PREVALENCE OF CAPRINE AND BOVINE FASCIOLIASIS IN SLAUGHTER HOUSES WITHIN KADUNA METROPOLIS

BY

OPEYEMI ADEBIMPE OKE

DEPARTMENT OF BIOLOGICAL SCIENCES,
AHMADU BELLO UNIVERSITY, ZARIA,
NIGERIA.

MAY, 2016.

PREVALENCE OF CAPRINE AND BOVINE FASCIOLIASIS IN SLAUGHTER HOUSES WITHIN KADUNA METROPOLIS

By

OpeyemiAdebimpeOKE, BSC (ED) BIOLOGY, (OAU) 2009
M.Sc/SCIE/3367/2011-2012

A DISSERTATION SUBMITTED TO THE SCHOOL OF POSTGRADUATE STUDIES, AHMADU BELLO UNIVERSITY, ZARIA IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF MASTER OF SCIENCE DEGREE IN EDUCATIONAL BIOLOGY

DEPARTMENT OF BIOLOGICAL SCIENCES,
FACULTY OF SCIENCE,
AHMADU BELLO UNIVERSITY, ZARIA,
NIGERIA.

MAY, 2016.

DECLARATION

I declare that the work in this dissertation entitled ‘Prevalence of bovine and caprine fascioliasis in slaughter houses within Kaduna metropolis’ has been carried out by me in the Department of Biological SciencesAhmadu Bello University,Zaria Nigeria under the supervisions of Dr.S.A.Luka and Mr.D.A.Adebote.The information derived from the literature has been duly acknowledged in the text and in a list of references provided. No part of this thesis was previously presented for another degree or diploma at this or any other Institution.

Opeyemi OKE ______________________ ________________________

Signature Date

CERTIFICATION

This dissertation entitled **"PREVALENCE OF CAPRINE AND BOVINE FASCIOLIASIS IN SLAUGHTER HOUSES WITHIN KADUNA METROPOLIS."** By **Opeyemi OKE** meets the regulations governing the award of the degree of Master of Science in Educational Biology of the Ahmadu Bello University, and is approved for its contribution to knowledge and literary presentation.

Dr.S.A Luka ____________________ ____________________
Chairman, Supervisory Committee Signature Date
Department of Biological Sciences,
Ahmadu Bello University, Zaria

Mr D.A Adebote ____________________ ____________________
Member, Supervisory Committee Signature Date
Department of Biological Sciences,
Ahmadu Bello University, Zaria

Prof. A. K. Adamu ____________________ ____________________
Head,Departmentof Signature Date
Biological Sciences,
Ahmadu Bello University, Zaria.

Prof. K.Bala ____________________ ____________________
Dean, School of Postgraduate Studies Signature Date
Ahmadu Bello University, Zaria.

DEDICATION

This work is dedicated to the faithful and loving God,the author of all wisdom and the reason for my existence.To my loving husband David Olajide and to my wonderful children:Isaac Oluwaseyi and Isabel Oluwaseun. I am eternally grateful to God to have you all.

ACKNOWLEDGEMENT

Unto God to whom all good and perfect gift comes from do I owe this thanksgiving, for this giftI say thank you JESUS. Believing by faith that he will grant me a glorious and expected end in my academic pursuit and journeys through life, Amen.

My profound gratitude goes to my team of supervisors, Dr.S.A.Luka and Mr D.A. Adebote for their immense tutoring and leading in the course of this research work, I thank them for their patience, motivation, enthusiasm, and immense knowledge. Their guidance and constructive criticisms helped me throughout the time of my research and writing of this thesis. I could not have imagined having better supervisors and mentors for my M.Sc study.

Besides my supervisors, I would like to thank other lecturers in the Department,Prof S.A.Abdullahi,Dr W.S Japhet,Prof S.JOniye every of my erudite scholars in the department for their support and contributions.Prof J Ajanusi and MrAmadi of the Faculty of Veterinary Medicine, Ahmadu Bello University Zaria,thanks for your contributions. I thank all my course mates, for their encouragement, insightful comment, and constructive questions and contributions.

My appreciation goes to the staff of various abattoir units and their leaders for their permission for sample collections,MrAmadi the chief Laboratory Technologist of the Parasitology and Entomology Department, Faculty of Veterinary Medicine for direction in the course of microscopic analysis.

My heartfelt appreciation goes to my husband, David Olajide, to whom this work is dedicated, You have been a constant source of love, care, support, encouragement and strength all these years. I would like to express my heart-felt gratitude to my family members Pastor and Deaconess Oke who are my parents, my siblings, GbemisolaAkintola,Dr.A.O.Oke,Itunuoluwa and to my wonderful in-laws, Elder and MrsFakorode,OluOluwakorode,Wale Fakorode for being there for me. Your spiritual, financial, moral and emotional support have helped me through. I say a big thank you.

To my collengues and friends, those mentioned and others not mentioned,Isaiah,Yemi,Bunmi,Nurat,MrsOyelere, Alex Jatau,MrsKindness,MrIbrahim,MrDavid,Mrs. Salomi Kennedy, Mr. Clement Yaro; I appreciate you all.You all will not miss your reward.Amen.

ABSTRACT

Prevalence and intensities of hepatic parasites in bovine (*Bostaurus*) and caprine (*Capra hircus)*animal hosts at slaughter were evaluated at the Sabo-Tasha and Kakuri abattoirs within Kaduna, Nigeria. Intact gall bladders of randomly selected slaughtered animals of known sexes and ages belonging to both species were obtained from butchers at the abattoirs. Bile samples from the gall bladders were subjected to the centrifugal sedimentation technique of eggs isolation. Eggs of encountered parasites were examined microscopically, counted and identified by the aid of pictorial keys. Of the total of 250 caprine host species,28(11.20%) were positive with the eggs of *Fasciola gigantica* while 127/250 (50.80%) of bovine host species had the parasites. Eggs of other parasites observed in the animals were those of *Dicrocoelium dendriticum* in 33/250(13.20%) of caprine and 80/250 (32.00%)of bovine hosts. Single parasite infections occurred in 56(22.40%) and 99(39.60%) caprine and bovine hosts respectively; while mixed parasite infections occurred in 8(3.20%) and 111(44.40%) caprine and bovine hosts respectively. Age-specific intensities of caprine *Fasciola* and *Dicrocoelium*infections were statistically significantly different (P>0.05). Age-specific intensities of bovine *Fasciola* infection were also statistically significantly different (P<0.05; 7-9years >4-6years > 1-3 years). The respective mean values for cattle aged <1-3, <4-6 and 7-9 years being ±62.26, ±296.31 and ±965.25. Intensities of bovine *Dicrocoelium* infection were statistically significantly higher (P<0.05) in animals aged 7-9 years than in the lower age groups. Intensities of *Fasciola* and *Dicrocoelium* eggs in bovine hosts were significantly higher (P<0.05) in animals slaughtered and examined at the Sabo-Tasha than Kakuri abattoir. The study concluded that caprine and bovine hepatic parasite infections were high in the study location and recommends intensification of control measures to stem the tide.

TABLE OF CONTENTS

Page

LIST OF TABLES

Pages

LIST OF PLATES

Pages

LIST OF APPENDICES

ABBREVIATIONS

ELISA	Enzyme-Linked Immunosorbent Assay
ERCP	Endoscopic Retrograde Cholangiopancreatography
FAO	Food and Agricultural Organization
MRCP	Magnetic Resonance Cholangiopancreatography
PCR	Polymerase Chain Reaction
RNA	Ribonucleic Acid
Rpm	Revolutions per Minute

CHAPTER ONE

1.0 INTRODUCTION

1.1 Introduction

Fasciolosis is an important helminth disease caused by two species of trematodes belonging to the genus *Fasciola* commonly known as liver flukes*: Fasciola hepatica* and *Fasciola gigantica*. Fascioliasis may be categorized as plant borne - trematode infection with zoonotic potential (Mas-Coma *et al.*,2005). In Europe and America, *Fasciola hepatica* is the major species of concern, but the distribution of both species overlap in many areas of Africa and Asia (Mas-Coma *et al.*, 2005).

Fascioliasis is a well known parasitic disease because of its veterinary importance and the great losses it causes in livestock production. Fascioliasis or liver rot is a parasitic disease of the liver of ruminants such as bovine, sheep, caprine, deer, horses, buffalos and other herbivores (Alonge and Fasanmi, 1979). Fascioliasis was only considered as a secondary human disease by public health officials, with only approximately 2000 human cases reported between 1970 and 1990 worldwide (Chen and Mott, 1990). The public health importance of human fascioliasis has, however increased in recent years, as more human cases were increasingly reported. The total estimated number of people infected was 2.4million in 61 countries and the number at risk were more than 180million throughout the world (FAO,2008). Surveys in several regions indicate that there are areas with true endemic human fascioliasis ranging from low to high prevalence and intensity (Mas-Coma *et al.*,1998). Recent estimates suggest that up to 2.4million or even up to 17million people are infected with *Fasciola hepatica* in the world (Hopkins, 1992). *Fasciola* *hepatica* infects humans in all continents except Antarctica, thus, having the widest longitudinal and altitudinal distribution of the food-borne trematodiases (Mas-Coma *et al*., 2005). In contrast, *Fasciola gigantica* is more geographically constricted, occurring in the tropical regions of Africa, the Middle East and Asia where infection due to either species may occur (Keiser and Utzinger, 2009). Prior to 1980, fascioliasis was thought to be a zoonoses of only mild, sporadic or local importance. In the past 20 years however, a more complete understanding of disease epidemiology has emerged (Mas-Coma *et al*., 1999). The definitive host range is very broad and includes many herbivorous mammals including humans. The life cycle includes fresh water snails as intermediate hosts of the parasite (Yilmaz and Godekmerdan, 2004). Worldwide losses in animal productivity due to fascioliasis were estimated to be over US 3.2billion dollars per annum (Spithill *et al*., 1999).

The epidemiology and transmission characteristics of fascioliasis mean that the disease has a patchy distribution, with foci being related to the local distribution of snail intermediate host populations in freshwater bodies, as well as physical and climatic conditions. It is therefore not appropriate to refer to the characteristics of fascioliasis at country level, but rather to those in a given geographically and climatically homogenous area (Mas-Coma and Bargues, 1997). Helminth infections, which are often chronic and asymptomatic, on the field lead to retarded growth, delayed and reduced productivity and increased susceptibility to secondary infections (Ndarathi *et al*.,1987). *Fasciola gigantica* is a cosmopolitan parasite and occurs mostly in Africa (Muller, 1975). Studies carried out on fascioliasis in parts of Nigeria showed that the disease is endemic in bovine slaughtered at large abattoirs (Schillhorn Van veen *et al*.,1980). Livestock are major assets for resource-poor small holder-farmers and pastoralists throughout the world and internal parasites are recognized by the pastoralists communities as having an impact on health of the livestock (John *et al*., 2005). Bovine fascioliasis is a major public health problem in many areas of the world, especially in developing countries where there is poor sanitation, poor personal hygiene, poverty and poor animal husbandry (WHO, 2012). Millions of carcasses and livers are lost due to damages caused by *Fasciola* infections in animals such as cattle, sheep and goats (Okoli *et al.*,2000). The liver is an organ of metabolism in the body of animals; damage to it means impaired energy supply to the body and subsequent health problems (Fabiyi and Adeleye, 1982).

Fasciola species can also cause severe, even fatal diseases in humans (WHO, 1999). Humans become infected by ingesting metacercariae attached to aquatic or semi aquatic plants, drinking water contaminated with floating metacercariae and or ingest metacercariae attached to the surface of food or kitchen utensils washed with water contaminated with floating metacercariae.

Prevalence of fascioliasis is highest in areas where extensive ovine and bovine rearing occurs, and where a dietary practice which includes the consumption of raw aquatic vegetables are practiced. The need for temperate, slow-moving or standing water in *Fasciola hepatica* life cycle and transmission had previously kept infection limited to populations within well defined watershed boundaries (Savioli *et al*., 1999). However, urbanization, migration and developmental practices, such as dam building and irrigation, have increased the population at risk and incidence of human infection has increased significantly till date (Savioli *et al*., 1999). Intermediate hosts of *Fasciola hepatica* are fresh water snails of the family Lymnaeidae (Graczyk and Fred, 1999). The development of infection in definitive hosts is divided into two phases; the parenchymal (migratory phase) and the biliary phase (Graczyk and Fred, 1999). Clinical signs of fascioliasis are always closely associated with the amount of metacercariae ingested (Sinclair,1962). The disease is usually characterized by a chronic, sometimes acute or sub-acute inflammation of the liver and bile ducts accompanied by submandibular oedema, anaemia and death (Ogunrinade and Ogunrinade, 1980). In bovine and caprine hosts, the clinical manifestations are similar to that of sheep. However, acquired resistance to *Fasciola hepatica* infection is well known in adult cattle (Doyle, 1973). Importance of bovine fascioliasis includes economic and production losses due to infertility and weight loss of infected animals (Phiri *et al*., 2006).

In Nigeria, meat derived from bovine, ovine and caprine species provides major animal proteins for the people and incidentally ruminants are the definitive hosts of *Fasciola* parasite (Olusegun-Joseph *et al*.,2011).

1.2 Statement of Research Problem

Parasitic diseases of ruminants which include fascioliasis, dicrocoeliasis, cysticercosis, taeniasis and hydatidosis, which could be zoonotic, constitute a major economic and public health problem by lowering the productivity of bovine, ovine, caprine and other ruminants in addition to losses from condemnation of affected organs while humans can accidentally ingest the infective stages and become infected (Cook, 1989).

Beef serve as a source of animal protein for large population of people worldwide and cattle are often infested by *Fasciola* and other helminth parasites. The consequence of this is a gross shortage of dietary animal proteins (Ndarathi *et al*., 1987). Very poor meat inspection facilities and uncooperative attitude of butchers to meat inspectors have been reported in Nigerian abattoirs, thereby leading to poor disease diagnosis (Aladi, 1999).

Most of the available records on helminth infections of ruminants are based on the examination of faecal samples. This has numerous limitations, one of which is the low sensitive of the faecal examination techniques (Mistrel *et al*., 1972). In addition, quantification of *Fasciola* eggs in faeces may not reflect the worm burden, since the adult fluke are located in the bile duct. Reliance on results from only faecal examinations may therefore lead to an underestimation of intensity of infections and to adopt appropriate control measures (Oyeduntan *et al*., 2008). Reliance on data obtained from only direct faecal examination can obviously lead to gross underestimation of prevalence rates of *Fasciola* infection in ruminants in many areas of Nigeria, including Kaduna metropolis, located in the North Western part of the country.

1.3 Justification of the Study

Generally, parasitic infections impede livestock productivity; and detailed data on the epidemiology of infection is useful in designing control programmes that will increase productivity and ultimately, the amount of animal protein available for the human population. There is, however, lack of adequate epidemiological data on the prevalence of fascioliasis in ruminants based on direct bile examinations in Nigeria.

This study was therefore embarked upon to investigate the prevalence of bovine and caprine fascioliasis from selected abattoirs in Kaduna metropolis in order to provide updated knowledge on the epidemiology of the infection. Availability of this knowledge, it is hoped, may motivate the relevant agencies to institute appropriate control measures against fascioliasis in ruminants.

1.4 Aim

To conduct a parasitological survey on fascioliasis in bovine and caprine slaughtered in Sabo-Tasha and Kakuri abattoirs within Kaduna metropolis.

1.5 Objectives

The specific objectives of the study are:

- To determine the prevalence of *Fasciola gigantica* infection in bovine and caprine slaughtered at Kakuri and Sabo-Tasha abattoirs in Kaduna metropolis.
- To determine the intensity of *Fasciola gigantica* infection in bovine and caprine slaughtered at Kakuri and Sabo-Tasha abattoirs in Kaduna metropolis.

1.6 Hypotheses

1. Bovine and caprine slaughtered in Sabo-Tasha and Kakuri abattoirs are not infected by *Fasciola gigantica.*
2. Bovine and caprine slaughtered in Sabo-Tasha and Kakuri abattoirs have nil intensity of *Fasciola gigantica.*

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Introduction

Fasciolosis, is an important helminth disease caused by two species of trematode belonging to the genus *Fasciola* commonly known as liver fluke*: Fasciola hepatica* and *Fasciola gigantica*. Fascioliasis may be categorized as plant borne trematode infection with zoonotic potential (Mas-Coma *et al*., 2005). Fascioliasis has the widest geographic spread of any emerging vector-borne zoonotic disease and affects more than 51 countries worldwide. It is most prevalent in the Andean region, especially Bolivia and Peru(Marcos *et al*., 2008). Fascioliasis is a nearly-worldwide disease that affects people in Western Europe, Southeast Asia and the Caspian Sea region, and less commonly in Africa, Oceania and Eastern Europe. An estimated 17 million people are affected by fascioliasis (WHO, 1999). Fascioliasis has only more recently been recognized as a significant human disease, studies to determine the global morbidity caused by fascioliasis are ongoing. Indeed, most analyses of the global impact of fascioliasis focus on the economic impact caused by infections in domesticated herd animals. Depending on the disease prevalence in a herd, these losses could be significant. The direct economic impact of fascioliasis is the increase in condemnation of the liver, meat, but the far more damaging effects are decreased animal productivity, lower calf birth weight and reduced growth in affected animals (Hillyer, 2005).

2.2 Historical Background

Eggs of *Fasciola hepatica* have been found in mummies, which shows that human infections were occurring at least as early as Pharaonic times (Farag). Indeed, *Fasciola hepatica* was the first fluke or trematode to be reported. It was first discovered in sheep where it causes a more obvious burden. A French man, Jehan de Brie, made the earliest references to *Fasciola hepatica* and accurately recognized the source of infection in his 1379 publication, *Le Bon Berger* (The Good Shepherd). It was from this early text that *Fasciola hepatica*acquired its common name, sheep liver fluke. References to the sheep liver fluke appeared in animal husbandry texts several centuries until it was given its Latin name by Linnaeus in 1758 and it was not until the late 19^{th} century that *Fasciola hepatica* life cycle was elucidated and its role in humans was recognized. Since then, fascioliasis has become one of the most studied helminthic infections and holds a high-raking position in the minds of public health policy makers (Savioli *et al*., 1999).

2.3 Causative Agent

Fascioliasis is caused by two species of parasitic flatworms or trematodes that mainly affect the liver. It belongs to the group of foodborne trematode infections and is a zoonosis, meaning an animal infection that may be transmitted to human. The two species of trematodes that causes fascioliasis are *Fasciola hepatica* and *Fasciola gigantica* (WHO, 2012). The prevalence of *Fasciola gigantica* often overlaps with that of *Fasciola hepatica*, and the two species are so closely related in terms of genetics, behaviour, morphological and anatomical structures that it is notoriously difficult to distinguish them (Itagaki *et al*., 2011).

2.4 Scientific Classification and Structure of *Fasciola*

Fasciola hepatica

Kingdom: Animalia

Phylum: Platyhelminthes

Class: Trematoda

Sub-class: Digenea

Order: Echinostomida

Sub-order: Echinostomata

Family: Fasciolidae

Genus: *Fasciola*

Species: *Fasciola hepatica*

Fasciola gigantica

Kingdom: Animalia

Phylum: Platyhelminthes

Class: Trematoda

Sub-class: Digenea

Order: Echinostomida

Sub-order: Echinostomata

Family: Fasciolidae

Genus: *Fasciola*

Species: *Fasciola gigantica* (en.wikipedia.org/wiki/Fascioliasis)

The two species of trematode that cause fascioliasis (*Fasciola hepatica* and *Fasciola gigantica*) are leaf-shaped worms, large enough to be visible to the naked eye as seen in plate 1. Adult *Fasciola hepatica* measures 20–30 mm x 13 mm; adult *Fasciola gigantica* measure 25–35 mm x 12 mm) (WHO, 2012). *Fasciola hepatica* is one of the largest flukes of the world, reaching a length of 30mm and a width of 13mm. It is leaf-shaped, pointed posteriorly and wide anteriorly, although the shape varies somewhat. The oral sucker is small but powerful and is located at the end of a cone-shaped projection at the anterior end. The acetabulum (ventral sucker) is larger than the oral sucker and is anterior which allow it to attach to the lining of the biliary ducts. The tegument is covered with large and scale-like spines. The intestinal caeca are highly dendritic and extend to near the posterior end of the body. The testes are large and greatly branched, arranged in tandem behind the ovary. The smaller, dendritic ovary lies on the right side, coiling between the oviduct and the preacetabular cirrus pouch. Vitelline follicles are extensive, filling most of the lateral body and becoming confluent behind the testes (Keizer *et al.*, 2009).Each worm possesses ovaries and testes which are highly branched and allow for individual flukes to produce eggs independently (Plate 1) (Tolan, 2001).

The Egg - The eggs of *Fasciola hepatica* are operculated and average 140 µm in length and 75 µm in width (plate2) (Tolan, 2001).

Plate 1: Structure of adult *Fasciola hepatica* Source: en.wikipedia.org/wiki/Fascioliasis. Retrieved on 12/4/2014

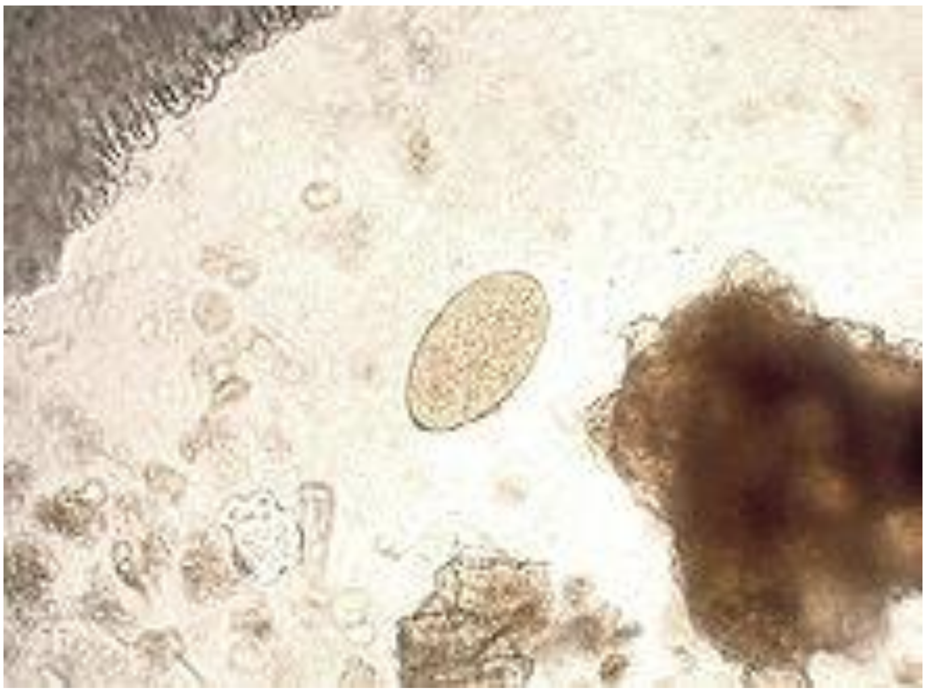


Plate 2: Egg of *Fasciola hepatica*. Source: CDC: Accessed 2014

2.5 Distribution

Fasciola gigantica causes outbreaks of fascioliasis in tropical areas of southern Asia, Southeast Asia and Africa. The geographical distribution of *Fasciola gigantica* overlaps with *Fasciola hepatica* in many African and Asian countries and sometimes in the same country (Soliman, 2008).*Fasciola hepatica* infects humans on all continents (except Antarctica), having the widest latitudinal, longitudinal and altitudinal distribution of the food-borne trematodiases (FBT) and other parasitic and vector-borne diseases (Mas-Coma, 2005). In contrast, *Fasciola gigantica* infection is more geographically constricted, occurring in the tropical regions of Africa, the Middle East and Asia, where infection due to either species may occur(Keiser and Utzinger, 2009).

2.6 Epidemiology

Fascioliasis, a food- or water-borne trematodiasis due to infection by *Fasciola hepatica* or *Fasciola gigantica*, is currently believed to affect as many as 17 million people worldwide, with 91.1 million individuals at risk of infection (Keiser and Utzinger, 2005). Contrary to early thinking, prevalence of veterinary disease is not predictive of prevalence of human disease in endemic regions (Mas-Coma, 2005). Prior to 1980, fascioliasis was thought to be a zoonosis of only mild, sporadic or local importance. In the past 20 years however, a more complete understanding of disease epidemiology has emerged(Mas-Coma *et al*., 1999). An estimated 2 million cases of fascioliasis have been reported worldwide, and incidence has apparently increased since 1980(Haseeb *etal.*,2002). *Fasciola hepatica* typically occurs worldwide in temperate regions, except Oceania. *Fasciola gigantica* causes outbreaks in tropical areas of Southern Asia, Southeast Asia, and Africa (Youn, 2009). Infection is most prevalent in regions with intensive sheep and cattle production. Miracidia require temperate water to hatch and develop. *Fasciola hepatica* occurs mainly in sheep-rearing areas of temperate climates, particularly in parts of Central and South America, Europe, China, Africa and the Middle East (Mahanty *et al.*, 2006). Sporadic cases have also been reported in the United States (Mas-Coma, 2005). Ninety one million people are at risk worldwide(Keiser and Utzinger, 2009).

Sheep and cattle are the most important definitive hosts of *Fasciola hepatica*; goats, buffalo, horses, camels, hogs, deer and rabbits can also be infected (Chan and Lam, 1987). Snails are intermediate hosts. Humans are incidental hosts and most often acquire infection by eating watercress grown in sheep-raising areas. Infection may be transmitted by other freshwater plants, including water lettuce, mint, alfalfa and parsley. Humans can also acquire infection by drinking contaminated water containing viable metacercariae (Chan and Lam, 1987). The incidence of animal and human infection rises during wet years because of an increased number of snails and longer survival of encysted cercariae (Arjona *et al.*, 1995).

In some areas, endemicity is almost 100 percent. In endemic regions very young children and women are most likely to be infected. There is a high incidence of co-infection with other parasites, especially echinococcosis (Arjona *et al.*, 1995).

2.7 Human Fascioliasis

Studies carried out in recent years have shown human fasciolosis to be an important public health problem (Chen and Mott, 1990). Human fasciolosis has been reported from countries in Europe, America, Asia, Africa and Oceania. The incidence of human cases is on the increase in 51 countries of the five continents (Esteban *et al.,* 1998). A global analysis shows that the expected correlation between animal and human fasciolosis only appears at a basic level. High prevalence in humans are not necessarily found in areas where fasciolosis is a great veterinary problem. For instance, in South America, hyperendemics and mesoendemics are found in Bolivia and Peru where the veterinary problem is less important, while in countries such as Uruguay, Argentina and Chile, human fasciolosis is only sporadic or hypoendemic (Esteban *et al.,* 1998).

In Europe, human fasciolosis occur mainly in France, Spain, Portugal, and the former USSR (Esteban *et al.,* 1998). France is considered an important human endemic area. A total of 5,863 cases of human fasciolosis were recorded from nine French hospitals from 1970 to 1982.Concerning the former Soviet Union, almost all reported cases were from the Tajik RepublicRecently, serological survey of human fasciolosis was performed in some parts of Turkey. The zero prevalence of the disease 3.01% in Antalya Province and between 0.9 and 6.1% in Isparta Province in the Mediterranean region of Turkey. In other European countries, fasciolosis is sporadic and the occurrence of the disease is usually combined with travelling to endemic areas (Esteban *et al.,* 1998).

In North America, the disease is very sporadic. In Mexico, 53 cases have been reported(Mas-Coma *et al.,*2005). In Central America, fasciolosis is a human health problem in the Caribbean islands, especially in zones of Puerto Rico and Cuba. Pinar del Río Province and Villa Clara Province are Cuban regions where fasciolosis was hyperendemic(Mas-Coma *et al.*,1999). In South America, human fasciolosis is a serious problem in Bolivia, Peru, and Ecuador. These Andean countries are considered to be the area with the highest prevalence of human fasciolosis in the world (Mas-Coma *et al.*, 1999). Well-known human hyperendemic areas are localized predominately in the high plain called altiplano. In the northern Bolivian altiplano,up to 72% and 100% prevalence were detected in some communities coprological and serological survey, respectively(Mas-Coma *et al.*,1999). In Peru, *Fasciola hepatica* in humans occurs throughout the country. The highest prevalence was reported in Arequipa, Mantaro Valley, Cajamarca Valley and Puno region(Mas-Coma *etal.*,2005). In other South American countries like Argentina, Uruguay, Brazil, Venezuela and Colombia, human fasciolosis appear to be sporadic, despite the high prevalence of fasciolosis in cattle (Mas-Coma *et al.*,2005).

In Asia, most human cases were reported in Iran, especially in the Gīlān Province, on the Caspian Sea. It was mentioned that more than 10,000 human cases were detected in Iran. In eastern Asia, human fasciolosis appears to be sporadic. Few cases were documented in Japan, Koreas, Vietnam and Thailand (Mas-Coma *et al.*, 2005).

In Australia, human fasciolosis is very rare (only 12 cases have been documented). In New Zealand, *Fasciola hepatica* has never been detected in humans(Mas-Coma *et al.*, 1999).

In Africa, human cases of fasciolosis, except in northern parts, have not been frequently reported. The highest prevalence was recorded in Egypt where the disease is distributed in communities living in the Nile Delta(Mas-Coma *et al.*, 2005).

2.8 Animal Fasciolosis

Countries where fasciolosis in livestock were repeatedly reportedinclude:

- Europe: United Kingdomn, Ireland, France, Portugal, Spain, Switzerland, Italy, Netherlands, Turkey, Germany, Poland (en.wikipedia.org/wiki/Fasciolosis).
- Asia: Russia, Thailand, Iraq, Iran, China, Vietnam, India, Nepal, Japan, Korea, Philippines (en.wikipedia.org/wiki/Fasciolosis).
- Africa: Kenya, Zimbabwe, Nigeria, Egypt, Gambia, Morocco (en.wikipedia.org/wiki/Fasciolosis).
- Australia and the Oceania: Australia, New Zealand(en.wikipedia.org/wiki/Fasciolosis).
- Americas:United States, Mexico, Cuba, Peru, Chile, Uruguay, Argentina, Jamaica, Brazil (en.wikipedia.org/wiki/Fasciolosis).

2.9 Fascioliasis in Nigeria

Researchers have attempted to provide data on economic and public health aspects of fascioliases in Nigeria. Estimates of economic losses due to fascioliases had been mainly derived from records of organ and carcass condemnation (Biu *et al.*, 2006). Meat derived from cattle, sheep and goats provides major sources of animal protein for the populace of Nigeria (Ekwunife and Eneanya,2006). Biu *et al.* (2006) observed that in Maiduguri abattoir, 41.7% of condemned organs consisted mainly of livers, were due to fasciolosis.

Hassan and Anwo (2011) from Isheri-Olofin in Ogun state reported 45.16% prevalence among slaughtered cattle at the abattoir. Likewise, Magaji *et al* ((2014) reported 27.68% prevalence of infections due to *Fasciola gigantica* in slaughtered cattle at Sokoto abattoir. FAO (2008) put the incidence of *Fasciola gigantica* in cattle in Nigeria at 60%, total loss for Nigeria was calculated to be 32.5million pounds sterling, though this represents a substantially lower estimate according to FAO.

2.10 Life Cycle and Transmission

The life cycle of *Fasciola* is complex (plate 3). It involves a final host (where the adult worm lives), an intermediate host (where the larval stages of the worm develop) and a carrier (entailing suitable aquatic plants)(WHO, 2012).

The process starts when infected animals (cattle, sheep, buffaloes, donkeys and pigs also horses, goats, dromedaries, camels, llamas and other herbivores) defecate in fresh-water sources. Since the worm lives in the bile ducts of such animals, its eggs are evacuated in faeces and hatch into larvae that lodge in a particular type of water snail which is *Radixauricularia*(the intermediate host)(WHO, 2012).

Once in the snail, the larvae reproduce and eventually release more larvae into the water. These larvae swim to nearby aquatic or semi-aquatic plants, where they attach to the leaves and stems and form small cysts (metacercariae). When the plants, with the small cysts attached, are ingested, they act as carriers of the infection. Watercress and water-mint are good plants for transmitting fascioliasis, but encysted larvae may also be found on many other salad vegetables. Ingestion of free metacercariae floating on water (possibly detached from carrier plants) may also be a possible mode of transmission (plate 3) (WHO, 2012).

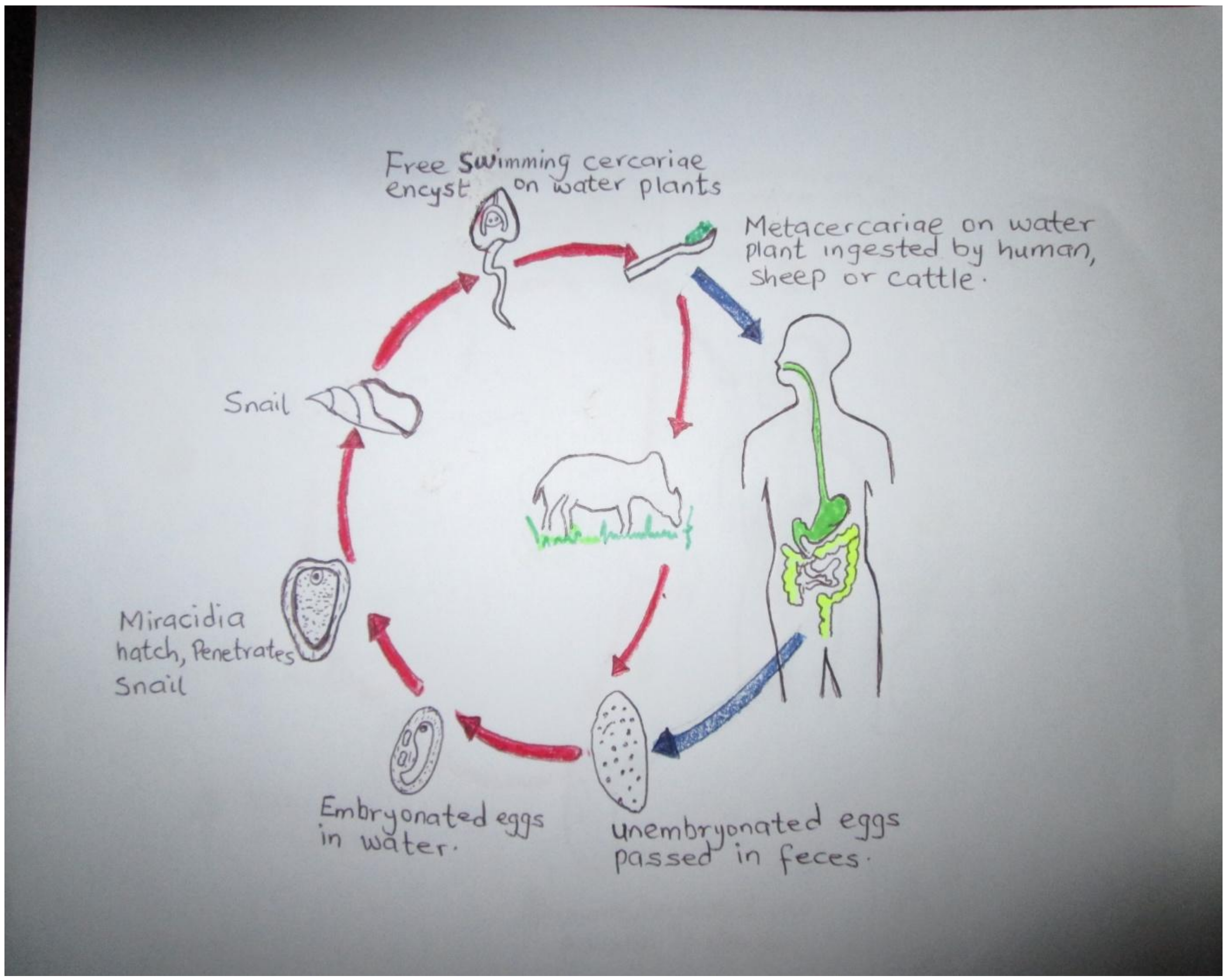


Plate 3: Life cycle of *Fasciola gigantica.*

2.10.1 Intermediate hosts

As with other trematodes, *Fasciola* develops in a molluscan intermediate host. Species of the freshwater snails from the family Lymnaeidae are well known for their roles as intermediate hosts in the life cycle of *Fasciola gigantica*. However, throughout the years an increasing number of other molluscan intermediate hosts of *Fasciola gigantica* have been reported (Soliman, 2008). The species of *Fasciola* can become adapted to new intermediate hosts under certain conditions at least based on laboratory trials. The most important intermediate host for *Fasciola gigantica* is *Radix auricularia*. However, other species are also known to harbour the fluke including *Lymnaea rufescens* and *Lymnaea acuminata* in the Indian Subcontinent; *Radix rubiginosa* and *Radix natalensis* in Malaysia and in Africa respectively; and the synonymous *Lymnaea cailliaudi* in East Africa (Soliman, 2008). Other snails also serve as natural or experimental intermediate such as *Austropeplea ollula*, *Austropeplea viridis*, *Radix peregra*, *Radix luteola*, *Pseudosuccinea columella* and *Galba truncatula* (Correa *et al.*, 2010).The Australian *Lymnaea tomentosa* (host of *Fasciola hepatica*) was shown to be receptive to miracidia of *Fasciola. gigantica* from East Africa, Malaysia and Indonesia(Soliman, 2008).

2.10.2 Definitive hosts

Fasciola gigantica is a causative agents (together with *Fasciola hepatica*) of fascioliasis in ruminants and in humans worldwide (Soliman, 2008).The parasite infects cattle and buffalo and can also be seen regionally in goats, sheep, and donkeys (Soliman, 2008).

2.10.3 Source of human transmission

Human *Fasciola hepatica* infection is determined by the presence of the intermediate snail hosts, domestic herbivorous animals, climatic conditions and the dietary habits of man (Chen and Mott, 1990). Sheep, goats and cattle are considered the predominant animal reservoirs. While other animals can be infected, they are usually not very important for human disease transmission. On the other hand, some authors have observed that donkeys and pigs contribute to disease transmission in Bolivia (Mas-Coma *et al.*,1998). In France, nutria (*Myocastor coypus*) was confirmed as a wild reservoir host of *Fasciola hepatica* (Menard *et al.*,2001). Humans are infected by ingestion of aquatic plants that contain the infectious cercariae. Several species of aquatic vegetables are known as a vehicle of human infection. In Europe, *Nasturtium officinale* (common watercress), *Nasturtium silvestris*, *Rorippa amphibia* (wild watercress), *Taraxacum dens leonis* (dandelion leaves), *Valerianella olitoria* (lamb's lettuce), and *Mentha viridis* (spearmint) were reported as a source of human infections.(Mas-coma *et al*., 1999) In the Northern Bolivian Altiplano, some authors suggested that several aquatic plants such as bero-bero (watercress), algas (algae), kjosco and tortora could act as a source of infection for humans (Bjorland *et al.,* 1995), because *Fasciola hepatica* cercariae also encyst on water surface, humans can be infected by drinking of fresh untreated water containing cercariae (Chen and Mott 1990). In addition, an experimental study suggested that humans consuming raw liver dishes from fresh livers infected with juvenile flukes could become infected (Taira *et al.*, 1997).

2.11 Pathogenesis

The development of infection in definitive host is divided into two phases: the parenchymal (migratory) phase and the biliary phase. The parenchymal phase begins when encysted juvenile flukes penetrate the intestinal wall. After the penetration of the intestine, flukes migrate within the abdominal cavity and penetrate the liver or other organs. *Fasciola hepatica* has a strong predilection for the tissues of the liver (Behm and Sangster, 1999). Occasionally, ectopic locations of flukes such as in the lungs, diaphragm, intestinal wall, kidneys and subcutaneous tissue can occur (Boray, 1969). During the migration of flukes, tissues are mechanically destroyed and inflammation appears around migratory tracks of flukes. The second phase (the biliary phase) begins when parasites enter the biliary ducts of the liver (Plate 4). In biliary ducts, flukes mature, feed on blood, and produce eggs. Hypertrophy of biliary ducts associated with obstruction of the lumen occurs as a result of tissue damage (Boray, 1969).

2.12 Clinical Signs

2.12.1 In animals

Clinical signs of fasciolosis are always closely associated with the infectious dose (amount of ingested metacercariae). In sheep, as the most common definitive host, clinical presentation is divided into 4 types:(Sinclair, 1962).

- **Acute Type I Fasciolosis**: infectious dose is more than 5000 ingested metacercariae. Sheep suddenly die without any previous clinical signs. Ascites, abdominal haemorrhage, icterus, pallor of membranes, weakness may be observed in sheep.

- **Acute Type II Fasciolosis**: infectious dose is 1000-5000 ingested metacercariae. As above, sheep die but briefly show pallor, loss of condition and ascites.
- **Subacute Fasciolosis**: infectious dose is 800-1000 ingested metacercariae. Sheep are lethargic, anaemic and may die. Weight loss is a dominant feature.
- **Chronic Fasciolosis**: infectious dose is 200-800 ingested metacercariae. Asymptomatic or gradual development of bottle jaw and ascites (ventral oedema), emaciation, weight loss.

In blood, anaemia, hypoalbuminemia and eosinophilia may be observed in all types of fasciolosis(Behm and Sangster, 1999). Elevation of liver enzyme activities, such asglutamate dehydrogenase (GLDH), gamma-glutamyl transferase (GGT), and lactate dehydrogenase (LDH), are detected in subacute or chronic fasciolosis from 12-15 week after ingestion of metacercariae (Anderson *et al*., 1981). In goats and cattle, the clinical manifestations are similar to sheep. However, acquired resistance to *Fasciola hepatica* infection is well known in adult cattle (Haroun and Hillyer, 1986). Calves are susceptible to disease but an infective dose in excess of 1000 metacercariae is usually required to cause clinical fasciolosis. In this case the disease is similar to sheep and is characterized by weight loss, anaemia, hypoalbuminemia and (after infection with 10,000 metacercariae) death (Anderson *et al*.,1981).

In sheep and sometimes cattle, the damaged liver tissues may become infected by *Clostridium* bacteria *Clostridium. novyi* type B. The bacteria will release toxins into the bloodstream resulting to Black Disease which is usually fatal. As *Clostridium.novyi* is common in the environment, black disease is found wherever populations of liver flukes and sheep overlap.

2.13 Resistance To Infection

Mechanisms of resistance have been studied by several authors in different animal species. These studies may help to better understand the immune response to *Fasciola hepatica* in host and are necessary in development of vaccine against the parasite. It has been established that cattle acquire resistance to challenge infection with *Fasciola hepatica* and*Fasciola gigantica* when they have been sensitized with primary patent ordrug-abbreviated infection (Haroun and Hillyer,1986). Resistance to fasciolosis was also documented in rats. On the other hand, sheep and goats are not resistant to re-infection with *Fasciola hepatica* (Chauvin *et al.*, 1995). However, there is evidence that two sheep breeds, in particular Indonesian thin tail sheep and Red Maasai sheep, are resistant to *Fasciola gigantica* (Robert *et al.*, 1997). No reports concerning resistance in humans are available.

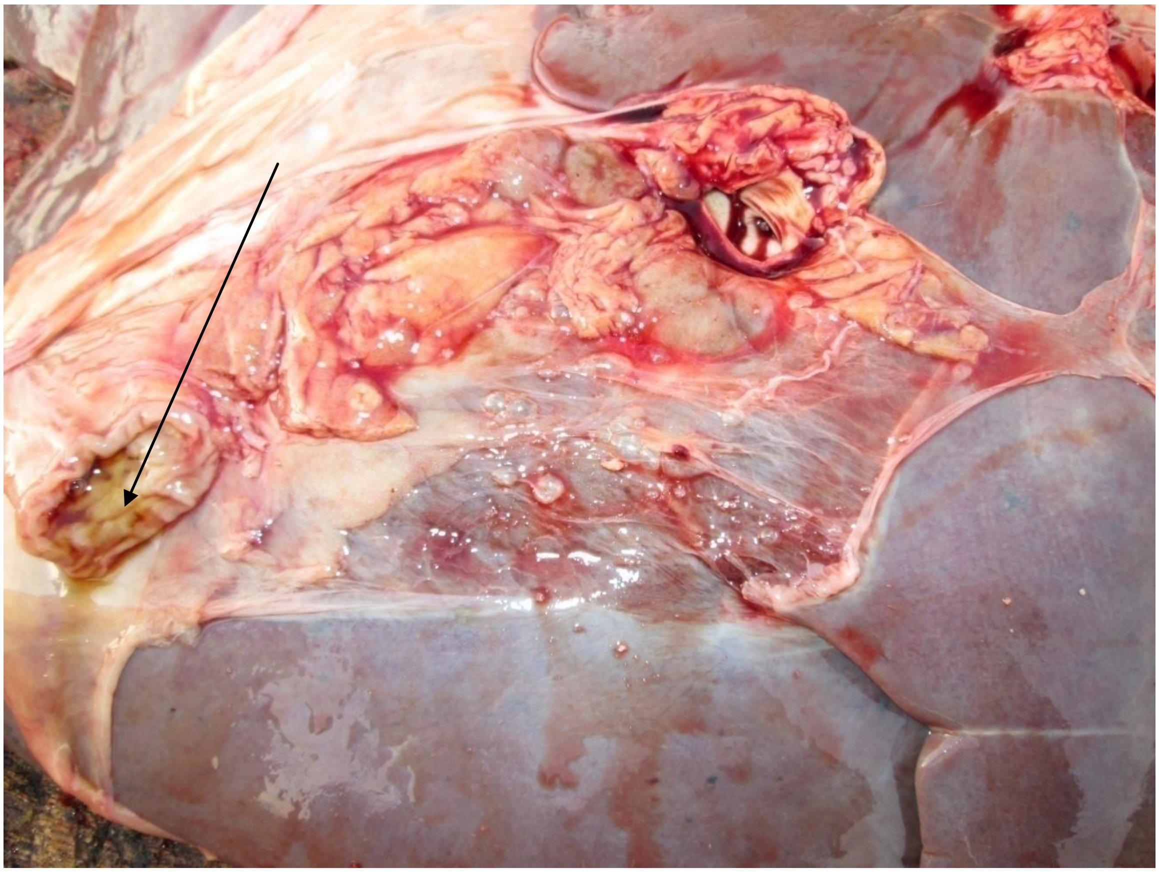


Plate 4: Biliary duct of the liver (Liver section; Bovine)

.

2.14 Diagnosis

Clearly, the appropriate diagnostic approach to the patient with possible fascioliasis depends first and foremost on a high index of suspicion, as well as the stage of the infection and the resources and expertise available(Mas-Coma *et al*.,1999).

In humans, diagnosis of fasciolosis is usually achieved parasitologically by finding the fluke eggs in stool and immunologically by ELISA and Western blot. Coprological examinations of stool alone are generally not adequate because infected humans have important clinical presentations long before eggs are found in the stools. Moreover, in many human infections, the fluke eggs are often not found in the faeces, even after multiple faecal examinations (Chen and Mott, 1990). Furthermore, eggs of *Fasciola hepatica*, *Fasciola gigantica* and *Fasciolopsis buski* are morphologically indistinguishable (Hillyer, 1988). Therefore, immunonological methods such ELISA and enzyme-linked immunoelectrotransfer blot, also called Western blot, are the most important methods in diagnosis of *Fasciola hepatica* infection. These immunological tests are based on detection of species-specific antibodies from sera. The antigenic preparations used have been primarily derived from extracts of excretory/secretory products from adult worms, or with partially purified fractions(Hillyer, 1999). Recently, purified native and recombinant antigens have been used, e.g. recombinant *Fasciola hepatica* cathepsin L like protease(O'Neil *et al.,*1998). Methods based on antigen detection (circulating in serum or in faeces) are less frequent. In addition, biochemical and haematological examinations of human sera support the exact diagnosis (eosinophilia, elevation of liver enzymes). Ultrasonography and biopsy of liver and gallbladder punctuate can also be used. False fasciolosis (pseudofasciolosis) refers to the presence of eggs in the stool resulting not from an actual infection but from recent ingestion of infected livers containing eggs. This situation (with its potential for misdiagnosis) can be avoided by having the patient follow a liver-free diet several days before a repeat stool examination (Hillyer, 1999).

In animals, intravital diagnosis is based predominantly on faecal examination and immunological methods. However, clinical signs, biochemical and haematological profile, season, climate conditions, epidemiological situation and examinations of snails must be considered. Similarly to humans, faecal examinations are not reliable. Moreover, the fluke eggs are detectable in faeces 8–12 weeks post-infection. In spite of that fact, faecal examination is still the only used diagnostic tool in some countries. While coprological diagnosis of fasciolosis is possible from 8-12 weeks post-infection (WPI) *Fasciola hepatica*- specific antibodies are recognized using ELISA or Western blot from 2-4 weeks post-infection (Dumenigo *et al*., 2000). Bile examination is known to give a higher percentage of positive findings (Oyeduntan *et al*., 2008).

2.14.1 Diagnostic imaging

In the appropriate clinical context, imaging studies can be very useful to confirm the diagnosis of fascioliasis (Koc *et al*., 2009). Typical findings include subcapsular haemorrhage; parenchymal nodular, ill-defined lesions, which may coalesce into tortuous or tubular tracks; filling defects in the biliary tract; and subcapsular, peribiliary, or periportal indistinct nodules and abscesses. Ultrasound, computed tomography, magnetic resonance imaging, and, to a lesser extent, nuclear medicine scanning all have a role. Endoscopic retrograde cholangiopancreatography and magnetic resonance cholangiopancreatography (MRCP) have been found to be useful in chronic disease when the adult worms occupy the biliary tree (Koc *et al*.,2009). In Nigeria, bile examination was used in the investigation of fascioliasis in slaughter house (Hassan and Anwo, 2011; Magaji *et al.,* 2014)

2.14.2 Histopathology

Rarely, a biopsy is performed and a tissue diagnosis is made, most often when the diagnosis has not been considered. With Endoscopic Retrograde Cholangiopancreatography (ERCP) removal of a worm, diagnosis can be confirmed as above. In ectopic fascioliasis, removal of the migrating metacercariae is diagnostic, as well. A liver biopsy typically reveals necrosis, acute and chronic inflammatory changes, debris and occasionally fragments of migrating larvae (Cantisani *et al.,*2010).

2.15 Current Control Strategy

Fascioliasis in humans is so poorly characterized compared to the infection in animals. Control measures are difficult to devise and implement. However, an understanding of the human health impact of the disease has increased and, support for control measures has increased as well.This effort is buoyed by the World Health Organization's (WHO) designation of fascioliasis as an extremely neglected disease(Mas-Coma *et al.,*2009). Due to the fact that the infection can be difficult to detect and can be transmitted in so many ways, control of fascioliasis has represented a significant challenge. The role of domestic and wild animal reservoirs, coexistence of the various *Fasciola* and snail species, and varying types of endemic situations have rendered the creation of a universal control strategy unrealistic (Mas-Coma *et al.,* 2009).

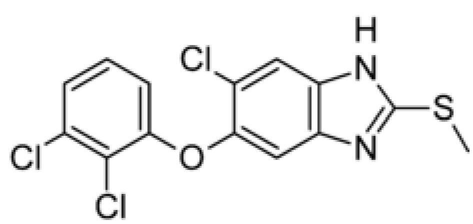
Current WHO guidelines recommend that hospitals maintain stockpiles of triclabendazole and that further classification studies regarding the nature of fascioliasis endemicity be undertaken in each endemic country; which vary widely in geography and socio-economic status from endemic France to hyperendemic Peru and Bolivia(WHO, 1999).

2.16 Treatment and Prevention

2.16.1 In humans

For high efficacy and safety, triclabendazole (Egaten) in dose of 10–12 mg/kg is the drug of choice in human fasciolosis (Savioli *et al*.,1999). Praziquantel treatment is ineffective (Price *et al*.,1993). There are case reports of nitazoxanide being successfully used in human fasciolosis treatment in Mexico (Rossignol *et al*.,1998). There are also reports of bithionol being used successfully (Rossignol *et al*.,1998).

More recently, Mirazid, an Egyptian drug made from myrrh, has been investigated as an oral treatment of trematode-caused ailments including fascioliasis.



Formula of triclabendazole (Mitchell, 1998).

2.16.2 In animals

A number of drugs have been used to control fasciolosis in animals. Drugs differ in their efficacy, mode of action, price and viability. Fasciolicides (drugs against *Fasciola* spp.) fall into five main chemical groups (Fairweather and Boray, 1999).

- **Halogenatedphenols**: bithionol (Bitin), hexachlorophene (Bilevon), nitroxynil (Trodax)
- **Salicylanilides**: closantel (Flukiver, Supaverm), rafoxanide (Flukanide, Ranizole)
- **Benzimidazoles**: triclabendazole (Fasinex), albendazole (Vermitan, Valbazen), mebendazol (Telmin), luxabendazole (Fluxacur)
- **Sulphonamides**: clorsulon (Ivomec Plus)
- **Phenoxyalkanes**: diamphenetide (Coriban)

Triclabendazole (Fasinex) is considered as the most common drug due to its high efficacy against adult as well as juvenile flukes. Triclabendazole is used in the control of fasciolosis of livestock in many countries. Nevertheless, long-term veterinary use of triclabendazole has caused appearance of resistance in *F. hepatica*. In animals, triclabendazole resistance was first described in Australia, later in Ireland and Scotland and more recently in the Netherlands(Moll *et al.*,2000). Considering this fact, scientists have started to work on the development of new drugs. Recently, a new fasciolicide was successfully tested in naturally and experimentally infected cattle in Mexico. This new drug is called 'Compound Alpha' and is chemically very similar to triclabendazole (Ibarra *et al*.,2004).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Study Area

The study was conducted in Kaduna metropolis, capital of Kaduna State; a state in north central Nigeria with a population of about 6,066,562 according to the 2006 population census (Canback, 2008). Kaduna is located on Latitude 10^{0}30’N and Longitude 07^{0} 21’E, with a total area of 46,053 km^2(17.781 sq metre), ranking 3rd in size out of the 36 States in Nigeria. Kaduna is inhabited by Fulani, Hausa, Bajju, Kadara, Jaba, Kagoro, Gbagyi and Kataf tribesmen alongside people from other ethnic groups in Nigeria like the Yoruba, Igbo, Igala, Idoma e.t.c. as well as foreign nationals.

The climatic condition of Kaduna is tropical continental with a distinct rainy and dry season. The rainy season occurs from May to October and the dry season from November to April. There are two main abattoirs (Sabo-Tash and Kakuri) within Kaduna metropolis where several of the cattle and goats are slaughtered on a daily basis. These two abattoirs constituted the sampling units for the study.

3.2 Sample Size Determination

The sample size for this study was 500 samples and included the bile samples of goats and cattle in the study area. This sample size was arrived at based on the assumption of normal approximation, using sample size calculation formula according to Naings *et al*. (2006).

$$n = \frac{Z^2 P(1-P)}{d^2}$$

Where; n = Sample size

Z = statistical level of confidence

P = Expected prevalence and

d = precision

For the 95% level of confidence, Z value is 1.96. P which is the expected prevalence was derived from previous study (Olusegun-Joseph *et al.*, 2011) which was 52.1% (0.52) and d at 0.05.

Substituting the values in the formula:

$$n = \frac{(1.96)2 \times 0.52(1-0.52)}{(0.05)2}$$

$$n = \frac{3.84 \times 0.25}{0.0025}$$

$$n = \frac{0.96}{0.0025}$$

$$n = 384$$

The least sample size is 384, but for the purpose of this study,500 samples were collected.

3.3 Sample Collection

Weekly visits were paid on rotational basis to the two slaughter houses for the purpose of collecting bile samples from slaughtered cattle and goats between 0700hours and 1000hours each sampling day. Samples were collected over a period of 12 weeks. During bile collection, the sex and age of the respective animals whose bile were collected were determined by examination of external genitalia and dentition respectively. This involves checking their teeth arrangement and their reproductive organs located outside the animal's body. Intact gall bladders removed from randomly selected cattle and goats slaughtered at the two abattoirs were collected from butchers. The bile samples from gall bladders of caprine and bovine were emptied by incision made with scissors into properly labelled small plastic bowls with lids and preserved with few drops of 10% formalin. The samples were transported to the Department of Parasitology and Entomology, Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria for the determination of prevalence and intensity of *Fasciola gigantica* and other hepatic parasitic infections.

3.4 Microscopic Examination of Bile for *Fasciola* species and Egg Count

Bile samples emptied into small plastic bowls was poured into separate beakers and mixed thoroughly by shaking for random distribution of eggs. About 10ml of each bile sample was introduced into a labelled centrifuge tube and centrifuged at 1,500 rpm for ten minutes. Two to three drops of water was added to the sediments to make it measure up to 1ml.This is for the purpose of having a uniform volume for the bile samples and also to make it easily observable under the microscope. Using a micro-pipette, 0.1ml of each sample was introduced on a slide covered with cover slip and examined under X40 magnification of a research microscope for the presence of *Fasciola gigantica* eggs. Identification of *Fasciola* eggs was based on the pictorial keys by Soulsby(1982).The total number of *Fasciola* eggs was determined by calculating the dilution factor and recorded. Dilution factor is calculated thus:

$$0.1ml = x\ eggs$$

$$1ml = \frac{x\ no\ of\ eggs\ \times 1}{0.1}$$

$$= \frac{x\ no\ of\ eggs\ X10}{1}$$

$$= 10x$$

Therefore for each sample, number of eggs found is multiplied by 10 which is the dilution factor to get actual number of eggs per 10ml per bile sample.

3.5 Statistical Analysis

Data obtained was analyzed using SPSS software for windows version 16.0 statistical package. Student’s t-test was used to test for significant differences in parasite intensities between abattoirs and significant difference between caprine age categories. Differences in parasite intensities among bovine age categories were assessed using ANOVA. Significance was determined at (P > 0.05) that is 95% level of significance.

CHAPTER FOUR

4.0 RESULTS

4.1 Prevalence of Caprine and Bovine Fascioliasis

The prevalence of caprine and bovine fascioliasis obtained in this study is presented in Table 4.1. A total of twenty eight (28) of the 250 samples of slaughtered caprine hosts were positive for *Fasciola gigantica* eggs, giving a prevalence of 11.20%. A total of one hundred and twenty seven (127) of the two hundred and fifty (250) bile samples obtained from slaughtered bovine hosts were positive for *Fasciola gigantica* eggs, giving a prevalence of 50.80%. Of the 250 caprine host sampled, 185 were males and 65 were female. A total of 21(11.35%) of the male caprine host were positive for *Fasciola* eggs, also 7(10.77%) of the female caprine sampled had *Fasciola* infection as presented in Table 4.1. Out of the 250 bovine host sampled, 116 were males and 134 were females. Of the 116 male bovine examined, 55(47.4%) had infection due to *Fasciola*. Of 135 female bovine sampled, 72(53.7%) were positive for *Fasciola* eggs.

4.2 Pattern of Infection of Caprine and Bovine With *Fasciola gigantica* and Other Parasites

Table 4.2 shows the pattern of single and mixed parasitic infections based on egg detections in bile samples of caprine and bovine slaughtered at abattoirs within Kaduna, Nigeria. Of the 185 male caprine examined, 37(20.00%) had single infection, 17(9.19%) of which were due to *Fasciola* and 20(10.81%) due to *Dicrocoelium.* A total of 17 (26.15%) of the 65 female caprine examined had single parasite, 4(6.15%) due to

Table 4.1: Gender based prevalence of caprine and bovine fascioliasis in slaughter house within Kaduna, Nigeria.

Animal Species	Sex	Number Examined	Number Infected	Prevalence (%)
Caprine (*Capra hircus)*	Male	185	21	11.35
	Female	65	7	10.77
	Total	250	28	11.20
Bovine (*Bos taurus)*	Male	116	55	47.40
	Female	134	72	53.70
	Total	250	127	50.80

Table 4.2: Pattern of detection of egg of *Fasciola* and other helminthes in bile samples obtained from slaughter houses in Kaduna, Nigeria.

Animal species	Egg type(s)	Number(%) of positive samples per sex		
Capra hircus (Caprine)	Single parasite infections	Male (n = 185)	Female (n = 65)	Total
	Fasciola gigantica	17 (9.19)	4 (6.15)	21 (8.40)
	Dicrocoelium dendriticum	20 (10.81)	13 (20.00)	33 (13.20)
		37 (20.00)	17 (26.15)	54 (21.60)
	Mixed infections			
	F. gigantica, D. dendriticum	4 (2.16)	3 (4.62)	7 (2.80)
Bos taurus (Bovine)	Single parasite infections	Male (n = 116)	Female (n = 134)	Total
	Fasciola gigantica	4 (3.45)	13 (9.70)	17 (6.80)
	Dicrocoelium dendriticum	40 (34.48)	40 (29.85)	80 (32.00)
		44 (37.93)	53 (39.55)	97 (38.80)
	Mixed infections			
	F. gigantica, D. dendriticum	51 (43.97)	59 (44.03)	110 (44.00)

Fasciola gigantica and 13(20.00%) due to *Dicrocoelium spp*. In all, *Dicrocoelium spp* was responsible for 61.1% (33/54) of single infection in the sampled caprine. Mixed infections with two parasites were observed in 4(2.16%) of the 185 male caprine and 3(4.62%) of the 65 female caprine examined. A total of 7(2.80%) caprine belonging to both sexes were infected with mixed infections of two parasites predominated by *Fasciola gigantica* and *Dicrocoelium dendriticum*. Of the 116 male bovine examined, 44(37.93%) had single infection, 40(90.9%) of which were due to *Dicrocoelium spp* alone. A total of 53(39.55.%) of 134 female bovine had single infection, 40(74.1%) of which were due to *Dicrocoelium spp*. In all 97(38.80%) of the 250 bovine hosts sampled had single infection, with *Dicrocoelium spp* causing 82.5% (80/97) of the cases. Overall, the prevalence rate of *Dicrocoelium spp* in the sampled bovine was 32%. Eggs of *Fasciola* and *Dicrocoelium* are presented in Plate 5. Mixed infections with two parasite egg types occurred in 51(43.97%) of the 116 male bovine sampled, while the prevalence of mixed infections in the 134 female bovine sampled was 44.03%. Thus, a total of 110(44.00%) of the 250 bovine hosts examined had mixed infections, mainly due to with *Fasciola gigantica* and *Dicrocoelium dendriticum.*

4.3 Age-Specific Prevalence and Intensity of Caprine and Bovine Fascioliasis and other Parasites

The caprine encountered in this study fell into four in Table 4.3. Seven (5.38%) and 9(6.92%) of the 130 caprine aged >1-<2 were infected by *Fasciola gigantica* and *Dicrocoelium spp* respectively. The respective prevalence values (%) for caprine in the >1-<2 year age group were 11.67 and 19.17 (Table 4.3). The intensities of *Fasciola gigantica* eggs in bile samples of infected caprine ranged from 3.71 eggs per ml of bile in

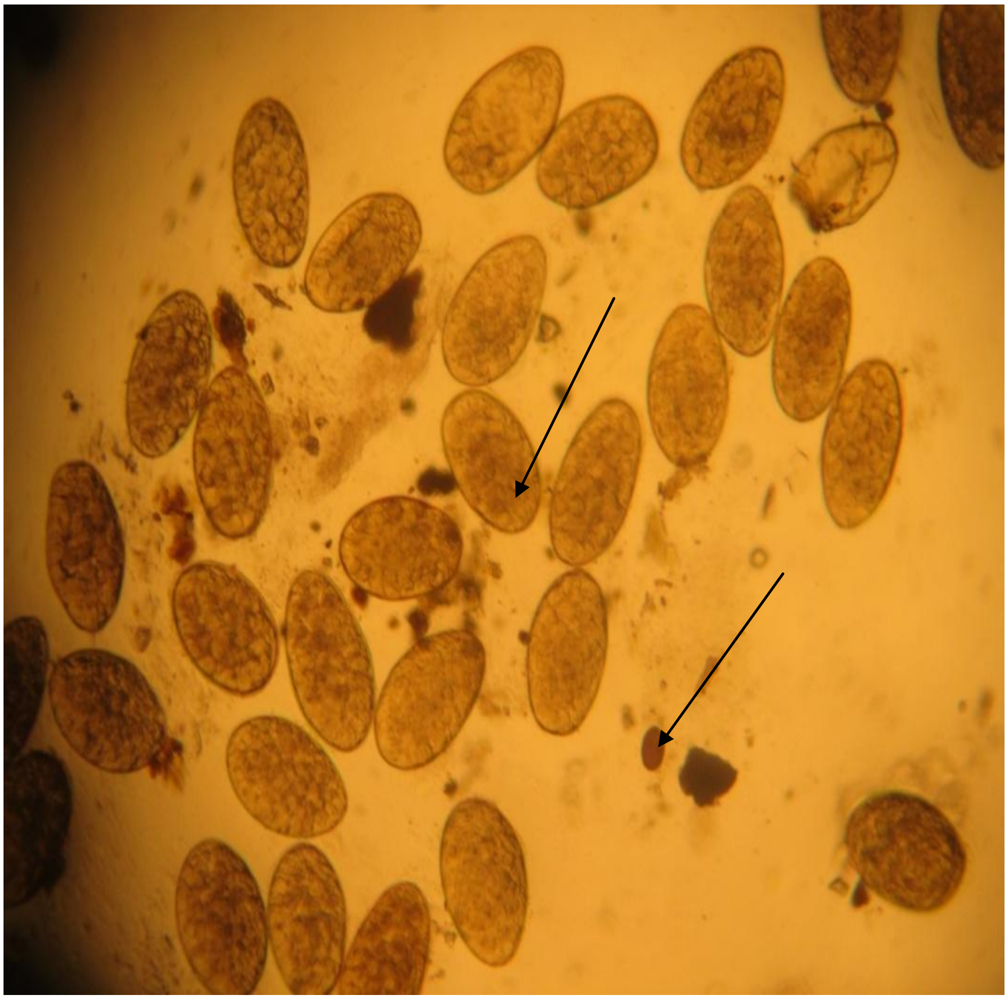


Plate 5: Egg of *Fasciola gigantica* (A) and *Dicrocoelium dendriticum* (B)

Magnification=X400

those aged >1-<2 years to 7.29 eggs per ml of bile in those aged >2-<3 years. The intensities of *Fasciola gigantica* eggs in bile samples of caprine belonging to the age categories did not differ significantly (P>0.05). The intensities of *Dicrocoelium dendriticum* eggs in bile samples of infected goats ranged from 20.91 eggs per ml of bile in those aged >2-<3 years to 41.22 eggs per ml of bile in those aged >1-<2 years. The intensities of *Dicrocoelium dendriticum* eggs in bile samples of caprine belonging to the age categories did not differ significantly (P>0.05) (Table 4.3).

The bovine examined in this study fell into three age categories; those aged 1-3 years, those aged 4-6 years and those aged 7-9 years. The modal age group was 4-6 years where 87/156 (55.77%) was infected by *Fasciola gigantica*, but the highest prevalence 16/27 (59.26%) was obtained in those aged 7-9 years and the least prevalence of *Fasciola gigantica*, 27/67 (40.30%), was obtained amongst the slaughtered bovine aged 1-3 years. The intensities of *Fasciola gigantica* eggs in bile samples of infected bovine increased with age and bovine ranged from 62.26 eggs per ml in those aged 1-3 years to 965.25 eggs per ml in those aged 7-9 years. The intensities of *Fasciola gigantica* eggs in bile samples of bovine belonging to the age categories differed significantly (P<0.05) in the trend 7-9years>4-6years>1-3years (Table 4.3).

The modal age group of bovine infected by *Dicrocoelium spp* was 4-6 years, with 122/156 (78.21%) prevalence. Fifty out of 67 bovine (74.63%) aged 1-3 years were positive for *Dicrocoelium spp* while the least prevalence 18/27 (66.67%) was obtained amongst those aged 7-9 years. The intensities of *Dicrocoelium dendriticum* eggs in bovine bile samples ranged from 360.62 eggs per ml of bile in those aged 1-3 years to

Table 4.3: Age specific prevalence and intensity of caprine and bovine fascioliasis and associated parasitosis in slaughter houses within Kaduna, Nigeria

Animal species (type)	Age categories (years)	Number examined	*Fasciola*		*Dicrocoelium*	
			Number positive (Prevalence %)	Intensity ±SD	Number positive (Prevalence%)	Intensity ±SD
Capra hircus (caprine)	<1- 1	0	0 (0.00)	0.00 ±0.00	0 (0.00)	0.00 ±0.00
	>1- <2	130	7 (5.38)	3.71^{c}± 3.99	9 (6.92)	41.22^{b} ± 105.61
	>2- <3	120	14 (11.67)	7.29^{c} ± 8.15	23 (19.17)	20.91^{b}± 36.34
	>3 and above	0	0 (0.00)	0.00 ± 0.00	0 (0.00)	0.00 ± 0.00
Bos taurus (bovine)	1 – 3	67	27 (40.30)	62.26^{c} ± 95.30	50 (74.63)	360.62^{b} ±284.18
	4 – 6	156	87 (55.77)	296.31^{b}±414.32	122 (78.21)	406.5^{b} ±400.09
	7 – 9	27	16 (59.26)	965.25^{a} ±466.35	18 (66.67)	714.39^{a}±495.90

Notes: Column intensities (means) of same animal species and parasite followed by same superscript are not significantly different (P > 0.05).

714.39 eggs per ml in those aged 7-9 years. Intensities of *Dicrocoelium dendriticum* eggs were significantly ($P<0.05$) higher in those aged 7-9 years than in the other two lower age categories; however there was no significant difference ($P>0.05$) between these two lower age categories.

4.4 Site-Specific Prevalence and Intensity of Bovine Fascioliasis and Other Parasitosis

Table 4.4 shows the site-specific prevalence and intensity of bovine fascioliasis and other parasitosis in Kakuri and Sabo-Tasha slaughter houses within Kaduna, Nigeria. Of the 250 bovine examined, 127(50.80%) were infected with *Fasciola gigantica* in both slaughter houses. Prevalence of *Fasciola gigantica* infection ranged from 24/57 (42.11%) in bovine slaughtered at Kakuri abattoir to 103/193 (53.37%) in those slaughtered at the Sabo-Tasha abattoir. The intensities of *Fasciola gigantica* eggs ranged from 131.13 eggs per ml of bile in bovine slaughtered at Kakuri abattoir to 393.55 eggs per ml of bile in those slaughtered at the Sabo-Tasha abattoir. The intensity of *Fasciola gigantica* eggs was significantly higher ($P<0.05$) in bovine slaughtered at the Sabo-Tasha abattoir than those slaughtered at the Kakuri abattoir (Table 4.4). The prevalence of *Dicrocoelium dendriticum* in bovine slaughtered at both slaughtered houses in Sabo-Tasha and Kakuri was 193/250 (77.20%). Prevalence of *Dicrocoelium dendriticum* in bovine slaughtered at the Sabo-Tasha abattoir was 145/193 (75.13%) while the prevalence in those slaughtered at the Kakuri abattoir was 48/57 (84.21%). The intensities of *Dicrocoelium dendriticum* eggs ranged from 215.85 eggs per ml of bile in bovine slaughtered at Kakuri abattoir to 491.73 eggs per ml of bile in bovine slaughtered at the Sabo-Tasha abattoir (Table 4.4).

Table 4.4: Site specific prevalence and intensity of bovine fascioliasis and associated parasitosis in slaughter houses within Kaduna, Nigeria

Location	Number examined	Number (%) with *Fasciola*	*Fasciola* intensity ±SD	Number (%) with *Dicrocoelium*	*Dicrocoelium* intensity ±SD
Kakuri abattoir	57	24 (42.11)	131.13^{b}±183.34	48 (84.21)	215.85^{b}±208.86
Sabo abattoir	193	103 (53.37)	393.55^{a}± 488.79	145 (75.13)	491.73^{a}±416.23
Total	250	127 (50.80)	343.96	193 (77.20)	423.12

Note: Column intensities (means) followed by same superscript are not significantly different (P > 0.05)

The intensities of *Dicrocoelium dendriticum* eggs was significantly higher ($P<0.05$) in bovine slaughtered at the Sabo-Tasha abattoir than those slaughtered at Kakuri abattoir (Table 4.4).

CHAPTER FIVE

5.0 DISCUSSION

This study has revealed that caprine and bovine species slaughtered at abattoirs within Kaduna metropolis were infected with *Fasciola gigantica*, the causative agent of fascioliosis. The eggs of this parasitic trematode were present in the bile samples of 11.20% and 50.80% of slaughtered caprine and bovine hosts respectively. This is an indication that *Fasciola gigantica* remains an endemic parasitic trematode infection in small and large ruminants in the northern part of Nigeria which is domain of the livestock resources of the country. The prevalence obtained by examination of bile samples for egg within the predilection sites of the parasite (biliary duct of the liver) especially in dead hosts could better represent true positive confirmations over coprology method. Ulayi *et al.* (2007) and Hassan and Anwo (2011) also adopted bile examination in their investigations of slaughtered cattle at Zaria (Kaduna State) and Isheri-Olofin (Ogun State), Nigeria respectively. The marked differences in observed prevalence between both species of animals could be due to differences in exposure risks to infective metacercariae of the trematode, pertaining to different husbandry practices employed in raising the two categories of animals. Majority of small ruminants (especially caprine) are being raised as semi-extensive sedentary village livestock subjected to seasonal confinement and their distributions mirror those of human settlements (Kosgey *et al.*,2008; Umunna *et al.*,2014). Conversely, most of the bovine in Nigeria are owned by nomadic Fulani and their stocks are raised under the pastoral system of production, involving grazing associated with long distance movements in search of pasture (Nuhu, 1988; Daodu *et al.*, 2009; Aliyu *et al.,* 2014). Bovine raised under the pastoral system of production are exposed to variable ecological settings where they graze, contaminate the environment with parasites' egg- laden faeces and could ingest metacercariae- infested pasture. These cycles of exposures could heighten infectivity that would culminate in the high prevalence observed in bovine hosts in this study. The husbandry practices associated with caprine production were diversified but mainly homestead and the animals in most cases were not subjected to long distance trekking in search of pasture which could reduce their chances of contracting metacercariae; hence the lower observed prevalence. Caprine fascioliasis is rarely investigated compared to the numerous studies on bovine fascioliasis in Nigeria and other worldwide foci, despite the broad definitive host range of the parasite that cuts across several herbivorous mammals including humans. The caprine *Fasciola gigantica* prevalence of 11.20% obtained in this study is higher than the 0.28% obtained in Maiduguri by Mbaya *et al.*(2010) but lower than the 32.9% recorded for the species in Argentina by Cuervo *et al.* (2013). The prevalence of bovine fascioliasis obtained in this study was higher than 37.1% that was reported by Ulayi *et al.* (2007) and also higher than 23.3% prevalence reported by Njoku (2011) from Imo abattoirs, Nigeria also higher than 13.62% prevalence reported by Usip *et al*. (2014) from Akwa-Ibom abattoir and higher than 27.68% prevalence reported by Magaji *et al*. (2014) from Sokoto abattoir. This may be due to different agro-climatic conditions (Bhutto *et al.,* 2012) and also difference in resistance to infection because of the host breed and grazing habits (Tasawar *et al.*, 2007). Also, difference in infection may have been influenced by the varying ecological and climatic conditions of the area where they might have grazed upon before getting to the abattoir (Aladi, 1999). *Fasciola gigantica* is a zoonotic parasite and its high prevalence in animals that occur close to humans might suggest possible risk of human exposure. Humans usually acquire *Fasciola* infection through the consumption of uncooked metacercariae - infested vegetables. Badaki *et al.* (2005) reported a 19.3 percent prevalence of *Fasciola* among school children in parts of northern Nigeria which is higher than 11.20 percent prevalence obtained for caprine in this study. This portend the zoonotic importance of *Fasciola* species in Nigeria and the need to monitor and reduce the burden of the parasite in domesticated animals so as to check the plausibility of elevated contamination of human vegetable food items with infective stages of the parasite.

Polyparasitism, especially of *Fasciola gigantica* and *Dicrocoelium dendriticum*, were observed in both animal hosts (Table 2). The overall prevalence of *Fasciola gigantica* in mixed infection in bovine at the studied abattoirs is comparable to that which was reported by Biu *et al.* (2006) at Maiduguri abattoir. The observed low prevalence of single infection with *Fasciola gigantica* in male bovine and female bovine conforms to the low prevalence of single infection of *Fasciola gigantica* with 15.52% reported by Idowu *et al.* (2006) in Abeokuta, Nigeria and Phiri *et al.* (2006) in Zambia. Single infection with *Dicrocoelium dendriticum* in bovine reported in this study was 32.09% and that of caprine was 13.29%. That of the bovine conforms to 30.16% reported by Hassan and Anwo, (2011) from Isheri Olofin abattoir in Ogun-State, Nigeria. The prevalence of both single and mixed parasitic infections was low in caprine, this is due to the fact that most of the goats brought to the slaughter houses were intensively raised and might not have travelled far along the trade route to have ingested metacercariae on vegetations. Another reason could be that caprine and ovine do not support worm burden, they die if highly infected before they are bought to the abattoir. Prevalence of single and mixed *Fasciola* infection in bovine was higher in female than male, so also was the intensity. The prevalence conforms to the prevalence, recorded by Affroze *et al* (2013) with 41.3% in females and 13.8% in males respectively; It also agrees with the results of Onyeabor and Wosu (2014) who reported rates of 17.82% and 27.82% in male and female bovine in Abia State. This is a sharp contrast with the studies earlier reported by Idris and Mandara (2005); and Obadiah (2010), who reported a higher infection rate among the male bovine than the female bovine from Gwagwalada abattoir, Federal Capital Territory, Abuja, Nigeria and Jalingo abattoir, Taraba state, Nigeria, respectively. The possible explanation could be that both sexes move together in search of food and water and therefore possibility for both sexes to be equally exposed to the risk of the infection is high (Idris and Madara 2005). This variation could also be due to the intrinsic factors (genetics, physiology and immunology) and extrinsic factors (environment and management practises) that exist between male and female bovine. The high prevalence of single infection of bovine and caprine with *Dicrocoelium spp* over prevalence of single infection with *Fasciola* for bovine and caprine is noteworthy. This conforms with the higher prevalence of *Dicrocoelium* eggs over *Fasciola* eggs recorded by Hassan and Anwo (2011). This may be due to the fact that the second intermediate host for *Dicrocoelium* is the brown ant, a terrestrial invertebrate, which the host can readily ingest during upland grazing, in contrast to *Fasciola* metacercariae which might only be ingested during dry season low-land grazing along the river banks.

Caprine and bovine of different age range were brought to the slaughtering houses (Table 3), considerably high rate of infection was found in adult bovine. Age-specific intensities of bovine *Fasciola* infection were statistically significantly different ($p < 0.05$) among the age group. Intensities of bovine *Dicrocoelium* and *Fasciola* infection were statistically significantly higher ($p < 0.05$) in animals aged 7-9 years than the ones aged 1-3. Higher percentage prevalence in bovine aged 7-9 over percentage prevalence in bovine aged 1-3 reported in this study agrees with prevalence of 33.3% in bovine >3 over 11.0% in bovine <3 that was reported by Affoze *et al* (2013). High prevalence observed in the adult bovine from this study could be due to the fact that adult bovine must have grazed on contaminated areas while travelling through their trade route (FAO,1999). The reason could also be due to the intake of parasites for a longer period and their grazing habit close to water-logged areas. Bovine *Dicrocoelium* infections were not statistically significantly different ($p > 0.05$) in intensities between animals aged 1-3 years and those aged 4-6 years**.** Also, it was observed that the bovine within the age groups of 1-3years and 4-6years were slaughtered and analyzed more than older ones between age group 7-9.The reasons might be that the beef of younger bovine are tender and more palatable. Infection was both high and severe in adult bovine than the young bovine as seen in table 3. This suggest that there could be resistance to infection by young bovine or their intestine are too tender for worm attachment (Hill and Onabamiro, 1961). Keyyu *et al* (2005) also reported that the high infection rate in older animals might be associated with age and consequently longer exposure with time. Young bovine are not subjected to long distant movement along their trade route and this may have reduced their chances of picking the infection while grazing (FAO, 1999).

Statistically, from this study, the intensity of both *Dicrocoelium* and *Fasciola* egg were significantly higher in bovine slaughtered at the Sabo-Tasha abattoir than those slaughtered at Kakuri abattoir (Table 4). This could be as a result of the fact that most of the bovine brought to Sabo-Tasha abattoir have acquired infections from the various sources from where they were brought from before getting to the abattoir. During the sample collection, it was observed that high intensity of *Fasciola* and *Dicrocoelium* eggs was found in gall bladder that is larger with more volume of bile. Olusegun-Joseph (2011) also reported that the intensity of *Fasciola* eggs found significantly correlated with the size of the gall bladder. Thus the claim made by the butchers at the abattoir while samples were collected about the size of gall bladder being indicative of *Fasciola* infection was true to a large extent: since an infected liver is often enlarged, so also, the gall bladder that is closely linked to it will be enlarged. The large-sized gallbladder obtained with *Fasciola* eggs present in them could have been due to obstruction of the bile ducts caused by the flukes leading to the accumulation of the bile fluid. Although in this study liver samples were not analyzed directly, it is expected that result obtained using gall bladder contents is an indication of liver infections. Cawdery *et al*. (1977) had suggested that a prevalence rate up to 25% in indicative of a level of infection in which most of the animals affected would have had their liver damaged, rendering them unfit for human consumption. This often times result to production loss due to mortality; lower production of meat, milk and wool; reduced weight gain; metabolic disease and impaired fertility (Mason, 2004). The value of the losses resulting from this disease runs into millions of naira (Ogunrinade and Ogunrinade, 1980). Apart from direct economic losses associated with the disease, other non quantifiable losses are also experienced. Also, high prevalence of bovine fascioliasis recorded in this study could elevate human exposure to risk infection as it has already being reported among school children in Northern Nigeria (Badaki *et al.*,2010). The prevalence rate of 50.80% in bovine fascioliasis recorded place a probability that each liver supplied to the market is unfit for public consumption at 50% (0.5). This is consistent with the general observation that some of the bovine brought to the abattoir for slaughter were apparently diseased and weak. Some barely made it to the abattoir alive.

CHAPTER SIX

6.0 CONCLUSIONS AND RECOMMENDATIONS

6.1 Summary

Prevalence of *Fasciola gigantica* infections in bile samples of goats and cattle slaughtered in abattoirs within Kaduna metropolis, Nigeria was high with values of 11.20% and 50.80% for caprine and bovine hosts respectively from this study. There were incidences of single and mixed infection of *Fasciola gigantica* with other parasites, like *Dicrocoelium dendriticum*. Mixed infections with *Fasciola gigantica and Dicrocoelium dendriticum* with prevalence of 2.80% in caprine and 43.60% in bovine was obtained respectively.

6.2 Conclusions

The result of this study clearly indicates that prevalence of liver fluke infections remains high in the study area. The overall prevalence of trematodes parasites eggs in gall-bladder observed in bovine (50.80%) and caprine (11.20%) was high. The findings of this present study imply that bovine and caprine fascioliasis is still a problem to livestock in the study area. Therefore the need to investigate the pattern of prevalence trematodes' infections in cattle and goats slaughtered at the abattoir cannot be over-emphasized.

4.5 Recommendations

Based on the findings from this study, more enlightenment is needed to enhance knowledge, attitudes and practice among cattle and goats herdsmen for better management of trematodes infections. Treatment of the grazing and slaughtering locations with molluscides to reduce the incidence of the intermediate host which are the snails. Control of Fascioliasis should be done which should involve actions such as detailed study of the epidemiology of the disease in ruminants and snail intermediate hosts and determining well defined intervals for anthelmintic treatment of infected animals. Abattoir record keeping should be reviewed to provide information about disease of animal. The Federal Government in conjunction with Health and Agricultural Agencies should also assist to provide adequate veterinary health enlightment programme for early detection of infected animals by cattle and goats rearers in the study area.

Appendix

Appendix 1: Gall bladder with the bile content inside

Appendix 2: Bile samples in labeled plastic containers

REFERENCES

Adedokun,O.A.,Ayinmode,A.B. and Fagbemi,B.O.(2008).Seasonal prevalence of *Fasciola gigantica* infection among the sexes in Nigerian cattle.*Veterinary Research* ,**2** (10):12-14.

Affroze, S., Begun, N., Islam, M.S., Rony, S.A., Islam, M.A. and Mondal, M.M.H. (2013). Riskfactors and gross pathology of bovine liver fluke infection at Netrokona district, Bangladesh. *Journal of Animal Science Advances,* **3** (2): 83-90.

Aladi,N.O.(1999).Current trends in the production, handling and sales of meatin Nigeria. *Bachelor of Agriculture Thesis, Federal University of Technology Owerri*,71pp.

Aliyu, A.A., Ajogi, I.A., Ajanusi, O.J. and Reuben, R.C. (2014). Epidemiological studies of*Fasciola gigantica* in cattle in Zaria, Nigeria using coprology and serology. *Journal ofPublic Health and Epidemiology,* **6** (2): 85-91.

Alonge,D.O.and Fasanmi,E.F.(1979).A survey of abattoir data in Northern Nigeria. *Tropical Animal Health and Production,* **1**:57-62.

Anderson, P.H.,Matthews, J.G., Berrett, S., Brush, P.J. and, Patterson, D.S. (1981).Changes in plasma enzyme activities and other blood components in response to acute and chronic liver damage in cattle.*Research in Veterinary Science,***31**: 1–4.

Arjona, R., Riancho, J.A. and Aguado, J.M. (1995). Fascioliasis in developed countries: a review of classic and aberrant forms of the disease. Medicine (Baltimore); **74**:13.

Badaki, J.A., Shittu, K. and Akogun, O.B. (2005).Geohelminthiasis amongst school children intwo local government areas of Taraba State, Nigeria. *Journal of Tropical Biosciences,* **5**:38-41.

Behm, C.A. and Sangster, N.C.(1999).Pathology, pathophysiology and clinical aspects.In: *Fasciolosis* Dalton, J.P. (Ed).CAB International Publishing, Wallingford, pp. 185–224.

Bhutto, B., Arijo, A., Phullan, M.S. and Rind, R. (2012).Prevalence of fascioliasis in buffaloesunder different agro-climatic areas of Sindh Province of Pakistan. *International Journal of Agriculture and Biology,* **14**: 241-245.

Biu,A.A. and Babangana,M.(2004).Fascioliasis:Post mortem worm count and faecal analysis in cattle of Borno State, Nigeria. *Journal of Science and Technology Research*,**3**(3):4-6.

Biu, A.A., Ahmed, M.I. and Mishelia, S.S. (2006). Economic assessment of losses due to parasitic diseases common at the Maiduguri Abattoir.*African Scientist,***7**:1-3.

Bjorland, J., Bryan, R.T., Strauss,W., Hillyer, G.V.,Mcauley,J.B. (1995). An outbreak pf acute fascioliasis among Aymara Indians in Bolivian Altiplano.*Clinical Infectious Disease,* **21** (5): 1228-1233.

Boray, J.C. (1969).Experimental fascioliasis in Australia.. *Advances in Parasitology,***7**: 95–210..

Canback Dangel (2008). Canback Global Income Distribution Databack. C-GIDD.

Cantisani,V.,Cantisani, C. and Martele, K. (2010). Diagnostic imaging in the study of human hepatobiliary fascioliasis.*Radiology Medline* **115**:82-83.

Cawdery,H.M.J., Strickland,L.K.and Crowe,P.J.(1977).Production effects of liver fluke in cattle,the effect on live weight gain,feed intake and food conversion efficiency in beef cattle.*British Veterinary Journal*,**133**:145-159.

Chan, C.W. and Lam, S.K. (1987). Diseases caused by liver flukes and cholangio carcinoma:.*Bailliereis Clinical Gastroenterology*, **1 (2)**:297-.318

Chauvin, A., Bouvet, G. and Boulard, C. (1995).Humoral and cellular immune responses to *Fasciola hepatica* experimental primary and secondary infection in sheep. *International Journal Parasitology,* **25** (10): 1227–1241.

Chen, M.G.and Mott, K.E. (1990). Progress in assessment of morbidity due to *Fasciola hepatica* infection: A review of recent literature. *Tropical Diseases Bulletin,* **87:**R1–R38.

Cook,B.R.(1989).The epidemiology of *Echinococcus granulosus* in Great Britain.*Annals of Tropical Medicine and Parasitology*, **83** (1):51-61.

Cuervo, P., Sidoti, L., Fantozzi, C., Neira, G., Gerbeno, L. and Sierra, R.M. (2013).*Fasciolahepatica* infection and association with gastrointestinal parasites in Creole goats fromwestern Argentina.*Revista Brasileira de Parasitologia Veterinaria,* **22**(1):53-57.

Daodu, M.O., Babayemi, O.J. and Iyayi, E.A. (2009). Herd composition and managementpractices of cattle production by pastoralists in Oyo area of southwest Nigeria.*LivestockResearch for Rural Development,* 21(5), Article ♯ 66. Retrieved August 17, 2015 from http://www.lrrd.org/lrrd2115/daod21066.html.

Doyle, J. J. (1973).The relationship between the duration of a primary infection and the subsequent development of an acquired resistance to experimental infection with *Fasciola hepatica* in cattle. *Research in Veterinary Science,* **14**: 97- 103.

Duménigo, B.E., Espino, A.M., Finlay, C.M. and Mezo, M. (2000).Kinetics of antibody-based antigen detection in serum and faeces of sheep experimentally infected with *Fasciola hepatica*. *Veterinary Parasitology,* **89** (1–2): 153–161.

Ekwunife,C.A.and Eneanya,C.I. (2006).*Fasciola gigantica* in Onitsha and Environs. *Animal Research International,***3** (2):448-450.

En.wikipedia.org/wiki/Fascioliasis

Esteban, J.G., Bargues, M.D.and Mas-Coma, S. (1998). Geographical distribution, diagnosis and treatment of human fascioliasis: a review. *Research and Review. Parasitology*, **58** (1):13–42.

Fabiyi,J.P. and Adeleye,G.A.(1982).Bovine fascioliasis on Jos Plateau, Northern Nigeria with particular reference to economic importance: *Bulletin of Animal Health and Production in Africa,* **30** (1):41-43.

Facey, R.V. and Marsden, P.D.(1960). Fascioliasis in man: an outbreak in Hampshire. *Britain Medline Journal,* **2**:619–625.

Fairweather, I. and Boray, J.C. (1999).Fasciolicides: efficacy, actions, resistance and its management. *Veterinary Journal,* **158** (2): 81–112.

Fitzgerald, P.R and Mansfield, M.E.(1969). Economic significance of coccidiosis in calves. *Journal of Parasitology*, 55:39.

Food and Agricultural Organization.(1999).Report of expert consultationin helminth infections of livestock in developing countries. Rome, Italy, 23-27.

Food and Agricultural Organization.(2008).Distribution and impact of helminth diseases of livestock in developing countries. http://www.fao/docrp/004/t0584e/T0584E03.htm>

Graczyk,T.K.and Fried,B. (1999). Development of *Fasciola hepatica* in the intermediate host.In: *Fascioliosis* Dalton, J.P(Ed.)CAB International Publisher. Wallingford, Oxon United Kingdom, Pp 31-46.

Haroun, E.T.and Hillyer, G.V. (1986). Resistance to fascioliasis-a review. *Veterinary Parasitology.* **20** (1–3): 63–93.

Haseeb, A.N., El-Shazly, A.M., Arafa, M.A and Morsy, A.T. (2002).A review on fascioliasis in Egypt. *Journal of Egypt Society of Parasitology,* **32**(1): 317-354.

Hassan,A.A. and Anwo,O.O.(2011).Pattern of trematode infection in gall bladder from cattle slaughtered at Isheri-Olofin, Ogun-State, Nigeria. *The Zoologist*, **9**:25-30.

Hill,D.W. and Onabamiro,S.D.(1961).Sited in Enyenihi, United Kingdom .*Bulletin of Animal Health and Production in Africa*, **23** (3):289.

Hillyer, G.V. (1988).Fascioliasis and fasciolopsiasis. In: *Laboratory diagnosis of infectious diseases: principles and practices*, Springer-Verlag New York Incorporation Pp856–862.

Hillyer, G.V. (1999). Immunodiagnosis of human and animal fasciolosis. In: *Fasciolosis*. Dalton, JP.(Ed).CAB International Publisher, Wallingford Oxon, United Kingdom.

Hillyer, G.V. (2005).*Fasciola* antigens as vaccines against fascioliasis and schistosomiasis. *Journal of Helminthology*, **79** (3): 241-247.

Hopkins, D.R.(1992).Homing in on helminths. *American Journal of Tropical Medicine and Hygiene*,**46**:626-634.

Ibarra, F., Vera, Y.and Quiroz, H. (2004).Determination of the effective dose of an experimental fasciolicide in naturally and experimentally infected cattle.*Veterinary Parasitology*, **120** (1–2): 65–74.

Idowu,O.A.,Olorode,N.,Idowu,A.B. and Sam-wabo,S.O.(2006). Fascioliasis: Prevalence, protein content and attitude of meat sellers to infected livers of slaughtered cattle in Abeokuta. *Nigerian Journal of Parasitology*,**28**:125-128**.**

Idris, H.S. and Madara, A.A. (2005). Vector competence and prevalence of *Fasciola gigantica* in cattle slaughtered in Gwagwalada abattoir, Abuja Nigeria. *Biological and Environmental Science Journal for the Tropics*, **1** (2): 48-52.

Itagaki, T., Ichinomiya, M., Fukuda, K., Fusyuku, S. and Carmona, C. (2011). Hybridization ecperiments indicate incomplete reproductive isolating mechanism between *Fasciola hepatica* and *Fasciola gigantica*. *International Journal of Parasitology*, 138(10): 1278-1284.

John, B.G.,Johan,H. and Peter J.W.(2005).Ethno-veterinary plant preparations as livestock dewormers, practises, popular beliefs, pitfalls and prospects for the future. International Livestock Research Institute (ILRI), Nairobi, Kenya.

Keiser, J. and Utzinger, J. (2005). Emerging foodborne trematodiasis. *Emerging Infectious Diseases*, **11**:1507-1514.

Keiser, J., and Utzinger, J. (2009).Food-borne trematodiasis. *Clinical Microbiology Review*, **22**:466-483.

Keyyu, J.U., Monrad, J., Kyvsgaard, N.C and Kassuku, A.A. (2005). Epidemiology of *Fasciola gigantica* and a amphistomes in cattle on traditional, small-scale dairy and large-scale dairyfarmsin the southern highlands of Tanzania. *Tropical Animal Health and Production*, **37**: 303-314.

Koc,Z.,Ulusan,S. andTokmalc.N. (2009).Hepatobiliary fascioliasis: Imaging characteristics with a new finding. *Diagnostic Radiologist***15**: 247-251.

Kosgey, I.S., Rowlands, G.J., van Arendonk, J.A.M. and Baker, R.L. (2008).Small ruminant production in smallholder and pastoral extensive farming systems in Kenya. *Small Ruminant Research,* **77**: 11-24.

Magaji, A.A., Ibrahim, K., Salihu, M.D., Saulawa, M.A., Mohammed, A.A. and Musawa, A.I.(2014). Prevalence of fascioliasis in cattle slaughtered in Sokoto Metropolitan Abattoir, Sokoto, Nigeria. *Advances in Epidemiology*: 1-5.

Mahanty, S., Maclean, J.D and Cross, J.H. (2006).Liver, lung and intestinal fluke infections. In: *Tropical Infectious Diseases: Principles, Pathogens and Practice, 3rd ed.*

Marcos,L.A., Terashima, A. and Gotuzzo,E. (2008).Update on hepatobiliary flukes:Fascioliasis opisthorchiasis and clonorchiasis. *Current Opinion Infectious Disease* **21**:523

Mas-Coma, S. (2005). Epidemiology of fascioliasis in humans endemic areas.*Journal of Helminthology*,**79** (3):207-216.

Mas-Coma, S. and Bargues, M.D.(1997).Human liver fluke:A review. *Research inParasitology*, **57**:144-225.

Mas-Coma, S., Bargues, M.D. and Esteban, J.G. (1999). Human fasciolosis. In: *Fasciolosis*. Dalton J.P (Ed.)CAB International Publisher, Wallingford, Oxon, United Kingdom, 411–434.

Mas-Coma, S., Bargues, M.D. and Valero,M.A. (2005).Fascioliasis and other plant-borne trematode zoonoses. *International Journal of Parasitology,* **35** (12): 1255–1257.

Mas-Coma, S., Bargues, M.D and Valero, M.A. (2009).*Fasciola lymnaeids* and human fascioliasis with a global overview on disease, transmission, epidemiology, evolutionary genetics, molecular epidemiology and control.*Advanced Parasitology*, 69:141-146.

Mas-Coma, S., Esteban, J.G. and Bargues, M.D. (1999).Epidemiology of human fascioliasis.A review and proposed new classification,**77**:340-346.

Mas-Coma, S., Rodriguez, A., Bargues, M.D.,Valero, M.A.,Coello, J. and Angles, R. (1998).Secondary reservoir role of domestic animals other than sheep and cattle in fascioliasis transmission on the northern Bolivian Altiplanz. *Research and Review Parasitology*, 57: 39–46.

Mason,C. (2004). Fasciolosis associated with metabolic disease in a dairy herd and its effects on health and productivity. *Cattle Practice,* **12** (1): 7–13.

Mbaya, A.W., Shingu, P. and Luka, J. (2010). A retrospective study on the prevalence of *Fasciola* infection in sheep and goats at slaughter and associated economic losses fromcondemnation of infected liver in Maiduguri abattoir, Nigeria. *Nigerian Veterinary Journal,* **31** (3): 174-178.

Menard, A., Agoulon, A., L'Hostis, M., Rondelaud, D.,Collard, S. and Chauvin, A. (2001). Myocastor coypus as a reserviour host of *Fasciola hepatica* in France.*Veterinary Research,* **32** (5):499-508.

Mistrel,S.C.,Das,D.N.,Patnik, K.C. and Mohapatra,H.C.(1972).Incidence of gastro-intestinal parasites and technique for the recovery of their ova or cysts in the faeces.*Indian Veterinary Journal,***49**:140-145.

Mitchell, G.B., Maris, L. and Bonniwell, M.A. (1998).Triclabendazole-resistant liver fluke in Scottish sheep. *Veterinary. Records,***143** (14): 399-401

Molina, E.C., Gonzaga, E.A. and Lumbao, L.A. (2005). Prevalence of infection with *Fasciola gigantica* and its relationship to carcass and liver weights, and fluke and egg counts inslaughter cattle and buffaloes in southern, Philippines. *Tropical Animal Healthand Production,* **37**(3): 215-221.

Moll, L., Gaasenbeek, C.P. Vellema, P. and Borgsteede, F.H. (2000). Resistance of *Fasciola hepatica* against triclabendazole in cattle and sheep in Netherlands. *Veterinary. Parasitology,***91** (1–2): 153–158.

Muller,R.,(1975).*Worms and Disease: A Manual of Medical Helminthology*: Heinemann Medical Books, London.

Naings, L.,Winn, T. and and Rusli, B.N.(2006).*Sample size calculations for prevalence studies*. Available at http://www.kck.usm.my/ppsg/stats resources.html

Ndarathi,C.M.,Waghela,S.and Semenye,P.P.(1987). Helminthiasis in Massa Ranches in Kenya. *Bulletin of Animal Health and Production in Africa,* **37**:205-208.

Njoku,R.F.(2007). Ecological studies on some human and animal trematodes in some parts of Imo State, Nigeria, Ph.D thesis, Imo State University, Owerri, Nigeria.

Njoku,R.F. (2011). Bovine fascioliasis among cattle slaughtered in some abattoir in Imo state Nigeria. *World Rural Observations*,**3**(1): 82-86.

Nuhu, S. (1988). Research and development in pastoral production systems in Nigeria: Past, present and future. In proceedings of a National Conference on Pastoralism in Nigeria, Kaduna.

Obadiah, S.E.(2010). Preliminary studies on fascioliasis in cattle slaughtered at Jalingo abattoir, Tarabe state Nigeria. *Nigerian Journal of Science, Technology and Environmental Education*, **3** (1): 143-146.

O'Brien, D.J. (1998). Fasciolosis: a threat to livestock. *Irish Veterinary Journal*, **51**:539–541.

Ogunrinade, F.A. and Ogunrinade,B.I (1980). Economic importance of bovine fascioliasis in Nigeria. *Tropical Animal Health and Production*, **12** (3): 155–160.

Okoli,I.C.,Agoh, E.C.,Okoli,G.C.,Idemili,G.C. and Umeisiobi,D.O.(2000).Bovine and caprine fascioliasis in Enugu State, Nigeria. *Bulletin of Animal Health and Production in Africa*,**48**:7-11.

Olusegun- Joseph, T.S., Doherty, V.F. and Edeghagha,B.O.(2011).Prevalence of fascioliasis in cattle slaughtered at Zaria abattoir. *Journal of Environmental Issues*, **1**: 50-56.

Omowaye, S.O., Idachaba, O.S and Falola, O.O.(2012). The prevalence of parasitic infections in bile of cattle slaughtered in Jos abattoir, Plateau State Nigeria. *Global Journal of Bio-Science and Biotecnology,* **1** (1): 121-123.

Onyeabor, A and Wosu, M. (2014). Prevalence of Bovine Fascioliasis observed in three major abattoirs in Abia state, Nigeria. *Journal of Veterinary Advances*, **4**(11): 752-755.

O'Neill, S.M., Parkinson, M., Strauss,W., Angles, R.and Dalton, J.P. (1998). Immunodiagnosis of *Fasciola hepatica* infections in human population in the Bolivan Altiplano using purified cathepsin L.cysteine proteinase. *American Journal of Tropical, Medicine and Hygiene*,**58** (4): 417–423.

Oyeduntan, A.A., Adekunle, B.A. and Fagbemi, O.B.(2008).Seasonal prevalence of *Fasciola gigantica:* Infection among the sexes of Nigerian cattle. *Veterinary Research*,**2**(1):12-14.

Phiri, I.K., Phiri, A.M. and Harrison, L.J. (2006).Serum antibody isotype responses of *Fasciola*-infected sheep and cattle to excretory and secretory products of *Fasciola* species. *Veterinary Parasitology*, **141** (3–4): 234–242.

Price, T.A., Tuazon, C.U. and Simon, G.L. (1993). Fascioliasis: case reports and review. *Clinical Infectious Disease,* **17** (3): 426–430.

Raji, M.I.O., Ibrahim, Y.K.E. and Ehinmidu, J.O. (2010).Physico-chemical characteristics and heavy metals levels in drinking water sources in Sokoto metropolis in Northwestern Nigeria. *Journal of Applied Science and EnvironmentalManagement*.**14** (3): 81-85.

Roberts, J.A., Estuningsih, E., Wiedosari, E. and Spithill, T.W. (1997).Acquisition of resistance against *Fasciola gigantica* by Indonesian thin tail sheep.*Veterinary Parasitology,***73** (3–4): 215–224.

Rossignol, J.F., Abaza, H. and Friedman, H. (1998).Successful treatment of human fascioliasis with nitazoxanide. *Transactions of the Royal Society of Tropical Medicine and Hygiene,* **92** (1): 103–104.

Savioli, L., Chitsulo, L. and Montresor, A. (1999).New opportunities for the control of fascioliasis. *Bulletin of the World Health Organization,***77** (4): 300-

Schillhorn Vanveen, T.W., Folaranmi, D.B.S., Usman, S. and Ishaya, T. (1980).Incidence of liver fluke infections (*Fasciola gigantica* and *Dicrocoelium hospes*) in ruminants in Northern Nigeria. *Tropical AnimalHealthand Production,* **12**:97-104.

Sinclair, K.B. (1962). Observations on the clinical pathology of ovine fascioliasis.*British Veterinary Journal,* **118**: 37–53.

Soliman, M. F. M. (2008). Epidemiological review of human and animal fascioliasis in Egypt.*The Journal of Infection in Developing Countries***2**(3): 182-189.

Soulsby, E. J. L. (1982). *Helminths, arthropods and protozoa of domesticated animals*. (7th edition) Bailler Tindall, London,809pp.

Spithill, T.W., Smooker, P.M. and Copeman, D.B. (1999).*Fasciola gigantica*: epidemiology, control, immunology and molecular biology. In: *Fasciolosis*. Dalton, J.P (Ed.) Wallingford, Oxon, UK: CAB International Publisher, 465–525.

Taira, N., Yoshifuji, H., Boray, J.C. (1997). Zoonotic potential of infection with *Fasciola spp* by consumption of freshly prepared raw liver containing immature flukes. *International Journal Parasitology,***27**(7): 775-779.

Tasawar,Z.,Minir,U., Hayat, C.S. and Lashari,M.H.(2007).The prevalence of *Fasciola hepatica* in goats around Multan.*Pakistan Veterinary Journal*, **27**(1):5-7.

Tolan, R.W.Jr. (2011). Fascioliasis due to *Fasciola hepatica* and *Fasciola gigantica* infection: an update on this neglected tropical disease. *LaboratoryMedicine,***42** (2):107-117.

Ulayi,B.M.,Umaru-Sule,I.B. and Adamu,S.(2007). Prevalence of *Dicrocoelium hospes* and *Fasciola gigantica* infections in cattle slaughtered in Zaria Nigeria. *Journal of Animal Veterinary Advances,***6**(9): 1112-1115.

Umunna, M.O., Olafadehan, O.A. and Arowona, A. (2014).Small ruminant production and management systems in urban area of southern Guinea savanna of Nigeria. *Journal of Agriculture and Food Science,* **2**(2):107-114.

Usip,L.P.E, Ibanga, E.S, Edoho,H.J, Amadi,E.C and Utah,E.(2014). Prevalence of Fascioliasis and the economic loss of condemned liver due to *Fasciola* infection in cattle slaughtered at three abattoirs in Eket Urban, Akwa Ibom State of Nigeria. *Global Advanced Research Journal of Food Science and Technology*,**3**(2): 54–75

WHO.(1999).Report of WHO information consultation of schistosomiasis control.Geneva,December1998 WHO/CDS/SIP/99.2.

WHO. (2008).Control of Neglected Tropical Disease(NTD).*Issue 10:fasciola@who International*,**6**(9):

WHO. (2012). Global burden of human foodborne trematodiasis: A systematic review and meta-analysis. *The Lancet Infectious Disease*, 12(3): 210-221.

Yilmaz, H. and Gödekmerdan, A. (2004).Human fasciolosis in Van Province, Turkey. *Acta Tropica*,**92**(2): 161–162.

Youn, H.(2009). Review of zoonotic parasites in medical and veterinary fields in the Republic of Korea. *Korean Journal of Parasitology*, **47**(1):133-141.