# OCCURRENCE AND ANTIMICROBIAL SUSCEPTIBILITY PROFILES OF *DERMATOPHILUS CONGOLENSIS* ISOLATES FROM SLAUGHTERED CATTLE IN ABUJA, NIGERIA

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**AUGUST, 2018**

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**BY**

**Kingsley OPUTEH DVM (ABU) 2009**
**(P14VTPM8034)**

**A DISSERTATION SUBMITTED TO THE SCHOOL OF POSTGRADUATE STUDIES, AHMADU BELLO UNIVERSITY, ZARIA**

**IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF THE DEGREE OF MASTER OF SCIENCE IN VETERINARY MICROBIOLOGY**

**DEPARTMENT OF VETERINARY MICROBIOLOGY,**
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**AUGUST, 2018**

## DECLARATION

I declare that the work in this dissertation entitled **"Occurrence and Antimicrobial Susceptibility Profiles of *Dermatophilus congolensis* Isolates from Slaughtered Cattle in Abuja, Nigeria"**has been performed by me in the Department of Veterinary Microbiology, Faculty of Veterinary Medicine, Ahmadu Bello University Zaria under the supervision of Dr.K.A. Majiyagbe and Dr. J. Adamu. The informations derived from the literature areduly acknowledged in the text and a list of references provided. No part of this dissertation has been presented for another degree or diploma at this or any other institution.

Kingsley OPUTEH---------- ----------

Signature Date

## CERTIFICATION

This dissertation entitled, **"OCCURRENCE AND ANTIMICROBIAL SUSCEPTIBILITY PROFILES OF *DERMATOPHILUS CONGOLENSIS* ISOLATES FROM SLAUGHTERED CATTLE IN ABUJA, NIGERIA"** by Kingsley OPUTEH, meets the regulations governing the award of the degree of Master of Science of Ahmadu Bello University Zaria and is approved for its contribution to scientific knowledge and literary presentation.

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## DEDICATION

This dissertation is dedicated to my ma, Mrs Victoria Oputeh whose arms around me have kept me going through the years.

## ACKNOWLEDGEMENTS

I wish to express my profound gratitude to God Almighty to whom all praises are, for His infinite mercies, blessings, guidance, and protection all through my years of study, granting strength, perseverance and courage to complete this study despite all odds, to my parents and all my siblings for their tremendous prayers and support throughout my life especially during the period of this study.

My sincere gratitude to my supervisors Dr.K.A. Majiyagbe, (who sadly passed away before the completion of this study; may his gentle soul continue to rest in peace) and Dr. J. Adamu for their encouragement, guidance, patience and supervision throughout the period of the research, and also for scrutinizing the work meticulously, diligently and painstakingly putting me through this wonderful learning experience, and also for their invaluable contributions to making this work a success.

I wish to appreciate Dr. Alfa of the Agricultural and Rural Development Secretariat FCTA, Dr. Zakari and Mr James of the Karu abattoir, Dr. J. Dalis, Mr Y. Sadiq and Mr Emmanuel of the Dermatophilosis Research Department, N.V.R.I Vom, Mrs Salamatu and Mr. Dodo of Veterinary Microbiology Department, Faculty of Veterinary Medicine ABU, Zaria for all their selfless efforts in making sure this work comes to a successful completion.

I cannot forget to thank my friends for all their support and encouragement through the years especially Drs. E.R. Edeh, A.F. Ogundele, D.E. Awai and I.A. Igbaver (for assisting with the statistical analysis for this work).Finally I want to sincerely thank Miss Juliet Kuyet Bulus for always being a real pillar of support.

**ABSTRACT**

Antibiotic resistance has long been recognized as a major problem in the treatment of bacterial infections including dermatophilosis. This study was carried out to determine the occurrence and antimicrobial susceptibility profiles of *Dermatophilus congolensis*isolates obtained from slaughtered cattle at the Karu abattoir in the Federal Capital Territory Abuja, and to determine the rate of isolation of the bacterium during the wet and dry periods respectively. One hundred and thirty five (135) skin scabs were obtained from animals suspected to be infected with *Dermatophilus congolensis*based on observed clinical signs. Ninety two (68.1%) of these suspected cases occurred during the wet period (August to October) while 43 (31.9%) were in the dry period. The scab samples were subjected to both direct examination with methylene blue staining and culture for isolation on blood agar to determine and confirm the presence of *D. congolensis*. The *Dermatophilus*isolates obtained were tested against eight commonly used antibiotics namely; Augmentin, Cloxacillin, Gentamicin, Cefuroxime, Ceftriaxone, Ceftazidine, Ofloxacin and Erythromycin. Following direct examination, all 135 skin scab samples were found to be positive for *D. congolensis* with characteristic branching filamentous rods, however only 53 (39.3%) yielded typical *D. congolensis* isolates via culture and isolation.Out of the 53 isolates tested to eight commonly used antibiotics, the majority (94.3%) of the isolates were sensitive to Ofloxacin, 42 (79.2%) to Ceftazidine, 26 (49.1%) and 21 (39.6%) to Gentamicin and Ceftriaxone respectively. On the other hand, all the isolates(100%) were resistant to Cefuroxime, Erythromycin, Cloxacillin and Augmentin at concentrations up to 250µg/ml. The study established the presence of *D. congolensis* among slaughtered cattle in the region.There were no statistical association in the rate of *D. congolensis* isolation between the breeds encountered or the sex of the animals examinedwith the rate of detection of the organism. However the highest caseof the infection was recorded during the rainy period.In conclusion there is a need for clinicians to carry out antimicrobial susceptibility testing on isolates of *D.congolensis* to

determine the most effective antibiotics against specific isolates for the best treatment outcome.

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## LIST OF ABBREVIATIONS

º Degree

% Percent

£ British Pound

µl Microliter

µm Micrometer

BA Blood Agar

BHI Brain Heart Infusion

Bw Body weight

CBPP ContagiousBovine Pleuropneumonia

CIE Counter immunelectrophoresis

CLSI Clinical Laboratory Standard Institute

Cm Centimeter

$CO_2$ Carbondioxide

D.C *Dermatophilus congolensis*

E East

ELISA Enzyme Linked Immunosorbent Assay

EUCAST European Committee on Antimicrobial Susceptibility Testing

FAO Food and Agriculture Organization

FAT Florescent Antibody Test

FCT Federal Capital Territory

FCTA Federal Capital Territory Authority

g Gram

hr Hour

ITA International technology association

Kg Kilogram

MB Methylene blue

MBC Minimum bactericidal concentration

MIC Minimum inhibitory concentration

mg Milligram

Min Minutes

ml Millititer

mm Millimeter

N North

NA Nutrient agar

N.V.R.I NationalVeterinary Research Institute

PCR Polymerase chain reaction

# CHAPTER ONE

# INTRODUCTION

## 1.1 Background of the Study

Dermatophilosis is an acute, subacute or chronic skin disease that affects a wide range of domestic and wild animal species as well as humans and is characterized by the development of an exudative epidermitis followed by scab formation, alopecia and thickening of the skin. (Zaria, 1993).

The disease was first reported by Van-Saceghem (1915) in cattle in the Belgian Congo (Shoorijeh *et al*., 2008). Even though the disease affects a wide variety of animals and occasionally humans, the most commonly affected species are cattle, sheep and horses. Dogs and cats are rarely affected in many parts of the world (Dejene *et al*., 2012).

The disease in cattle and sheep is commonly called cutaneous streptotrichosis and mycotic dermatitis, respectively and in horse rain scald.Other local names exist including Senkobo skin disease in Central Africa, Kirchi in Nigeria, and Saria in Malawi. Dermatophilosis is a name common to the disease in all species (Radostits *et al*., 2007).

The causative agent of dermatophilosis is *Dermatophilus congolensis* whichis classified in the Family *Dermatophilaceae*, Order *Actinomycetales*. It is a pleomorphic, Gram positive, non-acid-fast, aerobic and facultative anaerobic bacterium. It has two characteristic morphologic forms: filamentous hyphae and motile zoospores (Zaria and Amin, 2004).

The zoospores can remain dormant in the absence of adequate stimulus such as moisture, and in this state they are highly resistant to dessication (Roberts, 1963). Motile zoospores are not very resistant to physical and chemical changes such as pH, osmotic changes and dessication; these conditions normally result in decreased viability after a few hours outside the scab lesions, but in favourable substrate such as the persistently moistened skin of a suitable host,

they lose their motility, and germinate (Zaria and Amin, 2004).It can be cultured on or in most ordinary laboratory media but optimal growth occurs on blood agar and brain heart infusion agar (Zaria, 1993).

Dermatophilosis occurs in all areas of the world and can be epizootic in tropical and subtropical areas of the world (Radostits *et al*., 2007;Dalis *et al*., 2010). Survey of large number of cattle in Africa revealed prevalence rates approaching 15% with a 100% infection rate in some herds at the time of peak seasonal prevalence (Gebreyohannes and Gehresselassie, 2013). In temperate climates the disease is usually sporadic but can still have considerable economic importance where predisposing factors persists (Radostits *et al*., 2007). High prevalence in sheep flock occurs in high and medium rainfall areas (Aitken, 2007).

Transmission occurs from the carriage lesions by contact from the face of one animal to the fleece or skin of another, and from the feet to the skin during mounting. Transmission can be direct or indirectly through contaminated water or grass. Insect transmission which has been demonstrated with flies and ticks is believed to be a principal means of spreading zoospores (Quinn *et al*., 2002).

There is also breed differences in susceptibility to Dermatophilosis. In Africa, the N'dama and Muturu cattle breeds and native sheep are resistant, while Zebu, White Fulani, and European breeds are susceptible(Radostits *et al*., 2007). Within breeds, differences in susceptibility are also apparent and genetic markers have been identified in Zebu. (Radostits *et al*., 2007).

A provisional diagnosis of dermatophilosis can be made on clinical signs but should be confirmed by the demonstration of the organism by microscopic examination of Giemsa- or Gram-stained smears prepared from the lesions, by histopathological examination of biopsies of lesions or by cultivation and identification of the organism (Zaria and Amin, 2004).

The most important aspect of control involves removal of factors predisposing to infection especially control of ticks and other arthropod vectors as well as good husbandry and management practices to ensure avoidance of prolonged wetness and macerations.

With the disease that occurs in tropical areas and associated with tick infestation parenteral treatment with antibiotics, can be used and should be used in conjunction with Acaricides (Stewart, 1997).

Dermatophilosis leads to great economic losses in African countries (Samui and Hugh-Jones, 1990; Woldemeskel, 2000)due to inferior wool and leather quality (Edwards, 1985; Alley *et al.,* 1987), death and culling (Yeruham *et al.*,2000a), decreasedmilk production and increase in somatic cell count (Yeruham *et al.*,2000b), decrease in semen quality (Sekoni, 1993)and the treatment expenses incurred during the management of the disease. In addition to its economic importance, the disease plays a role in public health and can be transmitted to humans (Harman *et al.,* 2001; Burd *et al.,* 2007).

There is no single treatment specific for dermatophilosisbut antibiotic treatment is the one which is most widely used and effective for cure from the clinical signs and to reduce the complications due to secondary infections. Several antibiotics were found to be effective with variable levels of success.

## 1.2 Statement ofthe Research Problem

Dermatophilosis is considered by the Food and Agriculture Organization (FAO, 1982) annual health services report as one of the four major bacteriological diseases affecting cattle and other animals in the tropics, consequently affecting the development of the livestock industry in these regions especially Africa(DeRyke *et al.,* 1991). The other diseases include contagious bovine pleuropneumonia (CBPP), haemorrghagic septicaemia and brucellosis (DeRyke *et al.,* 1991). The economic effects of dermatophilosis have been described as enormous,thisis mainly due to the fact there is high incidence of the disease in tropics and

subtropics especially Africa and the infection leads to severe losses in different sectors of animal production causing reduced quality of hides and skin as well decreased meat production especially in its chronic form(DeRyke *et al.,* 1991).

The economic losses by dermatophilosis were described by Lloyd as chiefly due to two factors; the downgrading of hides and secondly lowering of meat production, but other economic losses have also been reported which include low milk production, failure in reproduction and loss of draught cattle (Lloyd, 1971).

The economic importance of the disease is estimated in millions of dollars, due to the loss in productivity in terms of work by infected draft oxen(Oppong, 1976), wool, hides and skin (Woldemeskel,2000), decreased milk and meat production (Dalis *et al.,* 2007), a failure in reproduction in cows with severe vulva infection (Ate*et al.,* 2007) and stud bulls with severe leg lesion making them unable to mount (Iliyasu*et al.,* 2015).

In addition, dermatophilosis has placed restrictions on the establishment of high producing exotic cattle breeds and also affected the upgrading of indigenous breeds by cross breeding them with exotic breeds because this breed of cattle are highly susceptible to the infection(Lloyd, 1971).

The treatment of dermatophilosis still remains a matter of great concernowing to the recurrence of the disease and the difficulties to cure it using antibiotics by the parenteral route (Nath *et al.,* 2010). The focus is still on topical treatment of dermatophilosis, so many other treatments were tested mixing several natural drugs, regardless of the risk of toxicity and without scientific protocol, but none of them gave complete healing without recurrence (Nwufoh, 1985; Ali-Emmanuel *et al.,* 2003).

Problem of antibiotic resistance due to indiscriminate use of antibiotics has long been recognized in most bacterial infection including dermatophilosis leading to failure in treatment.

Although the prevalence and epidemiology of the disease in some parts of Nigeria has been established but there is lack of information on the occurrence and prevalence of the disease in thefederal capital territory (FCT) particularly among slaughtered cattle bearing in mind that it is a zoonotic disease.

### 1.3 Justification of the Study

Although, there are varying estimations of cattle population in Nigeria ranging between 10and 15million (Bourn *et al.,* 1986; Ademosun, 1987; Adesehinwa *et al.,* 2004) the mean average of the nation's cattle population was put at 13.9 million as at 1990 (Blench, 1998), clearlyruminants play significant roles in the social and economic wellbeing of Nigerians in various ways. Economically the animals serve as source of income earning to major ruminants' dealers- sellers of live animals and butchers/meat sellers; generates employments and creates markets for larger number of people who explore the animals' product and by-products for economic gains. Meat constitutes the foremost animal product that is highly explored by the Nigerian households, particularly for direct consumption and as such, the ruminants, especially cattle, constitute the major and cheapest source of meat consumption for most households in the country (ITA, 2004), about 1million cattle are annually slaughtered for meat in the country. This suggests heavy dependence on cattle for meat consumption by households in the country. In a country in which malnutrition is an ever increasing problem and where the need for high quality protein is critical, the control of dermatophilosis could make an important contribution (Lloyd, 1976).

Many researchers as well as clinicians have documented difficulty in treatment of this infection (Zaria, 1993; Ali-Emmanuel*et al.,* 2003) and resistance of the organism to various antibiotics (Kruger *et al.,* 1998; Mannan *et al.,* 2009) as different strains of the organism have shown different and diverse antimicrobial susceptibility patterns. The study is therefore aimed at providing information on the antimicrobial susceptibility profiles of the strains of

*D.congolensis* isolated from the FCT and also provide information on the current status of the disease among sedentary herds and cattle brought for slaughter at the Karu abattoir which will eventually assist clinicians in selection of the most suitable antibiotics for treatment and control of the infection.

## 1.4 Aim and Objectives of the Study

### 1.4.1 Aim of the study

To determine the occurrence and antimicrobial sensitivity profiles of *D congolensis* isolates from cattle in the Federal Capital Territory (FCT) Abuja, Nigeria.

### 1.4.2 Objectives of the study

1. To determine the rate and level of occurrence of dermatophilosis among cattle population in Abuja during dry and wet periods.
2. To isolate *D. congolensis*from cattle presented for slaughterand those from sedentary herds in Abuja.
3. To determine antibiotic susceptibility profiles (*in vitro*)of the *Dermatophilus*isolates obtained from cattle in FCT, Abuja.

## 1.5 Research Questions

1. To what extent does occurrence of dermatophilosis among cattle populations in Abuja differs between wet and dry periods?
2. What could be the rate of isolating *D. congolensis* from cases of dermatophilosis in cattle in FCT Abuja?
3. What are the antibiotic susceptibility profiles ofthe *D. congolensis* strains isolated from cattle in FCT Abuja?

# CHAPTER TWO

# LITERATURE REVIEW

## 2.1 Aetiology of Dermatophilosis

*Dermatophilus congolensis* is a Gram-positive, coccobacillary actinomycete that causes an exudative dermatitis in a variety of species, notably in ruminants and horses, although rare infections occur in cats, dogs and humans. (Yager and Scott, 1993; Ambrose, 1996; Burd *et al.*, 2007). Infection is generally restricted to the skin and subcutaneous tissue, although uncommon involvement of lymphoid tissue in ruminants and cats has been described (Jones, 1976; Gibson *et al.*,1983).

The classification of the causative agent of dermatophilosis was chaotic in the early period of the discovery of the disease. The first description of the organism was reported by Van-Saceghem(1915) who found the lesion to contain a branching, filamentous organism which divided both transversely and longitudinally to form spores, he named the organism *Dermatophilus congolensis.*

The organism associated with dermatophilus infection in sheep was initially called *Actinomyces dermatonomus*(Bull, 1929). Because of the close relationship of the sheep pathogen with the agent of bovine cutaneous Streptothricosis, another name was later suggested as *Actinomyces congolensis* by Hudson (1937). The second organism causing Strawberry foot-rot in sheep was first isolated and named as *Dermatophilus pedis* (*Polysepta pedis*) by Thompson (1954). These ovine mycotic dermatitis pathogens and the cattle pathogen, *D. congolensis* was earlier considered to be morphologically and culturally different, they are now believed to be the same organism which are reclassified in a new family *Dermatophilaceae* in the order of *Actinomycetales.* The family has a single genus *Dermatophilus* (Austwick, 1958). The three organisms were however, found to be one species and designated all under the name *Dermatophilus congolensis* (Gordon, 1964; Roberts, 1965; Stewart, 1972).

## 2.2 Morphology of *Dermatophilus congolensis*

*Dermatophilus congolensis* is a gram positive, pleomorphic, non-acid fast, aerobic actinomycete. It has two characteristic morphologic forms: filamentous hyphae and motile zoospores. The hyphae are characterized by branching filaments (1-5μm in diameter) that ultimately fragment by both transverse and longitudinal septation in to pockets of coccoid cells(Andrew *et al*., 2003). The coccoid cells mature in to flagellated ovoid (0.6-1μm in diameter)zoospores,the dormant zoospores are resistant to dessication and may remain viable even after heating at $100^0$C for up to 30minutes, but the zoospores become motile 24hr after re-establishment of warm, humid conditions and can migrate from moistened scab debris (Roberts, 1963b). The migration of the zoospores maybe accelerated as a result of negative chemotaxis from their endogenous carbondioxide. The mycelial elements encountered in the lesions consist of coccoid cells arranged longitudinally in a symmetrical chain, which may consist of a single row or of thick pillars of up to eight parallel rows of coccoid cells (Chodnik, 1956). Occasionally, some asymmetrical and irregular scattered cocci are seen in some filaments.

### 2.2.1 Growth requirements of *Dermatophilus congolensis*

Gordon (1976) reported that strains of *Dermatophilus congolensis* grew well aerobically at $36^0$C on various media types, while others have different experience under the same condition. It was reported that growth under aerobic condition was poor, but was improved with anaerobiosis and was best under micro-aerophilic condition (Macadam and Haalstra, 1971). Memery (1966) reported that, carbon dioxide ($CO_2$) exerted no significant influence on the growth of the organism. Macadam and Haalstra (1971) and Roberts (1963b) however found out that the growth of the organism was much more stimulated in the presence of $CO_2$.

The optimum growth temperature for *Dermatophilus congolensis* was found by most workers to be $37^0$C (Van Sacegham 1916; Harris 1948; Bugyaki 1959; Macadam and Haalstra

1971).Masters *et al* (1995) however reported $27^0$C to be the optimum growth temperature for isolates from turtles,which may reflect adaptation to poikilothermic hosts in a coolaquatic environment.The organism grows on most ordinary laboratory media but best on blood agar (BA), Brain heart infusion (BHI) agar and Tryptone broth; abundant growth has been reported on Loeffler serum medium and Dorset's egg medium (Macadam and Haalstra, 1971). Memery (1966) reported no growth on coagulated egg albumin medium and growth on chick embryo caused death after four days. No growth was reported on Sabouraud dextrose agar, Potato dextrose agar, McConkey agar, Deoxycholate citrate agar and Czapeks solution medium (Pridham *et al*., 1957; Ainsworth and Austwick, 1959; Bugyaki, 1959; Memery, 1966; Macadam and Haalstra 1971).

### 2.2.2 Cultural characteristics of *Dermatophilus congolensis*

Culturally, many workers reported great variations in the colonial appearance of the organism. A single strain often gives rise to diverse forms on different media under varying conditions (Zaria, 1993). Gordon (1976) described a wide range of colonial appearance suggesting strain differences. At 24 hours aerobic culture on blood agar plates incubated at $37^0$C, it shows typicaltiny round or irregular grayish-white, conical, very rough granular colonies usually tough and adherent and pitting the medium (Macadam and Haalstra, 1971; Gordon, 1976; Masters *et al*., 1995; Buenviaje *et al*.,1997; Samuel *et al*., 1998). The colonies are surrounded by a zone of beta hemolysis. Short aerial hyhae were observed by Macadam and Haalstra (1971). On further incubation, the colour of the colonies changed to yellowish and then orange, smooth, opaque and large in size, with increased zone of beta hemolysis (Austwick, 1958; Macadam and Haalstra, 1971; Gordon, 1976). The report on pigment production by different strains also varies. Macadam and Haalstra (1971) however found that most strains tested formed a pale yellow pigment after four days of incubation.

Strain variation in the rate of growth was observed more under microaerophilic condition than in aerobic condition and this variation crossed host and species line (Gordon, 1976). Masters *et al*. (1995) reported the use of different techniques to study the morphology of the organism. These include transmission electron microscopy to study flagella, mature filaments after negative staining with Indian ink as described by Cowan and Steel (2004), and capsular thickness from measurement of photographic prints.

In liquid media, growth appears as thick sediment with a clear supernatant fluid. Some strains form a hard adherent ring, while other strains form a flocculent ring at the surface. Other strains were reported to form small, hard, discrete colonies that were submerged and adherent to wall of the tube. The sediment was generally flocculent either granular or flaky, but in some, it was ropy or taffy-like (Gordon, 1976; Macadam and Haalstra, 1971). Some strains have been reported to produce shiny deposits (Thompson, 1954; Macadam and Haalstra, 1971).

### 2.2.3 Biochemical characteristics of *Dermatophilus congolensis*

Bacteriological examination of various strains isolated from various animal species demonstrated only slight variation in the biochemical reactions of *D. congolensis* strains. Generally, the organisms have little saccharolytic activity with the production of acid from glucose and fructose within 2 days of incubation (Macadam and Haalstra, 1971; Gordon, 1976), laevulose was also reported to be acidified by the majority of the strains tested but less intensely acidified (Macadam and Haalstra, 1971). Maltose, galactose, mannitol and sucrose were acidified by a few strains, most of which were isolated from cattle, sheep and goats, and trehalose was fermented by only two sheep strains while lactose was fermented by one strain from sheep and cattle (Van Saceghem, 1934; Macadam and Haalstra, 1971). These sugars were not, however, fermented by the strains tested by Gordon (Gordon, 1976). None of the *D. congolensis* strain examined produce acid in xylose, dulcitol, sorbitol, salicin adonitol,

aesculin, arabinose, erythritol, inositol, inulin raffinose or rhaminose (Macadam and Haalstra, 1971; Gordon, 1976).

All strains examined were found to produce catalase (Macadam and Haalstra, 1971;Gordon, 1976; Oppong, 1976). There was no strain reported to produce indole and none gave methyl-red and voges-prauskeur positive reactions (Macadam and Haalstra, 1971;Gordon, 1976). The majority of the strains were found not to reduce nitrate except few strains were reported to give a positive reaction (Bugyaki, 1959; Macadam and Haalstra, 1971).*D. congolensis* failed to produce hydrogen sulphide (Macadam and Haalstra, 1971) but a positive reaction was recorded by Bugyaki (1959). Some strains of the organism were reported to hydrolyse urea within 24hr and starch within 1 week (Gordon, 1976). The proteolytic properties of *D.congolensis* varied among the strain examined by Gordon (1976).

The exhibition of minor biochemical and colonial variation observed by various workers could not be correlated or ascribed to be associated with host species origin or permit the separation of the species, but were described to be due to variation in media; techniques and methods of reaching results in respective laboratories. For example, Memery (1966) added 10% serum to his carbohydrate media and observed acid formation in some of the sugar substrates reported elsewhere as non-reactive. These differences among strains of all origins did not permit separation into distinct biochemical types and their microbiological characteristics remain unchanged.

## 2.3 Epizootiology

Generally it is believed that the interaction of multiplepredisposing and some precipitating factors under widely differing environmental conditions play an important role in the epizootiology of the disease (Zaria, 1993). Predisposing factors such as rainfall, humidity (moisture), high ambient temperature, ectoparasites, vegetation, skin pigmentation, methods of husbandary, intercurrent diseases, malnutrition, stress and heredity have been reported to

play an important role in the course, spread and resolution of the disease in susceptible animals (Zaria, 1993). The effect of climate is in fact one of the most prominent epizootiological features of bovine dermatophilosis, and animals in which the disease regresses are usually re-infected repeatedly in successive wet seasons(Lloyd, 1971).

### 2.3.1 Geographical distribution

Dermatophilosis occurs in all areas of the world and can be epizootic in tropical and subtropical areas of the world (Radostits *et al*., 2007; Dalis *et al*., 2010) which are known to be areas of relatively low altitude; subject to high ambient temperatures and high torrential rain patterns.

The disease was first described by Van Saceghem (1915) in the Belgian Congo, Central Africa and has since been considered endemic, often associated with frequent outbreaks in other parts of Africa and the world as a whole. The disease has been reported in cattle, sheep, goats and horses in West Africa such as Senegal (Memery, 1960); Nigeria (Lloyd, 1971); Ghana (Oppong, 1976); and Gambia(Macadam, 1977). In Central Africa, the disease was described mainly as a disease in cattle and other animals (Chamoiseau, 1973).In East Africa, Dermatophilosis has been reported in the Uganda (Bwangamoi, 1968);Kenyan region (Bwangamoi, 1969); Sudan (Obeid, 1976) affecting cattle, sheep, goat and camels.

In southern Africa the disease has been reported in cattle in countries such as Angola, Botswana, Malawi, Mozambique, Namibia, Zambia, Zimbabwe and South Africa (Samui, 1991) and in Madasgascar (Buck, 1948).Native sheep in tropical Africa appears to possess some degree of innate resistance to the disease, which appears to be responsible for fewer cases of the disease reported in sheep and goats. (Hyslop, 1980).

In Europe, dermatophilosis has been reported in the UK and British Isle (Ainsworth and Austwick, 1975) in cattle, sheep and horses; in cattle and man in Isreal (Nobel *et al*., 1975); and in Holland, it's been reported in horses (Sloet, 1989); in pigs in Bulgaria (Verdes *et al.*,

1990). It has also been reported in France (Bussieras *et al.,* 1978) and Norway (Ulvund, 1975).Dermatophilosis infection has also been reported in the USA in cattle, wildlife animals and man (Dean *et al.,* 1961; Kaplan and Johnston, 1966; Erickson, 1975) and in Canada mostly in cattle and horses (Searcy, 1968).

In the carribbean, the disease has been described in Martinique, Guadaloupe St Martins Island, Antigua and St Lucia. (Morrow and Compton, 1989)

The disease was also reported in cattle in Puerto Rico and Costa Rica in Central America. InSouth America it has been reported in Brazil, Argentina, Chile, Uruguay andColombia (Moreira and Barbosa, 1976). Dermatophilosis has been reported in Australia, Papua New Guinea and New Zealand in various animals including horses, man, crocodiles and turtles (Masters *et al.,*1995; Kaminski, 1971).

In Asia, the disease has been recorded in countries including in Hong Kong (Yip *et al.,* 1973) and India (in cattle and camels)(Kharole *et al.,* 1975) and Banglandesh (Noorudin and Khaleque, 1986). It has also been reported in polar bear in South Korea (Eo and Kwon, 2014).

Because dermatophilosis is not a globally reportable disease, its prevalence may be much greater in Asia and indeed other countries than often assumed (Zaria, 1993).

### 2.3.2 Animal host affected

Dermatophilosis infection is known to affect a wide variety of domestic and wild animals, including terrestrial, aquatic mammals like seals (Frese and Weber, 1971) and reptiles. (Theonly non-mammalian host so far encountered are the Australianbearded lizards *(Amphibolurus barbatus)* (Montali *et al.,* 1975) and marble lizard*(Calotes mystaceus)* (Anver *et al,* 1976). Masters *et al*. (1995) also described the infection in turtles and tortoise in Australia. Among domestic animals, the infection occurs most commonly in ruminants especially cattle, sheep, goats and horses (Ford *et al.,* 1974). It has also been reported in mules, donkeys and pigs (Momotani *et al.,* 1983). However certain breeds of cattle such as N'dama and Muturu in Africa(Bida and Dennis, 1967) and the Creole cattle breeds in Guadeloupe (Martinez, 1991) are resistant to the infection, on the other hand European breeds with long hair coat showed increased susceptibility (Hyslop, 1980).

The disease has been described in camels (Gitao *et al.,* 1990). Camel dermatophilosis was found to be one of the most serious skin problems faced by camel herders and in several camel rearing areas (Yared *et al.*, 2015).The infection however is rarely seen in dogs and cats (Jones,1976; Chastin *et al.*, 1976).

The disease has been known to affect a wide range of wild animals and the bacterium has been recovered from lesions in these animals including; deer, wood chuck, stripe skunk, raccoon, giraffe and antelope (Salkin *et al.,* 1981). It has also been described in buffaloes, chamois, zebras, small rodents and monkeys (Hyslop, 1980).Also in captive polar bears (Newman *et al.*, 1975;Eo and Kwon, 2014).

Birds are generally resistant to dermatophilosis, but experimental infection was demonstrated in the white leghorn breed (Sansi, 1972).

Mice are also thought to be relatively resistant but infection has been reported following disruption of the skin barrier (Chastin *et al.,* 1976).

In man, the disease has been recognized in several forms; such as pitted keratolysis (Woodgyer, 1985). Burd *et al.* (2007) also described pustular dermatitis associated with *D. congolensis* in a 15 year old girl.Ramanathan *et al.*(2010) also isolated the bacteria from the oesophagus of a woman in the United States of America.

### 2.3.3 Transmission

The natural habitat of *D. congolensis* is unknown. It is probably thought of a saprophyte in the soil, however attempts to isolate the organism from the soil has been unsuccessful (Kahn, 2010). There is no satisfactory evidence yet that *D. congolensis* multiplies in the soil or is able to survive for long periods of time in the soil (Macadam, 1976; Atia, 1980).

The principal source of infection for Dermatophilosis is the infected animals, including the healthy carrier and the apparently recovered animals(Jubb *et al*., 1992). In endemic areas, up to 50% of apparently healthy cattle may be carrier of the bacterium, while persist in the ostie of hair follicles (Jubb *et al*., 1992). Generally it is believed that close contact between animals undoubtedly favours mechanical transmission for example, direct sheep to sheep and sheep to lamb transmission has been reported by Thomas (1957) and LeRiche (1968). Also transmission from cows to bulls and also calves as a result of rubbing has been recorded (Oppong, 1976).

*Dermatophilus congolensis* is not highly invasive and does not normally breach the barriers of healthy skin (Gebreyohannes and Gebresselassie, 2013). These barriers include the sebaceous gland on the body of sheep and the physical barrier of the wool. On the feet and face these barriers are easily and commonly broken by abrasive terrain or thorny and spiny forage and food stuffs,*Dermatophilus congolensis* may infect these lesions i.e the broken skin and the organism may be transmitted onto these wounds mechanically by feeding flies to result in minor infection on the face and feet. This mode of transmission of the disease is common in most herds and minor lesions are evident at the junction of the haired and non-

haired areas of the nares and of the claws and dewclaws(Kahn, 2010). These minor lesions are ofno clinical significance to the animal except that they provide a source of more serious infection when other areas of the skin surface are predisposed to infection (Kahn, 2010).

Transmission occurs from the carriage lesions by contact from the face of one animal to the fleece or skin of another, and from the feet to the skin during mounting. Dermatophilosis is transmitted by the coccoid forms, which results from the multidimensional division of the hyphae known as a zoospore. The zoospore is motile and released when the scabs are exposed to moisture. Transmission can be direct or indirectly through contaminated water or grass. Insect transmission which has been demonstrated with flies and ticks is believed to be a principal means of spreading zoospores (Quinn *et al*., 2002).

Dipping and spraying against ectoparasite has been reported also as a means of indirect transmission of the infection as scabs deposited in the dips can serve as source of infection to susceptible animals as the organism can survive in certain acaricides contaminated by infected scabs (Zaria, 1993).Transmission has been attributed frequently to the activities of some birds that alight on the back of animals especially the ox-pecker bird (Bida and Dennis, 1967).

Many workers have reported apparent congenital infection in calves and lambs from different areas of the world, which may suggest a placental transmission of the organism (Roberts and Valley, 1962; Egerton, 1964; Dennis, 1966; Dillman, 1967).

### 2.3.5 Predisposing factors to dermatophilosis

*2.3.5.1 Environmental and management factors*

*Cattle*: In temperate zones, outbreak in herds and severe disease in individuals are uncommon but can occur associated with high rainfall with attack rate of 50%(Gebreyohannes and Gebresselassie, 2013). The use of periodic showers or continual misting to cool cattle during hot periods is a risk factor for infection in dairy herds (Radostits *et al*., 2007). In tropical

zone, climate is the most important risk factor in tropical and subtropical regions. For example, rain fall can act indirectly to increase the range and activity of potential arthropod vectors. These arthropod vectors are important in the endemic tropical and subtropical areas than in temperate zones (Jubb *et al*., 1992; Radostits *et al*., 2007). The disease has highest incidence and severity during the humid and high rainfall season. The seasonal occurrence is associated with concomitant increase in tick and insect infestation (Radostits *et al*., 2007).

Tick infestation, particularly with *Amblyomma variegatum*, *Hyalomma asticum*and *Boophilus microplus* is strongly associated with the occurance of extensive lesions of Dermatophilosis, which can be minimized by the use of acaricides (Oppong, 1976). The lesions of Dermatophilosis on the body does not occur at the predilection sites for ticks and it is thought that the importance of tick infestation relates to a tick produced immune suppression in the host rather than mechanical or biological transmission (Kahn, 2010).

There is a particular tendency for lesions to occur on the rump and back in female and males probably due to the introduction of infection through minor skin abrasions caused by mounting, other penetrating lesions caused by ear tags or biting flies can also result in minor slesions. Intercurrent disease, stress and trauma to the skin produced by thorny bushes can act as risk factors (Radostits *et al*., 2007).

*Sheep:* Prolonged wetting of the fleece is the major risk factor and lead to emulsification of the wax barrier and maceration of skin surface with disruption of the stratum corneum. A prolonged and heavy rain is sufficient to do this especially if followed by warm and humid weather that retards drying of the fleece(Aitken, 2007). Increased environmental humidity and temperature, as distinct from wetting of the skin, does not appear to promote the development of lesions. Moisture releases infective zoospores from carriage lesions and these may be carried mechanically by flies which are attracted to the wet wool. The motile

zoospores are aided in their movement to the skin surface by the moisture of the fleece and their positive chemotactic response to carbon dioxide at the skin surface (Aitken, 2007).

A protracted wetting period of the fleece can also occur following dipping, jetting, or spraying of sheep for external parasites when these procedures are conducted at periods greater than 1-2 months after shearing; the incidence of mycotic dermatitis in sheep has been shown to increase with the time period between shearing and dipping. Shearing also destroy the barriers of the skin and cuts may become infective mechanically by flies, physically by tight yarding after shearing, and mediate infection in dips when sheep are dipped immediately following shearing. The resultant lesion does not spread over the body but provide a significant source of infection for other sheep in the flock when management or climatic circumstances lead to a high degree of flock skin susceptibly (Aitken, 2007).

*Horse*: Biting flies (*Stomxys calcitrans*) are thought to act as mechanical vectors of the infection and House fly *(Musca domestica*) can carry infection. Skin damage from trauma or from ectoparasites can predispose disease as does wetting from rainfall or from frequent washing (Reed *et al*., 2004).

*2.3.5.2Host factor*

There is breed differences in susceptibility to Dermatophilosis. In Africa, the N'dama and Muturu cattle breeds and native sheep are resistant, while Zebu, White Fulani, and European breeds are susceptible (Chodnik, 1956). Within breed differences in susceptibility are also apparent and genetic markers have been identified in Zebu (Maillard *et al.*, 2003). Susceptibility in cattle can be influenced by genetic selection. Sheep that have strong or medium wool strains are most susceptible. Open fleeced sheep and sheep with a low wax and high suint content in their fleece are more prone to infection (Radostits *et al*., 2007).

*2.3.5.3Pathogen factor*

*Dermatophilus congolensis* does not live well off the body and in the normal environment, and is susceptible to the external influences of pH and moisture fluctuations. In the laboratory it can survive for four years in sterile broth culture and for at least 13 years in dry scab material (Radostits *et al.*, 2007).

## 2. 4 Pathogenesis

The natural skin serves as effective barrier to infection. It has been suggested though that ostia of hair follicles may provide portal of entry (Hyslop, 1980). However Minor trauma, or maceration by prolonged wetting, allows establishment of infection and multiplication of the organism in the epidermis.*D.congolensis* infection occurs when the organism overcomes the three skin barriers protecting the uncornified epidermis hair or fleece, sebous wax and stratum corneum (Roberts, 1963). The skin basement membrane is another important barrier against dermal invasion by *D. congolensis*. (Amakiri, 1973). Garcia *et al*, (2004) identified and characterized an alkaline ceramidase gene from strains of*Dermatophilus congolensis* which is thought to play in the cleavage of ceramide; a major end product of the differentiation and keratinization process of the mammalian epidermis and a major component of the permeability barrier and water reservoir of the skin having a direct and indirect effects to assist *Dermatophilus* penetration of the epidermis.

When the organism overcomes the barriers, it multiplies deep in the epidermis which becomes separated from the dermis as a result of infiltration with exudates. The organism is not often found in the dermis except following rupture of hair follicles. The hyphae of *D.congolensis* are reported to be invasive and exert mechanical force to enable them penetrate the epidermal cells (Roberts, 1965).

The formation of typical pyramidal shaped crust is caused by repeated cycles of invasion in to the epidermis by hyphae, bacterial multiplication in the epidermis, rapid infiltration of neutrophils, and regeneration of epidermis. The organism in the scab is the source for repeated and expanding invasions which occurs until immunity develops and the lesion heals. The scab then separates from the healed lesion but is still held loosely in place by hair fibers. In sheep, the extensive maceration of the skin that can occur with prolonged fleece wetting can result in extensive skin lesions under the fleece. In cattle, tick infestation suppresses immunity function and promotes the spread of the lesion. Secondary bacterial infection may occur and give rise to extensive suppuration and severe toxemia (Jubb *et al*., 1992).

## 2.5 Clinical Signs

Dermatophilosis is seen in animals at all ages and both sexes are also susceptible to infection (Haward, 1996) generally in all affected animals species, the disease is characterized by the appearance of a proliferative and exudative dermatitis with subsequent formation of scab under which the hair or fleece tends to break or be matted together (Zaria, 1993). In a typical dermatophilosis lesion, the local lesion appears as an area of matted hair or fleece which may sometimes be detached together with a moist crust and leave a raw red exudative area (Hyslop, 1980).

In cattle, the lesion commences as a circumscribed moist patch, often with raised or matted hairs, giving a characteristic ‘’paint brush‘’ appearance. Discrete lesions occur in the initial stages which coalesce to form large areas of hyperkeratotic scab and crust. Distribution of the gross lesion usually correlates with the predisposing factors that reduce or permeate the natural barrier of the integument. Typical lesions consists of circular, dome shaped scab 2-9cm in diameter. Scab may be of variable thickness and on removal show a concave underside coated in thick, yellowish exudates, leaving a row, bleeding epidermis (Andrew *et al*., 2003). Death usually occurs particularly in calves because of generalized disease with or

without secondary bacterial infection and secondary fly or screw worm infestation (Kahn, 2005).

In sheep, the majority if not all, present a normal appearance and the most notable features are scabby lesions on the ears and muzzle area particularly in lambs (Austwick and Davis, 1958). Sometimes intense pruritus may be observed but it was only a transient phase lesion which also appeared on the lip and on the leg from the coronets readily seen in hairless areas. But in the fleece of woollen sheep, lesions are not commonly visible because they are obscured by the fleece but crusts can be palpated as hard mass at the surface of the skin. Heavy mortalities can occur in very young lambs where there can be extensive lesions over the body (Aitken, 2007).

Lesions in horses are similar to those in cattle, the hairs are matted together over the lesion and an exudative dermatitis produces a firm mat of hairs and debris just above the skin surface. If this hair is plucked the entire structure may lift off, leaving a characteristic ovoid, slightly bleeding skin area (Reed *et al*., 2004).

In wild animals, extensive encrusted lesions were evident on the snout, around the eyes, under the chin, on the axillae, wrist, ankles and digits of the front and hind legs. Alopecia is also common. (Zaria, 1993).

In man, the lesions are usually located and appear as pustules on the hand and occasionally, the feet and scab-like lesion developed at the site of inoculation which itched intensely for days (Zaria, 1993). It has been reported to cause pitted keratolysis called “keratoma plantare sulcatum”. (Zaias and Rebell, 1965; Woodgyer, 1985). The organism has also been reported to cause leukoplakia in man(Bunker *et al*., 1988) and has been isolated from the oesophagus in a 53 year old woman (Ramanathan *et al.,* 2010).The disease in man usually heal spontaneously without any treatment or with minimal treatment but a case of chronic nodular

disease has been reported in an 8 year old boy who suffered from immune deficiency (Hyslop, 1980).

## 2.6 Pathology

### 2.6.1 Clinical pathology

The causative organism may be isolated from scraping or a biopsy section and is much easier to isolate from acute case than chronic ones (Jubb *et al*., 1992). Typical branching organisms with double row of zoospores can be seen in a stained impression smear made directly from the ventral surface of a thick scab preserved firmly on to a slide(Jones *et al*., 1996). The organism can be also demonstrated by fluorescent antibody test (FAT), enzyme linked immuno sorbent assay (ELISA) and counter immuno electrophoresis (CIE) also been used to detect serological evidence of infection with *Dermatophilus congolensis* (Jones *et al*., 1996).

Grossly, the disease in cattle may be quite severe and is characterized by formation of raised papular and vesicular lesions causing the hair above them to be raised in a characteristics paint brush like manner (Bida and Dennis, 1976; Oduye 1976; Njoku and Alafiatayo, 1984). The disease may progress to the exudative phase producing crust coupled with the formation of thick multi strata scabs (Abu-Samra, 1980).

Histopathological changes in the early stage of infection in animals include the appearance of exudate with a moderateinflux of neutrophils and some lymphocytes at the base of the stratum corneum. Small vesicles form that cause separation ofthe cornified layers. Other histopathological changes includecongestion, edema, and thickening of the stratum spinosum.Langerhans cells may be present and possibly play a role in bacterial antigen presentation and initiation of the host immuneresponse in animals and humans (Ellis *et al*., 1989; Towersey *et al*., 1993). The formation of thick scabs in animal infections is caused by repeated cyclesof invasion and multiplication of the organism in the epidermis, rapid

infiltration by neutrophils, regeneration of the epidermis and invasion of the regenerated epidermis (Zaria, 1993; Ambrose *et al.*, 1999).

Meanwhile microscopic study showed that the infected epidermis becomes acanthotic, which is attributed to accumulation of the malpighian cells of the epidermis (Zlotnik, 1955) and is in turn covered superficially by several layers of laminated leucocytes and debris. There is parakeratosis and micro-abscessation (Oduye, 1976) and sometimes with marked dermal reaction consisting of a few plasma cells, and Lymphocytes with pockets of neutrophillic cellular infiltration (Schulz, 1955; Chodnik, 1956; Tuker, 1966; Oduye 1976).

In animals that die, there is extensive dermatitis, sometimes a secondary pneumonia, and often evidence of intercurrent disease (Radostits *et al*., 2007).

Clinical biochemistry was studied in cattle to define the systematic effect of infection where serum magnesium, copper, calcium, zinc, sodium, and potassium levels were not affected (Gbodi, 1980). There is an increase in fibrinogen, a fall in cholesterol and in calcium potassium ratio and changes in the protein component reported by Gaulier *et al*. (1972).

## 2.7 Diagnosis

The diagnosis of dermatophilosisis is primarily based on the clinical signs, that is, appearance of lesions on the body of infected animals and the demonstration of the organism in a direct microscopic examination of Geimsa stained smears prepared from the scab lesions. But, confirmatory diagnosis of the disease may be achieved by culturing the organism on bacteriological media and identification of the organism (Zaria, 1993).

The characteristic segmenting appearance seen in tissue is often not seen in gram stained smears; these may at times show cocci only, but they usually show gram positive branched filamentous organisms. Motile zoospores can be shown after growth in tryptose broth. Definitive identification is made on the unique appearance in tissues, or the following tests:

catalase positive; urase positive; glucose, fructose and maltose positive; indole negative; gelatin positive; sucrose, salicin, and xylose positive (Cullimore, 2000).

Pin point colonies surrounded by small zones of beta hemolysis are evident after twenty four hour incubation at $37^0$C. After incubation for three to four days, colonies are considerably larger. They may be wrinkled or smooth, convex, and varying in color from grayish white to bright orange (Dalis *et al*., 2010).

Polymerase chain reaction (PCR) can be carried out for the detection of the *Dermatophilus congolensis* genome isolated from the suspected samples (Shaibu *et al*., 2010).

### 2.7.1 Differential diagnosis

Differential diagnosishas been stressed to exclude fungal infection caused by dermatophytes especially *Trichophyton* species and infestation with skin parasites such as *Demodex* species and *Psoroptes* species (Laidet, 1977; Weber, 1977).

### 2.7.2 Microbiological methods

*2.7.2.1 Isolation and Identification of D. congolensis*

The organism has a characteristic microscopic appearance, it is septate, and its branching filament become longitudinally as well as transversely divided to form ribbons of spherical or ovoid cocci, each about 0.5μm in diameter, in multiple rows. (Gebreyohannes and Gebresselassie, 2013).

The isolation of the organism by culture is usually facilitated by moisturizing scab or crust lesions with distilled water, the material is soaked for 3hrs, the suspension is transferred into $CO_2$ jar for 20min. and the fluid incubated under 20% $CO_2$ for 48 h (Haalstra, 1965; Searcy, 1968).

*2.7.2.2 Cultural characteristics*

Pin point colonies surrounded by small zones beta hemolysis are evident after twenty four hour incubation at $37^0$C. After incubation for three to four days, colonies are considerably

larger. There may be wrinkled or smooth, convex, and varying in colour from greyish white to bright orange (Dalis *et al*., 2010).

### 2.7.3 Molecular diagnostic method

The difficulty in the isolation and conventional biochemical characterization of the organism from skin scabs of infected animals has led to the search for a simpler, rapid and specific method for the diagnosis and identification of the disease and its agent (Shaibu *et al.,* 2010). WengXing *et al.* (2009) established the use of Polymerase Chain Reaction(PCR) for the detection of dermatophilosis in sheep in China. Shaibu *et al.*(2010) and Oladunni *et al.* (2015) have also used PCR to detect the organism from ruminants in Nigeria. Recently Garcia *et al.* (2013) have also used a real-time PCR to detect *D. congolensis* DNA in both cultures and clinical animal samples which have the ability to quantify and detect the pathogen more rapidly than conventional PCR.

## 2.8 Treatment

The treatment of dermatophilosis has been very difficult and is still a challenge to clinicians and researchers. Part of the difficulty has been attributed to some factors such as the predilection site for the organism i.e. the upper epidermis which is relatively avascular thus not readily reached by medication, topically or via parenteral means (Zaria, 1993). Also the large number of strains of *D. congolensis* have contributed to the difficulty of treatment of the dermatophilosis infection (Coleman, 1967).

The most important aspect of therapy involves removal of predisposing factors to the infection. Most conditions that result in cutaneous maceration must be avoided to give the skin an opportunity to dry out (Smith, 2009). The area where the infected animals have been kept should be either disinfected or abandoned (Smith, 2002).

The organism is susceptible to many antibiotics including penicillin and chloromycetin (George, 1965). Treatment is effective with topical application of ointments, sprays, and dips

in the early stages (George, 1965). Bacteriostatic and bacteroicidal action of dipping compounds containing various antibiotics on *Dermatophilus congolensis* were tested *in vitro* (Prester, 1973). The organism was also susceptible *in vitro* to large numbers of antibiotics (Abu-Samra *et al*.,1976), but drugs active *in vitro* were not necessarily effective *in vivo*.

For cattle, with the disease that occurs in temperate areas Tetracycline (5mg/kg body weight (b.w.)) repeated weekly as required is recommended and long acting oxytetracycline (20mg/kg b.w.) in one injection is recommended (Radostits *et al*., 2007). With the disease that occurs in tropical areas and associated with tick infestation parenteral treatment with antibiotics, can be used and should be used in conjunction with Acaricides (Stewart, 1997).

In sheep, bacterial dips are used, but have limited efficacy as topical treatments do not penetrate the scab to the active lesion and are more appropriate for control. Antibiotics that are effective include procaine penicillin combined with streptomycin at doses of 70,000 units/kg and 70mg/kg respectively. Treatment appears effective in wet weather and it should be given prior to shearing (Aitken, 2007).

In case of horses topical therapy is most commonly used with removal from whatever is causing prolonged wetness of the skin. Scabs can be removed by grooming under sedation and the lesions treated topically daily with povidone-iodine or chlorhexidine until the lesion heal. Severe cases can be treated daily for three days with penicillin at 20,000 units/kg alone or in combination with streptomycin at 10mg/kg (Reed*et al*., 2004).

Antibiotic sensitivity also varies with the isolate, geographical area and species of the affected animals (Tresamol and Saseendranath, 2013). Even though dermatophilosis is reported to be sensitive to various antibiotics, several workers have reported resistance to several antibiotics. Tresamol and Saseendranath (2013) reported variable resistance of the bacterium to Penicillin, Mannan *et al.* (2009) recorded resistance to Ciprofloxacin; Kruger *et al.* (1998) to Enrofloxacin and Co-trimaxole while Amor *et al.* (2011) reported resistance to

quinolones from an isolate obtained from a human case. In Nigeria, Oladunni *et al.* (2015) reported high resistance to Streptomycin and Cetriaxone while Dikwa and Zaria (2003) recorded resistance to Sulphafurazole.

Various workers have reported varying degrees of success with the use of different so called medicinal plant extracts such as *Senna alata, Lantana camara and Mitracarpus scaber* (Ali-Emmanuel *et al.*, 2003), *Aloe vera* (Hill *et al.*, 2005) and some essential oils (Yardley, 2005). In Nigeria, a formulation, Lamstreptocide developed at the National Veterinary Research Institute, Vom containing fatty acids (palmitic, stearic and oleic acids) and some transitional metals have been reported to be successful in treatment of mild and acute cases of the disease (Zaria, 1993).

Roberts and Graham (1966) found that local treatment of lumpy wool disease in sheep in Australia was of no value because the scab material could not be removed without severe injury to the animals. They also pointed out that in chronic infection there is continual invasion of the follicle sheath, so that even when the scab material is removed, the invading hyphae are at a depth beyond the reach of the topical agents. Topical treatment is laborious and small lesions are so easily missed. Moreover, under range conditions of which many cattle are kept, facilities for handling individual animals may not be available especially in Africa so that treatment is only restricted to very severe cases leaving the others as reservoir for the disease (Lloyd, 1971).

## 2.9 Control and Prophylaxis

The control of dermatophilosis in developing countries especially those in the tropical and subtropical regions remain a great problem. Although it has been proved that protection of livestock against excessive wetting was highly successful when the number of animals is small, but it is completely impractable under the extensive system of husbandry (Hyslop, 1980).The most effective method to control dermatophilosis is by tick control (Plowright,

1956; Martinez, 1991) and the reduction of other ectoparasites especially fly population. (Zaria, 1993).Other methods include good husbandry and good hygiene such as clearing, draining of contaminated parasites dip, destruction of contaminated implements. (Hyslop, 1980).

Chronically infected animals should be culled to prevent them from acting as reservoirs and establishment of resistant breeds.

Vaccinations have been largely unsuccessful in large scale field trials. In experimental vaccine trials, vaccinated animals showed more rapidly healing following challenges with *D.congolensis* than did the control group. It was concluded that vaccination would both be uneconomical and of dubious efficacy (Coleman, 1967). Several workers such as: Jubb *et al*., (1992); Krauss *et al*., (2003); Kahn, (2010);Radostits *et al*., (2007); Awad *et al*., (2008) and Smith, (2009)concluded that the following methods are important in the prevention and control of dermatophilosis;

- Avoidance of skin trauma and management practices that promote transmission.
- Treatment with antibiotics.
- Infected animals must be carefully groomed to remove crusts that contain the organism.
- The crust must be appropriately disposed to prevent further contamination of the environment.
- Establishment of animal breeds resistant to *Dermatophilus congolensis*.
- For humans, use of protective clothing, gloves and personal hygiene particular those working or handling animals.

### 2.10 Economic and Zoonotic Importance of Dermatophilosis

In Africa the disease in cattle causes great losses and many deaths, and the disease ranks as one of the four major bacteriological diseases with equivalent importance to Contagious

Pleuro Pneumonia and Brucellosis. Goats in the same area also suffer a high incidence. Losses are from direct animal loss, decreased work ability of affected oxen, reproductive failure from vulval infection or infection on the limbs of males preventing mounting, death from starvation of calves of dams with udder infection, loss of animal meat and milk production, and down grading of the hides (Radostits *et al.,* 2007).

Dermatophilosis in humans is rare and there have been few cases reported, most of them concerning people with activities related to the handling of cattle in which it is consider as an occupational disease. However, it has been suggested that there must be both environmental and intrinsic factors of the host that would trigger a predisposition of the host to develop an infection as many individuals handle sick animals every day without becoming infected (Zaria, 1993; Harman *et al.,* 2001) The clinical appearance is protean and includes cases of abscesses, furunculosis, eczematoid exudative lesions, cracked intertrigo and folliculitis (Erickson, 1975; Burd, 2007). Humans who are exposed to infected animals or contaminated animal products (example, slaughter house workers, butchers, hunters, dairy farmers, veterinarians) may acquire the infection (Kamininski and Suter, 1995). In humans, Dermatophilosis appear as eczemoid cells, multiple pustules, or even furuncles, localized predominantly on the hands and forearms. In most cases, the lesions heal spontaneously within two to three weeks (Krauss *et al*., 2003).Although it's been suggested that cases of human infection may be underestimated and possibly underdiagnosed (Amor *et al.,* 2011).

## 2.11 Current State of Research

Shaibu and Ayinzat (2015) reported that different researchers have tried to establish the protein profiles of isolates from different animal species with the aim of differentiating the isolates by the use of polyacrylamide gelelectrophoreses (PAGE) and sodium dodecyl sulphate polyacrylamide gel electrophoreses (SDS-PAGE) (Gogolewski*et al.,* 1992; Masters *et al.,* 1995; Kruger *et al.,*1998;Makinde and Gyles, 1999;Shaibu, *et al*., 2011).

Similarlyresearchers continue to use other methods such as polymerase chain reaction (PCR), (Buenviaje*et al.,* 2000; WengXing*et al.*,2009;Shaibu, 2010) and cloning of serine protease gene, (Mine and Canegie, 1997). Larasa*et al.* (2002) reported the use of a simple Random Amplified polymorphic DNA (RAPD) genotyping method for field isolates of *Dermatophilus congolensis* and suggested that using this technique; they found genotypic variation between isolates, which correlated with host species. Larasa*et al.* (2002) also used other methods in their attempt to type isolates of *Dermatophilus congolensis* by evaluation of RAPD and Pulse Field Gel Electrophoreses (PFGE) techniques for molecular typing of *Dermatophilus congolensis* and concluded that both methods were good for molecular typing. Researchers have also used restriction enzymes to restrict the DNA of the organisms for genotyping of isolates (Faibra, 1993; Master *et al.,* 1995).

Since researchers began attempt to understand the genome and sequences o*f Dermatophilus congolensis,* attempt to sequence the entire genome has not been achieved until recently when a series of shotgun sequences were made culminating in a master record of a whole genome sequence of the organism. Scientists started by sequencing the organism in bits and genes and partial sequences of the genes they could identify (Shaibu and Ayinzat, 2015). Yang and Woese, (1993) sequenced *Dermatophilus congolensis* small subunit ribosomal RNA sequence. Three years later,Normand *et al.*(1996) sequenced the *Dermatophilus congolensis* 16S ribosomal RNA (16S rRNA) gene andStackebrandt and Schumann, (2000) the partial sequence of the same gene and Shaibu*et al.* (2011) sequenced partial sequences of *D. congolensis* 16S ribosomal RNA (16S rRNA) gene from cattle sheep and goats in Nigeria. Mine and Caniege, (1997) sequenced a serine protease antigen. Garcia, *et al.*(2004) sequenced an agac alkaline ceramidase gene and also serine protease gene (nasp gene). Kyrpedis*et al.*(2013) did a *Dermatophilus congolensis* DSM 44180=NBRC 105199 strain

DSM 44180, whole genome shotgun sequencing project, which is the latest work on the sequencing genome of the organism (Shaibu and Ayinzat, 2015).

## 2.12 Dermatophilosis Status in Nigeria

The prevalence of 6.2% and 2.5% in cattle during rainy and dry season respectively had been reported by Lloyd (1971) whereas Bwangamoi (1976) found a prevalence of 11%, 5.5%, and 2.5% in cattle, sheep and goats, respectively in Nigeria during the rainy season. In another study, prevalence rate were 11.98% in herd in northern Nigeria and 1.2% in the abattoir samples in Ibadan Western Nigeria (Adetunji *et al*., 2000). Dikwa and Zaria (2003) reported 4.5% prevalence in Borno state while Oladunni *et al.* (2015) reported a prevalence of 8.4% in a study conducted in Abeokuta. Ikpeze (2007) in his study reported that 12% prevalence of infection in Nigeria's estimated 10.8 million cattle and 10% prevalence in draught cattle.

The antibiotic sensitivity profiles of isolates from Nigeria have been studied by some researchers with varying reports, Oladunni *et al.* (2015) reported high resistance to Streptomycin and Cetriaxone while Dikwa and Zaria (2003) recorded resistance to Sulphafurazole while Denthe (2013) reported high susceptibility to ciprofloxacin with isolates from sokoto state.

# CHAPTER THREE

## Materials and Methods

### 3.1 Study Area

Abuja, the Federal capital territoryis located on $9^0$4'N and $7^0$29'E. The city is bordered by Niger, Kaduna, Nassarawa and Kogi states. At the 2006 census, the city had a population of 776, 298 (FRN, 2009). According to the United Nations, Abuja grew at annual rate of 139.7% between 2000 and 2010. As at 2015, the city was still experiencing an annual growth of at least 35% (Boumphery, 2010). The unofficial metropolitan area of Abuja has a population of well over three million and comprises the fourth largest metropolitan area in Nigeria, surpassed only by Lagos, Kano and Ibadan (Boumphery, 2010). Abuja is divided into six local area councils namely; Abuja municipal area council (AMAC), Bwari, Gwagwalada, Kuje, Kwali and Abaji area councils. The FCT falls within the Guinea forest-savanna mosaic zone and features a tropical wet and dry climate. The rainy seasons begins from April and ends in October. There are four abattoirs in the city, Karu, Kubwa, Deidei and Gwagwalada abattoirs. It is estimated that about 150 cattle and 750 sheep and goats are being slaughtered at the Karu daily and ₦5 million naira generated at the karu abattoir monthly (FCDA).

### 3.2 Study Design

The target population were all cattle presented to the abattoir for slaughter and selected small holder herds in the FCT. The Karu abattoir was selected because it had the highest average slaughter figure of over 80 cattle per day in the FCT. The sedentary herds were selected based on herds already identified by the Veterinary Services Department of the Agricultural and Rural Development Secretariat of the Federal Capital Territory Authority (FCTA) for a planned vaccination campaign. A total of 25 sedentary herds were examined, 8 at the Bwari and 17 at Gwagwalada area councils.

A cross-sectional study was carried out to identify the level of occurrence of dermatophilosis among cattle brought for slaughter at the Karu abattoir and selected sedentary herds in the FCT. The period of the study covered 7 months from August, 2016 to February, 2017 to cover the wet (August to October) and dry periods (November to February) of the year.

### 3.3 Collection of Scab Samples

Scabs from animals suspected to be infected with *D.congolensis*based on clinical signs were collected at every market day from the Karu abattoir. Despite weekly observations from the selected small holder herds no scabs were obtained because none of the cattle showed clinical signs of dermatophilosis. Skin scabs were collected into sterile containers and properly labelled, with the dates of collection, cattle breed and sex, as well as the site of lesion noted. Samples were properly stored at room temperature prior to transportation to the Diagnostic Bacteriology laboratory in the Department of Veterinary Microbiology ABU Zaria and subsequently to the Dermatophilosis Research Laboratory at the National Veterinary Research Institute, Vom for culture and isolation.

### 3.4 Preparation of Media used for the Isolation.

10% sheep blood agar, Brain Heart Infusion broth and nutrient agar were used for the isolation and preservation of isolates for identification and for antimicrobial susceptibility testing of the isolates.

#### 3.4.1 Blood agar (OXOID; Basingstoke, United Kingdom).

The blood agar served as an all-purpose and is the best medium recommended for the isolation of *Dermatophilus congolensis* (Macadam and Haalstra, 1971). The medium was prepared by suspending 28g of the commercial powder in 1 liter of distilled water. The mixture were allowed to dissolve and then boiled for 15 minutes at $121^0$C by autoclaving. It was then allowed to cool to $50^0$C before the blood was added. It was then dispensed into the Petri dishes to solidify and then incubated overnight at $37^0$C for 24 hours.

### 3.4.2 Brain heart infusion broth

The brain heart infusion broth served as the medium for preserving the pure isolates and as broth for antibiotic sensitivity testing. The medium was prepared by dissolving 37g of the commercial powder in 1 litre of distilled water. This was thoroughly mixed and dispensed into bijou bottles then sterilized by autoclaving at $121^0$C for 15minutes.

### 3.4.3 Nutrient agar

Nutrient agar is an all-purpose medium that supports growth of wide range of microbes. It served as the medium for antibiotic sensitivity testing. The medium was prepared by adding 23g of the nutrient agar powder to 1 liter of distilled water, the mixture was then sterilized by autoclaving at $121^0$C for 15minutes, and then cooled to about $50^0$C, thoroughly swirled and dispensed into petri dishes.

## 3.5 Laboratory Analysis

All the scabs samples obtained were subjected to three laboratory analysis

- Direct examination
- Culture and Isolation
- Antimicrobial susceptibility testing

### 3.5.1 Direct examination of scab samples for *D.congolensis*

Direct examination of the skin scrapping was carried out using standard technique (Awad *et al.*, 2008)modified by staining with methylene blue stain. This involved placing a sterile drop of water on a clean glass slide with the suspected skin scab held with a pair of forceps and mixed with the sterile water to make a smear. The smear was then allowed to dry in air, heat fixed and stained with Methylene blue (MB) stain. The stained smear was then examined with high power (x100) objective lens of the microscope with oil immersion for the presence of filamentous organism with horizontal and vertical septation typical of *D. congolensis*.

### 3.5.2 Culture for *Dermatophilus congolensis*

Isolation of *D congolensis* was carried out using the modified Haalstra's technique as described by Awad *et al.* (2008).A small amount of scab material was grinded up, placed in a screw capped bottle,moistened with 5 ml sterilized distilled water and allowed to stand open for three and half hours on the bench. Then the bottle were opened and the opened bottle transferred to candle jar with a commercial $CO_2$ gas pack within the jar to obtain 20% $CO_2$ tension (so the motile zoospores were chemotactically attracted to the $CO_2$ enhanced atmosphere and move to the surface of distilled water). After 30 minutes, the bottle was carefully removed and a sterile bacteriological loop was used to seed the surface fluid on blood agar plates and incubated for 48-72 hours in10 % $CO_2$ chamber. The plates were then examined for small, grayish yellow, beta-haemolytic colonies that are pitted into the medium typical of *D congolensis*. These colonies were then sub-cultured on to blood agar plates and the pure isolates from the secondary culture were then inoculated into Brain heart infusion broth in duplicates and kept for further studies. Smears werethen made from suspected colonies growing on each plate then gram stained and examined with oil emersion objective of the microscope for Gram positive filamentous organism that divide vertically and horizontally consisting of three or more rows of cocci typical of *D. congolensis*.

### 3.5.3 Antimicrobial sensitivity testing of *Dermatophilus congolensis* isolates

The agar disc diffusion method was used in accordance with the Clinical and Laboratory Standards Institute (CLSI) guidelines (CLSI, 2007) as described by Oladunni *et al.* (2015). *Dermatophilus congolensis* isolates were tested for antimicrobial sensitivity to eight antimicrobial discs - Ceftazidine (30µg), Cefuroxime (30µg), Gentamicin (10µg), Ofloxacin (5µg), Ceftriaxone (30µg), Cloxacillin (5µg), Augmentin (30µg), Erythromycin (5µg) obtained from Abtek Biologicals limited (United Kingdom). These antimicrobials were chosen because of their importance in treating bacterial infections in human and animals which have not yielded to other antimicrobial agents.

*3.5.3.1 Procedure for Antimicrobial Sensitivity Testing (Disc diffusion)*

Two discrete colonies of each tested isolates were suspended in a sterile bijou bottles containing 5mls of BHI broth and incubated for 48 hours. Using micropipette, 200µl suspensions of the organism from the broth were then inoculated on to Nutrient agar plate. With a sterile spreader, the inocular suspension was spread evenly on Nutrient agar. It was then allowed to dry up before the antimicrobial-impregnated disc were applied using a sterile forcep with aseptic precautions. Within 15 minutes after the disc were applied, the plates were inverted and placed in a $CO_2$ jar and then incubated at $37^0$C for 24- 48 hours. The plates were then examined for the development of zone of inhibition.

*3.5.3.2 Broth dilution*

Following the result from the disc diffusion method, antibiotics to which the isolates were found to be resistantfrom the antibiotic concentrations of the disc were further tested using the broth dilution method and increasing concentrations of up to 250µg/ml,to determine if increased concentrations may improve their sensitivity against the isolates.

Pure cultures of *D congolensis* from the BHI broth were diluted in sterile saline and standardized using a commercial sensititre nephelometer to 0.5 McFarland standard and used as the stock inoculum amounting to$1.5 \times 10^8$ colony forming unit per ml.

Commercial antibiotics for human use were which were obtained as follows; augmentin and cefuroxime GlaxoSmithKline (UK), Erythromycin from Arbor pharmaceuticals (USA) and cloxacillin from Lifescience PVT ltd (India) were diluted to required concentrations in sterile normal saline. Each antibiotic diluted to 250µg/ml. thereafter 2-folds serial dilutions (125µg/ml, 62.5 µg/ml, 31.5 µg/ml and 15.6µg/ml) were carried to 5 dilutions to cover a therapeutically achievable range for each agent.

A volume of bacterial suspension (1ml) equal to volume of diluted antimicrobial solution was added to the broth in test tubes and incubated at 37°C for 48 hours. Positive (broth containing inoculum only) and Negative (sterile broth) controls were also incubated (Eucast, 2003).

### 3.6 Data Analysis

The rate of detection and isolation of*Dermatophilus* species as well as their antibiotic sensitivity profiles were presented in tables as percentages. Association between variables (sex and breed against the rate of detection) was tested using the Chi-square test using Stata Version 12 statistical package. $P \leq 0.05$ were assumed to be significant.

# CHAPTER FOUR

# RESULTS

## 4.1 Direct Examination and Culture of Samples

From August 2016 to February 2017, a total of six thousand, seven hundred and twenty (6,720) cattle from the Karu abattoir, three hundred and sixty seven (367) cattle from 8 sedentary herds at Bwari Area Council and 708 from 17 herds at Gwagwalada Area Councilwere examined for typical skin scab lesionssuspected to be infected with dermatophilosis. Of these, one hundred and thirty-five (135) skin scabs samples were obtained from cattle suspected to be clinically affected by dermatophilosis. All 135 samples were obtained from the abattoir as no animals from the sedentary herds presented with skin scab lesions that could be suspected to be dermatophilosis.

The 135scab samples collected were analysed for the presence of *Dermatophilus congolensis*by direct smear method and cultural isolation methods. Of the total samples, 135 samples (100%) were positive by the direct smear method (Plate I) and fifty-three (53) (39.3%) yielded positive growth on culture with typical*D. congolensis* colonies (Plate II),giving an overall occurrence rate forbovine dermatophilosis during the seven month period at the Karu abattoir as 2% and isolation rate of 0.79%.

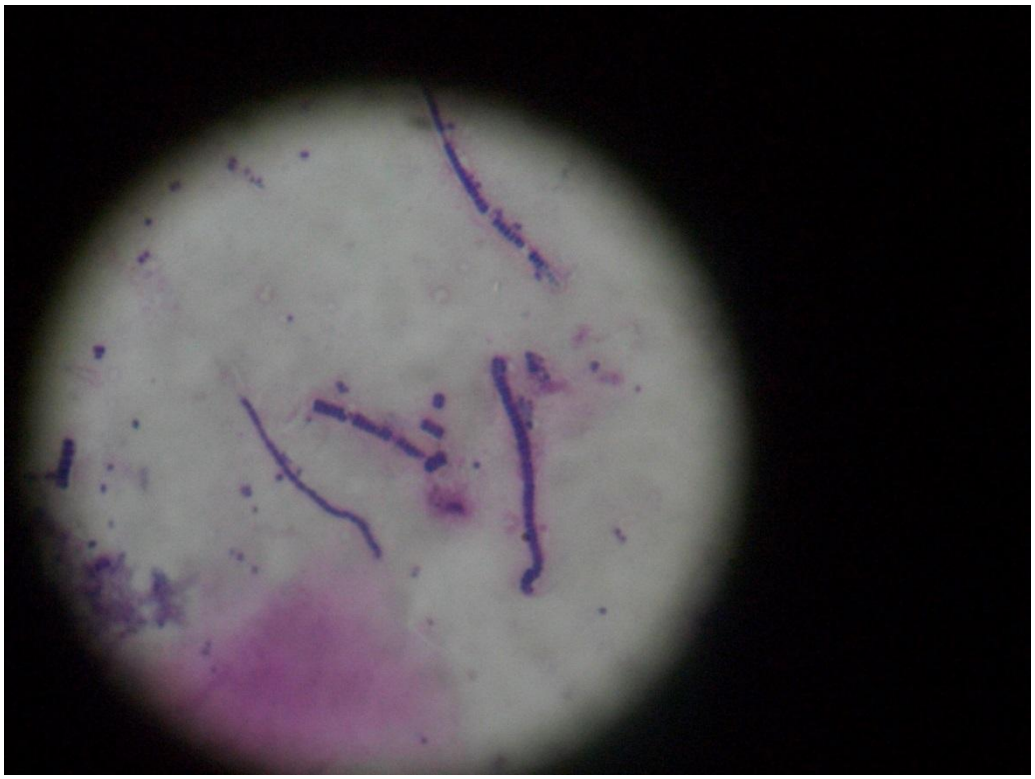


**Plate I: Direct smear of scabs stained with methylene blue showing filamentous cells characteristic of *D. congolensis***



**Plate II: Culture of *D.congolensis*on blood agar showing typical smalldry round raised β-hemolytic colonies adherent to the medium.**

### 4.1.1 Monthly cases of dermatophilosis in cattle from Karu abattoir FCT, Abuja

The monthly occurrence of dermatophilosis was higher in the months of August, September and October with 44, 26and 22 cases respectively, followed by November(16), the least number of cases were in the months of December (6) and February (9), in January, only 12 cases were observed (Tables 1 and 2).

### 4.1.2 Breed distribution of cases of dermatophilosis from slaughtered cattle from Karu abattoir, FCT, Abuja

Sixty four (47.4%) of the *D. congolensis* isolates were from the White Fulani breed, thirty eight (28.1%) from the Red Bororo and thirty three (24.4%) from the Gudali breed (Table 3).

### 4.1.3 Sex distribution of cases of dermatophilosis from slaughtered cattle from Karu abattoir, FCT, Abuja

The distribution of *Dermatophilus congolensis* by sex also revealed that 77 (57%) were from female animals, and 58 (43%) were from male animals (Table 4).

**Table 1:Monthly occurrence ofdermatophilosis among cattlepresented for slaughter at the Karu abattoir, FCT, Abuja**

| Month | Number of cattle | Number of positive cases | Occurrence rate (%) |
|---|---|---|---|
| August 2016 | 962 | 44 | 4.57 |
| September 2016 | 948 | 26 | 2.74 |
| October 2016 | 972 | 22 | 2.26 |
| November 2016 | 956 | 16 | 1.67 |
| December 2016 | 976 | 6 | 0.61 |
| January 2017 | 967 | 12 | 1.24 |
| February 2017 | 939 | 9 | 0.96 |
| Total | 6720 | 135 | 2.0 |
| $\chi^2$=0.4970 | P=0.481 | df=6 | |

**Table 2: Monthly rate of isolation of *D. congolensis* from cattle presented for slaughter at the Karu abattoir, FCT, Abuja**

| Month | Number of cattle | Number Isolates | Isolation rate (%) |
|---|---|---|---|
| August 2016 | 962 | 19 | 1.98 |
| September 2016 | 948 | 12 | 1.27 |
| October 2016 | 972 | 8 | 0.82 |
| November 2016 | 956 | 5 | 0.52 |
| December 2016 | 976 | 4 | 0.41 |
| January 2017 | 967 | 3 | 0.31 |
| February 2017 | 939 | 2 | 0.21 |
| Total | 6720 | 53 | 0.79 |

**Table 3: Breed distribution of cases of dermatophilosis and isolation rate for *D. congolensis* among cattle presented for slaughter at the Karu abattoir, FCT, Abuja**

| Breed | No of cattle examined | No of positive cases | Detection rate (%) | No of isolates | Isolation rate (%) |
|---|---|---|---|---|---|
| White Fulani | 3495 | 64 | 1.83 | 24 | 0.69 |
| Red bororo | 1814 | 38 | 2.1 | 16 | 0.88 |
| Gudali | 1411 | 33 | 2.33 | 13 | 0.92 |
| Total | 6720 | 135 | 2.00 | 53 | 0.79 |
| $\chi^2$ = 0.2124; | P= 0.899 | df= 2 | | | |

**Table 4: Sex distribution of cases of dermatophilosis and isolation rate for *D. congolensis* among cattle presented for slaughter at the Karu abattoir, FCT, Abuja**

| Sex | No of cattle examined | No of positive cases | Detection rate (%) | No of isolates | Isolation rate (%) |
|---|---|---|---|---|---|
| Male | 2928 | 58 | 1.98 | 19 | 0.65 |
| Female | 3792 | 77 | 2.00 | 34 | 0.90 |
| Total | 6720 | 135 | 2.00 | 53 | 0.79 |
| $\chi^2$ = 1.8020 | P= 0.179 | df= 1 | | | |

### 4.2 Antibiotic Sensitivity Profiles of *Dermatophilus congolensis* Isolates from the Karu Abattoir, FCT, Abuja

Out of the fifty-three (53) isolates tested to eight commonly used antibiotics, the majority of the isolates 50(94.3%) were sensitive to Ofloxacin, 42 (79.2%) of the isolates were sensitive to Ceftazidine, 26 (49.1%) and 21 (39.6%) to Gentamicin and Ceftriaxone respectively. On the other hand all, the isolateshad a hundred percent (100%) resistance to Cefuroxime, Erythromycin, Cloxacillin and Augmentin(Table 5).

**Table 5: Results of antibiotic sensitivity testingof *D. congolensis*isolates from cattle presented for slaughter at the Karu abattoir, FCT, Abuja**

| | ***D. congolensis* isolates (n=53)** | |
|---|---|---|
| **Antibiotic** | **Sensitive (%)** | **Resistant (%)** |
| Ceftazidine | 42 (79.2%) | 11 (20.8) |
| Cefuroxime | 0 (0%) | 53 (100%) |
| Gentamicin | 26(49.1%) | 27(50.9%) |
| Ceftriaxone | 21 (39.6%) | 32 (60.4%) |
| Erythromycin | 0(0%) | 53(100%) |
| Cloxacillin | 0(0%) | 53(100%) |
| Ofloxacin | 50(94.3%) | 3(5.7%) |
| Augmentin | 0(0%) | 53(100%) |

**Table 6: Resistance patterns of *D. congolensis* isolates from cattle presented for slaughter at the Karu abattoir, FCT, Abuja to various antibiotics**

| Number of antibiotics | Frequency | Percentage (%) |
|---|---|---|
| Resistance to four antibiotics | 53 | 100 |
| Resistance to five antibiotics | 32 | 64.4 |
| Resistance to six antibiotics | 27 | 50.9 |
| Resistance to more than six antibiotics | 11 | 20.8 |




**Plate III: Antibiotic sensitivity test (Disc diffusion) of isolates of *D. congolensis*on nutrient agar showing susceptibility to ofloxacin and gentamicin.**

### 4.2.1 Broth dilution

Following the result from the disc diffusion test, Cefuroxime, Erythromycin, Cloxacillin and Augmentin to which the isolates were completely resistant to, were subjected to further testing and the results read as turbidity of the broth with sputum-like growth which is the characteristic appearance of the *D congolensis* in BHI broth.

Even with the highest concentrations of the antimicrobials (250µg/ml) used, isolates were however found to be resistant to the tested antimicrobials exhibiting turbidity and growth in the broth (Plate IV).

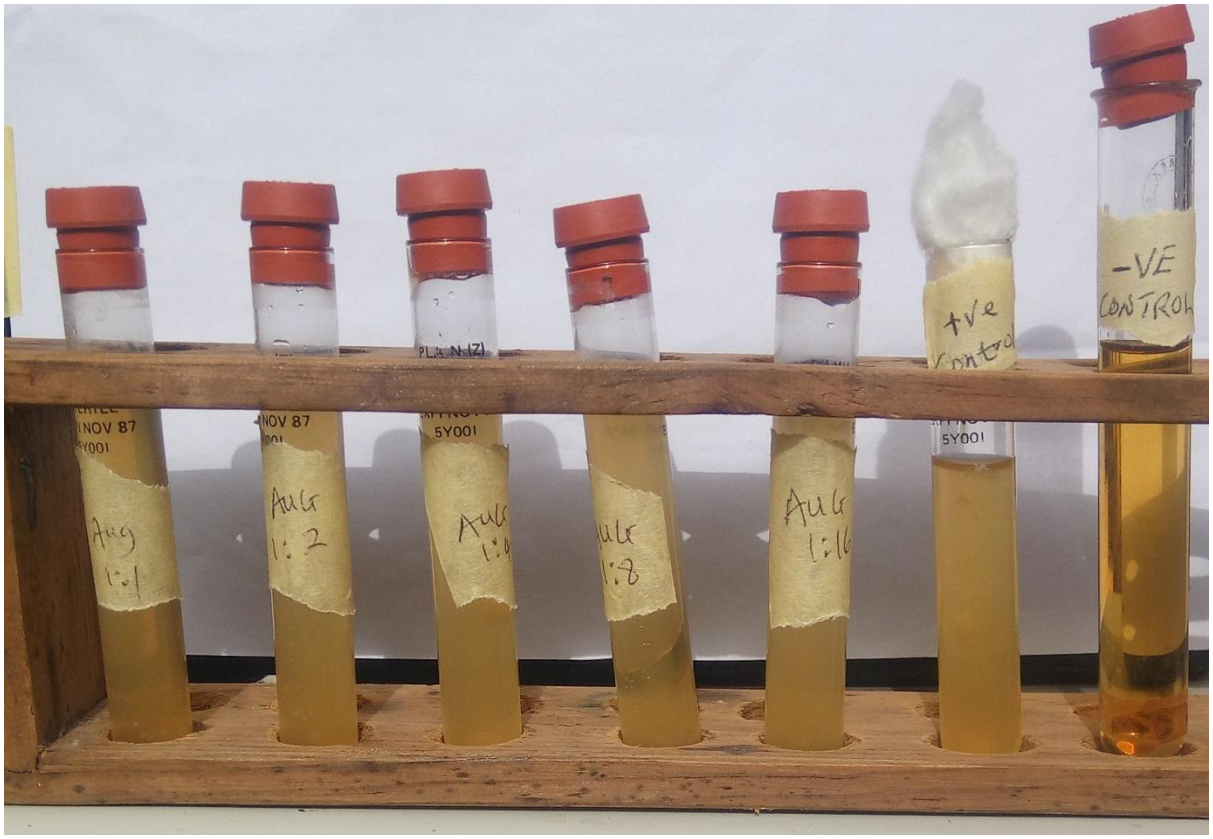



**Plate IV: Antibiotic sensitivity testing (Broth dilution) of isolates of *D. congolensis*showing resistance to Augmentin**

# CHAPTER FIVE

## DISCUSSION

The characteristic clinical manifestation of dermatophilosis coupled with identification of the organismby direct examination of stained scabs and isolation from culture established the occurrence of dermatophilosis among slaughtered cattle at the Karu abattoir FCT, Abuja.

From the study it is evident that dermatophilosis exist and is also prevalent in the study area. Skin scabs were obtained only from cattle brought for slaughter during the seven months period of the study.The apparent lack of samples from the sedentary herds was due to the fact that, the farmers culled all animals suspected to be affected by dermatophilosis so as to prevent spread of the infection to other in contact animals. This is however different from study conducted by Denthe (2013) who observed a dermatophilosis prevalence of 0.8% among sedentary herds in Sokoto state. The 39.2% isolation rate obtained in the present study is similar to results obtained by Oladunni *et al.*(2015) where they obtained 78% positive result on direct examination but only 32.9% positive by culture and isolation method in a study conducted inAbeokuta, and that of Denthe (2013) who was only able to isolate the organism from 42.4% of their samples. This relatively lower rate of isolation of the organism may not be unconnected to the fact that *D. congolensis* has been described as having generally poor competitive ability under laboratory conditions (Williams and Cross, 1971).This is probably due to the fact that Colonies of *D. congolensis* from this study took between 48 to 72 hours to grow on sheep blood agar under microaerophilic incubation at $37^0$C. Because of the delay in its growth and also because of the tiny pinpoint appearance of the colonies, the rapidly growing and less fastidious bacteria like *Staphylococcus* spp. may outgrow *D. congolensis* on blood agar.

From the 6,720 animals studied within the seven months period of the study, an overall occurrence of 2% was obtained which is at variant with various other studies in different regions of the country. Lloyd and Ojo (1976)reported 4.4% for Kano State in the north

central and 4.5% reported by Dikwa and Zaria (2003) for Borno State in the north eastern Nigeria, 0.2% reported by Denthe for Sokoto state in 2008 and 8.4% reported by Oladunni *et al.*,(2015) for Abeokuta in Ogun State, South West Nigeria while Dalis *et al.*, (2004) reported a prevalence of 5.1% in Jos and Zaria.

With the exception of Denthe with 0.6%, the prevalence from this study was significantly lower than figures obtained from other studies, this is probably due to improvement in management practices especially tick and other ectoparasites control and also due to earlier recognition of the disease thereby leading to improved successes in treatment. And in the case of Oladunni *et al.*, the higher prevalence recorded for the south maybe due to higher rainfall obtained from the region (South west).

In the present study, there was a slightly higher rate of isolation of *Dermatophilus* species from female (2.0%) cattle than their male (1.98%) counterparts. However, statistically there was no significant difference, and this is in agreement with the report of some workers (Moule and Sutherland, 1947; Abu-Samra, 1974; Macadam, 1976) that both male and female animal have been found to be equally susceptible to dermatophilosis. This was contrary to findings of Lloyd, (1976) that reported males were more susceptible than female in his study without any convincing reason.

Though all three breeds of cattle (White Fulani, Red Bororo and Gudali) encountered in this current study have been shown to be susceptible to dermatophilosisasreported by Zaria, (1993).

High rainfall and humidity are recognized and documented predisposing factors in the proliferation of dermatophilosis, as reported by several researchers (Macadam, 1964; Lloyd, 1971; Oppong, 1996; Dalis *et al*., 2004; Radostits, 2007). In the current study more cases of dermatophilosis occurred within the months of August (4.57 %) and September (2.74 %), which represent the months with the highest rainfall in the study area; which also coincides

with the period with the highest arthropod activities which has been shown to be an important factor in the proliferation of the disease (Radostits, 2007). It has also been suggested that rain may act by leaching off the inhibitory substance of the skin bacteria below their bacteriostatic level, thereby allowing *Dermatophilus congolensis* to proliferate and precipitating clinical dermatophilosis (Kingali *et al*., 1990). This might be the reason for the high rate of occurrence in the months of August and September. Also with increased rainfall, invariably there is increase in vegetation with the attendant thorns which may increase the chances of bruises and wounds which is also an important predisposing factor in the establishment of the infection among susceptible animals. Increased moisture in the rainy seasons also plays an important role in the maturation of filamentous hyphae into motile zoospores which are the infective form of the bacterium (Lloyd, 1971; Oppong, 1976; Zaria and Amin, 2004).

Sensitivity of *D. congolensis* isolates to many antibiotics has been reported by many workers (Gillum *et al.,* 1988; Towersey *et al.,* 1993). Jordon and Venning (1995) observed many antibiotics including erythromycin, spiramycin, penicillin G, ampicillin, chloramphenicol, streptomycin, Amoxicillin, tetracyclines, and novobiocin to be highly effective in the treatment of dermatophilosis. Minimum inhibitory concentrations (MIC) and minimal bactericidal concentrations (MBC) of antimicrobials on *D. congolensis* isolates determined by Hermoso de Mendoza *et al* (1995) revealed that erythromycin, spiramycin, penicillin G, ampicillin, chloramphenicol, streptomycin, Amoxicillin, tetracyclines and novobiocin had high serum concentrations and were shown to have potential use for the treatment of dermatophilosis.

The most sensitive antibiotics in the present study were the quinolone, ofloxacin and the third generation cephalosporin, ceftazidine. Out of 53 isolates, 50 isolates were sensitive to ofloxacin (94 per cent) and 42 isolates (80 per cent) to ceftazidine and only three were resistant to ofloxacin and eleven to ceftazidine.These are relatively newer antibiotics which

are not really employed in the treatment of animal bacterial infections in the study area. This finding is contradictory to the findings of most of the workers, who reported resistance of *D.congolensis* to quinolones. Mannan *et al* (2009) found that only 10 percent of the isolates from Bangladesh were sensitive to ciprofloxacin and Kruger *et al* (1998) found resistance of few isolates of *D. congolensis* to enrofloxacin. Amor *et al* (2011) found antibiotic resistance to quinolones for an isolate from a human case. Howeverthe result obtained in the present study is similar to reports by Oladunni *et al.,* 2015 and Tresamol and Saseendranath (2013); who reported a similar pattern of sensitivity (98.7%) to the quinolones, ciprofloxacin and enrofloxacin.

In the present study, 26 (50%) of the isolates were sensitive to Gentamicin, and 21 (40%) were sensitive to Ceftriaxone, this is similar to Oladunni *et al.,* 2015 who reported a 39.2% sensitivity to ceftriaxone but contradictory to Tresamol and Saseendranath (2013) who reported 97.3% and 84% sensitivity to Gentamicin and Ceftriaxone respectively.

However there was maximum resistance to the other cephalosporin (Cefuroxime), the penicillin antibiotic (Cloxacillin and Augmentin) and the macrolide,Erythromycin. This is similar to reports of Tresamol and Saseendranath (2013) who reported some resistance to the penicillins but contradictory to the results obtained by Oladunni who reported 92.2% sensitivity to Augmentin and 83.2% sensitivity to Cloxacillin. Mannan*et al.,* 2009 reported 30% sensitivity to Erythromycin which is contradictory to the complete resistance obtained in this study.

The variation of antibiotic sensitivity and resistance pattern on some group of antibiotic exhibited by the isolates of *Dermatophilus congolensis* species might be owing to genetic variation, chromosomal or plasmid alteration of the organisms and repeated and preferential use of antibiotics, which may differ from region to region and country to country (Mannan *et al.,* 2009)

# CHAPTER SIX

# CONCLUSION AND RECOMMENDATIONS

## 6.1 Conclusions

It can be concluded from the study that dermatophilosis exists and is prevalent among breeds of cattle presented for slaughter at the Karu abattoir. Out of 6,720 animals examined during the period of study, 135(2%) showed and were confirmed to be affected by dermatophilosis.

Although there were more cases of the infection observed in the female animals, this difference is not significant statistically therefore from this study dermatophilosis is seen to affect both male and female animals equally.

Majority of the cases of dermatophilosis were observed in the month of August (4.57 %) and September (2.74 %) which corresponded with the period of peak rainfall and high humidity in the study area.

This study tested antibiotics which are not routinely used for the treatment of animal infections in the study area rather used for treatment of bacterial infection especially in humans to ascertain their effect *in vitro* against *Dermatophilus congolensis.*

The study also showed that majority of *Dermatophilus congolensis* were sensitive to only a few antibiotics; Ceftazidine (79.2 %) and Ofloxacin (94.3 %) which are relatively new generation antibiotics and were very resistant to other antibiotics tested (Augmentin, Cloxacillin, Erythromycin and Cefuroxime). Variable results were however obtained with Gentamicin and Ceftriaxone.

To our knowledge, this appears to be the first study on the occurrence of dermatophilosis in cattle in the study area and is a confirmation of the presence of the disease among cattle slaughtered at the Karu abattoir with an attendant economic loss in both livestock and leather industries.

## 6.2 Recommendations

1. Identification and antimicrobial susceptibility test should be made for every *Dermatophilus congolensis*isolates for effective treatment.
2. There is need for further studies to sequence the whole genome of the organism as well as annotation of the genome to determine the number and function of the genes and also to carry out field trials of antibiotics using *in vitro* results for possible treatment of dermatophilosis in the study area.
3. There is need to improve veterinary extension service to educate farmers, butchers and public on the zoonotic importance of the disease, improve management system in terms of ticks control and reduce indiscriminate use of antibiotic which may likely lead to drug resistance.

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