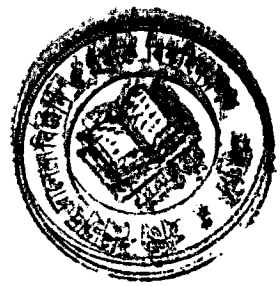
COMPARATIVE EFFICACY OF PIPERAZINE CITRATE AND PINEAPPLE *(Ananus comosus)* LEAVES EXTRACT ON ASCARIASIS IN INDIGENOUS CHICKENS IN PALASHBARI UPAZILLA OF GAIBANDHA DISTRICT

A Thesis

By

Mst. Kamrunnaher Akter
Registration No. 1105123
Semester: July/2012 – December/2012



MASTER OF SCIENCE (M.S.)

IN

PHARMACOLOGY

DEPARTMENT OF PHYSIOLOGY AND PHARMACOLOGY
HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY
UNIVERSITY, DINAJPUR, BANGLADESH

COMPARATIVE EFFICACY OF PIPERAZINE CITRATE AND PINEAPPLE *(Ananus comosus)* LEAVES EXTRACT ON ASCARIASIS IN INDIGENOUS CHICKENS IN PALASHBARI UPAZILLA OF GAIBANDHA DISTRICT

A Thesis

By

Mst. Kamrunnaher Akter
Registration No. 1105123
Semester: July/2012 – December/2012

Submitted to the
Department of Physiology and Pharmacology
Hajee Mohammad Danesh Science and Technology University, Dinajpur
in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE (M.S.)

IN

PHARMACOLOGY

DEPARTMENT OF PHYSIOLOGY AND PHARMACOLOGY
HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY UNIVERSITY, DINAJPUR-5200

DECEMBER, 2012

COMPARATIVE EFFICACY OF PIPERAZINE CITRATE AND PINEAPPLE *(Ananus comosus)* LEAVES EXTRACT ON ASCARIASIS IN INDIGENOUS CHICKENS IN PALASHBARI UPAZILLA OF GAIBANDHA DISTRICT

A Thesis

By

MST. KAMRUNNAHER AKTER
Registration No. 1105123
Semester: July/2012- December/2012

Approved as to style and content by

(Dr. Fahima Binthe Aziz)
Supervisor

(Dr. Md. Nazrul Islam)
Co-supervisor

Dr. Md. Bazlar Rashid
Chairman, Examination Committee
And
Chairman
Department of Physiology and Pharmacology
Hajee Mohammad Danesh Science and Technology University, Dinajpur-5200

DECEMBER, 2012

Dedicated
To
My Beloved Parents
and
Husband

ACKNOWLEDGEMENT

At first all praises and deepest sense of gratitude are due to Almighty ALLAH, the supreme creator of this universe, who enabled the author to complete this thesis.

The author sincerely desires to express her heart felt respect, deepest sense of gratitude, sincere appreciation and profound indebtedness to her respected teacher and Research Supervisor, ***Dr. Fahima Binthe Aziz,*** *Assistant professor , Department of Physiology and Pharmacology, Faculty of Veterinary and Animal Science (FVAS), Hajee Mohammad Danesh Science and Technology University (HSTU), Dinajpur for her constant supervision, scholastic guidance, uncompromising principles, helpful advice, constant inspiration, affectionate feeling, radical investigation and constructive criticism in all phases of this study and preparing the manuscript.*

The author finds it a great pleasure in expressing her heartfelt gratitude and immense indebtedness to her honorable and respected Research Co-Supervisor, ***Dr. Md. Nazrul Islam,*** *Assistant professor, Department of Pathology and Parasitology, Faculty of Veterinary and Animal Science (FVAS), Hajee Mohammad Danesh Science and Technology University (HSTU), Dinajpur for his sympathetic supervision, kind co-operation, inspiration and valuable suggestion, constructive criticism and valuable advice for the completion of the research work and preparation of the thesis.*

The author also expresses her deepest sense of gratitude to her respected teacher, ***Dr. Md. Bazlar Rashid,*** *Assistant professor and Chairman, Department of Physiology and Pharmacology and* ***Dr. Md. Mahmudul Hasan,*** *Lecturer, Department of Physiology and Pharmacology, Hajee Mohammad Danesh Science and Technology University, Dinajpur, for their encouragement and cordial co-operation throughout the course of the study.*

The author takes this opportunity in expressing her heartfelt thanks to Dr. Md. Anisur Rahman, Dr. Md. Rashed Kamal, Dr. Md. Khalesur Rahman, Dr. Parbati Paul, Dr. Umme Kulsum and Dr. Ayatullah for their help, constant inspiration and encouragement.

The author is also thankful to laboratory technician & all stuff of the Department of Physiology and Pharmacology, Hajee Mohammad Danesh Science and Technology University, Dinajpur, for their technical assistant during the experiment.

The author expresses heartfelt gratitude and gratefulness to her beloved parents, brothers, sisters, friends and well wishers for their blessing, inspiration, sacrifice and moral support which opened the gate and paved the way to her higher studies and also provided her with the best of everything in her life.

Finally, the author falls short of vocabulary in expressing her ever gratefulness to her husband Prodhan Mahbub Ibna Seraj for his sacrifice, love, patience, overwhelming support and inspiration.

The author

December, 2012

ABSTRACT

An experiment was carried out in the laboratory of the department of Physiology and Pharmacology, Hajee Mohammad Danesh Science and Technology University, Dinajpur and Upazila Livestock Office, Palashbari, Gaibandha, during the period from July to November/2012. 45 chickens were randomly divided into 3 groups (A, B and C). Group A was kept for control, Group B was treated with Piperazine citrate (Pipervet®) and Group C was treated with the aqueous extract of Pineapple leaves. These two were screened for assessment their comparative efficacy against ascariasis in indigenous chickens. Pineapple leaves was used at a dose of 1gm/kg bwt for seven consecutive days and piperazine citrate at a dose of 200mg/kg bwt as a single dose. Efficacy was recorded as compared to control on the basis of fecal egg count. Piperazine citrate showed 100% efficacy within 14 days of treatment. On the other hand, Pineapple leaves showed 80% efficacy on 28th day of treatment. Post mortem worm count revealed that pineapple leaves were considerable effective against worm burden. Hematological analysis revealed that the aqueous extract of leaves did not show any adverse on blood parameter of treated chickens.

LIST OF CONTENTS

LIST OF CONTENTS (Contd.)

LIST OF TABLES

LIST OF FIGURES

LIST OF PICTURES

LIST OF ABBREVIATIONS

bwt	:	Body weight
FAO	:	Food and Agriculture Organization
Cu mm	:	Cubic millimeter
DLS	:	Department of Livestock Services
EPG	:	Egg Per Gram
ESR	:	Erythrocytes Sedimentation Rate
et al.	:	Associates
Fig.	:	Figure
gm	:	Gram
Hb	:	Hemoglobin
i.e.	:	That is
J.	:	Journal
Kg	:	Kilogram
lit	:	Litre
Ltd	:	Limited
mg	:	Milligram
ml	:	Milliliter
mm^3	:	Cubic millimeter
No	:	Number
PCV	:	Packed Cell Volume
SE	:	Standard Error
TEC	:	Total Erythrocyte Count
TLC	:	Total Leukocytes Count
µg	:	Microgram
%	:	Percent
&	:	And
@	:	At the rate of
<	:	Less than
>	:	Greater than
±	:	Plus minus

CHAPTER I

INTRODUCTION

CHAPTER I

INTRODUCTION

Poultry is a promising sector in Bangladesh which is increasing day by day. Poultry production is hindered by many problems among which various diseases namely bacterial, viral and parasitic infections are the most important (Ojok, 1993). In fact poultry of Bangladesh are parasitized by various parasites (Sarkar, 1976).The Agro-ecological and geo-climatic condition of Bangladesh favors the survival, multiplication, spread and perpetuation of animal parasites. Moreover, management and managerial system play an important role in high degree of occurrence of parasitic disease in Bangladesh. Both ecto and endo-parasites affect the poultry health. Islam and Shaikh (1967) reported that internal parasites are present in chickens of Bangladesh. There are different types of internal parasites such as roundworms (*Ascaridia spp*, *Heterokis spp*), tapeworms (*Railleitina spp*) etc. The parasites because of their adverse effects leads to lower productivity, retarted growth rate and death of animals (FAO, 1974: Barger, 1982: Steel and Symons, 1982: Sykes, 1982: Holmes, 1986: Sykes, 1994).

Among the parasites next to the coccidian, *Ascaridia galli* infection in chicken is considered to be of great importance and the rate of infection in Bangladesh varies from just over 30% to nearly 80%. The rate of infection is higher in male than female and in young than adults (Islam, 1966; Sarker, 1977; and Mondal and Qadir, 1991). It is an intestinal worm and chickens under three months of age are mostly susceptible. Both in rural and farm conditions ascarids infection is important in Bangladesh (Islam and Shaikh, 1967; Sarker, 1977; Haq, 1986). *Ascaridia galli* causes extensive economic losses in different ways such as loss of weight gain, meat production, egg production and death of birds (Kamal, 1989).

There are various types of anthelmintics which are imported from abroad and are expensive, having side effects and they are not equally active on all stages of lifecycle of parasites. The indiscriminate use of anthelmintics made the parasites to be resistant against the drug, which have been reported by experts throughout the world including Bangladesh (Hannan *et al.*, 2001). A good spectrum of effective anthelmintics is available in the market. Albendazole, fenbendazole, ivermectin, livamisole, piperazine etc are the widely used anthelmintics. Among them piperazine citrate is widely used for the treatment of ascariasis in chickens.

Piperazine was first introduced as an anthelmintic in 1953. A large number of piperazine compounds have anthelmintic action. Mode of action of piperazine is generally by paralysing parasites, which allows the host body to easily remove or expel the invading organism (The Merck index, 11th edition).

People of Bangladesh who rare indigenous poultry by scavenging system are mainly farmers. They live under poverty line. They have no ability to buy modern medicine for the treatment of the livestock because of high price of the drug and their poor economic condition. As a result their livestock and poultry die from any disease. Furthermore, frequent use of these anthelmintics increased the resistant population of nematodes (Waller *et al.*, 1987). So if we become able to develop the traditional system of medicine in Bangladesh, it will be highly beneficial for the farmers and for the overall improvement of the livestock sector.

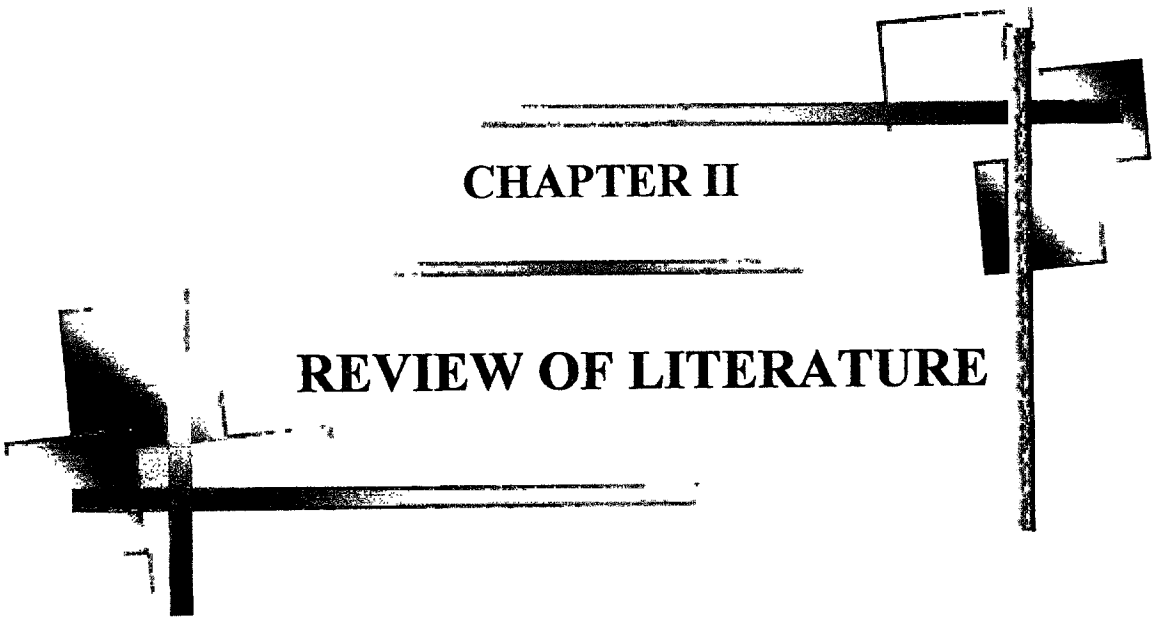
There are several indigenous medicinal plants (Nath *et al.*, 1983; Mostofa, 1983) have anthelmintics action and used against both ecto and enodparasites in Bangladesh (Mostofa, 1983; Mannan *et al.*, 1997) and India (Dutta and Hazarika, 1976). Pharmacological actions along with therapeutic trial of these plants may be studied experimentally, which might prove worthy of medical

value. Medicinal plants are being used traditionally in this country as folk medicine. World Health Organization (WHO, 1993) has recognized the necessity for investigation and mobilization of ancient medicinal practice to fulfill the primary health care of the animals and realises that the traditional system of medicine may play an important role in the development of livestock of the third world countries.

Neem, tobacco plant, barbados lilac, betel leaf, pineapple, jute, turmeric, garlic, devil's tree, papaya are the important medicinal plants. Pineapple leave extract is used as anti-inflammatory, anti bacterial and also as anthelmintic agent (Mostofa, 1983 and Amin *et al.*, 2009). Rural people more prefer pineapple as an anthelmentic (Mostofa, 1983; Mannan *et al.*, 1997; Sidvas, 1980 and Nath *et al.*, 1983). Therefore, if we use indigenous medicinal plant like pineapple leave as anthelmintic instead of imported drugs the country will be benefited and can save her hardly earned foreign currency.

Considering the above facts the present study was undertaken with the following objectives:

- To compare the efficacy of Pineapple leaves and Piperazine citrate on ascariasis in indigenous chickens.
- To compare the control of ascariasis in indigenous chickens by the application of Pineapple leaves and Piperazine citrate.



CHAPTER II

REVIEW OF LITERATURE

CHAPTER II

REVIEW OF LITERATURE

Many research works have been carried out by numerous researchers to evaluate the efficacy of Piperazine citrate and Pineapple leaves as anthelmintics in chickens. Attempts have been made to reflect some of these works, which are directly related with present study.

Morphology of the parasites

One of the most common parasitic disease of chickens is ascariasis which is mainly caused by a roundworm *Ascaridia galli*. Adult male worms are about 5 to 7.6 cm and females are about 11.6 cm long. Thus they can be easily seen with naked eye. Heavily infected birds may show droopiness, emaciation and diarrhea. The primary damage is reduced efficiency of feed utilization, but death has been observed in severe infections. (Rahman *et al*, 1996)

Taxonomic position of the parasites

Kingdom: Animalia

Phylum: Nematoda

Class: Secernentea

Subclass: Spiruria

Order: Ascaridida

Family: Ascarididae

Genus : *Ascaridia*

Species : *Ascaridia galli*

Sources and transmission of the parasites

Transmission of the worms involves the adult parasitic worm laying eggs, possibly as many as hundreds of thousands a day, that are then passed out of the infected bird in its faeces. This faeces is then eaten by another bird allowing the eggs to enter the diner. Inside the new host bird the larvae, which are actively infective after a couple of weeks, hatch and then attach themselves to the mucosa of the large intestine. Resistance to the parasites may appear by three months old though.

The eggs of the ascarid worm, complete with the embryonic larvae inside, are very robust indeed. They can survive in the environment for a couple of years and they are not destroyed by disinfectant. This means that prevention of the spread of the disease focuses on killing the adult worms. This can be achieved by thoroughly sterilising an area before a new flock of birds is put into it.

Life cycle of *Ascaridia galli*

Egg passed in the feces develop to the infective stage containing second stage larvae in about 8-10 days under optimal conditions of temperature and moisture. These infective eggs are fairly resistant to adverse conditions but is rapidly killed by heat and desiccation. In moist cool conditions they may remain viable for several months (7 months). Infection of final host occurs through ingestion of eggs with food or water, earth worms may ingest eggs and act as mechanical/transport host. The eggs hatch in the duodenum liberating second stage larvae; remain in the duodenum for about 8 days and then enter the mucosa of the duodenum. Larvae either in lumen or mucosa moult to third-stage larvae by 8 days. Moulting to fourth-satge occurs on the 14-15th day, for the lumen dwelling one's, in mucosal forms moulting occurs irregularly, second moult may be delayed even after 17th day after infection.

After 18th day lumen dwelling one's grow rapidly and reach to maturity an lay eggs by 5-6 weeks after infection. Prepatent period may be longer in mucosa dwelling one's or in birds above 3 months of age. Occasionally larvae may be found in erratic sites such as the liver and lungs and even adults may be found in hen's egg (Rahman, 1996).

2.1 Ascariasis in poultry

Matur *et al.* (2010) examined gastrointestinal tracts of 500 local and exotic breeds of chickens collected from the Gwagwalada Market, Nigeria for helminth parasites. Of these parasites, *Ascaridia galli* was found to be the most prevalent (51.60%) among the chickens. They also found that Females harbored more parasites than males.

Hassouni and Belghyti (2006) examined gastrointestinal tracts of 300 chickens from three villages of the Gharb region, Morocco, for adult helminths during 2002-2005. Helminth species found were: *Notocotylus gallinarum* (prevalence 0.7%), *Hymenolepis carioca* (3.7%), *Raillietina echinobothrida* (5.7%), *Hymenolepis contaniana* (7%), *Raillietina tretragona* (9.3%), *Raillietina cesticillus* (12%), *Capillaria obsignata* (6%), *Subulura brumpti* (15.3%), *Heterakis gallinarum* (10%), *Cheilospirura hamulosa* (2.7%), *Dispharynx nasuta* (5.3%), *Ascaridia galli* (9%) and *Tetrameres sp.* (3.3%). The prevalence and mean intensity of helminth infections did not differ significantly between male and female chickens.

Rabbi *et al.* (2006) studied gastrointestinal helminths infection in different types of poultry. During routine examination, total six species of helminth parasites were recorded, of which three species were nematodes such as *Ascaridia galli, Heterakis gallinarum* and *Capillaria annulata*; two species were cestodes such as *Raillietina tetragona* and *Amoebotaenia sphenoides*.

Prevalence of different species of gastrointestinal helminths was highest in backyard poultry (100%) followed by layer (48.75%) and broiler (3.75%). prevalence of the recovered parasites from backyard poultry were observed such as the prevalence of *R. tetragona* (100%) was the highest followed by that of *A. galli* (87.50%) and *H. gallinarum* (80%).

Martin *et al.* (2005) proved that *Ascaridia galli* is a common nematode found in the intestine of domesticated chickens. The objectives of the study were to conduct a coprological and serological survey on the prevalence of ascaridiosis in laying hens of commercial farms. Serological tests revealed a seroprevalence of 21.8% (range 7.6-95%). No positive serum samples were detected in the same farms with previous negative coprological analysis. Western blot analyses confirmed the results obtained by the enzyme-linked immunosorbent assay (ELISA) tests.

Kilpinen *et al.* (2005) showed that the effect of infections with *Ascaridia galli* (roundworm) on the behavior and health of laying hens . six groups of 15 pullets (Isa brown) were kept in indoor pens from 18 weeks of age *A.galli* resulted in lower weight gain but no significant changes were seen in blood variables or behavioural activities.

Gauly *et al.* (2005) examined ninety white chickens (Lohmann LSL) reared ender helminth-free conditions and divided into five groups. Four groups were artificially infected with 250 embryonated *Ascaridia galli* eggs at the age of 6, 12, 18, or 24 weeks. Ten birds were kept as uninfected controls. Six and 10 weeks after infection (p.i.), individual fecal egg counts (FEC) were performed. The birds were slaughtered after the second sampling and their gastrointestinal tracts were examined for the presence of adult *A. galli*. The FEC increased from the first to second sampling significantly in all the infected groups. The highest increase was shown in the group infected at 12

weeks of age, whereas the increase in the other groups was relatively moderate.

Irungu *et al.* (2004) studied on 456 indigenous chicken intestinal specimens from various towns in Kenya to determine the occurrence and distribution of helminth parasites in the intestinal tract of the birds. Of the specimens examined , 414 had parasites whereas the remaining 42 had none, which is an infection rate of 90.78%. The main species of helminthes found in the intestines were *Raillietina sp.* (47.53%), *Heterakis gallinarum*(21.33%), *Ascaridia galli* (10.03%), *Strongyloides avium* (9.96%), *Choanotaenia infundibulum* (4.61%), *Cotugnia digonopora* (3.6%), *Capillaria sp.* (1.5%) and *Syngamus trachea*(0.40%).

Idi *et al.* (2004) conducted two experiments to compare the effect of chickens age on resistance to primary and secondary infections with *Ascaridia galli.* The results from these experiments demonstrate that the chickens age only partially influences resistance to *A. galli* infection.

Permin *et al.* (2002) determined the prevalence of ecto-, endo- and haemoparasites in free-range chickens from the Goromonzi District, Zimbabwe. Fifty young and 50 adult birds were selected randomly. All chickens harboured ecto- and endoparasites, and 32% were infected with heamoparasites. Eight different ectoparasites were identified; the more prevalent ones had the following prevalences (young, %; adult, %) : *argas persicus* (6; 14), *Cnemidocoptes mutans* (6; 32), *Echidnophaga gallinacean* (72; 74), *Goniocotes gallinae* (0; 22), *Allodapa suctoria* (76; 72), *Ascaridia galli* (48; 24), *Gongylonema ingluvicola* (28; 56), *Heterakis gallinarum* (64; 62) and *Tetrameres Americana* (70; 62).

Eshetu *et al.* (2001) examined a total of 267 rural scavenging chickens from October 1998 to August 1999 in four woredas (districts) of the Amhara Region, Ethiopia. Of these chickens, 243 (91.01%) were found to harbour one to nine different helminth parasites and 24 (8.99%) were free of helminth parasites. A significant difference ($P < 0.01$) was found between the prevalence rates of helminth parasites in the different agro-ecological zones; the highest prevalence was observed in the lowland areas. This suggests that agro-ecology has a major influence on the distribution of helminth parasites. Nematodes recovered included *Heterakis gallinarum* (17.28%), *Subulura brumpti* (17.60%), *Ascaridia galli* (35.58%), *Cheilospirura hamulosa* (0.75%) and *Dyspharynx spiralis* (2.62%). The principal cestode species encountered were *Raillietina echinobothrida* (25.84%), *Raillietina tetragona* (45.69%), *Raillietina cesticillus* (5.62%), *Amoebotaenia sphenoides* (40.45%), *Davainea proglottina* (1.12%) and *Choanotaenia infundibulum* (4.49%).

Poulsen *et al.* (2000) conducted a cross-sectional study to determine the prevalence and species of gastro-intestinal helminths and haemoparasites in 100 chickens kept under extensive management systems in Ghana, West Africa. All the examined chickens (100%) were infected with gastro-intestinal helminths; a total of 18 species were detected. The species and their prevalences were: *Acuaria hamulosa* (25%), *Allodapa suctoria* (20%), *Ascaridia galli* (24%), *Capillaria spp.* (60%), *Choanotaenia infundibulum* (13%), *Gongylonema ingluvicola* (62%), *Heterakis gallinarum* (31%), *H. isolonche* (16%), *Hymenolepis spp.* (66%), *Raillietina cesticillus* (12%), *R. echinobothrida* (81%), *R. tetragona* (59%), *Strongyloides avium* (2%), *Subulura strongylina* (10%), *Tetrameres fissispina* (58%), *Trichostronygylus tenuis* (2%), and finally one unidentified acanthocephalan (1%) and one unidentified trematode (1%).

Mushi *et al.* (2000) examined thirteen adult indigenous chickens from Oodi, Kgatleng district, Botswana for helminth parasites. Two species of nematodes, *Ascaridia galli* and *Heterakis gallinarum*, and species of the cestode genus *Raillietina*, were recovered. *A. galli* and *H. gallinarum* were the most commonly seen parasites. The nematode *A. galli* occurred concurrently with *Raillietina spp.*

Permin *et al.* (1999) conducted a cross-sectional prevalence study of gastrointestinal helminths in Danish poultry production systems on 268 adult chickens selected at random from 16 farms in Denmark from October 1994 to October 1995. The trachea and the gastrointestinal tract of each bird was examined for the presence of helminths. In the free-range/organic systems the following helminths were found: *Ascaridia galli* (63.8%), *Heterakis gallinarum* (72.5%), *Capillaria obsignata* (53.6%), *Capillaria anatis* (31.9%) and *Capillaria caudinflata* (1.5%). In the deep-litter systems: *A. galli* (41.9%), *H. gallinarum* (19.4%) and *C. obsignata* (51.6%). In the battery cages: *A. galli* (5%) and *Raillietina cesticillus* or *Choanotaenia infundibulum* (3.3%). In the broiler/parent system: *C. obsignata* (1.6%), and finally for the backyard system: *A. galli* (37.5%), *H. gallinarum* (68.8%), *C. obsignata* (50.0%), *C. anatis* (56.3%) and *C. caudinflata* (6.3%).

Hemalatha *et al.* (1987) studied the helminthic infection in domestic fowls reared on deep litter and cage system and found that *Ascaridia galli* infection was 37% reared under a case system and 40% on deep litter.

Steiner and Davis (1981) mentioned that good hygiene and sanitation are the prerequisite for prevention and control of ascariasis in chickens and other birds.

Haider *et al.* (1980) conducted a survey on helminth parasites of broiler chickens in and around Lahore, Pakistan. The authors reported that the most common nematodes were *Ascaridia galli, Heterakis gallinae* and *Subulura brunpti.*

Velichkin *et al.* (1976) found that the typical incidence of *Ascaridia spp* and their incidence was recorded from 40% to 80% with the intensity up to 10-12 parasites per bird and *Heterakis spp* was 46-85% with 20 parasites per bird for birds in the field house without runs, while the figure was only 10-15% for both parasites for birds in cages.

Bose (1972) studied the incidence of *Ascaridia galli* in hen's egg in osria, India.

Busa and Hernanez (1970) described the ecology, biology and epidemiology of helminth of domestic fowls in cuba with *Ascaridia galli* infection and recorded highest infection in chickens of litter than caged.

2.2 Prevalence of Ascariasis in Chickens throughout the world

Hussen *et al.* (2012) conducted a cross-sectional survey on gastrointestin al helminthes on 124 chickens raised under traditional management system in two selected districts namely Ada'a and Adamitulu of Eastern Shewa zone, Ethiopia. Of these chickens, 111 (89.5%) were found to harbor one of the five different helminth parasites and 13 (10.48%) were free of helminths parasites. The major nematode species encountered were *Heterakis gallinarum* 47 (37.9%), *Ascaridia galli* 40 (32.0%), *Gongylonema ingluvicola* 32 (25.8%), *Dispharynx nasuta* 5 (4.0%), *Heterakis isolonche* 11 (8.9%), *Allodapa suctoria* 9 (7.3%), *Capillaria anatis* 4 (3.2%) and *Heterakis dispar* 8 (6.5%). The major cestode species recovered from chickens were *Raillietina echinobothrida* 79 (63.7%), *Raillietina tetragona* 70 (56.5%), *Raillietina*

cesticillus 50 (40.3%) and *Choanotaenia infundibulum* 17 (13.7%), *Davainea proglottina* 10 (8.1%), *Hymenolepis contaniana* 22 (17.7%) and *Hymenolepis carioca* 7 (17.7%).

Rayyan *et al.* (2010) examined Gastrointestinal tracts of 100 commercial growers and 90 free-range indigenous chickens collected from Gaza city for helminthes during 2007–2008. The commercial chickens were free of helminthes while the indigenous chickens were encountered five helminthes, three nematodes and two cestodes .The nematodes identified were *Ascaridia galli* (75.6%), *Heterakis gallinarum* (68.9%) and *Capillaria spp.* (2.2%). The cestodes were *Raillietina echinobothrida* (57.8%) and *Choanotaenia infundibulum* (26.7%). No trematode was found.

Nnadi and George (2010) conducted a cross-sectional survey on parasites of chickens in selected villages in the Subhumid Zones of South-Eastern Nigeria. A total of 1038 chickens comprising of 468 chicks, 207 growers and 363 adults were examined during the house to house survey for ectoparasites, gastrointestinal helminths and coccidia infections. The finding showed that 41% were infected with ectoparasites with lice, fleas, and mites having prevalence rates of 62.2%, 35.7% and 2.1%, respectively. Helminths and coccidia had prevalence of 35.5% each. Among the helminthes *Ascaridia galli* was the most dominant species (17.2%). Generally, there was a significantly higher helminth infestation relative to the ectoparasites ($P < .05$), high prevalence of mixed infections and absence of tick infestation.

Mukaratirwa and Khumalo (2010) collected A total of 79 chickens from 4 rural localities and processed to detect the presence of helminth parasites and their prevalences. The most prevalent nematode species across the 4 localities were *Heterakis gallinarum* (prevalence range 80-94.4 %), *Gongylonema ingluvicola* (43.3-86.7 %), *Tetrameres americana* (53.3-66.7 %) and

Ascaridia galli (22.2-43.8 %) and for cestode species *Raillietina tetragona* (16.7-40 %) and *Skrijabinia cesticillus* (3.3-13.3 %) were the most prevalent in that order.

Kose *et al.* (2009) conducted a research on 200 broilers collected from 10 modern farms, 200 layer hens from different 10 modern farms and on 100 free ranging backyard chickens for determining the prevalence of the gastro-intestinal helminths in chicken. According to the necropsy findings, none of gastro-intestinal helminths was found in broiler chickens. In total 4 modern layer farms, *Choanotaenia infundibulum* was detected in 23 (11.5%) chickens out of 200. A total of 58 (58%) free-range backyard chickens were infected with various helminth species, which were: *Heterakis gallinarum* 38%, *Ascaridia galli* 19%, *C. obsignata* 15%, *Capillaria caudinflata* 13%, *Trichostrongylus tenuis* 3%, *Subulura differens* 2%, *Choanotaenia infundibulum* 14%, *Raillietina echinobothrida* 6%, *R. cesticillus* 3%. The predominantly seen helminth species was *H. gallinarum* (38%) and the rarely seen species was *S. differens* (2%). In total 9 species were detected, which 6 of them were nematodes and 3 of them were cestodes. In infected chickens, minimum one and maximum 3 helminth species were detected. No trematode species was detected in necropsied and fecal examinated chickens. A total of 81 (16.2%) chickens out of 500 were found infected with various helminth species in necropsy and fecal examination. The helminth infection rates and species numbers in modern farms obtained less than in free-ranging systems.

Pinckey *et al.* (2008) studied on prevalence of gastrointestinal parasites in Grenada, West Indies. This study was done to establish the species and frequency of gastrointestinal parasites in 106 free-range chickens aged between 1 to 2 years, and collected from 13 households. Three nematodes and one tapeworm were identified: *Gongylonema ingluvicola* (29.2%); *Ascaridia*

galli (10.3%); *Heterakis gallinarum* (4.7%); *Capillaria contorta* (2.83%); *Raillietina tetragona* (38.6%).

Phiri *et al.* (2007) examined gastrointestinal tracts of 125 free-range chickens in Zambia for helminthes revealed a 95.2% prevalence rate. The species and their prevalences were: *Allodapa suctoria* (85.6%), *Tetrameres americana* (80.8%), *Ascaridia galli* (28.8%), *Gonglonema ingluvicola* (50.4%), *Raillietina spp.* (81.6%) and *Heterakis gallinarum* (32.8%). No trematodes or *Syngamus trachea* were found. Mixed infections accounted for 88.2% as compared to 7.2% of single infections.

Romanenko *et al.* (1985) reported that at the age of 4-5 months 100% of chicken at two farms were infected with helminthes including *Ascaridia spp* 4%, *Heterakis spp.* 10%, *Capillaria spp* 33%. There was a wide variation on the prevalence and intensity of each infection for age group for 4-24months.

Samad and Rahman (1985) investigated the incidence of gastrointestinal parasitic infection in domestic fowls and recorded *Ascaridia galli* as 44.82%, *Heterakis gallinae* 58.28%, *Capillaria annulata* 8.24%, *Capillaria columbae* 16.38%, *Ascaridia spiralis* 16.28%, *Subulura brumpti* 1.91% and *Subulura avium* 1.11%.

Hayat and Hayat (1983) described intestinal parasites of chicken in Faisalabad district of Pakistan and recorded that out of 793 chicken, 22.9% were infected with *Ascaridia galli*, 11.9% with *Subulira brumpti* and 2.2% with *Heterakis gallinae*.

Kaushik and Decoranj (1968) examined 620 chickens in Uttar Pradesh, India. Helminthes were present upto 70% of which *Ascaridia galli* 41% and *Heterakis gallinae* 10.5%.

Durriani and chauhan (1965) surveyed the intestinal parasites of chicken in Lyallpur district, West Pakistan and found 79.5% infection. They recorded as 48.5% infection with tapeworm, 22.66% with ascarids and 37.36% with caecal namatodes.

2.3 Prevalence of Ascariasis in Chickens in Bangladesh

Sarker *et al.* (2009) studied on prevalence of ascariasis in village poultry at five upazilas under feni district. They found that Highest prevalence of Ascariasis was in Daganbhuiyan upazila (90.47%) and lowest was in Parshuram upazila (81.57%) and the infection rate on the basis of age was at the age of 2-4 months.

Rabbi *et al.* (2006) studied on gastrointestinal helminths infection in different types of poultry in Mymensingh. They found that prevalence of different species of gastrointestinal helminths was highest in backyard poultry (100%) followed by layer (48.75%) and broiler (3.75%) which was statistically significant ($p < 0.01$). Backyard poultry was significantly ($p < 0.05$) 168.21 and 4106.67 times more susceptible to helminth infection than layer and broiler respectively. But layer was 24.41 times more susceptible to helminth infection than broiler. In backyard poultry, all six species of helminth parasites were found. A statistically significant ($p < 0.05$) variation in the prevalence of the recovered parasites from backyard poultry were observed such as the prevalence of *R. tetragona* (100%) was the highest followed by that of *A. galli* (87.50%) and *H. gallinarum* (80%).

Akhtar (1987) examined 50 domestic fowls in Bangladesh and recorded the infection of *Strongyloides avium, Capillaria contorta, Capillaria anulata, Capillaria columbae, Heterakis gallinarum, Heterakis beramporia, Heterakis parisis* and *Ascaridia galli.*

Haq (1986) examined 400 domestic fowls in Bangladesh under rural condition and recorded *Ascaridia galli* as 45%, *Heterakis gallinae* 80%, *Capillaria annulata* 10%, and *Capillaria columbae* 15%.

Hassain (1967) examined 200 fowls of Comilla, East Pakistan, of which 187 were found to be infected with helminthes, *Raillietina tetragona* was common cestode and *Ascaridia galli* was the common nematode, one specimen of Acanthocephala, but trematode was not found.

Islam and Shaikh (1967) examined 400 domestic fowls in Mymensingh, East Pakistan and recorded *Heterakis gallinae* as 85%, *Ascaridia galli* 50%, *Capillaria annulata* 10%, *Capillaria columbae* 20%, *Acuaria spiralis* 12%, *Amorbotaneia sphenoides* 25%, *Hymenelepsis carioca* 40%, *Raillietina tetragona* 35%, and *Catatropis verrucosa* 15%.

2.4 Efficacy of piperazine citrate against ascariasis

Jacques *et al.* (2012) studied on comparative effects of piperazine citrate and papaya seeds on *Ascaridia galli* in layers harco. After the treatment (state 2), there was a difference highly significant between the treatments with an efficiency rate of -50% for the witness group; 80% for the treatment 1; -14% for the treatment 2; 4% for the treatment 3; 16% for the treatment 4 and 65% for the treatment 5. Papaya seeds have reduced significantly the number of eggs per gram (EPG) comparing to the witness group. But that reduction remains lower than the one of citrate of piperazine (2.5g/L of the solution) in *Ascaridia galli*'s eggs reduction. In water treatment of chickens infected by *Ascaridia galli*, Piperazine citrate is recommended; but the papaya seeds can play also an important role, reducing the eggs of this worm in poultry's feaces.

Begum *et al.* (2010) conducted a research on comparative efficacy of leaves extract of neem and bishkatali with patent drugs piperazine and levamisole against ascariasis of indigenous chicken. Authors found that neem leaves extract have anthelmintic activity and could be use in poultry ascariasis. Bishkatali have mild anthelmintic action but showed toxic sign.

Hafiz and Bhattacharyya (2009) examined about comparative efficacy of piperazine and ivermectin against ascariasis in chickens. They found that the both drugs were effective against ascariasis in chicken.

Hoque *et al.* (2006) conducted a study on comparative efficacy of piperazine citrate, levamisole and pineapple leaves extract against naturally infected ascariasis in indigenous chickens. They observed that pineapple leaves extract showed less effectiveness in reducing parasite count in postmortem examination than piperazine citrate and levamisole but its use may be encouraging because of less adverse effects. In all treated groups total erythrocyte count (TEC), hemoglobin estimation (Hb) and packed cell volume (PCV) significantly ($p < 0.01$) increased and erythrocyte sedimentation rate (ESR) and total leukocyte count (TLC) significantly ($p < 0.01$) decreased. Body weight was also found to be increased following administration of piperazine citrate, levamisole and pineapple leaves extract.

Duong *et al.* (1997) reported high effectivity of piperazine against *Toxacara vitulorum* in calves and young buffaloes.

Maqbool *et al.* (1995) studied the incidence and chemotherapy of *Ascaridia galli* infection in chicken. They made trials with mebendazole and piperazine. He found that mebendazole@ 100mg/kg body weight and piperazine@ 200mg/kg body weight were 100% and 96% effective against *Ascaridia galli.*

Verma *et al.* (1991) studied the efficacy of piperazine adipate, levamisole Hcl, pyrantal pamoate and found that levamisole, piperazine, pyrantel pamoate were effective 92%, 82%, 54% against L4 respectively and 96%, 89%, 71% respectively against adult *Ascaridia galli* in poultry.

Nilson and Alderin (1988) studied the efficacy of piperazine HCl against *Ascaridia galli* in domestic fowls and found these to be effective 90%-100% in removing worms.

Malakhov (1988) stated that piperazine adipate was given at 0.5 g on two successive days to fowls on three premises, one of which had used piperazine for 20 years, one had used it occasionally and the third had not used it at all. Groups of 15 fowls were taken for postmortem examination 10days after treatment. Anthelmintic efficacy was greatly reduced on the first premises.

Sinha *et al.* (1987) studied 99.68 and 99.91% efficacy of fenbendazole and piperazine citrate respectively against ascariasis in calves. Here piperazine citrate was given at 20mg/kg body weight.

2.5 Efficacy of pineapple leaves against ascariasis

Begum *et al.* (2010) investigated prevalence of ascariasis and comparitave efficacy of pinapple Leaves extract with patent drug piperazine against ascariasis of Poultry at five villages under mymensingh district. The efficacy of indigenous medicinal plants used in poultry ascariasis is highly encouraging, pineapple leaves may be used as an alternative anthelmintic in the treatment of ascariasis in indigenous poultry.

Patra *et al.* (2010) conducted a study on comparative anthelmintic efficacy of pineapple and neem leaves in broiler chickens experimentally infected with *Ascaridia galli*. Neem and pineapple leave powder treatment caused cent

percent evacuation of the worms on 28th and 56th day post treatment respectively.

Amin *et al.* (2009) examined *in vitro* anthelmintic efficacy of 20 indigenous medicinal plants against gastrointestinal nematodes of cattle. Water extracts of 20 indigenous plants(neem, tobacco plant, barbados lilac, betel leaf, pineapple, jute, turmeric, garlic, devil's tree, papaya, lime tree, dodder, white teak, conessi tree, bitter gourd, sweet basil, white verticillia, pomegranate, sage, chaste tree) showed potential in vitro activities against adult parasites. Out of these, 20 plant extracts, 10 plants (neem, tobacco, barbados lilac, betel leaf, pineapple, jute, turmeric, garlic, dodder and bitter gourd) showed 100% efficacy against adult worms, 4 plants (devil's tree, papaya, white verticillia and chaste tree) showed 90-98% and others (lime tree, white teak, conessi tree, sweet basil, pomegranate and sage) showed below 90%.

Sujon *et al.* (2008) studied on ten (10) indigenous medicinal plants against gastroinstestinal nematodes of goats. A relatively higher efficacy was recorded in ethanol extract of neem treated animals in comparison to other plants extracts. The results obtained in this study showed that ethanol extract of Labanga, Neem, Karolla and Pineapple at the dose of 100mg/kg showed a significant and potent antinematodal effect. Within these ten (10) plants 4 showed more than 70% efficacy at a concentration of 100mg/mkg.

Khalid *et al.* (2005) studied the effects of indigenous medicinal plants (neem and pineapple) against gastrointestinal nematodiasis in sheep. Sheep were treated with neem and pineapple (10% water extract of leaves). A significant ($p<0.01$) reduction of EPG count was found on 7th, 14th, 21st and 28th day of neem(47.03, 46.27, 41.82 and 37. 60%) and pineapple (41.13, 39.27, 36.32 and 32,18%) treated sheep. The body weight was increased significantly ($p< 0.01$ and $p< 0.05$) in neem and pineapple treated sheep.

Islam *et al.* (2005) studied the comparative efficacy of some modern anthelmintics and pineapple leaves in calves infected with Ascarid parasites. On the basis of fecal egg count of ascaris in calves, ivermectin was found to be more effective(100%), followed by piperazine citrate (100% at 28th day of post-treatment), albendazole (92.95% at 28th day of post-treatment) and pineapple leaves extract (51.215 at 7th day). Total erythrocyte count (TEC), Hemoglobin (HB) and Packed cell volume(PCV) values significantly ($p<0.05$) increased while Erythrocyte sedimentation rate(ESR) significantly ($P<0.05$) decreased following treatment. Body weight of treated calves were significantly ($P<0.05$) increased than the control.

Githiori *et al.* (2004) evaluated the anthelmintic efficacy of 7 plants used as dewormers by farmers and pastoralists in Kenya. Thus 3 commercial anthelmintics and 7 plant preparations were tested in lambs infected with 5000 or 3000 L3 *Haemonchus contortus* in 4 experiments. In the first experiment, ivermectin, levamisole and albendazole were tested in 46 lambs. Seven plant preparations of *Hagenia abyssinica, Olea europaea* var. *africana, Annona squamosa, Ananas comosus, Dodonea angustifolia, Hildebrandtia sepalosa* and *Azadirachta indica* were tested in151 lambs in 3 experiments. Plant preparations *O.europea* and *A.indica* compared with controls, no significant reductions in FEC were observed for any of the treated groups either 2 or 3 weeks post-treatment. Lambs treated with *A.squamosa* and *A.comosus* were slaughtered 4 weeks post-treatment. No significant differences were observed in mean TWC or number of eggs per female worm between treated animals and the controls. No significant improvements in weight gain were observed in treated lambs.

Islam *et al.* (2004) studied the comparative efficacy of Pineapple leaves extract with three modern anthelmintics; ivermectin, albendazole and piperazine citrate mainly on the basis of fecal egg count reduction against

ascariasis. 25 calves naturally infected with ascarids were divided into five equal groups. Group A, B, C and D were treated with ivermectin (200mg/kg ; sc), albendazole (7.5mg/kg; PO), piperazine citrate (200mg/kg ; PO) and pineapple leaves extract (1gm/kg; PO) respectively and group E was kept as untreated control. The efficacy of ivermectin, albendazole, piperazine citrate and pineapple leaves extract on the basis of faecal egg count was found to be 100%, 83%, 100% and 33% respectively. The mean body weight of the treated calves were increased significantly in comparison to control group. Total erythrocyte count (TEC), hemoglobin (Hb) and packed cell volume (PVC) increased whereas erythrocyte sedimentation rate (ESR) decreased significantly.

Hordegen *et al.* (2003) divided forty-eight helminth-free lambs into eight groups (A-H) of six animals. Groups A-G were infected artificially with 10,000 third stage larvae of *Haemonchus con*tortus and 20,000 third stage larvae of *Trichostrongylus colubriformis*, whereas group H remained uninfected. Thirty days post-infection the lambs were treated orally with a single dosage of one of the following products: group A with 3mg/kg body weight (BW) of an aqueous ethanol extract (70%, v/v) of the seeds of *Azadirachta indica* A. juss syn. *Melia azedarach* L. (Meliaceae); group B with 1g/kg BW of a raw powder of the leaves of *Ananas comosus (L.)* Merr. (Bromeliaceae); group C with 0.3 mg/kg BW of an aqueous ethanol extract of a 1:1 mixture (g/g) of *Vernonia anthelmintica* (L.) Willd.(Asteraceae) seeds and Embelia ribes burm (Myrsinaceae) fruits; group D with 183 mg/kg BW of an aqueous ethanol extract of the whole plants of Fumaria parviflora Lam.(Fumariaceae); group E with 28mg/kg BW of an aqueous ethanol extract of the seeds of Caesalpinia crista L. (Caesalpiniaceae); group F with 25mg/kg bwt of pyrantel tartrate and group G with 50% ethanol. Group H remained untreated. Only the ethanol extract of *F. parviflora* caused a strong reduction

of the fecal egg counts (100%) and a 78.2% and 88.8% reduction of adult *H. contortus* and *T. colubriformis* on day 13 post-treatment.the extract was as effective as the reference compound pyrantel tartrate. Therefore, the ethanol extract itself or single constituents of *F. parviflora* could be a promising alternative source of anthelmintic for the treatment of gastrointestinal trichostrongylids in small ruminants.

Akhtar *et al.* (2000) reviewed anthelmintic activity of medicinal plants with particular reference to their use in animals in the indo-pakistan subcontinent. A wide variety of plants are naturally available in the Indo-Pakistan subcontinent which possess narrow or broad spectrum anthelmintic activities.

Khatun *et al.* (1995) reported the anthelmintic activities of water extract of pineapple leaves @ 100, 150 and 200mg/kg body weight were detected with 53.00, 57.00 and 59.41% efficacy rate, respectively against gastrointestinal nematodes in goats. These efficacy rates (53-59%) with pineapple leaves were found inferior in comparison to 98% efficacy rate obtained with fenbendazole. The live body weight, total erythrocytic count and haemoglobin value were found significantly ($p<0.01$) increased at 21st day of post-treatment.

Mostofa (1983) reported *Ananus sativus* (Eng. pineapple) plants, *Glycosmis pantaphylla* (Beng, matkhila) bards etc. are the medium spectrum anthelmintics for the gastrointestinal nematodiasis in cattle.

2.6 Effects of ascariasis on poultry health

Katoch *et al.* (2012) studied on impact of gastrointestinal helminths on body weight gain in backyard chickens in subtropical and humid zone of Jammu, India. One hundred growing chickens, aged 40 days were randomly assigned to two groups (treated and untreated controls) of 50 birds each. The birds in treated group were given fenbendazole at 7.5 mg per kg body weight in

drinking water, while the birds in other group served as untreated controls. At the end of the 90 days of the field trial, the mean body weight gain of untreated controls was 1232.2 ± 7.28 g (13.7 g/day) compared with 1617.6 ± 5.43 g (18.0 g/day) in the treated group. It was associated with a significantly ($P < 0.05$) higher mean worm burden (32.92 ± 6.12) in untreated controls than the treated group (2.46 ± 1.14).

Phiri *et al.* (2007) investigated effects of helminthoses on weight gain in 100 growing chickens randomly assigned to treatment (levamisole) and untreated control groups. There was a significant mean (+/- SEM) weight gain (grams) of 812.8 +/- 51.4 in the treatment group and 623 +/- 57.4 in the control group ($p < 0.01$). The mean (+/- SEM) worm burdens from the control group and the treatment group were 96.3 +/- 5.61 and 22.05 +/- 2.61, respectively.

Permin *et al.* (2006) reported three experiments carried out to examine the consequences of concurrent infections with *Ascaridia galli* and *Escherichia coli* in chickens raised for table egg production. Characteristic pathological lesions including airsacculitis, peritonitis and/or polyserositis were seen in all groups infected with *E. coli*. Furthermore, a trend for increased mortality rates was observed in groups infected with both organisms which, however, could not be confirmed statistically. The mean worm burden was significantly lower in combined infection groups compared to groups infected only with *A. galli*. It was also shown that combined infections of *E. coli* and *A. galli* had an added significant negative impact on weight gain.

Rabbi *et al.* (2006) studied on the gross pathological lesions produced by gastrointestinal helminths in different types of poultry. Gross pathological lesions were found only in backyard poultry. Pathological changes were detected in case of *A. sphenoides* and *H. gallinarum* infection. In *A. sphenoides* infection petechial hemorrhages were observed in the mucosa of

the duodenum. On the other hand, tiny, white, circumscribed nodules of about 2-3 mm of diameter were found in the caecal mucosa in case of *H. gallinarum* infection.

Islam *et al.* (2005) observed the effects of some modern anthelmentics and pineapple leaves on certain blood parameters and body weight gain in calves infected with Ascarid parasites. They found that total erythrocyte count (TEC), hemoglobin (Hb) and packed cell volume (PCV) values significantly ($p<0.05$) increased while erythrocyte sedimentation rate (ESR) significantly ($p<0.05$) decreased following treatment. Body weights of treated calves were significantly ($p<0.05$) increased in treated calves than the control.

CHAPTER III

MATERIALS AND METHODS

CHAPTER III

MATERIALS AND METHODS

The experiment was conducted at the Department of Physiology and Pharmacology, Hajee Mohammed Danesh Science and Technology University, Dinajpur and Upazila Livestock Office, Palashbari, Gaibandha, during the period from July to November/2012. To complete the research work following steps were followed:

3.1 To study the proportional incidence of ascariasis in indigenous chickens

A total of 500 native chickens were examined ranging from 2 to 7 months of age from different villages of Palashbari Upazila of Gaibandha district. Prevalence was recorded on the basis of

a) Male/Female infection rate,
b) Incidence rate in different villages of Palashbari Upazila of Gaibandha district and
c) Age-wise infection rate.

For prevalence study fecal samples were collected from the cloaca of chickens in polythene bags early in the morning. Samples were sent quickly after collection to the laboratory for examination. For assessing prevalence, direct smear method and Stoll's ova counting technique were used for fecal examination.

3.2 Collection of chickens

60 chickens were collected from the local market. Among them, 45 chickens having infection with eggs of *Ascaridia galli* parasite were selected for this experiment. The chickens were allowed to take rest for 7 days for adaptation. The experiment was carried out in Upazila Livestock Office, Palashbari, Gaibandha. The age and body weight of all selected chickens ranged from 60 to 210 days and 300 to 500 gm respectively. The chickens were supplied with normal diet and water.

3.3 Fecal egg counts

For determination of infectivity, fecal samples were collected and eggs were counted by Stoll's ova counting technique and direct smear method through microscope in the laboratory.

3.3.1 Egg counting by Stoll's ova counting method

Experimental procedures and calculation

3 gms of feces was thoroughly mixed with 42 ml of NaOH (sodium hydroxide) solution in 50 ml cylinder. Then 0.15 ml was withdrawn with a graduated pipette and transferred to a glass slide and covered with a 22 X 40 mm cover glass. Number of eggs were counted under the lower power of microscope. Number of ova counted was multiplied by the factorial 100 to determine the number of ova per gram of faeces. (Rahman *et al*, 1996)

3 gm + 42.00 cc = Total volume 45.0ml

3 gm faeces in 45.0 ml suspension

1 gm faeces in 15.0 ml suspension

0.15 ml equivalent to 15.0 ml ÷ 100

3.3.2 Direct smear method

Experimental procedures

A drop of water was placed on the centre of a clean glass slide. A small amount of faeces was detached from the given sample with the help of a tooth-pick and spread out to form a thin smear. Then slide was covered with cover slip and placed under the low power objective (10x) of a microscope for examination. At least three slides from each faecal sample were examined. (Rahman *et al*, 1996)

3.4 Selection and collection of drugs/chemicals

Pipervet® (piperazine) and Pineapple (*Ananus comosus*) leaves were selected. Pineapple leaves (*Ananus comosus*) were collected from the horticulture garden of Hajee Mohammed Danesh Science and Technology University. Then 10% water extract of Pineapple leaves was prepared freshly, 20 gm Pineapple leaves (*Ananus comosus*) was extracted in mortar and pastle and the extract was made upto 20ml by adding distilled water and filtered and administered orally by dropper.

Pipervet® (piperazine) was purchased from local market.

3.5 Evaluation of the comparative efficacy of modern anthelmintics piperazine citrate and Pineapple leaves (*Ananus comosus*) extract against ascariasis in chickens

3.5.1 Experimental design

All the 45 chickens randomly divided into 3 groups (A, B and C) for assessing the efficacy of modern anthelmintics Piperazine and Pineapple leaves extract against ascariasis in chickens.

Group A : Control (without treating)

Group B : Piperazine (Pipervet®) treated group. The drug was administered orally @ 200mg/kg bwt by dropper as a single dose.

Group C : Pineapple leaves extract treated group. This was administerded orally @ 1 gm/kg bwt by dropper by consecutive seven days.

All the chickens of treated and control groups were closely observed for 28 days after treatment and following parameter were studied.

Layout of the Experiment

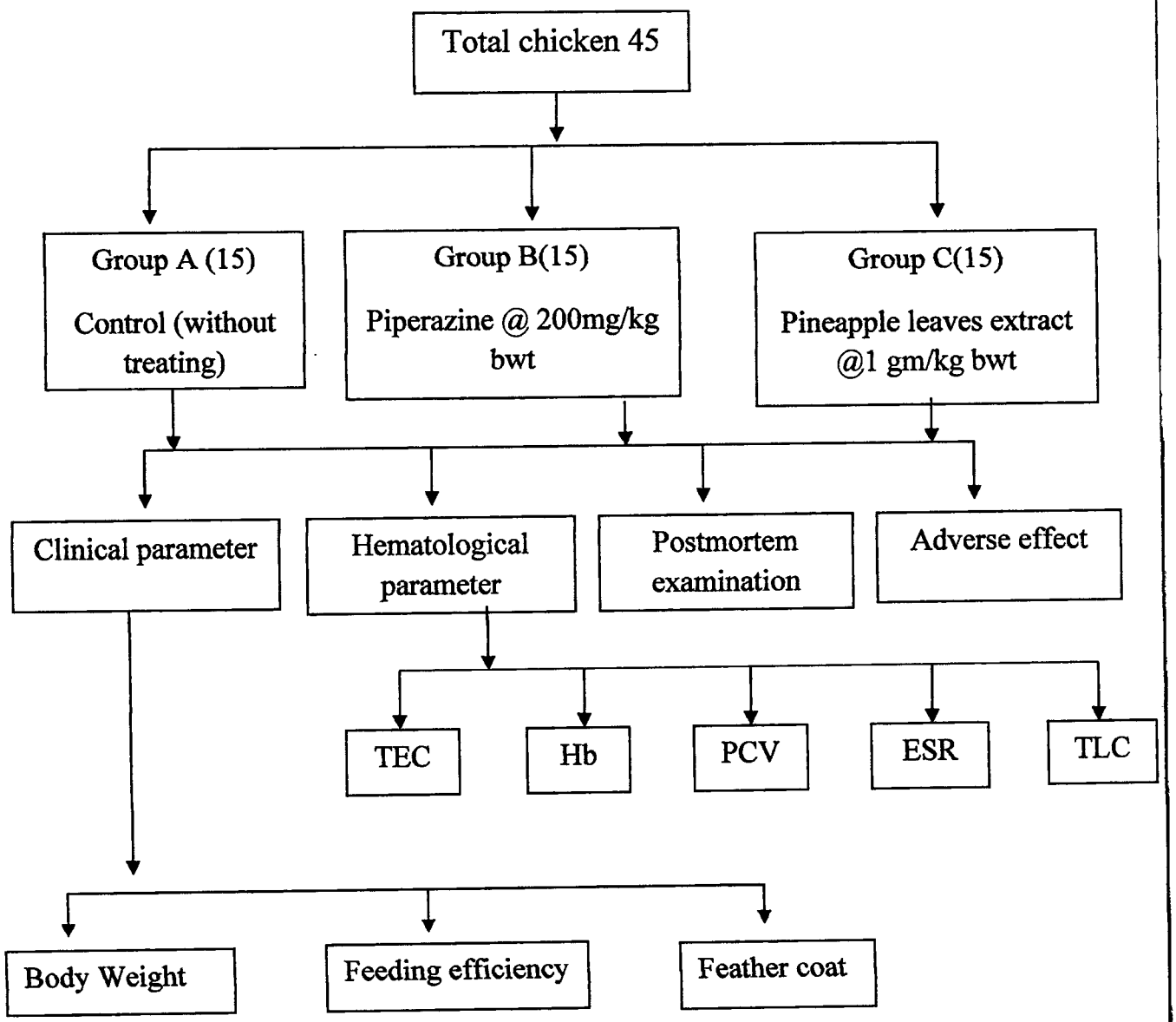


Fig. 1. Layout of the experiment

3.5.2 Clinical examination

a) The effect of the Pineapple leaves extract and Pipervet® (Piperazine) on body weight was recorded before and during administration of drugs.

b) The effect of the Pineapple leaves extract and Pipervet® (Piperazine) on feeding efficiency was recorded before and during administration of drugs.

c) The effect of the Pineapple leaves extract and Pipervet® (Piperazine) on feather coat was observed before and during administration of drugs.

d) Chickens under trial and control groups were weighed with Electric weighing machine. The weight of each chicken was taken before feeding in the morning , in noon and afternoon. The average of these three weights was calculated and recorded.

Mean live weight of each group of chicken on 7^{th}, 14^{th}, 21^{st} and 28^{th} days were recorded with the procedures described above.

3.5.3 Hematological parameters

Blood samples were collected from the wing vein of chicken of both control and treated groups at pre-feeding and during feeding (28 days) period at 7 days interval to study the effect of the Pineapple leaves extract and Pipervet® (Piperazine). The following parameters were observed:

a. Total erythrocyte count (TEC)
b. Hemoglobin estimation (Hb)
c. Packed cell volume (PCV)
d. Erythrocyte sedimentation rate (ESR)
e. Total leukocyte count (TLC)

Total erythrocytes count (TEC), Erythroctes sedimentation rate (ESR), Packed cell volume (PCV) and Total leukocyte count (TLC) were performed as per methods described by Schalm (1967). Hemoglobin estimation was perfomed as per method described by Coffin (1955).

3.5.3.1 Total erythrocyte count (TEC)

a) The tip of the dry and clean red pipette was placed on the blood sample.
b) The blood was sucked up until it reached the exactly 0.5 mark and carefully wiped the tip of the pipette with a piece of absorbent cotton.
c) Then the tip of the pipette placed immediately in the diluting fluid and filled the pipette exactly upto 101 mark.
d) The number tube around the tip of the pipette was stretched and held with thumb and finger at each end.
e) The contents of the pipette was shaked thoroughly with 8 knot or twisting motion for 1-2 minutes.
f) Then the counting chamber with cover glass was placed under the microscope and made visible the finely rolled area with low power objective.
g) After discarding 2 or 3 drops, a small drop from the pipette was placed to the end of the polished surface of the counting chamber containing the ruling and allowed the space to fill the area under the cover glass.
h) The chamber was allowed to stand for 2 minutes to settle the erythrocytes and counted the cells on the four corner squares and one center square with high power objective(100x).
i) The number of RBC was calculated as follows:
j) Number of RBC = No. of cell counted x 10000
k) The result was expressed in million/ cu.mm.

3.5.3.2 Determination of Hemoglobin

Following procedures were taken for determination of Hemoglobin.

a) N/10 HCl was placed into the perfectly clean and dry diluting tube upto 2 mark.
b) Then the blood sample was drawn into the sahli pipette exactly upto 20 mark and the side of pipette wiped to remove sticking blood to its outside with absorbent cotton.
c) The blood of the pipette was transferred immediately into the diluting tube containing N/10 HCl acid and rinsed the pipette 2 or 3 times by sucking water into the pipette and added these washing to the solution in the tube.
d) Using the dropper, water was added drop by drop, each time mixing the solution with a stirrer until the color of the solution matched the standard.
e) After 5 minutes of first noting time, the result was read in day light from the scale of the measuring tube by observing the graduation mark at the lower edge of the meniscus at the top of the liquid column.
f) The result was expressed in gm%.

3.5.3.3 Packed cell volume determination (PCV)

The wintrobe hematocrit tube was filled up with well-mixed blood by special loading pipette upto 10 mark. The tubes were centrifuged at 3000 rpm for half an hour, then the reading was taken.

3.5.3.4 Determination of Erythrocyte sedimentation rate (ESR)

Following procedures were taken for the determination of ESR.

a) The blood sample was drawn in the loading pipette.
b) Tip of the pipette was inserted to the chemically cleaned and dry hematocrit tube and expelled the blood slowly by pressure on the rubber bulb withdrawing the pipette and thus filled the tube with blood exactly upto "0" or "100" mark. Care was taken so that there was no bubble in the tube.
c) The filled tube was placed in a special rack in exact vertical position and waited for one hour.
d) After passing the required time, the fall of RBC column from the scale at the top of the tube was noted.
e) The result was expressed in mm/1st hour.

3.5.3.5 Total leukocyte count (TLC)

The principles involved in enumeration of Total Leukocyte Count were almost same to those of erythrocytes. Here the leukocyte diluting fluid was N/10 HCl solution. Well mixed blood was drawn upto the 0.5 mark of white blood cell pipette. The diluting fluid was filled up to the 11 mark of the pipette and the contents were thoroughly mixed for 2 minutes. 2-3 drops of content were discarded and counting chamber was then filled in the same way as in the red blood cell count. The counting chamber was placed under the microscope and examined under low power objective (10x). The leukocytes in the 4 large squares (each 1 square mm.) of the counting chamber were counted x 50 and expressed the result in thousand per cu.mm.

3.5.4 Counting of parasites by Postmortem examination

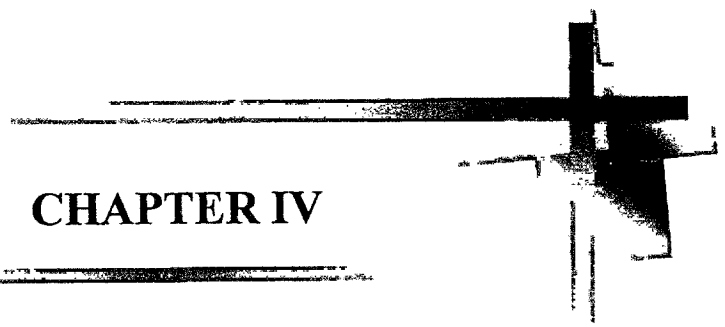
Before treatment three chickens from each group were also slaughtered to count the number of parasites (Ascarids) and to see if there were any pathological changes present. After treatment three chickens from each group were slaughtered to count number of parasites (Ascarids) and to see if there were any pathological changes present on 14^{th} day of treatment. That was also done on 28^{th} day of treatment. There was no significant pathological change in any internal organs of the chicken of the treated groups. On the other hand, in the control (untreated) group there were presence of parasites (*A. galli*) on 14^{th} and 28^{th} day and ulcerative lesion in the intestine.

3.5.5 Adverse Effects

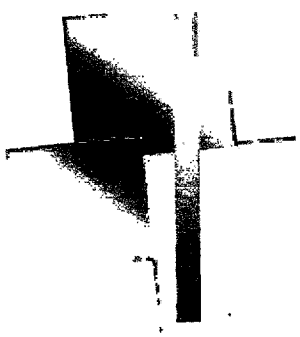
After Treatment there were no adverse effects on treated chickens.

3.5.6 Statistical analysis

The data were analyzed statistically between control and treated groups of chicken by the well-known *student's test* ('t' test).



CHAPTER IV



RESULTS

CHAPTER IV

RESULTS

A total of 500 chickens from different villages of Palashbari Upazila of Gaibandha district were examined to study the (a) proportional incidence of Ascarid nematodes infection. In addition one patent drug namely Pipervet® (Piperazine) and one indigenous medicinal plant Pineapple leaves extract were given orally to study (b) the comparative efficacy of these drugs against ascariasis by observing the effect of these two drugs on clinical (Body weight, presence or absence of parasite, feed intake and water consumption) and haematological parameters (TEC, Hb, PCV, ESR and TLC).

4.1 Prevalence

4.1.1 Male/female infection rate

Out of 500 chickens 365 were hens and 135 were cocks. Among the female and male birds 292 (80%) and 119 (88.15%) were found infected with Ascarid nematodes respectively.

4.1.2 Area wise infection rate

The proportional incidence of ascariasis in chickens in different villages of Palashbari Upazila of Gaibandha district is summarized in Table-1. The highest (93%) and lowest (74.74%) proportional incidence of ascariasis were recorded in village Andua and Nuniagari respectively. However, the prevalence in villages- Beradanga (76.36%), Bengulia (85.45%), Katuli (78.18%), Shimulia (90%) and Gridharipur (82.35%) were also recorded. A chi-square test revealed a significant variation in the proportional incidence rates of the infection among different villages.

Table- 1. Proportional incidence of ascariasis in chickens of different villages of Palashbari Upazila of Gaibandha district

Name of the places (Village)	No. of chicken examined	No. of chicken infected	% of infected chicken
(1) Beradanga	55	42	76.36%
(2) Bengulia	55	47	85.45%
(3) Katuli	55	43	78.18%
(4) Andua	100	93	93%
(5) Shimulia	60	54	90%
(6) Gridharipur	85	70	82.35%
(7) Nuniagari	95	71	74.74%
	Total=500	Total=420	Total infection = 84%

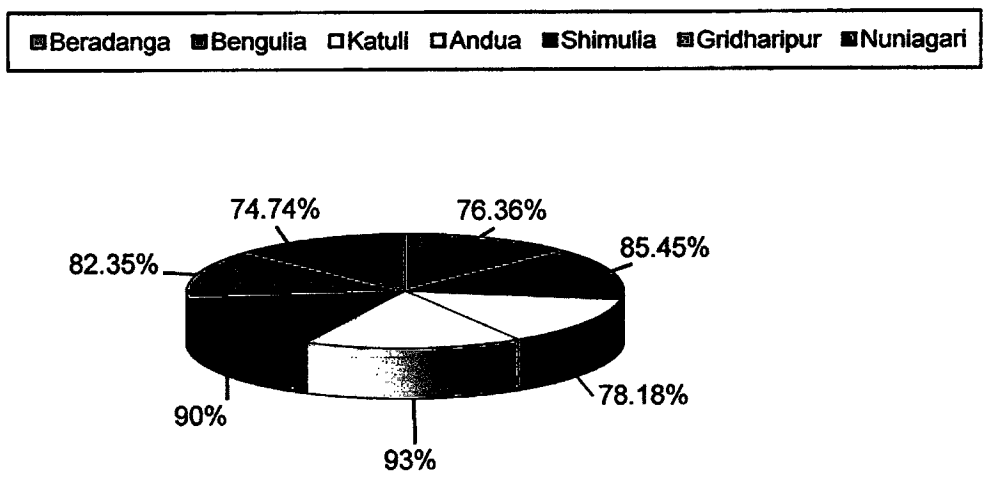


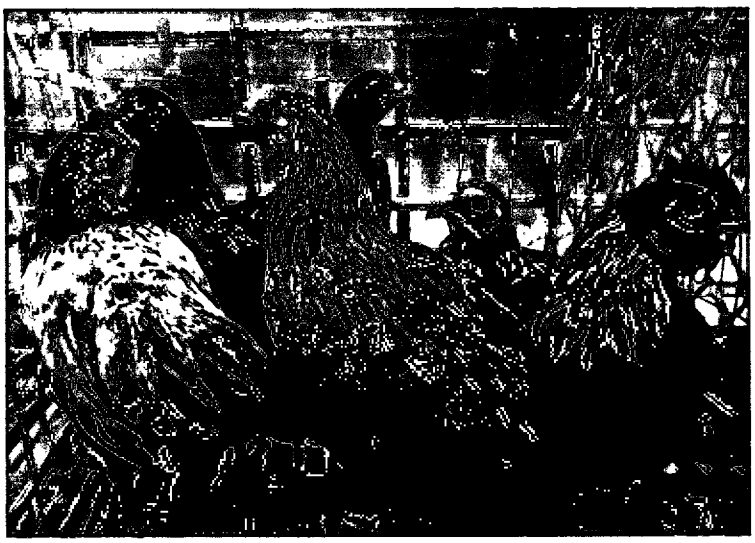
Fig. 2. Proportional incidence of ascariasis in chickens of different villages of Palashbari Upazila of Gaibandha district

4.1.3 Age wise infection rate

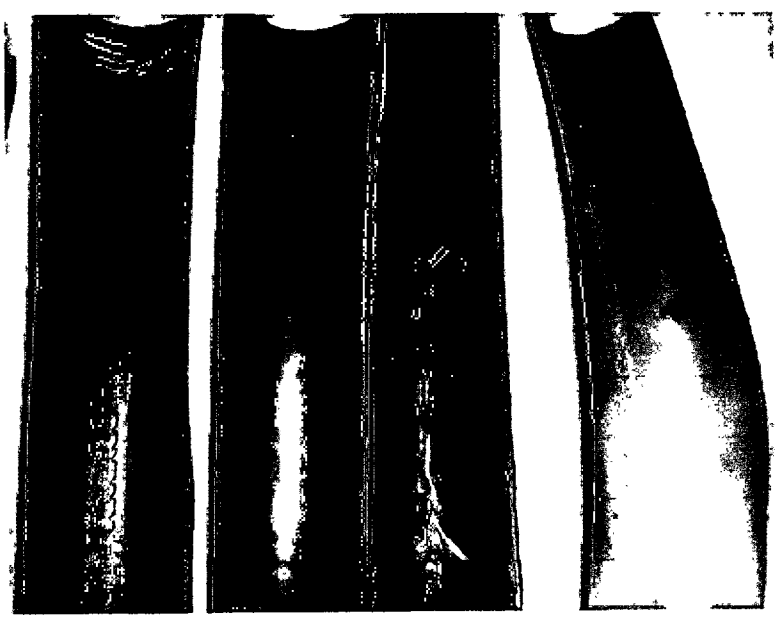
Proportional incidence of ascariasis in chickens of various age groups of 60 days to 90 days is shown in Table – 2. A significant higher incidence (95.26%) of the infection was recorded in chickens between 60-90 days of age, followed by 92.78% and 55.38% between age group of 3-5 months and 5-7months respectively.

Table 2. Proportional incidence of ascariasis at various age groups of chickens of Palashbari Upazila of Gaibandha district

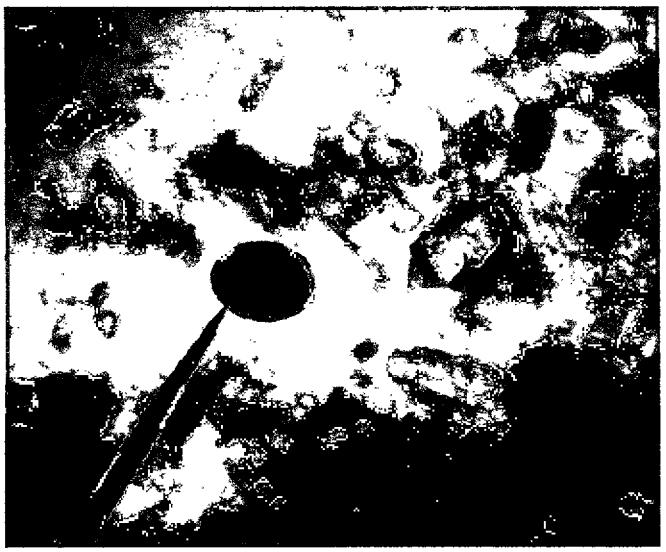
Category	Category-1 60-90 days (2-3 months)	Category-2 91-150 days (above 3-5 months)	Category-3 151-210 days (above 5-7 months)	Total
Number of chicken examined	190	180	130	500
Number of infected chicken examined	181	167	72	420
Rate of infection (%)	95.26	92.78	55.38	84



Picture 1: Chicken for experiment



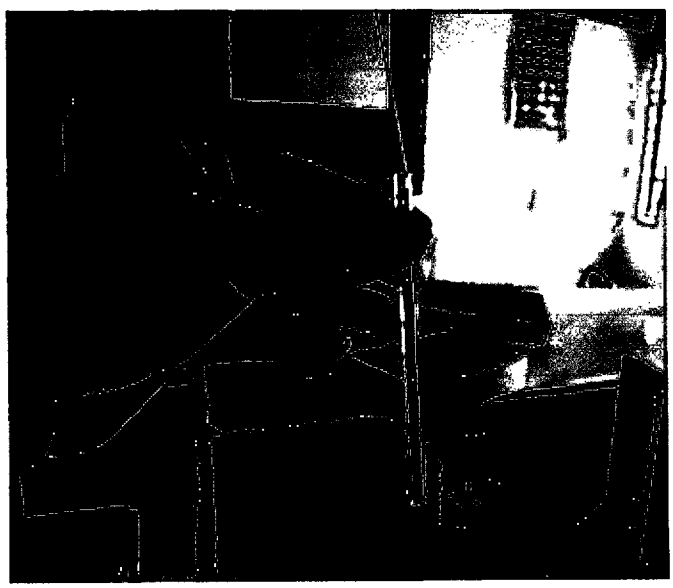
Picture 2: Leaves of Pineapple (*Ananus comosus*)



Picture 3: Egg of *Ascaris spp*

Picture 4: Collection of blood from wing vein of chicken

Picture 5: Blood for hematological test



Picture 6: Determination of Packed cell volume

4.2 Studies on the comparative efficacy of patent drug Pipervet® (piperazine) and Pineapple leaves as anthelmentics against ascariasis in chickens.

4.2.1 Clinical examination

4.2.1.1 Effect on body weight

The effect of one patent drug piperazine and one indigenous medicinal plant Pineapple (*Ananus comosus*) leaves on body weight was observed for 28 days at 7 days interval. Mean body weight of each group of chickens prior to treatment and after treatment on 7^{th}, 14^{th}, 21^{st} and 28^{th} days was calculated in gram and percentage of live weight gain/loss in gram was shown in the Table no. 3.

On day '0' the mean value of initial body weight of group B treated with pipervet® was 414.90±3.94 (gm.) and on the 28^{th} day of post treatment, the mean value of body weight was 432.267±2.01 (gm.). The increased body weight was significant ($p<0.05$) in comparison to their pre-treatment ('0' day) value.

Similarly, on day '0' the mean value of initial body weight of group C treated with Pineapple leaves extract was 430.03±5.29 (gm.) and on the 28^{th} day of post treatment, the mean value of body weight was 446.096±5.26. The increased body weight was significant ($p<0.05$) in comparison to their pre-treatment ('0') value.

Table 3. Effects of Pipervet® and Pineapple leaves extract on body weight in chicken

Group of chicken	Drug, dose and route	Pre-treatment (gm.)	After drug administration (Post-treatment) (gm.)				Mean weight gain (%)
		0day	7th day	14th day	21st day	28th day	
A	Control (untreated)	402.466±3.65	398.241±3.88	405.53±3.58	403.413±3.54	406.06±3.46	0.89
B	pipervet® @ 200mg/kg bwt orally as a single dose	414.90±3.94	417.845±4.60	425.157±2.25	425.6127±1.63	432.267*±2.01	4.19
C	Pineapple leaves extract @ 1gm/kg bwt orally for seven consecutive days	430.03±5.29	431.99±5.92	437.79±5.19	439.074*±5.56	446.096*±5.26	3.74

Values given above the represent the mean ± SE of 5 chickens

* Significantly increased ($p<0.05$)

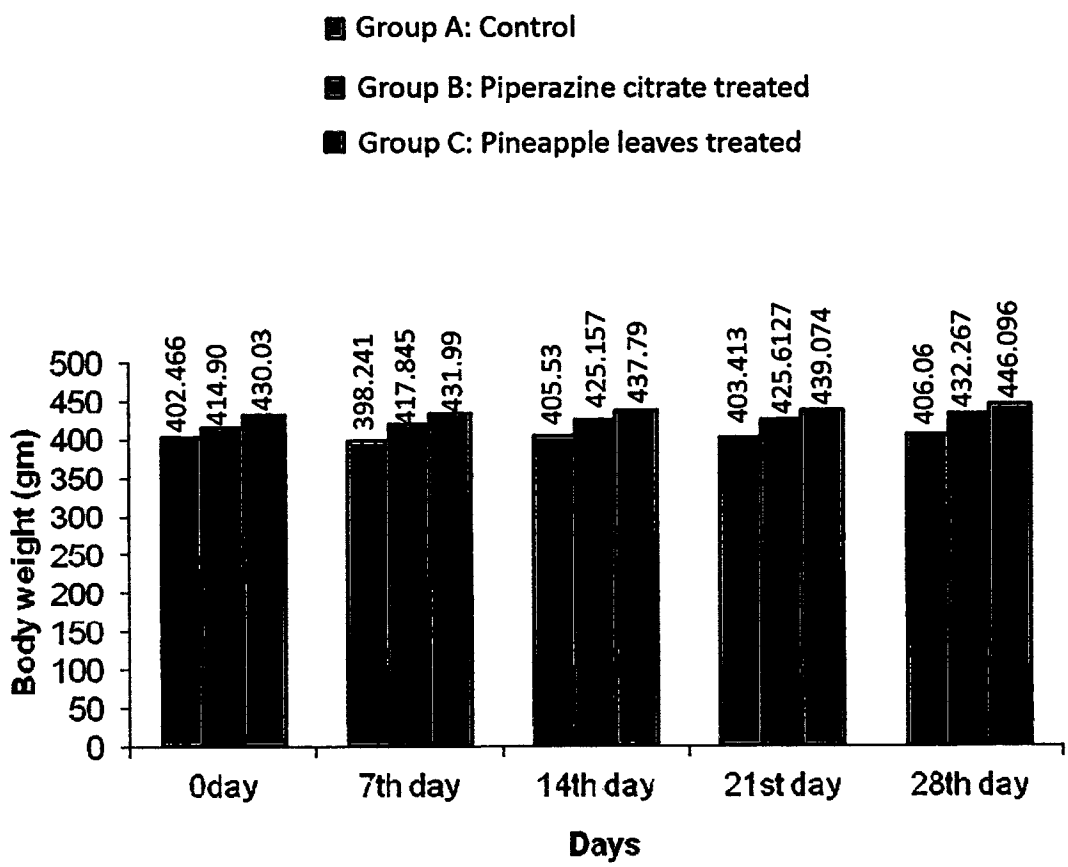


Fig. 3. Effects of Pipervet® and Pineapple leaves extract on body weight in chicken

4.2.2 Fecal egg count

The results of the efficacy of pipervet® and Pineapple leaves extract against ascariasis in village poultry are shown in the Table 4.

Reduction of EPG count was found on 14th and 28th day in the group of chickens of B and C. On the other hand EPG count was found increased in the group A (control).

Table 4. Effects of pipervet® and Pineapple leaves extract on fecal egg count

Group of chicken	Drug and dose	Pre-treatment (No.)	Post-treatment (No.)	
		0 day	14th day	28th day
A	Control (untreated)	300 ± 11.07	320 ±11.42	340 ± 13.96
B	Pipervet®@ 200mg/kg bwt orally as a single dose	260 ± 10.68	0	0
C	Pineapple leaves extract@ 1gm/kg bwt orally for consecutive 7 days	300 ± 11.07	140 ± 7.40	60 ± 7.40

Values given above the represent the mean ± SE of 5 chickens

4.2.3 Hematological parameters

4.2.3.1 Effect on total erythrocyte count (TEC)

The effect of oral administration of one patent drug (Pipervet®) and one indigenous plant (Pineapple leaves) on TEC was determined on the 7th, 14th, 21st and 28th day after treatment. The values are shown in Table no 5.

The oral administration of Pipervet® (200mg/kg bwt) and Pineapple leaves extract (1 gm/kg bwt) increased the number of erythrocytes of chickens in group B and C respectively. The highest number of cells was recorded on day 28th dat after application of drugs.

Table 5: Effects of Pipervet® and Pineapple leaves extract on Total Erythrocyte Count (million/cu mm) in indigenous chickens

Group of chicken	Drug and dose	Pre-treatment (million/ cu mm)	After drug administration(post-treatment) (million/cu mm)			
		0 day	7th day	14th day	21st day	28th day
A	Control	3.20±0.32	3.17±0.46	3.056±0.34	3.12±0.30	2.96±0.19
B	Pipervet® @ 200mg/kg bwt orally as a single dose	3.38±0.14	3.44±0.16	3.52±0.26	3.55±0.12	3.63±0.22
C	Pineapple leaves extract @ 1 gm/kg bwt orally for consecutive 7 days	3.53±0.26	3.55±0.26	3.56±0.265	3.61±0.29	3.7±0.13

Values given above the represent the mean ± SE of 5 chickens

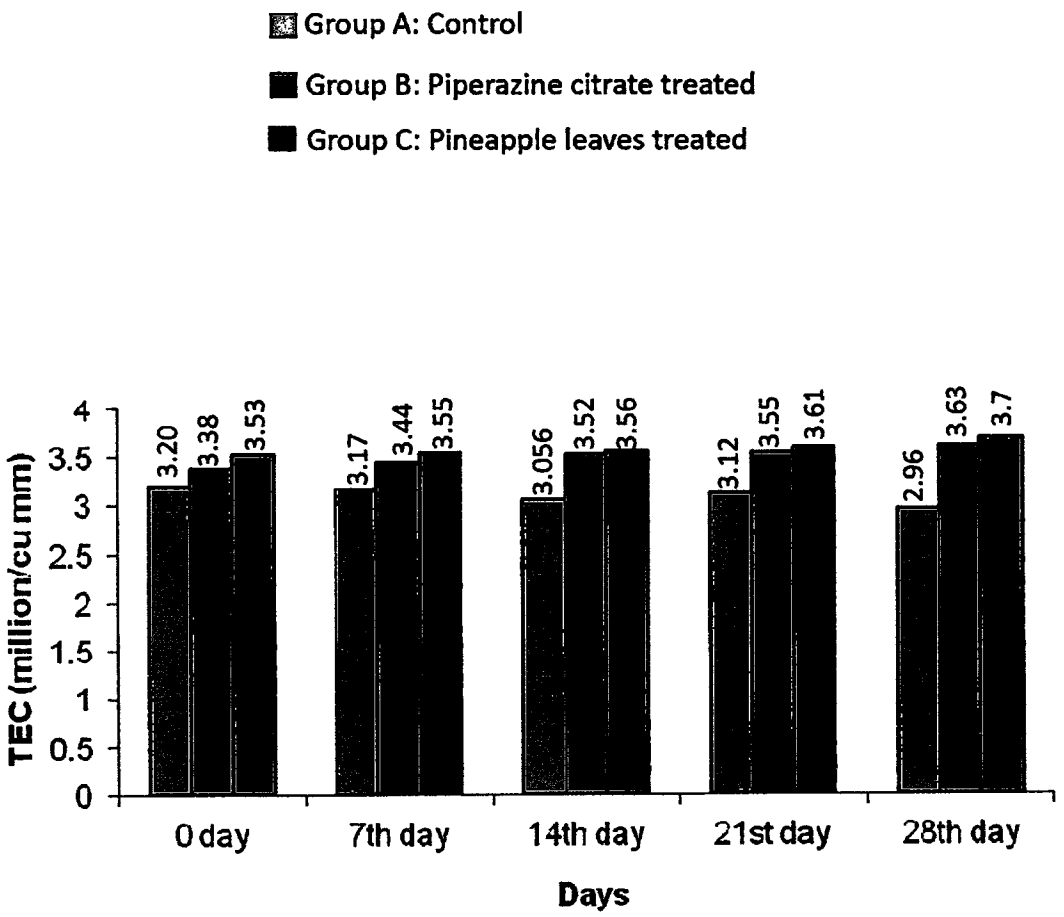


Fig. 4. Effects of Pipervet® and Pineapple leaves extract on TEC in chicken

4.2.3.2 Effect on Hb estimation

The effect of Pipervet® and Pineapple leaves extrac on Hb estimation in chickens was shown in Table 6.

The oral administration of Pipervet® (200mg/kg bwt) and Pineapple leaves extract (1 gm/kg bwt) increased the the Hemoglobin level in chickens. The increase of Hemoglobin level was highest on 28th day after treatment.

Table 6: Effects of Pipervet® and Pineapple leaves extract on Hemoglobin (gm%) in indigenous chickens

Group of chicken	Drug and dose	Pre-treatment (gm%)	After drug administration(post-treatment) (gm%)			
		0 day	7th day	14th day	21st day	28th day
A	Control	9.14±0.68	8.96±0.57	8.9±0.50	8.88±0.51	8.82±0.50
B	Pipervet ® @ 200mg/kg bwt as a single dose	9.42±0.33	9.42±0.49	9.44±0.34	9.5±0.47	9.52±0.55
C	Pineapple leaves extract @ 1 gm/kg bwt for consecutive 7 days	9.48±0.54	9.52±0.47	9.6±0.38	9.6±0.35	9.8±0.53

Values given above the represent the mean ± SE of 5 chickens

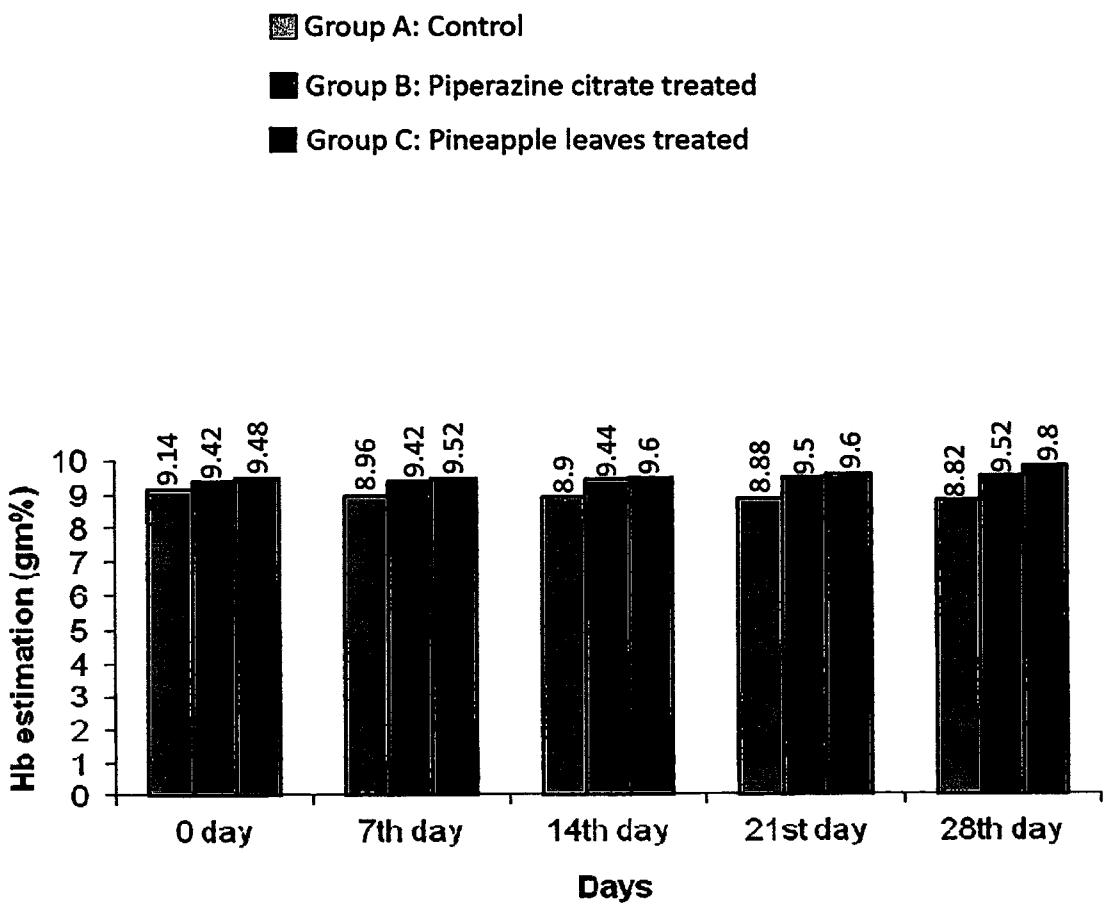


Fig. 5. Effects of Pipervet® and Pineapple leaves extract on Hb estimation in chicken

4.2.3.3 Effect on Packed cell volume (PCV)

The oral administration of Pipervet®(200mg/kg bwt) and Pineapple leaves extract (1 gm/kg bwt) in ascarid infected chicken showed a significant effect on PCV. The increase of PCV values was highest on 28th day after treatment. The effect on PCV was shown on Table 7.

Table 7: Effects of Pipervet® and Pineapple leaves extract on Packed Cell Volume (% 30 minutes) in indigenous chickens

Group of chicken	Drug and dose	Pre-treatment (% 30 minutes)	After drug administration(post-treatment) (% 30 minutes)			
		0 day	7th day	14th day	21st day	28th day
A	Control	20.46±0.88	20.02±0.78	19.8±0.78	19.12±0.93	18.22±0.69
B	Pipervet ® @ 200mg/kg bwt as a single dose	19.32±0.91	19.48±0.81	20.84±0.78	22.52±0.77	22.72±0.36
C	Pineapple leaves extract @ 1 gm/kg bwt for consecutive 7 days	19.36±0.88	20.86±0.84	20.92±0.78	21.64±0.78	22.38±0.65

Values given above the represent the mean ± SE of 5 chickens

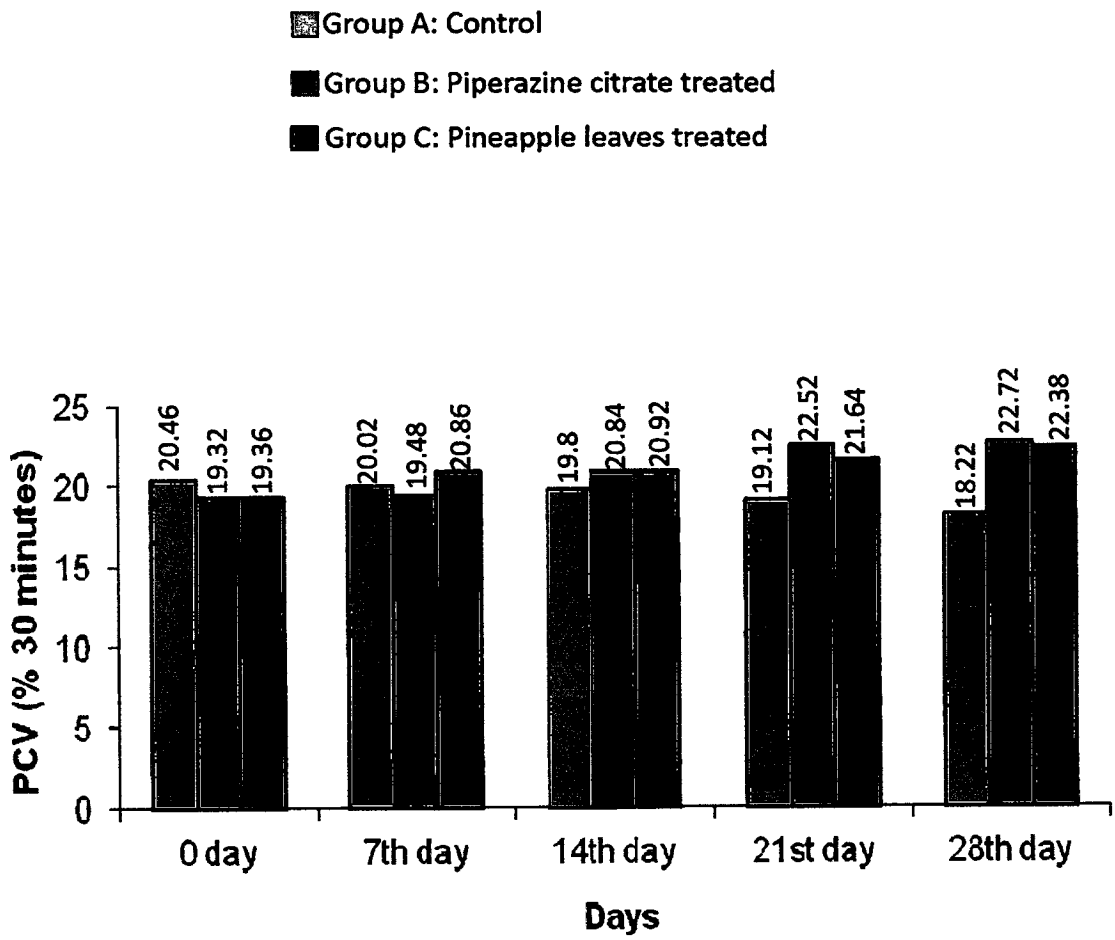


Fig. 6. Effects of Pipervet® and Pineapple leaves extract on PCV in chicken

4.2.3.4 Effect on Erythrocyte Sedimentation Rate (ESR)

The oral administration of Pipervet® (200mg/kg bwt) and Pineapple leaves extract (1 gm/kg bwt) reduced the ESR of chickens . The highest reduction of ESR was found on 28th day after treatment. The effect on ESR was shown in Table 8.

Table 8: Effects of Pipervet® and Pineapple leaves extract on Erythrocyte Sedimentation Rate (mm/1st hour) in indigenous chickens

Group of chicken	Drug and dose	Pre-treatment (mm/1st hour)	After drug administration (post-treatment) (mm/1st hour)			
		0 day	7th day	14th day	21st day	28th day
A	Control	0.62±0.39	0.56±0.41	0.6±0.32	0.58±0.29	0.48±0.29
B	Pipervet® @ 200mg/kg bwt as a single dose	0.52±0.36	0.5±0.32	0.46±0.34	0.48±0.29	0.42±0.29
C	Pineapple leaves extract @ 1 gm/kg bwt for consecutive 7 days	0.5±0.27	0.44±0.34	0.48±0.29	0.42±0.36	0.4±0.32

Values given above the represent the mean ± SE of 5 chickens

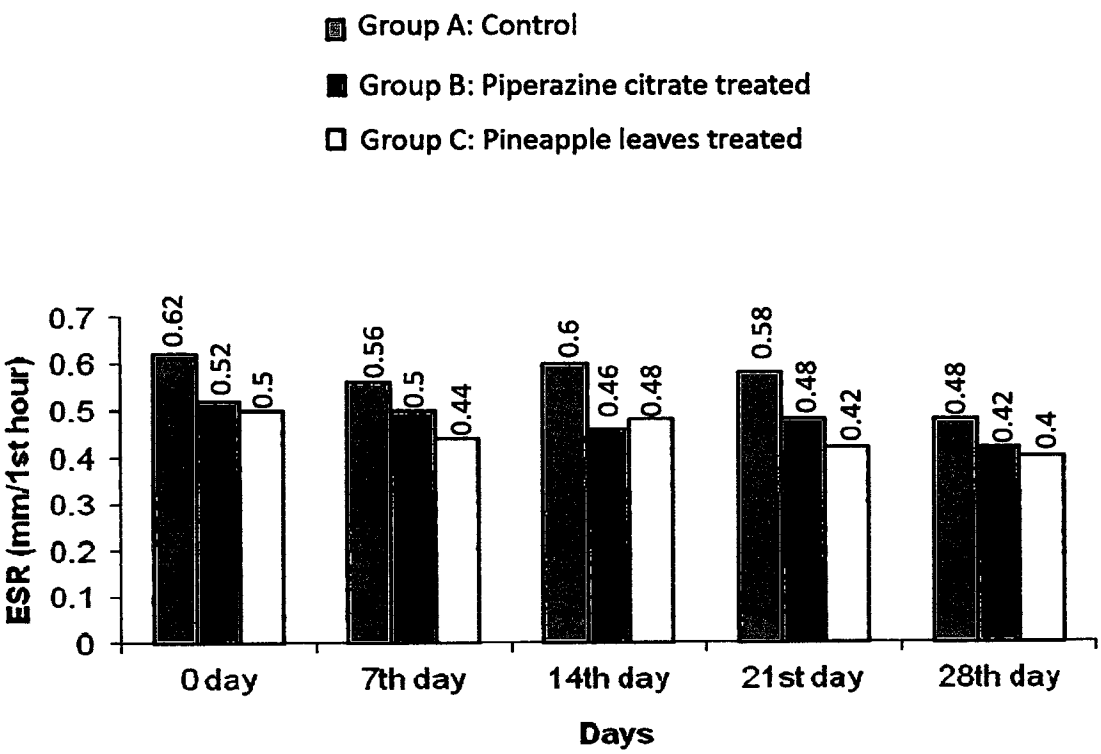


Fig. 7. Effects of Pipervet® and Pineapple leaves extract on ESR in chicken

4.2.3.5 Effect on Total Leukocyte Count (TLC)

The effect of one patent drug Pipervet® and one indigenous medicinal plant Pineapple leaves extract on Total leukocyte count was shown in Table 9 .

The oral administration of the pervet® (200mg/kg bwt) and Pineapple leaves extract (1 gm/kg bwt) reduced the TLC of chicken. The highest reduction of TLC was found on 28th day after treatment.

Table 9: Effects of Pipervet® and Pineapple leaves extract on Total Leukocyte Count ($10^3/mm^3$) in indigenous chickens

Group of chicken	Drug and dose	Pre-treatment ($10^3/mm^3$)	After drug administration (post-treatment) ($10^3/mm^3$)			
		0-day	7th day	14th day	21st day	28th day
A	Control	10.23±0.69	10.32±0.75	10.37±0.71	10.41±0.64	10.49±0.81
B	Pipervet®@ 200mg/kg bwt orally as a single dose	7.97±0.79	7.93±0.67	7.91±0.60	7.88±0.39	7.84±0.75
C	Pineapple leaves extract@ 1gm/kg bwt orally for consecutive 7 days	7.66±0.64	7.60±0.45	7.56±.30	7.53±0.28	7.43±0.39

Values given above the represent the mean ± SE of 5 chickens

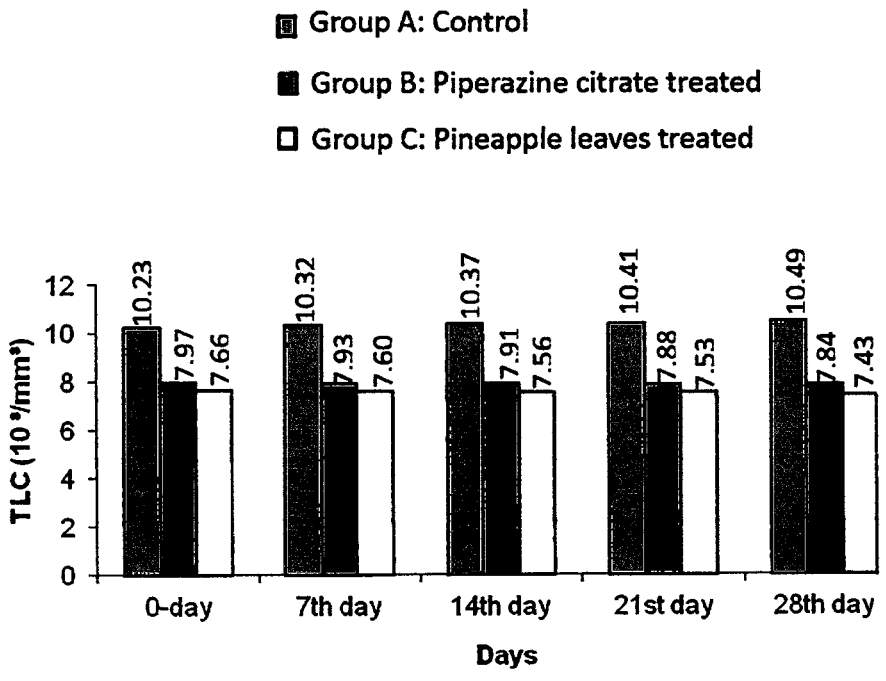


Fig. 8. Effects of Pipervet® and Pineapple leaves extract on TLC in chicken

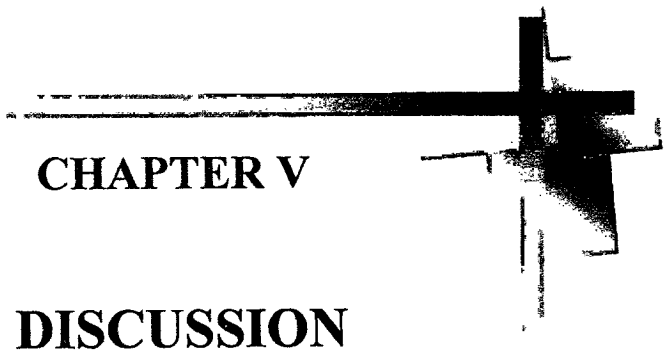
4.2.4 Postmortem examination

One patent drug Pipervet ® and one indigenous medicinal plant namely Pineapple leaves extract were given orally in chickens of groups B and C respectively and observed for 28 days. Before treatment three chickens from each group were also slaughtered to count the number of parasites (Ascarids) and to see if there were any pathological changes present . After treatment three chickens from each group were slaughtered to count number of parasites (Ascarids) and to see if there were any pathological changes present on 14th day of treatment. That was also done on 28th day of treatment. There was no significant pathological change in any internal organs of the chicken of the treated groups. Table 10 shows the effect of Pipervet® and Pineapple leaves extract on number of parasites in chickens.

Table 10. Effects of Pipervet® and Pineapple leaves extract on number of parasites in chickens.

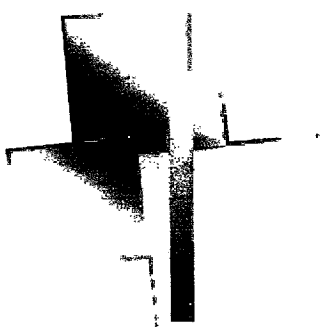
Group of chicken	Drug and dose	Pre-treatment (No.)	After drug administration (post-treatment) (No.)	
		0 day	14th day	28th day
A	Control	10±1.41	14±1.41	15±1.90
B	Pipervet®@200mg/kg bwt orally as a single dose	9±1.73	0	0
C	Pineapple leaves extract @ 1gm/kg bwt orally for consecutive 7 days	8±1.63	3±1.32	1±1.31

Values given above the represent the mean ± SE of 3 chickens



CHAPTER V

DISCUSSION



CHAPTER V

DISCUSSION

In this research work following parameters were studied

1. The proportional incidence of ascariasis in indigenous chickens in various villages of palashbari thana of Gaibandha district.
2. Comparative efficacy of one patent drug namely Pipervet® and one indigenous plant, Pineapple leaves (*Ananus comosus)* extract against ascariasis in indigenous chickens.
3. Effect of Pipervet® and Pineapple leaves extract as anthelmintics on feed intake, feather coat and body weight.
4. Effect of Pineapple leaves extract and Pipervet® on hematological parameters.
5. Post mortem examination of chicken to study the number of parasites present and adverse effects of Pipervet® and Pineapple leaves extract.

5.1 The proportional incidence of ascariasis in chicken

In this research work chicken from 60 days (2 month) to 210 days (7 months) of age of different villages of Palashbari thana of Gaibandha district were examined and overall rate of infection of ascariasis was found 84%. The rate of infection was found highest in village Andua (93%) and lowest in Nuniagari (74.74%). The rate of infections were 95.26% in 60 to 90 days (2 to 3 months) of age, 92.78% in 91 to 150 days (above 3 to 5 months) of age and 55.38% in 151 to 210 days (above 5 to 7 months) of age of chickens. In our country, more or less similar prevalence of Gastro-intestinal nematodiasis in chickens have been reported earlier by Sarker *et al.* (2009) who found that Highest prevalence of Ascariasis was in Daganbhuiyan upazila (90.47%) and lowest was in Parshuram upazila (81.57%) under Feni district. Rabbi *et al.*

(2006) recorded the prevalence of *R. tetragona* (100%) was the highest followed by that of *A. galli* (87.50%) and *H. gallinarum* (80%) in backyard poultry. Haq (1986) recorded *Ascaridia galli* as 45%, *Heterakis gallinae* 80%, *Capillaria annulata* 10%, and *Capillaria columbae* 15%. Samad *et al.*(1985) investigated incidence of gastro-intestinal parasitic infection in domestic fowls and recorded 6 parasites namely, *Ascaridia galli* (44.83%), *Heterakis gallinae* (58.38%), *Capillaria anulata* (8.34%), *Capilaria columbae* (16.38%), *Ascaridia spiralis* (16.28%) and *Subulura bumpti* (1.91%). Islam and Shaikh (1967) found the infection rate with *Heterakis gallinae, Ascaridia galli, Capillaria columbae* 85%, 50%, 10% and 20% respectively. The highest rate of infection was found in 60 to 90 days (2 to 3 months) of age group fowls. Similar findings have reported by Sarker *et al.* (2009), Gauly *et al.* (2005), Romanenko *et al.* (1985).

In aboard more or less similar prevalence of Gastro-intestinal nematodiasis in fowls have also been recorded which was supported by Hussen *et al.* (2012) , Rayyan *et al.* (2010), Nnadi and George (2010), Mukaratirwa and Khumalo (2010), Matur *et al.* (2010), Kose *et al.* (2009), Pinckey *et al.* (2008), Phiri *et al.* (2007), Martin *et al.* (2005), Irungu *et al.* (2004), Permin *et al.* (2002), Eshetu *et al.* (2001), Poulsen *et al.* (2000), Permin *et al.* (1999), Hemalatha *et al.* (1987), Samad and Rahman (1985), Hayat and Hayat (1983), Velichkin *et al.* (1977), Kaushik and Deorain (1968), and Durriani and chauhan (1965).

From the above study it was evident that management and age factors played an important role in ascariasis in chicken. This is why control program should be given priority.

5.2 Comparative efficacy of Pipervet® (Piperazine) and Pineapple leaves on Ascariasis in indigenous poultry

5.2.1 Clinical parameters

(a) Severity of infestation i.e., number of egg of parasites

In this study, orall administration of piperazine citrate at recommended dose (200mg/kg bwt) was found to be 100% effective within 14 days of treatment against ascariasis in indigenous chicken. The efficacy was assessed on the basis of reduction of fecal egg count. More or less similar findings were observed by Jacques *et al.* (2012), Begum *et al.* (2010), Hafiz and Bhattacharyya (2009), Hoque *et al.* (2006), Maqbool *et al.* (1995), Verma *et al.* (1991), Nilson and Alderin (1988) and Sinha *et al.* (1987) who reported 90%-100% efficacy of piperazine citrate against ascariasis in chickens.

Oral administration of Pineapple leaves extract at recommended dose (1gm/kg bwt) was found to be about 53% effective within 14 days of treatment and 80% effective within 28 days of treatment. These results are in agreement with earlier reports of Patra *et al.* (2010), Amin *et al.* (2009), Sujon *et al.* (2008), Islam *et al.* (2005), Khalid *et al.* (2005) and Khatun *et al.* (1995).

On comparing the efficacy of piperazine citrate and pineapple leaves extract on the basis of reduction of fecal egg count of *ascaridia spp*, piperazine citrate was found to be best drug and was 100% effective within 14 days of oral administration. On the other hand, the efficacy of Pineapple leaves extract was slow which was upto 80% on 28th day of treatment. The better efficacy of piperazine citrate than pineapple leaves extract was also reported by Begum *et al.* (2010), Hoque *et al.* (2006) and Islam *et al.* (2004).

(b) Effect on feeding efficiency

The oral administration of recommended doses of Pineapple leaves extract (1gm/kg bwt) and Pipervet® (200mg/kg bwt) were given to chicken of group C and B respectively and group A was kept untreated and control. The anthelmintics significantly increased feeding efficiency of the chickens of group B and C after drug administration. But feeding efficiency was decreased gradually in chickens of control group A.

(c) Effect on feather coat

The feather coats of all treated chickens with Piperazine citrate and Pineapple leaves extract were observed smooth and shiny at the end of post-treatment period. Rough and discoloured feather coats were due to severe parasitic infestation.

(d) Effect on body weight

The oral administration of recommended doses of Pipervet® (200mg/kg body weight) and Pineapple leaves extract (1gm/kg body weight) were given to chicken of group B and C respectively and group A was kept untreated and control. The anthelmintics significantly increased the percentage of mean body weight of chicken of groups B and C as 4.19% and 3.74% respectively on 28^{th} day after treatment. But percentage of body weight gain of the control group was very negligible as 0.89% on 28^{th} day. In chicken of group B and C, body weight gain might be due to proper absorption and metabolism of feed nutrients. Because at that time the chicken of group B and C were free from parasites. But in the chicken of group A, body weight was not increased significantly because they were suffering from parasitic infestation. Parasites interfere with absorption of feed nutrients. As a result, the body weight of chicken of group A was decreased gradually. The present findings support the

earlier observation of Hoque. M. E *et al.* (2006), Khalid *et al.* (2005), Islam *et al.* (2005), Islam *et al.* (2004), Khatun *et al.* (1995).

(e) Post mortem examination

Reduction of parasite count was found on 14th and 28th day in the group of chicken of B and C. Number of parasites was found to be '0' on 14th day in group B after Pipervet® administration. Pineapple leaves extract as anthelmintics decreased the number of parasites in group C on day 14th and 28th. The highest reduction of number of parasites was recorded on 28th day of treatment. On the other hand, number of parasites were increased day by day in control group A. This findings support the earlier observation made by Gauly *et al.* (2005), Malakhov (1988).

5.2.2 Hematological parameters

The administration of recommended doses of Pipervet® (200mg/kg bwt) and Pineapple leaves extract (1gm/kg bwt) increased the TEC, Hb and PCV of chicken of group B and C respectively. On the other hand, there were reduction of TLC and ESR values in the group B and C. But in control group A, TEC, Hb and PCV were decreased and TLC and ESR values were increased. In case of group B and C, TEC, Hb and PCV were increased due to partial or fully destruction of parasites by anthelmentic action. But in the group A, TEC, Hb and PCV were increased due to effect of parasitic infestation on Hematopoitic system.

Changes in hematological parameters were also reported by other researchers such as Hoque *et al.* (2006), Islam *et al.* (2005) and Khatun *et al.* (1995).

CHAPTER VI

SUMMARY AND CONCLUSION

CHAPTER VI

SUMMARY AND CONCLUSION

This experiment was carried out in the laboratory, Department of Physiology and Pharmacology, Hajee Momahmmad Danesh Science and Technology University , Dinajpur during the period from July/2012 to November /2012, to determine the proportional incidence of ascariasis in chicken of different villages of Palashbari Upazila of Gaibandha district and the comparative efficacy of Piperazine citrate and Pineapple leaves extract against ascariasis in indigenous poultry mainly on the basis of presence or absence of egg of Ascarid parasites on feces of chicken. Attempt were also made to study the effects of Piperazine citrate and Pineapple leaves extract on some clinical (severity of infestation, feeding efficiency and body weight) and hematological (TEC, Hb, PCV, ESR and TLC) parameters in chicken.

Fecal samples were examined to observe the incidence of ascariasisin chicken and average infection rate was found 84%. The rate of infection was studied in various age groups i.e. 60 to 90days, 91 to 150 days and 150 to 210 days of age group. The highest infection rate 95.26% was found in 60 to 90 days of age group.

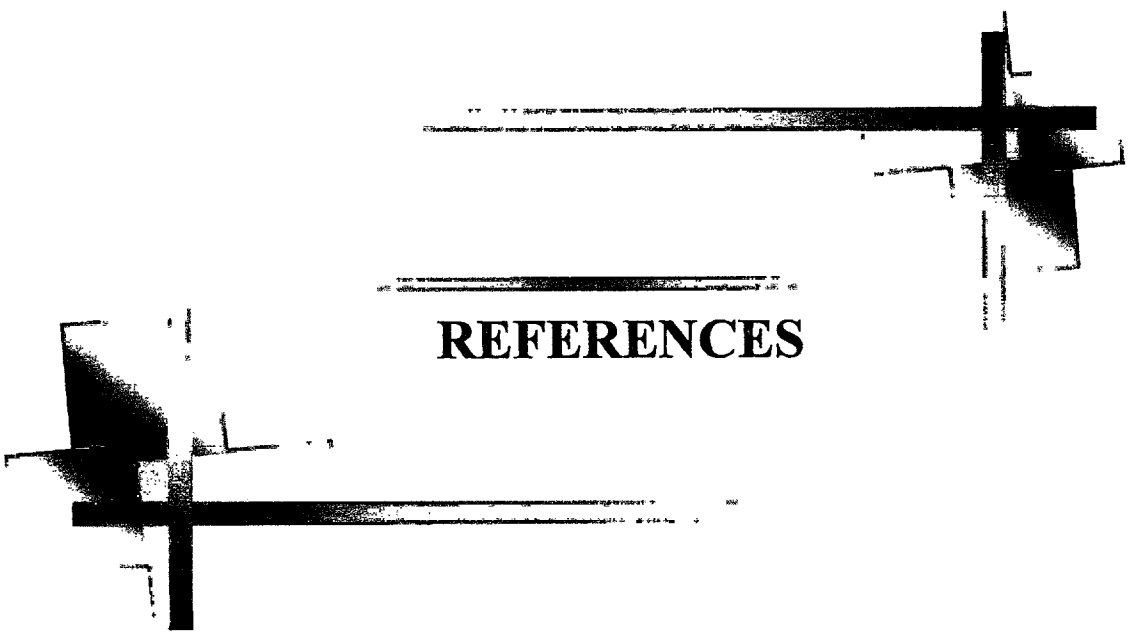
A total 60 of chickens were examined for the presence of Ascarid parasites by feces examination. Out of 60 chickens, 45 chickens having ascarid positive were randomly divided into 3 equal groups (group A, B and C), each group consisting of 15 chickens.

The chickens of group B and C were treated with recommended (200mg/kg bwt) dose of Piperazine citrate and recommended (1gm/kg bwt) dose of Pineapple leaves extract orally respectively. The chickens of group A were kept for control (untreated).

On the basis of fecal egg count, the efficacy of Pineapple leaves extract was 80% after 28 days of treatment whereas Piperazine was 100% effective after 14 days of treatment against ascariasis in chickens. After treatment, both group of treated and also control groups of chickens were kept for 28 days and clinical and hematological parameters were investigated at 7 days interval ('0' day, 7th day, 14th day, 21st day and 28th day of treatment). Number of parasites were counted after post mortem examination.

In this study, the hematological parameters i.e., TEC, Hb and PVC were increased in chickens after treatment with Piperazine citrate (200mg/kg bwt) and Pineapple leaves extract (1gm/kg bwt), whereas ESR and TLC values were decreased slightly in all treated groups. But TEC, Hb and PVC were decreased and ESR and TLC were increased in chickens of group A.

Among Piperazine citrate and Pineapple leaves extract, Piperazine citrate was found to be best drug against ascariasis in poultry showing 100% efficacy. Pineapple leaves also showed highly encouraging efficacy as an alternative anthelmintics. But piperazine citrate is not cost effective and available for rural people. From this point of view, pineapple leaves is considering more suitable one anthelmintic for the treatment of ascariasis in indigenous poultry to rural people. But this study is preliminary one considering small population of chicken. So further research works must be carried out to needed to evaluate the adverse effects, bio-chemical analysis of Pineapple leaves against ascariasis in chickens.



REFERENCES

REFERENCES

Ahmed, S. and Islam, N. (1985). Backyard poultry development project in 100 villages. Proceeding of the 1st Conference of Bangladesh Animal Husbandry Association. 23-24 February 1985, *Bangladesh Agricultural Research Council*, Dhaka, Bangladesh.

Akhtar, H. (1987). Gastro-intestinal nematodes from domestic chicken. *Bangladesh Journal of Zoology*. **15(2):** 155-159.

Akhtar, M. S.; Iqbal, Z.; Khan, M. N. and Lateef, M. (2000). Anthelmintic activity of medicinal plants with particular reference to their use in animals in the Indo-Pakistan subcontinent. *Small Ruminant Research.* **38 (2) :** 99-107.

Amin, M. R.; Mostofa, M.; Awal, M. A. and Sultana, M. A. (2008). Effects of garlic, turmeric and betel leaf against gastrointestinal nematodes in cattle. *Bangladesh Journal of Veterinary Medicine.* **6 (1)**: 115–119.

Amin, M. R.; Mostofa, M.; Hoque, M. E. and Sayed, M. A. (2009). In vitro anthelmintic efficacy of some indigenous medicinal plants against gastrointestinal nematodes of cattle. *Journal of Bangladesh Agriculture University.* **7(1) :** 57–61.

Barger, I. A. (1982). Helminth parasites and animal production. In: Symons, L.E.A., A.D. Donald and J.K. Dineen (Eds), Biology and control of endoparasites. *Academic press, Sydney*. pp : **133-155.**

BBS, (1995). Statistics Division. Ministry of Planning. Government of the Peoples Republic of Bangladesh.

Begum, S.; Mostofa, M.; Alam, A. K. M. R.; Hossain, M. F.; Barman, V. G.; Ali, A. A. M. and Mian, R. (2010). Comparative efficacy of leaves extract of neem and bishkatali with patent drugs piperazine and levamisole against ascariasis of indigenous chicken. *International Journal of BioResearch* . **1 (5) :** 17-20.

Begum, S.; Mostofa, M.; Alam, A. K. M. R.; Tanjim, M.; Ali, A. A. M.; Islam, M. N. and Das, S. (2010). Prevalence of ascariasis and comparitave efficacy of pinapple leaves extract with patent drug piperazine against ascariasis of poultry at five villages under mymensingh district. *International Journal of BioResearch.* **1 (5) :** 41-44.

Bose, S. K. (1972). Incidence of Ascaridia galli in hen's egg. *OrissaVeterinary Journal.* **7(1):** 33-34

Busa, V. and Hernandez, N. L. (1970). Ecology Biology and Epizootiology of helminths of the domestis fowl. *Revista Cubana de ciencias veterimarias.* **1:** 41-59. *Heminthological Abatract* vol. 41(9). Abstract No.2827.

Coffin, D. L. (1955). Manual of Vterinary Clinical Pathology (3rd edn). *Comstock Publishing Association,* Inc. New York, USA.

Durrani, M. Z. and Chauhan, A. H. (1965). Data on the incidence of intestinal parasites of poultry in Lyalpur district .*16th All Pakistan Science Conference.* Page A-68.

Dutta, P. K. and Hazarica, R. N. (1976). Chemotherapy of stephenofilerial dermatitis in bovines. *Indian Veterinary Journal.* **53** : 221-224.

Eshetu, Y.; Mulualem, E.; Ibrahim, H.; Berhanu, A. and Aberra, K. (2001). Study of gastro-intestinal helminths of scavenging chickens in four rural districts of Amhara region,Ethiopia. *Revue Scientifique Et Technique-Office International Des Epizooties.* **20 (3) :** 791-796.

FAO, (1974). Pakistan Livestock Sector Survey Report No. 32/74/Pak/7.FAO/World Bank Cooperative programme. **P: 11.**

Gaulya, M.; Homannb, T. and Erhardt, G. (2005). Age-related differences of Ascaridia galli egg output and worm burden in chickens following a single dose infection. *Veterinary Parasitology.* **128(1-2) :** 141-148.

Githiori, J. B.; Hoglund, J.; Waller, P. J. and Baker, R. L.(2004). Evaluation of anthelmintic properties of some plants used as livestock dewormers against Haemonchus contortus infections in sheep. *Parasitology.* **129(Pt 2) :** 245-53.

Hafiz, A. and Bhattacharyya, H. K.; (2009). Comparative efficacy of piperazine and ivermectin against ascariasis in chickens. *Indian Journal of Animal Research.* **43 (3) :** 213-214.

Haider, S. S.; Shaikh, A. L. L.; Saeed, M. M. and Khan, T.M. (1980). A study on the helminth parasites of broiler chickens in and around Lahore. *Pakistan Journal of Science.* **32(1-2):** 44,46.

Hannan, A. S. M. A.; Mostofa, M.; Haque, M. A. and Alim, M. A. (2001). Efficacy of ivomec® pour on against gastrointestinal nematodes, lice and ticks in goats. *Bangladesh Veterinary Journal.* **18 :** 95-98.

Haq, M. S. (1986). Studies on helminthes infections of chicken under rural condition of Bangladesh. *Bangladesh Veterinary Journal.* **20 (2-4) :** 55-60.

Hassain, A. (1967). Comparative efficacy of modern anthelmintves against intestinal nematodiasis in cattle with that of neem seed (Azadiarehta India). M.Sc. (Vet. Sc.) Thesis, Dept. of Pharmacology, Bangladesh Agriculture University, Mymensingh.

Hassouni, T. and Belghyti, D. (2006) . Distribution of gastrointestinal helminths in chicken farms in the Gharb region—Morocco. *Parasitol Research.* **99 (2) :** 181-3.

Hayat, B. and Hayat, C. S. (1983). Incidence of intestinal parasites of chicken in Faisalalad district. *Pakistan Veterinary Journal* . **3(4):** 165-167.

Hemalatha, E. A.; Rahman, S. A. and Jagannath, M. A. (1987). Helminthic infection in domestic fowls reared on sheep litter and cage system. *Mysore Journal of Agricultural Science.* **21 (3):** 338-341. *Poultry Abstract.* Vol. 15(1) Abstract No. 207, P. 25.

Holmes, P. H. (1986). Pathogenesis of trichostrongylosis. *Veterinary Parasitol.* **18:** 89-101.

Hoque, M. E.; Mostofa, M.; Awal, M. A.; Choudhury, M. E.; Hossain, M. A. and Alam, M. A. (2006). Comparative efficacy of piperazine citrate, levamisole and pineapple leaves extract against naturally infected ascariasis in indigenous chickens. *Bangladesh Journal of Veterinary Medicine.* **4 (1) :** 27–29.

Islam, K. R.; Farjana, T.; Begum, N. and Mondal, M. M. H. (2008). In vitro efficacy of some indigenous plants on the inhibition of Development of eggs of ascaridia galli (digenia: nematoda). *Bangladesh Journal of Veterinary Medicine.* **6 (2) :** 159–167.

Islam, S. A.; Mostofa, M.; Awal, M. A. and Khan, K. A.(2004) . Efficacy of pineapple leaves extract compared with modern anthelmintics against ascariasis in calves. *The Bangladesh Veterinarian.* **21(1):** 9-13.

Islam, S.A.; Rahman, M. M.; Hossain, M. A.; Chowdhury, M. G. A. and Mostafa, M. (2005). Comparative efficacy of some modern anthelmintics and pineapple leaves with their effects on certain blood parameters and body weight gain in calves infected with ascarid parasites. *Bangladesh Journal of Veterinary Medicine.* **3 (1) :** 33-37.

Jacques, D. T.; Aléodjrodo, E. P.; Napoléon, A. M.; Pamphile, T. and Issaka, Y. (2012). Comparative effects of piperazine of citrate and papaya seeds on Ascaridia galli in layers harco. *International Journal of Biosciences.* **2 (2) :** 72-76.

Kamal, A. H. M. (1989). Pathological investigation on the mortality of chickens in Bangladesh Agricultural University Poultry Farm. M.Sc. (Vet. science) Thesis, Department of Pathology, Bangladesh Agricultural University, Mymensingh.

Katoch, R.; Anish Yadav; Godara, R.; Khajuria, J. K.; Borkataki, S. and Sodhi, S. S. (2012). Prevalence and impact of gastrointestinal helminths on body weight gain in backyard chickens in subtropical and humid zone of Jammu, India. *Journal Of Parasitic Diseases.* **36 (1) :** 49-52.

Kaushik, R. K. and Decoranj, V. P. S. (1968). Studies on helminth infection of poultry in Uttar Pradesh in India. *Ceylon Veterinary journal.* **16(2):** 56-58.

Khalid, S. M. A.; Amin, M. R.; Mostofa, M.; Choudhury, M. E. and Uddin, B. (2005). Effects of indigenous medicinal plants (neem and pineapple) against gastrointestinal nematodiasis in sheep. *International Journal of Pharmacology.* **1 (2) :** 185-189.

Khatun, M.; Awal, M. A.; Mostofa, M. and Rashid, M. S. H. (1995). Comparative efficacy of pineapple leaves with Fenbendazole against gastro-intestinal nematodes in goats. *Bangladesh Veterinary Journal.* **29:** 75-78.

Kilpinen, O.; Roepstorff, A.; Permin, A.; Norgaard, N. G.; Lawson, L. G. and Simonsen, H. B. (2005). Influence of Dermanyssus gallinae and Ascaridia galli infections on behavior and health of laying hens (Gallus gallus domesticus). *British poultry Science.* **46(1) :** 26-34.

Kose, M.; Sevimli , F. K.; Kozan, E. K. And Çiçek, H. S. (2009). Prevalence of Gastrointestinal Helminths in Chickens in Afyonkarahisar District, Turkey. *Kafkas Univ Vet Fak Derg.* **15 (3) :** 411-416.

Malakhov, Av.; Korchagin, A. I.; Pevneva, V.D. and Nadykto, M.V.;(1988). Tolerance of Ascaridia galli to piperazine. *Veterinariya,* Moscow, No. **7,** 38-39.

Mannan, M. A.; Rafiq, K.; Mostofa, M. and Hason, Q. (1997). Comparative efficacy of Ivomec pour on, Neguvon Ointment and Neem-tobacco herbal preparation against naturally occurring hampsore Lesion in cattle. *Bangladesh Veterinary Journal.* **31 (3-4):** 119-122.

Maqbool, A.; Arshad, M. J.; Qaiser, S. and Hashmi, H. A. (1995). Incidence and chemotherapy of Ascaridia galli infection in chickens. *Indian Veterinary Journal.* **72(11):** 1145-1147.

Martin-Pacho, J. R.; Montoya, M. N.; Aranguena, T.; Toro, C.; Morchón, R.; Marcos-Atxutegi, C. and Simon, F. (2005). A coprological and serological survey for the prevalence of Ascaridia spp. in laying hens. *Journal of Veterinary Medicine. B, Infecioust Disease and Veterinary Public Health.* **52(5)** : 238-42.

Matur, B. M.; Dawam, N. N.; and Malann, Y. D. (2010). Gastrointestinal Helmith Parasites of Local and Exotic Chickens Slaughtered in Gwagwalada, Abuja (FCT), Nigeria. *New York Science Journal.* **3 (5).**

Mondal, M. M. H. and Qadir, A. N. M. A. (1991). Some epidemiological aspects of ascarids infection in chicken under the existing situation of Bangladesh Agricultural University Poultry Farm. *Bangladesh Agricultural University Research Progress* **5:** 332-336.

Mostofa, M. (1983). Efficacy of some indigenous medicinal plants against gastrointestinal nematodiasis in cattle and their comparative activity with that of Nemafax. M.Sc (Vet. Sc.) Thesis. Department of Pharmacology, BAU.

Mukaratirwa, S. and Khumalo, M. P. (2010). Prevalence of helminth parasites in free-range chickens from selected rural communities in KwaZulu-Natal province of South Africa. *Journal of the South African Veterinary Association.* **81(2) :** 97-101.

Mushi, E. Z.; Binta, M. G.; Chabo, R. G.; Ndebele, R. and Thibanyane, T. (2000). Helminth parasites of indigenous chickens in Oodi, Kgatleng District, Botswana. *Journal of the South African Veterinary Association.* **71 (4) :** 247-8.

Nath, K.; Rajagopal and Garg, A. K. (1983). Washed neem seed kernel cake as a cattle feed. *The Journal of Agricultural Science.* **101 (2): 323-326.**

Nilson, O. and Alderin, A.(1988). Efficacy of Piperazine dihydrochloride against Ascaridia galli in the domestic fowl. *Avian pathology.* **17** : 495-500.

Nnadi, P. A. and George, S. O. (2010). A cross-sectional survey on parasites of chickens in selected Villages in the subhumid zones of south-eastern Nigeria. *Journal of Parasitology Research.* **4(1):** 1-6

Ojok, L. (1993). Diseases as important factor affecting increased poultry production in Uganda. *Der Tropenland Wirt Zeitschrift fur die landwirtschaft in din Tropen and Subtropen.* **94 :** 37-44.

Patra, G.; Lyngdoh, W. M.; Ali, M. A.; Prava, M. and Chanu, Kh. V. *et al.* (2010). Comparative Anthelmintic Efficacy of Pineapple and Neem Leaves in Broiler Chickens Experimentally Infected with Ascaridia galli. *International Journal of Poultry Science.* **9 (12) :** 1120-1124.

Permin, A.; Bisgaard, M.; Frandsen, F.; Pearman, M.; Kold, J. and Nansen, P. (1999). Prevalence of gastrointestinal helminths in different poultry production systems. *British Poultry Science.* **40 (4):** 439-43.

Phiri, I. K.; Phiri, A. M.; Ziela, M.; Chota, A.; Masuku, M. and Monrad, J. (2007). Prevalence and distribution of gastrointestinal helminths and their effects on weight gain in free-range chickens in Central Zambia. *Tropical Animal Health Production.* **39 (4):**309-15.

Pinckney, R. D.; Coomansingh, C.; Bhaiyat, M. I.; Chikweto, A.; Sharma, R. and Macpherson, C. N. L. (2008). Prevalence of gastrointestinal parasites in Grenada, West Indies. *West Indian Veterinay Journal.* **8 (1):** 23-26.

Poulsen, J.; Permin, A.; Hindsbo, O.; Yelifari, L.; Nansen, P. and Bloch, P. (2000). Prevalence and distribution of gastro-intestinal helminths and haemoparasites in young scavenging chickens in upper eastern region of Ghana, West Africa. *Preventive Veterinary Medicine.* **45 (3-4) :** 237-45.

Rabbi, A. K. M. A.; Islam, A.; Majumder , S.; Anisuzzaman and Rahman, M. H. (2006). Gastrointestinal helminths infection in different types of poultry. *Bangladesh Journal of Veterinary Medicine.* **4 (1):** 13–18.

Rahman, M. H.; Ahmed, S. and Mondol, M. M. H. (1996). Introduction to Helminth Parasites of Animals and Birds in Bangladesh. RS. Tahsina Mostofa (1st edn.). pp. 16, 18, 105-106

Rayyan, A.; Al– Hindi, A. and Al–Zain, B. (2010). Occurrence of gastrointestinal helminthes in commercial and free-range chickens in gaza strip, Palestine. *Egypt Poultry Science.* **30 (II):** 601-606.

Romanenko, P. T.; Troenko, T. A.; Zaremba, I. A. and Kuzyakin, A. V. (1985). Age variation in helminth infection on chicken's farm and factory faems in the Roostov region. *Poultry Abatract.* **11(10):** 249-250.

Samad, M. A. and Rahman, A. (1985). Incidence of gastro-intestinal parasitic infection in domestic fowls of Bangladesh. *Poultry Adviser.* **18(10) :** 33-38.

Sarkar, A. J. (1976). The prevalence of avian diseases in Bangladesh Agricultural University Farm. *Bangladesh Veterinary Journal.* **10:** 61-66.

Sarker, R. R., Mostofa, M.; Awal, M. A.; Islam, M. S. and Mian, R. (2009). Comparative efficacy of the selected indigenous medicinal plants with a patent drug levamisole against ascariasis in village poultry. *Bangladesh Journal of Veterinary Medicine.* **7 (2) :** 320 – 324.

Sarker, R. R.; Mostofa, M.; Awal, M. A.; Islam, M. S. and Mamun, S. A. (2009). Prevalence of ascariasis in village poultry at five upazilas under feni District. *Bangladesh Journal of Veterinary Medicine.* **7 (1) :** 293 – 295.