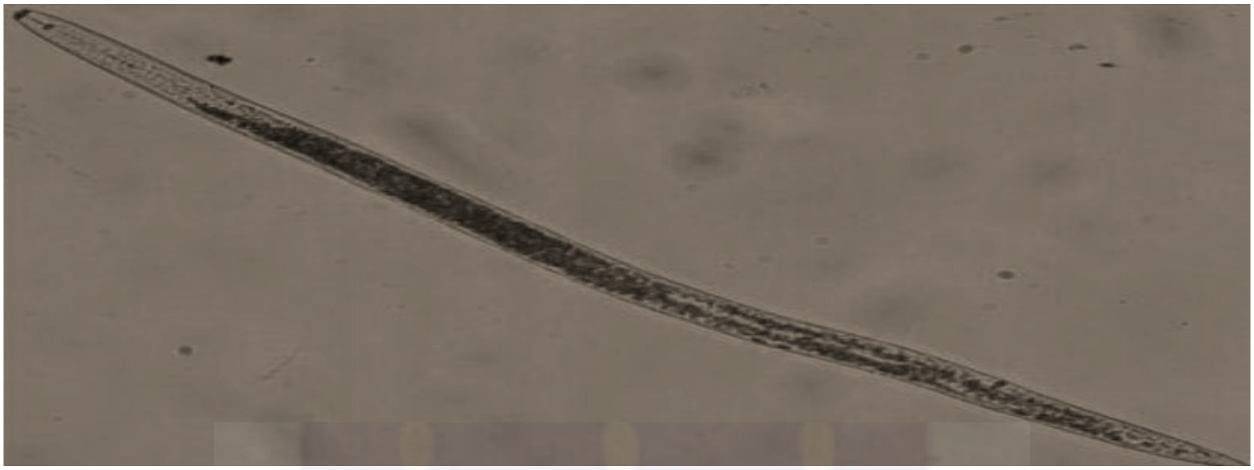


Figure 17. *Pratylenchus* spp.

4.3.6.1 *Scutellonema bradys*.

Scutellonema bradys was identified to the species level based morphological features of the female and males.

Female *Scutellonema bradys*

Body straight to slightly arcuate when relaxed; annules about 1.6 µm wide near middle; lateral fields about one-fifth body-width, areolated at phasmids and anteriorly, sometimes irregularly areolated on mid-body and tail. Lip region knob-like, offset by a constriction, with a labial disc and 6 to 8 annules lacking longitudinal striations (Fig. 18A). Cephalic sclerotization strong. Spear well developed with large oval to rounded basal knobs bearing flattened, indented or irregular anterior surfaces; anterior tapering portion a little less than half spear length. 0-3 annules anterior to excretory pore and close to oesophago-intestinal junction. Hemizonion 1 annule long, about 8 annules behind the excretory pore. Oesophageal glands elongate, overlapping intestine dorsally and dorso-laterally; nucleus of dorsal gland anterior to those of subventrals. Vulva a transverse slit with conspicuous cuticular thickenings towards ends (Fig. 18B). Tail variable with obtusely rounded striated terminus.

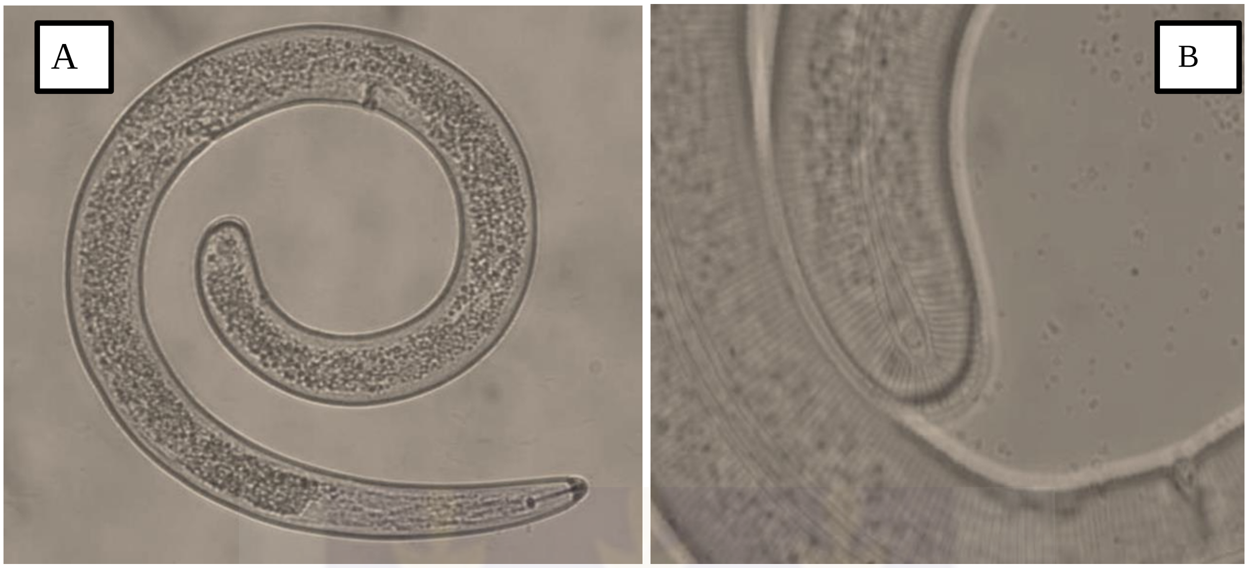


Figure 18. (A) *Scutellonema bradys* (female) (B) Tail region of *Scutellonema bradys* female

4.3.6.2 *Male*

Similar to female. Bursa large, crenate, enclosing tail. Spicules slightly cephalated and ventrally arcuate, with large distal flanges. Capitulum (telamon) prominent, about 10 µm long. Cuticular, non-protoplasmic terminal portion of tail 11-16 µm long (Fig. 19 A and B).

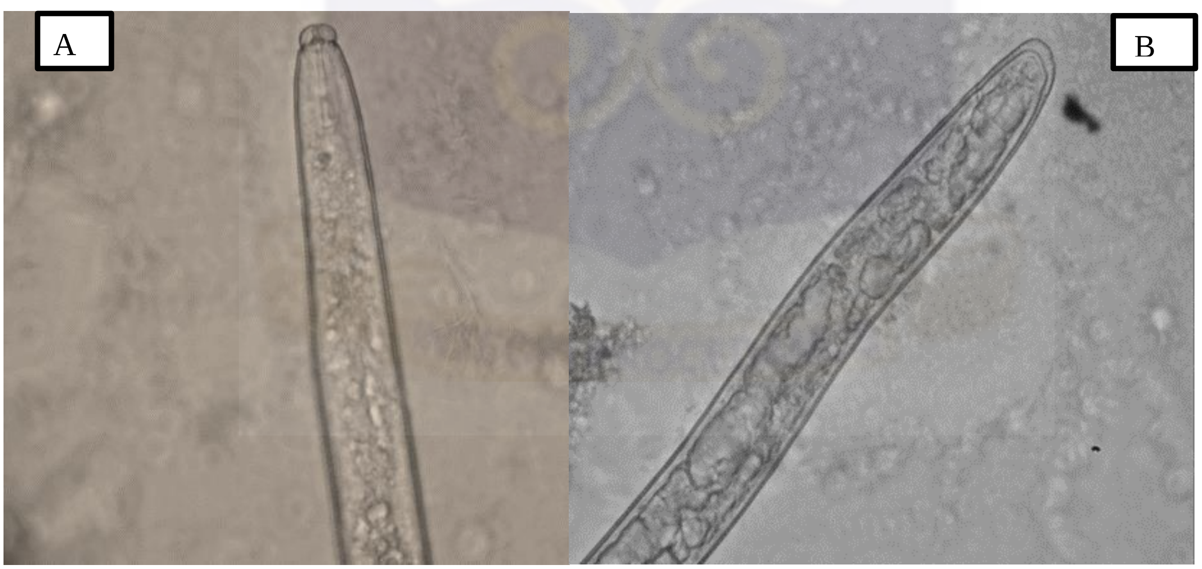


Figure 19: Head region (A) and Tail region- male (B) of *Scutellonem bradys* - male

4.3.7 *Trichodorus* spp.

The body is cylindrical, ventrally curved, tapering increasingly from the mid body to the head and strongly to loosely J shaped (Fig. 20). Tail terminus gently tapering, bluntly rounded with cuticular thickenings, rounded lip region and terminus flat at the tip.

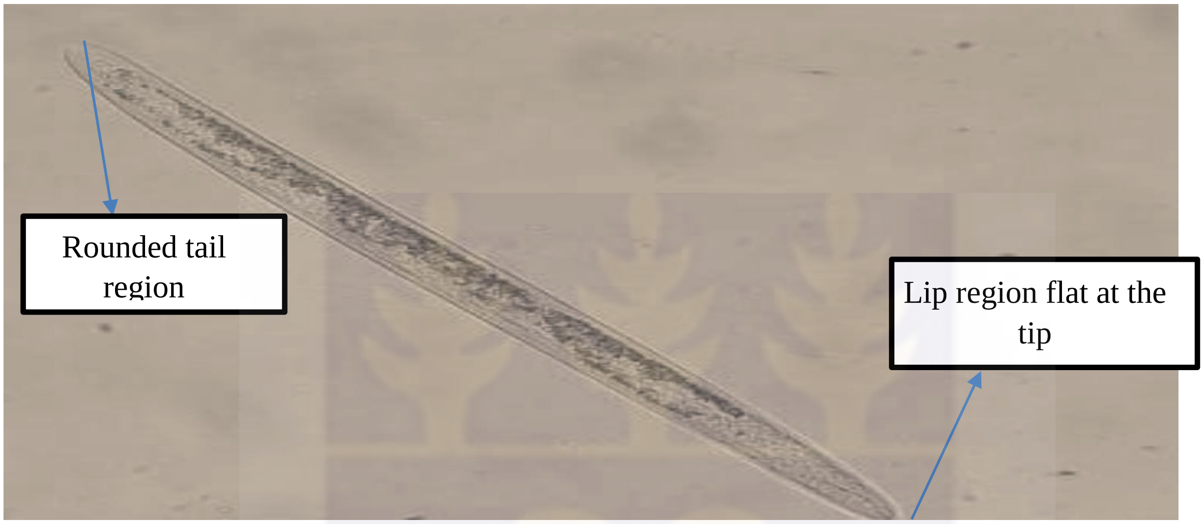


Figure 20: *Trichodorus* spp.

4.3.8 *Tylechorhynchus* spp.

Lip region set off by constrictions or continuous. Phasmids conspicuous and located behind anal region. It has a strong stylet with heavy basal knobs (Fig. 21). Vulva is located near the mid body. Tail is blunt and conoid. Stylet well developed with conspicuous basal knobs.

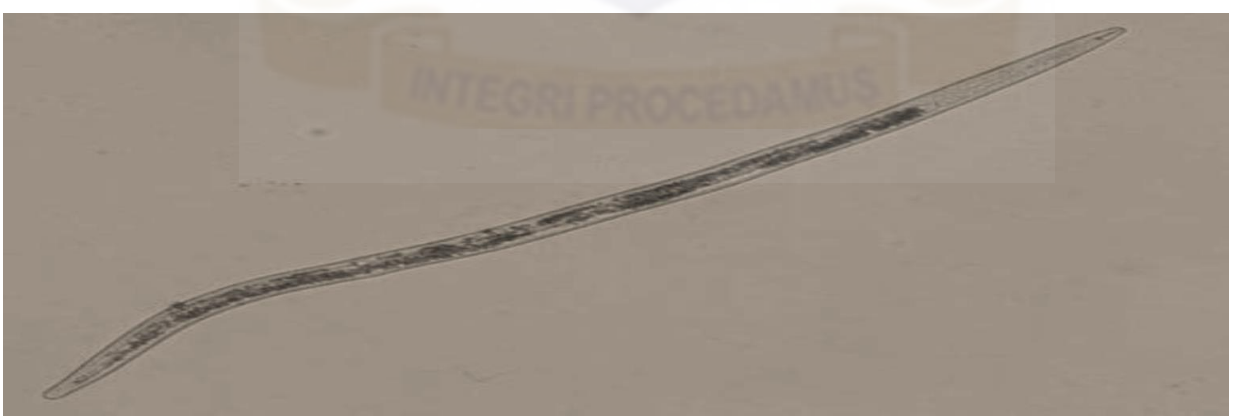


Figure 21: *Tylenchorhynchus* spp.

4.3.9 *Tylenchus* spp.

Tylenchus spp differ from differ from meloidogyne by the presence of a stronger stylet, a more posterior excretory pore and the absence of pharyngeal overlap. Tail is elongated and attenuated. Head flattened with caudal alae. Vey slender body and body width very variable.

4.4 Density of plant-parasitic nematodes associated with yam and soil around the rhizosphere in the guinea savanna and transitional agro ecological zones of Ghana

Generally, nine nematode genera were encountered in both the Guinea Savanna and Transitional agro ecological zones. They include *Pratylenchus* spp. *Scutellonema bradys, Meloidogyne* spp. *Helicotylenchus*s spp., *Tylenchus* spp., *Tylenchorhynchus* spp., *Hoplolaimus* spp., and *Rotylenchus* spp. Higher population of nematodes (5,545) were extracted from yam rhizosphere soil in the Guinea-savanna agro-ecological zone compared to nematode numbers in the Transitional rain forest (Fig. 22). The highest and least nematode genera identified in the Guinea-savanna agro- ecological zone soils were *Scutellonema bradys* (1,295) and *Tylenchorhynchus* spp. (96) respectively. Within the Transitional forest zone however, *Pratylenchus* species (1,101) and *Rotylenchulus reniformis* (96) were the highest and least nematode genera identified in soil samples. H*oplolaimus* spp. was the only nematode genera not present in the Transitional rain forest although, was identified in the Guinea avannah soils.

Three nematode genera (M*eloidogyne* spp., S*cutellonema bradys*, and P*ratylenchus* spp.) were isolated from yam peels during the rainy season for both the Guinea savannah and Transitional rain forest agro-ecological zones.

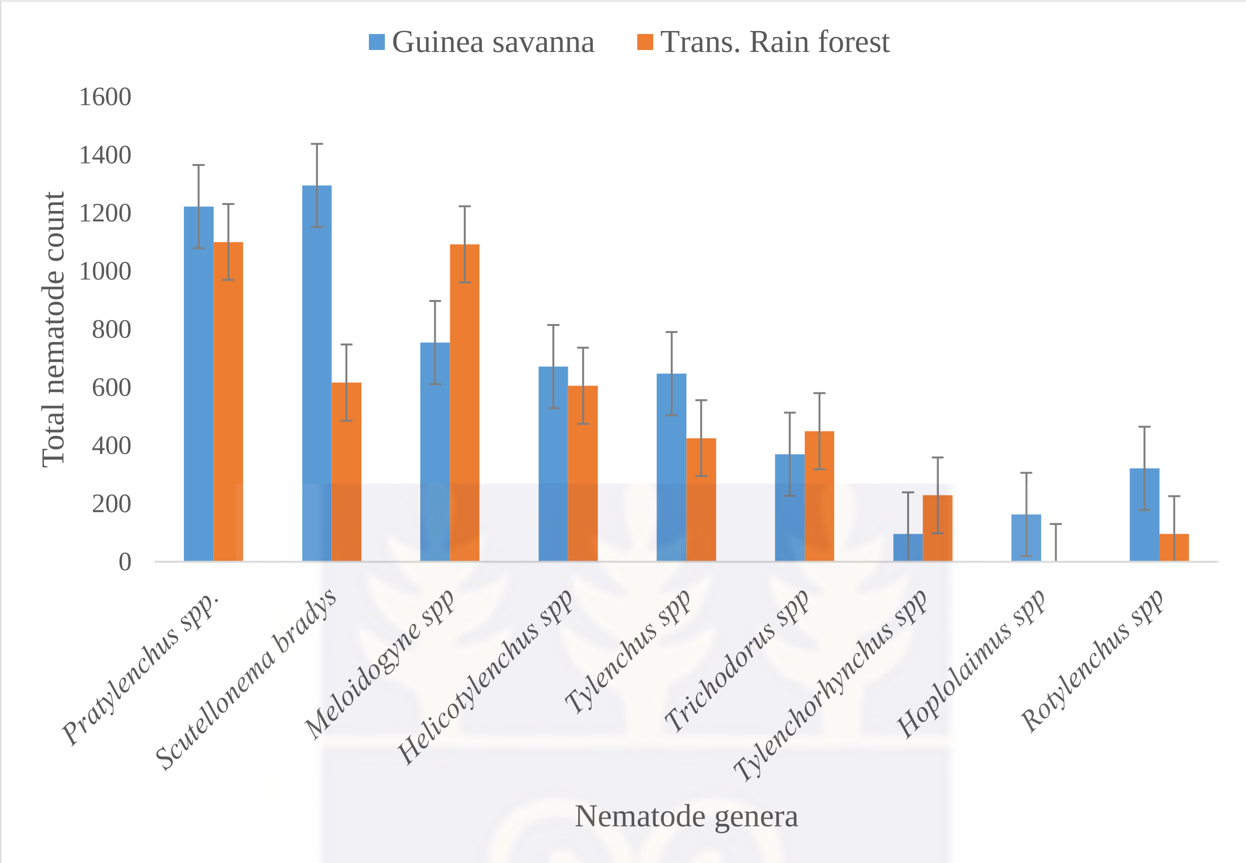


Figure 22. Population densities of nematodes extracted from 200 cc of soil around the rhizosphere of yam plants in the Guinea savanna and Transitional rain forest agro-ecological zones (Vertical bars represent standard error).

The highest nematode genera were *Pratylenchus* spp. for both agro-ecological zones (Fig. 23). However, for yam tubers sampled in storage, eight nematode genera each were isolated for the yam for both the Guinea savanna and the Transitional rain forest agro-ecological zones. They include, *Pratylenchus* spp., *Meloidogyne* spp., *Scutellonema* spp., *Tylenchorhynchus* spp., *Hoplolaimus* spp., *Trichodorus* spp., *Helicotylenchus* spp. and *Tylenchus* spp.

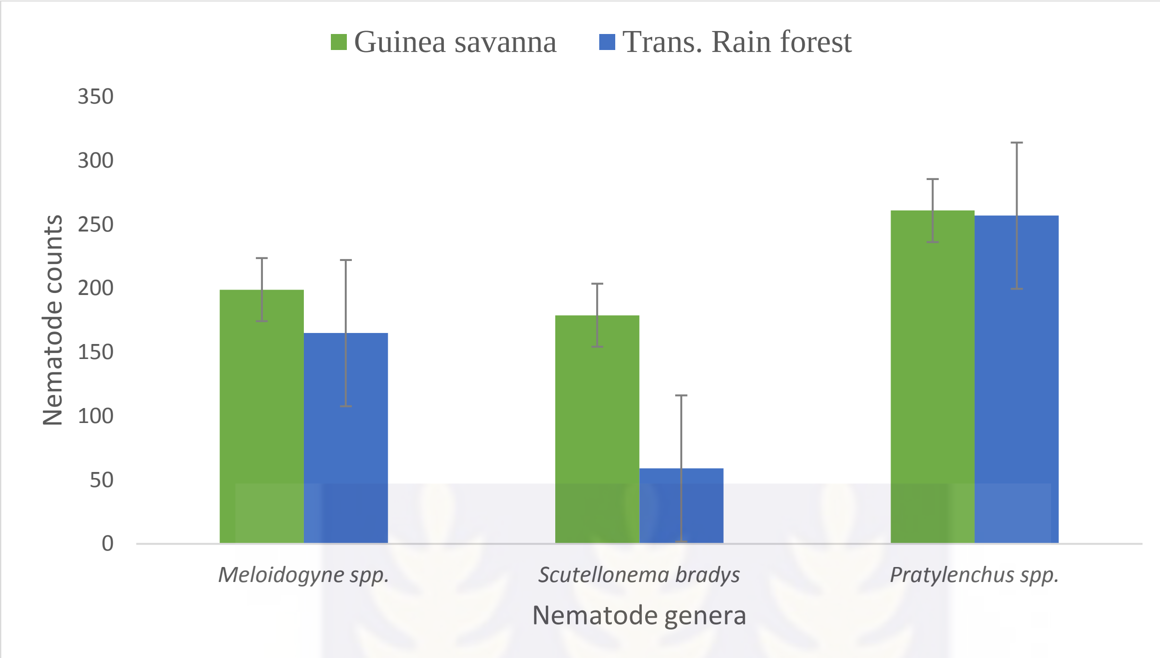


Figure 23. Density of nematode genera extracted from yam tuber peels in the field of the Guinea savanna and transitional rain forest agro-ecological zones during the rainy season (Vertical bars represent standard error).

A higher number of nematodes were isolated from the yams obtained in storage compared to those obtained during the rainy season (Fig. 24) *Scutellonema bradys* (1686) and *Pratylenchus* spp (1577) were the highest recorded nematode in yam tubers from the Guinea savanna and Transitional agro ecological zones respectively.

4.5 Relative abundance and occurrence of plant parasitic nematode associated with yam in the Guinea savanna and transitional rain forest ecological zones

Plant-parasitic nematodes were detected in all soil and yam samples but at varying relative abundance and frequencies of occurrence. Nine nematode genera were isolated from the various samples from the ten districts in the two AEZs.

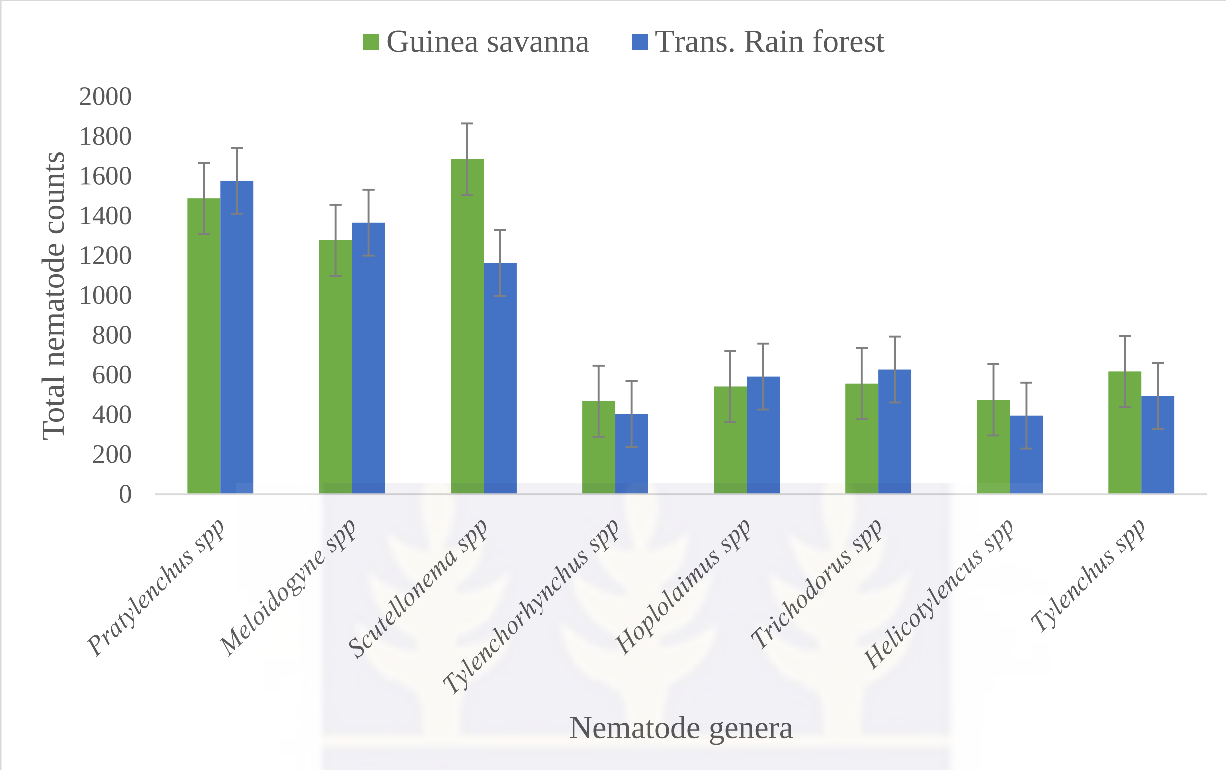


Figure 24. Densities of nematodes extracted from stored yam tubers in storage from the guinea savanna and Transitional agro-ecological zones (Vertical bars represent standard error).

They include *Prtaylenchus* spp., *Scutellonema* spp., *Meloidogyne* spp., *Helicotylenchus* spp., *Tylenchus* spp., *Tylenchorhynchus* spp., *Rotylenchus* spp., *Trichodorus* spp. and *Hoplolaimus* spp. (Table 16). *Pratylenchus* spp., *Scutellonema* spp., *Meloidogyne* spp., *Helicotylenchus* spp., *Tylenchus* spp and *Trichodorus* spp occurred at high frequencies across both AEZs. Although plant-parasitic nematodes (PPNs) occurred across all sampled districts in these AEZs, soils from Atensu and Kanshegu had the highest PPN occurrence. *Pratylenchus* spp. and *Hoplolaimus* spp. had the highest and least relative abundances of 22.9% and 1.6% respectively (Table 16). *Tichodorus* spp., *Rotylenchulus* spp. and *Tylenchorhynchus* spp. were present in the soils, but at lower relative abundance of 8.1%, 4.1%, and 3.1% respectively.

Table 16. Occurrence and relative abundance of plant parasitic nematodes in the yam rhizosphere soil in the AEZs during the rainy season

Nematode Genera	**Freq. of occurrence**	**Percent Freq. rating***	**Nematode pop/200 cc soil**	**Relative Abundance (%)****
Pratylenchus spp.	10	100	2324	22.9
Scutellonema spp.	10	100	1912	18.8
Meloidogyne spp.	10	100	1848	18.2
Helicotylenchus spp.	10	100	1278	12.6
Tylenchus spp.	9	90	1074	10.6
Trichodorus spp.	7	70	821	8.1
Rotylenchulus spp.	4	40	418	4.1
Tylenchorhynchus spp.	4	40	325	3.2
Hoplolaimus spp	3	30	163	1.6

*n/N X100 (n = frequency of individual nematode occurrence. N = Sample size (10)
**In/TN (In = Individual genera in all samples. TN = Total population of all nematode genera)

Nematode genera that were isolated from yam tubers in the field during the rainy season in the various districts within the two AEZs include *Pratylenchus* spp., *Meloidogyne* spp. and *Scutellonema* spp. All yam tubers (100%) collected during the rainy season had *Pratylenchus* spp present in them at a higher relative abundance of 46.3% in the two AEZs (Table 17). *Meloidogyne* spp. and *Scutellonema bradys* was present at moderate frequencies 32.4% and 21.2% respectively. During the dry season when yam tubers were in storage, eight nematode genera were extracted from sampled yam tubers across all ten districts in the two AEZs. They include *Pratylenchus* spp., *Scutellonema* spp., *Meloidogyne* spp., *Tylenchus* spp., *Hoplolaimus* spp., *Trichodorus* spp., *Helicotylenchus* spp. and *Tylenchorhynchus* spp. (Table 18).

Table 17. Occurrence and relative abundance of plant parasitic nematodes associated with yam tubers during the rainy season in the two AEZs

Nematode Genera	Freq. of occurrence	Percent Freq. rating*	Nematode pop/10 g yam peels	Relative Abundance (%)**
Pratylenchus spp.	10	100	520	46.3
Meloidogyne spp.	10	100	364	32.4
Scutellonema spp.	10	100	238	21.2

*n/N X100 (n = frequency of individual nematode occurrence. N = Sample size (10))
**In/TN (In = Individual genera in all samples. TN = Total population of all nematode genera)

All eight nematode genera had 100% frequency rating in the yam tubers sampled from storage across all ten districts in the two AEZs. *Pratylenchus* spp., *Scutellonema* spp. and *Melodogyne* spp. however had higher relative abundance of 22.3%, 20.8% and 19.2% respectively. *Trichodorus* spp., *Hoplolaimus* spp., *Tylechus* spp., *Tylenchorhynchus* spp. and *Helicotylenchus* spp. occurred at moderate relative abundance of 8.6%, 8.3%, 8.1%, 6.4%, and 6.3% respectively.

Table 18. Occurrence and relative abundance of plant parasitic nematodes associated with yam tubers in storage during the dry season in the two agro ecological zones.

Nematode Genera	Freq. of occurrence	% Freq. rating*	Nematode pop/10 g peels	Relative Abundance (%)**
Pratylenchus spp.	10	100	3065	22.3
Scutellonema spp.	10	100	2849	20.8
Meloidogyne spp.	10	100	2643	19.2
Trichodorus spp.	10	100	1185	8.6
Hoplolaimus spp.	10	100	1134	8.3
Tylenchus spp.	10	100	1112	8.1
Tylenchorhynchus spp.	10	100	872	6.4
Helicotylecus spp.	10	100	870	6.3

*n/N X100 (n = frequency of individual nematode occurrence. N = Sample size (10))
**In/TN (In = Individual genera in all samples. TN = Total population of all nematode genera)

A total of 1,122 nematodes specimens were encountered from yam tubers in the field during the rainy season in the 2 AEZs during the survey. Three (3) genera of nematodes were extracted from the yam peels in both ecological zone (Table 19). They include *Scutellonema* spp., *Meloidogyne* spp. and *Pratylenchus* spp. Kanshegu in the Guinea savanna ecological zone had the highest population of nematodes from the yam peels during the rainy season followed by Atrensu in the Transitional rain forest region. Afrefreso in the transitional zone was the locality less populated with nematodes followed by Kaswurape in the Guinea savanna ecological zone.

Kanshegu was the most heavily infested with an overall mean number of 365 nematodes per 10 g of yam tuber peels during the rainy season. The lowest population of nematodes (13) were isolated from yam tubers obtained from Nokwarease. The highest population of *pratylenchus* spp. was recorded in Atrensu. Similarly, the highest population of *Scutellonema bradys* was recovered from Kanshegu. During the rainy season, *Scutellonema bradys*. was not encountered in all yam tubers sampled from Ayayo and Nokwarease, both in the Transitional rain forest agro ecological zone.

Table 19. Mean nematode population in yam tubers (5 g of yam peels) in the field during the rainy season

Community	**Mean nematode densities from 10g of yam peel in the rainy season**			
	Meloidogyne spp.	*Scutellonema* spp.	*Pratylenchus* spp.	Overall mean
Choo	16	8	48	72
Shebo	38	24	72	134
Kpachi	28	38	44	110
Kaswurape	9	11	12	32
Kanshegu	108	98	85	365
Promposo	14	6	24	44
Ayayo	54	0	90	144
Atrensu	86	45	119	250
Afrefreso	6	8	18	32
Nokwareasa	5	0	8	13

When populations of different PPN genera in the soil during the rainy season were evaluated for all ten districts, the population densities ranged from 773 to 1495 nematodes per 200 cc of soil (Table 20).

A total of 10,163 nematodes specimens were encountered from the soil around the rhizosphere of yam in the 2 ecological zones during the survey. Nine genera of plant parasitic nematodes were encountered in the soils around the rhizosphere of yam roots in the two ecological zones (Table 20). Nematode populations were highest in the soils from Atrensu in the Transional rain forest region. The lowest population of PPN were extracted from yam rhizophere soils of Ayayo also in the transitional rain forest zone. Ayayo also recorded the lowest *S. bradys* population compared to all other districts in the two agro ecological zones.

Meloidogyne spp. had the highest mean density of 406 nematodes per 200cc of soil in Ayayo in the Transitional AEZ. However, *Pratylenchus* spp. with an overall mean population density of 2324 nematodes was the predominant genera across all ten districts in the two AEZs. Ayayo recorded the least mean population density of *Pratylenchus* spp. in the soils across the ten districts. Similarly, Kanshegu recorded the highest population of *Scutellonema bradys*. compared to all nine districts in the two ecological zones. Ayayo recorded the least population of *Scutellonema* spp. among all the ten districts in the two AEZs. However the highest mean population density of *Meloidogyne* spp. was recorded in Ayayo. *Hoplolaimus* spp. was only recorded in 3 districts in the guinea savanna AEZ out of the ten districts in the two AEZs.

Table 20. Mean nematode population in soils (200cc of soil) of yam field from the Guinea savanna and transitional ecological zones of Ghana

Community	Mean nematode densities of 200 cc of soil from the rhizosphere of yam									Overall mean
	***Pratylenchus* spp.**	***Scutellonema bradys***	***Meloidogyne* spp.**	***Helicotylenchus* spp.**	***Tylenchus* spp.**	***Trichodorus* spp.**	***Tylenchorhynchus* spp.**	***Hoplolaimus* spp.**	***Rotylenchus* spp.**	
Choo	192	318	178	125	142	120	0	90	85	1250
Shebo	271	154	192	215	154	153	0	0	48	1187
Kaswurape	156	274	145	96	116	98	0	0	0	773
Kpachi	242	195	112	56	88	0	0	25	55	885
Kanshegu	362	354	128	180	148	0	96	48	134	1450
Promposo	266	54	184	186	118	176	0	0	96	1080
Ayayo	40	10	406	15	0	20	0	0	0	125
Atrensu	340	273	134	245	195	186	122	0	0	1495
Afrefreso	240	132	117	96	65	68	86	0	0	954
Nokwarease	215	148	252	64	48	0	21	0	0	804

4.5.1 Nematode populations in soils and yam peels during rainy season

Regression analysis revealed a weak positive relationship ($R^2 = 0.188$) between the population of PPN in the soil and those obtained in the yam tuber peels during the rainy season (Fig. 25). The results showed that the model only explains 18.8% of the variability in nematode populations in the soil and the response data (nematode population in yam tuber peels) around the mean.

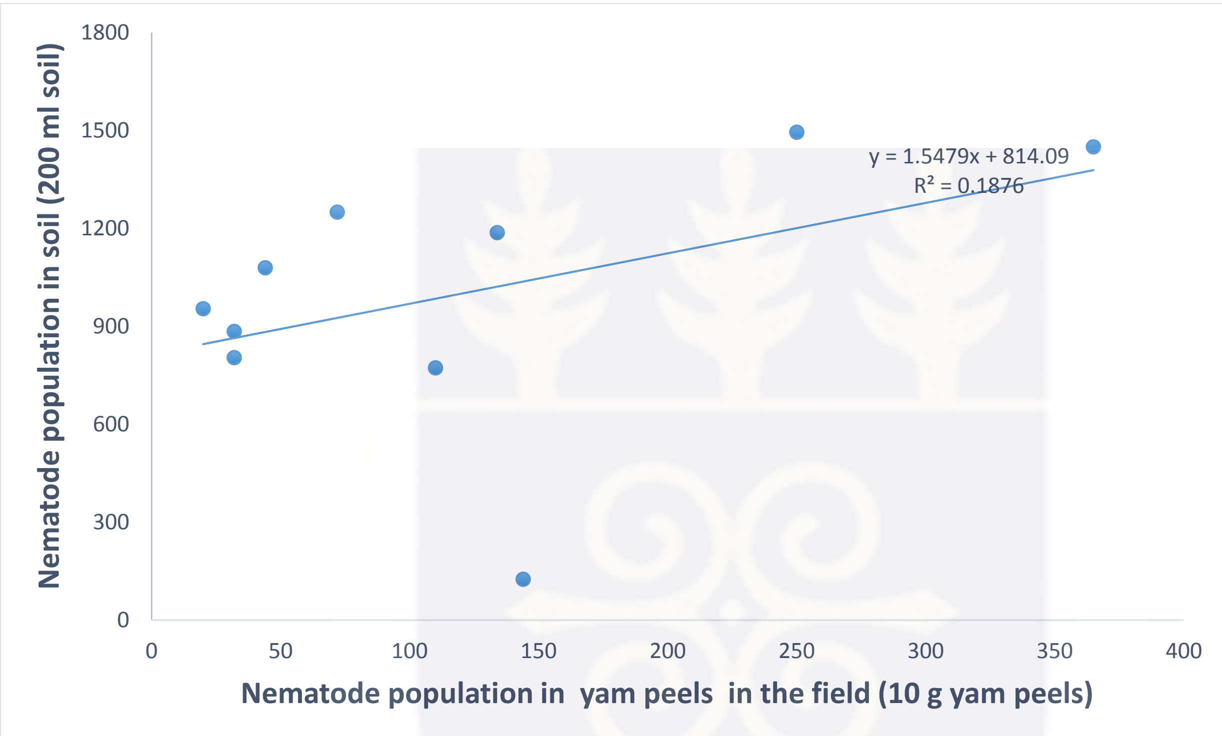


Figure 25. Relationship between plant parasitic nematodes in the field (soil) and yam tubers harvested from same field in the Guinea savanna and Transitional agro ecological zo

An evaluation of yam tubers in storage for PPN recovered 8 nematode genera in the ten districts among the two AEZs. They include, *Pratylenchus* spp., *Meloidogyne* spp., *Scutellonema* spp., *Tylenchorhynchus* spp., *Hoplolaimus* spp., *Trichodorus* spp., *Helicotylenchus* spp. and *Tylenchus* spp. The population densities of PPN recovered ranged from 770 to 1960 nematodes per 10 g of yam tuber peels (Table 21).

Table 21. Mean nematode population (10 g of yam peels) from yam tubers in storage

COMMUNITY	Mean nematode densities of 10g of yam tuber peels during the dry season								Overall mean
	Pratylenchus spp.	*Meloidogyne* spp.	*Scutellonema* spp.	*Tylenchorhynchus* spp.	*Hoplolaimus* spp.	*Trichodorus* spp.	*Helicotylecus* spp.	*Tylenchus* spp.	
Choo	361	282	378	126	131	114	103	142	1637
Shebo	176	112	185	22	42	35	46	152	770
Kaswurape	185	216	285	86	95	76	81	64	1088
Kpachi	342	283	372	113	147	178	98	122	1655
Kanshegu	424	384	466	121	127	154	147	138	1961
Pramposo	261	254	242	76	108	118	96	123	1278
Ayayo	282	364	194	87	104	123	74	66	1294
Atrensu	265	238	346	98	134	185	62	142	1470
Afrefreso	421	278	187	84	126	107	117	111	1431
Nokwarease	348	232	194	59	120	95	46	52	1146

A total of 13,730 nematodes were recovered from the peels of yam tubers in storage for the two ecological zones. *Scutellonema* spp. recorded the highest mean population density of 466 nematodes per 10 g of yam tuber peels among all eight nematode genera in the two AEZs. The predominant genera of nematodes in stored yam tubers among all ten districts was *Pratylenchus* spp. with a population of 3,065 nematodes while *Helicotylencus* spp. recorded the lowest mean population density of 870 nematodes among all eight nematode genera recovered from the tubers.

Also, Kanshegu and Kpachi both in the Guinea savanna ecological zone were the localities with the highest densities (1961 and 1655) respectively of plant parasitic nematodes extracted from stored yam tubers sampled. The lowest population of *S. bradys* was encountered in the tubers sampled from Shebo.

4.5.2 Nematode populations in soils and yam peels in storage during dry season

Further analysis also revealed a significant weak relationship ($R^2 = 0.06$) between the population of PPN in the soil and those recovered from the yam tuber peels in storage during the dry season (Fig. 26). The results also showed the the model only explains 6% of the variability in the nematode populations in the soil and in the yam peels during the dry season.

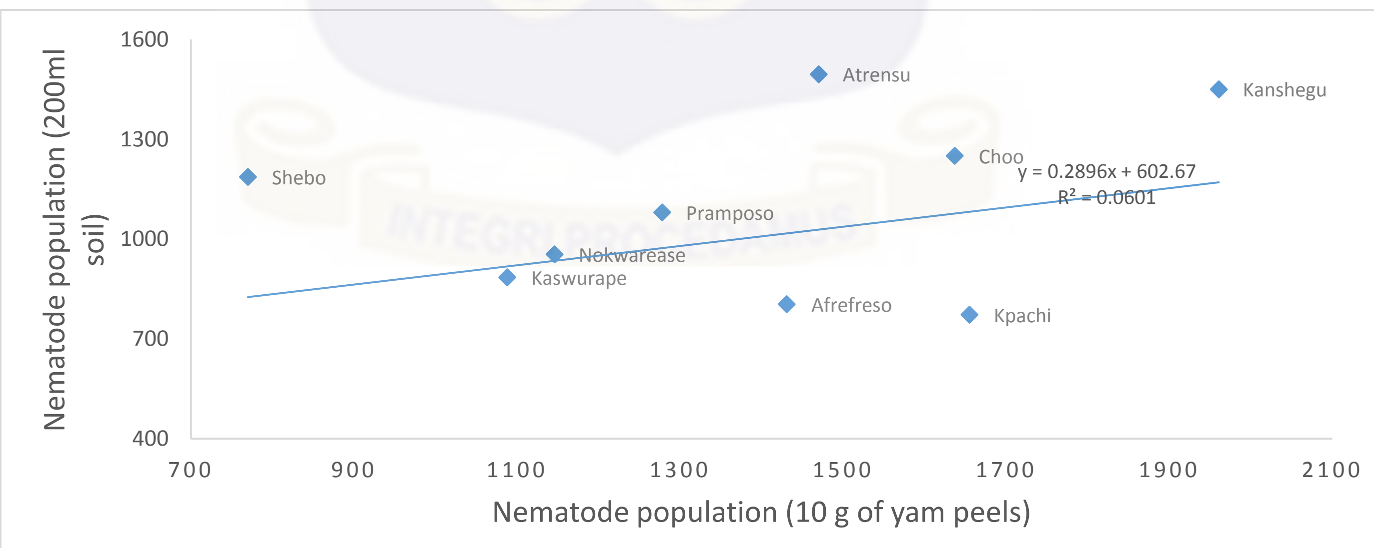


Figure 26. Relationship between density of plant parasitic nematodes in the field (soil) and nematodes in stored yam tubers harvested from the same field in the two agro ecological zone.

4.6 Genera diversity of nematodes in the Guinea savanna and the Transitional rain forest agro ecological zones

There was significant ($p < 0.05$) differences in the diversity of nematodes genera extracted from yam rhizosphere soils across the two AEZs. Simpson diversity indices indicates slight variations among the nematode genera from the different communities. The diversity was highest in Choo followed closely by Shebo (Fig. 27). There was however no significant ($p < 0.05$) difference in the diversity of these two populations.

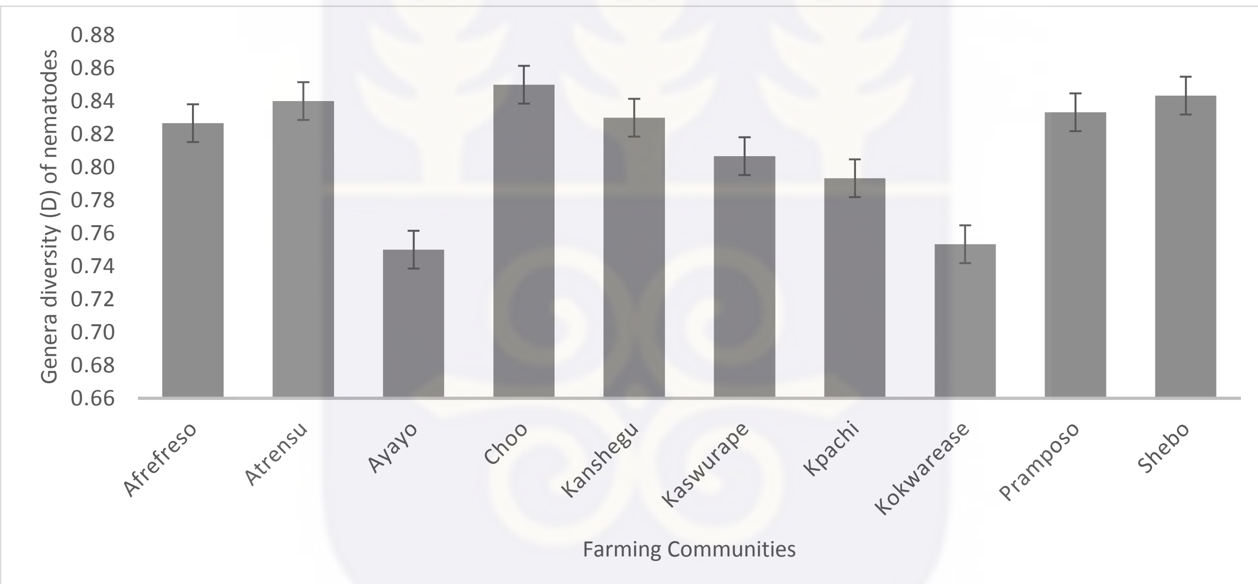


Figure 26. Genera diversity of PPN extracted from soils in the Guinea savanna and Transitional rain forest AEZs (Vertical bars represent standard error).

The genera diversity also indicated slight variations in the nematode genera occurring in stored yam tubers in these communities. Atrensu and Kpachi recorded the highest diversity (0.85) in nematode genera extracted from yam tuber peels. Ayayo, Choo, Kanshegu and Kaswurape all had a diversity index of 0.84 (Fig. 28). Simpson diversity indices for nematode genera extracted from yam tuber peels during the dry season were generally high indicating very high diversity of all nematode genera (Fig. 28).

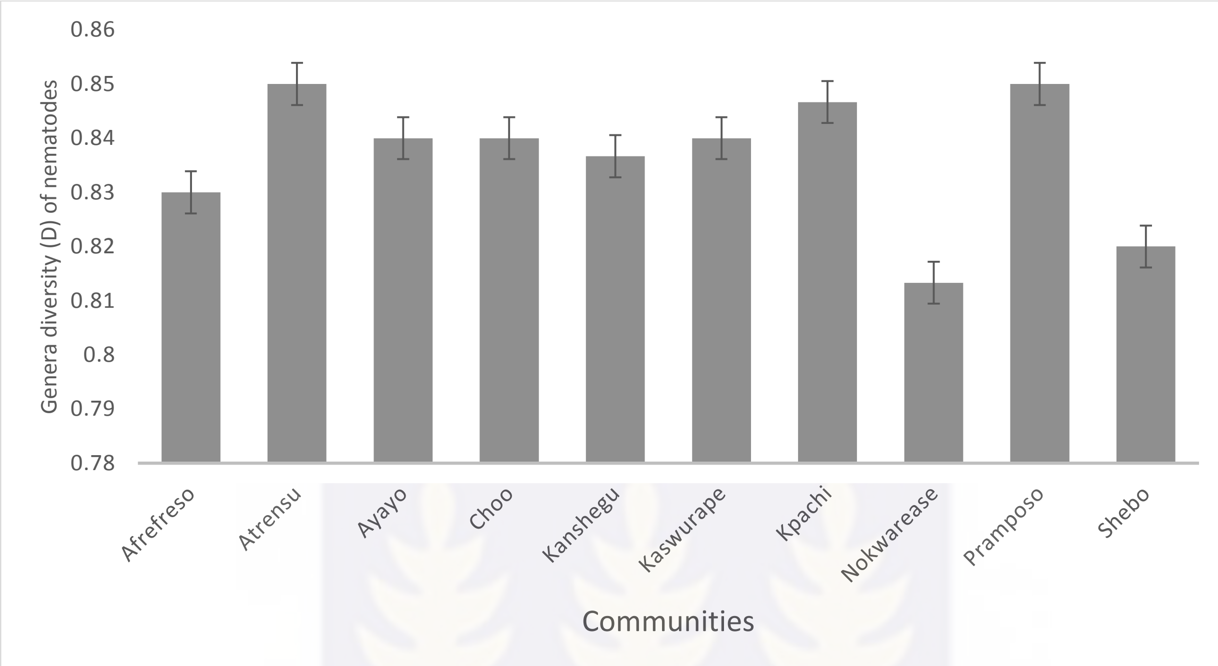


Figure 27. Genera diversity of PPN extracted from yam tuber in storage in the Guinea savanna and Transitional rain forest AEZs (Vertical bars represent standard error).

4.7. Morphometric observation of *Scutellonema bradys* populations

4.7.1 Morphometric observations of *Scutellonema bradys* female population from the Guinea savanna AEZ

Seventeen morphometric characteristics were measured on the female *S. bradys* populations sampled from the Guinea savanna ecological zone (Table 22). The total body length of the Guinea savanna female populations ranged from 865.5 µm to 1012.8 µm (mean: 886.5 µm). Populations from Shebo had the shortest average body length (879 µm) while those of Kpachi had the longest average body length (924 µm). The ‘a’ ratio of the Guinea savanna female population ranged from ranged from 19.4 µm to 30 µm (mean: 24.6 µm). Females fro Shebo (19.4 µm) and those of Kpachi (30 µm) had the highest and lowest a ratio respectively.

Table 22. Morphometrics of scutellonema bradys females from the Guinea savanna ecological zone. Measurements (µm) and ratios are in the form mean ± standard deviation (range)

Location	Shebo	Kaswurape	Kpachi	Kanshegu	Choo
L	378±2.8 (868.5 -989.7)	386.5±35.3 (879.4 - 985.9)	394.9 ± 59.0 (869.3 - 1012.8)	368.5±72.2 (895.3 - 995.5)	912.0 ± 56.8 (891.8 - 988.9)
A	24.9±2.8 (19.4 -28.3)	26.2±2.4 (22.2 - 30.2)	27.0±2.1 (23.2 - 30.0)	26.5±2.6 (21.8 -29.0)	25.6 ± 2.3 (21.7 - 29.0)
C	98.7±13.6 (70.5 -114.1)	100.7±12.5 (87.7 - 120.8)	108.3±12.0 (90.6 - 125.7)	105.8±12.4 (81.3 - 117.1)	105.3 ± 9.3 (90.3 - 116.8)
c'	0.5±0.1 (0.4 - 0.6)	0.5±0.1 (0.4 - 0.6)	0.5±0.1 (0.4 - 0.6)	0.6±0.1 (0.5 - 0.7)	0.5 ± 0.1 (0.4 - 0.6)
S	1.5±0.2 (1.2 - 1.6)	1.5±0.1 (1.3 - 1.7)	1.5±0.2 (1.3 - 1.7)	1.4±0.2 (1.1 - 1.8)	1.4 ± 0.1 (1.3 - 1.5)
Stylet length	22.5±2.3 (19.4 - 25.4	21.3±2.1 (17.1 - 23.6)	23.7 ± 2.9 (18.7 - 28.0)	21.1±3.0 (16.2 - 23.2)	20.6 ± 1.0 (18.6 - 22.1)
Diameter at mid body	29.1±2.4 (26.3 -31.5)	26.7±1.7 (24.5 - 29.3)	28.1±2.3 (25.0 - 31.1)	28.6±1.9 (25.7 - 31.8)	28.6 ± 1.5 (26.4 - 31.8)
Tail length	7.4±1.0 (5.7 - 8.9)	7.2±1.0 (5.7 - 8.9)	7.0±1.0 (5.3 - 8.9)	7.2±0.9 (6.0 - 8.9)	7.0 ± 0.8 (5.9 - 8.3)
Lip region diameter	9.7±1.1 (7.4 - 11.3)	7.4±0.8 (6.2 - 8.5)	3.8±0.9 (7.4 - 9.9)	3.3±1.6 (5.8 - 10.0)	7.1 ± 0.8 (6.0 - 8.8)
Diametre at head region	15.5±0.7 (14.3 - 16.6)	14.6±1.1 (12.4 - 16.2)	15.4±1.4 (13.0 - 18.2)	15.4±1.1 (14.1 - 17.3)	14.3±1.3 (12.4 - 16.4)
Diametre at tail region	14.2±0.7 (13.3 - 15.4)	14.4±1.1 (12.4 - 16.3)	13.2±1.6 (11.3 - 16.1)	12.3±1.3 (10.3 - 14.3)	13.3±1.0 (11.5 - 14.7)
Stylet knob height	3.8±0.5 (3.3 - 4.5)	2.7±0.4 (2.4 - 3.7)	3.4±1.1 (2.1 - 5.3)	2.6±0.6 (2.0 - 3.8)	4.2±0.5 (3.6 - 4.8)
Stylet knob width	4.7±0.4 (3.8 - 5.1)	3.7±0.7 (2.8 - 4.5)	5.1±1.3 (2.2 - 7.0)	4.1±0.4 (3.1 - 4.7)	3.5±0.8 (2.5 -4.9)
Lip region height	4.5±0.6 (3.4- 5.2)	3.5±0.6 (2.4 - 4.3)	3.4±0.6 (2.8 - 4.8)	2.7±0.6 (2.0 - 3.8)	4.2±0.7 (3.4 - 5.6)
Position of vulva to anterior	360.0±34.4 (304.5 - 398.7)	351.4±28.1 (318.5 - 393.7)	368.7±39.9 (319.7 - 438.5)	352.3±36.5 (314.6 - 423.0)	330.5±38.4 (90.3 - 116.8)
Scutellum height	5.4±0.9 (5.4 - 7.7)	5.6±1.2 (4.4 - 8.2)	5.7±0.5 (5.0 - 6.5)	7.7±0.7 (6.8 - 8.5)	5.8±0.8 (2.6 - 3.6)
Scutellum width	3.2±0.9 (2.3 - 5.3)	3.7±0.6 (2.8 - 4.8)	2.9±0.4 (2.2 - 3.5)	3.1±0.7 (2.0 - 3.8)	2.9±0.4 (4.6- 6.7)

a= body length / diameter at mid body, c= body length / tail length, c' = tail length / diameter at tail region, s= stylet length / diameter at head region

Also, female populations from the Guinea savanna ecological zone has stylet length ranging from 16.2 µm to 28 µm (mean: 18.4 µm). Populations from Kanshegu had the shortest average stylet length (16 µm) while that of Kpachi had the longest average stylet length (28 µm). The 'c' ratio of the Guinea savanna female population ranged from 70µm to 125µm. Females from Shebo had the lowest average 'c; ratio whiles that of Kpachi recorded the highest average 'c' ratio. The average tail length of the female population from the Guinea savanna zone ranged from 5.3µm to8.9µm (6.4 µm). Populations from Kpachi had the longest average tail length (8 µm) while that of Kaswurape had the shortest average tail length (5.7 µm). The greatest body diameter of the female population also ranged from 24 µm to 32 µm (mean: 28.2 µm). Female from Kaswurape had the smallest average body diameter (25.3 µm) while that Choo had the highest average body diameter (33.2 µm). The scutellum height of the female populations ranged from 2.6 µm to 8 µm (mean: 5.3 µm). Female populations from Choo has the shortest average scutellum height (3.2 µm) whiles that of Kanshegu recorded the longest average scutellum height (7.2 µm). The position of vulva to anterior of the female populations from the Guinea savanna zon ranged from 286 µm to 468 µm (mean: 324.6 µm). Populations from Choo had the shortest average postion of vulva to anterior (287.8 µm) whiles populations from Kpachi had the longest postion of vulva to anterior (427.5 µm). The average diameter at the tail region of the female population ranged from 10 µm to 16.3 µm (mean: 13.2 µm). Populations from Kaswurape had the largest average diameter at the tail region (15.2 µm) whiles that of Kanshegu had the smallest average diameter at the tail region (14.2 µm).

4.7.2 Morphometric observations for female *Scutellonema bradys* population from the Transitional AEZ

The morphometrics of female *S. bradys* populations collected from th Transitional rain forest agro ecological zone are presented in Table 23. The average total body length of S. bradys female populations from the Transitional rain forest AEZ ranged from 846.3 µm – 1041.8 µm (mean: 928.6 µm). Female *S. bradys* populations from Atrensu has the shortest average body length (852.4 µm) while female populations from Afrefreso has the longest average body length (928.6 µm). Female *S. bradys* populations from the Transitional rain forest AEZ has their 'a' ratio ranging from 19 µm – 30 µm (mean: 23.4 µm). Female populations from Ayayo has the smallest average 'a' ratio (20.2 µm) while female populations from Pramposo has the largest average 'a' ratio (27.6 µm).

The Transitional rain forest male *S. bradys* populations has their 'c' ratio ranging from 73.6 µm – 121 µm (mean: 93.4µm). The lowest 'c' ratio (76.4 µm) was recorded in populations from Nokwarease while the highest 'c' ratio was recorded in populations from Afrefreso (116.3 µm). Also, female *S. bradys* populations from the Transitional rain forest AEZ has their stylet length ranging from 17.3 µm – 25.4 µm (mean: 20.3µm). Female populations from Pramposo has the shortest average stylet length (18.0 µm) whiles female from Ayayo has the longest average stylet length (23.2 µm). The Transitional rain forest zone female *S. bradys* populations has their mid body diameter ranging from 25.2 µm – 34.7 µm (mean: 29.4µm). The smallest average body diameter for the female population was recorded in the Afrefreso populations (26.8µm) while the largest body diameter was recorded in the Ayayo populations (30.1µm). Female *S. bradys* populations from the Transitional rain forest zone has their average position of vulva to anterior ranging from 286.5 µm - 442.7 µm (mean: 324.6 µm). Populations from Ayayo has the shortest average position of vulva to anterior (296.7 µm). Female populations from Nokwarease has the longest position of vulva to anterior (356.8 µm). Also, female *S. bradys* populations from the Transitional agro ecological zone has their scutellum height ranging from 3.3 µm – 7.4 µm (mean: 4.6 µm). Female populations from Pramposo has the shortest average scutellum height (3.7 µm) while the population Atrensu has the highest scutellum height (5.3 µm)

Table 23. Morphometrics of female *scutellonema bradys* from the Transitional rain forest ecological zone. Measurements (µm) are in the form of mean ± standard deviation (range)

Location	Afrefreso	Atrensu	Pramposo	Ayayo	Nokwarease
L	896.5±42.9 (873.5 - 989.8)	854.0±47.7 (846..3 - 1041.8)	853.9±69.6 (851.5 - 998.6)	863.0±51.6 (857.4 - 968.2)	912.3±5.5(868.9 - 987.2)
A	27.0±2.02 (23.3 - 29.3)	25.1±2.5 (21.2 - 29.4)	26.1±2.7 (20.9 - 30.0)	26.4±3.7 (19.2 - 29.7)	24.5±1.9(22.1 - 27.6)
C	96.6±15.7 (75.9 - 121.0)	97.0±11.3 (88.8 - 114.8)	104.2±11.5 (86.2 - 114.3)	101.3±7.0 (90.5 - 116.4)	96.3±13.8(73.6 - 116.2)
c’	0.5±0.1 (0.4 - 0.6)	0.5±0.1 (0.4 - 0.6)	0.5±0.1 (0.4 - 0.6)	0.5±0.1 (0.4 - 0.6)	0.5±0.1(0.4 - 0.6)
S	1.4±0.1 (1.3 - 1.7)	1.5±0.1 (1.3 - 1.7)	1.4±0.2 (1.1 - 1.7)	1.5±0.1 (1.3 - 1.8)	1.3±0.1(1.1 - 1.5)
Stylet length	21.7±1.8 (18.4 - 24.8)	22.9±1.7 (20.7 - 24.8)	20.1±1.9 (17.3 - 23.2)	23.1±1.7 (20.6 - 25.4)	21.9±1.7(19.6 - 24.3)
Diameter at mid body	28.3±2.4 (25.3 - 29.1	29.6±2.2 (26.2 - 34.0)	29.0±1.8 (27.3 - 31.8)	28.9±3.1 (26.1 - 34.7)	31.8±1.9 (29.0 - 34.3)
Tail length	8.1±1.2 (6.1 - 9.6)	7.3±0.8 (6.3 - 8.7)	7.3±0.6 (6.3 - 8.3)	7.4±0.7 (6.4 - 8.2)	8.2±0.9(6.9 - 9.5)
Lip region diameter	8.7±0.4 (8.2 - 9.3)	7.9±1.2 (5.9 -10.0)	7.1±0.8 (6.3 - 78.9)	8.6±1.1 (6.6 - 9.8)	8.6±0.7(7.3 - 9.6)
Diametre at head region	15.4±1.3 (13.7 - 17.8)	15.4±1.0 (14.7 - 17.4)	14.8±0.8 (13.2 - 15.7)	15.6±1.0 (13.7 - 17.0)	16.7±1.2(14.8 - 18.4)
Diametre at tail region	14.2±2.6 (8.9 - 17.8)	14.6±1.3 (12.6 - 16.1)	14.6±0.9 (12.9 - 15.6)	15.2±0.8 (13.8 - 16.5)	15.6±0.7(14.2 - 16.4)
Stylet knob height	2.7±0.5 (2.2 - 3.5)	2.8±0.5 (2.1 - 3.5)	3.1±0.6 (2.6 - 3.5)	3.0±0.6 (2.2 - 4.1)	3.6±0.5(3.9 - 5.2)
Stylet knob width	4.8±1.0 (3.4 - 6.5)	4.2±0.5 (3.7 -4.9)	4.2±0.5 (3.0 - 4.6)	4.1±0.6 (3.2 - 4.8)	4.6±0.5(3.9 - 5.2)
Lip region height	3.7±0.8 (2.1 - 5.0)	2.9±0.4 (2.2 - 3.4)	3.2±0.5 (2.3 - 3.7)	3.9±0.6 (2.5 - 4.7)	3.2±0.4 (2.6 - 3.6)
Position of vulva to anterior	363.2±34.1 (315.3 - 415.6)	361.0±42.7 (297.9 - 436.3)	371.0±34.4 (311.7 - 421.7)	347.7±45.9 (286.7 - 426.2)	385.7±45.4(312.3 - 442.2)
Scutellum height	5.9±0.7 (4.8 - 7.2)	6.4±0.9 (4.7 - 7.7)	4.1±0.7 (2.6 - 4.8)	2.8±0.5 (2.2 - 3.6)	4.3±0.5(3.7 - 5.2)
Scutellum width	2.9±0.3 (2.5 - 3.3)	3.3±0.5 (2.4 - 4.2)	3.2±0.7 (2.2 - 4.6)	6.3±1.3 (4.6 – 5.2)	3.7±0.6(2.5 - 4.3)

a= body length / diameter at mid body, c= body length / tail length, c^1= tail length / diameter at tail region, s= stylet length / diameter at head region

4.7.3 Morphometric observations of *Scutellonema bradys* male populations from the Guinea savanna AEZ

Morphometeric observations *S. bradys* male populations from the Guinea savanna agro ecological zone are presented in Table 24 below. Male *S. bradys* populations from the Guinea savanna zone has their average total body length ranging from 820.6 µm – 1040 µm (mean: 871.4 µm). Male *S. bradys* populations from Choo has the shortest average body length (859.7 µm) while male *S. bradys* populations from Kpachi has the verage longest body length (882.8 µm).

The *S. bradys* male populations from the Guinea savanna AEZ has their 'a' ranging from 20.4 µm – 29.2 µm (mean: 24.7 µm). The smallest average 'a' average was recorded in populations from Kanshegu (22.0 µm) while the highest average highest 'a' was recorded in populations from Choo (26.6 µm). The Guinea savanna population also has a 'c' range of 78.0 µm- 124.4 µm (mean: 91.5 µm). Populations from Shebo has the smallest average 'c' ratio (87.6 µm) while populations from Choo has the highest average 'c' (100.6 µm). Male *S. bradys* populations from the Guinea savanna AEZ has their average stylet length ranging from 16.5 µm – 28.6 µm (mean 21.2 µm). Male populations from Kaswurape has the shortest average stylet length (18.3 µm) while populations from Choo has the longest average stylet length (25.8 µm). The male *S. bradys* populations from the Guinea savanna agro ecological zone has the diameter at mid body ranging from 24.5 µm – 30.6 µm (mean: 27.4 µm). Populations from Choo has the smallest average mid body diameter (24.8 µm) while populations from Kanshegu has the greatest mid body diameter (29.2 µm).

24. Morphometrics of male *scutellonema bradys* from the Guinea savanna ecological zone. Measurements (µm) are in the form of mean ± standard deviation (range)

Location	Shebo	Kaswurape	Kpachi	Kanshegu	Choo
L	368.4 ± 55.1(820.6 - 954.3)	381.3 ± 34.8(876.4 -992.6)	382.8 ± 40.4(984.2 -1040.4)	318± 44.(842 – 988.4.8)	359.7 ± 39.0 (845.6 - 1014.5)
a	25.2 ± 2.1(22.3 - 28.2)	23.8 ± 0.9(22.3 - 24.7)	23.1 ± 0.9(21.9 - 24.1)	22.5 ± 2.2(20.1 - 25.0)	26.6 ± 2.4 (24.6 -29.4)
c	37.6 ± 11.4(81.8 - 100.6)	94.1 ± 18.7(78.0 - 118.5)	96.6 ± 12.0(82.7 - 109.5)	97.0 ± 12.9(80.9 - 111.0)	100.6 ± 15.8 (81.9 - 124.4)
c'	0.5 ± 0.1(0.4 - 0.5)	0.5 ± 0.1(0.5 - 0.6)	0.4 ± 0.1(0.4 - 0.5)	0.5 ± 0.1(0.4 - 0.5)	0.5 ± 0.1 (0.4 - 0.5)
s	1.2 ± 0.1(1.1 - 1.2)	1.2 ± 0.1(1.1 - 1.3)	1.2 ± 0.1(1.1 - 1.3)	1.2 ± 0.1(1.1 -1.3)	1.4 ± 0.2(1.2 - 1.6)
Stylet length	18.9 ± 1.2(17.4 - 20.3)	18.3 ± 1.5(16.2 - 19.6)	18.6 ± 1.2(17.4 - 20.5)	18.2 ± 1.1(16.9 - 19.7)	25.8 ± 1.6 (24.4 -28.5)
Diameter at mid body	26.6 ± 1.4(24.5 - 28.1)	27.8 ± 1.2(25.9 - 28.7)	28.7 ± 1.6(26.7 - 30.6)	29.2 ± 1.0(27.9 - 30.3)	25.8 ± 1.6 (24.4 - 28.5)
Tail length	7.7 ± 0.9 (6.3 - 8.5)	7.2 ± 1.1(5.6 - 8.4)	6.9 ± 0.8(5.8 - 7.7	6.8 ±0.8(5.9 - 7.6)	6.9 ± 0.8 (5.8 - 7.7)
Lip region diameter	6.6 ± 0.5(5.9 - 7.1)	6.4 ± 0.4(5.9 - 7.1)	7.6 ± 0.8(6.7 - 8.4)	7.5 ± 1.0(6.4 - 8.8)	7.0 ± 0.5(6.4 - 7.8)
Diametre at head region	16.1 ± 1.2(14.1 - 17.3)	15.2 ± 1.5(13.7 - 17.7)	15.3 ± 0.7(14.3 - 16.2)	15.6 ± 0.6(14.8 - 16.4)	15.4 ± 1.0 (14.4 - 16.8)
Diametre at tail region	16.9 ± 0.9 (15.5 - 17.7)	13.8 ± 0.8(12.5 - 14.3)	15.4 ± 0.6(14.7 - 15.9)	15.0 ± 0.4(14.6 - 15.6)	14.5 ± 0.6 (13.8 - 15.6)
Stylet knob height	2.9 ± 0.3 (2.6 - 3.4)	2.6 ± 0.3 (2.3 - 3.1)	3.3 ± 0.5(2.7 - 3.7)	3.4 ± 0.7 (2.7 - 4.4)	2.9 ± 0.8 (2.2 - 4.2)
Stylet knob width	5.4 ± 0.7 (4.5 - 6.5)	4.7 ± 0.6(4.2 - 5.3)	5.0 ± 0.2(4.8 - 5.3)	4.5 ± 0.7(3.6 - 5.5)	4.5 ± 0.6 (3.8 - 5.3)
Lip region height	3.5 ± 0.5 (2.8 - 4.2)	3.6 ± 0.5 (2.9 - 3.7)	4.0 ± 0.7(3.1 - 4.8)	3.9 ± 0.3(3.7 - 4.5)	4.0 ±0.5 (3.5 - 4.5)
Spicule length	27.4 ± 1.0 (26.4 - 29.7)	29.0 ± 1.0 (28.8 -33.3)	29.7 ± 1.5(28.9 - 32.2)	28.9 ± 0.9(27.9 - 31.2)	27.9 ± 0.6 (26.5 - 31.8)
Scutellum height	5.0 ± 0.5 (4.4 - 5.7)	5.6 ± 0.6 (4.8 – 6.2)	6.2 ± 0.6(5.7 - 6.8)	5.5 ± 0.6(4.9 - 6.4)	5.2 ± 0.7 (4.3 - 5.9)
Scutellum width	3.1 ± 0.3 (2.7 - 3.4)	3.1 ± 0.8 (2.3 - 4.3)	3.8 ± 0.4(3.4 - 4.4)	3.0 ± 0.6(2.6 - 3.7)	3.1 ± 0.7(2.1 - 3.7)

a= body length / diameter at mid body, c= body length / tail length, c'= tail length / diameter at tail region, s= stylet length / diameter at head regio

S. bradys male populations from the Guinea savanna zone has their average tail length ranging from 5.6 µm – 8.5 µm (mean: 7.2 µm). The shortest average tail length was recorded in population from Kanshegu (6.8 µm) while the average longest tail length for the *S. bradys* male populations was recorded in the Shebo populations (7.7 µm). *Scutellonema bradys* male populations from the Guinea savanna zone has their average spicule length ranging from 26.4 µm – 33.3 µm (mean: 28.8 µm). Male populations from Shebo has the shortest average spicule length (27.4 µm) while populations from Kpachi has the longest average spicule length (29.7 µm) Male *S. bradys* populations from the Guinea savanna agro ecological zone has a scutellum height ranging from 4.4 µm – 6.8 µm (mean: 5.6 µm). The average shortest scutellum height was recorded in populations from Shebo (5.0 µm) while the longest average scutellum height was recorded in populations from Kpachi (6.2 µm).

4.7.4 Morphometric observations of *Scutellonema bradys* male populations from the Transitional rain forest AEZ

The morphometric observation from *S. bradys* male population from the Transitional rain forest agro ecological zone are presented in Table 25. Male *S. bradys* populations from the Transitional rain forest zone has their total body length ranging from 834.9 µm – 1023.1 µm (mean: 905.7 µm). Male populations from Ayayo has the shortest average body length (845.1 µm) while that of Nokwarease has the longest average body length (942.7 µm). The male *S. bradys* populations has an 'a' ratio ranging from 22.1 µm – 29.5 µm (mean: 25.2 µm). The lowest average 'a' ratio was recorded in populations from Pramposo (24.1 µm). Male populations from Afrefreso has the highest average 'a' ratio (27.0 µm).

Table 25. Morphometrics of male *scutellonema bradys* from the Transitional rain forest ecological zone. Measurements (µm) are in the form of mean ± standard deviation (range)

Location	Afrefreso	Atrensu	Pramposo	Ayayo	Nokwarease
L	915.5 ± 11.3 (894.8 - 981.2)	868.1 ± 30.8 (858.3 - 1018.7)	912.0 ± 45.5 (896.5 - 994.3)	845.1 ± 43.5 (834.9 - 976.5)	942.7±60.1 (879.5 - 1023.1)
a	27.0 ± 1.0(25.4 - 28.1)	24.5 ± 1.4(23.0 - 26.5)	24.1 ± 2.3(22.1 - 26.7)	25.7 ± 2.9(22.5 - 29.5)	25.5±2.4(22.4 - 27.7)
c	115.5 ± 16.4(92.5 - 130.1)	98.0 ± 10.1(83.0 - 107.0)	110.2 ± 15.4(90.7 - 128.7)	100.2 ± 12.4(91.5 - 121.5)	98.5±14.4(83.0 - 113.9)
c'	0.4 ± 0.1(0.4 - 0.5)	0.4 ± 0.1(0.4 - 0.5)	0.4 ± 0.1(0.3 - 0.5)	0.4 ± 0.1 (0.4 - 0.5)	0.5±0.1(0.4 - 0.6)
s	1.3 ± 0.1(1.2 - 1.5)	1.0 ± 0.1(1.0 - 1.1)	1.2 ± 0.2(1.1 - 1.4)	1.2 ± 0.1 (1.1 - 1.2)	1.1±0.1(1.0 - 1.2)
Stylet length	20.8 ±1.1(19.5 - 22.5)	17.8 ± 0.9(16.4 - 18.9)	19.0 ± 1.6 (17.4 - 21.8)	18.5 ±1.5(16.6 - 20.1)	18.7±0.7(17.8 - 19.7)
Diameter at mid body	26.0 ± 0.9(24.7 - 27.0)	28.6 ± 1.9(26.6 - 31.8)	27.6 ± 0.9(26.5 - 28.6)	26.4 ± 1.5(24.6 - 28.6)	30.1±1.6(28.6 - 32.4)
Tail length	6.2 ± 0.9(5.4 - 7.5)	7.2 ± 0.5(6.8 - 7.9)	6.1 ± 0.7(5.2 - 6.9)	6.8 ± 0.6(5.8 - 7.5)	7.8±0.7(6.9 - 8.6)
Lip region diameter	6.9 ± 0.4(6.6 - 7.4)	9.3 ± 0.7(8.5 - 10.3)	7.3 ± 0.7(6.5 - 8.3)	8.4 ± 0.8(7.5 - 9.6)	7.5±0.7(6.7 - 8.4)
Diametre at head region	15.5 ± 0.8(14.7 - 16.7)	17.1 ± 1.1(15.4 - 18.4)	15.7 ± 1.2(14.6 - 17.5)	16.1 ± 1.2(14.4 - 17.7)	17.7±0.6(16.9 - 18.2)
Diametre at tail region	13.9 ± 0.9(12.5 - 14.7)	16.4 ± 1.5(14.5 - 17.6)	14.5 ± 0.8(13.5 - 15.7)	15.2 ± 0.9(14.5 - 16.7)	14.8±0.7(13.7 - 15.4)
Stylet knob height	3.1 ± 0.4(2.7 - 3.5)	3.1 ± 0.5(2.7 - 3.7)	3.3 ± 0.3(2.8 - 3.5)	2.8 ± 0.4(2.4 - 3.4)	3.4±0.6(2.7 - 4.1)
Stylet knob width	4.8 ± 0.5(4.2 - 5.4)	4.6 ± 0.4(4.3 - 5.4)	4.6 ± 0.7(3.6 - 5.7)	4.1 ± 0.5(3.5 -4.8)	5.3±0.6(4.6 - 6.2)
Lip region height	3.4 ± 0.5(2.7 - 3.8)	4.0 ± 0.7(3.1 - 4.8)	4.0 ± 0.4(3.6- 4.5)	3.8 ± 0.8(2.6 - 4.6)	4.1±0.4(3.7 - 4.6)
Spicule length	28.3 ± 0.9(25.0 – 34.5)	29.4 ± 0.9(27.8 - 34.9)	28.6 ± 1.2(26.5 - 31.2)	29.4 ± 0.7(28.6 - 34.4)	28.7±0.7(27.7 - 31.4)
Scutellum height	6.0 ± 0.5(5.7 - 6.6)	4.4 ± 0.9(3.4 - 5.7)	5.2 ± 0.7(4.5 -6.2)	4.8 ± 0.5(4.3 - 5.7)	6.1±1.1(4.5 - 7.4)
Scutellum width	3.6 ± 0.8(2.8 -4.8)	3.5 ± 0.7(2.7 - 4.5)	3.0 ± 0.4(2.8 - 3.5)	3.0 ± 0.4(2.6 - 3.7)	4.8±0.7(3.8 - 5.6)

a= body length / diameter at mid body, c= body length / tail length, c'= tail length / diameter at tail region, s= stylet length / diameter at head region

The Guinea savanna male *S. bradys* population has a 'c' ratio ranging from 83.0 µm – 130.1 µm (mean: 108.7 µm). Male populations from Atrensu has the lowest average 'c' ratio (98 µm) while male populations from Afrefreso has the highest average 'c' ratio (115.5 µm). Male *S. bradys* populations from the Guinea savanna zone has their average stylet length ranging from 16.4 µm – 22.5 µm (mean: 19.1 µm). Populations from Atrensu has the shortest average stylet length (17.8 µm). Afrefreso populations has the longest average stylet length (20.8 µm). The Guinea savanna male *S. bradys* populations has their diameter at mid body ranging from 24.7µm – 32.4 µm (mean: 28.4 µm). Male S. bradys populations from Afrefreso has the smallest mid body diameter (26.0 µm) while male populations from Nokwarease has largest mid body diameter (30.1 µm).

Scutellonema bradys male populations from the Guinea savanna AEZ has their average tail length ranging from 5.2µm – 8.6 µm (mean: 6.8 µm). The populations from Afrefreso has the shortest average tail length (5.4 µm) while male populations from Nokwarease has the longest average tail length (7.8 µm). The Guinea savanna *S. bradys* male populations has their average spicule length ranging from 25.0 µm – 34.9 µm (mean: 28.9 µm). Male populations from Afrefreso has the shortest average spicule length (28.3 µm). The longest average spicule length was recorded in populations from Atrensu (29.4 µm). *Scutellonema bradys* male populations from the Guinea savanna agro ecological zone has their scutellum height ranging from 3.4µm – 7.4µm (mean: 5.5 µm). Male populations from Atrensu has the shortest average scutellum height (4.4 µm) while male populations from Nokwarease has the longest average scutellum height (6.1 µm).

4.8.1 Principal component analysis of ten *Sctellonema bradys* female populations

The first two principal components accounted for 51.0 % of the total variance of morphometric characters for the ten *S. bradys* female populations (Table 26). Factor 1 (F1) is dominated by high positive weights for total body length, position of vulva to anterior, tail length and ratio a. Factor 2 (F2) is dominated by high positive weights for stylet length and ratio 's'.

Table 26. Eigenvectors and eigenvalues of principal components derived from nematode morphometric characters for ten *Scutellonema bradys* female populations from two AEZs

	Principal components	
Variables	**F1**	**F2**
Total body length	**0.59**	0.136
Stylet length	-0.067	**0.631**
Tail length	**0.337**	-0.219
Position of vulva to anterior	**0.543**	0.072
Scutellum height	-0.115	0.008
Diametre at tail region	0.181	-0.127
a	**0.415**	0.279
s	-0.145	**0.66**
Eigenvalue	2.274	1.789
Variability (%)	28.427	22.365
Cumulative %	28.427	50.792

a=body length/body diameter, s=stylet length/diameter at head region, F1=Factor 1, F2=Factor 2

In the PC analysis, variations in female *S. bradys* species from the ten populations is explained by total body length, position of vulva to anterior, tail length, stylet length, ratios 'a' and 's'. When factors 1 and 2 were plotted on a biplot, the result showed that variations in factor 1 is as result of high values for total body length, position of vulva to anterior and a ratio and low positive values for diameter at tail region and tail length (Fig. 29). Variations in factor 2 was also as a result of high values for stylet length and ratio ‘a’ and negative value for scutellum width.

4.8.2 Relationship between morphometric characters of female *Scutellonema bradys*

A correlation matrix of eight female morphometric variables revealed a strong positive correlation between position of vulva to anterior and total body length, ratio ‘a’ and ratio ‘s’ and stylet length (Table 27). There was also weak positive correlations between tail length and total body length, position of vulva to anterior and tail length, diameter at tail region and tail length, ratio a and position of vulva to anterior. It also showed negative correlations between ratio s and tail length, diameter at tail region and scutellum height.

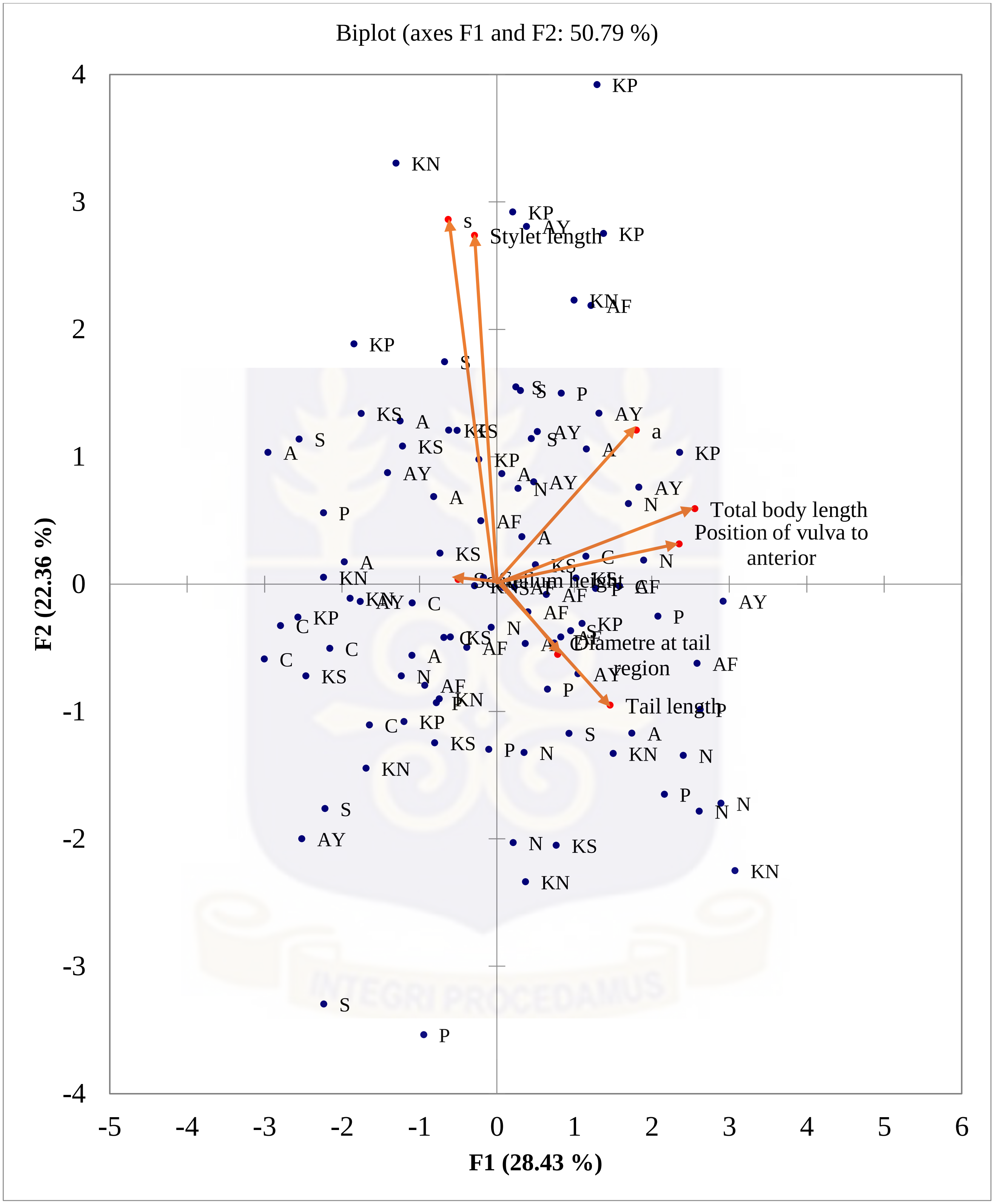


Figure 28. Biplot showing relationships between *Scutellonema bradys* morphometric characters *Scutellonema bradys* female populations

Table 27. Correlation matrix (Pearson (n) of *Scutellonema bradys* female morphometric variables

Variables	Total body length	Stylet length	Tail length	Position of vulva to anterior	Scutellum height	Diametre at tail region	a	s
Total body length								
Stylet length	-0.017							
Tail length	**0.333**	-0.072						
Position of vulva to anterior	**0.677**	0.038	**0.244**					
Scutellum height	-0.067	-0.089	-0.073	-0.058				
Diametre at tail region	0.047	-0.015	**0.258**	0.173	**-0.353**			
A	**0.602**	0.056	0.014	**0.373**	0.021	-0.065		
S	-0.029	**0.705**	**-0.244**	-0.132	0.014	-0.049	0.105	

Values in bold are different from 0 with a significance level alpha=0.05

4.8.3 Relationship between variables and factors of *Scutellonema bradys* female populations

A correlation between factors and *S. bradys* female morphometric variables shows that F1 has a stronger positive correlation for tail legth, total body length, and postion of vulva to anterior (Table 28). Factor 2 (F2) also has stronger positive correlation for stylet length.

Table 28. Correlations between *Scutellonema bradys* female morphometric variables and principal components

Variables	F1	F2
Total body length	0.889	0.182
Stylet length	-0.1	0.844
Tail length	0.508	-0.293
Position of vulva to anterior	0.818	0.097
Scutellum height	-0.174	0.011
Diametre at tail region	0.273	-0.17
A	0.626	0.373
S	-0.218	0.883

4.8.4 Principal component analysis for ten *Scutellonema bradys* male populations from the two AEZs

Principal component analysis performed on *S. bradys* male populations from all ten districts in the two AEZs shows that the first four factors account for 83.5% of the variability in the variables that were measured (Table 29). Factor 1 (F1) is dominated by negative weights for stylet length, ratio ‘s’ and ‘c’, and a positive weight for tail length. Factor 2 (F2) is also dominated by high positive weights for diameter at tail region, stylet knob width and scutellum width.

Table 29. Eingenvectors and eingenvalues of factor analysis derived from nematode morphometric characters for populations of *Scutellonema bradys* males from different locations in the Guinea savanna and the Transitional rain forest agro-ecological zones of Ghana.

	Principal components	
Variables	**F1**	**F2**
Total body length	-0.033	0.391
Stylet length	**-0.407**	0.093
Diametre at head region	0.259	**0.558**
Stylet knob width	-0.033	**0.425**
Scutellum width	-0.015	**0.401**
Tail length	**0.521**	-0.144
c	**-0.489**	0.29
s	**-0.504**	-0.284
Eigenvalue	2.4	1.74
Variability (%)	30.002	21.744
Cumulative %	30.002	51.746

A biplot displaying *S. bradys* populations projected on the plane of F1 and F2 shows that variations in the male S. bradys populations is explained by tail length, diameter at tail region, scutellum width, stylet knob width and total body length (Fig. 30). The distribution of male *S. bradys* species on the first two principal axis accounts for 51.75% of the total variation and is as a result of differnces in tail length and diameter at tail region.

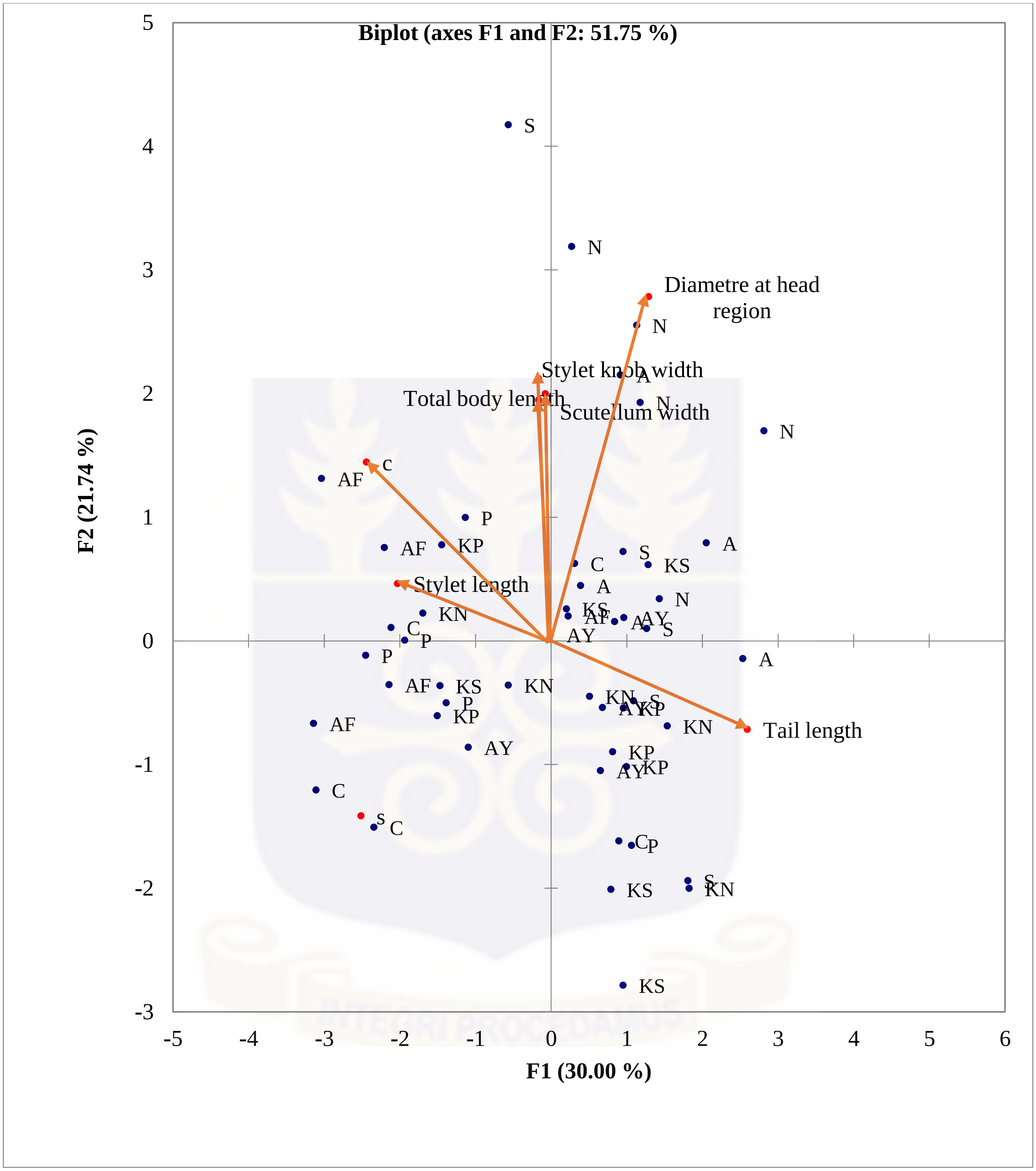


Figure 29. Biplot showing relationships between *Scutellonema bradys* morphometric variables *S. bradys* male populations for F1 and F2

4.8.5 Correlation matrix among *Scutellonema bradys* male morphometric variables

The correlation matrix among the morphometric variables shows a strong positive correlation between ratio ‘s’ and stylet length (Table 30). There were weak positive correlations between the ratio ‘c’ and total body length, stylet knob width and scutellum width with negative correlations between ratio ‘s’ and diameter at head region, and ratio ‘c’ and tail length.

Table 30. Correlation matrix (Pearson (n) of *Scutellonema bradys* male morphometric variables

Variables	**Total body length**	**Stylet length**	**Diameter at head region**	**Stylet knob width**	**Scutellum width**	**Tail length**	**c**	**s**
Total body length								
Stylet length	-0.042							
Diametre at head region	0.241	0.100						
Stylet knob width	0.090	0.181	0.206					
Scutellum width	-0.096	0.116	0.213	**0.432**				
Tail length	0.037	-0.265	0.155	0.030	-0.072			
C	**0.432**	0.216	-0.058	-0.007	-0.001	**-0.873**		
S	-0.135	**0.685**	**-0.618**	0.068	-0.036	-0.307	0.225	

Values in bold are different from 0 with a significance level alpha=0.05

4.8.6 Relationship between variables and factors in male *Scutellonema bradys* populations

The correlation between factors and *S. bradys* male morphometric variables also shows that F1 has a strong positive correlation for tail length (0.807) and negatively correlated with stylet length (-0.631) and ratio 'c' (-0.758) . Factor 2 (F2) has strong positive correlation for diameter at head region, stylet knob width, scutellum width and total body length (Table 31).

Table 31 Correlations between *Scutellonema bradys* male morphometric variables and principal components

Variables	F1	F2
Total body length	-0.051	**0.515**
Stylet length	**-0.631**	0.123
Diametre at head region	0.401	**0.736**
Stylet knob width	-0.051	**0.561**
Scutellum width	-0.024	**0.529**
Tail length	**0.807**	-0.189
c	**-0.758**	0.383
s	**-0.781**	-0.374

CHAPTER FIVE

5.0 DISCUSSION

Field survey and analysis to assess knowledge and perception of *Dioscorea* species farmers in the Guinea savanna and Transitional rain forest ecological zones revealed that majority of the yam farmers (93.3%) were males. This could probably be because of the physical nature of the cultural practices involved in yam production. Despite similar labour requirements and costs, female participation in yam production in Western Nigeria increased as result of increasing returns (Manyong *et al.*, 1996). Most *D.* species farmers in both agro-ecological zones had no or very little formal education because, most individuals with secondary or tertiary education seeked for jobs in the cities and thus, leave the farming in the hands of those who have little or no formal education. Sesay *et al.* (2013) revealed that, farming in Sierra Leone remained in the hands of individuals with little or no formal education, because the highly educated moved to the cities for white colar jobs. The lack of knowledge about nematodes may have contributed to the high prevalence of nematode diseases in both agro-ecological zones.

Most farmers in the two agro ecological zones recycled their planting materials. They obtained their plating materials from their own farms with a few getting theirs from family members and the market because of the high cost of seed yam (Nweke *et al.*, 1991; MiDA, 2010). Farmers usually produced their own planting materials, and occasionally obtained some from their neighbours and markets (Bridge, 1987; Kwoseh *et al.*, 2005). The yam tuber serves as the major source of inoculum for the yam nematode, and its movement from one location to the other contributes to the the spreads of nematodes within and between farms.

Dioscorea species cultivars grown by farmers in the two ecological zones were 'Pona,' 'Lareboko,' 'Fuseinbilla', 'Fimo', 'Asobayere', 'Afishe', Matches, 'Lobere', 'Gunguma' and Deddii in decreasing order of popularity. 'Pona' and 'Pareboko' even though were perceived by most farmers to be highly susceptible to nematode infection, still ranked highest in terms of production by farmers in both zones. This was because, these two cultivars had high market value in both ecological zones. Studies have revealed that 'Pona' and 'Lareboko' were the most preferred cultivars both in the domestic and export markets (MiDA, 2010). All *Dioscorea* species were however perceived to be susceptible to nematode infection albeit at different levels (Bridge, 1982; Kwoseh, 2000; Coyne *et al.,* 2006).

Majority of the farmers have resorted to continuous cultivation of their fields even though this was done by rotation with crops such as maize, garden eggs, cassava, cowpea, pigeon pea, millet, sorghum, pepper, tomatoes and soybean. This continuous cultivation has become the major practice especially in the transitional rain forest zone. This practice has intensified due to increasing population in most farming communities and households. The practice has also aggravated the nematodes problem both in the field and in storage mainly due to majority of crops rotated and intercropped to yam were susceptible hosts of nematodes. Intensification of production systems due to unavailability of farming lands aggravates the nematode problems in yam production (Plowright and Kwoseh, 1998; Kwoseh, 2000; Kwoseh *et al.*, 2005).

Different crop components were also planted by farmers and there were various yam/crop mixtures been practiced in both ecological zones. Farmers grew these crop mixtures because they matured early, for food security and for income before yam was harvested. Coursey and Booth (1977) reported that yams were intercropped to other crops to minimize risks associated with diseases, price variability, and to ensure all year round availability of food.

The vulnerability of yam to nematode parasitism reduce tuber quality and yields through root and tuber gallings and lesions, together with dry and soft rots (Jatala and Bridge, 1990) These also depends on the species of plant- parasitic nematode present in the soil. Farmers estimated losses in storage to be about 25 % and 20% in the Guinea savanna and Transitional zones respectively. Quantities of losses were also observed to have a positive correlation with the length of storage. Kwoseh *et al.*, (2005) estimated loses in storage due to nematode parasitism in Ghana to be about 30% and 21% in the Guinea savanna and Transitional forest zones, respectively. In Nigeria, losses of between 25% to 75% due to *Scutellonema bradys* infection have been recorded within storage periods of 16 weeks (Nwauzor, 1982). Storage losses of up to 80 % have also been reported in Nigeria (Hahn *et al.*, 1989).

Farmers in the two agro-ecological zones produced *Dioscorea* species mainly for consumption (food security), income generation, and purchasing of assets. Failure by farmers to meet their expected yields affects their abilities to settle their wards school fees, medical bills, redeem outstanding loans, and acquire assets. In order to overcome nematode problems associated with yam, research should be geared towards generating and maintaining sustainable healthy seed systems so that farmers can have greater access to seed material that will result in more productivity.

A large number of PPNs were recovered from both the rhizosphere of yam and the yam tubers in the field and in storage. These nematodes were found in all ten districts across the two AEZs. Intensive and mixed cropping might be responsible for the relatively rich nematode genera where yams were sometimes intercropped or rotated with cassava, pigeon pea and cowpea (Luc, *et al.*, 2005).

The upsurge in human population has resulted in intensification of agriculture which promotes favorable conditions for certain species of plant parasitic nematodes (Wallace, 1971; Tiyagi *et al.*, 1987; Abd- Elgawad *et al.*, 2007). Plant parasitic nematodes have been reported to constitute serious impediments to yam production in various parts of the world (Adesiyan *et al.*, 1975; Hahn *et al.*, 1989; Abebe and Geraert, 1995; Weber *et al.*, 1995; Kwoseh *et al.*, 2005; Adegbite *et al.*, 2006; Baimey, 2006; Kumar and Singh, 2010). In our study, high numbers of PPNs were encountered in the soil and in the yam tubers in storage during the dry season. Only three nematode genera, *Meloidogyne* spp., *Pratylenchus* spp and *Scutellonema bradys* were, however, encountered in the yam tubers in the field during the rainy season and in lower densities in all ten districts across the two AEZs. Almost all the nematodes genera that were encountered in the soil during the rainy season, were also recovered from the yam tubers in storage during the dry season, and in relatively higher numbers. *Pratylenchus* spp. was the most common PPN in yam tubers both in the storage and in the field and soils. This nematode was found to have a high frequency of occurrence in samples from all AEZs. *Pratylenchus* spp. were previously identified to have a higher relative abundance in 13 nematode genera in soils planted to yam within the Transitional rain forest region of Ghana and Uganda (Mudiope *et al.*, 2001; Osei *et al.*, 2004). The high populations of *Pratylenchus* spp. in nearly all the studied areas in both AEZs makes this an important nematode of yam. Most farmers used crops that were hosts to *Pratylenchus* spp. and *S. bradys* in their crop rotation cycles and as intercrops. Forty nine (49%) of farmers intercropped their yam with cassava, cowpea, and pigeon pea. *Pratylenchus* spp. and *S. bradys* have been found to be associated with pigeon pea, cowpea, and cassava (Luc *et al*., 2005). Ten genera of plant-parasitic nematodes have been identified to be associated with yam in Ogun and Osun states of Nigeria (Bridge and Page, 1984; Adegbite *et al*., 2005). In their study, they identified *Scutellonema* spp., *Meloidogyne* spp., and *Pratylenchus* spp. as the most important nematodes limiting yam production. Caveness (1982) found 28 plant-parasitic nematodes associated with yams in West and Mid-West, East and North of Nigeria and listed four genera, (*Scutellonema* spp., *Pratylenchus* spp., *Meloidogyne* spp. and *Rotylenchulus* spp.). Adesiyan and Odihirin (1977), identified six genera of plant parasitic nematodes (*Scutellonema* spp., *Pratylenchus* spp., *Meloidogyne* spp., *Helicotylenchus* spp., *Criconemoides* spp and *Xiphinema* spp.) in Mid-West State of Nigeria.

A comparison of *S. bradys* in our study with others described in the literature shows some similarities with e.g., body length (820µm – 1040µm), 'a' ratio (19µm - 30µm) and stylet length (16µm - 29µm) (Goodey, 1952, Sher, 1964, Siddiqi, 1972, Germani *et al.*, 1985, Van de Berg *et al*., 2013, Humphery *et al*., 2014). Body was loose when relaxed and slightly arcuate (Siddiqi, 1972). Scutellum was rounded at the level of anus. The tail is rounded, measuring 6.5µm – 8.5µm in length. Stylet was described as well developed with oval basal knobs (Germani *et al.*, 1985). Spicules were slightly cephalated and ventrally arcuate and about 29µm - 33µm length (Siddiqi, 1972). The lip region of the nematodes were highly variable (Ali *et al.*, 1973) and also varied in their height and width (Van den Berg and Heyns, 1973).

Principal component analysis could not enable the separation of ten *S. bradys* female and male populations based on a combination of 8 morphometric characters. The best morphometric characters for the separation of *S. bradys* populations (body length, stylet length, tail length, position of vulva to anterior, scutellum height, diameter at head region, ratios 'a', 'c' and 's') corresponded with most characters commonly used to separate *S. bradys* (Humpherey *et al.,* 2014; Van de Berg *et al.,* 2013). For the females and male, 79.4% and 83.5% respectively, of the variation could be explained by differences in eight morphometric characters.

The result of the study has serious implications as it has shown that most farmers in the two agro ecological zones lack or have little knowledge about about plant parasitic nematodes. Seed yam has been observed as the major inoculum of nematodes associated with yam (Kwoseh *et al*., 2005; Baimey, 2006). The occurrence and effect of nematodes associated with yam on the yam tubers is not cultivar specific (Coyne *et al.,* 2006). The quantity of yam tuber losses as a result of nematode infection is dependent on the length of storage of the tubers (Hahn *et al.*, 1989; Coyne *et al.,* 2006; 2011). *Pratylenchus* species and *Scutellonema bradys* were found to be the most predominant nematode genera associated with yam in the two agro ecological zones and therefore poses serious impediment to yam production in these two agro ecological zone (Osei, *et al.*, 2004; Adegbite *et al.*, 2005)

CHAPTER SIX

6.0 CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

- The result of the study indicates that yam farmers in the Guinea savanna and Transitional rain forest AEZs were well aware of the presence of nematodes parasitism and the physical damage they caused to their yams in the fields and in storage. They were however ignorant about the causal organisms and the spread of the disease within and among farms. Although farmers believed nematode parasitism cannot be controlled, some used cultural methods to manage nematode parasitism. Nematode parasitism impacted negatively on the livelihood of farmers.
- Nematode genera extracted and identified from both Guinea savanna and the transitional rain forest agro ecological zones were ; *Pratylenchus* spp., *Meloidogyne* spp., *Scutellonema bradys*, *Helicotylenchus* spp., *Tylenchorhynchus* spp., *Trichodorus* spp., *Rotylenchus* spp., *Tylenchus* spp. and *Hoplolaimus* spp., *Pratylenchus* spp., *Scutellonema* spp. and *Meloidogyne* spp.
- Nematode populations were found to be higher in the Guinea savanna ecological zone both in the soil (6258 per 200 cc of soil) and yam tuber peels (7824 per 10 g of yam peels) as compared to that of of Transitional zone (4941 per 200 cc of soil) and (5959 10g yam tuber peels).
- The *Scutellonema* species found in Ghana is *S. bradys* as it shares similar morphological and morphometric characters with other *S. bradys* from other yam growing regions in West Africa.

- Principal component analysis of the morphometric variables could however not distinctly detect variations among the 10 female and male populations of *S. bradys* from the two AEZs.

6.2 Recommendations

- Education of yam farmers in the two Agro ecological zones on the causes and mode of spread of nematode parasitism could change farmers' perception and contribute to effective management of nematode parasitism.
- Other multivariate techniques for example, canonical discriminant analysis must be explored to identify presence of morphometric variations in populations.
- The use of scanning electron microscope should be explored to further aid in the identification of species through their morphology.
- In order to validate these morphology-based methods and to detect variations within populations if present, molecular characterization of the yam nematode from these populations should be carried out.

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APPENDICES

APPENDIX 1: QUESIONNAIRE

SURVEY ON NEMATODES ASSOCIATED WITH YAM PRODUCTION IN THE GUINEA SAVANNA AND TRANSITIONAL RAIN FOREST AEZs OF GHANA

Documentation of farmers' knowledge, perceptions, and experiences concerning occurrence and management of nematodes associated with yam production.

QUESTIONAIRE NO ……….

A. Demographic characteristics

1. Name of farmer ……………………………………………….
2. Sex: Male [1] Female [2]

3. Age of respondent:

< 20	=	1
21 – 30	=	2
31 – 40	=	3
41 – 50	=	4
> 50	=	5

4. Level of education: Primary [1] Middle sch. /JHS [2] Secondary [3] Tertiary [4] none [5]

5. Name of district ……………………………

6. Name of locality …………………………..

7. How long have you been farming yam? 1 – 5yrs [1]: 6 – 10yrs [2]: 11 – 15yrs [3] >15 [4]

8. Size of farm, year of cultivation and source of planting material

Year of cultivation	Size of farm (Acre)	Source of planting materials
2011		
2012		
2013		
2014		

Codes: Size of farm: 0-1 acre [1], 1- 2 acres [2], 3-4 acres [3] 5-8 acres [4] more than 8 acres [5]

Source of planting materials: own farm [1] family member [2], market [3], others (specify)…….

9. What are the varieties of yam grown in your farm?

Variety	Proportion of farm

Code for proportions: < 1 acre [1], 1-2 acres [2], 3-5 acres [3], >5 acres [4]

B. Land use intensity

10. How long have you cultivated this land 1-3 years [1] 4-6 years [2] 7-10 years [3] 11- 15years [4] more than 15 years [5]

12. What type of crop(s) have you planted on this piece of land in the past five (5) years? .

Yam [1]

Cassava [2]

Pigeon pea [3]

Maize [4]

Others (specify)………………………………

13. How long have you planted yam on this piece of land: 1-3 years [1], 4-6 years [2] 7-10 years [3], more than 10 years [4]

14. Have you rotated your yam crops before Yes [1] No [1]

15. If yes, with what crop(s) ………………………

16. What was the period of rotation: 1-2 year [1], 3-5 years [2], More than 5years [3]

17. Have you practiced land fallow in the past? Yes [1], No [2]

18. If yes, how long was the fallow period? 1 year [1], 2-4 years [2], 5-10 years [3]

C. **Farmers knowledge, perception and experiences concerning occurrence of nematodes on farm and in storage.**

19. Do you experience any pests and disease problems in your yam crops during the growing season?

Yes [1] No [2]

20. Do you know what nematodes are? Yes [1] No [2]

21. Which of your varieties do you observe these symptoms more: ………………………………

22. Which of these symptoms and signs do u observe in your yam field?

Symptom/Sign	Yes [1] / No [2]

1. 2. Necrotic streaks or lesions on roots	
3. Reduced rooting system	
4. Internal rotting or discolorations of vegetative organs	
5. Dwarfing	
6. Stunting and resetting	

23. What are the conditions of the soil in your field in which these signs/symptoms are prevalent?

a. Lowland (Waterlogged) [ ] b. Well drained soil [ ] c. Sandy - clayey [ ]

d. Sandy soil [ ] e. Loamy soil [ ] f. Clayey soil [ ]

24. Where do you store your yam after harvest?

a. In a pit at the farm []

b. Yam barn at home/farm []

c. In a store at home []

d. In a farmstead []

25. Do you experience any loses in storage due to pests and diseases? Yes [1] No [2]

26. If yes, what signs/symptoms do you observe in the stored tubers?

Sign/Symptom	Yes [1] / No [2]
1. Hardened and dried tissues with varying discolorations	
2. External cracks on skin of tubers	
3. Significant weight loses in tubers	
4. Light yellow lesions below the outer skin of the tuber	
5. Rotting of the outer 1 – 2cm layer.	
6. Flaking off- of external coverings exposing patches of dark brown	
7. General decay of tubers	

Codes for response: Yes [1] No [2]

D. Farmers knowledge, perception and experiences concerning control/ management

27. How does nematodes spread? [A] Infected tubers [B] Soil [C] Heat [D] Prolong drought

28. Are you able to manage / control nematode infestation? a. [Yes] b. [No]

29. If Yes, what are some of the management practice used in your farm?

Management Practice	Yes/ NO

1. Crop rotation	
2. Use of Nematicides	
3. Land rotation	
4. Use of organic/ inorganic fertilizers	
5. Others (specify)	

Response codes: Yes [1] No [2]

30. If No, give reasons. A. [no reason] b. [I do not know how to control them] c. Other, (please specify)………………………………......................................

E. **Farmers knowledge, perception and experiences concerning economic importance of nematodes**

31. What are some of the reasons why you cultivate yam? ……………………………………..

32. Estimated loses as result of nematode infestation.

Year	Farm size (acres)	Expected yield (100 tubers)	Quantity harvested (100 tubers)	Quantity stored (100 tubers)	Percentage loses (%) due to nematodes
2011					
2012					
2013					
2014					

33. Where actual yields higher than expected yield, give reasons:

No reason [1], Favorable rainfall [2], Use of inorganic/ organic fertilizers [3], High soil fertility [4], Others (specify)……….

34. Please explain the effect of nematode parasitism on the following:

I. Money to pay children's school fees………………………………………………………..

II. Money to repay loans…………………………………………………………………………

III. Money to buy assets…………………………………………………………………………

IV. Money to pay medical bills………………………………………………………………

V. Your relationship with extension officers…………………………………………………

VI. Your relationship with your spouse and neighbours…………………………………………..

APPENDIX II: Pictures of infected yam tubers used to administer the questionnaire

APPENDIX III: ANOVA for Simpson diversity index for nematode counts

Variate: soil Simpson diversity index

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Community	9	0.03558667	0.00395407	79.08	<.001
Residual	20	0.00100000	0.00005000		
Total	29	0.03658667			

Variate: Rainy season yam tuber Simpson div. index

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Community	9	0.2283867	0.0253763	27.89	<.001
Residual	20	0.0182000	0.0009100		
Total	29	0.2465867			

Variate: yam tubers instorage Simpson div. index

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Community	9	0.00406667	0.00045185	45.19	<.001
Residual	20	0.00020000	0.00001000		
Total	29	0.00426667			