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## Incidence of Gastro-Intestinal Nematodes of Cattle in Rajshahi and Anthelmintic Efficacy of Medicinal Plants and Patent Drugs against Them

Sultana, Mst. Rokeya

University of Rajshahi

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### INCIDENCE OF GASTRO-INTESTINAL NEMATODES OF CATTLE IN RAJSHAHI AND ANTHELMINTIC EFFICACY OF MEDICINAL PLANTS AND PATENT DRUGS AGAINST THEM



## THESIS SUBMITTED FOR THE DEGREE OF DOCTOR OF PHILOSOPHY (Ph. D.) IN THE DEPARTMENT OF ANIMAL HUSBANDRY AND VETERINARY SCIENCE UNIVERSITY OF RAJSHAHI, RAJSHAHI-6205 BANGLADESH

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# Dedicated To My Beloved Parents

#### **DECLARATION**

I hereby declare that the thesis entitled "Incidence of gastro-intestinal nematodes of cattle in Rajshahi and anthelmintic efficacy of medicinal plants and patent drugs against them" is the results of my own and original investigation to the Department of Animal Husbandry and Veterinary Science, Faculty of Agriculture, University of Rajshahi, Bangladesh under the Supervision of Professor Dr. Md. Jalal Uddin Sarder, Department of Animal Husbandry and Veterinary Science, University of Rajshahi and Professor Dr. Mahbub Mostofa, Co-supervisor, Department of Pharmacology, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh in fulfillment of the requirement for the degree of Doctor of Philosophy.

I further declare that this research work has not been submitted in part or in full previously for any academic degree in this university or any university.

April, 2014

(Mst. Rokeya Sultana)

Candidate

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The Author

#### **ABSTRACT**

Sultana M. R. 2014. Ph.D thesis. Incidence of gastro-intestinal nematodes of cattle in Rajshahi and anthelmintic efficacy of medicinal plants and patent drugs against them, Department of Animal Husbandry and Veterinary Science, University of Rajshahi, Bangladesh page 1-327.

The experiment was performed in the Department of Animal Husbandry & Veterinary Science, Faculty of Agriculture and University of Rajshahi in collaboration with the Department of Pharmacology, Physiology, and Parasitology, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh. The prevalence of gastro-intestinal nematodes was observed in 400 randomly selected cattle during the period from June, 2011 to May, 2012 in greater Rajshahi in Bangladesh. The prevalence of Ascaris sp., Strongyles (Haemonchus sp., Trichostrongylus sp., Oesophagostomum sp. and Mecistocirrus sp.), Bunostomum sp., Trichuris sp. Strongyloides sp. and mixed infection were 6.5%, 25.5%, 2.25%, 5% 5% and 10.5%, respectively. The seasonal prevalence of gastro-intestinal nematodes was significantly highest in rainy seasons (June-August) 79.8% followed by autumn (September-November) 57.7%, summer (March-May) 44.3% and winter seasons (December-February) 34.8%. The incidence of gastro-intestinal nematodes was highest in Rajshahi district followed by Natore district, Naogaon district and Chapai Nawabgoni district and the intradistrict prevalence of gastro-intestinal nematodes in cattle was ranged from 38-62.5%. The incidence of gastro-intestinal nematodes in cattle was highest 134 (67.0%) in 12 to <24 months old cattle and lowest 22 (36.7%) in 1 to < 6 months old cattle and highest incidence was also observed in female. The incidence of gastro-intestinal nematodes was more in cross breed than local breed. The incidence of gastro-intestinal nematodes was highest in L×F genotype followed by in L×F×SL genotype, L×SL genotype and local breed. The incidence of gastrointestinal nematodes was highest in the month of July. Screening of 10 indigenous medicinal plants (Neem, custard apple, papaya, garlic, bitter gourd, jute, chaste tree, pineapple, pomegranate and betel leaf) of Bangladesh having reported anthelmintic activity and to determine the comparative efficacy in-vitro. Within these 10 plants, 3 showed 100%, 2 showed 90% and others showed 60-70% efficacy in water extract at the concentration of 100 mg/ml against adult worms. On the other hand, 5 showed 100% efficacy, 3 showed 90% and others showed 80% efficacy in ethanol extract at the concentration of 100 mg/ml among these 10 plants against adult worms. Ethanol extracts were more effective than water extracts against adult worms. Among 10 tested plants 2 medicinal plants (neem and korolla) and 2 patent drugs (Ivertin® and Levavet® were administered against gastro-intestinal nematodes in cattle for in vivo trail. Twenty five (25) naturally parasitized female cattle of Binodpur, Rajshahi, were selected for efficacy study of 2 indigenous medicinal plants and 2 patent drugs in-vivo. The animals were divided into five (5) groups (A, B, C, D, and E), each group consisting of five (5) cattle. Ethanol extract of leaves of neem @100 mg/kg b. wt and bitter gourd @100 mg/kg b. wt were administered orally to the cattle of group B and C respectively. Cattle of group D and E were treated with ivermectin (Ivertin®) @ 0.2 mg/kg body weight pour on and levamisole (Levavet®) @ 7.5 mg/kg b. wt. respectively. Cattle of group A was kept as infected control group. Prior to trials (day 0), initial body weight, total egg count of gastro-intestinal nematodes, biochemical, hematological and clinical parameters were examined and recorded. During the experimental period, faecal samples were examined on 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day. Clinical (body weight), haematological (TEC, Hb, PCV, TLC, DLC, MCV, MCHC and MCH) and biochemical parameters (ALT and AST) were also observed on 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day for the determination of effects of neem, bitter gourd, Ivertin® and Levavet®. In-vivo neem showed highest egg reduction among tested plants and ivermectin pour on showed 100% egg reduction. Anthelmintic treatment showed increased values than control for the TEC, Hb, PCV, neutrophil count of DLC, MCHC and MCH, decreased values for TLC and eosinophil count of DLC, very little changed values for monocyte count and lymphocyte count of DLC and no change was found in ALT and AST in cattle. G.I. nematodiasis is a great problem in Bangladesh especially in greater Rajshahi and the data generated from this study revealed that Ivermectin is drug of choice, levamisole is a good drug and as an alternative, farmers could use neem and bitter gourd with good therapeutic value.

#### **ABBREVIATIONS**

% = Percentage

& = and

/ = Per

< = Less than

> = Greater than

 $\pm$  = Plus-minus

° = Degree

 $\mu$  = Micron (s)

 $\mu g = Microgram$ 

ANOVA = Analysis of variance

b. wt. = Body weight

BAU = Bangladesh Agricultural University

BBS = Bangladesh Bureau of Statistics

C = Celsius

cm = Centimeter(s)

d = Day(s)

DF = Degree of freedom

DLS = Department of Livestock Services

DMRT = Duncan's Multiple Range Test

e.g. = For example

EPG = Eggs per gram

et.al. = Associate

Etc. = Et cetera (L) and other

F = Friesian

FAO = Food and Agriculture Organization

Fig. = Figure

FY = Fiscal Year

g = Gram(s)

GDP = Gross Domestic Product

gm = Gram(s)

Hb = Haemoglobin

i.e. = That is

i.u = International unit

Id = Identification

j. = Journal

Kg = Kilogram(s)

L = Local

m = Month(s)

M.S. = Master of Science

mg = Milligram(s)

mill. = Million

min = Minutes(s)
ml = Millilitre
mmol = Millimole

n or N = Number of observation

nmol = Nanomole no. = Number

PBS = Phosphate Buffered Saline

PCV = Packed Cell Volume

pp = Page

RBC = Red Blood Cell
SE = Standard Error

Sig. = Significance

SL = Shahiwal

SPSS = Statistical package for the social sciences

USA = United States of America

WBC = White Blood Cell

yrs = Years

#### **CONTENTS**

	Page No.
Acknowledgement	i-ii
Abstract	iii
Abbreviation	iv-v
Contents	vi-xxii
CHAPTER-1: INTRODUCTION	1-13
CHAPTER-2: REVIEW OF LITERATURE	14-94
2.1 Prevalence of Gastro-intestinal Nematodes	14
2.2 Anthelmintic Effects of Indigenous Medicinal Plants	55
2.3 Anthelmintic Effects of Ivermectin	71
2.4 Anthelmintic Effects of Levamisole	84
CHAPTER-3: MATERIALS AND METHODS	95-164
3.1 Collection and Examination of Faecal Samples	95
3.1.1 Selection of animals	95
3.1.2 Procedure of feces examinations	96
3.2 Collection and Identification of Indigenous Medicinal Pla	ints having
Anthelmintic Activity	100
3.3 Description of Indigenous Medicinal Plants	100
3.3.1 Local name: Neem	100
3.3.2 Local name: Ata, Nona	102
3.3.3 Local name: Papa	104
3.3.4 Local name: Rashun	105
3.3.5 Local name: Korolla	108
3.3.6 Local name: Pat	109
3.3.7 Local name: Nishinda	111

3.3.8 Local name: Anaros	112
3.3.9 Local name: Dalim	113
3.3.10 Local name: Pan	115
3.4 Preparation of Plants Powder and Extract	117
3.4.1 Processing of plant materials	117
3.4.2 Preparation of plants powder	114
3.4.3 Preparation of plants extract	122
3.4.4 Preparation of stock solution	127
3.5 Collection and Maintenance of Adult Gastro-intestinal Nematod	les in
the Laboratory	127
3.6 <i>In vitro</i> Screening of Plant Extracts for Anthelmintic Activity	128
3.7 Selection of Research Site for <i>in-vivo</i> Trials	139
3.8 Selection of Cattle for <i>in-vivo</i> Trials	139
3.9 Collection of Two Indigenous Medicinal Plant and Drugs for in	vivo
Experiment	140
3.9.1 The selected medicinal plants	140
3.9.2 The patent drugs	141
3.9.2.1 <i>Ivermectin</i>	142
3.9.2.2 <i>Levamisole</i>	145
3.10 Experimental Design for <i>in vivo</i> Trials	146
3.11 The Test Parasites	148
3.12 Faecal Sample Examination	148
3.13 Determination of Efficacy of Different Treatment	150
3.14 Determination of Haematological Parameters	151
3.14.1 Total erythrocyte count (TEC)	151
3.14.2 Haemoglobin (Hb) content	152
3.14.3 Packed cell volume (PCV)	153

3.14.4 Total leukocyte count (TLC)
3.14.5 Differential leukocyte count (DLC)155
3.14.6 Mean corpuscular volume (MCV)156
3.14.7 Mean corpuscular haemoglobin concentration (MCHC)156
3.14.8 Mean corpuscular haemoglobin (MCH)156
3.15 Determination of Biochemical Parameters157
3.15.1 Determination of alanine aminotransferase (ALT) level157
3.15.2 Determination of aspartate aminotransferase (AST) level158
3.16 Measurement of Body Weight160
3.17 Statistical Analysis
CHAPTER 4: RESULTS165-223
4.1 Incidence of Gastro-intestinal Nematodiasis in Cattle
4.1.1 The Overall incidence of gastro-intestinal nematode parasites of
400 cattle in greater Rajshahi166
4.1.2 Average incidence of gastro-intestinal nematodes in relation with
age of cattle in greater Rajshahi167
4.1.3 Incidence of gastro-intestinal nematodes species in relation with
age of cattle in greater Rajshahi168
4.1.4 Incidence of gastro-intestinal nematodes with the sex of cattle in
greater Rajshahi170
4.1.5 Effect of breed on incidence of gastro-intestinal nematodes of
cattle in greater Rajshahi171
4.1.6 Incidence of gastro-intestinal nematodes in relation with genotype
of cattle in greater Rajshahi
4.1.7. Incidence of gastro-intestinal nematodes in cattle in relation
with seasons in greater Raishahi

4.2.2.2 In vitro anthelmintic efficacy of 5% (50mg/ml) concentration of
ethanol extracts of 10 indigenous medicinal plants against adult
gastro-intestinal nematodes of cattle186
4.2.2.3 In vitro anthelmintic efficacy of 10% (100 mg/ml) concentration
of ethanol extracts of 10 indigenous medicinal plants against
adult gastro-intestinal nematodes of cattle187
4.2.2.4 Comparative anthelmintic efficacy of different concentrations of
ethanol extracts of 10 indigenous medicinal plants against adult
gastro-intestinal nematodes of cattle189
4.3 Results Obtained from <i>in vivo</i> Screening
4.3.1 Efficacy of two (2) indigenous medicinal plants and two (2) patent
drugs against natural gastro-intestinal nematodes in cattle192
4.3.2. Effects of two (2) indigenous medicinal plants and two (2) patent
drugs on haematological parameters in cattle195
4.3.2.1 Total erythrocyte count (TEC, million/cu.mm.)
4.3.2.2 <i>Haemoglobin content (Hb, g %)</i> 196
4.3.2.3 Packed cell volume (PCV, %)
4.3.2.4 Total leukocyte count (TLC, thousand/cu.mm.)201
4.3.2.5. Differential leukocyte count (DLC, %)
4.3.2.5.1. <u>Eosinophil count (%)</u>
4.3.2.5.2 <u>Neutrophil count (%)</u>
4.3.2.5.3 <u>Monocyte count (%)</u>
4.3.2.5.4 <u>Lymphocyte count (%)</u>
4.3.2.6 Mean corpuscular volume (cubic micron)211

4.3.2.7 Mean corpuscular haemoglobin concentration (%)	213
4.3.2.8 Mean corpuscular haemoglobin (micro micro gram)	215
4.3.3. Effects of two (2) indigenous medicinal plants and two (2)	patent
drugs on some serum biochemical parameters in cattle	217
4.3.3.1 Alanine aminotransferase level of serum (U/I)	217
4.3.3.2 Aspartate aminotransferase level of serum (U/I)	219
4.3.4. Effects of two (2) indigenous medicinal plants and two (2)	patent
drugs on body weight (Kg) in cattle	221
CHAPTER 5: DISCUSSION	.224-247
5.1 Prevalence of Gastro-intestinal Nematodiasis in Cattle	224
5.2 Screening of Plants for <i>in vitro</i> Anthelmintic Activity	228
5.3 Efficacy of two Indigenous Medicinal Plants and two Patent	
Drugs against Natural Gastro-intestinal Nematodes in Cattle	230
5.4 Effects of two Indigenous Medicinal Plants and two Patent D	rugs
on Haematological Parameters in Cattle	233
5.4.1 Total erythrocyte count (TEC, million/cu.mm.)	233
5.4.2 Haemoglobin content (Hb, g %).	234
5.4.3 Packed cell volume (PCV, %)	236
5.4.4 Total leukocyte count (TLC, thousand/cu.mm.)	237
5.4.5 Differential leukocyte count (DLC, %)	238
5.4.5.1 <i>Eosinophil count (%)</i>	238
5. 4.5.2 <i>Neutrophil count</i> (%)	239
5.4.5.3 <i>Monocyte count</i> (%)	240
5.4.5.4 <i>Lymphocyte count</i> (%)	241
5.4.6 Mean corpuscular volume (cubic micron)	242

5.4.7 Mean corpuscular haemogtobin concentration (%)	242
5.4.8 Mean corpuscular haemoglobin (micro micro gram)	243
5.5 Effects of two Indigenous Medicinal Plants and two Patent	
Drugs on some Serum Biochemical Parameters in Cattle	244
5.5.1 Alanine aminotransferase level of serum (U/I)	244
5.5.2 Aspartate aminotransferase level of serum (U/I)	245
5.6 Effects of 2 Indigenous Medicinal Plants and 2 Patent Drug	s on Body
Weight (Kg) in Cattle	246
CHAPTER-6: SUMMARY AND CONCLUSIONS	248-254
6.1 Summary	248
6.2 Conclusions	253
CHAPTER-6: REFERENCES	255-303
APPENDICES	304-327

#### LIST OF TABLES

Table No.	Title	Page no.
Table 1:	Ten indigenous medicinal plants used for studied.	100
Table 2:	Protocol of the study for in vivo trial	148
Table 3:	Overall incidence of nematode parasites in 400 cattle in greater Rajshahi	166
Table 4:	Incidence of GI nematodes in relation with age in cattle.	167
Table 5:	Incidence of GI nematodes species in different age of cattle at study area	169
Table 6:	Incidence of gastro-intestinal nematodes in between sex of cattle in study area.	170
Table 7:	Effect of breeds on incidence of gastro-intestinal nematodes in cattle of greater Rajshahi	171
Table 8:	Incidence of gastro-intestinal nematodes in different genotype of cattle in study area.	172
Table 9:	Incidence of gastro-intestinal nematodes in cattle in greater Rajshahi among different seasons	173
Table 10:	The influence of month in a year on incidence of gastro- intestinal nematodes of cattle in greater Rajshahi	175
Table 11:	Incidence of Gastro-intestinal nematodes in different districts of greater Rajshahi	177
Table 12:	In vitro anthelmintic efficacy of 2.5 % (25 mg/ml) concentration of water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	179
Table 13:	In vitro anthelmintic efficacy of 5% (50mg/ml) concentration of water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	180
Table 14:	In vitro anthelmintic efficacy of 10% (100 mg/ml) concentration of water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	182
Table 15:	Comparative anthelmintic efficacy of different concentrations of water extracts of 10 indigenous medicinal	183

Table No.	Title	Page no.
	plants against adult gastro-intestinal nematodes of cattle	
Table 16:	<i>In vitro</i> anthelmintic efficacy of 2.5 % (25mg/ml) ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	185
Table 17:	In vitro anthelmintic efficacy of 5% (50mg/ml) ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	186
Table 18:	<i>In vitro</i> anthelmintic efficacy of 10% (100 mg/ml) ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	188
Table 19:	Comparative anthelmintic efficacy of different concentrations of ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	190
Table 20:	The effects of Neem leaves (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) against G.I nematodes in Cattle.	193
Table 21:	Analysis of variance for EPG of Cattle by the influences of different drugs during experimental periods.	194
Table 22.	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on total erythrocyte count (million/cu.mm.) in cattle.	196
Table 23:	Analysis of variance for total erythrocyte count (million/cu.mm.) in cattle by treated with various effective drugs during experimental periods	197
Table 24.	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on haemoglobin content (g %) in cattle.	198
Table 25:	Analysis of variance for haemoglobin content (g %) in cattle by the used of different drugs during experimental periods.	199
Table 26:	The effects of neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on packed cell volume (%) in cattle.	200
Table 27:	Analysis of variance for packed cell volume (%) in cattle	201

Table No.	Title	Page no.
	among the different drugs.	
Table 28:	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on total leukocyte count (thousand/cu.mm.) in cattle.	202
Table 29:	Analysis of variance for total leukocyte count (thousand/cu.mm.) in cattle by the different drugs.	203
Table 30:	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on eosinophil count (%) of differential leukocyte count in cattle.	204
Table 31:	Analysis of variance for eosinophil count (%) of differential leukocyte count in cattle treated with different drugs.	205
Table 32:	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on neutrophil count (%) of differential leukocyte count in cattle.	206
Table 33:	Analysis of variance for neutrophil count (%) of differential leukocyte count in cattle treated with different drugs	207
Table 34:	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on monocyte count (%) of differential leukocyte count in cattle.	208
Table 35:	Analysis of variance for monocyte count (%) of differential leukocyte count in cattle by the treatment with different drugs.	209
Table 36:	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on lymphocyte count (%) of differential leukocyte count in cattle.	210
Table 37:	Analysis of variance for lymphocyte count (%) of differential leukocyte count in cattle treated with different drugs.	211
Table 38:	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular volume (cubic micron) in cattle.	212

Table No.	Title	Page no.
Table 39:	Analysis of variance for mean corpuscular volume (cubic micron) in cattle by different drugs	213
Table 40:	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular haemoglobin concentration (%) in cattle.	214
Table 41:	Analysis of variance for mean corpuscular haemoglobin concentration (%) in cattle treated with different drugs.	215
Table 42:	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular hemoglobin (micro micro gram) in cattle.	216
Table 43:	Analysis of variance for mean corpuscular hemoglobin (micro micro gram) in cattle by the treatment with different drugs.	217
Table 44:	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on alanine aminotransferase level of serum (U/I) in cattle.	218
Table 45:	Analysis of variance for alanine aminotransferase level of serum (U/I) in cattle in relation with different drugs.	219
Table 46:	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on aspartate aminotransferase level of serum (U/I) in cattle.	220
Table 47:	Analysis of variance for aspartate aminotransferase level of serum (U/I) in cattle in relation with different drugs.	221
Table 48:	The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on body weight (Kg) in cattle.	222
Table 49:	Analysis of variance for body weight (Kg) in cattle in relation with different drugs in different days of treatment.	223

#### LIST OF FIGURES

Figure No.	Title	Page no.
Fig 1:	Graphical presentation of incidence of gastro-intestinal nematode parasites of cattle in greater Rajshahi	167
Fig 2:	Graphical representation of Incidence of GI nematodes in relation with age of cattle in study area	168
Fig 3:	Graph shows the influence of age on different gastro- intestinal nematodes of cattle in study area	169
Fig 4:	Graph represents the incidence of gastro-intestinal nematodes in different sex of cattle in study area	170
Fig 5:	Graph showing incidence of gastro-intestinal nematodes in different sex of cattle in greater Rajshahi	171
Fig 6:	Graph showing incidence of gastro-intestinal nematodes in different genotypes of cattle in study area	173
Fig. 7:	Graphical presentation of the effects of seasons on incidence of gastro-intestinal nematodes in study area	174
Fig. 8:	Graph shows the influence of months in a year on incidence of gastro-intestinal nematodes of cattle in greater Rajshahi	176
Fig. 9:	Graphical representation of incidence of Gastro-intestinal nematodes among the districts of greater Rajshahi	177
Fig. 10:	Graphical presentation of anthelmintic efficacy of 10 indigenous medicinal plants of 2.5% water extract against adult gastro-intestinal nematodes in cattle	179
Fig. 11:	Graphical presentation of anthelmintic efficacy of 5% (50 mg/ml) water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	181
Fig. 12:	The graph represents the anthelmintic efficacy of 10% (100mg/ml) water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	182
Fig. 13:	Graph showing the efficacy of water extract of 10 indigenous medicinal plants against gastro-intestinal nematodes of cattle in different concentration	184

Figure No.	Title	Page no.
Fig. 14:	Graphical representation of anthelmintic efficacy of 2.5% (25 mg/ml) ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	185
Fig. 15:	Graphical presentation of anthelmintic efficacy of 5% (50 mg/ml) concentration of ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	187
Fig. 16:	Graph showing anthelmintic efficacy of 10% (100 mg/ml) concentration of ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	189
Fig. 17:	Graphical representation of anthelmintic efficacy of different concentrations of ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle	190
Fig. 18:	Graph is showing the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) against G.I nematodes in Cattle.	194
Fig. 19:	The graphical representation of the effects of various effective drugs on total erythrocyte count (million/cu.mm.) in cattle.	196
Fig. 20:	Graphical representation of the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on haemoglobin content (g %) in cattle.	198
Fig. 21:	Graph showing the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on packed cell volume (%) in cattle.	200
Fig. 22:	Graph represents the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on total leukocyte count (thousand/cu.mm.) in cattle.	202
Fig. 23:	Graph shows the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on eosinophil count (%) of differential	
	leukocyte count in cattle.	204

Figure No.	Title	Page no.
Fig. 24:	Graphical presentation of the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on neutrophil count (%) of differential leukocyte count in cattle.	206
Fig. 25:	Graph represents The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on monocyte count (%) of differential leukocyte count in cattle.	208
Fig. 26:	Graph showing the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on lymphocyte count (%) of differential leukocyte count in cattle.	210
Fig. 27:	Graphical representation of the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular volume (cubic micron) in cattle.	212
Fig. 28:	Graphical presentation of the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular haemoglobin concentration (%) in cattle.	214
Fig. 29:	Graph shows the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular hemoglobin (micro micro gram) in cattle.	215
Fig. 30:	Graphical representation of the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on alanine aminotransferase level of serum (U/I) in cattle.	218
Fig. 31:	Graph represents the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on aspartate aminotransferase level of serum (U/I) in cattle.	220
Fig. 32:	Graph showing the effect of medicinal plants on body weight (kg) in cattle	222

#### LIST OF PLATES

Plate No.	Title	Page no.
Plate 1.	Egg of Haemonchus (40X)	98
Plate 2.	Egg of Bunostomum (40X)	98
Plate 3.	Egg of Oesophagostomum (40X)	98
Plate 4.	Egg of Trichuris (40X)	99
Plate 5.	Egg of Trichostronyglus (40X)	99
Plate 6.	Egg of Strongyloides (40X)	99
Plate 7.	Neem (Neem tree / Margosa tree, Azadirachta indica)	101
Plate 8.	Ata, (Custard apple, Annona reticulata)	103
Plate 9.	Papa, (Papaya / Papu / Melon tree, Carica papaya)	104
Plate 10.	Rashun, (Garlic, Allium sativum)	106
Plate 11.	Korolla, (Bitter gourd, Momordica charantia)	108
Plate 12.	Pat, Tosa, (Jute, Corchorus oletorious)	110
Plate 13.	Nishinda, (Chaste Tree, Vitex negundo)	111
Plate 14.	Anaros, (Pineapple, (Ananas comosus)	112
Plate 15.	Dalim, (Pomegranate, Punica granatum)	114
Plate 16.	Pan, (Betle Leaf, Piper betle)	116
Plate 17.	Hot-air Oven	117
Plate 18.	Small pieces of leaves in hot-air oven	117
Plate 19.	Electric Grinder	118
Plate 20.	Powder of Neem (Leaves)	118
Plate 21.	Powder of Ata (Leaves)	118
Plate 22.	Powder of Papaya (Leaves)	119

Plate No.	Title	Page no.
Plate 23.	Powder of Garlic	119
Plate 24.	Powder of Korola (Fruit)	119
Plate 25.	Powder of Jute (Leaves)	120
Plate 26.	Powder of Nishyinda (Leaves)	120
Plate 27.	Powder of Pineapple (Leaves)	120
Plate 28.	Powder of Dalim (Leaves)	121
Plate 29.	Powder of Betel (Leaves)	121
Plate 30.	Measurement of plant powder with balance and weight box	123
Plate 31.	Electric Stirrer	123
Plate 32.	Filtration of plant extract	124
Plate 33.	Neem leaves extract	124
Plate 34.	Ata leaves extract	124
Plate 35.	Papaya leaves extract	125
Plate 36.	Garlic extract	125
Plate 37.	Korolla leaves extract	125
Plate 38.	Jute leaves extract	125
Plate 39.	Nishyinda leaves extract	126
Plate 40.	Pineapple leaves extract	126
Plate 41.	Dalim leaves extract	126
Plate 42.	Betel leaves extract	126
Plate 43.	Petri-dishes containing parasites	128
Plate 44.	In-vitro trial of Neem extract	129
Plate 45.	In-vitro trial of Ata extract	130
Plate 46.	In-vitro trial of Papaya extract	131

Plate No.	Title	Page no.
Plate 47.	In-vitro trial of Garlic extract	131
Plate 48.	In-vitro trial of Korolla extract	133
Plate 49.	In-vitro trial of Jute extract	134
Plate 50.	In-vitro trial of Nishinda extract	135
Plate 51.	In-vitro trial of Anaros extract	136
Plate 52.	In-vitro trial of Dalim extract	137
Plate 53.	In-vitro trial of Pan extract	138
Plate 54.	Experimental Cattle	139
Plate 55.	Powder of Neem	140
Plate 56.	Powder of Korolla	140
Plate 57.	Extract of Neem	141
Plate 58.	Extract of Korolla	141
Plate 59.	Levamisole (Levavet)®	141
Plate 60.	Ivermectin (Ivertin)®	141
Plate 61.	Fecal sample	150
Plate 62.	Examination of fecal sample by microscope	150
Plate 63.	Treated with Ivertin® (Pour-on) in experimental cattle	150
Plate 64.	Collection of blood from the experimental cattle	158
Plate 65.	Collected blood sample	159
Plate 66.	Examination of blood sample by a compound light microscope at laboratory of BAU	159
Plate 67.	Examination of biochemical parameter by Reflotron Plus Machine	160
Plate 68.	Measurement of body weight	160
Plate 69.	In vitro trial of ethanol extract of Neem	191
Plate 70.	In vitro trial of ethanol extract of Korolla	191

#### **CHAPTER-1**

#### INTRODUCTION

Bangladesh lies in the northeastern part of south Asia. Bangladesh is a topical country and geographically it stands between latitudes 20°34' and 26°38' north and latitudes 88°01' and 92°41' east. The country is bordered by India on the east, west and north and by the Bay of Bengal on the south. There is also a small strip of frontier with Burma on the southeastern edge. The land is a deltaic plain with a network of numerous river and canals. The country consists of low, flat and fertile land except the hilly regions in north east and southeast. Bangladesh has a tropical monsoon climate characterized by wide seasonal variations in rainfall, high temperatures, and high humidity. Regional climatic differences in this flat country are minor. Three seasons are generally recognized: a hot, muggy summer from March to June; a hot, humid and rainy monsoon season from June to November; and a cold, dry winter from December to February. Average winter temperature ranges from 7°C-13°C and average summer temperature ranges from 24°C-30°C (BBS, 2010). April is the hottest month in most parts of the country. January is the coolest month, when the average temperature for most of the country is 16°C - 20°C (61°F - 68 °F) during the day and around 10 °C (50 °F) at night. The climate is one of the wettest in the world; most places receive more than 1,525 mm of rain a year, and areas near the hills receive 5,080 mm (BBS, 2010). Most rain falls during the monsoon (June-September) and little during the dry season (November-February).

Chapter 1 Introduction \( \mathre{\infty} \)

Bangladesh is a densely populated agro-based developing country with an estimated population of about 156 million, most of which are suffering from malnutrition (BBS, 2010). Protein deficiency is a major problem in this regard. About 73 percent of this population lives in rural areas (BBS, 2010). So the development of this country depends greatly on the development of rural people. The economy of Bangladesh mostly depends on agriculture. Agriculture is the economic backbone of Bangladesh with approximately 80% people who depends on it directly or indirectly for their subsistence. The livestock is an important sub-sector, which considered being the backbone of agriculture (Anonymous, 1985). Livestock being one of the major components of agriculture plays a vital and crucial role in the traditional subsistence farming and national economy. Livestock sub-sector contributed to solve the economical problems of small and marginal farmers and played important role in poverty alleviation. Among them cattle are one of the indexes of civilization and great source of wealth for any nation.

Bangladesh is a small country but it has a large number of population. The amount of land per farmer is less so it is easier for them to establish animal farm in large and small scale rather than cultivation of crop as a source of livelihood. There is a lot of demand for the livestock products and by-products throughout the country. The contribution of the livestock sub-sector to Gross Domestic Product (GDP) during FY 2011-12, from this sub-sector was 2.50 percent (DLS, 2012). Livestock Sector plays a significant role in milk and meat production and source of hides and skin. The supply of animal origin food is disproportionately low against high demand. The current intake per head of animal protein in Bangladesh is less than 2 gm per day, against the recommendation of 28 gm per day.

Chapter 1 Introduction ∠ 3

Similarly, Milk availability per head is approximately 30 ml per day against the recommendation of 250 ml (FAO, 2008). Milk production needs to be grown by 4.2 to 5.6 percent annually to meet the increasing demand (Hossain and Bose, 2000). The average per capita meat consumption is 42.1 (kg/year) in the world. In developed and developing country it is 84.9 (kg/year) and 31.1 (kg/year), respectively (FAO, 2009). The demand of meat in Bangladesh is 6.37 million metric ton per year. The per capita daily meat requirement is 120 g but the per capita daily availability is 22.0 g (DLS, 2010). The deficit of meat is more than 80% in 2010 and this demand will increase to 3.96 million metric ton and the deficit will be 78% in 2015 (BBS, 2009). People of Bangladesh get less than one fifty animal proteins of the required recommendation (Chawdhuri *et al*, 1993), which indicates the shortage in supply of meat for human consumption. It is therefore, recommended that increasing meat production can fulfill the demand of people.

The livestock population in Bangladesh comprises of 23.03 million cattle, 1.35 million buffaloes, 23.26 million goats, 2.98 million sheep, 228.035 million chickens and 42.677 million ducks (BBS, 2010). From cattle, we get 173 thousand metric tons of beef and 782 thousand metric tons of milk per year. Furthermore, the country gets 18 thousand metric tons of butter and ghee and 32 thousand metric tons of excellent type of hides from cattle (Anon, 2001a). Livestock provides milk, meat, skin, fuel, organic fertilizer and draft power. It generates 13% of total foreign currency and provides full time employment to about 30% and partial employment to about 50% of the rural population (Alam, 1993). The skin and hides of domestic animals are important commercial material if they are raised properly and protected against parasitic infestation particularly

Chapter 1 Introduction ≥ 4

ectoparasites. Leather is a surplus and export commodity of Bangladesh. About 10% of the available leathers are required to meet domestic demands and the rest 90% is exported. Skin and hides earn 9% of the total foreign currency (Jabbar, 1985). Cattle provide a main draft power in agriculture, especially in cultivation. Cattle are also used for transportation, threshing and crushing of crops. Not only that, cow dung is a good bio-fertilizer, which increases the productivity in land without affecting the environment. Dry cow dung is a popular fuel in the rural areas of Bangladesh, thus it saves the green forest. Bone and teeth are used for making comb. Other industrial and medicinal bio-products are manufactured from horns, hooves, fats, blood etc. However, the present production of cattle is not sufficient for the large population of our country. As such to improve our present situation simultaneously with national economy, it has become essential to increase animal population and their products.

Parasitism is an important limiting factor that is responsible for deteriorating the health and productivity of livestock. The agro-ecological and geo-climatic conditions of Bangladesh are highly favorable for the growth and multiplication of parasites. As a result, about 50% apparently healthy cattle population has been demonstrated to be affected with different species of parasites (Garrels, 1975). The geo-climatic conditions together with the water logged and low lying areas in Bangladesh are conductive to parasitic diseases in domestic ruminants (Qadir, 1982). In fact, cattle of Bangladesh are affected by various types of helminthes parasites (Rahman and Mondal, 1983). In Bangladesh there are many constrains in cattle production, among them malnutrition and parasitism are the major limiting factor (Jabber and Green, 1983). Endoparasites are

Chapter 1 Introduction \( \noting 5

those organisms living within their hosts, in the gut, body cavity, liver, lungs, gall bladder and blood or within the internal cavities, tissues or cell of the host. Such forms nearly always live a completely parasitic existence. Since they totally depend upon their host, endoparasitism is also referred to as infection. Infection with Gastro-intestinal nematodes is regarded as one of important factor causing productivity loss. The losses due to parasitism take place in the form of morality, poor general health condition, retarded growth, lower draft power, decrease in the production of milk and meat (Faiz, 1972). ADB report (1984) clearly mentioned that the loss of productivity of animals in terms of morality, milk, meat, generation loss and other productive traits due to parasitism (50%) in Bangladesh.

The prevalence of parasitic infection depends on ecology, geographical and climatic condition prevailing in Bangladesh (Hossain et al. 2004) Gastro-intestinal nematodes (Haemonchus, Trichostrogylus and Strogylus) cause impaired digestion and also affect the absorption of minerals particularly the Calcium and Phosphorus (Speedy, 1992). Infections by gastro-intestinal helminth parasites in livestock are among the most common, which are considered as economically important diseases of grazing livestock (Perry et al., 2002). They are characterized by lower outputs of animal products (meat, milk, hides and skins), manure and traction, which all impact on the livelihood of small holder farmers (Perry and Randolph, 1999). The greatest losses associated with nematode parasite infections are sub-clinical and economic assessments have showed that financial costs of internal parasitism are enormous (McLeod, 1995). Afazuddin (1985) estimated an annual economic loss of taka 0.1 million due to various parasitic diseases in cattle in military farm,

Chapter 1 Introduction &

Savar, Dhaka. On the other hand, Ghosh (1988) reported an annual loss of Tk. 54.11 million in Bangladesh due to parasitic diseases, specially abomasal nematodes such as *Haemonchus* spp., *Mecistocirrus digitatus*, *Trichostrongylus axei*, *Capillaria* spp., *Trichuris* spp. etc. They are known to cause severe morbidity and mortality in livestock. These nematodes cause anorexia, reduced feed intake, lower activity of some intestinal enzymes and subsequently leading to diarrhoea, anemia, reduced weight gain and milk production, poor general health condition and even mortality (Soulsby, 1982).

The production performances of these cattle are very low in Bangladesh because of wide spread occurrence of pathogenic parasites. Parasitic diseases are considered important in causing enormous economic losses through morbidity and mortality in livestock. Among the parasitic diseases, gastro-intestinal nematodes such as *Haemonchus contortus*, *Trichostrongylus* spp., *Cooperia* spp., *Oesophagostomum columbianum*, *Trichuris* spp. and *Strongyloides papillosus* are most common in Bangladesh (Qadir, 1981; Rahman and Mondal, 1983). Among different species of gastro-intestinal nematodes Toxocara vitulorum is extremely harmful to the calves including morbidity and mortality. Rahman and Mondal (1983) recorded highest load of Toxocara vitulorum in one year old calf. Elsa (1968) reported highest load of Toxocara vitulorum in Nigeria.

The principal feature of nematode infection is as follows; both the adult and the fourth larval stage of *Haemonchus* spp. *Bunostomum* spp. *Trichuris* spp and *Mecistocirrus* spp. suck blood and in addition move and leave wounds with hemorrhagic lesions into the abomasums and

Chapter 1 Introduction & 7

small intestine. The average blood loss has been calculated as 0.05 ml/parasite/day and blood first appears in the feces 6 to 12 days after infection (Clark *et al.* 1962). Whereas *Trichuris* spp. and *Capillaria* spp. causes irritation leading to diarrhoea. Animal may die without showing sign of anemia and hydremia in Haemonchus infection. In chronic cases, it causes bottle jaw. Sheep with 500 *Haemonchus* spp loss 250 ml of blood per day (Urquhart, 1996). These nematodes in the small intestine may cause severe damage to the intestinal mucous membrane. Strongyloides larvae damage the endothelium and form thrombus, cause marked inflammation and thickening of the arterial system especially the cranial mesentery artery and its branches. Clinical signs are intermittent diarrhoea and in some cases loss of appetite and retarded growth (Bashir, 2009).

Studies of Gastro-intestinal nematode were mostly limited to incidence and some extent seasonal variations of incidences (Hirani, 2006). Very limited studies were made on the epidemiology of these nematode which is emphasized as a priority need for implementing control measures. Parasitism is the major cause hindering the development of livestock population in the country (Shahiduzzaman *et al.*, 1999). Several studies have indicated the incidence of different parasitic diseases and their seasonal prevalence in cattle in Bangladesh (Rahman and Razzak, 1973). Gastro-intestinal nematodes are serious problems for ruminants, especially young animals. Debanth *et al.* (1995) suggested that 50% calves up to 1 year of age died due to Gastro-intestinal parasites that cause digestive disturbances and malnutrition leading to calf mortality. Different helminth infections are responsible for about 54.22% calf mortality in Bangladesh. Strongyles are another harmful group of bovine

Chapter 1 Introduction ≥ 8

parasites due to their feeding habit of development process in the digestive system (Shahiduzzaman *et al.*, 1999).

Gastro-intestinal parasitosis adversely affect the nutritional status of cattle and is associated with economic losses in terms of lowered fertility, reduced work capacity, involuntary culling, retarded weight gains lowered milk production, treatment costs and mortality in heavily parasitized animals (Fikru *et al.*, 2006). Hann and Bekur (1991) estimated annual losses of US \$ 2 billion due to mortality and decreased production in animals caused by endoparasites in Sub-Saharan Africa. The agroecological and geo-climatic conditions are favorable for high prevalence of helminthiasis in Bangladesh (Samad, 2000 and 2001)

In Bangladesh, among many causes, parasitism is thought to be a major cause that hinders, the development of livestock population (Jabber and Green, 1983). Unlike bacterial and viral diseases and the diseases caused by parasites are of great importance. Parasitic diseases are also emphasized for their pathogenicity and economic importance in animals by the experts both from the government and nongovernmental organizations. In developed countries, the data on epidemiology of various helminthiasis are published in an efficient manner as an aid to combat infections more effectively. In contrast, in developing countries, little published information and data on the epidemiological aspect of helminth infections exists. Helminth parasitism, especially, gastro-intestinal parasitism is one of the major health problems severely limiting the animal productivity in dairy animals and the significant production losses, which may run into millions of rupees (Shah and Chaudhry, 1995). The problem is neglected due to its chronic and insidious nature (Sanyal, 1998).

Chapter 1 Introduction & 9

Medicinal plants are among the most important natural resources of a country. Bangladesh has also been enriched with this resource. Due to favorable climate, heavy monsoon rain and fertile land plants grow abundantly in forests, roadsides and around house yards. Many of the food, vegetable, beverages, spices and ornamental plants, which are grown in this country contain medicinally useful chemical constituents and constitute important items of crude drugs or therapeutic agents of various medicinal preparations. More than 500 wild and cultivated medicinal plants of this country have so far been enumerated with information on their medicinal properties and uses (Yusuf et al., 1994). But all axe not being used due to lack of knowledge and technology. Besides, ignorance of our people and relevant physicians could also be held responsible for this negligence. Many drugs are now derived from medicinal plants and their constituents are used in many countries of the world; even in our neighboring countries like India, Pakistan, Ceylon, Thailand, China and Japan. Such drugs and raw materials are not only used in their countries but also are exported abroad.

Medicinal plants have been used from the times immemorial for the treatment of various diseases in man and animals. A large number of plants are scattered throughout Bangladesh. Some plants have been identified by the scientist but some have not been identified. These plants could become natural sources of new medicine. However, 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 practices to fulfill the primary health care systems of the man and animals and realizes that the traditional system of medicine may play an important role in the development of livestock of the third world countries.

Chapter 1 Introduction ≥ 10

In this context, investigations on indigenous medicinal plants might contribute to develop effective but low-cost herbal anthelmintics. The "ayurvedic" and "unani" systems of medicine have used several hundreds of plants to cure many diseases in Bangladesh from time immemorial. However, these are mostly used in crude forms and their pharmacological preparations, dosages and mode of action are not based on strong scientific evidence. Until today very little works (Mostofa, 1983; Begum, 1997) have been performed in our country to investigate *in vivo* anthelminite properties of medicinal plants.

Primitive people learned the medicinal values of plants from intuition and observation of the behaviours of the animals through trial and error. They discovered the efficacy of certain plants for certain ailments and they passed this knowledge on to its neighbors and descendents (Concha, 1982). Many unregistered physicians in the Indo-Pakistan sub-continent use various herbal preparations for the treatment of animals. Various pharmaceutical companies in India, Indonesia and Thailand manufacture a variety of indigenous herbal preparations. However, in Bangladesh, these resources have been still very little explored and as such, our farmers and animal owners are deprived of the benefits of using these herbal preparations.

Imported synthetic anthelmintics have long been considered the only effective way of controlling parasitic infections. However, as these are very expensive and unavailable to farmers in rural areas, livestock producers are not interested to use these anthelmintics. Furthermore, some serious disadvantages of using those anthelmintics, notably the development of resistance to helminth parasites (Waller and Prichard, 1985; Lans and Brown, 1998) against various anthelmintic compounds and classes, as well

Chapter 1 Introduction \( \sime \) Introduction

as their residues and toxicity problems (Kaemmerer and Butenkotter, 1973). For these reasons, interest in the screening of medicinal plants for their anthelmintic activity has remained of great scientific interest despite extensive use of synthetic chemicals in modern clinical practices all over the world (Akhtar *et al.*, 2000). Plant remedies were also extensively used as anthelmintics in the developed world before the era of broad spectrum synthetic drugs (British Veterinary Codex, 1953). Many currently available therapeutic compounds are plant derived and/or synthetic analogues derived from those compounds (Farnsworth *et al.*, 1985).

In developed countries the principle of controlling parasitic diseases are based on pasture and barn management and protective treatment (Rodistis *et al.*, 2000). Whereas in Bangladesh, where animals are mainly maintained in mixed farming system with virtually no pasture land for grazing, these methods have limitation to control parasites. To minimize the effects of parasites on animal, different types of anthelmintics are being used all over the world. But due to their indiscriminate use some researchers have expressed their opinion on the development of resistance to anthelmintics in some nematodes. For that, efforts have been made for the development of new effective anthelmintics. Since then various groups of anthelmintics with narrow and broad spectrum activities have been discovered. Ivermectin and Levamisole hydrochloride are the latest broad spectrum anthelmintics.

Control of parasitic diseases have been mainly based on regular anthelmintic treatment in Bangladesh. Because of high economic cost and unavailability of anthelmintics, poor farmers cannot afford to purchase allopathic medicine. Furthermore, frequent use of these anthelmintics increases the resistant population of nematodes (Waller, 1987). On the other hand, herbal

Chapter 1 Introduction ≥ 12

medicine which are equally active but compatible to the economic status of our people as because they are produced from the plants grown in our country and prepared by native technology very cheaply (Khalid *et al.*, 2005). By considering the medicinal value of plants and herbs, government has given special emphasis on the plantation of medicinal plant along with forest and fruit trees. Substantial amounts of foreign exchange can be saved and earned by commercial production of medicinal plants in a country and by exporting them to other countries. In this context, investigations on indigenous medicinal plants might contribute to develop effective but low-cost herbal anthelmintics.

Existence of human on the earth is partly dependent on existence of animals and birds because man depends on them for food, cloths, energy and for recreation. Although the herbal medicine were being used in the human medicine from the ancient period but it is yet not been used equally in veterinary medicine. Due to misuse/abuse of various harmful chemicals, their residues are accumulated in the animal bodies, which can enter into the human body through food chain, and various complexities develop in the human beings. However, the foodstuffs of livestock treated by the herbal medicine are not expected to produce such type of side effects. Considering these points the ethnoveterinary medicine would be safe and environment would be friendly.

From the foregoing discussions, it is assumed that parasitic infection is one of the major impediments for growth and development of cattle in Bangladesh. Until today, very little works have been performed in our country to investigate the anthelmintic properties of indigenous medicinal plants in cattle.

Chapter 1 Introduction ≥ 13

## **OBJECTIVES**

Considering all these constraints, this pioneering work has been under taken in this country with the following objectives:

- To study the prevalence of gastro-intestinal nematodes of cattle in greater Rajshahi.
- To asses the anthelmintic efficacy of 10 indigenous medicinal plants *in vitro*.
- To study the comparative anthelmintic efficacy of two selected medicinal plants with trial of two patent drugs on cattle *in vivo*.
- To study the effect of experimental drugs on clinico-hematobiochemical parameters of cattle.

## **CHAPTER-2**

## **REVIEW OF LITERATURE**

Parasitic infestation is a common problem in animals. Cattle are subjected to a variety of parasitic infections of which gastro-intestinal parasitic infections are of great economic importace. The medicinal plants have now been recognized as potant anthelmintic all over the world. There are many reports published on the use of plant materials against gastrointestinal nematodiasis. Several species of gastro-intestinal nematodes have been reported to be controlled/treated by the application of medicinal plants such as powder, extract and oil as potential source of anthelmintics. Various research works have been done on anthelmintic activity of ivermectin and levamisole on sheep, goat and cattle. So it is somewhat difficult to review and compile all the published information. It can also be mentioned here that there are very limited research works conducted particularly on the use of medicinal plants as anthelmintics in Bangladesh. Attempts have been made to reflect some of these works, which are directly related with the present study. Available literatures on key factors that influence the prevalence of parasitic infections of cattle are reviewed below. The contribution made by numerous research workers has been complied under the following sub-headings.

## 2.1 Prevalence of Gastro-intestinal Nematodes

Eysker and Ogunsusi (1980) found higher prevalence of gastro-intestinal helminthiasis during the rainy season among the cattle population in Nigeria.

**Qadir** (1981) demonstrated the seasonal influence on the GI nematodes infection Six genera of nematodes goats. recorded Trichostrongylus, Haemonchus, Oesophagostomum, Trichuris, Strongyloides and Bunostomum (arranged in order of predominance). The peak infection period of *Haemonchus* spp. extended from June to September and that of Trichostrongylus spp. Oesophagostomum from June to November, and August to January, respectively.

Malan et al. (1982) observed the seasonal incidence of helminth parasites of cattle in South Africa and reported that worm burdens in tracer calves reached in peak levels during January to March after high rainfall from November to February. Dry weather of winter and spring were associated with low worm burdens.

**Dakshinkar** et al. (1982) determined the incidence of helminth infections in ruminants of Nagpur, India. The authors observed that the prevalence of nematode was highest during rainy and summer seasons especially among crossbred cattle.

Chowdhury et al. (1983) recorded 54.6% Haemonchus contortus, 15% Ostertagia ostertagi and 50% Trichostrongylus axei infestation in cattle of Azad Kashmir, Pakistan.

Motalib and Alam (1983) identified Copperia punctata, Oesophagostomum Haemonchus placei, Trichostrongylus axei Mecistocirrus radiatum, digitatus, Bunostomum phlebotanum and Strongyloides papillosus parasites in cattle, of which Cooperia spp. and Oesophagostomum sp. were found to be the predominant species.

Rahman and Mondal (1983) examined 450 cattle and recorded the infection: Mecistocirrus digitatus following nematodes (44%),Trichostrongylus axei (45.3%), Oesophagostomum radiatum (43%), Haemonchus contortus (10.6%), and Haemonchus similes (22%). Heavy infection with *Mecistocirrus digitatus* was recorded in cattle between 2-3 years of age.

Bordas and Migron (1983) studied the abomasal parasites in Norma Charolais cattle in France and reported that Ostertagia spp. and *Trichostrongylus* spp. was low from April to October with a peak in June-July.

Assoku (1983) examined faeces of 570 cattle of both sexes and different ages in Ghana and showed a high incidence (45.6%) of helminth infection, of which 96.92% was the result of single helminth infection. Seven nematodes, two trematodes, two cestodes and one protozoa were identified. Haemonchus contortus was the most common helminth (35.6%) followed by Moniezia benedini (29.7%); Cooperia punctata and Dicrocoelium hospes, Trichostrongylus longispiculus, Oesophagostomum radiatum, Cooperia punctata, Moniezia benedini and Eimeria auburenesis were reported for the first time in cattle from Ghana.

Costa and Vieira (1984) reported *Haemonchus contortus*, *Trichostrongylus* colubriformis, Oesophagostomum columbianum and Strongyloides papillosus were the most harmful nematodes of goats in Ceara, Brazil.

Overend (1984) studied the prevalence of abomasal Trichostrongylidiasis in 68 dairy cows of 2-13 years age in Northern Victoria and Australia. He noted that abomasal Trichostrongyles were found in 93% of animals examined. The author also reported that there was no significant correlation between total Trichostrongylid burden and age of cow or the time of year examined.

**Afzal** et al. (1984) found helminth oval larvae in faecal samples of 820 (46.24%) of 1773 buffaloes and 304 (41.87%) of 726 cattle in Pakistan. The helmiths recorded were: Fasciola spp., Moniezia expansa, Neoascaris vitulorum, Haemonchus spp. Trichuris spp., Dictyocaulus spp. and *Moniezia benedini*.

**Afazuddin** (1985) detected 3.39% ascariasis, 7.11% fascioliasis and 19.72% nematodiasis (excluding ascariasis) in cattle at Savar Military Farm, Dhaka.

Al-Dulaimi et al. (1985) demonstrated that the rate of infection of some parasites such as *Haemonchus contortus* increased towards spring. In contrast, rates of infection with some other parasites such as Ostertagia spp. and *Nematodirus filicollis* were found to increase towards autumn.

Omra-Opyene (1985) examined 2227 cattle of Northern Kenya during the dry and rainy seasons and reported that strongyles was serious in young stock especially in post weaning age and gastro-intestinal parasitism was a predominant problem in the rainy season.

Ramos and Gutierres (1985) conducted a study to examine 143 weaned calves in Brazil, between 1977 and 1981 to detect population dynamics of Trichostrongyles. Species identified were: Cooperia punctata (100%), Cooperia oncophor (9 1%), Cooperia memasteri (75%), Trichostrongylus axei (100%) and Trichostrongylus longispicularis (2.1%). Infection with Cooperia spp. was significantly influenced by rainfall. Trichostrongylus infection was highest in the summer and autumn, and was significantly influenced by the minimum and maximum temperature.

**Ambrosi** et al. (1986) by a faecal examination of 88 cattle farms in Itally, reported strongylosis, dicrocoeliasis, paramphistomiasis and coccidiosis to be widespread, while fasioliasis, moniezia infection, strongyloidosis, trichuriasis and ascaridiasis were less common. Capillariasis occurred rarely and dictyocaulosis was absent. Vegetation of the area was considered to favour the spread of endoparasites.

Nakazawa (1986) described a modification of the Wisconsin sugar centrifugal floatation technique for faecal examination of nematode eggs and intestinal nematodes were detected in the abomasum and upper small intestine of 56% of 150 cattle examined in Hokkaido, Japan Ostertagia ostertagi was most prevalent (47%) followed by Mecistocirrus digitatus (29%), Ostertagia punctata (1%), Haemonchus placei (1%) and Nematodirus helvetinus (1%). In a further survey 74% of 231 cattle were found to be infected with Ostertagia sp. (62.7%), Oesophagostomum sp. (23.2%), Trichuris sp. (17.3%), Mecistocirrus sp. (3.9%), Trichostrongylus sp. (3.5%), Cooperia sp. (1.2%), Moniezia eggs, Eimeria ocysts and eggs of *Toxocara* sp. were also present in 1.7%, 59.7% and 41.1% of faeces respectively.

**Haque** (1986) studied the effect of season on the infectivity of gastrointestinal parasites in Bangladesh and reported the maximum rate of infection in rural condition and in rainy season.

Guimaraes et al. (1987) carried out postmortem examination of 83 goats between April 1982 and April 1984 in Brazil. The percentage of infection were H. contortus-82%, T. colubriformis-80%, T. axe-39%, T. longispicularis-2%, C. curticei-30%, C. punctata-6%, C. Pectinata-6% O. columbianum-42%, O. radiatum-2% and T. ovis- 12%.

**Ahmad and Ansari (1987)** examined the gastro-intestinal tracts of 479 goats and 342 sheep from January 1984 to December 1985. 70.2-75.5% of goats harboured Haemonchus contortus, 67.5-72.4% Oesophagostomum columbianum, 42.6-43.7% Bunostomum trigonocephalum and 51.5-57.8% Trichuris ovis. The corresponding values in sheep were H. contortus 79.2-81.0%, O. columbianum 63.7-74.5%, B. trigonocephalum 53.5-54.7% and T. ovis 45.4-52.8%. The prevalence of H. contortus was highest in June to November, and peaked in October at 92.0%. The prevalence of B. trigonocephalum peaked in March and April at 50.0-54.8%.

Nooruddin et al. (1987) recorded 60.30% (32 out of 53) Trichuris infection in calves at Bangladesh Agricultural University Dairy Farm.

Okafor (1987) conducted a field survey on the occurrence of intestinal nematodes in 335 goats and 117 sheep in the Imo State of Nigeria and found an overall 78% and 77% infection respectively. The author found H. contortus as the prevalent species in goats with 35.52% infection, 0. columbianum in 37.61%. S. papillosus in 6.56% T. colubriformis in 6.26%. Relationship was found between the age of the animals and nematode species as well as the season and rate of infection.

Pal and Balakrishnan (1987) observed the incidence of gastro-intestinal parasites of cattle in the Andamans. Percentage incidence ranged from 23% with Fasciola sp. to 0.4% with Oesophagostomum sp. Paramphistomum sp., Ascaris sp., Strongyloides sp., Strongylus sp., Trichuris sp., coccidia,

Ancylostoma sp. and Haemonchus sp. were also found. Analysis of the pooled data showed a bimodal distribution of infection annually, with maximum incidence occurring in March and December.

Marnu et al. (1987) conducted a study to observe the monthly and seasonal fluctuations in abomasal nematodes by examining 191 abomasii from different abattoirs of Austria during October 1980 to September 1981 and reported that pastured animals showed a higher intensity of infection than stable animals.

Amin and Samad (1987) examined 1324 diarrhoea cattle at Kaliakaur, Gazipur and reported highest prevalence of gastro-intestinal nematode infection in cattle during summer (12.72%) followed by that in autumn (11.69%), spring (10.35%) and lowest in winter (9.37%).

**Gahutu** (1988) described the prevalence of gastro-intestinal nematodes in cattle in June (wet season) 1988 at Kibungo, Rwanda, and compared with results obtained in the dry season (August-November) of 1986. Over-all prevalence was 70.73% in 1986 but >76% in 1988. The prevalences of the major identified species were *Haemonchus placei* (57.7%), Oesophagostomum radiatum (46.5%), Trichostrongylus axei (31.0%) and Cooperia punctata (22.9%). Bunostomum phlebotomum, Ostertagia ostertagi, Nematodirus spathiger, Strongyloides papillosus, Toxocara vitulorum and Moniezia expansa were present at prevalences <10%.

Mishra (1988) studied gastro-intestinal parasitic burden in Sikkim local goats. Cestode proglottids were detected in June and September-November, adult amphistomes in May-August, adult nematodes in May-October, and lungworm larvae from May-December. Faecal egg counts

of nematodes (Strongyloides, Haemonchus Contortus, Bunostomum trigonocephalum, Oesophagostomum columbianum, Trichocephalus ovis, Moniezia and Stilesia) were highest in June-November; coccidian oocysts were most numerous in July-December.

Redl et al. (1988) observed 55 of 58 slaughtered cattle from abattoirs in Hungary were infected with gastro-intestinal nematodes. Prevalence was Ostertagia ostertagi 91.3%, Grosspiculagia podjapolskyi 55.1%, Cooperia punctata 51.7%, Trichostrongylus axei 48.2%, Haemonchus contortus 46.5%, C. oncophora 36.2%, Nematodirus helvetianus 10.3% and Bunostomum phlebotomum 8.6%.

**Lepojev** et al. (1989) reported the prevalence of helminths in sheep. Gastro-intestinal nematodes were found in 100%, Strongyloides papillosus in 67.5%, Trichuris ovis in 32.5%, Moniezia spp. in 62.5%, Dictyocaulusfilaria in 15.0%, Protostrongylus rufescens in 55.0%, Muellerius capillaris in 45.0% and Neostrongylus linearis in 15.0%.

Charles (1989) autopsied 3-4 goats at every month for gastro-intestinal nematodes in Brazil. All goats were found to be infected with one or more of the following species: H. contortus in 96.9%, S. papillosus in 95.4%, O. columbianum in 87%, T. axei in 74.1%, T. colubriformis in 74.1% and T. ovis in 44.3%. Total worm burden present in the farm animals were highest during late rainy early dry season (January-February). The acquisition of nematodes tracer goats occurred mainly from mid-rainy to early dry season (January-June).

Eslami and Hoseini (1989) investigated faecal materials from 360 cattle and 96 calves and the following helminth eggs were reported to be present: Nematodirus spp., Trichuris spp., and Fasciola hepatica.

Yadav and Tandon (1989) examined goats between March 1986 and February 1987 in the East and West Khasi Hills Districts of Meghalaya State, India. 86.8% were infected with one or more species of nematodes. Prevalence of *Haemonchus contortus* infection was highest (52.7%), followed by Bunostomum trigonocephalum (41.7%), Oesophagostomum columbianum (3 8.4%), Trichuris globulosa (24.3%), O. aspersum (19.6%) and T. ovis (3.5%). Goats killed during the 1-year period were also monitored; the prevalence and overall mean egg values were highest (92%) in October (following the period of heavy rains), and lowest (47%) in May.

Pandey (1990) examined 304 abomasum of sheep in Zimbabwe and found 70% infection with *Haemonchus contortus*, to reach a peak in February-March.

Quesada et al. (1990) observed 3921 sheep and 375 goats in 104 flocks in the district of Filiano in Potenza province, Italy, were studied in the autumn and winter of 1985-1986. Faecal samples were collected from 20 flocks. Infection with gastro-intestinal nematodes (Ostertagia, Haemonchus, Trichostrongylus, Strongyloides papillosus, Nematodirus, Cooperia. Chabertia ovina, Bunostomum trigonocephalum, Qesophagostomum and Trichuris) was found in all 20 flocks, with lungworms (Cystocaulus ocreatus, Muellerius capillaris, Protostrongylus rufescens, Dictyocaulus filaria) in 15, with Moniezia in 11, with Fasciola hepatica in 15, with Dicrocoelium dendriticum in 18, with paramphistomes in 2 and with coccidia (Eimeria intricata, E. ovina, E. ovinoidalis, E. parva and E. marsica) in 17.

Rahman and Abmed (1991) investigated the prevalence of gastrointestinal parasites of cattle in different place of Bangladesh by faecal examination. The average rate of infection in animal of different project were observed as follows: 17.6% Fasciola spp., 43.35% Paramphistomum spp., 7.1% Toxocara spp., 26.35% Strongyloids spp., 5.8% Trichuris spp., 5.72% Schistosoma spp. and 0.6% Capillaria spp.

Hossain (1991) examined 10 diseased buffaloes in 138 clinical cases during the period form September, 1989 to June, 1990. The rate of infection with *Toxocara* sp was the highest (34.9). Beside he examined 358 faecal samples and recorded the following gastro-intestinal parasites: Haemonchus spp., (21%). Cooperia spp., (8.9%), Trichuris spp. (1%) and Bunostomum sp. (4.7%).

Matto and Bail (1991) performed post-mortem examination of a total of 1097 buffaloes which revealed 13 species of helminths. In 69 animals from Bareilly, Uttar Pradesh, the prevalence of cestodes trematodes and nematodes infecton were 60.8%, 97.1%, and 60.8% respectively. The prevalence of these parasites in the 20 animals from Patna, Bihar were 60%, 40% and 95% respectively, and in the 20 animals from Ludhiana, Indian Punjab were 100%, 50% and 33% respectively. Gastrothylax crumenfer was the most common parasitic infection in all these areas.

Pal and Qayyum (1992) examined the gastro-intestinal tracts of 53 Kaghani goats from Sweat Valley, Pakistan from September 1990 to January 1991 and recovered H. contortus in 9.43%, O. ostertagi in 81.13%, O. cincumcincta in 66.03% and T. axei in 50.94% of the abomasums; T. colubriformis in 73.58% from the small intestine and O. venulosum in 22.64%, T. ovis in 39.62% and T. globulosa in 11.32% from the large intestine.

Chowdhury et al. (1993) recorded 3.7% Fasciola spp., 19.8% Paramphistomum, 2.9% schistosomes, 77.2% strongylids, 6.6% ascaris, 7.4% strongyloides, 11.0% Trichuris sp. and 8.8% Capaillaria sp. infections in calves at Savar.

Fritsehe et al. (1993) worked on seasonal epidemiology of gastrointestinal nematodes of small ruminants in Gambia and the authors observed the nematodes inflicting small ruminants were, in order of prominence; Trichostrongylus colubriformis (96%), Oesophagostomum columbianum (82%), Haemonchus contortus (67%), Strongyloides papillosus (55%), Gaigeria pachyscelis (3 8%), Cooperia spp. (49%) and Trichuris ovis (12%).

Maingi et al. (1993) determined the prevalence of gastro-intestinal helminths by using the number of nematode eggs and identification of nematode larvae in faecal cultures. The authors reported 83% kids, 93% immature and 93% adult goats were infected with nematodes. Nematode genera identified were *Haemonchus*, Trichostrongylus, Cooperia, Oesophagostomum, Nematodirus and Strongyloides, in that order of prevalence.

Nguyen and Nguyen (1994) examined 459 goats in Hi Tay and Hoa Binh provinces and found Fasciola gigantica in 40%, Paramphistomum cervi in 27%, Moniezia expansa in 36%, H. contortus in 36% and

in 15% Other *Trichostrongylus* spp. goats. parasites were Oesophagostomum, Strongyloides, Trichuris, Bunostomum and Cooperia.

Pandey et al. (1994) examined 32 goats of Zimbabwe for their gastrointestinal nematode burden and found all of the goats were infected. The 4 dominant species, Haemonchus contortus, Trichostrongylus axei, T. colubriformis and Oesophagostomum columbianum were present in 88-97% of the animals. Three other nematodes, Strongyloides papillosus, Bunostomum spp. and Trichuris spp., occurred in 9%, 3% and 21%, respectively of the goats.

**Joshi** (1994) stated that parasitic gastroenteritis is one of the major causes of productivity loss in goats in Nepal. The major parasites recovered in the gut wash of on slaughtered goat from the affected herd were Haemonchus contortus (49.4%), Trichostrongylus spp. (43.5%), followed by Oesophagostomum venulosum (4.2%), O. cercumcincta (2.5%) and Bunostomum trigonocephalum (0.1%).

Garcia-Rameo al. (1994)reported 90.93% et prevalence Trichostrongylids from Galici Spain, with a mean of 759 was per animal. Single infections were less frequent than mixed infections. Nine tichostronglids identified were: Ostertagia ostertagia, Ostertagia lyrata, Haemonchus contortus, Cooperia punctata, Ostertagia circumcinera, Ostertagia oncophora, Cooperia memasteri, Trichostrongylus axei, Nemotodirus helventianus. Ostertagia ostertagi are the most important because both its prevalence (79.79%) and mean parasitic burden (709) were higher. In March-April and October-December periods, all animals were infected. The highest intensity of infection was between July and November. Ostertagia spp. and Trichostrongyles were the predominant genera during this period.

**Bonfoh** et al. (1995) reported that the epidemiological study in Togo revealed the presence of 8 species of gastro-intestinal nematodes in sheep descending and goats. In order of prevalence these Trichostrongylus Spp. (92% and 93% in sheep and goat, respectively), Haemonchus contortus (80% and 88%), Strongyloides papillosus (64% and 70%), Cooperia curticei (58% and 30%), Oesophagostomum columbianum (39% and 42%), Gaigeria pachyscelis (42% and 32%) and Trichuris ovis (3% and 5%).

Githigia et al. (1995) studied the prevalence and economic significance of *Oesophagostomum* spp. and other helminth parasites of ruminants in Kenya and reported that condemnation of cattle carcass due to helminth comprised 11.8% of all cattle slaughtered.

Mostofa et al. (1996) reported the prevalence of gastro-intestinal nematodes belonging to the genera *Haemonchus* (24%), *Trichostrongylus* (10%), Oesophagostomum (12%), Trichuris (4%), Strongyloides (4%), *Mecistocirrus* (6%) and *Ostertagia* (2%) in sheep.

Sanyal et al. (1996) reported that parasitic gastroenteritis dominated by haemonchosis, is a major constraint to profitable sheep and goat production in India.

Saithanoo et al. (1996) stated that gastro-intestinal nematodes particularly *Haemonchus contortus* were common and a major constraint to goat production in southern Thailand.

Alam (1997) found 32.04% Fasciola spp., 36.19% Paramphisiomum spp., 7.83% Haemonchus spp., 4.47% Ascariasis spp. and 2.98% Bunostomum spp. in cattle at Homna Thana in Comilla District.

Qureshi et al. (1997) observed faecal samples collected from 500 male and female adult buffaloes in Charsaddah District in the North-Western Valley of Pakistan. 233 (46.6%) were infected with nematodes: Trichostrongylus (23.40%), Oesophagostomum (18.91%), Haemonchus (13.80%), Neoascaris (13.14%), Ostertagia (9.61%), Bunostomum (6.41%), Strongyloides (6.09%) and Mecistocirrus (5.12%). Overall rate of infection was higher in males than in females.

Thilakan and Satianesan (1997) collected 732 faecal samples from cattle, buffaloes and goats in India. The prevalence of helminth parasites was 77.5% in goat, 38.3% in cattle and 31.9% in buffaloes. Haemochus contortus was the most prevalent species in cattle, buffaloes and goats.

Eslami and Tahmsebi (1997) examined 63 intestinal tract and 1208 carcasses of indigenous cattle in Iran. Haemonchus placi and Ostertagia ostertagi were found in abomasum.

**Crespo** (1998) surveyed for nematodes during 2 periods in the dry season in 130 cattle. Faecal examination revealed eggs of *Strongyloides* spp. in 37 (28.46%) cattle. Eggs of *Trichuris* spp. and other GI nematodes were also detected. Following the necropsy of 20 cattle, Haemonchus placei (32%), Cooperia punctata (52%) and Oesophagostomum radiatum (60%) were identified in the abomasii, small intestine and caecum respectively.

EI-Khalid (1998) examined the digestive tract, liver and lungs of 15 calves in Syria. Six species of nematodes were identified in the abomasum. Ostertagia ostertagi, Trichostrongylus axei and Haemonchus contortus were most common.

**Berrag and Cabaret** (1998) stated that gastro-intestinal and pulmonary nematode infection decrease goat productivity in Moroccan semi-arid condition. Within a herd of goat the authors found that the infection percentage of gastro-intestinal nematode was Teladorsagia in 26%, Trichostrongylus in 19%, Nematodirus in 12%, Trichuris in 24%, Oesophagostomum in 12%, Haemonchus in 5% and Chabertia in 1%.

**Lima** (1998) observed tracer calves to assess the seasonality of infections of gastro-intestinal parasites in beef cattle extensively raised at a farm in the State of Minas Gerais, Brazil. Tracer calves acquired infections during all months of the year, however, highest worm burdens were observed in the rainy season. The following nematode species were recovered from tracer calves: Cooperia punctata, C. spatulata, C. pectinata, Haemonchus similis, H. placei, Oesophagostomum radiatum, Trichostrongylus colubriformis, T axei, Bunostomum phlebotomum and Trichuris discolor. Cooperia was most prevalent, representing 74.4% of the total of all nematodes. This was followed by *Haemonchus* 19.2%, Oesophagostomum 4.5%, Trichostrongylus, Trichuris and Bunostomum, represented less than 1% of the total.

**Jugessur** et al. (1999) reported that gastro-intestinal parasitism could be one of the causes of death that led to a drastic fall in the goat population. 55.4% of the herds had egg counts above 100 eggs of faeces for the 4 common genera of nematodes, namely Haemonchus, most Trichostrongylus, Strongyloides and Dictyocaulus.

Herani et al. (1999) investigated 929 faecal samples of cattle and buffaloes in India from February, 1995 to January, 1997 which revealed that 38.86% animals were positive for gastro-intestinal parasites. Strongyles (7.53%), Trichuris spp., (0.97%), Coecidia (7.86%), Amphistomes (21.85) and *Moniezia* spp. (0.11%) were detected in their study.

Leo poldino et al. (1999) collected faecal samples from 486 cattle in the Triangulo Minerio reason of Minas Gerais between April, 1994 and June, 1995 and were examined for helminth eggs and larvae. Samples from 337 (69.34%) of the animals were positive for helminth eggs (Strongyles, Strongyloides papillosus, Moniezia spp.). Larvae of 7 genera (Cooperia spp., Trichuris spp., Trichostrongylus spp., Haemonchus spp., Ostertagia spp., Oesophagostomum spp. and Diclyocaulus viviparus) were detected.

Vassilev (1999) examined faecal samples from 12472 cattle between November, 1992 and December, 1994 in Zimbabwe. The most prevalent parasites were: Cooperia spp. (35%), Trichostrongylus spp. (33%), Haemonchus spp. (18%),*Oesophagostomum* (11%)and spp. Strongyloides papillosus (3%).

Katoch et al. (2000) observed seasonal incidence of gastro-intestinal nematodes in goats. The highest egg was recorded in the rainy season and the lowest in the summer. The highest incidence of *Trichuris* (40%) was found in the winter, followed by the rainy season (17.5%). Abomasal counts showed the highest incidence of *Haemonchus* (80%) in the rainy season and the lowest (26.9%) in the summer.

Yildirim et al. (2000) reported the prevalence of helminth infections in cattle raised in barns in Kayseri province. On coprological examination,

the types of eggs found in cattle were *Paramphistomum* spp. (14.5%), Fasciola spp. (7.5%), Dicrocoelium dendriticum (9%), Monjezia spp. (1%), Strongylidae (12%) and Toxocara vitulorum (0.5%). Identification of infective larvae from the faecal cultures showed that a majority of eggs in cattle faeces were contributed by nematodes belonging to Ostertagia spp. (35%), Cooperia spp. (15%), Oesophagostomum spp. (15%) and Bunostomum spp. (13%). These were followed by Nematodirus spp. (7%), Haemonchus spp. (4%) and Trichostrongylus spp. (4%). Unidentified larvae were detected as 7%.

Holland et al. (2000) observed the faecal samples between January, 1999 and January, 2000. The following helminth were identified: Toxacara vitulorum, Cooperia punctata, Cooperia pectinata, Oesophagostomum radiatum Trichostrongylus axei, Haemonchus spp., Fasciola spp. and Paramphistomum spp. Six (8%) samples were collected from young calves.

Agneessens et al. (2000) examined the abomasii and faecal samples of 121 dairy cows in Belgium for nematode infection from November, 1997 to October, 1998. Nematodes were present in the abomasii of 110 animals. Ostertagia ostertagi were found in all 110, Trichostrongylus were seen in 65 cases.

**Al-Farwachi** (2000) reported internal parasites in cattle in Mosul from 1997- 1998 by faecal examination. He observed that 21.5% animals were infected with internal parasites. The result of faecal examination and egg culture identified 10 genera of nematodes of which, Ostertagia spp. and. Haemonchus spp. were common.

Borghteede et al (2000) examined abomasii and faecal samples for nematode infection from 125 dairy cows between November, 1997 and October, 1998. The prominent nematodes were: Trichostrongylus spp. (29%), Oesophagostomum spp. (23%), Cooperia punctata (20%), Cooperia oncophora (4%),Haemonchus contortus (2%)and Bunostomum phlebotomum (1%).

Angulo et al (2001) determined the prevalence and annual parasitic population were 33% and 7.28 EPG, respectively, and the prevalence (%) and monthly parasitic population (EPG) were 45.6% and 12.4, 10% and 3.4, 42.8% and 7.1, 25% and 4.9, 16% and 2.56 and 42.8% and 11.3 for January, February, March, April, May, June July, August, September, October, November and December, respectively. The result indicate a significant effect of the months of the year on the variables studied (P<0.01)

Landim et al (2001) compared the contemporary prevalence of nematodiasis in calf in Brazil. The gastro-intestinal nematodes found in the mucosae were carefully removed, identified and counted. The nematodes found were: Cooperia punctata (n=161), Haemochus contortus (n=675). C. pectinata (n=497), Oesophagostomum radiatum (n=348), H. similis (n=186), Trichostrongylus axei (n=110), Trichuris discolor (n=47) and Capillaria bovis (n=1).

Belem et al. (2001) observed parasites of abomasii, small and large intestines of 94 cattle in Burkina Faso. They identified 9 different helminth species such as: Cooperia punctata, Cooperia pectinata, Haemonchus contortus, Trichostrongylus colubriformis, Bunostomum phlebotomum, Moniezia expansa, Avitellina spp., Oesophagostomum radiatum and Trichuris spp., Cooperia spp. were the most prevalent (89.4%) followed by H. contortus (66%) and Oesophagostomum radiatum (42.6%).

**Koroglu et al.** (2001) studied the digestive organ of 75 cattle from Elazg and Turkey. Forty nine cattle (65.34%) were infected with nematodes belonging to 19 species. A total of 6909 nematodes (average 141 nematodes per cattle) were collected. The nematodes most frequently observed were: Ostertagia ostertagi (63.26%), Cooperia oncophora (32.65%), *Haemonchus contortus* (10.2%) and *Trichuris* spp. (10.2%).

Githigia et al. (2001) investigated the impact of gastro-intestinal nematodes on health and production of goat in Kenya and concluded that the gastro-intestinal nematodes cause production losses, weight loss and mortalities in goats. Haemonchus contortus was the main nematode infecting the goats in this area.

**Arslan and Mohammed (2001)** reported that total infestation of parasitic helminths was 68.5% in sheep. The highest rate of infestation was by larvae of Ostertagia spp. followed by Trichostrongylus, Haemonchus and Cooperia, respectively. The study also revealed the presence of various percentages of infestations with three species of lungworms which included Dictyocaulus, Muellerius and Protostrongylus spp., in addition to one tapeworm (Moniezia).

**Lindqvist** et al. (2001) reported the prevalence of nematode infections in sheep. The most prevalent species were *Haemonchus contortus*, Teladorsagia circumeincta, Trichostrongylus axei, T. colubriformis and Chabertia ovina and infections progressively increased during summer in lambs grazing on permanent pastures. H. contortus was found in 37% of the flocks. Nematodirus battus was recorded for the first time in Sweden during the course of this study.

Koroglu et al. (2001) reported prevalence of gastro-intestinal nematodes of sheep were 89.33% in Elazig. The nematodes most frequently observed Marshallagia marshalli (66.67%),were Ostertagia circumcincta (62.67%), O. occidentalis (42%), O. trifurcata (24.67%), Trichuris ovis (28.67%), Trichuris skrjabini (20.67%), Nematodirus abnormalis (18.67%), Teladorsagia davtiani (14%) and N. filicollis (13.33%). Marshallagia marshalli and Ostertagia were observed throughout the year. The infestation rate of *Ostertagia* was highest during the months of June, July, October, November and December. For Marshallagia marshalli, infestation rate was at its lowest between the months of March and September. Trichostrongylus was not observed during the month of February, but they were present in low numbers during the rest of the year. Nematodirus was not observed during the month of May, but they were present throughout the year with peak rates in August and October.

Maingi et al. (2001) determined the prevalence of gastro-intestinal nematodes infection in sheep of Nyandarua District in central Kenya. Haemonchus contortus was the most prevalent nematode in faecal cultures during the rainy season (56%) and dry season (62%), followed by Trichostrongylus sp. (28% and 21%, respectively), Oesophagostomum sp. (12% and 9%, respectively) and *Cooperia* sp. (4% and 8%, respectively).

Pal et al. (2001) reported the prevalence of gastro-intestinal parasites in cattle and buffaloes from Chattisgarh region. They revealed that 23.3% were positive for various gastro-intestinal parasitic infections of cattle and buffalo. The prevalence of Amphistomes (18.2%), Strongyles (14.2%), Trichuris spp. (6.6%), Eimeria spp. (3.3%), Toxocara sp. (2.4%), Fasciola sp. (1.2%) and Moniezia spp. (0.6%) was recorded. The prevalence rate of the parasitic infections was higher among the younger animals (<6 months) than among adults.

Sarker (2001) reported the prevalence of nematode infestation in indigenous cattle breed in Mymensingh. The most common nematodes observed were *Haemonchus* spp. (16.66%), *Trichostrongylus* spp. (6.66%), Trichuris spp. (6.66%), Oesophagostomum spp. (6.66), Strongyloides spp. (8.33) and mixed infection (11.66).

Waruiru et al. (2001) observed the epidemiology of gastro-intestinal nematodes of dairy cattle in Central Kenya. The study revealed that Haemonchus placei, Trichostrongylus axei, Cooperia spp. and Oesophagostomum radiatum were responsible for parasitic gastroenteritis and that H. placei was the predominant nematode present in the young cattle. The total worm burdens in the animals were highest during the rainy season (March-June and October-December) and lowest during the dry seasons (July-September and January-February).

Abebe and Esayas (2001) surveyed caprine gastro intestinal helminthosis in Eastern part of Ethiopia. The authors reported 100% prevalence at post mortem examination in goats respectively. The nematodes recovered were Haemonchus spp. (96.8%), Bunostomum spp. (35.2), Strongyloides spp. (43.6%), Oesophagostomum spp. (70.8%) in goat.

Barry et al. (2002) stated the presence of the helminth species in order of prevalence: Haemonchus contortus (94%), Trichostrongylus colubriformis (84%), Oesophagostomum columbianum (75%), Cysticercus tenuicollis (71%), T. axei (70%), Cooperia sp. (55%), Trichuris ovis (55%), Moniezia sp. (39%), Gaigeria pachyscelis (39%), Strongyloides papillosus (25%) and Paramphistomum sp. (12%) in goats. Gastrointestinal nematode burdens also showed seasonal variations, with a peak in the rainy season between July and October.

Bueno et al. (2002) reported the prevalence of nematode infestation in meat breed ewes in southeast region of Brazil. The most common nematodes observed were *Haemonchus* spp. (72.6%), *Trichostrongylus* spp. (22.9%) and *Cooperia* spp. (4.5%).

Crespo et al. (2002) carried out parasitological studies on 93 oxen slaughtered at Bissau abattoir in the Republic of Guinea-Bissau by the end of the dry (May/June) and rainy seasons (October/November). Parasites obtained were classified as Trematoda, Nematoda and Pentastomida and their prevalences were 96.77%, 77.41% and 1.08%, respectively. The prevalences according to genera were: Dicrocoelium (51.61%), Fasciola (1.08%), Schistosoma (19.35%), Setaria (33.33%) and Linguatula (1.08%). Paramphistomatidae prevalence was 81.72%. Egg output in faeces was positive for several helminths in addition to trematodes: gastro-intestinal nematodes 50.54%, Ascaris Trichuris 2.15% and Strongyloides 1.08%.

Gokcen and Guclu (2002) determined the prevalence of gastro-intestinal nematodes in cattle in the province of Konya. 22% cattle were infected with various nematodes. The cattle were infected with one, two, three and four different species at rates of 72.73%, 13.65%, 4.54%, and 9.08%, respectively. Haemonchus contortus (8%), Ostertagia marshalli (4%), O. ostertagi (3%), O. trifurcata (2%), O. circumcincta (1%), and Trichostrongylus axei (1%) were collected from the abomasums. Cooperia oncophora (3%), Nematodirus filicollis (1%), and Setaria cerivi (1%) were collected from the small intestine. Trichuris oviis (3%), T. discolor (2%), T. globulosa (1%), Oesophagostomum radiatum (1%), O. columbianum (1%) and Chabertia ovina (1%) were collected from the caecum and colon.

Piskorova et al. (2002) reported the prevalence of nematode infestation was 67.1% and 62% for the goats and roe deer respectively. In the goat faeces, the nematodes observed were *Cooperia* spp. (31.3%),Nematodirus spp. (16.3%), Chabertia spp. (13.6%) and Bunostomum spp. (6.1%). While the nematodes observed in the faecal samples from the control roe deer were Trichostrongylus spp. (40%), Ostertagia spp. (32.5%), *Chabertia* spp. (32.5%) and *Cooperia* spp. (27.5%).

Sahoo et al. (2002) observed the prevalence of helminthic infection was and 55.48% in stall-fed and grazing cattle respectively. Amphistomes (40.30%) were predominant, followed by strangles (2.61%), Fasciola (2.11%), Ascaris (1.40%), Strongyloides (1.11%), Trichuris (0.9%) and *Moniezia* spp. (0.6%) in grazing cattle. Prevalence of Amphistomes, Strongyles, Fasciola. Ascaris, Strongyloides, Trichuris, and Moniezia spp. were 3.89, 5.0, 0.56, 3.89, 1.67, 1.67 and 5.6% respectively in stall-fed cattle. Helminthic infection was recorded throughout the year with little seasonal variation, i.e., highest during the rainy (51.8%) followed by the winter (50.7%) and summer (47.6%) seasons.

Diakou and Papadoponlos (2002) studied 600 faecal samples, collected from cattle in Greece and the following parasites were detected: Entamobia bovis (0.5%), Strongyloides (41.67%), Neoascaris vitulorum (3.33%), *Trichuris* spp. (0.83%) and *Paramphistomum cervi* (4.66%).

Borkovcova and Chladek (2002) examined faecal samples collecting from 31 calves and cows from October, 2000 to September, 2001 in Czech Republic. The prevalence varied from 0% in January to 35% in September and the following eggs of nematodes were detected: Ostertagia spp., Cooperia spp., Oesophagostomum spp. and Bunostomum spp.

Palampalle et al. (2002) examined faecal sample collecting from 450 cattle and 420 buffaloes for the detection of gastro-intestinal nematode infection. It was found that cattle and buffaloes harbored 7 and 5 Strongyles respectively. Among them *Trichostrongylus* spp., *Haemonchus* spp. and *Bunostomum* spp. were commonly found.

Waruiru et al. (2002) detected infective nematode larvae on pasture from July, 1995 to June, 1996 in Kenya. The number of larvae on pasture was directly related to the rainfall pattern. Haemonchus sp. was the predominant genus, followed in decreasing order by Trichostrongylus spp., Cooperia spp., Oesophagostomum spp. and Bunostomum spp.

Achi et al. (2003) studied helminthological postmortem examinations of 48 cattle from the savannah area of Cote-devoir were performed between September 1996 and August 1997. In the gastro-intestinal tract, six species of nematodes have been found. The most frequent (prevalence of 50% to 98%) were Haemonchus sp., Cooperia punctata, Cooperia pectinata, Oesophagostomum radiatum, Trichostrongylus axei and

Bunostomum phlebotomum. All parasites and faecal egg counts showed significant seasonal variations with a peak in March and a second peak in June-August during the highest rains period.

**Islam** (2003) reported the prevalence of gastro-intestinal nematodes in buffalo was Strongyloides spp. (9.6%), Haemonchus spp. (21.15%), Trichostrongylus spp. (7.6%), Oesophagostomum spp. (9.6%) and mixed infection (25%).

Eisa and Esmail (2003) reported the prevalence rate of naturally infected with gastro-intestinal nematodes was 28.9% in sheep in Egypt. The most prevalent nematodes recovered were Trichostrongylus spp. (33.9%), Haemonchus spp. (30.6%), Ostertagia spp. (18.2%), and Nematodirus spp. (17.4%).

**Keyyu** *et al.* (2003) determined the prevalence of gastro-intestinal nematode infections in indigenous Zebu cattle in the lower plain (lowland zone) of the southern highlands of Tanzania. The results indicated that 97.2% of the examined animals were infected. The prevalence of nematodes were: Haemonchus placei-84.7%, H. similes-5.6%, Oesophagostomum radiatum-79.2%, Cooperia pectinata-55.6%, C. punctata-44.4%, Bunostomum phlebotomum-5.6%, Trichuris globulosa-5.6% and *Trichostrongylus* colubriformis-1.4%. The highest burdens occurred at the end of the rainy/early dry season, while the lowest burdens were found at the end of the dry/early rainy season. Immature cattle (<3 years) had significantly higher worm burdens than mature cattle.

Ha and Vu (2003) observed on parasitism in goats in USA. The authors reported the incidence of infection in native goats was 88.9%. which

parasites like involved gastrointestinal Fasciala spp. (14.2%)paramphistomum spp. (23.5), and Haemonchus spp. (76.1%). Mainly 4 parasites infected Most of the goats

**Soca et al.** (2003) determined that 100% of the animals (young cattle) were parasitized by Haemonchus, Cooperia, Oesophagostomum and Ostertagia spp. Haemonchus was the most prevalent (61.4%). EPG showed a decrease of 64.6% during the first samplings until May, while slight increases were observed in June and September-October during higher rainfall levels.

Vasquez (2003) conducted a study in between 1991-1996 to determine the types of gastro-intestinal nematodes present and also to find out the seasonal variation in the prevalence of these parasites in cattle in Argentina. Lots of 40 weaned heifers aged 6-8 months were examined in each year. The most frequent parasites were: Cooperia spp., Ostertagia spp. and *Haemonchus* spp.

Ngeole et al. (2003) identified the species of parasites infecting cattle in the high land area of Cameroon. A total of 100 cattle from January to June 2000 were examined. The most prevalent parasites were: Haemonchus contortus (91%), Trichostrongylus spp (67%) and *Oesophagostomum radiatum* (17%).

Niddhi et al. (2003) studied the prevalence of caprine gastrointestinal parasitism in 277 goats based on qualitative and quantitative examination of faeces collected during April to August 2000. The author suggested that overall prevalence rate of gastrointestinal parasitism was 37

(49.46%). The author also suggested that *H. contortus* relatively higher in Jamunapari goat (52.63%) than Barbary (42.86%).

Lee Keo Hua et al. (2003) randomly collected faecal samples from calf, heifer and dairy cattle and examined to study the present status of endoparasitism in cattle in Taiwan. A total of 7 parasites such as; *Toxocara* vitulorum. **Trichuris** Strongyloides spp., papillosus, Trichostrongylus colubriformis, Haemonchus contortus, Oesophagostomum radiatum and Eimeria bovis were identified. The infection rates in calf, heifer and dairy cattle were 46.0%, 50.3% and 40.6% respectively.

Tsotetsi and Mbati (2003) studied the intensities, seasonal incidence and distribution of helminth parasites that occur in cattle of various breeds in Tanzania. The study was conducted from March 2000 to May 2001. Haemonchus spp. and Oesophagostomum spp. were dominant nematode genera found to be infecting the animal.

Rehbein et al. (2003) necropsized fifteen Holstein-Friesian bulls and reported that the most prevalent gastro-intestinal helminth was: Ostertagia ostertagi (100%; 1355; 130-4590) followed by Cooperia oncophora (86.7%; 343; 20-14800), Oesophagostomum radiatum (86.7%; 3.7; 1-42), Trichostrongylus axei (80%: 65; 50-910), C. punctata (73.3%; 14.8; 10-140) and *Trichuris discolor* (73.3%; 4.8; 1-108).

Pandit et al. (2004) conducted a survey on gastro-intestinal helminth parasites in cattle of Kashmir, under two different management practices. Out of 741 and 1121 animals examined from organized and unorganized sectors, 72.46 and 75.02% harbored parasitic infection, respectively. Haemonchus spp., Ostertagia spp., Mecistocirrus spp., Nematodirus spp.,

Oesophagostomum spp., Bunostomum spp., Trichostrongylus spp., Trichuris spp. and Strongyloides spp. were the identified nematodes. A significant (P<0.05) effect of season on the incidence of helminth infections was observed in both stocks.

**Mbae** et al. (2004) studied the prevalence of gastro-intestinal nematodes in small ruminants in semi-arid Turkana District of Kenya. Faecal shedding was higher in young animals compared to adults and the infection was heavier in sheep. The faecal egg counts were significantly higher during the wet season for both sheep and goats. Haemonchus contortus was the main nematode encountered in coprocultures (goats 73%. 62.8%). Other encountered sheep nematodes included Trichostrongylus axei, Trichostrongylus colubriformis, Bunostomum trigonocephalum, Oesophagostomum columbianum and Trichuris ovis.

Pratibha et al (2004) observed the effect of season on the incidence of Toxocara vitulourum in cow and buffalo calves in patna and its surroundings. The authors found incidence in cow (38.3%) and buffalo calves (41.7%) were highest during monsoon.

Bhattacharyya and Ahmed (2005) examined 546 faecal samples collected from cattle and buffaloes of different ages, breeds and both sexes in India from March, 2002 to February, 2003. The samples were examined for the presence of helminth ova using sedimentation technique. It was showed that 387 (70.88%) samples were positive for heiminth ova. The incidence of infection was slightly higher in buffaloes (72.95%) compared to cattle (68.60%). The highest prevalence of infection was in the monsoon (82.31%), followed by winter (76.88%). The most dominant species were: Ascaris sp. (39, 7.14%), Strongyloides

(32, 5.86%), Trichuris spp. (15, 2.75%), Strongylus sp. (13, 2.3 8%), Haemonchus spp. (11, 2.0 1%) and Bunostomum spp. (10, 1.83%).

Muraleedharan (2005) reported that 18.22% cattle and 20.86% buffaloes were infected by nematodes in Karnataka during drought period. Strongyles were the most common nematodes. Neoascaris infection was slightly higher in buffaloes than in cattle.

Diakou et al. (2005) investigated the level of parasitism in high producing dairy cattle in well managed intensive farms. Faecal samples were collected from 105 Friesian cattle from 5 dairy farms in Thessaloniki. In this study, 50 animals (47.6%) were found to be infected with gastro-intestinal nematodes such as, Strongyloides egg in 10 (9.5%) and *Trichuris* spp in 2 (1.9%).

**Tendon** et al. (2005) conducted a study to access the spectrum of helminth parasites in mithun (Bos frontal is) and other co-existing wild (B. gaurus) and domestic livestock in Bhutan and India. It was observed that nematodes with direct life cycle such as Trichuris spp., Strongyloides spp., Toxocara spp., Ascaris spp., Oesophagostomum spp., Bunostomum spp., Mecistocirrus spp., Capillaria spp., Cooperia spp., Ostertagia spp. and *Dictyocaulus* spp. were most predominant among these animals.

**Jager** et al. (2005) studied prevalences of Glardia duodenalis, Strongyloides papillosus and strongyles in a German upland area in German Angus (GA) and German Simmental (OS) stickler of beef cattle herds covering two winter housing periods and the grazing season between them. G. duodenalis was observed with a maximum prevalence of 38% in 4 weeks old calves, a cumulative incidence of 58% 9 weeks after birth and with generally low

intensities. Strongyloides papillosus was common with a cumulative incidence of 53% 9 week after birth and occurred independent of the housing system. Mean strongyles egg prevalence was 50% with 50-100 EPG by means throughout the grazing season.

**Keyvu** et al. (2005) determined the prevalence of gastro-intestinal (GI) nematodes in traditional, small-scale dairy and large-scale dairy cattle farms in Iringa district, Southern highlands of Tanzania. The predominant nematodes were Cooperia spp. (51.6%), Oesophagostomum radiatum (35.7%), and *Haemonchus placei* (10.2%). The worm burden in tracers was mainly composed of *Cooperia* spp. (83%) in large-scale dairy farms, while O. radiatum was dominant (60.8%) in traditional farms. Faecal egg counts (FEC) and tracer worm counts were generally low and FEC peaked only in calves and weaners/yearlings. Adults and all age groups in small-scale dairy farms had very low FEC throughout the year. Pasture larval counts, FEC and tracer worm counts peaked towards the end of the rainy season.

Muraleedharan (2005) reported that 18.2% cattle and 20.9% buffaloes were infected by nematodes in Karnataka during drought period. Strongyles were the most common nematodes. *Neoascaris* infection was slightly higher in buffaloes than in cattle.

Shashi et al. (2005) examined for the presence of helminthes in 750 goat intestine samples collected from abattoirs located in 3 major localities (i.e., 250 smaple each from Jajra, Vikasnagar and Gullar ghati) of the city of Dehradun Uttaranchal, India. The author cited cestode was detected in 50, 60 and 100% of the samples from Majra, Vikasnagar and Gullar ghati, respectively. The author alos cited *Oesophagostomum columbianum* was present in 30, 30 and 70%, while Trichuris trichiura was identified in nil, 10 and 30% of the sample from Majra, Vikasnagar and Gullar ghati, respectively.

**Keyyu** et al. (2006) determined the prevalence of gastro-intestinal (GI) nematodes in communally grazed traditional cattle, zero-grazed smallscale dairy cattle and intensively grazed large-scale dairy cattle. The prevalence of GI nematodes in traditional, large-scale dairy and smallscale dairy cattle was 67%, 44.4% and 37%, respectively, with the highest faecal egg counts in calves. The prevalence of GI nematodes varied greatly among villages and farms.

**Pedreira** et al. (2006) observed the presence of gastro-intestinal nematode parasites infecting sheep in Galicia (NW Spain), an area with Atlantic climate where sheep production is replacing cattle due to the Agricultural Community Politics of the European Union. From September 2001 to November 2002, 1710 faecal samples were randomly collected from 49 sheep farms and examined by using the flotation technique to determine the prevalence of gastro-intestinal nematode parasites. The sheep-level prevalence was 100%, and the genera identified were Chabertia, Cooperia, Haemonchus, Nematodirus, Oesophagostomum, Teladorsagia, Trichostrongylus and Trichuris.

Hirani et al. (2006a) reported month and season wise prevalence of gastro-intestinal parasites in adult cattle in Gujarat. Examination of 1503 faecal samples revealed the overall infection up to 44.18%. Three major parasitic infections were detected such as amphistomes (16.23%), strongyles (12.11%) and coccidia (9.12%). Prevalence of gastro-intestinal parasitism was highest during summer (51.83%) followed by monsoon

(45.22%) and the lowest in winter (35.97%). The highest prevalence was observed during September and the least in December.

Hirani et al. (2006b) examined 1073 faecal samples of adult cows in Gujarat for two consecutive years (August, 1995 to July, 1997) and reported parasitic infestation rate was 45.76%. The predominant parasites found were: Amphistomes (17.89%), Strongyles (14.17%) and Coccidia (7.46%). Slightly highest infection was found during the month of September and in the rainy season.

Repossi-Junior et al. (2006) studied the prevalence of gastro-intestinal helminthosis in calves and suggested the control measures against these infections in Brazil. About 222 calves of 1-18 months old were examined and divided among the 12 dairy farms. Result revealed that the prevalence of Trichostrongylidea eggs (Haemonchus sp. and Cooperia sp.), Trichuris sp., Moniezia sp., Strongyloides papillosus and Coccidial oocysts in adult and calves of these farms were: 100 and 66%, 100 and 57.8%, 50 and 8.2%, 25 and 1.8°/o and 33.3 and 7.8% respectively.

Fikru-Reassa et al. (2006) reported the prevalence and risk factors associated with gastro-intestinal parasitism in Western Oromia during 2003-2004. A total of 257 cattle were examined in the study using standard coprological procedure. This study showed that 69.6% cattle were infected. Strongyles were the most prevalent parasites encountered in the area.

Murphy et al. (2006) conducted a research work to study the prevalence of gastro-intestinal nematode infection in culled cows in Ireland. Abomasii were collected from 30-68 culled beef and dairy cows during autumn, 2002 and summer, 2003 respectively. Ostertagia ostertagi were found in the abomasii of only three (10%) samples in autumn and in 38 (57%) samples in summer. The majority of positive animal had low burdens of O. ostertagi but in a few individual in the summer had a moderate infection (5000-1000) of adult worm. A proportion of the cows in the summer were also co-infected with small number of Trichostrongylus axei, Cooperia oncophora predominated in the recoveries from the larval cultures although O. ostertagi were also recovered.

**Kaewthamasorn and Worangsamee** (2006) determined baseline data on the prevalence of gastro-intestinal parasites of beef cattle in Thailand and investigated the factors associated with the prevalence of parasitic infections. A total of 207 faecal samples were collected during the summer of 2005. The overall prevalence of gastro-intestinal infection was 61% (126/207). The most common helminth infection in this study were rumen flukes 28% (58), followed by Strongyles 27% (55), Strongyloides 1% (2) and *Trichuris* sp. 1% (2). The high rate of parasitic infection in this area might be related to the poor management by the farmers.

Morgan et al. (2006) carried out a study on the agricultural restricting and gastro-intestinal parasitism in domestic ruminants in Kazahstan. The surveyed 505 cattle, sheep and goats for gastro-intestinal parasitism using coprological examination and a further 30 sheep and 4 goats using post mortem extraction of helminths. They reported that dominant helminth genera were *Marshallagia* sp., *Nematodirus* Trichostrongylus sp. Haemonchus sp. was also present but there was no relationship between fecal egg density and body condition score.

Mazid et al. (2006) recorded 94.67% sheep of Mymensingh district, Bangladesh harbour different species of helminth parasites in their digestive system. The authors identified six species of trematodes, namely, Fasciola gigantica, Schistosoma indicum, Paramphistomum cervi, Cotylophoron cotylophorum, Grastrothylax crumenifer and Homalogaster paloniae, two species of cestodes, namely, Moniezia expansa and Moniezia benedeni and three species of nematodes, namely, Haemonchus contortus, Oesophagostomum columbinanum and Trichuris ovis. The authors also recorded the higher occurrence in winter season (100%) followed by rainy (96%) and summer (88.57%). In relation to age, the occurrence of helminth parasites in younger (<1 year) and in old  $(\ge 1 < 2 \text{ years})$  was 76.09%. In relation to sex, the occurrence of helminth parasites was 100% in female and 78.57% in male sheep.

Gadre (2007) repoted that out of the 2288 animals examined, 1441 (62.96%) harboured helminthes infection. The infection rate was highest during post-monsoon (78.80%), followed by winter (63.44%), monsoon (62.34%) and summer (45.66%). The overall prevalence of nematode, trematode, cestode and mixed type of helminthes infection was 41.63, 11.11, 0.98 and 46.28%, respectively. The most common helminthes observed was *Paramphistomum* sp. (12.28%), followed by *Toxocara* sp. (10.97%), Moniezia sp. (8.96), Strongylus sp. (6.99%). Haemonchus sp. (5.98%), Fasciola sp. (3.81%), Schistosoma sp. (3.01%), Trichuris sp. (1.87%), Oesophagostomum sp. (1.00%) and Trichostrongylus sp. (0.96%). The infection rate in animals less than one year, between 1-3 years, between 3-6 years and more than 6 years was 81.38%, 59.95% and 63.16%, respectively. The sex wise prevalence in crossbred and local dairy animals was 63.98 and 61.45%, respectively.

Iqball et al. (2007) recorded Toxcara vitulorum, Fasciola hepatica Haemonchus contortus, Bunostomum phlebotomum, Ostertagia circumci, Oesophagostomum radiatum, and Trichostrongylus spp. through standard coprological examination procedure of dairy animals of Nestle milk collection areas of Punjab (Pakistan).

Asif et al (2007) recorded the overall prevalence of helminthiasis was 51% in cattle, 47% in buffaloes, 62% in sheep and 52% in goats, with nematodes being the most common helminths in an irrigated area of lower Punjab, Pakistan. The authors also reported the higher prevalence of helminths in young animals compared with adults in cattle (P<0.0001), buffaloes (P<0.0001), Sheep (P<0.059) and goats (P=0.010). The prevalence of different species of helminths also varied in different age groups, with *Toxocara vitulorum* being higher in calves than adults both in cattle and buffaloes (P<0.0001). Sex-wise prevalence of helminths was higher in males than females for buffaloes (P<0.0001) and sheep (P=0.014) in contrast to cattle and goats.

Chaudary et al. (2007) studied the prevalence and seasonal trend of the Haemonchus contortus in sheep and goats in the Potohar areas of northern Punjab, Pakistan from December 2004 to January 2006. The author represented highest EFLCs in goats in Islamabad, followed by Jhelum, Attock and chakwal districts. The author also observed that high prevalence of *H. contortus* in Potohar areas was due to favourable agroclimatic conditions that favour the development and survival of the freeliving stages of *H. contortus*.

Anwar (2008) studied the prevalence of helminth parasite in cattle. In the pre-monsoon, overall prevalence was 58.5% and in the post-monsoon it was 50.3%. Prevalence with different gastro-intestinal helminthes in the pre-monsoon period were; 30.1% with strongyles, 1.2% with Neoascaris sp., 1.8% with Strongyloides sp., 58.9% with Paramphistomes, 15.6% with Fasciola sp and 13.7% with intestinal schistosomes. In the postmonsoon period, prevalence with respective helminth groups/species were 15.8%, 1.2%, 3.4%, 21.4%, 11.3% and 5.2%.

Asif et al. (2008) collected a total of 338 fecal samples (8 from sheep 252) from goats) from August 2004 to December 2005 in and around twin cities of Rawalpindi and Islamabad and found 65.7% positive for endoparasites. The authors also mentioned the prevalence gastrointestinal parasites tended to be higher (p = 0.059) in sheep (72%) than in goats (63.7%). The endoparasite identified in sheep included Haemonchus (80.64%),Coccidia (51.61%),**Trichuris** (32.25%),Nematodiru (29.03%) and Fasciola (4.38%) while only Haemonchus (75%), Trichuris (62.5), and Coccidia (57.5%) were recovered from the fecal samples of goat.

Okaiyeto et al. (2008) conducted a survey on the prevalence of gastrointestinal parasites in nomadic sheep in 8 Local Government Areas (LGAS) of northern Nigeria and reported the mean helminth prevalence rates according to their species as *Haemonchus contortus* 49.9%, Cooperia curtecie 39.6%, *Oesophagostomum* 14.9% sp. and Trichostrongylus sp. 1.9%.

Gadahi et al. (2009) observed 63.50% samples positive for endoparasites collected from sheep and goats in and around Rawalpindi and Islamabad,

Pakistan. The authors also detected 53.33% and 66.45% samples were positive for gastrointestinal parasites in sheep and goats, respectively. The prevalence of identified parasites were-**Trichuris** (40%),Haemonchus (28.88%), Coccidia (27.77%), Nematodirus (11.11%) and Fasciola (4.44%) in sheep and Haemonchus (64.19%), Coccidia (43.87%), Trichuris (35.48%), Nematodirus (13%), Trichostrongylus (4.51%), Strongyloides (3.22%) and Fasciola (66%) in goats.

**Ijaz** et al. (2009) reported the overall infection rate of GIT helminthes in sheep was 70.67% in Lahore, Pakistan. The authors also recorded the higher infection rate of nematodes (50.67%) than trematodes (18.67%) and cestodes (1.34%).

Biu et al. (2009) conducted a faecal survey of ova/oocysts of gastrointestinal parasites of ruminants on the University of Maiduguri research farm and observed the prevalence rate as 47.0%, 54.0% and 58.0% for cattle, sheep and goats, respectively (P>0.05). The authors also observed more infection rate among younger ruminants (cattle: 50.0%; sheep: 54.7% and goats: 58.1%) compared to the older ruminants (cattle: 44.0%, sheep: 52.8% and goats: 62.7%) compared to the males (cattle: 52.0%; sheep: 46.8% and goats: 51.2%) (p>0.05).

Holzhauer et al. (2010) found a 2-month-old suckling calf had complaints of diarrhoea and roundworms near the perineum. Fecal examination showed the presence of a high number of round worm eggs of Toxocara vitulorum.

Nuruzzaman (2010) examined a total of 200 faecal samples and 50 abomasal sample of goat. Among them, 74.00% were infected with

stomach worm. The prevalence rate was higher in case of *Haemonchus* contortus (58.56%) than Trichostrongylus sp. (16.00%). In age groups, young aged in between 10-20 months (55.56%) were significantly susceptible (p<0.01) than that of kid aged <10 months (47.36) and adult aged >20 months (45.45%) in observation of fecal sample. The abomasums study showed that prevalence of stomach worm infection, young aged ≤20 months (84.61%) was higher than adult aged >20 months (61.21%) and almost similar in males (73.91%) and females (74.07%) goats (odd ratio 1.008), was also similar in faecal sample, males (46.93%), females (54.90%) (Odd ratio 1.38) stomach worm infection was in Black Bengal goat (72.08%) and Jamunapari (85.71%). where Jamunpari was higher (p<0.01) higher in poor body conditioned goat (93.75%) than normal body conditioned goat (67.63%). In this study, higher stomach worm per animal found in Haemochus contortus  $(6.02\pm0.0928)$  and lower in *Trichostrongylus* sp  $(0.04\pm0.14)$ .

Sangma (2010) prevalence of helminthes of sheep and its relation to age, sex, nutritional status, management system and flock size were studied at Tangail district, Bangladesh from July to October, 2010 examined a total of 190 sheep of which 154 (81.05%) were positive for one or more species of helminth parasites. Seven species of helminthes were identified, of them three species were trematodes, namely, Fasciola gigantica (8.42%), Paramphistomum spp. (44.21%) and Schistosoma indicum (3.68%); four species were nematodes, namely, Hookworm (18.95%), *Trichuris* spp. (2.11%), Strongyles (62.63%) and *Strongyloides* spp. (9.47%). No cestodes were identified. In the age groups, the prevalence was higher (p<0.01) in young sheep aged >1-2 year (92.75%) than in adult aged >2 years (83.33%) and in lamb aged  $\leq 1$  year (63.63%). Significantly (p<0.01) higher prevalence was recorded in female than male sheep. In relation to nutritional status and flock size, prevalence of helminthes were also significantly (p<0.01) higher in poor conditioned and large flock sized animals.

Hossain (2010) studied nematodes infection in cattle in Bangladesh Agricultural University dairy farm from November 2009 to May 2010 by fecal sample examination. Among 130 samples examined, 26 (20%) cattle were found infected with nematodes eggs of Toxocara vitulorum (4.62%), stomach worm (9.23%), Trichuris spp. (3.08%) and Capillaria spp. (3.08%). No mixed infection was recorded in this study. Prevalence of nematode infection in relation to age, sex, breed, season, and nutritional status were also studied. Relatively higher prevalence of nematode infection was observed in summer season (23.33%) than winter season (17.14%). In the age groups, calves (33.33%) were mostly susceptible to nematode infection followed by young (30%) and adult (12.5%). In case of breed, cross breed (23.33%) were found more susceptible to nematode infection than that of local breed (12.50%). The prevalence of nematode infection was also significantly higher in female (21.11%) than that of male (17.5%). In case of nutritional condition, prevalence was higher in poor body conditioned cattle (30%) than that of normal body condition cattle (13.75%). In case of nutritional condition, prevalence was higher in poor body conditioned cattle (30%) than that of normal body conditioned cattle (13.75%).

**Khan et al.** (2010) reported the significantly higher (P<0.05) prevalence of GI helminthes in sheep (44.17%, 371/840) than in other livestock of district Toba Tek Singh, Punjap, Pakistan, The authors identified

important helminth species which includes Fasciola gigantica, Fasciola hepatica, Haemonchus contortus, Toxocara vitulorum, Trichostrongylus Ostertagia spp., *Oesophagostomum* spp., spp., Cooperia spp., Strongyloides spp., Moniezia spp., and Trichuris spp. The authors also observed the significantly higher prevalence of GI helminthes except F. hepatica and F. gigantica in grazing animals, female (P<0.05) and young (P<0.05) of all the host species when compared with stall-fed animals, males and adults, respectively. Using ponds and rivers/canals as drinking water were found to have significant influence (P<0.05) on the prevalence of GI helminths.

Mamun et al (2011) reported that overall 61.02%. Buffaloes were found to be infected with gastro-intestinal parasites in Kurigram district. Nine species of gastrointestinal parasites were identified, of them four species were trematodes, namely, Fasciola gigantica (22.46%); Paramphistomum cervi (29.24%), Schistosoma indicum (1.27%). Schistosoma spindale (0.85%); three species were nematodes namely, Toxocara vitulorum (2.54%), Strongyles (0.85%), Strongyloides sp. (0.42%), two species were protozoa, namely, Eimeria sp. (3.39%) and Balantidium coli (37.29%). No cestodes were detected in his study.

**Siddiqua (2010)** carried out a parasitic investigation in 240 goats and 240 sheep of aged 2 year all in Ghatampur area of Kanpur, during the period from February 2000 to February 2005 and found 50.41% goats and 53.33% sheep were positive for various parasites. The author reported the prevalence of different helminthes, namely, *Moniezia* sp. (11.25%), Avitellina sp. (7.50%), Trichuris sp. (7.50%), Fasciola gigantica (5.4%), Strongloides papillous (5.41%), Haemonchus contortus (4.16%),

Oesophagostommum sp. (3.75%), Dicrocoelium dentriticum (2.91%) and Stilesia vitatta (2.50%) in goats and Moniezia sp. (11.66%), Avitellina sp. (8.75%), Fasciola sp. (6.25%), Oesophagostomum sp. (5.41%), Haemonchus sp. (4.58%), Strongyloides sp. (4.16%), Dicrocoelium sp. (3.33%) and *Stilesia* sp. (2.91%) in sheep. The authors observed the highest incidence in rainy season and lower in winter season but moderate in spring and summer season. The author also mentioned overall rate of infection was much higher in sheep (53.33%) than goats (50.41%).

**Biswas** (2012) examined a total of 497 buffaloes were among them 419 (84.90%) were found to be infected with gastro-intestinal parasites. Thirteen species of gastro-intestinal parasites were identified, of them four species were trematodes namely, Fasciola gigantica (25.40%), Paramphistomum cervi (41.40%), Schistosoma indicum (5.80%). Schistosoma spindale (2.40%); Six species were nematodes namely, stomach worm (9.70%), Toxocara vitulorum (3.0%), Trichuris sp. (3.0%) Strongyles (0.80%), Strongyloides sp. (2.0%), Capillaria sp. (0.40%), one species was cestode, *Moniezia* sp. (0.60%) and two species were protozoa, namely, Eimeria sp. (7.0%) and Balantidium coli (37.40%). In this study, prevalence of gastro-intestinal parasites in relation to age, sex, season dynamics and nutritional status were studied. Stlightly higher prevalence of parasite was observed in summer season (84.30%) followed by rainy (83.63%) and winter (81.16%) season. Females (87.53%) were more susceptible to gastro-intestinal parasitic infection than males (84.37%) and the odds ratio was 1.20. in case of age group, highest prevalence was found in adult (88.81%) followed by young (83.45%) and buffalo calves (68.05%) and which were statistically significant (p<0.05).

**Islam (2012)** conducted a survey on gastro-intestinal parasitic infections in cattle at Vangura upazila, Pabna district, Bangladesh during the period from July to November, 2012 using coproscopy. The effects age and sex on the prevalence of parasitic infections were determined. Among 170 fecal sample examined, 133 (78.24%) were found positive for parasitic infections and mean egg per gram of feces (EPG) was 403.01±31.82. The parasites identified on fecal examinations were snail borne trematodes namely, Fasciola gigantica (4.11%), amphistomes (40%), Schistosoma spp. (5%), nematodes namely, Haemonchus sp. (3.52%), Stronglyles (1.17%), Strongyloides (1.17%), Trichuris sp. (1.76%), Dictyocaulus sp. (0.58%) and cestodes Moniezia spp. (3.52%), and protozoal infections namely, *Balantidium coli* (41.76%) and *Eimeria* spp. (4.11%). Age between calf and adult cattle had significant (p<0.05) influence on the infections with the gastro-intestinal parasites, whereas the sex of the cattle had no significant effect. Lower infection with Moniezia spp. (3.33%) was found in young cattle than in the adults (4.08%). Higher rate of infections was also recorded in females (78.83%) than in the males (75.76%) which was statistically insignificant.

## 2.2 Anthelmintic Effects of Indigenous Medecinal Plants

Mostofa et al. (1983) observed the effect of water extract of Ananus sativus (Anaros) Calamus tennis (Bet) were 48.96% and 58% effective against gastro-intestinal nematodiasis in cattle.

Shilaskar and Parashar (1989) did an experiment on evaluation of indigenous anthelmintics. The *in vitro* anthelmintic activity of 15 extracts of various plants (Andrographis paniculata, Azadirachta indica, Butea frondosa, Caisalpiia crixta, Piper betle, Psoralia corylifolia, Swertia

chirata, Vernenia anthelmintica,) was evaluated against Ascaridia galli. Ether and alcoholic extracts of seeds of B. rondosa and V. anthelmintica, ether extracts of seeds of *C. crista*, successive alcohol extracts of seeds of P. corylifolia and essential oil from leaves of P. betle had highly significant anthelmintic activity.

**Palacpac** (1990) stated that the medicinal plants lagundi (*Vitex negundo*) ipil-ipil (Leucaena glauca), bitter gourd (Momordica charantia) and betal nut (Areca catechu) are used for the treatment of internal parasites in Philipines.

Garg et al. (1992) reported that the essential oil of Piper betle was more effective against tapeworms (Taenia solium) and hook worm the synthetic (Bunostomum trigonocephalum) than anthelmintic piperazine phosphate and hexyliresorcinols.

**Ahmed** et al. (1994) reported that neem seeds @ 100 mg/kg bwt. was 35.95-40% effective against gastro-intestinal nematodes in sheep. The body weight of nccm seeds treated sheep was increased (6.74%). The decreased live weight value was recorded in untreated sheep.

Satrija et al. (1995) tested the efficacy of papaya latex @ 2, 4, and 8 g/kg bwt.) againsi Ascaris suum in 16 pigs. The eggs per gram (epg) on days 0, 1, 5, and 7 were determined using a modified McMaster technique and the adult worms were collected and sexed at necropsy on day 7. The 4 g/kg and 8 g/kg bwt. treatments significantly decreased the epg produced (by 99%) and the number of adult worms by 80 and 100%, respectively.

**Khatun** et al. (1995) showed that anthelmintic efficacy of the aqueous leaf extract pineapple @100 mg/kg, @150 mg/kg and @200 mg/kg bwt. were 53.0%, 57.0% and 59.4%, respectively against gastro-intestinal nematodes in goats. The body weight, TEC and Hb value were increased significantly at 21st day of post-treatment.

**Mostofa** et al. (1995) studied the effects of neem (Azadirachta indica) seed @100 mg/kg and @150 mg/kg bwt. in goat and found 40% and 52.77% effective against gastro-intestinal nematodes in goat.

Aye-Than et al. (1996) was investigated the anthelmintic property of Myanniar pineapple. In the *in vitro* test, the pineapple juice was used at concentration equivalent to 20, 40 and 80 mg/ml in residual form, on 10live fresh Ascaris suum at each concentration, and the 2 higher concentration were found to produce immobilization of the worms leading to death within 16 hours at 40 mg/mi and 11.5 hours at 80 mg/mi.

**Hossain** et al. (1996) observed the effects of neem leaves @ 100mg/kg bwt. and neem seed kernels @ 150mg/kg bwt. in cattle. Seed kernel was more effective (46.60%) than leaves (9.09%) against gastro-intestinal nematodes. Neem leaves and neem seed kernels also increased body weight and Hb content of cattle.

Mostofa et al. (1996) stated that efficacy rate of imported anthelmintics between 80 and 100%. Thiophanate is reported to have reduced efficacy for nematodes, particularly H. contortus. Studies on seeds of the indigenous medicinal plant Neem (Azadirachta indica) have shown an anthelmintic efficacy of 35 to 53% against natural infections of gastrointestinal nematodes.

Chybowski (1997) studied the anthelmintic activity of garlic extracts and observed that extracts of *Allium salivum* and *Allium arsinum* were highly effective against free living nematode Rhabditis sp., larvae of Nippostrongylus brasiliensis and eggs of Ascaris suum.

Vieira et al. (1999) observed the anthelmintic efficacy of 9 plants (Allium sativum, Carica papaya, Musa acuminata, Carnavatia brasiliensis, Momordica charantia, Annona squamosa, Menta sp., Chenopodium ambrosioides and Hymenaea courbaril) against gastro-intestinal nematodes. Only Mentha sp. significantly reduced faecal egg output. In combination with 7.5 mg/kg closantel, neither of the plants or 3.8 mg/kg albendazole significantly reduced faecal egg output.

Singh and Nagaich (1999) reported that the aqueous seed extracts of Carica papaya caused 100% morality after incubation times of 9, 7 and 5h for Ascaridia galli and 7, 5 and 3 h for Heterakis gallinae at concentrations of 6, 8 and 12%, respectively. Controls retained normal appearance and motility.

Singh et al. (2000) reported that 2%, 4% and 6% garlic oil caused mortality in Ascaridia galli after exposure for 12, 10 and 8 hours, respectively and in *Heterakis gallinae* after exposure for 10, 8 and 6 hours, respectively. Garlic extract also reduced significantly the glucose uptake, glycogen content and oxygen consumption in both parasites.

**Arunachal** et al. (2002) observed the anthelmintic efficacy of aqueous extracts of neem leaves, seeds and bark in sheep. Neem leaves, seeds and bark were 53%, 49% and 38% infective against gastro-intestinal helminths in sheep.

**Brelin** (2002) reported that neem (Azadirachta indica) as an alternative anthelmintic for helminth control in small ruminants. Twelve sheep were divided into three groups where one was control and the other two received different amounts of fresh neem leaves; he found that the fresh neem leaves significantly reduced the numbers of H. contortus in the abomasums of the treated sheep.

**Rahman** (2002) examined the effects of aqueous extract of 7 plants in goat. Korolla showed highest anthelmintic effects (67%) than neem (62%), garlic (60%), betel leaves (58%), jute (42%) and katakhura (37%) on day 21 post-treatmept. Hatishur had no activity against gastrointestinal nematodes of goat on day 21 post-treatment. TEC, Hb content and PCV were increased in all treated goats on days 21 post-treatment. On the other hand, TEC decreased on 21 clay post-treatment.

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 contortus* 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 3 mg/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 1 g/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 28 mg/kg BW of an aqueous ethanol extract of the seeds of Caesalpinia crista L. (Caesalpiniaceae); group F with 25 mg/kg BW 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 faecal egg counts (100%) and a 78.2 and 88.8% reduction of adult H. contortus and T. colubriformis on day 13 posttreatment. 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 gastro-intestinal trichostrongylids in small ruminants.

Githiori et al. (2004) evaluated 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 africana, Annona squamosa, Ananas comosus, Dodonea var. angustifolia, Hildebrandtia sepalosa and Azadirachta indica were tested in 151 lambs in 3 experiments. Plant preparations O. europaea 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 efficacy of Pineapple leaves extract was compared with three modern anthelmintics; ivermectin, albendazole and piperazine citrate mainly on the basis of fecal egg count reduction against ascariasis. Calves (n=25) naturally with ascarids were divided into five equal groups. Groups A, B, C and D were treated with ivermectin (200) mg/kg; sc) albendazole (7.5 mg/kg PO) piperazine citrate (200 mg/kg; PO) and Pineapple leaves extract (1gm/kg; PO) respectively and group E was kept as untreated control. The efficacy of ivermectin, albendazole, 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 was increased significantly in comparison to control group. Total erythrocyte counts (TEC), haemoglobin (HB) and packed cell volume (PVC) increased whereas erythrocyte sedimentation rate (ESR) decreased significantly following each erythrocyte is suggested rate ivermectin, albendazole and piperazine citrate and Pineapple leaves extract effective against ascariasis in calves. Although efficacy of Pineapple leaves extract is not encouraging, this extract may be used as substitute for patent drugs.

Batatinha et al. (2004) examined the effects of garlic juice (Allium sativum) on gastro intestinal nematodes of goats. The percentage reductions in egg and larvae counts of the Strongyloidea were under 95%.

Rob et al. (2004) observed the anthelmintic efficacy of aqueous extracts of neem (Azadirachta indica) leaves in sheep. Neem leaves was 53.72% infective against haemonchosis in sheep. TEC, Hb content and PCV were also increased in neem treated sheep on days 28 post-treatment.

**Brahmachari** (2004) stated that neem (Azadirachta indica A. Juss.) has universally been accepted as a wonder tree because of its diverse utility. Multidirectional therapeutic used of neem have been known in India since the Vedic times. Besides its therapeutic efficacies, neem has already established its potential as a source of naturally occurring insecticide, pesticide and agrochemicals. Safe and economically cheaper uses of different parts of neem in the treatment of various diseases and in agriculture are discussed in this article. It further deals with the active chemical constituents of various neem formulations. Commercially available neem products are also mentioned along with their respective applications. Furthermore, evaluation of safety aspects of different parts of neem and neem compounds along with commercial formulation are also taken into consideration. Systematic scientific knowledge on neem reported so far is thus very useful for the wider interests of the world community.

Mostofa and Amin (2005) examined the effects of 10% water extract garlic on gastro-intestinal nematodes of sheep. A significant reduction 44.01%, 43.29%, 40.88% and 38.70% of EPG count was found on 7th, 14th, 21st and 28th day of garlic treated sheep. The EPG count of untreated control group was significantly increased about 7.88%, 11.45%, 20.15% and 30.26% on 7<sup>th</sup> 14<sup>th</sup> 21<sup>st</sup> and 28<sup>th</sup> day, respectively. After treatment with garlic, total erythrocyte count, haemoglobin content and packed cell volume were increased significantly in sheep. On the other hand total leukocyte count was decreased significantly in all treated sheep. The body weight was increased significantly in garlic treated sheep. On the other hand, body weight was decreased in untreated control group.

**Khalid** et al. (2005) stated that neem and pineapple (10% water extract of leaves) reduced significantly EPG count 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 EPG count of untreated control group were significantly increased about 7.88%, 11.45%, 20.15% and 30.26% on 14<sup>th</sup>, 21<sup>st</sup> and day, respectively. After treatment with necm and pineapple, total erythrocyte could (TEC), haemoglobin (Hb) content and packed cell volume (PCV) were increased significantly in sheep. On the other hand total leukocyte count (TLC) was decreased significantly in all treated sheep. The body weight was increased significantly in neem and pineapple treated sheep. On the other hand body weight was decreased in untreated control group.

Hoque et al. (2006) found the comparative efficacy of Piperazine citrate, Levamisole and Pineapple leaves extract as anthelmintics against ascariasis in chicken. The chickens were equally divided into 4 (A, B, C and D) groups. Group A was kept as infected control, while group B, C and D were treated with Piperazine citrate, Levamisole and Pineapple leaves extract, respectively. Although Pineapple leaves extract showed less effectivenss 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) significant (p< 0.01) increased and erythrocyte sedimentation rate (ESR) and total leukocyte country (TLC) significantly (p<0.01) decreased. Body weight was found to be increased following administration of Piperazine citrate, Levamisole and Pineapple leaves extract.

Bhalke et al. (2006) examined the bark of Annona reticulata was collected from Ahmednagar, Maharashtra, India, in December 2005, to determine the anthelmintic activity of its petroleum ether, ethylacetate and methanol extracts against adult Indian earthworms (Pheretima posthuma), which resemble, both anatomically and physiologically, the intestinal roundworm parasites of humans. The extract was applied at 20, 40 and 60 mg/ml. The bark extract showed a dose-dependent anthelmintic activity against the tested earthworms. Among the various extracts, the methanolic extract required less time for paralysis and death of the worms compared with the other extracts and the standard drug albendazole.

Sufian et al. (2006) found that the efficacy of albendazole, ivermectin and whole korolla fruit extract on body weight grain as well as on hematological parameters, changes after treating of naturally infested chicken. Accordingly, forty chickens that aged between 60 to 90 days were selected by examining fecal egg count. The chicken were equally grouped into 4 ('A', 'B', 'C' and 'D') and 'B', 'C' and 'D" were orally treated with albendazole, ivermectin and whole korolla fruit extract as per dosage schedule and Group-A was kept as control. The efficacy of oral administration of albendazole, ivermectin and whole korolla fruit extract were evaluated by observing body weight. The live body weight was increased in Chicken after administration of anthelmintics such as albendazole, ivermectin and whole korolla fruit extract in groups 'B', 'C' and 'D' respectively. But in control group 'A' body weight of chicken was slowly decreased. Increase values of TEC, Hb and PCV were observed the chicken after treatment with Almex® (50 mg/kg.b.wt.), Vermic® (500  $\square$  g/kg b.wt.) and whole korolla extract (1 gm/kg b.wt.).

Sujon et al. (2008) performed a detailed investigation with the aim to find out the indigenous medicinal plants having anthelminthic action. Ten (10) indigenous medicinal plants were primarily selected and the ethanol extracts were prepared for anthelminthic trial and determination of anthelminthic properties in vitro and in vivo against the gastro-instestinal nematodes in goat during the period from July 2006 to December 2006. Screening of ethanol extracts of selected plants showed the anthelmintic activity against gastro-intestinal nematodes at lower concentration (50 mg/ml). *In vivo* screening (by oral administration) of four plant extracts (ethanol) showed variable degree of efficacy in experimentally infected goats, as measured by faecal egg count reduction test. A relatively higher efficacy was recorded in ethanol extract of neem treated animals in comparison to other plants extracts. Ethanol extracts of korolla also showed significant efficacy. 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. These findings indicate that the adult gastro-intestinal nematodes are more vulnerable to selected indigenous plants. Within these ten (10) plants 4 showed more than 70% efficacy at a concentration of 100mg/mkg.

Amin et al. (2008) conducted the experiment in the Department of Pharmacology, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh, for a period of 28 days to study the effects of garlic (Allium sativum, Linn., @100mg/kg bwt.), turmeric (Curcuma longa, Linn., @100mg/kg bwt.)Â and betel leaf (Piper betle, Linn, @100mg/kg bwt) against natural gastro-intestinal nematodes in cattle. The effects of garlic, turmeric and betel leaf on some hematological parameters (TEC, Hb, PCV and TLC), biochemical parameters (ALT and

AST) and clinical parameter (body weight) were also observed. Twenty four (24) naturally parasitized cattle of BAU Dairy Farm, Mymensingh were randomly divided into four groups, each consisting of six (6) cattle. Water extract of bulbs of garlic were administered orally to the cattle of group A. Cattle of group B received orally water extract of rhizome of turmeric. Cattle of group C were treated orally with leaves of betel leaf. Cattle of group D was kept as infected control group. Fecal samples, body weight, hematological and biochemical parameters were examined before treatment and on 3<sup>rd</sup>, 10<sup>th</sup>, 17<sup>th</sup> and 28<sup>th</sup> day. A significant (p < 0.01) reduction of EPG count was found following administration of garlic (20.41-40.81%), turmeric (6.09-19.27%) and betel leaf (2.91-9.8%) in cattle. The EPG count of the control group (D) were significantly (p < 0.01) increased up to the last day of experimental period. After treatment with garlic and turmeric total erythrocyte count (TEC), hemoglobin (Hb) content and packed cell volume (PCV) were gradually increased significantly (p<0.01 and p<0.05) in cattle. Conversely, the total leukocyte count (TLC) were decreased significantly (p < 0.01 and p < 0.05) in treated cattle. On the other hand, TEC, Hb content and PCV were gradually increased significantly (p < 0.01 and p < 0.05) on day 3 and day 10 in betel leaf treated cattle but decreased on 17<sup>th</sup> and 28<sup>th</sup> day. Conversely, the total leukocyte count (TLC) were decreased significantly (p < 0.01) and p < 0.05) on day 3 and day 10 in betel leaf treated cattle but increased significantly (p < 0.01) on  $17^{th}$  and  $28^{th}$  day. The alanine aminotransferase (ALT) and aspartate aminotransferase (AST) level were not significantly changed in the cattle. The body weight was increased significantly (p < 0.01 and p < 0.05) in garlic, turmeric and betel leaf treated cattle. On the other hand, body weight was decreased in untreated

control group. The present study reveals that water extracts of garlic were moderately effective and turmeric and betel leaf were relatively less effective against gastro-intestinal nematodes in cattle.

Rahman et al. (2008) found that the aqueous extract of pineapple leaves patent anthelminics albendazole (Benazol®) and ivermectin (Vermic®) were assessed against acaridae in indigenous chicken. Among these, extract of pineapple laves was administered @ of 1 gm/kg body weight for seven consecutive days while albendazole @ full 500 mg/kg body weight for 3 days, and invermectin @ 0.2 mg/kg body weight s/c given as a single does. Results showed that pineapple leaves extract caused gradual increase of 100% efficacy up to day 28th of treatment. Haematological anlaysis revealed that the aqueous extract of pineapple leaves did not show any adverse effect on blood parameter in treated birds. But Benazol® and Vermic® were effective. Pineapple leaves is considering more suitable one. It should be given conclusion that pineapple leaves were effective against asscariasis in chicken.

Shahadat et al. (2008) examined anthelmintic efficacy of whole Korolla fruit (Momordica charantia) extract and ivermec® pour on was evaluated in vitro and in vivo on adult Ascaridia galli of indigenous chicken. The total trial chickens (60) were divided equally into 3 groups; group A as control, group B treated with Ivermec® pour on @ 500 µg/kg bwt by dropper through skin absorption for single dose and group C treated with 3% aqueous extract of Korolla. Freshly prepared aqueous extract of the Korolla fruit was performed as wormicidal properties against adult A. galli on in vitro and in vivo study. 3% aqueous extract of Korolla fruit was treated as higher efficacy against A. galli. The live body weight was increased in chicken after treatment in group B and C respectively but in control group body weight was slowly decreased. TEC (million/cu mm), Hb (gm %) and PCV (%30 minutes) were increased significantly in chickens of treated groups whereas ESR was increased in control groups.

Aziz (2008) observed the incidence of gastro-intestinal nematodes in calves and average infection rate was found to be 94%. The efficacy of administration of Levavet® (7.5 mg/kg bwt orally), Vermic® (200 µg/kg body weight) and Pineapple leaves extract (1 mg/kg body weight orally) were evaluated by observing body weight. The live body weight was increased significantly in calves after administration of anthelmintics e.g. Pineapple leaves extract, Vermic® and Levavet® in groups 'B', 'C' and 'D', respectively. Though, in control group 'A' the change of body weight was not considerable significant. Increased values TEC, Hb and PVC were observed in calves after treatment with Levavet® (7.5 mg/kg bwt), Vermic<sup>®</sup> (200 µg/kg bwt) and pineapple leaves extract (1 gm/kg bwt). Whereas ESR values decreased not significantly in all treated groups. But TEC, Hb and PVC were decreased and ESR was slightly increased in control group 'A'.

Amin et al. (2009) observed the prevalence of natural gastro-intestinal nematodes in cattle during the period from June, 2004 to May, 2005 in Sadar upazila of Mymensingh district. The prevalence of gastro-intestinal nematodes was 84.1% (rainy seasons-97%, summer-85.5% and winter seasons-69.8%). The prevalence of strongyles (Haemonchus Trichostrongylus sp., Oesophagostomum sp. and Mecistocirrus sp.), Bunostomum sp., Strongyloides sp., Trichuris sp. and Capillaria sp. were 63.9%, 26.3%, 21.5%, 17.3% and 24.5%, respectively. 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%.

Amin et al. (2010) studied the effects of neem, betel leaf, devil's tree, jute and turmeric against natural gastro-intestinal nematodes in sheep and on some hematological parameters (TEC, Hb and PCV) and body weight were studied. Thirty (30) naturally parasitized sheep were randomly divided into six groups (A, B, C, D, E and F), each consisting of five sheep. Ten percent water extract of leaves of neem, betel leaf, devil's tree and jute were administered orally to the sheep of group A, B, C and D, respectively. Sheep of group E was treated orally with 10% water extract of rhizome of turmeric. Sheep of group F was kept as infected control group. Fecal samples, hematological parameters and body weight were examined before treatment and on 7th, 14th, 21st and 28th day. A significant (p<0.01) reduction of EPG count was found following administration of neem (37.60-47.03%), betel leaf (6.43-14.00%), devil's tree (3.04-11.04%), jute (0.50-5.26%) and turmeric (0.46-8.30) in sheep. The EPG count of the control group (F) were significantly (p<0.01) increased up to the last day of experimental period. After treatment with neem, betel leaf, devil's tree, jute and turmeric total erythrocyte count (TEC), hemoglobin (Hb) content and packed cell volume (PCV) were increased significantly (p<0.01 and p<0.05) in sheep. The body weight was increased significantly (p<0.01 and p<0.05) in neem, betel leaf, devil's tree, jute and turmeric treated sheep. On the other hand, body weight was decreased in untreated control group. The present study reveal that 10% water extracts of neem was moderately effective and betel leaf, devil's tree, jute and turmeric were relatively less effective against gastro-intestinal nematodes in sheep.

Begum et al. (2010) conducted the experiment at the Department of Pharmacology, Bangladesh Agricultural University, Mymensingh, during the period from July to November 2009 to study the prevalence of ascariasis in village poultry at sadar upazila of Mymensingh district in Bangladesh. Survey was conducted on 10 village poultry. Prevalence of ascariasis was recorded on the basis of fecal egg counts in (80.47%) at the age of 3-4 months. The aqueous extract of Pineapple leaves and patent anthelmintic Piperazine (Piper-vet®) were assessed against ascariasis in indigenous chicken. Pineapple leaves was administered @ 1 gm/kg body weight and reduced EPG to 0 at 21st day and Piperazine reduced EPG to 0 at 7th day. Hematological analysis revealed that the aqueous extract of Pineapple leaves did not show any adverse effect on blood parameter in treated birds. Increase values of TEC, Hb and PCV were observed the chicken after treatment with Neem at dose rate 20mg/ml. Bishkatali leaves extract were incubated at the dose rate of 0.1-1.0 mg/ml to 1mg/ml. Piperazine were incubated at the dose rate of 0.1-4.0mg/ml to 4mg/ml. Levamisole were incubated at the dose rate of 0.1-0.6mg/ml to 0.6mg/ml.

Mian (2011) Conducted study to determine the comparative efficacy of Neem leaves powder and Ivermectin on the performance of body weight gain of bull. 1.5 years (15 calves) old calves were divided into three groups, I<sub>0</sub>, I<sub>1</sub> and I<sub>2</sub> which were supplemented with neem leaf powder@ 100 mg/kg bwt and Ivermectin@ 200 µg/kg bwt, sc. Weekly observations were recorded for live body weight, weekly gain in weight, weekly feed consumption, feed efficiency and blood parameters of calves for six months. All the treatment groups  $I_1$  (125) and  $I_2$  (106.01) recorded significantly higher means for live body weight than that of control  $I_0$ (60) group.

## 2.3 Anthelmintic Effects of Ivermectin

**Kashai** et al. (1984) carried out Ivermectin trials in cattle and pigs infected with endo-and ecto-parasites in Hungary and reported that a S/C infection of Ivermectin at 0.2 mg/kg b.w. cured 65.2% young cattle mixed strongly infection of 142 to 250 EPG, as do judged by fecal tests on day 19 after treatment. The weight gain of the treated group exceeded those in untreated groups by 6.8 kg. Ivermectin at the same doses reduce adult Ivermectin reduced Ascaris suum infection intensity by upto 99%. Ivermectin (as ivomec<sup>®</sup>) was also effective against psoroptic mange in cattle.

Corba et al. (1984) tested Ivomec® (As Ivermectin) as 1% injection on Czechoslovakia against natural gastro-intestinal nematode and pulmonary nematoda infection in sheep (400 µg/kg b.wt.) and in pig (300 µg/kg b.wt.) and killed the animal on 7 days post treatment of examination. He stated that in sheep Ivomec was 100% effective against Bunostomum trigonocephalum, Trichostrongylus spp. and Ostertagia spp. 77.80% against Muellerius casillaries. The cure rates for 10 pigs in poor condition were 99.2% for Metastrongylus sp. 98.2% for Ascaris suum 65% for O. dentatum and 64.6% for T. suis. Eggs were absent from faeces on day 3 after treatment. The heavy ectoparasitic infection presents (130 Hoemalopinus suis/animal) was also controlled. No side effects were observed following the therapy.

Nadykto et al. (1986) studied the efficacy of Ivomec against lung gastrointestinal nematode in sheep with gastro-intestinal infection in the Omsk region, USSR and reported that Ivomec was 100% effective against Dictyocaulus viviparus, Strongyloides, Nematodius and other Trichostrongylus and slightly less effective at higher dose. In a comparative study with Nilverm at the recommended doses the authors reported that Ivomec was more effective than Nilverm.

Arlian et al. (1988) reported that ivermectin treated rabbit have an increase level of haemoglobin, haematocrit and mean corpuscular hacmoglobin concentration (MCHC) in comparison to those of untreated controlled rabbit.

Oku et al. (1988) reported 99% effectiveness of ivermectin in the treatment of gastro-intestinal nematodes of calves at the dose rate of 200 μg/kg body weight.

**Zimerman** et al. (1988) reported 98% effectiveness of ivermeetin at the dose rate of 200µg/kg, body wt. against *Nematodius* spp. in calves. Effectiveness was measured by observing reduction of faecal egg output.

**Soll et al.** (1988) found 100% prophylaxis efficacy of ivermectin against nematode infestation in cattle. Ivermectin was given at 8mg/days for 120 days.

Shekhovtsov et al. (1988) observed 92.6% effectiveness of ivermectin against gastro-intestinal nematodes (Haemonchus contortus, Cooperia oncophora, and Ostertagia ostertagi) in sheep.

Vercruysse et al. (1988) found that the faecal out put of nematode egg was much lower in both treated groups (levamisole and ivermectin) of cattle during the study of efficacy of early season anthelmintic treatment against nematodes.

**Ponikarov** (1989) reported 100% effectiveness of Ivomec-F (ivermcctin) at the dose rate of 1ml/50kg body weight by giving subcutaneous injection against gastro-intestinal nernatodes.

Bauck et al. (1989) observed that ivermectin treated calves gained an increase average body weight per day in comparison to the control animals. The average daily weight gained by iverrnectin treated cattle was recorded as 0.08 kg for each individual animal.

**Abdalla (1989)** reported 100% efficacy of ivermectin be faecal egg count, against gastro-intestinal nematodes (Haemonchus SL Oesophagostomum V, Trichostrongylus W, strongyloides, Oestrus ovis Monizia, Cooperia, chaberitain) in sheep and cattle during the study of the effect of endoparasites on growth rate.

Shastri (1989) reported 97.5% and 92.3% efficacy of ivermectin against gastro-intestinal nematodes in goat.

Williams et al. (1989) observed the high effectivness of Ivermectin (Ivomec®) against gastro-intestinal nematodes in beef cattle. For this purpose, 16 cross breed beef calves were divided into four groups. Three

groups were treated with ivermectin and one group was treated with fenbendazole. Initially high faecal egg counts at wearing were more effectively reduced by ivermectin than fenbendazole.

Echevarria and Trinadade (1989) reported 100% effectiveness of ivermectin in removing Ostertagia circumcinta, Trichostrongylus axei, Trichostrongvlus colubriformis, Cooperia Nematodirus puntata, spathiger, Trichuris ovis and developing and early fourth stage larvae of Haemonchus contortus but only 59.4% and 85.7 effective in removing adult and maturing adult *H. contortus* respectively in sheep.

Gill et al. (1989) reported that infection of Neoascaris vitulorum. Trichostrongyloidae, Oesophagustomum sp. and Bunostomum were eliminated within one week of treatment with ivermectin in Buffalo. They also found that the mites disappeared within 2 weeks after treatment.

**Kennedy** (1990) studied the effect of Ivermectin on gaining body weight of calves in USA. Calves treated with Ivermectin indictable preparation (200μg/kg, S/C) significantly gained more weight than the control animals.

Bagherwal et al. (1991) studied the efficacy of ivermectin against naturally acquired nematodiasis in goats. For this purpose, 47 goats were examined for suspected parasitic infections, 43 (91.7%) had a mixed infection with Trichostrongylus V, Haemonchus spp. and Trichuris spp., A single subcutaneous injection of 0.2 mg/kg body weight of ivermectin in 37 animals was successful in eliminating the clinical signs and reducing the faecal egg count to 0 (zero) in 34 animals and to 8.2% (Trichuris spp.) in the other 3 to 5 days per infection. By 7 days all treated animals were negative.

Tada et al. (1992) reported the 100% efficacy of ivermectin against gastro-intestinal nematodes in sheep and goat of Zambia. Fourteen goats and 5 ewes were treated with subcutaneous injection of ivermectin at a dose rate of 200 µg/kg body weight. The animals were naturally infected with. Haemonchus contortus, Trichostrongylus, Oesophagostomum and Strongyloides V. The efficacy of ivermectin was 100% during faecal oval count on 7th and 14th day after treatment.

**Duncan and Forbes (1992)** reported that the mean live weight gain of ivermectin treated young beef cattle was significantly greater than oxfendazole treated group.

Basu et al. (1992) Studied the efficacy of Ivomec pour on (Ivermectin), 500 μg/kg body weight) as ideal anthelmintic for the treatment of gastrointestinal nematodes in Nigerian cattle. They observed a significant reduction in the average faecal egg counts at 5, 11, 29 and 56 post treatment days in the treated cattle in comparison to untreated control animals. The efficacy ranged from 96.0% at day 5 to 94.7% at day 56.

Anil Kumar and Joshi (1992) studied the anthelmintic activity of ivermectin and their effect on haemoglobin and body weight in sheep, naturally infected with gastro-intestinal nematodes. Ivermectin (at the dose rate of 200 µg/kg body weight subcutaneously) was 100% effective in the elimination of gastro-intestinal nematodes in 22 ewes. The EPG count was found negative in the treated animals following treatment with Ivermectin till 28 days. Hb and body weight measurements at day 0 (zero) and day 28th post treatment showed an improvement in the treated sheep.

Chandrawathani and Sani (1993) reported a higher weight gain of 0.28 and 0.25 kg/day following treatment with ivermectin and oxfendazole respectively in 8 to 12 months aged calves having *Haemonchus* contortus, Strongyloides papillosus, Moniezia and Toxocara infection than untreated controls. They used ivermectin and oxfendazole subcutaneously at the dose rate of 220 µg/kg body weight and 4.5 mg/kg body weight respectively.

**Taylor** et al. (1993) conducted an experiment to find out the effect of anthelmintics on gaining body weight of calves suffering from G.I. nematodes. They reported that ivermectin have a positive effect on gaining body weight in cattle.

**Pradhan** et al. (1993) reported the 99.8% and 42.2% average efficacy Ivermectin against *Neoascaris vitulorum* and non-Neoascaris infection respectively in calves, during the study of comparative efficacy of 2 anthelmintics in calves.

**Vassilov** (1993) found that treatment with ivermectin of 2 year old beef cattle were more effective (gain 25.5 kg over the control animals) than treatment with albendazole (gain 12.9 kg more weight than control animal) in the enhancement of productivity and protection from the effects of nematode infections.

Charles and Medeiros (1993) reported 85.5% mean efficacy of ivermectin against total worm population and 87.8% efficacy for Haemonchus contortus infection in goat.

Singh et al. (1993) reported the highest efficacy of Ivermectin among five anthelmintics in sheep naturally infected with gastro-intestinal nematodes. The authors recorded 97.91 % reduction strongyle egg out put in sheep treated with ivermectin.

Rossanigo and Silva (1993) treated one group of goats with ivermectin monthly (GI), another strategically for 2 months (G2) and lift another group as untreated control (G3) and observed live weight gain GI adults were 5.92 kg heavier than G2 adults and 4.21 kg heavier than G3 adults.

Chartier and Pors (1994) studied the efficacy of ivermectin against gastro-intestinal nematodes in goats by a faecal egg count reduction test (FECRT). Goats were treated with ivermectin at a dose rate of 0.2 mg/kg body weight orally.

Mukherjee et al. (1994) reported the 100% effectiveness of ivermectin against all types of nematodes (*Trichostrongylus* spp. *Strongyloides* spp. and *Nematodirus* spp.) in goats during a comparative study of 3 anthelmintic in Cashmere goats.

Singh et al. (1994) reported the 100% effectiveness of ivermectin in reducing the output of ova by the nematode worms in sheep and goats during a comparative study of 4 anthelmintics against natural nematode infection.

Smith (1994) reported the highly effectiveness of ivermectin against Cooperia spp., Dictyocaulus spp, Haemonchus spp, Ostertagia spp. and Nematodirus spp. (as judge by faecal egg counts and larval culture) in cattle and it resulted an average 20.7 kg weight gain.

Stacey et al. (1995) reported that the calves treated with Ivermectin had significantly gained higher weight than untreated calves.

Taylor et al. (1995a) studied the effect of ivermectin on body weight in cattle. Four groups were taken; one of the heavier and one of the lighter groups were treated with topical formulation of ivermectin. The treatment of die heavier group had only relatively minor parasitological effects and no effects on weight gains the treatment group of the lighter cattle resulted in increased weight gain due to effective nematode control.

**Taylor** et al. (1995, b) reported the effectiveness of ivermectin against Ostertagia ostertagi and Cooperia oncophora in cattle, during a combative. Study of early and mid grazing season suppressive anthelmintic treatment for first year grazing season calves and their effects on natural and experimental infection. Five groups of cattle were taken and groups 2 and 3 were treated by ivermectin and group 4 and 5 were treated with morantel. Over the 2 years observation period groups 2 and 4 showed significantly better weight gain than other groups.

Srivastava et al. (1995) reported the high effectiveness of ivermectin against gastro-intestinal nematodes of sheep, resistant to fenbendazole, tetramisole and oxyclozanide in sheep breeding farm, chamoli (Uttar Pradesh).

Partani et al. (1995) reported 98.96%-99.43% efficacy of ivermectin against a mixed infection with *Haemonchus longisitipes*, *Trichostrongylus* spp., Strongyloides papillosus, Nematodirus spp. and Trichuris spp. in camel during the study of a comparative efficacy of 5 anthelmintics.

Gogolewski et al. (1995) reported above 99% efficacy of ivermectin against all nematode species (Oesophagostomum columbianum,

Haemonchus contortus, Ostertagia circumcincta and Nematodirus *spathiger*) in sheep.

Stacey et al. (1995) reported that the calves treated with ivermectin significantly gained higher weight than untreated calves.

**Dorny** et al. (1995) reported more than 95% efficacy of ivermectin against strongyles infection of sheep during the study of the efficacy of four anthelmintic against strongyles infections of sheep.

**Dacastor and Cocuza** (1995) reported that ivermectin was highly effective at the dose rate of 200 µg/kg body weight against gastro-intestinal nematodes in goat.

Kochapakdec et al. (1995) reported that the ivermectin was highly effective against nematodes *Haemonchus* spp. and *Trichostronylus* spp. in goat in southern Thailand. In the study, the other drugs albendazole, fenbendazole, and levamisole showed resistance against nematodes (Haemonchus spp. and Trichostrongylus spp.).

Hang et al. (1995) reported more than 99% efficacy of ivermectin against Oestertagia oestertagi at 28 days after treatment during the study of persistent activity of ivermectin topical and moxidectin injection against Oestertagia ostertagi and Dictyocalus viviparus in calves.

**Purvis and Whittier (1996)** found the decreased fecal egg output after injection with Ivermectin in cattle. Treated heifers had gained greater weight than control heifers.

Waruiro et al. (1996) observed 99.3% efficacy of ivermectin against the thiabendazole resistant *Haemonchus contortus* in sheep during a comparative study of six anthelmintics. Ivermectin was given at the rate of 0.2 mg/kg body weight.

Bliss and Sizelove (1996) reported the comparative field efficacy of two, anthelmintics (Levamisole and Ivermectin) topical pour on formulation in cattle. Twenty-six naturally infected cattle and 433 naturally infected buffalo were selected for 2 field efficacies Animals in first treatment group of each study received levamisole pour on (Talator pour on Deworm). The 2<sup>nd</sup> group received ivermectin pour on (Ivomec) Tralaton treatment removed parasites and reduced faecal egg counts earlier than Ivomec. In both studies Cooperia W, Haemonchus spp., Trichostrongylus spp. complex with some *Nematodirus* spp. and *Oesophagostomum* spp. were present. Cooperia appeared to be the most prevalent parasite after treatment.

Vercruysse et al. (1996) reported the absence of clinical sign of parasitic gastroenteritis after treatment with ivermectin in calves. Here ivermectin was given at the dose rate of 500 µg/kg body weight.

Schnider et al. (1996) reported the effective reduction of gastro-intestinal nematode eggs out put after treatment with ivermectin in grazing cattle.

**Arantes** et al. (1996) observed more than 99% efficacy of ivermectin at the dose rate of 200 µg/kg body weight (Subcutaneously) against gastrointestinal nematodes (Adult Haemonchus placei, Trichostrongylus spp., Trichostrongylus culumbriformis Bunostomum phlebotomum, Cooperia punctata, and Oesphagostomum radiatum) in calves. Exanimation was done by slaughtering the cattle.

Pramanik et al. (1996) reported 100% efficacy of ivermectin against *Trichostrongylus Oesophagostomum* Haemonchus spp spp, Bunostomum spp, Trichuris spp, Gaigeria pachyscelis, and Strongyloides spp in goat on day 5 and 7-post treatment. Efficacy was determined by faecal egg count reduction.

**Ballwebner** et al. (1997) studied the average daily weight gain of cattle treated with ivermectin. The average daily weight gain of each individual was recorded as 0.152-0.272 kg compared to nontreated controls. The difference was statistically significant (p<0.05) in 3 of the 4 studies.

Imrul (1997) reported that the efficacy of ivermectin against gastrointestinal nematodiasis and ectoparasites like ticks and lice was 100% in cattle. Body weight of ivermectin treated cattle was increased by 6.205% on day 28-post treatment. Hb, PCV, and ESR values were changed significantly after treatment with ivermectin in cattle.

Ryan et al. (1997) conducted an experiment to determine the mean total weight gain of Ivermectin treated calves naturally infected with helminth The mean total weight gain was 33.9 kg greater for the Ivermectin treated calves than those of untreated control animals (p<0.02); a 34% increase. It was concluded that the Ivermectin S/C influenced the weight gain by limiting the impact of subclinical parasitism.

Yazwinski et al. (1997) reported 100% effectiveness of Ivermectin against infection. No adverse reaction was observed in trial animals.

Waruiru (1998) evaluated the efficacy of 4 groups of anthelmintics against naturally acquired nematode infections in 5 sheep and 4 goat herds. The drugs used were: levamisole @7.5 mg/kg (2 formulations), albendazole @5 mg/kg, refoxanide @7.5 mg/kg, ivermectin @0.2 mg/kg, mebendazole @ 15 mg/kg + levamisole @ 7.5 mg/kg and rafoxanide @11.25 mg/kg + levamisole @ 7.5 mg/kg. Ivermectin was 100% effective in all cases. Albendazole, levamisole, mebendazole+levamisole and rafoxanide+levamisole were >95% effective on all sheep and 3 goat farms; in the fourth goat herd the efficacies were 88.6%, 92.2%, 94.8% and 90.4%, respectively. The efficacy of rafoxanide ranged from 90.8% to 98.5% in sheep and from 77.5% to 95.9% in goats.

Hossen and Mostofa (1999) studied the comparative efficacy of ivermectin @ 200 µg/kg body weight (bwt) at a single subcutaneous injection, tobacco leaves (water extract) @ 50 mg/kg bwt orally thrice at 7 days interval and tobacco-copper sulfate formulation @ tobacco leaves 30 mg + Copper sulfate 30 mg/kg bwt orally thrice at 7 days interval orally resulted 100%, 54.55% and 62.79% reduction of fecal egg count respectively on 28<sup>th</sup> day of post-treatment.

Magona et al. (2000) examined the comparative efficacy of Wormicid Plus<sup>®</sup> (1.5% levamisole ± 8.0% bithionol), Vermitan<sup>®</sup>, and Ivomec<sup>®</sup> (ivermectin) against goat nematodes. Ivomec® Vermitan® and Wormicid® Plus maintained an efficacy of 100% for 7 weeks then dropped to 92, 98 and 70% respectively.

**Bauer** (2001) reported that albendazole (3.8 mg/kg) reduced the mean fecal egg counts by 82% and the mean worm counts of Haemonchus 22%, 10% by *Oestertagia* circumcincta by contortus and Trichostrongylus colubriformis by 41% compared with the controls in an Angora goat herd at northern Hesse in Germany. Ivermectin (0.2mg/kg) was 100% effective.

Singh et al. (2001) examined the efficacy of ivermectin, albendazole, and Nilzan® (Oxyclozanide+ Tetramisole) against CII nematodiasis in Alpine results of **EPG** clinical and oxyclozanide+tetramisole (Nilzan® 15m1/45kg) was the most effective against the gastro-intestinal nematodes, followed by ivermectin (200 mg/kg) and then by albendazole (5 mg/kg body weight).

Stevenson et al. (2002) observed the efficacy of Triclabendazole (Fasinex®) and ivermectin (Ivomec) against liver fluke (Fasciola hepatica) and gastro intestinal nematodes in cattle. Both treatments were highly efficacious (98%) against liver fluke. ivomec<sup>®</sup> groups were 97.6% and 94.2% on day 7, 98.9% d 91% on day 14, and 98.5% and 92.6% on day 21 against gastro-intestinal nematodes.

Hannan et al. (2003) examined that the sent percent efficacy of Ivermectin against ectoparasites (Lice and Ticks) had been evidenced on day 7 of treatment and sustained upto 28 days. Similarly, no nematodal

eggs were found on fecal examination after day '7' of treatment and sustained 28 day of the treatment in sheep.

Khalid et al. (2005) suggested that Vermico may be used as a broad spectrum anthelmintic against gastro-intestinal nematodiasis in sheep. After treatment with Vermic®, Total Erythrocyte Count (TEC), Hemoglobin (Hb) content and Packed Cell Volume (PCV) were increased significantly (P<0.01 and P<0.05) in sheep.

Avcioglu H. et al. (2011) reported that a study was made to investigate efficacy of eprinomectin pour-on against to Toxocara vitulorum in calves. In the study, 16 calves naturally infected with T. vitulorum were divided into two groups as treatment (eight calves) and control (eight calves). Eprinomectin (0.5 mg/kg, Eprinex, Merial) was given to treatment group calves, and eggs per gramme were determined in the faeces on the day of pre-treatment and the second, third, fourth, fifth, sixth, 14th and 28th days of post-treatment. No side effects associated with nervous, respiratory and gastro-intestinal systems were observed. In conclusion, eprinomectin was determined to be 100% effective against T. vitulorum. This is the first study to evaluate the efficacy of eprinomectin against a natural T. vitulorum infection in calves.

#### 2.4 Anthelmintic Effects of Levamisole

**Leguia** et al. (1979) stated that levamisole @4-5 mg/kg bwt. was 97.9% effective against gastro-intestinal nematodes in sheep.

Anderson and Lord (1979) reported the anthelmintic efficiency of oxfendazole, fenbendazole and levamisole against naturally acquired

infections of Trichostrongylus axei in cattle. Efficiency against T. axei exceeded 99% for all drugs.

Guerrero et al. (1984) studied the anthelmintic activity of pour on formulation of levamisole in calves. In the experiment, the efficacy of a single treatment of levamisole pour on @10 mg/kg of bwt. was determined, using a placebo pour on in a similar volume as a control. In the 2<sup>nd</sup> experiment, calves were treated with levamisole pour on @7.5, 10, or 12.5 mg/kg bwt. In the 3<sup>rd</sup> experiment, the anthelmintic efficacy of the optimal dose of levamisole pour on, @10 mg/kg bwt. was compared with that of injectable levamisole at the recommended rate @6 mg/kg bwt. In all 3 studies, levarmsole pour on, @10 mg/kg bwt., was efficacious (90% to 100%) against the adult forms of Trichostrongylus axei, Haemonchus placei, Ostertagia ostertagi, Cooperia spp, Oesophagostomum radiatum, *Nematodirus* spp and *Bunostomum* phlebotomum.

Vazquez et al. (1984) determined the efficacy of four commercial anthelmintics against gastro-intestinal nematodes of sheep. The reductions in Haemonchus spp., Cooperia spp., Oesophagostomum spp. and Trichuris spp. in treated animals were 82.5% for trichlorfon (@22 mg/kg); 90.7% for levamisole (@8 mg/kg); 91.8% for fenbendazole (@5 mg/kg) and 96.2% for albendazole (@5 mg/kg).

Beria Jaya et al. (1986) observed in west Java that weight gain in sheep is significantly increased when treated with levamisole against gastrointestinal nematodes.

Elliott (1987) investigated young goats harbouring 3, 7 and 36 day old experimental infections of gastro-intestinal nematodes, known to be susceptible to anthelmintics in sheep. The drugs and overall removals of Osteriagia circumcincta and Trichostrongylus spp. (T. colubriformis and T. vitrinus) respectively were: morantel citrate 69% and 68%; levamisole hydrochloride 55% and 99%; fenbendazole 98.5% and 99%; oxfendazole 99% and 99%.

Guha and Banerjee (1987) stated the efficacy of four anthelmintics in the treatment of gastro-intestinal nematodes of goats. Albendazole and thiophanate were 100% effective, levamisole and tetramisole 93.4% and 72.2% effective, respectively.

**Ghose (1987)** observed the efficacy of levamisole HCl in the treatment of gastro-intestinal nematodes in calves. Levamisole hydrochloride as 150 mg tablets given orally to calves @15 mg/kg bwt. was effective against natural infection with *Haemonchus*, *Bunostomum* and *Oesophagostomum*.

Bauer et al. (1988) observed the efficacy of nine anthelmintics (thiabendazoe, @50 mg/kg; mebendazole, @20 mg/kg; fenbendazole, @5 mg/kg; oxfendazole, @5 mg/kg; albendazole, @3.8 mg/kg; febantel, @5 mg/kg; pyrantel tartrate, @25 mg/kg; levamisole, @5 mg/kg and ivermectin, @0.2 mg/kg bwt.) against gastro-Intestinal nematodes in 10 Hessian sheep flocks using the egg count reduction test including larval cultures. Egg count reduction of at least 97% was observed in the two remaining flocks after treatment with each of these drugs with the exception of thiabendazole on one farm, unchanged egg counts persisted in some individual animals after medication with pro-benzimidazoles.

Narasinihan (1988) conducted field trial with levamisole HCl in the treatment of cattle and buffaloes infected with gastro-intestinal nematodes. 37 cows and 32 female buffaloes, naturally infected with gastro-intestinal parasites (Haemonchus sp., Bunostomum sp. and Oesophagostomum sp.) and belonging to a private dairy farm in the suburb of Madras City, India. The animals were divided into 2 groups. Group one consisted of 25 cows and 20 female buffaloes, randomly selected and treated with levamisole HC1 @7.5-10 nig/kg bwt, 12 cows and 12 buffaloes served as controls. On the 7th day post treatment, no nematode ova were detected in faeces of treated animals.

Charles et al, (1989) reported the efficacy of levamisole (@5 mg/kg bwt.), albendazole (@3.8 mg/kg bwt.) and fenbendazole (@ 15 mg/kg bwt.) against gastro-intestinal nematodes in goat. Post-mortem worm counts showed that the reduction in worm burdens was 57.4% in levamisole-treated animals, 71.1% in the albendazole group and 85.11/o in the fenbendazole group. Reductions for H. contortus were 80.2%, 87.9% and 83.90/o, respectively in the levamisole, albendazole and fenbendazole treated goats.

Mutafov et al. (1989) reported levamisole @5 mg/kg bwt. for 6 days were 95.7% effective against gastro-intestinal nematodes and 86.1% effective against lung worms in wild ruminants. Thiabendazole @50 mg/kg bwt for 8 days was 97.07% effective against gastro-intestinal nematodes.

**Redl** (1991) conducted ten field experiments on cattle infected naturally (with Coolieria, Ostertagia, Trichostrongylus, Haemonchus, Bunostomum, Oesophagostomum, Strongyloides) on 3 farms. A total of 477 cattle were treated once or twice with different anthelmintics (albendazole, oxfendazole, tetramisole, levamisole, ivermectin). Treatments were effective and the number of cattle infected with gastro-intestinal helminths and the level of egg production was greatly reduced. Body weight gain was higher in treated animals (1.3 to 17.0 kg) and daily body weight gain by 12-152 g compared with the untreated controls.

Sharma and Jagadish (1991) observed the efficacy of levamisole administered through different routes against gastro-intestinal nematodes in crossbred cattle. Levamisole administered orally @7.5 mg/kg bwt. Subcutaneously @7.5 mg/kg bwt. subcutaneously @7.5 mg/kg bwt. and topically pour-on formulation @10 mg/kg bwt. showed 100% efficacy against *Trichostrongylus* spp. and *Trichuris* spp. on day 7 after treatment in the 18 animals. The faecal egg count in the 6 untreated control animals increased sharply over the same period.

Quiroz-Romero et al. (1992) indicated the effectiveness of levamisole @3.18 mg/kg and @2.70 mg/kg bwt. in calves. PM studies 7 and 8 days after treatment was 100% 111d 85.7% against *Haemonchus* spp., 100% and 100% against Trichostrongylus axei, 100% and 99.36% against Cooperia spp. in the abomasums; 100% and 90.85% against Cooperia spp. in the small intestine; 100% and 100% against Oesophagostomum spp. and Trichuris spp. and 99.5% and 99.16% against D. viviparus respectively.

Pangui et al. (1993) used levamisole in the treatment of gastro-intestinal strongyles in West Africa. Levamisole was given in the form of a bolus to 4 N'Dama and 6 Zebu cattle. Egg counts remained constant for untreated animals, but decreased to 0 after 3 days in treated cattle. They concluded that levamisole @6.6 mg/kg bwt. was effective against all gastrointestinal nematodes (adults and larvae).

Chartier and Pors (1994) reported that levamisole at the dose rate of 11.4 mg/k-g bwt. was 82% effective against gastro-intestinal nematode infection in goats.

Alam and Mostofa (1995) stated the efficacy of levamisole and albendazole against gastro-intestinal nematodes in cattle. Levamisole and albendazole were 76.00% and 86.27% effective against gastro-intestinal nematodiasis, respectively in cattle.

Williams and Broussared (1995) found 90% to 100% efficacy of levamisole @10 mg/kg bwt. against gastro-intestinal nematodes in cattle. The overall efficacy of levamisole was generally high (93.0-100% against Haemonchus placei adults, Cooperia punctata C. spatulata adult males, Cooperia adult females, Oesophagostomum radiatum, В. spp. phlebotomum and D. viviparus adults).

Qureshi et al. (1997) studied incidence and chemotherapeutic efficacy of gastro-intestinal nematodes in dairy buffaloes in the North-Western of Pakistan. 46.6% infected with Valley were nematodes: Trichostrongylus (23.40%), Oesophagostomum (18.91%), Haemonchus (13.80%), Neoascaris (13.14%), Ostertagia (9.61 %), Bunostomum (6.41%), Strongyloides (6.09%), and Mecistocirrus (5.12%). Overall the rate of infection was higher in males than females. The efficacies of albendazole and levamisole were 96% and 98%, respectively.

Karim (1997) recorded 80% efficacy of levamisole hydrochloride (Emitrex ® Ethical Drug Lid, Bangladesh) at the dose rate of 7.5 mg/kg body weight against ascariasis in calves.

Gawor and Borecka (1999) observed the efficacy of levamisole 1.5% against gastro-intestinal nematodes in goats. On the 4<sup>th</sup> and 28<sup>th</sup> days after dosing, the efficacy was 98.3% and 76.3%, respectively. It is concluded that the treatment had a high, but short term efficacy against gastrointestinal nematodes in goats.

Magona and Musisi (1999) studied the efficacy of levamisole and albendazole against gastro-intestinal nematodes in goats. They found that albendazole @10 mg/kg bwt. had efficacy rates of 100% and levamisole 7.5 mg/kg bwt. had efficacy rates of 35% against gastro-intestinal nematodes in the goats.

Olaho-Mulani and Kimani (1999) evaluated the efficacy of injectable ivermectin an(levamisole hydrochloride against natural nematode infections (haemonchosis) in dromedary camels and found that one month after single treatment levamisole hydrochloride, 100% reduction in strongly faecal egg count there was only a 71% reduction for ivermectin.

Mattos et al. (2000) stated the efficacy of levamisole @8 mg/kg and 11 mg/kg bwt. and closantel @10 mg/kg bwt. against gastro-intestinal nematodes in naturally infected goats. The reduction in faecal egg count was 93.23% with levamisole @8 mg/kg, 91.25% with @11 mg/kg and 70.42% in the group treated with closantel. Levamisole was effective against Haemonchus spp., Ostertagia spp., Cooperia spp. Oesophagostomum spp. Closantel was not effective against Cooperia spp. or *Oesophagostomum* spp.

Chartier et al. (2000) conducted an experiment on goat and reported that repeated administration of levamisole hydrochloride at 10 hour intervals compared with a single administration does not alter the anthelmintic efficacy, bioavailability or milk residues. The repeated dosage regime does not afford any increase in the safety margin for levamisole hydrochloride in goats.

Gabriel et al. (2001) performed faecal egg count reduction test to assess tile anthelmintic efficacy of albendazole, levamisole and ivermectin against gastro-intestinal nematodiasis in small ruminants in South Africa. Albendazole resistance was found on 5 of the 6 farms. On each of the 4 farms where ivermectin gave less than 95% reduction in egg counts, there was resistance to albendazole as well. Levamisole showed an efficacy of 95% or higher on all 6 farms.

Michalski (2001) investigated the efficacy of levamisole and oxfendazole in goats with gastro-intestinal nematode infections. Faeces were collected on the 10<sup>th</sup>, 30<sup>th</sup> and 60<sup>th</sup> day after administration of levamisole @7.5 mg/kg bwt. SC and oxfendazole @5 mg/kg per os. Efficacy of levamisole was determined as 44.4%, 52.4% and 43.7%, respectively and of oxfendazole 100%, 62% and 56.2%, respectively.

**Keyvu** et al. (2002) determined anthelmintic efficacy by the faecal egg count reduction test. 10 days after treatment, the sheep treated with levamisole on the first farm had a 98% reduction in faecal egg count. The goats on the same farm had a 97% reduction in faceal egg count. At the second farm 10 days after treatment, sheep treated with levamisole had a 99.4% reduction in faecal egg count, whereas the sheep treated with albendazole only had a 59.4% reduction in faecal egg count.

**Khan et al.** (2003) stated that levamisole and albendazole were 97.22% and effective against gastro-intestinal nematodiasis in cattle. Body weight, total erythrocyte count (TEC) and haemoglobin (Hb) content were increased significantly but percentage of eosinophil was decreased significantly due to levamisole and albendazole treatment. Other hematological parameters (TLC, ESR, PCV, percentage of basophil, neutrophil, monocyte and lymphocyte) were not influenced appreciably.

Lateef et al. (2003) stated that levamisole causes 97-100% reduction in EPG of gastro-intestinal nematodes in sheep.

Shri-Kishan and Gupta (2004) observed efficacy of fenbendazole @7.5 mg/kg, morantel citrate @10 mg/kg and levamisole @7.5 mg/kg bwt. against natural gastrointestinill nematode infections in cattle in Haryana, India. It was shown that faecal egg count decreased significantly on 3 days after treatment with the three drugs and no parasitic eggs were detected on days 7 and 14 after treatment. However, a few eggs were seen on day 21 post-treatment. Haemoglobin and packed cell volume levels improved significantly and absolute eosinophil count significantly after treatment with fenbendazole, levamisole and morantel citrate.

Iqbal et al. (2004) reported that levamisole cause 99.2% reduction in EPG of gastro-intestinal nematodes in sheep.

**Hossain** et al. (2004) reported the efficacy of levamisole in sheep (n=10) with gastro-intestinal naturally infected nematodiasis and paramphistomiasis were treated with Levanid® (tetramisole HC1 BP (vet) -2g and oxyclozanide BP (vet) - 1.4g. The ACME laboratories Dhaka) at the dose rate of 23mg/kg body weight. Levanid was found to be 96.7% effective against gastro-intestinal nematodes and 93.4% effective against Paramphistomurn spp. Total erythrocyte count (TEC), haemoglobin (Hb) content and packed cell volume (PCV) values were increased significantly while erythrocyte sedimentation rate (ESR) significantly decreased following Levànid® treatment in sheep. Body weights of treated sheep were increased by 5.2% after 28 days of treatment. Levanid® may be used as a broad-spectrum anthelmintic against gastrointestinal nematodiasis and paramphistomiasis in sheep.

Iqbal et al. (2006) studied in vivo anthelmintic activity against Trichostrongylid nematodes in sheep. Levamisole (@7.5 mg/kg bwt.) exhibited 99.1 % reduction in EPG.

Hoque et al. (2006) found the comparative efficacy of Piperazine citrate, Levamisole and Pineapple leaves extract as anthelmintics against ascariasis in chicken. The chickens were equally divided into 4 (A, B, C and D) groups. Group A was kept as infected control, while group B, C and D were treated with Piperazine citrate, Levamisole and Pineapple leaves extract, respectively. Although Pineapple leaves extract showed less effectivenss 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) significant (p< 0.01) increased and erythrocyte sedimentation rate (ESR) and total leukocyte country (TLC) significantly (p<0.01) decreased. Body weight was found to be increased following administration of Piperazine citrate, Levamisole and Pineapple leaves extract.

#### **CHAPTER-3**

## MATERIALS AND METHODS

The aim of the present study was to evaluate the incidence of gastrointestinal nematodes of cattle in greater Rajshahi and anthelmintic efficacy of medicinal plants and patent drugs against them during the period from June 2011 to June 2013. Firstly, the prevalence of natural gastro-intestinal nematodes was observed in cattle during the period from June, 2011 to May, 2012 in greater Rajshahi in Bangladesh. Secondly, the study was on the screening of some indigenous medicinal plants for their anthelmintic properties against adult gastro-intestinal nematodes of cattle in vitro in the laboratory of the Department of Animal Husbandry and Veterinary Science, University of Rajshahi. Thirdly, the anthelmentic efficacy of some indigenous medicinal plants and patent drugs against naturally infected gastro-intestinal nematodes and their effects on some clinical, biochemical and hematological parameters in cattle were investigated in the laboratory of the Department of Animal Husbandry and Veterinary Science, University of Rajshahi and Department of Pharmacology, Parasitology and Physiology, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh, during the period from 1<sup>st</sup> June, 2012 to 28<sup>th</sup> June, 2012.

## 3.1 Collection and Examination of Faecal Samples

#### 3.1.1 Selection of animals

The area of Rajshahi, Natore, Chapai Nawabgonj and Naogaon district are called greater Rajshahi. A total 400 cattle were randomly selected from greater Rajshahi. Faecal samples collected from 400 randomly selected cattle of greater Rajshahi were examined to find out the prevalence of different

gastro-intestinal nematodes. Out of 400 faecal samples 200 from Rajshahi, 50 from Chapai Nawabgonj, 80 from Natore and 70 from Naogaon were collected. From the survey data total animal divided into different breeds according to their genetic composition viz. local (n = 85) and crossbreed (n = 315). The cattle were again divided in different genotype such as local (n = 85), L×SL (n = 143), L×F (n = 92) and L×F×SL (n = 80). The selected animal also classified on the basis of sex such as male (n = 237) and female (n = 163). The studied animal again divided according to various age group viz. 1 to <6 months (n = 60), 6 to <12 months (n = 40), 12 to < 24 months (n = 200) and > 24 months (n = 100). The cattle were divided into different season viz. summer (March-May, n = 97), rainy season (June-August, n = 104), autumn (September-November, n = 104) and winter (December-February, n = 95).

#### 3.1.2 Procedure of faeces examinations

Sufficient amount of faecal samples collected from the rectum of cattle by hand were kept in polythene bags and were brought to the laboratory of the Department of Animal Husbandry and Veterinary Science and examined by direct smear and floatation methods (Rahman *et al.*, 1996) and counted by McMaster egg counting technique (Soulsby, 1986) (Plate 1-6).

**Direct Smear Method:** A drop of water is placed on the centre of a clean glass slide. A small amount of faeces is detached from the given sample with the help of a tooth peck and spread out to form a thin smear (thick smears will prevent passing of light through it). This can be done gently drawing the coarse particles towards a side on the glass slide. A suitable cover slip may preferably be put over the smear and the slide is then placed under the low power objective of a microscope for examination.

Floatation Method: Principle of floatation method is to float ova or cysts in a media having higher specific gravity than the ova or cysts. Ova or cysts having specific gravity of 1.05-1.15 are allowed to float in a media having sp. Gr. of 1.120 to 1.630. Sodium chloride solution (sp.gr. 1.120) is used as media. Five to ten grams of faeces are thoroughly emulsified with a floatation fluid in a narrow tall cylinder. More floatation fluid is added to that the upper menisci of the fluid till at the brim of the cylinder. This is then allowed to stand for half an hour; a cover glass is applied to the surface of the fluid, is removed and placed with the wet side down on a clean slide for examination.

## Egg counting by McMaster method

Five (5) gms of faecal samples were taken in a beaker and then 45 ml of saturated salt solution was added and shaken to dilute the sample. A small amount of the diluted sample was withdrawn by a pipette and run into the counting chamber to fill in all the space. The slide was then put to stand for some times allowing the eggs to float under the surface of upper slide of the McMaster chamber. The slide was then examined under microscope using low power objective (10X, 40X) and eyepiece, (6X) and the total eggs within each ruled were counted. The number of eggs per gram of faeces was calculated by using the owing formula:

Number in one gram = 
$$\frac{\text{Number in two chamber}}{0.3} \times \text{dilution factor*}.$$

\*dilution factor = 
$$\frac{\text{Total volume of suspension in ml}}{\text{Total volume of faeces}}$$



Plate 1. Egg of *Haemonchus* sp. (40X)



Plate 2. Egg of Bunostomum sp. (40X)



Plate 3. Egg of *Oesophagostomum* sp. (40X)



Plate 4. Egg of *Trichuris* sp. (40X)



Plate 5. Egg of *Trichostronyglus* sp. (40X)



Plate 6. Egg of Strongyloides sp. (40X)

# 3.2 Collection and Identification of Indigenous Medicinal Plants having Anthelmintic Activity

Collection and identification of 10 indigenous medicinal plants having anthelmintic activity from different location of Bangladesh were performed (Table 1). The indigenous medicinal plants were preserved in the laboratory of the Department of Animal Husbandry and Veterinary Science, Faculty of Agriculture, University of Rajshahi, Bangladesh.

Table 1: Ten indigenous medicinal plants used for studied.

Sl. No.	Indigenous plants			
	Local name	Scientific name	<b>English name</b>	Part used
1.	Neem	Azadirachta indica	Neem	Leaves
2.	Ata	Annona reticulata	Custard apple	leaves
3.	Pepe	Carica papaya	Papaya	Leaves
4.	Rashun	Allium sativum	Garlic	Bulbs
5.	Korolla	Momordica charantia	Bitter Gourd	Fruits
6.	Deshi pat	Corchorus oletorious	Jute	Leaves
7.	Nishinda	Vitex negundo	Chaste Tree	Leaves
8.	Anaras	Ananas comosus	Pineapple	Leaves
9.	Dalim	Punica granatum	Pomegranate	Leaves
10.	Pan	Piper betle	Betel leaf	Leaves

## 3.3 Description of Indigenous Medicinal Plants

**3.3.1 Local name:** Neem

**English name:** Neem tree / Margosa tree

Scientific name: Azadirachta indica

**Habit and Distribution**: A small to large deep-rooted tropical tree with extremely bitter compound leaves, pinkish white small flowers in axillary

panicles and one-seeded drupe type of fruits, grows wild and planted all over the country (Plate 7).



Plate 7. Neem (Neem tree / Margosa tree, Azadirachta indica)

Chemical constituents: Various parts of the plant and the Nim oil contain triterpenoid bitter principles, saponins, flavonoids, tannins and alkaloids. The bitter principles include nimbidin, nimbin, nimbinine, 6-desacetylnimbinine, nimbidol, nimbolide and bakayanin. In addition to these, the leaves contain azadirachtin, salanin, meliantriol, margosopicrin, paraisine, azadinine, nimbinene, nimbolide, quercetin and its glycosides, beta-sitosterol, nhexacosanol, nonacosane, ascorbic acid and amino acids. Barks contain nimbolins A, B, organic acids, tannin, margosin and azadarin. Flowers contain essential oil, kaempferol, kaempferol glucoside, nimbosterin and N-nonacosane. Fruits contain resins, tannins,

triterpenoids, salanin and azadirachtin, melianone, oil and organic acids. Kernel contains triterpenoids, salanin, azadirachtin, oil and fatty acids. Seeds contain six tetranor-triterpenes and four new limonoids, 11-hydroxy-azadirachtin-B, 1-tigloyl-3-cetyl-azadirachtin, 1,2-diacetyl-7-tigloyl-12-hydroxylvilasinin and 23-desmethyl-limocin-B. Nim oil contains margosic acid. The isolation and structure elucidation of the antimalarial agent of the plant, gedunin, has been reported in 1989.

Properties and Uses: Every part of the plant is used medicinally. Various parts of the plant are used in inflammation of gums, gingivitis, sores, fevers (including malaria), spleen complaints, tumours, head scald, smallpox, diarrhoea and cholera. In addition, the leaves possess antiseptic properties and are used in boils, ulcers, eczema, ringworm and scabies. Aqueous and alcoholic extracts of the leaves and bark show good antibacterial activity. Extracts of leaves, bark, gum and seeds are used as remedies for scorpion-sting, snake-bite, as antiviral, antineoplastic and antifungal agents. The gum is a demulcent tonic and is useful in catarrhal affections. Flowers are used in atonic dyspepsia and general debility. Seed kernel produces anti-diabetic and antihyperlipaemic effects in alloxan diabetic rabbits. The oil is used in the treatment of ulcers, chronic skin diseases and rheumatism. Most of the constituents of the plant exhibit antibacterial and anti-inflammatory effects.

#### 3.3.2 Local name: Ata, Nona

**English Names:** Bullock's heart, Bull's heart, Custard apple.

Scientific Name: Annona reticulata Linn.

**Morphology:** A small deciduous or semi-deciduous tree with alternate elliptic leaves all along the lateral branches, greenish yellow flowers on

lateral peduncles, Fruits sub-globose, yellowish-red when ripe, edible with peculiar aroma (Plate 8).



Plate 8. Ata, (Custard apple, Annona reticulata)

**Distribution:** Trees are native to tropical America, particular West Indies. Naturalized and common cultivated in India; occasionally planted as a fruit tree in all parts of the country.

Chemical constituents: Leaves contain polyphenols, dopamine, salsolinol and coclaurine. Roots and root bark contains aporphine alkaloids, anonaine, oxoushinsunine, liriodenine, michelalbine, a benzyltetrahydro-isoquinoline base, reticuline, a phenolic base, and vitamin-C. Seeds contain a highly toxic resinous mixture and tannic acid. Stem, bark contains tannic acid. Fruits contain iodine, iron and starch.

The edible portion contains per 100g protein, 1.4; fat, 0.2 unit; minerals 0.7g; calcium, 5.0; phosphorus, 10.0; iron, 0.6; riboflavin, 0.07; vitamin-C, 5.0; and niacin, 0.6 mg carotene, 67 mg.

**Uses:** Leaves are used for treating inflammatory tumours. Bark is a powerful astringent, and is used as antidysenteric and vermifuge drug. Fruit pulp is sweet and edible, and is used in the treatment of diarrhoea and dysentery. The fruit and bark extracts are widely used as anthelmintics. The seed extract is toxic, insecticidal and acts as a parasiticide against lice.

## 3.3.3 Local name: Papa

English name: Papaya / Papu / Melon tree

Scientific name: Carica papaya

**Habit and Distribution**: A sub-herbaceous branchless tree with a crown of large segmented leaves having long pipe-like petioles, yellow flowers and large oblong fruits, commonly cultivated and planted in gardens throughout the country for its edible fruits, also grows naturally in wastelands (Plate 9).



Plate 9. Papa, (Papaya / Papu / Melon tree, Carica papaya)

Chapter 3

Chemical constituents: Young fruits and leaves are very rich in a

colourless, turning milky white on drying, latex, which contains a mixture

of a number of digestive enzymes, called papain. Latex also contains a

anticoagulant blood factor. immunosuppressive an enzyme,

chymopapain, and a number of medicinal enzymes. Leaves and roots

contain a number of alkaloids, which include nicotine, nicotinine,

myosmine, bis-piperidine, carpaine and dehydro-carpaine I & II. Leaves

also contain a glucoside, carposide and vitamins C and E. Fruit is a rich

source of vitamins, pectins and carotenoids. Seeds contain sulphur-

containing basic substance, carpasemine, carposide and carpaine. Roots

and bark also contain carposide and carpaine.

**Properties and Uses:** Fruits, latex and juice are digestive and are used in

intestinal irritation, habitual constipation and chronic dyspepsia,

diarrhoea (particularly the ripe fruits). Latex and juice of the green fruit

induce abortion. The fruit is also useful in treating bleeding piles and

enlarged spleen and liver. It is also regarded as having hypoglycaemic

properties. The latex has anthelmintic and anti-inflammatory properties

and is applied externally to speed the healing of wounds, ulcers, boils,

warts and cancerous turnours. Seeds are vermifuge, emmenagogue and

anthelmintic. Alcoholic and benzene extracts of seeds produce transient

and reversible antifertility effects and induces functional sterility in rats.

The blood anticoagulant factor of the latex inhibits clotting and the action

of thrombin and fibrinogen.

3.3.4 Local name: Rashun

**English name:** Garlic

**Scientific name:** Allium sativum

**Habit and Distribution:** A bulbuous low herb with white scale leaves and narrow, angular long green leaves and white flowers in terminal umbels, cultivated all over the country as a spice plant (Plate 10).



Plate 10. Rashun, (Garlic, Allium sativum)

Chemical constituents: Chief chemical constituent of garlic is an essential oil, which contains allyl propyl disuiphide, diallyl disulphide, dimethyl disuiphide and polysuiphides. The sulphur compounds include allicin, alum, cycloalliin, ajoene, allisatin I and II and sativins. It also contains anthocyanins, carbohydrates, proteins, amino acids, glycosides of kaempferol and quercetin, saponin-like substances, sterols (beta-sitosterol, cholesterol and campesterol), vitamins and polysaecharides. Four steroidal saponins. protoisoerubosicle, eriiboside-B, isoeruhoside-B. sativioside C, and two amino acids, adensine and tryptophan, have been isolated from the fresh bulbs. Vitamins A, B, C and aipha-tocopherol (vitamin E) have also been isolated from garlic (Chevallier, 19%). Garlic also contains prostaglandins  $A_2$  and  $F_1$ .

Properties and medicinal uses: The bulbs are acrid, bitter, sweet, astringent, salty, thermogenic, aperient, anodyne, oleaginous; aphrodisiac, anthelmintic, expectorant, febrifuge, diuretic, anticholesterol, antibacterial, antifungal and tonic. Garlic counters many infections, including those of the nose, throat and chest. It reduces cholesterol, helps circulatory disorders, such as high blood pressure, and lowers blood sugar levels. Garlic also offers significant protection against heart disease and strokes and reduces levels of serum lipids and is an effective drug for a number of other diseases, which include fever, rheumatism, diabetes, blood pressure and intestinal diseases. It is used as a stimulant, carminative, anthelmintic, hypoglycaemic, diuretic, aphrodisiac, tonic, alterative, hypotensive, antiinflammatory, anti-microbial and blood cholesterol reducing drug. Garlic is an antiseptic and expectorant, and is used in wounds, leprosy, coughs, piles, painful menstruation, abdominal pains in earache. It is given in indigestion, gas formation, headache, asthma and whooping cough. Its juice is regarded as a hair tonic. Garlic also possesses antiinflammatory, immune- enhancing and anticancer properties, it has been used successfully in the treatment of a female candidiasis patient with severe infestation of pathogenic Candida lusitaniae. Aqueous extract of garlic and its constituent allicin have been found to have significant activity against a large number of pathogenic bacteria. It also produces inhibitory effects on gram-positive germs of typhoid-paratyphoid-enteritis group. Allicin is a bactericide, fungicide and germicide and possesses antiturnour and hypoglycaemic properties. Garlic is used as a popular house-hold remedy for a variety of common ailments in addition to its use as a common condiment in cooking.

3.3.5 Local name: Korolla

English name: Bitter gourd

Scientific name: Momordica charantia

**Habit and Distribution:** A creeping or climbing weak annual herb with deeply lobed leaves, yellow epigynous flowers, ovate or oblong-ovate, tuberculed fruits, cultivated extensively as a vegetable crop throughout the country (Plate 11).



Plate 11. Korolla, (Bitter gourd, Momordica charantia)

Chemical constituents: Leaves have been reported to contain two acidic resins, a number of bitter substances, momordicines I, 11 and 111, three new cucurhitane triterpenoids, and gamma-aminobutyric acid. They also contain large quantities of essential amino acids, carotene, thiamine, riboflavin, niacin, ascorbic acid and minerals. Fruits contain a large number of steroidal compounds, saponins, two bitter cucurbitacin glycosides, momordicosides K and L, four non-bitter cucurbitacin

Chapter 3

glycosides, momodicosides F1, F2, G and I, and a mixture of

acylgiucosyl sterols. Fruits also contain the alkaloid momordicinc,

phenolic compounds, proteins, amino acids, vitamins of B group,

ascorbic acid, minerals and a neutral non-nitrogenous principle,

They also contain glucosides of beta-sitosterol and charantin.

stigmasterol. Seeds contain a purgative fixed oil consisting of esters of

stearic, oleic, linoleic and alpha-eleostearic acids, betasitosterol glucoside

and a number of triterpene glycosides. They also contain albumin,

globulin, glutelin, vitamin B, carotene and aipha-aminobutyric acid. The

entire plant has been reported to contain a trace amount of alkaloids,

saponins and orthophthalic acid. An insulin-like peptide has been

reported in this plant.

**Properties and medicinal uses:** Various parts of the plant are popularly

used as a remedy for diabetes mellitus. The unripe fruit is used mainly as

a treatment for late-onset diabetes. Leave and fruits are considered tonic,

stomachic, carminative, anthelmintic, febrifuge and cooling. They are

used in the treatment of rheumatism, gout, diseases of the liver and

spleen, jaundice and fever. Extract of the plant is used in colic, fever,

piles and leprosy, and as a vermifuge. Juice of fresh leaves is emetic,

antibilious and a mild purgative. Root is astringent and useful in

haemorrhoids. Juice of fruit is used in snakebite. Powdered fruit is said to

be useful in healing wounds and malignant ulcers.

3.3.6 Local name: Pat

English name: Jute

**Scientific name:** Corchorus oletorious

**Habit and Distribution:** An erect, slender, tall, soft-wooded, unbranched, annual plant with serrate-margined elliptic leaves and small yellow flowers and rounded angular dehiscent capsules enclosing small flat black seeds, extensively cultivated all over the country as a cash crop for its fibres (Plate 12).



Plate 12. Pat, Tosa, (Jute, Corchorus oletorious)

Chemical constituents: Leaves contain 3 bitter principles, corchoral, capsularol and capsularone, beta-sitosterol and its glucoside and a triterpenoid glucoside. Roots contain triterpines, corosin, oxocorosin, beta-sitosterol, corosilic and ursolic acid. Seeds contain cardiac glycosides, corchorin, capsularin, helveticoside, erysimoside, corchonisides A, 13 and C, and also strophanthidin and a bitter principle corchorin.

**Properties and medicinal uses:** Cardiac glycosides of seeds are very effective in acute and chronic cardiac insufficiency, peroxystic tachycardia and tachyarythrnia in oral dosing. Seeds are also purgative in action. Leaves are demulcent, stomachic, carminative, laxative, tonic, diuretic and useful in acute dysentery. Leaves and fiber are also act as

anthelmintics. Infusion of the leaves is used in atonic dyspepsia, liver disorders, fevers, some cases of chronic cystitis, gonorrhoea, dysuria, hepatic and intestinal colic and gastric catarrh.

#### 3.3.7 Local name: Nishinda

English name: Chaste tree

Scientific name: Vitex negundo

**Habit and Distribution:** A large shrub or small slender tree with 3-5 foliate compound odorous leaves and bluish-purple flowers in pedunculate branched tomentose cymes, grows naturally in all districts (Plate 13).



Plate 13. Nishinda, (Chaste Tree, Vitex negundo)

Chemical constituents: Leaves contain a pale greenish yellow essential oil. An alkaloid, nishindine, and a glucoside. Hentriacontane, sterols, beta-sitosterol, beta-sitosterol acetate, stigmasterol, ascorbic acid, p-hydroxybenzoic acid, carotene, and amino acids have also been isolated from this plant. Stem bark contains flavonoid glycosides of wogonin, aurosin, vitexin, myrecetin, also luteolin, leucodelphinidin, luecocyanidin rhammoside, beta-sitosterol, vanillic acid and p-hydroxybenzoic acid.

**Properties and Uses:** Leaves are antiparasitic, and used as alterative, vermifuge and anodyne. They very effectively reduce inflammatory swellings of joints in rheumatic attacks, relieve catarrh and headache. Juice of fresh leaves removes foetid discharges and worms from ulcers. Flowers are astringent and cooling, oil from flowers is applied to sinuses and scrofulous sores. Fruits are nervine stimulant, emmenagogue and vermifuge. Root is tonic, febrifuge, expectorant and diuretic. It regulates hormones, increases breast-milk production and possesses progesterogenic properties.

#### **3.3.8 Local name:** Anaros

English name: Pineapple

Scientific name: Ananas comosus

**Habit and Distribution:** A tufted stiff herb with a reduced sturdy stem, long sessile lanceolate leaves with spinous-serrate edges and flowers in a terminal cone-like dense, globose or oblong spiral inflorescence and succulent reddish yellow fruits, cultivated throughout the country, particularly in the hilly areas, for its delicious juicy fruit (Plate 14).



Plate 14. Anaros, (Pineapple, (Ananas comosus)

Materials and Methods  $\approx 113$ 

Chapter 3

Chemical constituents: Leaves and unripe fruit contain sterols and triterpenes, ergosterol peroxide, stigmastene-diol, beta-sitosterol, campesterol, stigmasterol and campestanol. Leaves of crown of fruits contain 5-hydroxytryptamine and a steroid triterpene. Stems contain a proteolytic enzyme, bromelin, starch, a trihydroxy terpenoid carboxylic acid, ananasic acid, and glyceryl esters of caffeic and coumaric acids. Fruit also contains polyphenols, phenolic acids, ascorbic and other fruit

acids, vitamins A and C and volatile flavouring constituents.

Properties and Uses: Extract of the leaves is used as a potent anthelmintic, diuretic, styptic and antiseptic. Fresh juice of leaves is an excellent digestive and a strong anthelmintic, and is applied to horny excrescence on the skin, leprosy and elephantiasis. Juice of unripe fruit is regarded as an abortifacient, exhibits significant antifertility activity in mice, antiscorbutic and anthelmintic. Juice of ripe fruit is diuretic, diaphoretic and aperient. Alcoholic extracts of unripe fruit and rhizome show antifertility activity in female albino rats. Bromelain is highly effective in the treatment of patients with pancreatic insufficiency and it has been shown to increase serum levels of a variety of antibiotics in different tissues and body fluids. Bromelain significantly inhibited the growth of tumour cells in cell cultures.

3.3.9 Local name: Dalim

**English name:** Pomegranate

Scientific name: Punica granatum

**Habit and Distribution:** A large shrub or small tree with spiny branches, oblong, opposite leaves, axillary, solitary red showy flowers and globose

many-celled fruits (berries) containing red sweet juice around the angular seeds, planted all over the country as a fruit plant (Plate 15).



Plate 15. Dalim, (Pomegranate, *Punica granatum*)

**Chemical-constituents:** Different chemical substances have been reported from various parts of the plant, particularly the bark, fruit rind and fruit the juice. These include alkaloids pelletierine, isopellelierine, pseudopelletierine and methylisopelletierine, sorbitol, mannitol, glucose, fructose, sucrose, isoquercetin, beta-sitosterol, friedelin, estrone, pectin, triterpenoids, ellagitannins (about 25%), citric acid (about 9%), oxalic acid, calcium, phosphorus, iron, sodium and potassium (Chevallier, 1996 and Said, 1996). Leaves contain betulic and ursolic acids, and betasitosterol. Stern bark contains D-mannitol, friedelin, oxime, oxime acetate, 2,4-dinitrophenyl hydrazone and pelletierine. Flowers contain sitosterol, ursolic, maslinic, asiatic, acetic, ellagic and gallic acids. Fruit pericarp contains tannin, ellagic, citric and ursolic acids. The presence of a hemorrhoidal compound has been reported from the pericarp. Fruit

Materials and Methods  $\approx 115$ 

Chapter 3

juice contains sucrose, pectin, carotenoids, anthocyanins, ascorbic acid,

vitamin C, thiamine and riboflavine. Root bark contains four alkaloids,

pseudopelletierine, pelletierine, isopelletierine and methyl pelletierine,

and three other basic components (Chopra et al., 1969). The plant has

also been reported to contain a hypoglycaemic agent, penta-O-galloyl-

beta-glucose along with ellagic acid and glucosides.

**Properties and medicinal uses:** All parts of the plant have astringent,

refrigerant, stomachic, anti-diarrhoeal and anthelmintic activities (Said,

1996). Root bark, stem bark and rind are astringent and anthelmintic,

specific for tapeworm due to their iso-pelletierine content (Chevallier,

1996). Rind of fruit is useful in the treatment of diarrhoea and dysentery.

Pulp of fruit is also astringent and stomachic. Extract of the fruit

inactivates several intestinal virus, herpes simplex virus and polio virus.

Fruit juice is nutritious and when drunk it produces cooling effect. Seeds

possess stomachic properties. Methanolic extract of fruit rind exhibits

significant antibacterial activity (Ghani, 2003).

3.3.10 Local name: Pan

**English name:** Betel leaf

Scientific name: Piper betle

Habit and Distribution: A stout twining climber with broadly ovate-

oblong or ovate-cordate leaves, tiny yellow-green flowers and small

spherical fruits, extensively cultivated as a cash crop throughout the

country (Plate 16).



Plate 16. Pan, (Betle Leaf, Piper betle)

Chemical constituents: Leaves contain up to 1% of an essential oil with burning taste, composed of cadinene, chavicol, chavibetol and cineole. They also contain an alkaloid, arakene, tannins, starch, sugars and diastases. In addition, they contain beta-carotene and alpha-tocopherol. Roots contain diosgenin.

Properties and Uses: Leaves are popularly used as carminative, astringent, stimulant and antiseptic drugs. They are also used in headache and coughs of children. Leaf-stalk is used as a suppository for rectal evacuation in children. Leaf, mixed with honey, is a remedy for coughs. Juice of leaf is used as an eye drop in painful affections and night blindness, and also to relieve cerebral congestion. Root induces permanent sterility in women. Extract of leaves exerts anti-tumour activity in carcinogenesis, and suppresses mutagenic and carcinogenic actions of tobacco-specific nitrosamines.

## 3.4 Preparation of plants powder and extract

Plant power and extracts used in this study were prepared by following methods:

## 3.4.1 Processing of plant materials

After collection and bringing them to the laboratory, all fresh indigenous medicinal plants were washed in running tap water and cut into small pieces. Firstly the plants material were dried in shade and then dried in the hot air oven at 55-60°C to gain constant weight (Plate 17 & 18).





Plate 17. Hot-air Oven

Plate 18. Small pieces of leaves in hot-air oven

## 3.4.2 Preparation of plants powder

Powders were prepared by pulverizing the dried indigenous medicinal plants with the help of electric grinder. A 25-mesh diameter sieve was used to obtain fine dust and preserved them into airtight plastic container, till their use for extract preparation (Plate 19-29).



Plate 19. Electric Grinder



Plate 20. Powder of Neem (Leaves)



Plate 21. Powder of Ata (Leaves)



Plate 22. Powder of Papaya (Leaves)



Plate 23. Powder of Garlic



Plate 24. Powder of Korolla (Fruit)



Plate 25. Powder of Jute (Leaves)



Plate 26. Powder of Nishyinda (Leaves)



**Plate 27. Powder of Pineapple (Leaves)** 



Plate 28. Powder of Dalim (Leaves)



Plate 29. Powder of Betel (Leaves)

## 3.4.3 Preparation of plants extract

Previously prepared plants powders were used for preparation of plants extract. Ten grams of each powder were taken in a 500 ml beaker and separately mixed with 100 ml of ethanol and 100 ml of water. Then the mixtures were stirred for 30 min by a magnetic stirrer (6000 rpm) and left as such for next 24 hrs. The extracts were then filtered through a fine cloth and again through filter paper (Whatman No. 1). The filtered material was taken into round bottom flask and then concentrated by evaporation of ethanol and water from filtrate in a water bath at 50°C till it reached the final volume of 10 ml. After the evaporation of ethanol and water from filtrate, the condensed extracts were preserved in tightly corked-labelled bottle and stored in a refrigerator until used for screening of anthelmintic activity (Plate 30-42).



Plate 30. Measurement of plant powder with balance and weight box

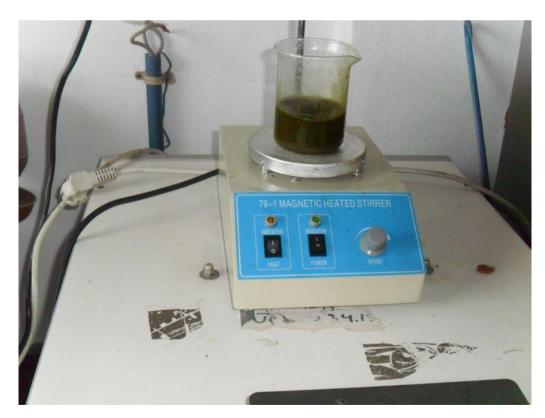


Plate 31. Electric Stirrer



Plate 32. Filtration of plant extract



Plate 33. Neem leaves extract



Plate 34. Ata leaves extract



Plate 35. Papaya leaves extract



Plate 36. Garlic extract



Plate 37. Korolla leaves extract



Plate 38. Jute leaves extract



Plate 39. Nishyinda leaves extract



Plate 40. Pineapple leaves extract



Plate 41. Dalim leaves extract



Plate 42. Betel leaves extract

## 3.4.4 Preparation of stock solution

Stock solutions of plant extracts were prepared by diluting the condensed extracts with water. Different concentrations of each category of plant extracts were prepared by dissolving them in the water prior to anthelmintic screening.

# **3.5** Collection and Maintenance of Adult Gastro-intestinal Nematodes in the Laboratory

Collections of parasites from abomasum were made by following standard procedure described by Taylor (1934), Bell (1957) and Rahman (2002). Viscera of cattle were collected from Saheb Bazar Slaughter House of City Corporation in Rajshahi. The abomasums were opened through its lesser curvature with the help of scissors. The contents were emptied in glass jars containing normal saline. The abomasi were thoroughly washed and cleaned off ingesta and put in a different jar containing normal saline and left for an hour or two to release the attachments of parasites from the wall of the abomasum. The mucosal surfaces of the abomasum were rubbed carefully between the fingers to remove any remaining worms adhering to the abomasal wall. Finally abomasl mucosa was examined with the help of a magnifying glass for any remaining parasites still adhering to the mucous lining of abomasum. The contents were washed several times with water and continued till the worms were free from debrises. The final wash was, made, with the normal saline. The sediments were examined in large petri-dishes over a black background. The parasites were collected with the help of curved needle and kept in normal saline. The parasites were also cleared off debris by brushing with camel hairbrush or shaking in normal saline. Then parasites were identified according to the keys given by Rahman et

al., (1996), Yamaguti (1958) and Yorke and Melpestone (1962). They were kept in petri-dishes containing normal saline (Plate 43). The petri-dishes containing parasites were kept in incubator at 38°C until required for experimental use.

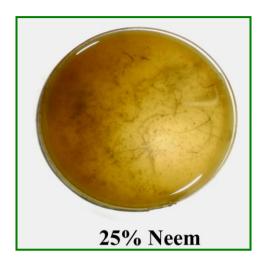


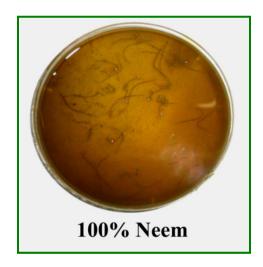
Plate 43. Petri-dishes containing parasites

#### 3.6 In vitro Screening of Plant Extracts for Anthelmintic Activity

In vitro screenings were performed to determine the anthelmintic activity of some indigenous medicinal plant extracts in the laboratory of the Department of Animal Husbandry & Veterinary Science, University of Rajshahi. Screening of water and ethanol extracts of plant at various concentration levels viz. 25 mg/ml, 50 mg/ml and 100 mg/ml were performed in the petri-dishes containing adult live stomach worms of cattle collected at slaughter house of Rajshahi City Corporation in PBS. 100 ml of PBS containing 10 adult worms (both male and female) were pipetted in 6 petri-dishes. 0.1 ml, 0.05 ml and 0.025 ml ethanol and water extracts were then added, respectively. The drug-parasite petri-dishes were incubated three hours at room temperature and the efficacy was counting the dead parasites and expressed in percentages (%) (Plate 44-53).







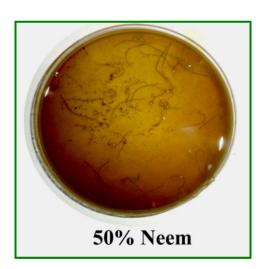
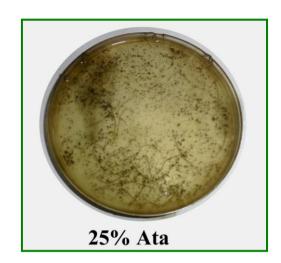
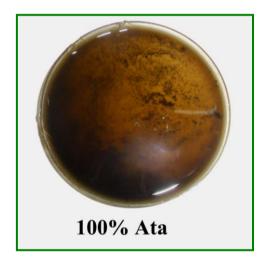


Plate 44. In-vitro trial of Neem extract







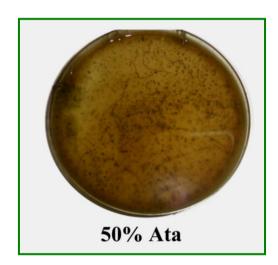


Plate 45. In-vitro trial of Ata extract









Plate 46. In-vitro trial of Papaya extract









Plate 47. In-vitro trial of Garlic extract

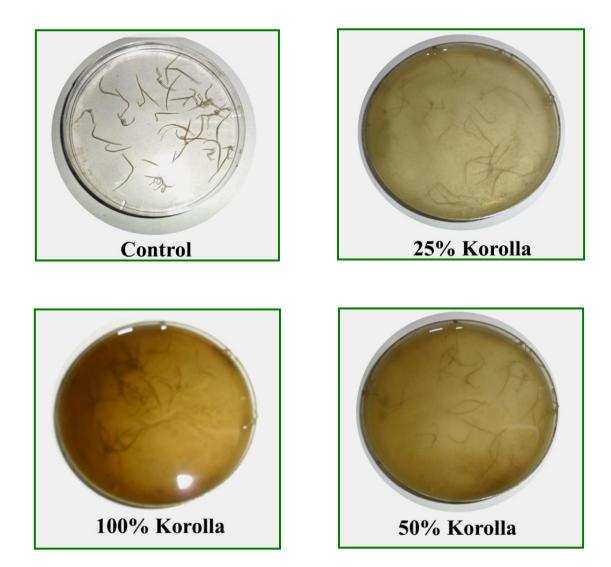


Plate 48. In-vitro trial of Korolla extract

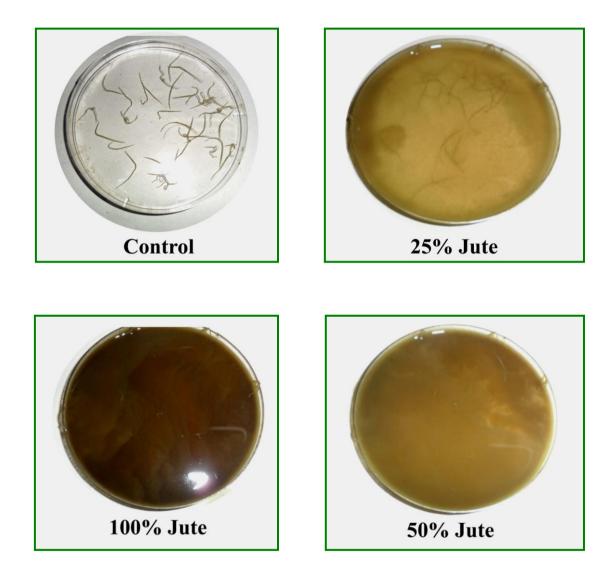


Plate 49. *In-vitro* trial of Jute extract







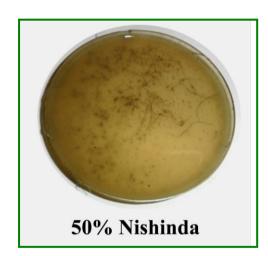


Plate 50. In-vitro trial of Nishinda extract







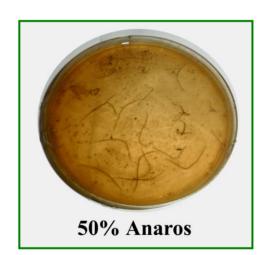


Plate 51. In-vitro trial of Anaros extract









Plate 52. In-vitro trial of Dalim extract







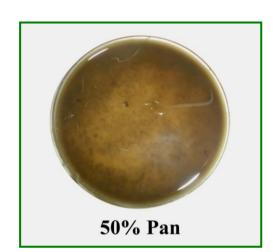


Plate 53. In-vitro trial of Pan extract

#### 3.7 Selection of Research Site for *in-vivo* Trials

This experiment was conducted at village of Binodpur, Rajshahi. The research was carried out during the period from 1<sup>st</sup> June, 2012 to 28<sup>th</sup> June, 2012.

#### 3.8 Selection of Cattle for in-vivo Trials

For this study, fifty cattle were selected which were suspected to be suffering from natural gastro-intestinal nematodes infection and they were marked at the ears by the numbered tag. All these cattle were maintained at the same altitude and under nearly identical conditions. They were kept in door at night and part of the day. All the cattle were fed with balanced rations which were composed of roughages and concentrates. Examination of faecal samples for gastro-intestinal nematodes egg counts by floatation method were carried out over a week prior to commencement of treatment. On the basis of faecal sample examination results, 25 cattle of both sexes were selected for this study and these cattle were confirmed cases infected with gastro-intestinal nematodes. The ages of cattle ranged from 1 to 2 years approximately (Plate 54).



Plate 54. Experimental Cattle

## 3.9 Collection of two Indigenous Medicinal Plant and Patent Drugs for *in vivo* Experiment

## 3.9.1 The selected medicinal plants

Among the 10 indigenous medicinal plants, two plants were selected for the experimental trials on the basis or highest efficacy on *in vivo* study (Plate 55-58). These two indigenous medicinal plants were as follows:

Sl. No.	Indigenous plants						
	Local name	Scientific name	English name	Part used			
1.	Neem	Azadirachta indica	Neem	Leaves			
2.	Korolla	Momordica charantia	Bitter Gourd	Fruits			



Plate 55. Powder of Neem



Plate 56. Powder of Korolla

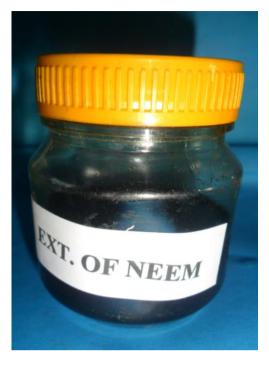


Plate 57. Extract of Neem



Plate 58. Extract of Korolla

## 3.9.2 The patent drugs

Two modern anthelmintics selected for the experiment were purchased from local market, Rajshahi. Levamisole (Levavet<sup>®</sup> 600 mg, Tablet preparation, The ACME Lab. Ltd.) and Ivermectin (Ivertin<sup>®</sup>, pour on formulation, The Chemist Lab. Ltd.) were selected for the experiment (Plate 59-60).



Plate 59. Levamisole (Levavet)®



Plate 60. Ivermectin (Ivertin)®

#### 3.9.2.1 Ivermectin

The Avermectins are a group of chemically related macroyclic lactones produced by fermentation of the actinomycete *Streptomyces avermitilis*. Avermectin is a complex of eight such fermentation products, each having nematocidal activity but, in contrast to the macrolide or polyene antibiotics, lacking significant antibacterial or antifungal activity. Ivermectin (Cevamec 1%®, Ivomec®, Oramec®, Heartgard® 30, Cardomec®, Eqvalan® Zimectrin® Mectizan®) is a semi-synthetic derivative of avermectin that has a broad spectrum of activity against a wide variety of arthropods and nematodes of wild and domestic animals and human

## **Chemistry**

Avermectin is a mixture of four major components (avermectin  $A_{1a}$ ,  $A_{2a}$ ,  $B_{1a}$ , and  $B_{2a}$ ) and four minor components recovered in smaller amounts (avermectin A1a, A2a, B La, and B2a). Of these, the B1a, components is recovered in greatest amount along with its  $B_{1b}$  minor homologue, ivermectin, derived from this mixture of  $B_1$  avermectins by saturation of the double bond between C-22 and C-23, consists of not less than 80 percent 22, 23- dihydroavermection B1a and not more than 20 percent 22,23- dihydroavermection  $B_{1b}$ .

Ivermectin (as its major component, 22, 23-dihyroavermectin  $B_{1a}$ ,) is an off- white powder that is highly lipophilic and hydrophobic. It dissolves in most organic solvents but is poorly soluble in water. It is stable at room temperature in non-acidic solutions but is degraded by UV light.

Chemical Structure of Ivermectin

#### Mode of action

It was originally thought that the macrolide endectocides increased the release of γ-aminobutyric acid (GABA) from synaptosomes of the nervous system. This, in turn, opened GABA-gated chloride channels. It is now known that these compounds open chloride channels in invertebrates via a specific binding site that is glutamate-gated, although the binding site apparently occurs in close anatomic proximity to GABA-gated sites, and the macrolide endectocides my potentiate GABA-gated sites as well. About 50 percent of the effect of a macrolide endectocide can be reversed with picrotoxin, a GABA antagonist active at the chloride channel. In nematodes, the synapse between inter-neurons and excitatory motor neurons is the primary site of action, whereas the myoneural junction is the primary site in arthropods. In either case the chloride ion influx lowers cell membrane resistance and causes a slight hyper polarization of the resting potential of postsynapitc of stimuli to muscles

is prevented resulting in a flaccid paralysis of affected parasites followed by their death or expulsion.

#### **Pharmacokinetics**

Fink and Porras (1989) summarize pharmacokinetics studies with ivermectin. The specific formulation used, the route to administration, and the animal species to which it is administered affect the pharmacokinetics of ivermectin. The biological half-life ( $t_{1/2}$ ) of ivermectin in plasma following IV administration of 300 µg/kg to cattle is 2.8 days. IV administration to sheep gives a similar biological half-life ( $t_{1/2}$  =2.7days) to that in cattle but a lower plasma concentration due to a greater volume of distribution in sheep than in plasma concentration due to a greater volume of distribution in sheep than in cattle (1.9 vs. 4.61/kg). Ivermectin is eliminated more rapidly in dogs ( $t_{1/2}$  =1.6-1.8 days).

Subcutaneous administration of the commercial formulation of ivermectin to cattle at dose rate of 200  $\mu$ g/kg results in a longer biological half-life ( $t_{1/2}$  =8 days) than IV administration due to slow absorption from the injection site. A peak plasma concentration (Cp) of 44 mg/ml occurs at 2 days after S/C injection. Clinically significant anthelmintic efficacy persists for approximately 2 weeks after S/C injection depending upon persists for oral dosing in sheep results in a  $t_{1/2}$  of 3-5 days.

Following administration, ivermectin residues are lowest in brain and highest in liver, bile, and fat (Chiu and Lu 1989). Depletion half-life wete 4.8 and 7.6 days for liver and fat, respectively, in cattle. Tissue redistribution patterns axe similar for similar for sheep, swine, and rats, but depletion half-lives for liver and fat are shorter in sheep and rats than

in cattle or swine. The parent drug is the major liver residue for 3, 5, 7 and 14 days dosing in rats, sheep swine, and cattle, respectively.

Fecal excretion is the main rout of elimination, accounting for 9.8% or more of excreted ivermectin, with the remainder appearing in the urine, except up to 5% of the dose may be excreted in the milk of lacerating animals.

#### 3.9.2.2 Levamisole

Levamisole, INN is a highly acceptable anti-nematodal drug because of its broad range of activity in a large number of hosts (sheep, cattle, pig horse, chicken, dog). Major advantages of levamisole are that it is effective against nematodes of the lungs and GI tract and can be administered to the skin as a pour on or by subcutaneous injection. It may also be administered in feed or by oral suspension bolus or gel. It has no activity against fluke's tapeworms or protozoa.

## Chemistry and relation to dl- tetrarnisole:

Levamisole is a I-isomer of dl- tetramisole. The later drug was introduced as an anthelmintic in 1966 and is a racemic mixture of two optical isomers: S (-) tetramisole (=I-tetramisole = levamisole) rotates flame polarized light to the left; R (+) tetramisole (=d-tetramisole) rotates light the right. It was found that the anthelmintic activity of the mixture rested almost solely with 1- isomer (levamisole).

$$N \sim N$$

Chamical structure of Levamisole

The chemical name of levamisole is (-) -2, 3, 5, 6 -tetraydro-6- phenylimidazo [2, I-b] thiazole. The marketed form is either its hydrochloride (bolus drench or paste) of its phosphate (Injectable form). Levamisole hydrochloride a white crystalline compound is highly soluble in water.

## Mode of action of levamisole hydrochloride:

Levamisole hydrochloride has a paralyzing action on nematodes. The paralysis is due to sustained muscle contraction. Levamisole acts as a ganglion stimulant (cholinomimetic). This conclusion is supported by the fact that levamisole induced contactions of *Ascaris suum* are blocked by the autonomic ganglion blocking agents mecamylamine and pempidine (Adams, 2001).

#### **Pharmacokinetics**

Absorption and excretion of levamisole is rapid following oral administration of the drug. Approximately 40% is excreted in urine in 12 hours. There after urinary excretion decreases and only another 8% are eliminated over the next 8 days. Elimination in feces over an 8-days period accounts for approximately 41% of the dose the bulk of which passes in 12-24 hours. A small amount is expelled in respired gases (Adams, 1995).

## 3.10 Experimental Design for in vivo Trials

On the basis of faeces examination, 25 (Twenty five) naturally parasitized cattle of Binodpur, Rajshahi were selected for the study. *In vitro* anthelmintic efficacy of 10 medicinal plants, among them best two were selected for *in vivo* experiment. The efficacy of two indigenous medicinal plants and two patent-drugs were determined against natural gastro-

intestinal nematodes. Cattle infested with gastro-intestinal nematodes were randomly divided into five (5) groups (A, B, C, D, and E), each group consist of five (5) cattle. Each ethanol extract of leaves of neem and fruits of korolla were administered (@ 100 mg/kg bwt.) orally to the cattle of group B and C respectively. Cattle of group D and E were treated externally with Ivermectin (pour on @200 µg/kg body weight) and orally with Levamisole (@7.5 mg/kg bwt.) respectively. Cattle of group A were kept as infected control without giving any treatment. Before trials, (pre-treatment/day 0) with these indigenous medicinal plants and patent drugs, initial body weight, total egg count of gastrointestinal nematodes, Physical and haematological parameters were examined and recorded. During the experimental period with treatment the faecal samples were examined on 7th, 14th 21th and 28th day. Clinical (body weight), biochemical (ALT, AST) and hacmatological (TEC, Hb, PCV, TLC DLC, MCV MCHC, MCH) parameters were also examined on day 7, 14, 21 and 28 for the determination of effects of neem, korolla, ivermectin and levamisole.

	Number		Time of days				
Group	of cattle	Treatment with	Pre-	Post-freatment			
	in each	dose	treatment				
	group		Day 0	Day 3	Day 10	Day 17	Day 28
A	5	Control	*	*	*	*	*
		(untreated)	♦ ♀	♦ ♀	<b>♦</b> ♀	<b>♦</b> ♀	<b>♦</b> ♀
			Ψ	Ψ	Ψ	Ψ	Ψ
В	5	leaves of neem	*	*	*	*	*
		(100 mg/kg	♦ ♀	<b>♦</b> ♀	<b>♦</b> ♀	<b>♦</b> ♀	<b>♦</b> ♀
		bwt.)	Ψ	Ψ	Ψ	Ψ	Ψ
С	5	fruits and seeds	*	*	*	*	*
		of bitter gird	♦ ♀	♦ ♀	<b>♦</b> ♀	<b>♦</b> ♀	<b>♦</b> ♀
		(100mg/kg	Ψ	Ψ	Ψ	Ψ	Ψ
		bwt.)					
D	5	Ivermectin	*	*	*	*	*
		(Ivertin® pour	<b>♦</b> ♀	<b>♦</b> ♀	♦ ♀	<b>♦</b> ♀	♦ ♀
		on 200 µg/kg	Ψ	Ψ	Ψ	Ψ	Ψ
		body weight)					
Е	5	Levamisole	*	*	*	*	*
		(Levavet ®)	<b>♦</b> ♀	<b>♦</b> ♀	♦ ♀	<b>♦</b> ♀	<b>♦</b> ♀
		(7.5mg/kg body	Ψ	Ψ	Ψ	Ψ	Ψ
		weight)					

Table 2: Protocol of the study for in vivo trial

#### 3.11 The Test Parasites

Gastro-intestinal nematodes were used as test parasites in this study. The most important gastro-intestinal nematodes of cattle in Bangladesh were strongyles (*Haemonchus* sp., *Trichostrongylus* sp., *Oesophagostomum* sp. and *Mecistocirrus* sp.), *Bunostomum* sp., *Strongyloide* sp., *Trichuris*, *Ascaris* sp. and *Capillaria* sp. etc.

## 3.12 Faecal Sample Examination

Sufficient amount of faecal samples were collected from the rectum of cattle by hand on day 0, 7, 14, 21 and 28 and were kept in polythene bags and were brought to the laboratory and were determined the eggs per

<sup>\*</sup> Faecal sample examination

<sup>♦</sup> Haematological tests (TEC, Hb, PCV, TLC, DLC, MCV, MCHC and MCH)

<sup>♀</sup> Biochemical tests (ALT and AST)

Ψ Clinical parameter (body weight)

gram (EPG) by faecal samples examination by using direct smear and floatation methods and counted by McMaster egg counting technique (Soulsby, 1986).

## Egg counting by McMaster method

## Materials required

- a) Compound microscope
- b) Saturated salt solution
- c) Pipette
- d) McMaster egg counting chamber
- e) Dropper
- f) Beaker
- g) Pestle and mortar
- h) Absorbent
- i) Weight balance

#### **Procedure**

Five (5) gms of faecal samples were taken in a beaker and then 45 ml of saturated salt solution was added and shaken to dilute the sample. A small amount of the diluted sample was withdrawn by a pipette and run into the counting chamber to fill in all the space. The slide was then put to stand for some times allowing the eggs to float under the surface of upper slide of the McMaster chamber. The slide was then examined under microscope using low power objective (10X, 40X) and eyepiece, (6X) and the total eggs within each ruled were counted. The number of eggs per gram of faeces was calculated by using the owing formula:

Number in one gram = 
$$\frac{\text{Number in two chamber}}{0.3} \times \text{dilution factor}^*$$
.

\*dilution factor =  $\frac{\text{Total volume of suspension in ml}}{\text{Total volume of faeces}}$ 

EPG of facces was counted on day 0 (pre-treatment) and on day 7, 14, 21 and 28 of post treatment. Faecal samples were counted from each animal of both treatment and control groups (Plate 61-63).



Plate 61. Fecal sample

Plate 62. Examination of fecal sample by microscope



Plate 63. Treated with Ivertin® (Pour-on) in experimental cattle

## 3.13 Determination of Efficacy of Different Treatment

Efficacies of different treatment were determined by faecal egg count reduction test as per formula mentioned below:

## $Efficacy = \frac{EPG \text{ prior to treatment-EPG post-treatment}}{EPG \text{ prior to treatment}} \times 100$

#### 3.14 Determination of Haematological Parameters

Collection of blood samples for the haematological examination, blood was collected with sterile syringe and needle from the jugular vein of cattle. Approximately 5 ml of blood was collected from jugular vein of each animal and was transferred immediately to a clean, dried glass vial containing anticoagulant (sodium citrate) at day 0 (pre-treatment) and 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day of post-treatment period and stored in ice-box for examination. The haematological studies were carried out at Physiology laboratory, BAU (Plate 64-67). The routine analysis of blood was carried out by the standard method as described by Coffin (1953) for following parameters:

- i. Total erythrocyte count (TEC)
- ii. Haemoglobin (Hb) content
- iii. Packed cell volume (PCV)
- iv. Total leukocyte count (TLC)
- v. Differential leukocyte count (DLC)
- vi. Mean corpuscular volume (MCV)
- vii. Mean corpuscular haemoglobin concentration (MCHC)
- viii. Mean corpuscular Haemoglobin (MCH).

#### 3.14.1 Total erythrocyte count (TEC)

#### **Materials required:**

- a) Microscope
- b) Haemocytometer
  - i) Counting chamber and cover glass
  - ii) White blood cell pipette with rubber tube

- iii) Red blood cell pipette with rubber tube
- c) Red blood cell diluting fluid (Hayem's solution)
- d) Filter paper
- e) Absorbent cotton
- f) Blood sample

**Procedure:** The TEC was determined as per method described by Coffin (1953). The tip of the red blood cell pipette was placed on the blood sample and was sucked up until reached 0.5 mark Then the tip of the pipette was placed in the diluting fluid (Hayem's solution) and filled the pipette to 101 mark. The content of the pipette was shaken with 8 knot for 1-2 min. After discarding 2-3 drops, a small drop from the pipette was placed to the counting chamber. The chamber was allowed to stand for a minute to allow the erythrocyte to settle. Then the cells were started to count with the high power objective (45X). The central squares of the counting chamber were used for erythrocyte count. Red blood cells were counted in the four corner squares and one centre square of the chamber. The number of RBC was calculated as follows:

Number of RBC = No. of cell count  $\times$  10000 and expressed the results as million/cu. mm.

### 3.14.2 Haemoglobin (Hb) content

#### **Materials required:**

- a) Hellige Haemometer
  - i) Comparator
  - ii) Diluting tube (special graduated)
  - iii) Sahli's pipette with rubber tube
  - iv) Stirrer

#### v) Dropper

- b) N/10 HCl solution
- c) Absorbent cotton
- d) Blood sample

Procedure: The Hb content was determined as per method described by Coffin (1953). N/10 HCI solution was taken in special graduated tube up to its 2 mark. The special Sahli pipette was filled with blood up to 20 marks and wiped its side with absorbent cotton. Immediately the blood of the pipette was transferred into the diluting tube containing N/10 HCI solute and rinsed the pipette 2-3 times by sucking water into the pipette and added these wasting to the solution in the tube. The tube was shacked until the blood was well mixed with N/10 HCI solution and H<sub>2</sub>O and the, mixture appeared uniformly dark brown in color. Using the dropper, water was added drop by drop each time mixing the solution with a stirrer until color of the solution matched with the standard. After 5 min of first noting time the result was read in daylight from the scale of measuring tube by observing the graduated mark at the lower edge of the meniscus at the top of the liquid column. The result was expressed in g%.

## 3.14.3 Packed cell volume (PCV)

#### **Materials required:**

- a) Wintrobe haematocrit tube
- b) Centrifuge machine
- c) A pipette
- d) Special loading tube
- e) Absorbent cotton
- f) Blood sample

**Procedure:** The PCV was determined as per method described by Coffin (1953). The blood was drawn into the special loading pipette. The tip of the pipette was inserted to the bottom of a clean, dry Wintrobe haematocrit tube. The pipette was slowly withdrawn but pressure was continued on the rubber bulb of the pipette so as to exclude air bubbles. The tip was filled exactly to the 10 mark. Then the tube was centrifuged at 3000 rpm for half an hour and the reading was taken. The result was expressed in percentage (%).

### 3.14.4 Total leukocyte count (TLC)

## **Materials required:**

- a) Microscope
- b) Haemocytometer
  - i) Counting chamber and cover glass
  - ii) White blood cell pipette with rubber tube
  - iii) Red blood cell pipette with rubber tube
- c) White blood cell diluting fluid (N/10 HCl acid)
- d) Filter paper
- e) Absorbent cotton
- f) Blood sample

**Procedure:** The principles involved in enumeration of total leukocyte count (TLC) were almost similar to those of erythrocytes. Here the leukocyte diluting fluid was N/10 HCI acid. Well mixed blood was drawn up to the 0.5 mark of white blood cell pipette. The diluting fluid was filled up to the 11 marks of the pipette and the contents were thoroughly mixed for 2 min. 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 objectives (10X). The leukocytes in the 4 large squares (each 1 square mm.) of the counting chamber were counted. The counting and calculation of leucocytes were performed as per methods described by Coffin (1953). The number of WBC was calculated as follows:

Number of WBC=No. of cell counted  $\times$  50 and expressed the results as thousand/cu.mm.

#### 3.14.5 Differential leukocyte count (DLC)

#### **Materials required:**

- i) Microscope
- ii) Modified Giemsa's stain
- iii) Absorbent cotton
- iv) Blood sample

**Procedure:** A thin smear of blood in glass slides was made and dried in the air. The smeared slide was then placed on a staring rack and modified Giemsa's stain was placed on the entire slide and allowed to reach for 3 mm. Then an equal amount of distilled water placed on the slide. The mixture was then allowed to remain on the slide for about 5 mm and air was blown over the slide, after 5 mm the slide was washed in a water container for few seconds. Then the slide was allowed to stand for drying. When the slide became dried, the prepared slide was adjusted under low power objective (l0x) of microscope to select the uniform or evenly distributed area. Then a small drop of immersion oil was placed on the stained surface on the slide and was examined under high power objective (100X) of microscope. The leukocytes were then counted in parallel strip method. The leukocytes were clearly identified and counted up to 100 cells. The result was read as per method described by Coffin

(1953). The different types of leukocyte were counted and expressed in percentage (%).

#### 3.14.6 Mean corpuscular volume (MCV)

The MCV is an average or mean volume of a single red blood cell expressed in cubic micron. It is calculated from the following two basic values:

- i) Red blood cell (RBC) count in million/mm<sup>3</sup> and
- ii) Packed cell volume (PCV) in 100 ml blood

$$MCV = \frac{PCV \times 10}{RBC \text{ count in million/mm}^3} \text{ (Coffin, 1953)}$$

#### 3.14.7 Mean corpuscular haemoglobin concentration (MCHC)

The MCHC represents the relationship between the red blood cell volume and its degree or percentage saturation with haemoglobin. The MCHC was calculated from the following formula:

$$MCHC = \frac{Hb}{PCV} \times 100 \text{ (Coffin, 1953)}$$

#### 3.14.8 Mean corpuscular haemoglobin (MCH)

The MCH, which is also determined indirectly, is the average haemoglobin content (weight of Hb) in a single red blood cell expressed in picagram/micro micro gram. It was calculated from the following basic values:

- i) RBC count in million/mm<sup>3</sup>, and
- ii) Hb in g %

$$MCH = \frac{Hb \times 10}{RBC \text{ count in million/mm}^3} \text{ (Coffin, 1953)}$$

#### 3.15 Determination of Biochemical Parameters

The biochemical parameters of serum [alanine aminotransferase (ALT) and aspartate aminotransferase (AST)] were determined by auto-analizer Reflotron® Plus (Boehringer Mannheim) according to the method described by Deneke and Rittersdorf (1984 and 1985).

#### **Preparation of serum**

Five (5) ml of blood was collected in the sterile glass test tube. The blood containing tubes were placed in standing position at room temperature for 6 hours. The tubes were then incubated overnight in the refrigerator (4°C). The serum samples were separated and centrifuged to get rid of unwanted blood cells where necessary. Analyses were done in a quickest possible time.

### 3.15.1 Determination of alanine aminotransferase (ALT) level Materials required:

- i) Reflotron® Plus
- ii) ALT test strip
- iii) Serum

**Procedure:** Serum was drawn with the help of capillary pipette (avoiding bubbles) up to red mark (30  $\mu$ l) and was placed as a drop to the centre of the red application zone (xx) of the ALT test strip taking care not to touch the application zone with the pipette tip. After opening the sliding cover the test strip was placed on to the guide within 15 seconds and the slide was forwarded until it locks into place. The sliding cover was closed. The ALT level was displayed on the monitor.

**Calculation:** The enzyme activity was shown for 37°C in U/l.

### **3.15.2 Determination of aspartate aminotransferase (AST) level Materials required:**

- i) Reflotron Plus
- ii) AST test strip
- iii) Serum

**Procedure:** Serum was drawn with the help of capillary pipette (avoiding bubbles) up to red mark (30  $\mu$ l) and was placed as a drop to the centre of the red application zone (xx) of the AST test strip taking care not to touch the application zone with the pipette tip. After opening the sliding cover the test strip was placed on to the guide within 15 seconds and the slide was forwarded until it locks into place. The sliding cover was closed. The AST level was displayed on the monitor.

**Calculation:** The enzyme activity was shown for 37°C in U/I.

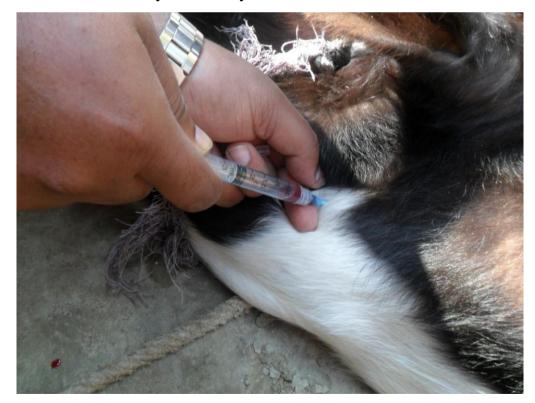


Plate 64. Collection of blood from the experimental cattle



Plate 65. Collected blood sample

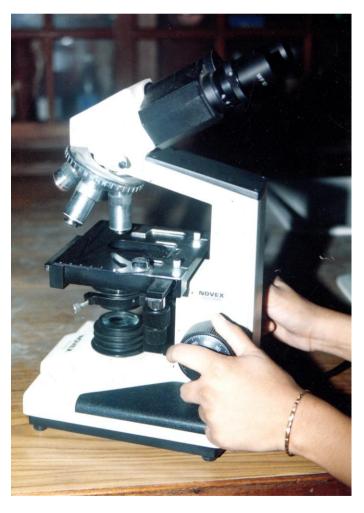


Plate 66. Examination of blood sample by a compound light microscope at laboratory of  ${\bf BAU}$ 



Plate 67. Examination of biochemical parameter by Reflotron Plus Machine

#### 3.16 Measurement of Body Weight

The weight of each cattle was taken and recorded. The result was expressed in Kg (Plate 68). The body weight of all experimental cattle was taken as per method cited by Samad (2001).

Body weight = 
$$\frac{\text{Length} \times (\text{Girth})^2}{300 \times 2.2} \text{ Kg}$$

Here,

Length = Length from the point of elbow to pin bone in inch Girth = Length of heart girth in inch.



Plate 68. Measurement of body weight

#### 3.17 Statistical Analysis

Data were statistically analyzed in accordance with the objectives of the study. Various statistical tools such as, percentages, incidence & chi-square test, mean and standard error had been adopted in this study. Mean of different data were then tested by Duncun Multiple Range Test (DMRT) by using SPSS programme computer package. Percentages of cattle with gastro-intestinal parasite were calculated to determine the incidence of gastro-intestinal nematodes for age, sex, season, breed, genotype, districts and time.

Incidence of parasite = 
$$\frac{\text{N of infected animal}}{\text{Total N of animal}} \times 100$$

The prevalence of gastro-intestinal nematodes was statistically analyzed by chi-square test in relation with age, sex, breed, genotype, seasons, districts and months.

### Model for $\chi^2$ test

The null and alternative hypotheses for chi square test in a contingency table are as follows:

H<sub>0</sub>: There is no association between the two variables. There is no relationship between gastro-intestinal nematodes and time, season, region, age, sex, breed and genotype of cattle.

H<sub>1</sub>: There is significant association between the two variables. There is relationship between gastro-intestinal nematodes and time, season, region, age, sex, breed and genotype of cattle.

If the calculated value is greater than the tabulated value than the hypothesis is reject and vice versa.

Chi square is the sum of the squared differences between observed (O) and the expected (E) data divided by the expected (E) data in all possible categories. It measures the agreement between experimentally obtained (observed) results and the expected results suggested by a theory or hypothesis.

Small values of chi square show good agreement and suggest that the hypothesis (null) is plausible and should not be rejected. Large values of chi square snow poor agreement and suggest that the basic hypothesis (null hypothesis) is probably wrong and should be

In contingency table problems we create an index that computes for each outcome cell

and then we sum this index over all cells. If O stands for observed count and E for expected count, the mathematical notation the formula looks like this:

$$\chi^2 = \sum_{i=1}^k \frac{(O-E)^2}{E}$$

$$= \frac{(O_1 - E_1)^2}{E_1} + \frac{(O_2 - E_2)^2}{E_2} + ... + \frac{(O_k - E_k)^2}{E_k}$$

This is chi square statistic. It represents the agreement between each individual expected value and the corresponding observed value.

Expected values are calculated as

$$E = \frac{(Row \ total) \ (Column \ total)}{Grand \ total}$$

$$= \frac{R \times C}{N}$$

That is expected frequency (E) for any cell of a  $r \times c$  table is obtained by multiplying the total (R) of the row to which it belongs by total of the column (O) to which it belongs and then dividing by the grand total (N).

If r is the number of rows and c is the number of columns in the contingency table, degrees of freedom of  $\chi^2$  statistic is calculated as

$$df = (no. of rows - 1) \times (no. of columns - 1)$$
$$= (r - 1) (c - 1).$$

The greater the value of  $\chi^2$  the greater would be the difference between observed and expected frequencies which yield p < 0.05. That is the difference could not arise due to fluctuations of sampling. This means, there is significant difference between the two variables.

#### Stepwise procedures for chi square test:

1. State the null hypothesis

H<sub>0</sub>: There is no association between the two variables

2. Determine the expected numbers for each observational class. Chi square should not be calculated if the expected value in any category is less than 5.

### 3. Calculate $\chi^2$

$$\chi^2 = \sum_{i=1}^k \frac{(O-E)^2}{E}$$

#### 4. Calculate degrees of freedom

$$df = (r-1)(c-1)$$

#### 5. Interpretation

If the p value for the calculated  $\chi^2$  with df (r-1)(c-1) is p < 0.05, reject  $H_0$  and conclude that there is significant association between the two variables.

### **CHAPTER-4**

#### **RESULTS**

Faecal samples were collected from 400 cattle from greater Rajshahi to find out the incidence of different gastro-intestinal nematodes. Screening of 10 indigenous medicinal plants of Bangladesh having anthelmintic activity was studied. A comparative study was conducted to evaluate the efficacy of two (2) indigenous medicinal plants (neem and korolla) and two (2) patent drugs ivermectin (Ivertin®) and levamisole (Levavet®); against natural gastro-intestinal nematode infections in 25 cattle. Each group consisted of 5 cattle. Ethanol extract of leaves of neem and korolla were administered orally @ 100 mg/kg b.wt. to the cattle of group B and C respectively. Cattle of group D and E were treated externally with ivermectin pour on preparation (Ivertin®) @ 200 µg/kg b.wt. and orally levamisole preparation (Levavet®) @ 7.5 mg/kg b.wt., respectively. Cattle of group A were kept as infected untreated control group i.e. without giving any treatment. Attempts were also made to investigate the effects of these indigenous medicinal plants (neem and korolla) and patent drugs (Ivertin® and Levavet®) on some hematological parameters (TEC, Hb, PCV, TLC, DLC, MCV, MCHC and MCH), biochemical parameters (ALT and AST) and clinical parameter (body weight) of cattle.

#### 4.1 Incidence of Gastro-intestinal Nematodiasis in Cattle

The incidence of gastro-intestinal nematodes in cattle of greater Rajshahi through a year is presented below. Incidence of Strongyles (*Haemonchus* sp., *Trichostrongylus* sp., *Oesophagostomum* sp. and *Mecistocirrus* sp.) infection was highest followed by *Ascaris* sp., *Bunostomum* sp., *Trichuris* sp., *Strongyloides* sp. and mixed infection. The incidence of gastro-

intestinal nematodes was more in cross breed than local breed. The incidence of gastro-intestinal nematodes was highest L×F genotype followed by L×F×SL genotype, L×SL genotype and local breed. The incidence of gastro-intestinal nematodes was highest in 12 to < 24 months old cattle followed by 6 to <12 months old cattle, > 24 months above and 1 to <6 months old cattle. The incidence of gastro-intestinal nematodes was more in female than male. Highest incidence of gastro-intestinal nematodes was found in rainy seasons (June-August) followed by autumn (September-November), summer (March-May) and winter seasons (December-February). The incidence of gastro-intestinal nematodes was highest in the month of July. The incidence of gastro-intestinal nematodes was highest in Rajshahi district followed by Natore district, Naogaon district and Chapai Nawabgonj district.

### 4.1.1 The Overall incidence of gastro-intestinal nematode parasites of 400 cattle in greater Rajshahi

The overall incidence of gastro-intestinal nematodes parasites of 400 cattle in greater Rajshahi is shown in Table 3 and Figure 1. Out of 400 cattle 219 (54.8%) were affected by different nematode parasites. Among the incidence, *Strongyles* were found highest 102 (25.5%) and *Bunostomum* sp. was lowest 9(2.25%).

Table 3: Overall incidence of nematode parasites in 400 cattle in greater Rajshahi

Name of Parasites	Number of Infected cattle	Percentage (%)
Ascaris sp.	26	6.5
Strongyles	102	25.5
Bunostomum sp.	9	2.25
Trichuris sp.	20	5.0
Stongyloides sp.	20	5.0
Mixed infection	42	10.5
Grand Total	219	54.75

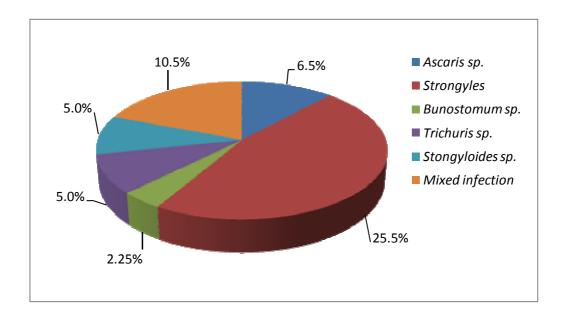


Fig. 1: Graphical presentation of incidence of gastro-intestinal nematode parasites of cattle in greater Rajshahi.

### 4.1.2 Average incidence of gastro-intestinal nematodes in relation with age of cattle in greater Rajshahi.

This infestation rate of gastro-intestinal parasites with their age was significantly (P<0.05) highest in 12 to <24 months old cattle 134 (67.0%) and lowest in 1 < 6 months old cattle 22 (36.7%) (Table 4, Figure 2 and Appendix I).

Table 4: Incidence of GI nematodes in relation with age in cattle.

Age	No. of total animal	No. of Infected Animal	Percentage (%) of infected animal	Chi square (χ²) calculated value	Chi square (χ²) tabulated value
1 to < 6 months	60	22	36.7		
7 to <12 months	40	25	62.5		
12 to <24 months	200	134	67.0	32.328	7.81
>24 months	100	38	38.0		
Total	400	219	54.8		

The calculated value of Chi square ( $\chi^2$ ) was 32.328. On the other hand the tabulated value of chi-square at (P<0.05) level of significance at 3 degree of freedom was 7.81. Since the calculated value of the test was higher than tabulated value, the null hypothesis may be rejected that means there is relation between gastro-intestinal nematode infections of cattle with their age.

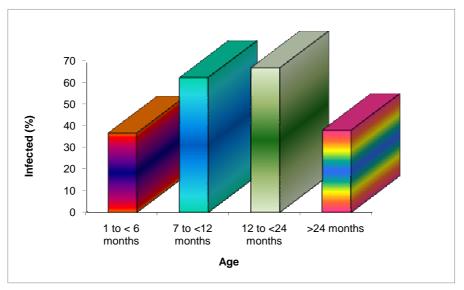


Fig 2. Graphical representation of Incidence of GI nematodes in relation with age of cattle in study area

### 4.1.3 Incidence of gastro-intestinal nematodes species in relation with age of cattle in greater Rajshahi.

The infestation rate of gastro-intestinal nematodes species in relation with age was observed and is shown in Table 5. Figure 3 and Appendix II. In 1 to <6 months old cattle *Ascaris* sp. infestation rate was significantly (P<0.05) was 33.3% followed by *Strongyles* 3.3%. The infestation of *Strongyles* was highest 25.0% in 7 to < 12 months old cattle followed by *Ascaris* sp. 15.0%, mixed infection 12.5%, *Trichuris* sp. 5.0% *Strongyloid* sp. 5.0%. The infestation of *Strongyles* was highest 35.0% followed by mixed infection 13.5% *Trichuris* sp. 7.5% *Strongyloid* sp.

7.0% and *Bunontomum* sp. 4.0% in 12 to <24 months old cattle. In age group of >24 months, the infestation of *Strongyles* was highest 20.0% followed by mixed infection 10% *Strongyloid* sp. 4.0%, *Trichuris* sp. 3.0% and *Bunontomum* sp. 1.0%. The calculated value of Chi square ( $\chi^2$ ) was 152.9. On the other hand the tabulated value of chi-square at (P<0.05) level of significance at 18 degree of freedom was 28.87. Since the calculated value of the test was higher than tabulated value, the null hypothesis may be rejected that means there is significant (P<0.05) effects of age on gastro-intestinal nematode infections of cattle.

Table 5: Incidence of GI nematodes species in different age of cattle at study area

			Name of p	parasite				Chi	Chi
Age of cattle (months)	Ascaris sp.	Strongyles	Bunosto- mum spp.	Trichuri s spp.	Strongy- loides	Mixed infection	Sub Total	square (χ²) calcula -ted value	square ( $\chi^2$ ) tabula- ted value
1 to <6 (Total no. 60)	20 (33.3%)	2 (3.3%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	22 (36.7%)		
7 to <12 (Total no. 40)	6 (15.0%)	10 (25.0%)	0 (0.0%)	2 (5.0%)	2 (5.0%)	5 (12.5%)	25 (62.5%)	152.9	28.87
12 to <24 (Total no. 200)	0 (0.0%)	70 (35.0%)	8 (4.0%)	15 (7.5%)	14 (7.0%)	27 (13.5%)	134 (67.0%)	132.9	20.07
> 24 (Total no. 100)	0 (0.0%)	20 (20.0%)	1 (1.0%)	3 (3.0%)	4 (4.0%)	10 (10.0%)	38 (38.0%)		

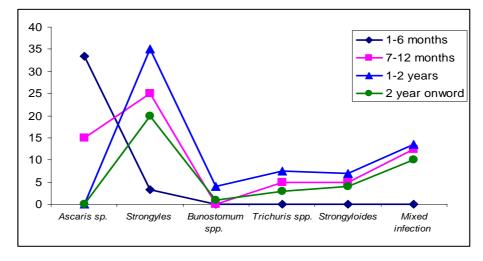


Fig. 3: Graph shows the influence of age on different gastro-intestinal nematodes of cattle in study area.

### 4.1.4 Incidence of gastro-intestinal nematodes with the sex of cattle in greater Rajshahi.

This infestation rate of gastro-intestinal parasites with their sex was significantly (P<0.05) higher in female cattle 147 (62.0%) than the male cattle 72 (44.2%). The data is presented in Table 6, Figure 4 and Appendix III.

Table 6: Incidence of gastro-intestinal nematodes in between sex of cattle in study area.

Sex	No. of total animal	No. of Infected Animal	Percentage of infected animal	Chi square (χ²) calculated value	Chi square (χ²) tabulated value
Female	237	147	62.0	10.10.5	201
Male	163	72	44.2	12.426	3.84
Total	400	218	54.8		

The calculated value of Chi square ( $\chi^2$ ) was 12.426. On the other hand the tabulated value of chi-square at (P<0.05) level of significance at 01 degree of freedom was 3.84. Since the calculated value of the test was higher than tabulated value, the null hypothesis may be rejected that means there is relation between gastro-intestinal nematode infections of cattle with their sex.

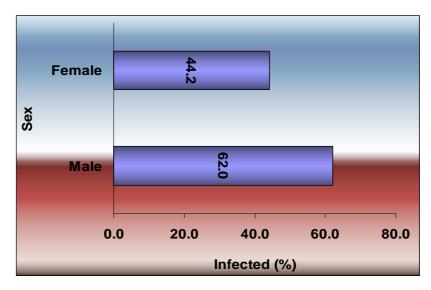


Fig. 4: Graph represents the incidence of gastro-intestinal nematodes in different sex of cattle in study area

### 4.1.5. Effect of breed on incidence of gastro-intestinal nematodes of cattle in greater Rajshahi.

Effect of breed on incidence of gastro-intestinal nematodes in relation with breed in cattle of greater Rajshahi is furnished in Table 7 and Fig. 5 and Appendix IV.

Table 7: Effect of breeds on incidence of gastro-intestinal nematodes in cattle of greater Rajshahi

Breed	No. of total animal	No. of Infected Animal	Percentage of infected animal	Chi square (χ²) calculated value	Chi square (χ²) tabulated value
Local	85	35	41.2		
Cross	315	184	58.4	8.027	3.84
Total	400	218	54.8		

This infestation rate of gastro-intestinal nematodes with their breed was significantly (P<0.05) higher in cross breed cattle 184 (58.4%) than in local breed cattle 35 (41.2%). The calculated value of Chi square ( $\chi^2$ ) was 8.027. On the other hand the calculated value of chi square at (P<0.05) level of significance at one degree of freedom was 3.84. Since the calculated value of the test was higher than tabulated value, the null hypothesis may be rejected that means there is relation between gastro-intestinal nematode infections of cattle with their breed.

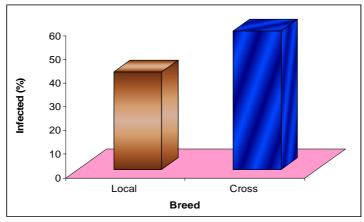


Fig. 5: Graph showing incidence of gastro-intestinal nematodes in different sex of cattle in greater Rajshahi

### 4.1.6 Incidence of gastro-intestinal nematodes in relation with genotype of cattle in greater Rajshahi.

This infestation rate of gastro-intestinal parasites with their genotype was significantly (P<0.05) highest in L×F genotype of cattle 56 (60.9%) and lowest in local cattle 35 (41.2%) (Table 8, Figure 6 and Appendix V).

Table 8: Incidence of gastro-intestinal nematodes in different genotype of cattle in study area.

Genotype	No. of total animal	No. of Infected Animal	Percentage of infected animal	Chi square (χ²) calculated value	Chi square (χ²) tabulated value
Local	85	35	41.2		
L×SL	143	82	57.3		
L×F	92	56	60.9	8.344	7.81
L×F×SL	80	46	57.5		
Total	400	219	54.8		

L = Local, SL = Shahiwal, F = Friesian,

The calculated value of Chi square ( $\chi^2$ ) was 8.344. On the other hand the tabulated value of chi-square at (P<0.05) level of significance at three degree of freedom was 7.81. Since the calculated value of the test was higher than tabulated value, the null hypothesis may be rejected that means there is relation between gastro-intestinal nematode infections of cattle with their genotype.

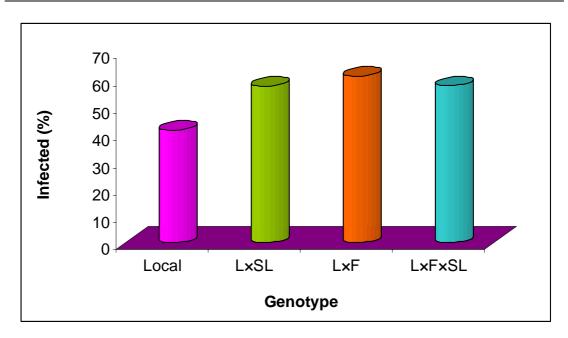


Fig. 6: Graph showing incidence of gastro-intestinal nematodes in different genotypes of cattle in study area

## 4.1.7 Incidence of gastro-intestinal nematodes in cattle in relation with seasons in greater Rajshahi.

The incidence of gastro-intestinal nematodes in relation with different seasons of greater Rajshahi is observed in Table 9, Figure 7 and Appendix VI.

Table 9: Incidence of gastro-intestinal nematodes in cattle in greater Rajshahi among different seasons

Season	No. of total animal	No. of infected animal	Percentage of infected animal	Chi square (\chi^2) calculated value	Chi square (\chi^2) tabulated value
Summer	97	43	44.3		
Rainy season	104	83	79.8		7.81
Autumn	104	60	57.7	46.331	
Winter	95	33	34.8		
Total	400	218	54.8		

The highest incidence was observed in rainy season 83 (79.8%) and lowest incidence was observed in winter 33 (34.8%). The calculated value of Chi square ( $\chi^2$ ) was 46.331. On the other hand the tabulated value of chi-square at (P<0.05) level of significance and three degree of freedom was 7.81. Since the calculated value of the test was higher than tabulated value, the null hypothesis may be rejected that means there is significant (P<0.05) effect of seasons on the incidence of gastro-intestinal nematode infections of cattle.

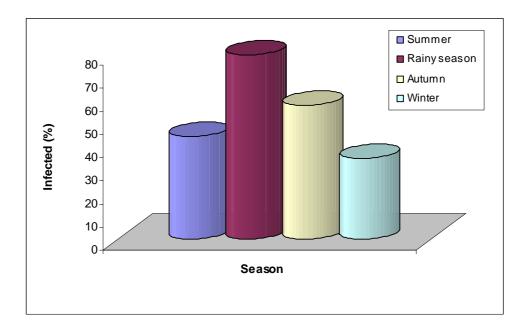


Fig. 7: Graphical presentation of the effects of seasons on incidence of gastro-intestinal nematodes in study area.

### 4.1.8 The influence of months in a year on incidence of gastro-intestinal nematodes in cattle in greater Rajshahi.

The incidence of gastro-intestinal nematodes in 400 cattle in greater Rajshahi during a year in relation with different months is summarized in Table 10, Figure 8 and Appendix VII. The highest incidence was observed

in July 30 (88.2%) and lowest incidence was observed in January 10 (33.3%). The calculated value of Chi square ( $\chi^2$ ) was 52.975. On the other

Table 10. The influence of month in a year on incidence of gastro-intestinal nematodes of cattle in greater Rajshahi

Month	Total no. of cattle	No. of infected cattle	ected of infected		Chi square (χ²) tabulated value
January	30	10	33.3		
February	32	11	34.4		
March	32	12	37.5		
April	35	15	42.9		
May	30	16	53.3		
June	35	27	77.1		
July	34	30	88.2	52.975	19.68
August	35	26	74.3		
September	34	23	67.6		
October	35	21	60.0		
November	35	16	45.7		
December	33	12	36.4		
Total	400	218	54.8		

hand the tabulated value of chi-square at (P<0.05) level of significance and eleven degree of freedom was 19.68. Since the calculated value of the test was higher than tabulated value, the null hypothesis may be rejected that means there is significant (P<0.05) effect of different months in a year on gastro-intestinal nematode infections of cattle.

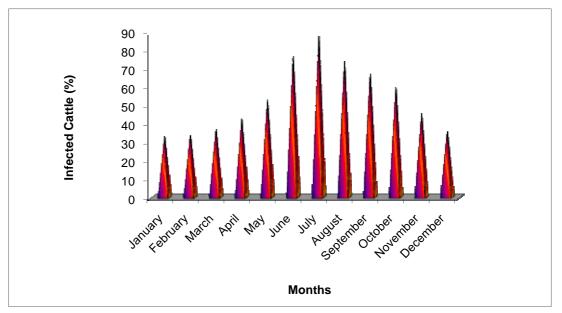


Fig. 8: Graph shows the influence of months in a year on incidence of gastrointestinal nematodes of cattle in greater Rajshahi

### 4.1.9 The incidence of Gastro-intestinal nematodes in cattle in relation with different districts of greater Rajshahi

The incidence of gastro-intestinal nematodes found in different districts of greater Rajshahi is presented in Table 11, Figure 9 and Appendix VIII. The highest incidence was observed in Rajshahi district 125 (62.5%) and lowest incidence was observed in Chapai Nawabgonj district 19 (38.0%). The calculated value of Chi square ( $\chi^2$ ) was 14.580. On the other hand, the tabulated value of chi-square at (P<0.05) level of significance and three degree of freedom was 7.81. Since the calculated value of the test was higher than tabulated value, the null hypothesis may be rejected that means there is significant (P<0.05) effect of districts on gastro-intestinal nematode infections of cattle.

Table 11: Incidence of Gastro-intestinal nematodes in different districts of greater Rajshahi

Name of District	Total Number	Infected Cattle	Percentage	Chi square (\chi^2) calculated value	Chi square (χ²) tabulated value
Rajshahi	200	125	62.5		
Chapai Nawabgonj	50	19	38.0		
Natore	80	45	56.2	14.580	7.81
Naogoan	70	30	42.9		
Total	400	218	54.8		

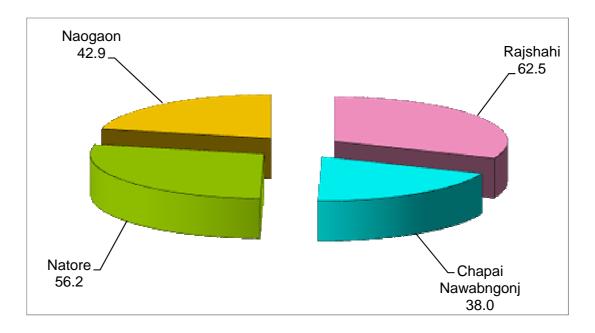


Fig. 9: Graphical representation of incidence of Gastro-intestinal nematodes among the districts of greater Rajshahi

#### 4.2. Results of in vitro Screening of Plants for Anthelmintic Activity

A large number of plants and plant materials were collected and processed for this experiment. Aqueous and ethanol extracts of ten selected plants were used for *in vitro* screening. The results obtained from *in vitro* screening of plants for anthelmintic activity are as follows:

## 4.2.1. *In vitro* screening of aqueous extracts of plants for anthelmintic activity

# 4.2.1.1 In vitro anthelmintic efficacy of 2.5 % (25 mg/ml) concentration of water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

Freshly prepared aqueous extracts of 10 indigenous medicinal plants (neem, custard apple, papaya, garlic, bitter gourd, jute, chaste tree, pineapple, pomegranate, betel leaf) and control group with PBS were screened *in vitro* in this study, at 2.5% (25 mg/ml) concentration by employing adult gastro-intestinal nematodes obtained from infected cattle. Within these 10 plants, 1 showed 30% efficacy against adult worms, 4 showed 20% and others showed 10% efficacy at the concentration of 25 mg/ml. The *in vitro* efficacy of water extracts of these 10 indigenous medicinal plants at 25 mg/ml concentration against adult gastro-intestinal nematodes of cattle are shown in the Table 12 and Fig. 10.

Table 12: *In vitro* anthelmintic efficacy of 2.5 % (25 mg/ml) concentration of water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

	Indigenous p	lants		Total	Number of	% of
Name of plants	Scientific name	English name	Part used	number of nematodes	dead GI Nematodes	dead parasite
Neem	Azadirachta indica	Neem	Leaves	10	3	30
Ata	Annona reticulata	Custard apple	leaves	10	1	10
Pepe	Carica papaya	Papaya	Leaves	10	1	10
Rashun	Allium sativum	Garlic	Bulbs	10	2	20
Korolla	Momordica charantia	Bitter Gourd	fruit & seeds	10	2	20
Deshi pat	Corchorus oletorious	Jute	Leaves	10	1	10
Nishinda	Vitex negundo	Chaste Tree	Leaves	10	1	10
Anaras	Ananas comosus	Pineapple	Leaves	10	2	20
Dalim	Punica granatum	Pomegranate	Leaves	10	1	10
Pan	Piper betle	Betel leaf	Leaves	10	2	20
PBS (Con	ntrol)			10	1	10

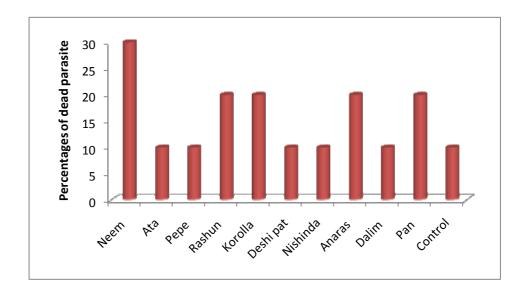


Fig. 10: Graphical presentation of anthelmintic efficacy of 10 indigenous medicinal plants of 2.5% water extract against adult gastro-intestinal nematodes in cattle

# 4.2.1.2 In vitro anthelmintic efficacy of 5% (50 mg/ml) concentration of water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

Freshly prepared aqueous extracts of 10 indigenous medicinal plants (neem, custard apple, papaya, garlic, bitter gourd, jute, chaste tree, pineapple, pomegranate, betel leaf) and control group with PBS were screened *in vitro* in this study, at 5% (50 mg/ml) concentration by employing adult gastro-intestinal nematodes obtained from infected cattle. Within these 10 plants, 2 showed 60% efficacy against adult worms, 3 showed 50%, 2 showed 40% and others showed 30% efficacy at the concentration of 50 mg/ml. The *in vitro* efficacy of water extracts of these 10 indigenous medicinal plants at 5% (50 mg/ml) concentration against adult gastro-intestinal nematodes in cattle are shown in the Table 13 and Fig. 11.

Table 13: *In vitro* anthelmintic efficacy of 5% (50mg/ml) water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

	Indigenous 1	Total	Number of	% of dead		
Name of plants	Scientific name	English name	Part used	number of nematodes	dead GI Nematodes	parasite
Neem	Azadirachta indica	Neem	Leaves	10	6	60
Ata	Annona reticulata	Custard apple	leaves	10	3	30
Pepe	Carica papaya	Papaya	Leaves	10	4	40
Rashun	Allium sativum	Garlic	Bulbs	10	5	50
Korolla	Momordica charantia	Bitter Gourd	fruit & seeds	10	6	60
Deshi pat	Corchorus oletorious	Jute	Leaves	10	3	30
Nishinda	Vitex negundo	Chaste Tree	Leaves	10	4	40
Anaras	Ananas comosus	Pineapple	Leaves	10	5	50
Dalim	Punica granatum	Pomegranate	Leaves	10	3	30
Pan	Piper betle	Betel leaf	Leaves	10	5	50
P	PBS (Control)			10	1	10

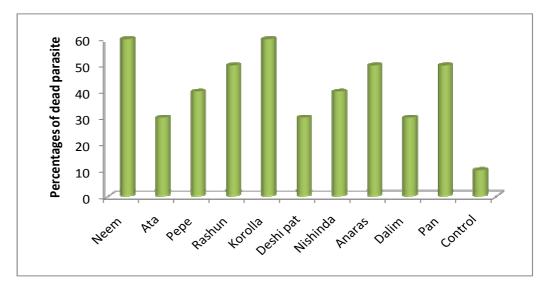


Fig. 11: Graphical presentation of anthelmintic efficacy of 5% (50 mg/ml) water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

# 4.2.1.3 In vitro anthelmintic efficacy of 10% (100 mg/ml) concentration of water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

Freshly prepared aqueous extracts of 10 indigenous medicinal plants (neem, custard apple, papaya, garlic, bitter gourd, jute, chaste tree, pineapple, pomegranate, betel leaf) and control group with PBS were screened *in vitro* in this study, at 10% (100 mg/ml) concentration by employing adult gastro-intestinal nematodes obtained from infected cattle. Within these 10 plants, 3 showed 100% efficacy against adult worms, 2 showed 90%, 2 showed 70% and others showed 60% efficacy at the concentration of 100 mg/ml. The *in vitro* efficacy of water extracts of these 10 indigenous medicinal plants at 10% (100 mg/ml) concentration against adult gastro-intestinal nematodes in cattle are shown in the Table 14 and Fig. 12.

Table 14: *In vitro* anthelmintic efficacy of 10% (100 mg/ml) concentration of water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

	Indigenous p	lants	Total	Number of dead/	% of	
Name of plants	Scientific name	English name	Part used	number of nematodes	nonmortile GI Nematodes	dead parasites
Neem	Azadirachta indica	Neem	Leaves	10	10	100
Ata	Annona reticulata	Custard apple	leaves	10	6	60
Pepe	Carica papaya	Papaya	Leaves	10	7	70
Rashun	Allium sativum	Garlic	Bulbs	10	9	90
Korolla	Momordica charantia	Bitter Gourd	fruit & seeds	10	10	100
Deshi pat	Corchorus oletorious	Jute	Leaves	10	6	60
Nishinda	Vitex negundo	Chaste Tree	Leaves	10	7	70
Anaras	Ananas comosus	Pineapple	Leaves	10	9	90
Dalim	Punica granatum	Pomegranate	Leaves	10	6	60
Pan	Piper betle	Betel leaf	Leaves	10	10	100
PBS(Control)				10	1	10

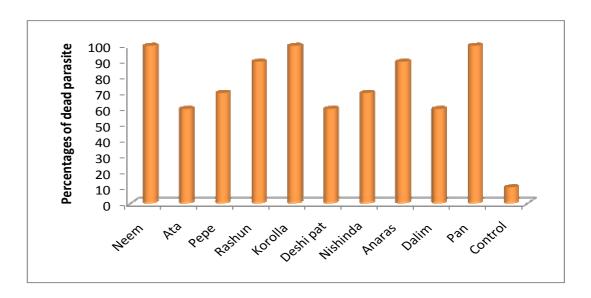


Fig. 12: The graph represents the anthelmintic efficacy of 10% (100mg/ml) water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

# 4.2.1.4 Comparative efficacy of different concentrations of water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

Freshly prepared aqueous extracts of 10 indigenous medicinal plants (neem, custard apple, papaya, garlic, bitter gourd, jute, chaste tree, pineapple, pomegranate, betel leaf) and control group with PBS were screened in-vitro in this study, at various concentrations by employing adult gastro-intestinal nematodes obtained from infected cattle. The efficacy of different plants at a concentration level of 25 mg/ml and 50 mg/ml were lower than that of concentration of 100 mg/ml. The in-vitro efficacy of different concentrations of water extracts of these 10 indigenous medicinal plants against adult gastro-intestinal nematodes in cattle are shown in the Table 15 and Fig. 13.

Table 15: Comparative anthelmintic efficacy of different concentrations of water extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

	Indigenou	ıs plants		Percentage (%) of non-motile adult worms at different concentrations of water extracts of plant			
Name of plants	Scientific name	English name	Part used	25 mg/ml	50 mg/ml	100 mg/ml	
Neem	Azadirachta indica	Neem	Leaves	30	30	100	
Ata	Annona reticulata	Custard apple	leaves	10	40	60	
Pepe	Carica papaya	Papaya	Leaves	10	50	70	
Rashun	Allium sativum	Garlic	Bulbs	20	60	90	
Korolla	Momordica charantia	Bitter Gourd	fruit & seeds	20	30	100	
Deshi pat	Corchorus oletorious	Jute	Leaves	10	40	60	
Nishinda	Vitex negundo	Chaste Tree	Leaves	10	50	70	
Anaras	Ananas comosus	Pineapple	Leaves	20	30	90	
Dalim	Punica granatum	Pomegranate	Leaves	10	50	60	
Pan	Piper betle	Betel leaf	Leaves	20	30	100	

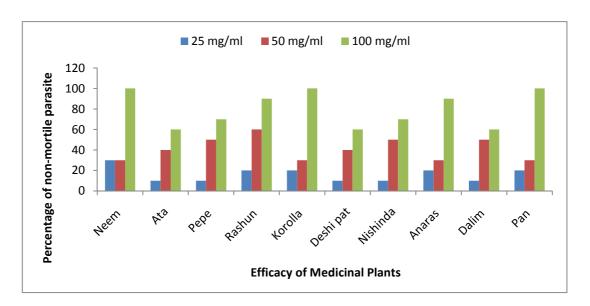


Fig. 13: Graph showing the efficacy of water extract of 10 indigenous medicinal plants against gastro-intestinal nematodes of cattle in different concentration.

### **4.2.2** *In vitro* screening of ethanol extracts of plants for anthelmintic activity

## 4.2.2.1 In vitro anthelmintic efficacy of 2.5% (25 mg/ml) concentration of ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

Freshly prepared ethanol extracts of 10 indigenous medicinal plants (neem, custard apple, papaya, garlic, bitter gourd, jute, chaste tree, pineapple, pomegranate, betel leaf) and control group with PBS were screened *in vitro* in this study, at 2.5% (25mg/ml) concentration by employing adult gastro-intestinal nematodes obtained from infected cattle. Within these 10 plants, 3 showed 50% efficacy against adult worms, 3 showed 40% and others showed 30% efficacy at the concentration of 25 mg/ml. The *in vitro* efficacy of ethanol extracts of these 10 indigenous medicinal plants at 25 mg/ml concentration against adult gastro-intestinal nematodes in cattle are shown in the Table 16 and Fig. 14. The result obtained from this screening almost similar as in the case of aqueous extracts of plants at 50 mg/ml concentration. However,

ethanol extracts were more effective against adult worms even at lower concentration level.

Table 16: *In vitro* anthelmintic efficacy of 2.5 % (25mg/ml) ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

	Indigenous p	lants	Total	Number of	Percentages	
Name of plants	Scientific name	English name	Part used	number of nematodes	dead GI Nematodes	of dead parasite
Neem	Azadirachta indica	Neem	Leaves	10	5	50
Ata	Annona reticulata	Custard apple	leaves	10	3	30
Pepe	Carica papaya	Papaya	Leaves	10	4	40
Rashun	Allium sativum	Garlic	Bulbs	10	4	40
Korolla	Momordica charantia	Bitter Gourd	fruit & seeds	10	5	50
Deshi pat	Corchorus oletorious	Jute	Leaves	10	3	30
Nishinda	Vitex negundo	Chaste Tree	Leaves	10	3	30
Anaras	Ananas comosus	Pineapple	Leaves	10	4	40
Dalim	Punica granatum	Pomegranate	Leaves	10	3	30
Pan	Piper betle	Betel leaf	Leaves	10	5	50
PBS(Cont	PBS(Control)			10	1	10

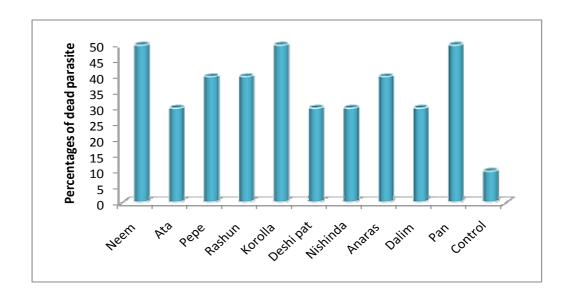


Fig. 14: Graphical representation of anthelmintic efficacy of 2.5% (25 mg/ml) ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

## 4.2.2.2 In vitro anthelmintic efficacy of 5% (50 mg/ml) concentration of ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

Freshly prepared ethanol extracts of 10 indigenous medicinal plants (neem, custard apple, papaya, garlic, bitter gourd, jute, chaste tree, pineapple, pomegranate, betel leaf) and control group with PBS were screened *in vitro* in this study, at 5% (50 mg/ml) concentration by employing adult gastro-intestinal nematodes obtained from infected cattle.

Table 17: *In vitro* anthelmintic efficacy of 5% (50mg/ml) ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

	Indigenous pl	Total	Number of	Percentages		
Name of plants	Scientific name	English name	Part used	number of nematodes	dead GI Nematodes	of dead parasite
Neem	Azadirachta indica	Neem	Leaves	10	10	100
Ata	Annona reticulata	Custard apple	leaves	10	6	60
Pepe	Carica papaya	Papaya	Leaves	10	7	70
Rashun	Allium sativum	Garlic	Bulbs	10	9	90
Korolla	Momordica charantia	Bitter Gourd	fruit & seeds	10	10	100
Deshi pat	Corchorus oletorious	Jute	Leaves	10	6	60
Nishinda	Vitex negundo	Chaste Tree	Leaves	10	7	70
Anaras	Ananas comosus	Pineapple	Leaves	10	9	90
Dalim	Punica granatum	Pomegranate	Leaves	10	6	60
Pan	Piper betle	Betel leaf	Leaves	10	10	100
PBS(Control)				10	1	10

Within these 10 plants, 3 showed 100% efficacy against adult worms, 2 showed 90%, 2 showed 70% and others showed 60% efficacy at the concentration of 50 mg/ml. The *in vitro* efficacy ethanol extracts of these 10 indigenous medicinal plants at 50 mg/ml concentration against adult gastro-intestinal nematodes in cattle are shown in the Table 17 and Fig. 15. The result obtained from this screening almost similar as in the case of aqueous extract of plants at 100 mg/ml concentration. However, ethanol extracts were more effective against adult worms even at lower concentration level.

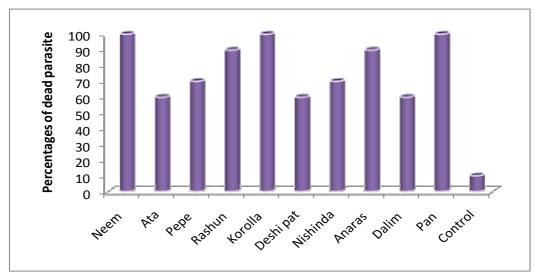


Fig. 15: Graphical presentation of anthelmintic efficacy of 5% (50 mg/ml) concentration of ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

## 4.2.2.3 In vitro anthelmintic efficacy of 10% (100 mg/ml) concentration of ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

Freshly prepared ethanol extracts of 10 indigenous medicinal plants (neem, custard apple, papaya, garlic, bitter gourd, jute, chaste tree, pineapple, pomegranate, betel leaf) and control group with PBS were screened *in vitro* in this study, at 10% (100 mg/ml) concentrations by employing adult gastro-intestinal nematodes obtained from infected

cattle. Within these 10 plants, 5 showed 100% efficacy against adult worms, 3 showed 90% and others showed 80% efficacy at the concentration of 100 mg/ml.

Table 18: *In vitro* anthelmintic efficacy of 10% (100 mg/ml) ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

	Indigenous	plants	Total number	Number of	Percentages		
Name of plants	Scientific name	English name	Part used	of nematodes	dead GI Nematodes	of dead parasite	
Neem	Azadirachta indica	Neem	Leaves	10		100	
Ata	Annona reticulata	Custard apple	leaves	10	8	80	
Pepe	Carica papaya	Papaya	Leaves	10	9	90	
Rashun	Allium sativum	Garlic	Bulbs	10	10	100	
Korolla	Momordica charantia	Bitter Gourd	fruit & seeds	10	10	100	
Deshi pat	Corchorus oletorious	Jute	Leaves	10	9	90	
Nishinda	Vitex negundo	Chaste Tree	Leaves	10	9	90	
Anaras	Ananas comosus	Pineapple	Leaves	10	10	100	
Dalim	Punica granatum	Pomegranate	Leaves	10	8	80	
Pan	Piper betle	Betel leaf	Leaves	10	10	100	
PBS(Control)				10	1	10	

The *in vitro* efficacy ethanol extracts of these 10 indigenous medicinal plants at 100 mg/ml concentration against adult gastro-intestinal nematodes in cattle are shown in the Table 18 and Fig. 16. However, ethanol extracts of plants at 100 mg/ml concentration were more effective

against adult worms than water extracts of plants at 100 mg/ml concentration level.

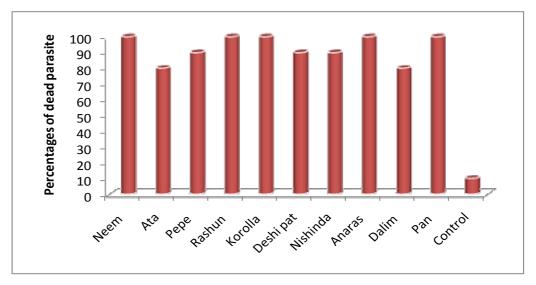


Fig. 16: Graph showing anthelmintic efficacy of 10% (100 mg/ml) concentration of ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

## 4.2.2.4 Comparative anthelmintic efficacy of different concentrations of ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

Freshly prepared ethanol extracts of 10 indigenous medicinal plants (neem, custard apple, papaya, garlic, bitter gourd, jute, chaste tree, pineapple, pomegranate, betel leaf) and control group with PBS were screened *in vitro* in this study, at various concentrations by employing adult gastro-intestinal nematodes obtained from infected cattle. The efficacy of different plants at 100 mg/ml concentration was highest among different concentration. The *in vitro* efficacy of different concentration of ethanol extracts of these 10 indigenous medicinal plants against adult gastro-intestinal nematodes in cattle are shown in the Table 19 and Fig. 17.

Table 19: Comparative anthelmintic efficacy of different concentrations of ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

	Indigenou	ıs plants		Percent non-motile adult worms at different concentrations of ethanol extracts of plant			
Name of plants	Scientific name	English name	Part used	25 mg/ml	50 mg/ml	100 mg/ml	
Neem	Azadirachta indica	Neem	Leaves	50	90	100	
Ata	Annona reticulata	Custard apple	leaves	30	60	80	
Pepe	Carica papaya	Papaya	Leaves	40	70	90	
Rashun	Allium sativum	Garlic	Bulbs	40	80	100	
Korolla	Momordica charantia	Bitter Gourd	fruit & seeds	50	90	100	
Deshi pat	Corchorus oletorious	Jute	Leaves	30	60	90	
Nishinda	Vitex negundo	Chaste Tree	Leaves	30	70	90	
Anaras	Ananas comosus	Pineapple	Leaves	40	80	100	
Dalim	Punica granatum	Pomegranate	Leaves	30	60	80	
Pan	Piper betle	Betel leaf	Leaves	50	70	100	

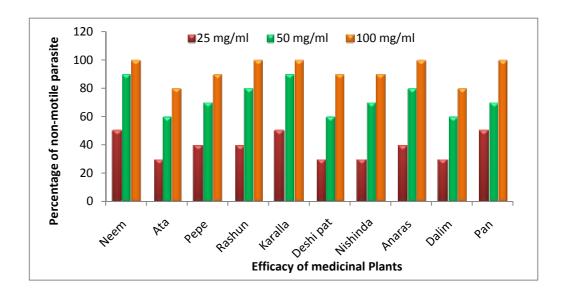


Fig. 17: Graphical representation of anthelmintic efficacy of different concentrations of ethanol extracts of 10 indigenous medicinal plants against adult gastro-intestinal nematodes of cattle

Chapter 4 Results 

Results 

191

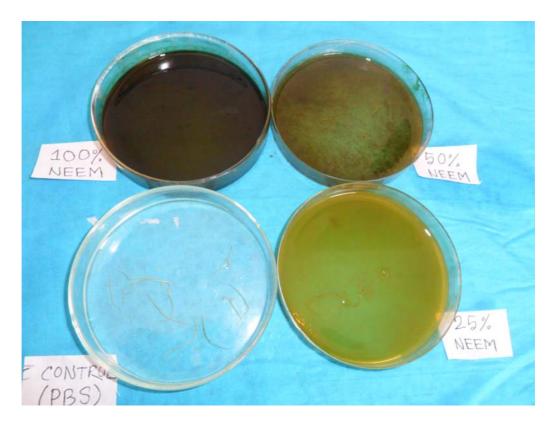


Plate 69. In vitro trial of ethanol extract of Neem

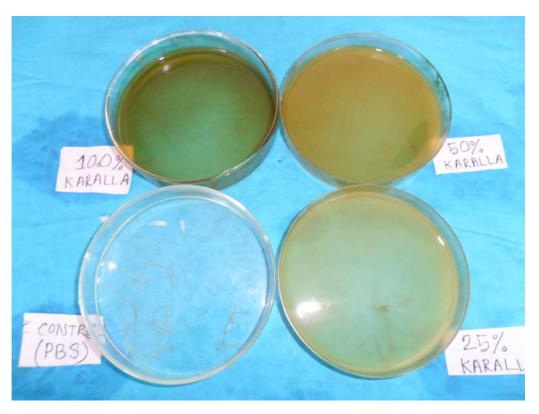


Plate 70. In vitro trial of ethanol extract of Korolla

#### 4.3 Results Obtained from in vivo Screening

For *in vivo* screening, only ethanol extracts of two selected plants were used, which were found to be most potential against adult gastro-intestinal nematodes *in vitro*. *In vivo* screening was performed to compare the efficacy of two selected ethanol extracts of plants with patent anthelmintics. The results of *in vivo* screening of different plants extracts and patent drugs against gastro-intestinal nematodes are presented below:

## 4.3.1 Efficacy of two (2) indigenous medicinal plants and two (2) patent drugs against natural gastro-intestinal nematodes in cattle

The effects of neem leaves (orally), Korolla (fruits), Ivermectin (Pour on formulation) and levamisole (orally) against gastro-intestinal nematodes in cattle are presented in Table 20- 21 and Figure 18 and Appendix IX. The number of ova varied from 660±47.33 to 870±43.59 at day 0 (pre treatment). The significant (P<0.05) lowest no. of EPG were found ivermectin and levamisole treated cattle (0 $\pm$ 0.00 and 0 $\pm$ 0.00 respectively) and highest in control group (710±36.74) at 7 days after treatment. The EPG was obtained minimum in ivermectin (0±0.00) treated cattle and maximum in control group (796±42.61) at 14 days after post treatment. The lowest no. of EPG in D and E groups (0±0.00 and 86±30.76 respectively) at 21 days after treatment and highest in control group (910±43.01). Significantly (P<0.05) lowest no. of EPG was found in group D and E (0±0.00 and 142±31.21 respectively) than the group of A, B, C at 28 days after treatment. Ivertin® and Levavet® were most effective, neem was found to be moderately effective and bitter gourd was less effective against gastro-intestinal nematodes in cattle The EPG counts of control group (A) were significantly (p<0.05) increased up to the last day of experimental period.

Table 20: The effects of Neem leaves (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) against G.I nematodes in Cattle.

		Pre- treatment	Post-treatment							
		Day 'O'	7 da	ıy	14 day		21 da	ay	28 day	
Group	Group Treatment (Mo		EPG (Mean ±SE)	EPG Increase /decrease (%)	EPG (Mean ±SE)	EPG Increase /decrease (%)	EPG (Mean ±SE)	EPG Increase /decrease (%)	EPG (Mean ±SE)	EPG Increase /decrease (%)
A	No Drug (Control)	660±47.33 <sup>b</sup>	710±36.74 <sup>a</sup>	-7.58	796±42.61 <sup>a</sup>	-20.61	910±43.01 <sup>a</sup>	-37.88	1050±52.44 <sup>a</sup>	-59.09
В	Neem leaves	870±43.59 <sup>a</sup>	410±50.99 <sup>b</sup>	+52.87	330±51.48 <sup>b</sup>	+62.07	444±50.36 <sup>b</sup>	+48.97	562±33.97°	+35.40
С	Korolla fruit	860±64.03°	490±57.88 <sup>b</sup>	+43.02	428±50.34 <sup>b</sup>	+50.23	560±65.50 <sup>b</sup>	+34.88	714±66.15 <sup>b</sup>	+16.98
D	Ivermectin 200mg/kg pouron	730±47.85 <sup>ab</sup>	0±0.00°	+100.00	0±0.00°	+100.00	0±0.00°	+100.00	0±0.00°	+100.00
Е	Levamisole 7.5 mg/kg (Orally)	662±33.38 <sup>b</sup>	$0\pm0.00^{c}$	+100.00	34±18.87°	+94.86	86±30.76°	+87.01	142±31.21 <sup>d</sup>	+78.55
Total		756.4±27.25	322±59	9.38	317.6±	61.37	400±69	9.80	493.6±7	9.95

The above values represent the mean±Standard Error (SE) of 5 cattle

'+' = Decrease

The values are mean±SE, a,b,c,d,e mean±SE with different supercript letters in same column deffers significantly with each other (P<0.05)

'-' = Increase

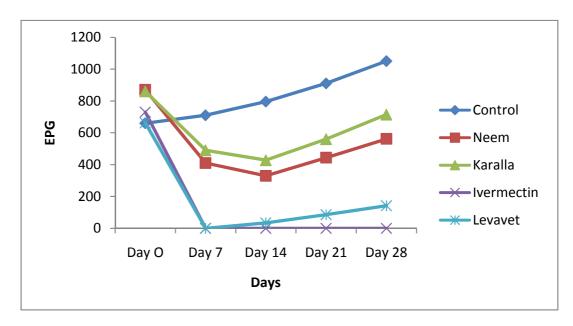


Fig. 18: Graph is showing the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) against G.I nematodes in Cattle.

Table 21: Analysis of variance for EPG of Cattle by the influences of different drugs during experimental periods.

Sou	rces of variation	Sum of Squares	df	Mean Square	F	Sig.
	Between Groups	212696.000	4	53174.000	4.567	.009
day0	Within Groups	232880.000	20	11644.000		
	Total	445576.000	24			
	Between Groups	1.969E6	4	492350.000	67.445	.000
day7	Within Groups	146000.000	20	7300.000		
	Total	2.115E6	24			
	Between Groups	2.113E6	4	528134.000	71.796	.000
day14	Within Groups	147120.000	20	7356.000		
	Total	2.260E6	24			
	Between Groups	2.731E6	4	682790.000	70.961	.000
day21	Within Groups	192440.000	20	9622.000		
	Total	2.924E6	24			
	Between Groups	3.650E6	4	912624.000	98.619	.000
day28	Within Groups	185080.000	20	9254.000		
	Total	3.836E6	24			

# 4.3.2. Effects of two (2) indigenous medicinal plants and two (2) patent drugs on haematological parameters in cattle

The results of the effects of two (2) indigenous medicinal plants and two (2) patent drugs on different haematological parameters are shown in the Table 22-43, Fig. 19-29 and Appendix X-XX.

### **4.3.2.1** *Total erythrocyte count (TEC, million/cu.mm.)*

Neem and bitter gourd and two patent drugs i.e. Ivertin® and Levavet® caused significant changes on total erythrocyte count (TEC). The TEC was increased significantly (p<0.05) after treated with neem, bitter gourd, Ivertin® and Levavet® in group B, C, D and E respectively. However, TEC was increased significantly (p<0.05) in Ivertin® treated cattle of group D. The TEC were decreased significantly (p<0.05) in control group A. The results of the effects of two (2) indigenous medicinal plants and two (2) patent drugs on total erythrocyte count in cattle are shown in the Table 22-23, Fig. 19 and Appendix X.

Table 22: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on total erythrocyte count (million/cu.mm.) in cattle.

Gro	Treatm	Pre- treatment	Post-treatment					
up	ent	Day 'O'	7 day	14 day	21 day	28 day		
		(Mean ±SE)	(Mean ±SE)	(Mean ±SE)	(Mean ±SE)	(Mean ±SE)		
A	No Drug (Control)	9.11±0.24 <sup>a</sup>	9.00±0.22 <sup>a</sup>	8.82±0.23 <sup>b</sup>	8.66±0.21 <sup>b</sup>	8.44±2.20°		
В	Neem leaves	8.31±0.23 <sup>a</sup>	8.37±0.23 <sup>a</sup>	8.66±0.20 <sup>b</sup>	8.92±0.21 <sup>b</sup>	9.04±0.19 <sup>b</sup>		
С	Korolla fruit	8.62±0.35 <sup>ab</sup>	8.66±0.37 <sup>a</sup>	8.82±0.35 <sup>b</sup>	8.99±0.35 <sup>b</sup>	9.09±0.35 <sup>b</sup>		
D	Ivermectin 200mg/kg pour-on	8.33±0.06 <sup>c</sup>	8.45±0.05 <sup>a</sup>	9.23±0.06 <sup>ab</sup>	10.11±0.05 <sup>a</sup>	10.61±0.04 <sup>a</sup>		
Е	Levamisole 7.5 mg/kg (Orally)	8.88±0.23 <sup>ab</sup>	9.00±0.22 <sup>a</sup>	9.65±0.17 <sup>a</sup>	10.18±0.06 <sup>a</sup>	10.50±0.05 <sup>a</sup>		
Total		8.65±0.11	8.68±0.11	9.04±0.11	9.37±0.15	9.54±0.19		

The above values represent the mean±Standard Error (SE) of 5 cattle

The values are mean $\pm$ SE, a,b,c mean $\pm$ SE with different supercript letters in same column deffers significantly with each other (P<0.05)

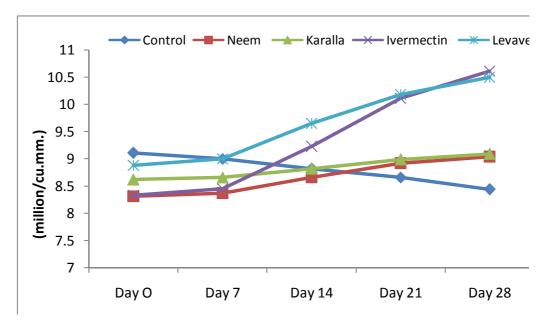


Fig. 19: The graphical representation of the effects of various effective drugs on total erythrocyte count (million/cu.mm.) in cattle.

Table 23: Analysis of variance for total erythrocyte count (million/cu.mm.) in cattle by treated with various effective drugs during experimental periods.

Sou	rces of variation	Sum of Squares	df	Mean Square	F	Sig.
	Between Groups	2.406	4	.602	2.023	.130
day0	Within Groups	5.946	20	.297		
	Total	8.353	24	Į.		
	Between Groups	1.764	4	.441	1.551	.226
day7	Within Groups	5.689	20	.284		
	Total	7.453	24			
	Between Groups	3.234	4	.809	3.083	.039
day14	Within Groups	5.246	20	.262		
	Total	8.480	24			
	Between Groups	10.292	4	2.573	11.277	.000
day21	Within Groups	4.563	20	.228		
	Total	14.855	24			
	Between Groups	18.691	4	4.673	22.275	.000
day28	Within Groups	4.196	20	.210		
	Total	22.887	24			

#### 4.3.2.2 Haemoglobin content (Hb, g %)

The haemoglobin (Hb) content was also increased significantly (p<0.05) after treated with neem, bitter gourd, Ivertin® and Levavet® in group B, C, D and E respectively. The results of the effects of two (2) indigenous medicinal plants and two (2) patent drugs on hemoglobin content in cattle are shown in the Table 24-25 Fig. 20 and Appendix XI.

Table 24: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on haemoglobin content (g %) in cattle.

G		Pre- treatment	Post-treatment					
Group	Treatment	Day 'O'	7 day	14 day	21 day	28 day		
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)		
A	No Drug (Control)	10.44±0.35 <sup>a</sup>	9.72±0.41 <sup>a</sup>	9.12±0.41 <sup>b</sup>	8.28±0.28 <sup>c</sup>	7.80±0.16 <sup>c</sup>		
В	Neem leaves	8.58±0.37 <sup>b</sup>	9.00±0.38 <sup>a</sup>	9.90±0.36 <sup>ab</sup>	10.70±0.24 <sup>b</sup>	11.48±0.21 <sup>b</sup>		
С	Korolla fruit	9.08±0.57 <sup>b</sup>	9.36±0.52 <sup>a</sup>	9.82±0.47 <sup>ab</sup>	10.40±0.43 <sup>b</sup>	11.06±0.36 <sup>b</sup>		
D	Ivermectin 200mg/kg pour-on	9.12±0.32 <sup>b</sup>	9.76±0.38 <sup>a</sup>	10.72±0.32 <sup>a</sup>	11.72±0.16 <sup>a</sup>	12.76±0.17 <sup>a</sup>		
Е	Levamisole 7.5 mg/kg (Orally)	9.72±0.41 <sup>ab</sup>	10.04±0.38 <sup>a</sup>	10.84±0.30 <sup>a</sup>	11.68±0.23 <sup>a</sup>	12.40±0.17 <sup>a</sup>		
Total		9.38±0.21	9.57±0.18	10.08±0.20	10.55±0.28	11.10±0.37		

The above values represent the mean±Standard Error (SE) of 5 cattle

The values are mean  $\pm$  SE, a,b,c mean  $\pm$  SE with different supercript letters in same column deffers significantly with each other (P<0.05)

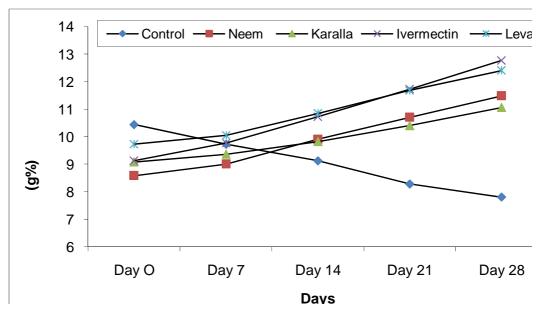


Fig. 20: Graphical representation of the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on haemoglobin content (g %) in cattle.

Table 25: Analysis of variance for haemoglobin content (g %) in cattle by the used of different drugs during experimental periods.

Sou	rces of variation	Sum of Squares	df	Mean Square	F	Sig.
	Between Groups	10.182	4	2.546	2.922	.047
day0	Within Groups	17.424	20	.871		
	Total	27.606	24			
	Between Groups	3.242	4	.810	.901	.482
day7	Within Groups	17.984	20	.899		
	Total	21.226	24			
	Between Groups	10.044	4	2.511	3.450	.027
day14	Within Groups	14.556	20	.728		
	Total	24.600	24			
	Between Groups	39.218	4	9.804	23.960	.000
day21	Within Groups	8.184	20	.409		
	Total	47.402	24			
	Between Groups	77.430	4	19.358	75.204	.000
day28	Within Groups	5.148	20	.257		
	Total	82.578	24			

## 4.3.2.3 Packed cell volume (PCV, %)

The packed cell volume (PCV) was increased significantly (p<0.0l) after treated with neem, bitter gourd, Ivertin® and Levavet® in group B, C,D, E, respectively. The results of the effects of two (2) indigenous medicinal plants and two (2) patent drugs on packed cell volume in cattle are shown in the Table 26-27, Fig. 21 and Appendix XII.

Table 26: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on packed cell volume (%) in cattle.

		Pre- treatment		Post-tre	atment	
Group	Treatment	Day 'O'	7 day	14 day	21 day	28 day
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)
A	No Drug (Control)	31.94±0.54 <sup>a</sup>	31.24±0.48 <sup>a</sup>	30.76±0.59°	29.74±0.38°	28.67±0.59 <sup>b</sup>
В	Neem leaves	30.22±0.82ª	30.94±0.70 <sup>a</sup>	31.64±0.60 <sup>bc</sup>	32.89±0.65 <sup>b</sup>	33.80±0.53 <sup>a</sup>
С	Korolla fruit	31.01±1.05 <sup>a</sup>	31.74±0.87 <sup>a</sup>	32.48±0.84 <sup>abc</sup>	33.51±0.71 <sup>ab</sup>	33.91±0.68 <sup>a</sup>
D	Ivermectin 200mg/kg pour- on	31.63±0.54 <sup>a</sup>	32.30±0.63 <sup>a</sup>	33.57±0.66 <sup>ab</sup>	34.43±0.62 <sup>ab</sup>	35.10±0.58 <sup>a</sup>
E	Levamisole 7.5 mg/kg (Orally)	32.26±0.63 <sup>a</sup>	32.87±0.47 <sup>a</sup>	34.04±0.39 <sup>a</sup>	34.73±0.42 <sup>a</sup>	35.34±0.42 <sup>a</sup>
Total		31.41±0.33	31.81±0.30	32.49±0.35	33.06±0.43	33.36±0.54

The above values represent the mean±Standard Error (SE) of 5 cattle

The values are mean $\pm$ SE, a,b,c mean $\pm$ SE with different supercript letters in same column deffers significantly with each other (P<0.05)

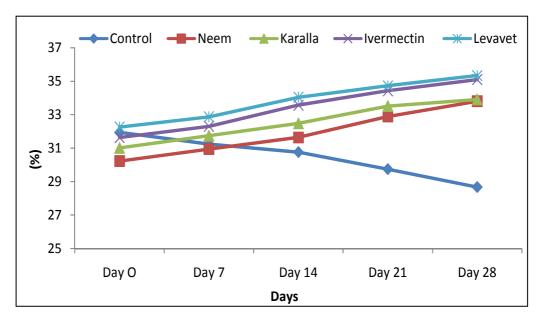


Fig. 21: Graph showing the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on packed cell volume (%) in cattle.

Table 27: Analysis of variance for packed cell volume (%) in cattle among the different drugs.

Sou	rces of variation	Sum of Squares	df	Mean Square	F	Sig.
	Between Groups	13.161	4	3.290	1.182	.349
day0	Within Groups	55.676	20	2.784		
	Total	68.836	24			
	Between Groups	12.250	4	3.063	1.445	.256
day7	Within Groups	42.394	20	2.120		
	Total	54.644	24			
	Between Groups	36.419	4	9.105	4.475	.010
day14	Within Groups	40.695	20	2.035		
	Total	77.114	24			
	Between Groups	79.625	4	19.906	12.028	.000
day21	Within Groups	33.100	20	1.655		
	Total	112.725	24			
	Between Groups	147.209	4	36.802	22.710	.000
day28	Within Groups	32.411	20	1.621		
	Total	179.621	24			

## 4.3.2.4 Total leukocyte count (TLC, thousand/cu.mm.)

The total leukocyte count (TLC) was decreased significantly (p<0.05) after treated with neem, bitter gourd, Ivertin® and Levavet® in group B, C, D, E respectively.

The TLC was increased significantly (p<0.05) in control group A. The results of the effect of two (2) indigenous medicinal plants and two (2) patent drugs on total leukocyte count in cattle are shown in the Table 28-29 Fig. 22 and Appendix XIII. The total number of lukocite (thousand /cu.mm.) had no significant effect (P>0.05) on days 0, 7 and 14 by except 21 and 28 days after treatment among the different drugs.

Table 28: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on total leukocyte count (thousand/cu.mm.) in cattle.

	_	Pre- treatment	Post-treatment					
Group	Treatment	Day 'O'	7 day	14 day	21 day	28 day		
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)		
A	No Drug (Control)	9.30±0.52	9.75±0.46	10.01±0.48	10.57±0.40 <sup>a</sup>	11.13±0.35 <sup>a</sup>		
В	Neem leaves	10.34±0.38	10.13±0.38	9.87±0.38	9.63±0.40 <sup>ab</sup>	9.35±0.42 <sup>b</sup>		
С	Korolla fruit	9.99±0.59	9.78±0.58	9.55±0.59	9.39±0.58 <sup>abc</sup>	9.18±0.54 <sup>bc</sup>		
D	Ivermectin 200mg/kg pour-on	10.18±0.21	9.97±0.21	9.46±0.26	9.21±0.22 <sup>bc</sup>	8.83±0.24 <sup>c</sup>		
Е	Levamisole 7.5 mg/kg (Orally)	9.20±0.41	9.16±0.27	8.68±0.27	8.19±0.32 <sup>c</sup>	8.06±0.23 <sup>c</sup>		
Total		9.80±0.20	9.75±0.17	9.51±0.19	9.39±0.22	9.31±0.25		

The above values represent the mean±Standard Error (SE) of 5 cattle

The values are mean $\pm$ SE, a,b,c mean $\pm$ SE with different supercript letters in same column deffers significantly with each other (P<0.05)

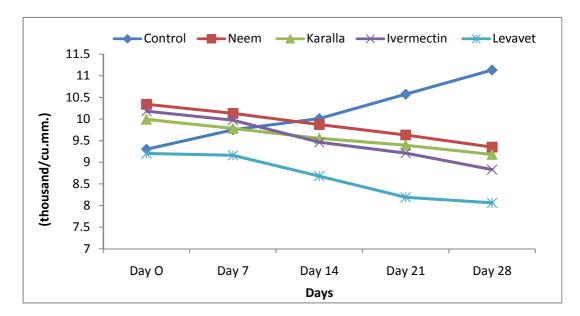


Fig. 22: Graph represents the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on total leukocyte count (thousand/cu.mm.) in cattle.

Table 29: Analysis of variance for total leukocyte count (thousand/cu.mm.) in cattle by the different drugs.

Sou	rces of variation	Sum of Squares	df	Mean Square	F	Sig.
	Between Groups	5.425	4	1.356	1.370	.280
day0	Within Groups	19.805	20	.990		
	Total	25.230	24			
	Between Groups	2.707	4	.677	.820	.528
day7	Within Groups	16.506	20	.825		
	Total	19.213	24			
	Between Groups	5.362	4	1.340	1.521	.234
day14	Within Groups	17.627	20	.881		
	Total	22.988	24			
	Between Groups	14.610	4	3.653	4.413	.010
day21	Within Groups	16.552	20	.828		
	Total	31.162	24			
	Between Groups	25.619	4	6.405	8.821	.000
day28	Within Groups	14.521	20	.726		
	Total	40.140	24			

### 4.3.2.5. Differential leukocyte count (DLC, %)

## 4.3.2.5.1. <u>Eosinophil count (%)</u>

The eosinophil count of differential leukocyte count was decreased significantly (p<0.05) after treated with neem, bitter gourd, Ivertin® and Levavet® in group B, C, D, E, respectively. The eosinophil count of differential leukocyte count was increased significantly (p<0.05) in control group A. The results of the effects of two (2) indigenous medicinal plants and two (2) patent drugs on eosinophil count of differential leukocyte count in cattle are shown in the Table 30-31 Fig. 23 and Appendix XIV.

Table 30: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on eosinophil count (%) of differential leukocyte count in cattle.

_	_	Pre- treatment	Post-treatment				
Group	Treatment	Day 'O'	7 day	14 day	21 day	28 day	
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	
A	No Drug (Control)	11.80±0.86	12.20±0.73	13.20±0.73 <sup>a</sup>	13.80±0.58 <sup>a</sup>	14.80±0.58 <sup>a</sup>	
В	Neem leaves	13.60±0.67	13.00±0.44	11.80±0.58 <sup>ab</sup>	10.80±0.58 <sup>b</sup>	$9.80\pm0.58^{b}$	
С	Korolla fruit	13.60±0.81	12.60±0.81	11.60±0.81 <sup>ab</sup>	10.20±0.58 <sup>b</sup>	9.00±0.44 <sup>bc</sup>	
D	Ivermectin 200mg/kg pour-on	13.60±0.81	12.60±0.81	11.40±0.92 <sup>ab</sup>	9.80±0.80 <sup>b</sup>	7.60±0.50°	
Е	Levamisole 7.5 mg/kg (Orally)	12.40±0.50	11.40±0.50	10.20±0.48 <sup>b</sup>	9.40±0.40 <sup>b</sup>	8.40±0.40 <sup>bc</sup>	
Total		13.00±0.34	12.36±0.30	11.64±0.36	10.80±0.40	9.92±0.56	

The above values represent the mean±Standard Error (SE) of 5 cattle

The values are mean $\pm$ SE, a,b,c mean $\pm$ SE with different supercript letters in same column deffers significantly with each other (P<0.05)

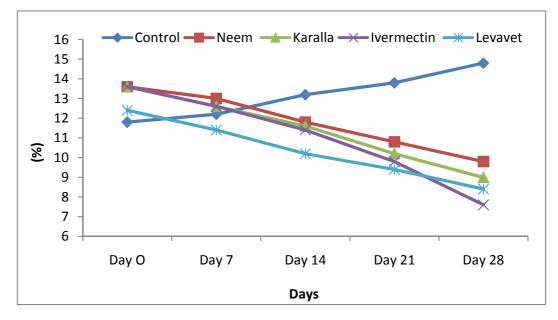


Fig. 23: Graph shows the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on eosinophil count (%) of differential leukocyte count in cattle.

Table 31: Analysis of variance for eosinophil count (%) of differential leukocyte count in cattle treated with different drugs.

Sou	rces of variation	Sum of Squares	df	Mean Square	F	Sig.
	Between Groups	14.400	4	3.600	1.295	.306
dag0	Within Groups	55.600	20	2.780		
	Total	70.000	24			
	Between Groups	7.360	4	1.840	.793	.543
dag7	Within Groups	46.400	20	2.320		
	Total	53.760	24			
	Between Groups	22.960	4	5.740	2.174	.109
dag14	Within Groups	52.800	20	2.640		
	Total	75.760	24			
	Between Groups	61.600	4	15.400	8.462	.000
dag21	Within Groups	36.400	20	1.820		
	Total	98.000	24			
	Between Groups	161.840	4	40.460	31.123	.000
dag28	Within Groups	26.000	20	1.300		
	Total	187.840	24			

### **4.3.2.5.2** Neutrophil count (%)

The results of the effect of two (2) indigenous medicinal plants and two (2) patent drugs on neutrophil count of differential leukocyte count in cattle are summarized in the Table 32-33, Fig. 24 and Appendix XV. The neutrophil count of differential leukocyte count was increased significantly (p<0.05) after treated with neem, bitter gourd, Ivertin® and Levavet® in group B, C, D, E, respectively. The neutrophil count was decreased significantly (p<0.05) in control group A.

Table 32: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on neutrophil count (%) of differential leukocyte count in cattle.

~	_	Pre- treatment		Post-treatment			
Group	Treatment	Day 'O'	7 day	14 day	21 day	28 day	
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	
A	No Drug (Control)	25.00±0.70 <sup>a</sup>	23.80±0.48 <sup>a</sup>	23.00±0.31 <sup>a</sup>	22.20±0.37 <sup>b</sup>	21.60±0.40 <sup>b</sup>	
В	Neem leaves	22.40±0.92 <sup>a</sup>	23.00±0.89 <sup>a</sup>	23.80±0.73 <sup>a</sup>	24.60±0.81 <sup>a</sup>	25.20±0.58 <sup>a</sup>	
С	Korolla fruit	22.80±1.24 <sup>a</sup>	23.20±1.06 <sup>a</sup>	24.20±1.06 <sup>a</sup>	24.80±0.96 <sup>a</sup>	25.40±0.92 <sup>a</sup>	
D	Ivermectin 200mg/kg pour-on	23.00±0.70 <sup>a</sup>	24.00±0.70 <sup>a</sup>	25.00±0.63 <sup>a</sup>	26.00±0.63ª	27.20±0.66ª	
Е	Levamisole 7.5 mg/kg (Orally)	23.00±1.00 <sup>a</sup>	23.60±0.81 <sup>a</sup>	24.40±0.81 <sup>a</sup>	25.40±0.81 <sup>a</sup>	26.00±0.70 <sup>a</sup>	
Total		23.24±0.43	23.52±0.34	24.08±0.34	24.60±0.40	25.08±0.47	

The above values represent the mean±Standard Error (SE) of 5 cattle

The values are mean $\pm$ SE, a,b mean $\pm$ SE with different supercript letters in same column deffers significantly with each other (P<0.05)

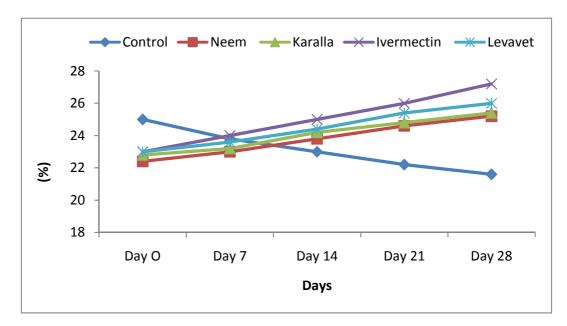


Fig. 24: Graphical presentation of the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on neutrophil count (%) of differential leukocyte count in cattle.

Table 33: Analysis of variance for neutrophil count (%) of differential leukocyte count in cattle treated with different drugs.

Sou	rces of variation	Sum of Squares	df	Mean Square	F	Sig.
	Between Groups	20.560	4	5.140	1.168	.354
dag0	Within Groups	88.000	20	4.400		
	Total	108.560	24			
	Between Groups	3.440	4	.860	.257	.902
dag7	Within Groups	66.800	20	3.340		
	Total	70.240	24			
	Between Groups	11.040	4	2.760	.972	.445
dag14	Within Groups	56.800	20	2.840		
	Total	67.840	24			
	Between Groups	42.000	4	10.500	3.750	.020
dag21	Within Groups	56.000	20	2.800		
	Total	98.000	24			
	Between Groups	87.840	4	21.960	9.548	.000
dag28	Within Groups	46.000	20	2.300		
	Total	133.840	24			

## 4.3.2.5.3 **Monocyte count (%)**

Neem, bitter gourd, Ivertin® and Levavet® were caused very little changes in monocyte count of differential leukocyte count in group B, C, D and E respectively. The results of the effects of two (2) indigenous medicinal plants and two (2) patent drugs on monocyte count of differential leukocyte count in cattle are shown in the Table 34-35, Fig. 25 and Appendix XVI. The monocyte count (%) of different leukocyte count in cattle had no significant effect (P>0.05) among the neem korolla, ivermectin and levamisole and no drug at 0, 7, 14, 21 days but had significant effect (P>0.05) at day 28 after treatment.

Table 34: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on monocyte count (%) of differential leukocyte count in cattle.

~	_	Pre- treatment	Post-treatment				
Group	Treatment	Day 'O'	7 day	14 day	21 day	28 day	
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	
A	No Drug (Control)	4.20±0.37	3.80±0.66	3.8±0.66	3.4±0.50	3.8±0.48 <sup>bc</sup>	
В	Neem leaves	4.0±0.31	3.8±0.37	4.2±0.66	4.2±0.48	5.0±0.00 <sup>a</sup>	
С	Korolla fruit	4.8±0.37	4.4±0.40	4.00±0.63	4.20±0.58	4.40±0.24 <sup>ab</sup>	
D	Ivermectin 200mg/kg pour-on	3.6±0.24	3.8±0.58	3.6±0.40	3.8±0.48	3.6±0.40 <sup>bc</sup>	
Е	Levamisole 7.5 mg/kg (Orally)	4.60±0.50	4.60±0.50	4.40±0.67	4.40±0.40	3.00±0.31°	
Total		4.24±0.18	4.08±0.22	4.00±0.26	4.00±0.22	3.96±0.20	

The above values represent the mean±Standard Error (SE) of 5 cattle

The values are mean $\pm$ SE, a,b,c mean $\pm$ SE with different supercript letters in same column deffers significantly with each other (P<0.05)

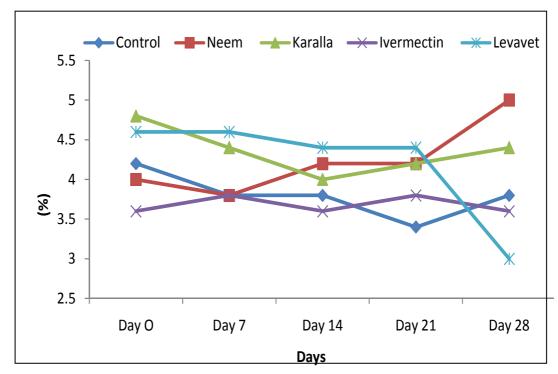


Fig. 25: Graph represents The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on monocyte count (%) of differential leukocyte count in cattle.

Table 35: Analysis of variance for monocyte count (%) of differential leukocyte count in cattle by the treatment with different drugs.

Sou	rces of variation	Sum of Squares	df	Mean Square	F	Sig.
	Between Groups	4.560	4	1.140	1.629	.206
dag0	Within Groups	14.000	20	.700		
	Total	18.560	24			
	Between Groups	3.040	4	.760	.567	.689
dag7	Within Groups	26.800	20	1.340		
	Total	29.840	24			
	Between Groups	2.000	4	.500	.263	.898
dag14	Within Groups	38.000	20	1.900		
	Total	40.000	24			
	Between Groups	3.200	4	.800	.645	.637
dag21	Within Groups	24.800	20	1.240		
	Total	28.000	24			
	Between Groups	11.760	4	2.940	5.250	.005
dag28	Within Groups	11.200	20	.560		
	Total	22.960	24			

## **4.3.2.5.4** Lymphocyte count (%)

Neem, bitter gourd, Ivertin® and Levavet® were caused very little changes in lymphocyte count of Differential Leukocyte Count in group B, C, D, and E respectively. The results of the effects of two (2) indigenous medicinal plants and. two (2) patent drugs on lymphocyte count of differential leukocyte count in cattle are summarized in the Table 36-37, Fig. 26 and Appendix XVII.

Table 36: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on lymphocyte count (%) of differential leukocyte count in cattle.

G		Pre- treatment	Post-treatment				
Group	Treatment	Day 'O'	7 day	14 day	21 day	28 day	
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	
Α	No Drug (Control)	59.40±0.40 <sup>b</sup>	60.20±0.37	60.00±0.89	59.00±0.83 <sup>b</sup>	59.80±0.73 <sup>b</sup>	
В	Neem leaves	61.20±0.80 <sup>a</sup>	60.20±0.86	60.60±0.92	61.00±0.54 <sup>ab</sup>	59.80±0.66 <sup>b</sup>	
С	Korolla fruit	58.80±0.73 <sup>b</sup>	59.40±0.67	59.80±0.66	$60.00\pm0.70^{b}$	60.00±0.89 <sup>b</sup>	
D	Ivermectin 200mg/kg pour-on	60.20±0.37 <sup>ab</sup>	60.20±0.73	61.20±0.58	62.40±0.50 <sup>a</sup>	63.40±1.07 <sup>a</sup>	
Е	Levamisole 7.5 mg/kg (Orally)	58.60±0.24 <sup>b</sup>	58.80±0.58	60.00±1.18	59.80±0.86 <sup>b</sup>	61.00±0.44 <sup>b</sup>	
Total		59.64±0.30	59.76±0.30	60.32±0.37	60.44±0.37	60.80±0.43	

The above values represent the mean±Standard Error (SE) of 5 cattle

The values are mean $\pm$ SE, a,b mean $\pm$ SE with different supercript letters in same column deffers significantly with each other (P<0.05)

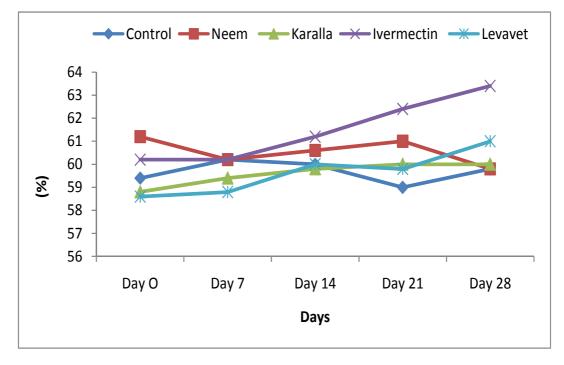


Fig. 26: Graph showing the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on lymphocyte count (%) of differential leukocyte count in cattle.

Table 37: Analysis of variance for lymphocyte count (%) of differential leukocyte count in cattle treated with different drugs.

Sou	rces of variation	Sum of Squares	df	Mean Square	F	Sig.
	Between Groups	22.960	4	5.740	3.727	.020
dag0	Within Groups	30.800	20	1.540		
	Total	53.760	24			
	Between Groups	8.160	4	2.040	.919	.472
dag7	Within Groups	44.400	20	2.220		
	Total	52.560	24			
	Between Groups	6.640	4	1.660	.432	.784
dag14	Within Groups	76.800	20	3.840		
	Total	83.440	24			
	Between Groups	34.160	4	8.540	3.416	.028
dag21	Within Groups	50.000	20	2.500		
	Total	84.160	24			
	Between Groups	47.200	4	11.800	3.758	.019
dag28	Within Groups	62.800	20	3.140		
	Total	110.000	24			

## **4.3.2.6** *Mean corpuscular volume (cubic micron)*

Neem, bitter gourd, Ivertin® and Levavet® were also caused little changes mean corpuscular volume (MCV) in group B, C, D, and E respectively. The results of the effects of two (2) indigenous medicinal plants and two (2) patent drugs on mean corpuscular volume in cattle are shown in the Table 38-39, Fig. 27 and Appendix XVIII.

Table 38: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular volume (cubic micron) in cattle.

G		Pre- treatment	Post-treatment			
Group	Treatment	Day 'O'	7 day	14 day	21 day	28 day
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)
A	No Drug (Control)	35.14±0.97 <sup>b</sup>	34.75±0.88 <sup>b</sup>	34.91±0.90	34.41±0.73 <sup>b</sup>	34.00±0.66 <sup>b</sup>
В	Neem leaves	36.35±0.16 <sup>ab</sup>	36.97±0.34 <sup>a</sup>	36.54±0.39	36.87±0.35 <sup>a</sup>	37.38±0.46 <sup>a</sup>
С	Korolla fruit	36.02±0.36 <sup>b</sup>	36.97±0.63 <sup>a</sup>	36.91±0.61	37.37±0.73 <sup>a</sup>	37.39±0.76 <sup>a</sup>
D	Ivermectin 200mg/kg pour-on	37.96±0.47 <sup>a</sup>	38.19±0.51 <sup>a</sup>	36.35±0.72	34.04±0.66 <sup>b</sup>	33.06±0.62 <sup>b</sup>
Е	Levamisole 7.5 mg/kg (Orally)	36.37±0.52 <sup>ab</sup>	36.56±0.52 <sup>a</sup>	35.28±0.29	34.10±0.22 <sup>b</sup>	33.63±0.31 <sup>b</sup>
Total		36.37±0.30	36.69±0.33	36.00±0.30	35.36±0.38	35.09±0.45

The above values represent the mean±Standard Error (SE) of 5 cattle

The values are mean $\pm$ SE, a,b mean $\pm$ SE with different supercript letters in same column deffers significantly with each other (P<0.05)

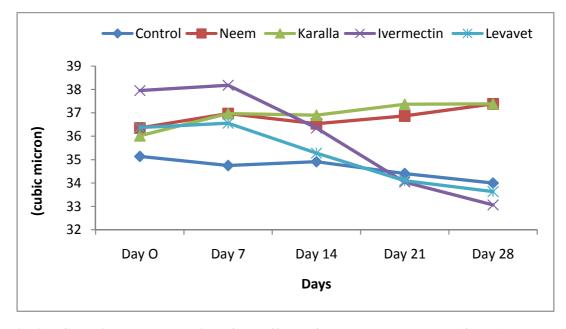


Fig. 27: Graphical representation of the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular volume (cubic micron) in cattle.

Table 39: Analysis of variance for mean corpuscular volume (cubic micron) in cattle by different drugs.

Sou	rces of variation	Sum of Squares	df	Mean Square	F	Sig.
	Between Groups	20.762	4	5.191	3.279	.032
day0	Within Groups	31.661	20	1.583		
	Total	52.424	24			
	Between Groups	30.852	4	7.713	4.271	.012
day7	Within Groups	36.116	20	1.806		
	Total	66.969	24			
	Between Groups	14.663	4	3.666	1.888	.152
day14	Within Groups	38.830	20	1.941		
	Total	53.493	24			
	Between Groups	52.801	4	13.200	7.927	.001
day21	Within Groups	33.306	20	1.665		
	Total	86.107	24			
	Between Groups	89.774	4	22.444	13.180	.000
day28	Within Groups	34.056	20	1.703		
	Total	123.830	24			

#### **4.3.2.7** *Mean corpuscular haemoglobin concentration* (%)

The mean corpuscular haemoglobin concentration (MCHC) was increased significantly (p<0.05) after neem, bitter gourd, Ivertin® and Levavet® treatment in group B, C, D, E respectively. The results of the effects of two (2) indigenous medicinal plants and two (2) patent drugs on mean corpuscular hemoglobin concentration in cattle are shown in the Table 40-41, Fig. 28 and Appendix XIX.

Table 40: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular haemoglobin concentration (%) in cattle.

		Pre- treatment		Post-treatment				
Group	Treatment	Day 'O'	7 day	14 day	21 day	28 day		
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)		
A	No Drug (Control)	32.70±1.07 <sup>a</sup>	31.11±1.23	29.65±1.22	27.83±0.84°	27.21±0.34°		
В	Neem leaves	28.34±0.54 <sup>b</sup>	29.36±0.71	31.64±0.45	33.07±0.22 <sup>ab</sup>	35.73±0.27 <sup>a</sup>		
С	Korolla fruit	29.17±0.93 <sup>b</sup>	29.53±0.91	30.52±0.76	31.71±0.77 <sup>b</sup>	33.15±0.67 <sup>b</sup>		
D	Ivermectin 200mg/kg pour-on	28.81±0.73 <sup>b</sup>	30.20±0.86	31.91±0.51	34.07±0.54 <sup>a</sup>	36.39±0.78 <sup>a</sup>		
Е	Levamisole 7.5 mg/kg (Orally)	30.09±0.80 <sup>b</sup>	30.51±0.79	31.83±0.68	33.63±0.52 <sup>a</sup>	35.09±0.34 <sup>a</sup>		
Total		29.82±0.46	30.14±0.40	31.11±0.36	32.06±0.53	33.52±0.71		

The above values represent the mean±Standard Error (SE) of 5 cattle

The values are mean $\pm$ SE, a,b,c mean $\pm$ SE with different supercript letters in same column deffers significantly with each other (P<0.05)

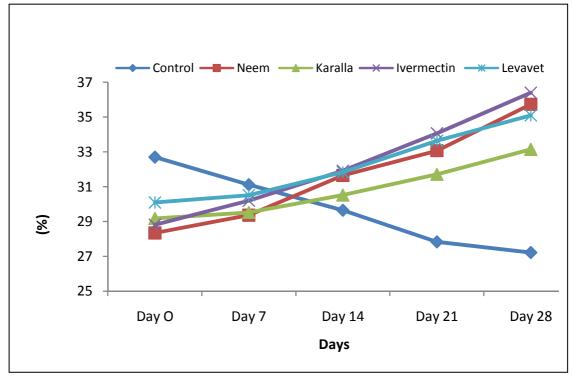


Fig. 28: Graphical presentation of the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular haemoglobin concentration (%) in cattle.

Table 41: Analysis of variance for mean corpuscular haemoglobin concentration (%) in cattle treated with different drugs.

	Sources of variance	Sum of Squares	df	Mean Square	F	Sig.
day0	Between Groups	59.925	4	14.981	4.312	.011
	Within Groups	69.492	20	3.475		
	Total	129.418	24			
day7	Between Groups	10.348	4	2.587	.614	.658
	Within Groups	84.314	20	4.216		
	Total	94.661	24			
day14	Between Groups	19.678	4	4.920	1.643	.203
	Within Groups	59.893	20	2.995		
	Total	79.572	24			
day21	Between Groups	127.450	4	31.863	16.762	.000
	Within Groups	38.017	20	1.901		
	Total	165.467	24			
day28	Between Groups	277.551	4	69.388	50.438	.000
	Within Groups	27.514	20	1.376		
	Total	305.065	24			

#### 4.3.2.8 Mean corpuscular haemoglobin (micro micro gram)

The mean corpuscular haemoglobin (MCH) was increased significantly (p<0.05) after neem, bitter gourd, Ivermectin (Ivertin®) and Levamisole (Levavet®) treatment in group B, C,D, E, respectively. The mean rpuscular haemoglobin were decreased significantly (p<0.05) in control group A.. The results of the effect of two (2) indigenous medicinal plants and two (2) patent drugs on corpuscular haemoglobin in cattle are shown in the Table 42-43, Fig. 29 and Appendix XX.

Table 42: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular hemoglobin (micro micro gram) in cattle.

~		Pre- treatment		Post-treatment				
Group	Treatment	Day 'O'	7 day	14 day	21 day	28 day		
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)		
A	No Drug (Control)	11.46±0.26 <sup>a</sup>	10.77±0.23	10.31±0.22 <sup>b</sup>	9.56±0.17°	9.24±0.07°		
В	Neem leaves	10.30±0.20 <sup>b</sup>	10.85±0.25	11.56±0.22 <sup>a</sup>	12.19±0.10 <sup>a</sup>	13.36±0.24 <sup>a</sup>		
С	Korolla fruit	10.50±0.29 <sup>b</sup>	10.90±0.21	11.26±0.29 <sup>a</sup>	11.85±0.30 <sup>ab</sup>	12.39±0.30 <sup>b</sup>		
D	Ivermectin 200mg/kg pour-on	10.94±0.37 <sup>ab</sup>	11.54±0.41	11.61±0.32 <sup>a</sup>	11.59±0.14 <sup>b</sup>	12.02±0.13 <sup>b</sup>		
Е	Levamisole 7.5 mg/kg (Orally)	10.94±0.30 <sup>ab</sup>	11.14±0.25	11.23±0.22 <sup>a</sup>	11.47±0.19 <sup>b</sup>	11.80±0.15 <sup>b</sup>		
Total		10.83±0.14	11.04±0.13	11.19±0.14	11.33±0.20	11.76±0.29		

The above values represent the mean±Standard Error (SE) of 5 cattle

The values are mean  $\pm$  SE, a,b,c mean  $\pm$  SE with different supercript letters in same column deffers significantly with each other (P<0.05)

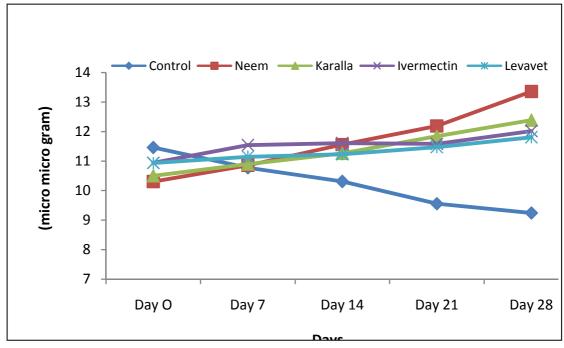


Fig. 29: Graph shows the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular hemoglobin (micro micro gram) in cattle.

Table 43: Analysis of variance for mean corpuscular hemoglobin (micro micro gram) in cattle by the treatment with different drugs.

	Source of variation	Sum of Squares	df	Mean Square	F	Sig.
day0	Between Groups	4.036	4	1.009	2.414	.083
	Within Groups	8.358	20	.418		
	Total	12.394	24			
day7	Between Groups	1.922	4	.480	1.231	.330
	Within Groups	7.808	20	.390		
	Total	9.730	24			
day14	Between Groups	5.454	4	1.364	4.041	.015
	Within Groups	6.748	20	.337		
	Total	12.202	24			
day21	Between Groups	21.155	4	5.289	28.126	.000
	Within Groups	3.761	20	.188		
	Total	24.916	24			
day28	Between Groups	46.728	4	11.682	61.033	.000
	Within Groups	3.828	20	.191		
	Total	50.557	24			

## 4.3.3. Effects of two (2) indigenous medicinal plants and two (2) patent drugs on some serum biochemical parameters in cattle

The results of the effects of two (2) indigenous medicinal plants and two (2) patent drugs on biochemical parameters of serum [alanine aminotransferase (ALT) and aspartate aminotransferase (AST) level] are shown in the Table 44-49, Fig. 30-32 and Appendix XXI-XXII.

## **4.3.3.1** Alanine aminotransferase level of serum (U/I)

The alanine aminotransferase (ALT) level of serum was not significantly changed) after treated with neem, bitter gourd, Ivermectin (Ivertin®) and Levamisole (Levavet®) in group B, C, D, E, respectively. The results of the effects of two (2) indigenous medicinal plants and two (2) patent drugs on alanine aminotransferase level of serum in cattle are shown in the Table 44-45, Fig. 30 and Appendix XXI.

Table 44: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on alanine aminotransferase level of serum (U/I) in cattle.

Group		Pre- treatment	Post-treatment					
	Treatment	Day 'O'	7 day	14 day	21 day	28 day		
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)		
A	No Drug (Control)	18.26±0.99	18.40±0.75	18.14±1.14	18.00±1.22	19.24±1.00		
В	Neem leaves	20.22±0.74	20.40± 1.16	19.58±0.67	20.32±1.15	19.22±0.94		
С	Korolla fruit	19.90±1.29	19.26±1.14	19.82±1.15	19.52±1.00	19.70±0.82		
D	Ivermectin 200mg/kg pour-on	20.14±0.56	19.48±1.10	19.40±0.80	19.56±0.77	19.48±0.92		
Е	Levamisole 7.5 mg/kg (Orally)	18.52±0.56	18.22±0.86	18.32±0.81	18.06±0.75	17.78±0.90		
Total		19.41±0.40	19.15±0.45	19.05±0.41	19.09±0.45	95.42±4.58		

The above values represent the mean±Standard Error (SE) of 5 cattle

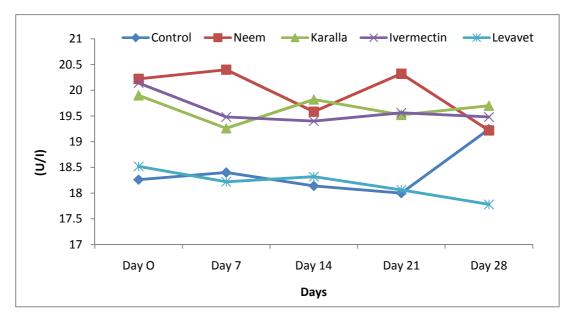


Fig. 30: Graphical representation of the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on alanine aminotransferase level of serum (U/I) in cattle.

Table 45: Analysis of variance for alanine aminotransferase level of serum (U/I) in cattle in relation with different drugs.

	-	Sum of Squares	df	Mean Square	F	Sig.
day0	Between Groups	17.718	4	4.430	1.153	.361
	Within Groups	76.860	20	3.843		
	Total	94.578	24			
day7	Between Groups	15.554	4	3.889	.751	.569
	Within Groups	103.568	20	5.178		
	Total	119.122	24			
day14	Between Groups	11.808	4	2.952	.676	.616
	Within Groups	87.305	20	4.365		
	Total	99.113	24			
day21	Between Groups	20.838	4	5.210	1.045	.409
	Within Groups	99.740	20	4.987		
	Total	120.578	24			
day28	Between Groups	11.398	4	2.849	.674	.618
	Within Groups	84.536	20	4.227		
	Total	95.934	24			

### 4.3.3.2 Aspartate aminotransferase level of serum (U/I)

The aspartate aminotransferase (AST) level of serum was not significantly changed) after treated with neem, bitter gourd, Ivermectin (Ivertin®) and Levamisole (Levavet®) in group B, C, D, E respectively. The results of the effects of two (2) indigenous medicinal plants and two (2) patent drugs on aspartate aminotransferase level of serum in cattle are shown in the Table 46-47, Fig. 31 and Appendix XXII.

Table 46: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on aspartate aminotransferase level of serum (U/I) in cattle.

Group		Pre- treatment	Post-treatment				
	Treatment	Day 'O'	7 day	14 day	21 day	28 day	
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	
A	No Drug (Control)	30.28±1.07	29.94±1.13	31.10±0.91	29.36±1.12	31.58±0.76	
В	Neem leaves	32.84±1.05	31.26±0.93	31.88±0.99	32.72±1.46	31.96±0.85	
С	Korolla fruit	31.74±1.30	33.14±1.10	32.46±0.67	31.02±1.25	31.82±0.81	
D	Ivermectin 200mg/kg pour-on	32.22±0.51	31.72±0.95	32.36±0.78	31.16±0.77	31.36±0.73	
Е	Levamisole 7.5 mg/kg (Orally)	30.50±0.68	31.94±1.01	31.60±0.64	29.80±1.29	30.06±0.88	
Total		31.52±0.44	31.60±0.47	31.88±0.35	30.81±0.55	31.36±0.36	

The above values represent the mean±Standard Error (SE) of 5 cattle

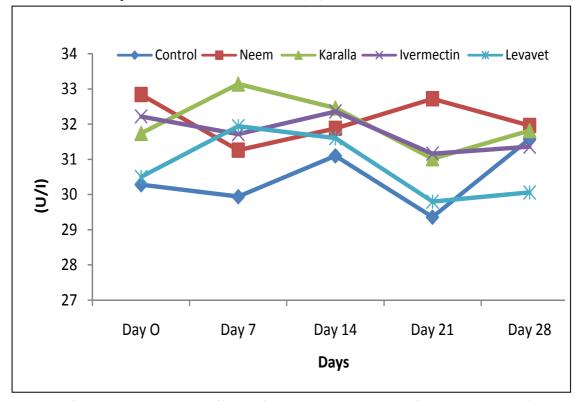


Fig. 31: Graph represents the effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on aspartate aminotransferase level of serum (U/I) in cattle.

Table 47: Analysis of variance for aspartate aminotransferase level of serum (U/I) in cattle in relation with different drugs

	_	Sum of				
		Squares	df	Mean Square	F	Sig.
day0	Between Groups	24.294	4	6.073	1.307	.301
	Within Groups	92.920	20	4.646		
	Total	117.214	24			
day7	Between Groups	26.864	4	6.716	1.275	.313
	Within Groups	105.356	20	5.268		
	Total	132.220	24			
day14	Between Groups	6.268	4	1.567	.481	.749
	Within Groups	65.152	20	3.258		
	Total	71.420	24			
day21	Between Groups	34.686	4	8.672	1.204	.340
	Within Groups	144.100	20	7.205		
	Total	178.786	24			
day28	Between Groups	11.550	4	2.887	.886	.490
	Within Groups	65.192	20	3.260		
	Total	76.742	24			

## 4.3.4. Effects of two (2) indigenous medicinal plants and two (2) patent drugs on body weight (Kg) in cattle

The body weight was increased significantly (p<0.05) after treated with neem, bitter gourd, Ivermectin (Ivertin®) and Levamisole (Levavet®) in group B, C, D, E, respectively. The results of the effects of two (2) indigenous medicinal plants and two (2) patent drugs on body weight in cattle are shown in the Table 48-49, Fig. 32 and Appendix XXIII.

Table 48: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on body weight (Kg) in cattle.

	<b>T</b>	Pre- treatment	Post-treatment				
Group	Treatment	Day 'O'	7 day	14 day	21 day	28 day	
		(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	(Mean±SE)	
A	No Drug (Control)	125.40±1.46	125.00±1.38	124.20±1.45	123.60±1.42 <sup>b</sup>	122.90±1.52 <sup>b</sup>	
В	Neem leaves	124.90±2.05	125.40±2.05	126.00±2.06	126.90±2.00 <sup>ab</sup>	127.80±1.89 <sup>a</sup>	
С	Korolla fruit	124.50±1.10	124.70±1.06	125.50±0.89	126.70±0.94 <sup>ab</sup>	127.60±0.87 <sup>a</sup>	
D	Ivermectin 200mg/kg pouron	126.80±1.12	127.30±1.12	128.20±1.07	129.10±1.06 <sup>a</sup>	130.40±0.90 <sup>a</sup>	
Е	Levamisole 7.5 mg/kg (Orally)	123.90±1.93	124.50±2.02	125.50±1.93	126.40±1.85 <sup>ab</sup>	127.40±1.81 <sup>a</sup>	
Total		125.10±0.67	125.38±0.68	125.88±0.68	126.54±0.71	127.22±0.77	

The above values represent the mean±Standard Error (SE) of 5 cattle

The values are mean $\pm$ SE, a,b mean $\pm$ SE with different supercript letters in same column deffers significantly with each other (P<0.05)

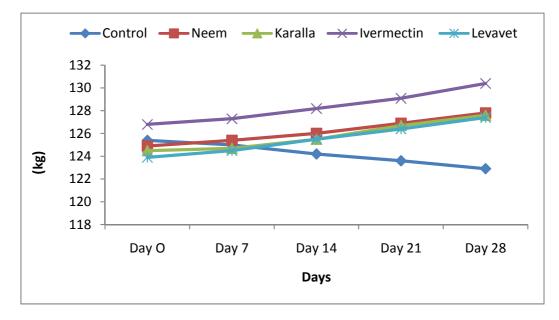


Fig. 32: Graph showing the effect of medicinal plants on body weight (kg) in cattle

Table 49: Analysis of variance for body weight (Kg) in cattle in relation with different drugs in different days of treatment.

	-	Sum of Squares	df	Mean Square	F	Sig.
day0	Between Groups	24.100	4	6.025	.477	.752
	Within Groups	252.400	20	12.620		
	Total	276.500	24	Į.		
day7	Between Groups	25.340	4	6.335	.500	.736
	Within Groups	253.300	20	12.665		
	Total	278.640	24			
day14	Between Groups	42.540	4	10.635	.879	.494
	Within Groups	242.100	20	12.105		
	Total	284.640	24			
day21	Between Groups	76.860	4	19.215	1.670	.196
	Within Groups	230.100	20	11.505		
	Total	306.960	24			
day28	Between Groups	146.440	4	36.610	3.388	.029
	Within Groups	216.100	20	10.805		
	Total	362.540	24			

#### **CHAPTER-5**

## **DISCUSSION**

Worldwidely, parasitism is an important factor that is responsible for the reducing the health and productivity of cattle (Hassan *et al.* 2004; Perry *et al.* 2002, Perry and Randolph, 1999). Many authors Qadir, 1981, Rahman 1983 studied that *Haemonchus contortus*, *Trichostrongylus* sp, *Cooperia* sp, *Oesphagostomum* sp, *Trichuris* sp, *Strongyloides* sp are most common among gastro-intestinal parasites in Bangladesh. More than 500 plants have medicinal importance in livestock and human of Bangladesh (Yusuf *et al.* 1994). Many authors have been studied the medicinal plants act on gastro-intestinal parasites. The present study have been found as new findings such which factors are very responsible for the gastro-intestinal parasites burden and identify the best medicinal plants as well as patent drugs. Under this discussion the present findings are compare with another authors broad and abroadly and interpretation the results.

#### 5.1 Prevalence of Gastro-intestinal Nematodiasis in Cattle

The prevalence of *Ascaris* sp., *Strongyles* (*Haemonchus* sp., *Trichostrongylus* sp., *Oesophagostomum* sp. and *Mecistocirrus* sp.), *Bunostomum* sp., *Trichuris* sp., *Strongyloides* sp. and *mixed infection* were 6.5%, 25.5%, 2.25%, 5% 5% and 10.5%, respectively. This result was in conformity with earlier reports made by Bhattacharyya and Ahmed (2005) who examined 546 faecal samples collected from cattle in India from March, 2002 to February, 2003. The most dominant species were: *Ascaris* sp. (39, 7.14%), *Strongyloides* (32, 5.86%), *Trichuris* spp. (15, 2.75%), *Strongylus* sp. (13, 2.3 8%), *Haemonchus* spp. (11, 2.0 1%) and *Bunostomum* spp. (10, 1.83%). Islam (2003) reported the prevalence

Chapter 5 Discussion 🗷 225

of gastro-intestinal nematodes in buffalo was Strongyloides spp. (9.6%), Haemonchus (21.15%),*Trichostrongylus* spp. (7.6%),spp. Oesophagostomum spp. (9.6%) and mixed infection (25%). This result was in conformity with earlier reports made by Islam (2012) in cattle and Biswas (2012) in buffaloes, Nuruzzaman (2010) in goat, Sangma (2010) in sheep, Hossain (2010) ) in cattle, Amin et al. (2009) in cattle, Anwar (2008) in cattle, Gadre (2007) in cattle, Hirani et al. (2006a,2006b) in cattle, Repossi-Junior et al. (2006) in cattle, Morgan et al. (2006) in ruminant, Kaewthamasorn and Worangsamee (2006) in cattle, Murphy et al. (2006) in cattle, Mbae et al. (2004) in cattle, Keyvu et al. (2005) in cattle, Achi et al. (2003) in cattle, Soca et al. (2003) in cattle, Yildirim et al. (2000) in cattle.

The incidence of gastro-intestinal nematodes was highest in 12 to <24 months old cattle followed by 6 to <12 months old cattle, >24 months old cattle and 1to < 6 months old cattle. In the age of 1to < 6 months the prevalence of gastro-intestinal nematodes was 36.7% (Ascaris sp. and Strongyles were 33.3% and 3.3% respectively), in the age of 6 to <12 months, the prevalence of gastro-intestinal nematodes was 62.5% Trichuris sp. Strongyloides sp. and mixed (Ascaris sp Strongyles infection were 15.0%, 25.0%, 5.0%, 5.0% and 12.5% respectively), in the age of 12 to <24 months, the prevalence of gastro-intestinal nematodes was 67.0% (Strongyles sp., Bunostomum sp., Trichuris sp., Strongyloides sp. and *mixed infection* were 35.0%, 4.0%, 7.5%, 7.0% and 13.5% respectively), in the age of >24 months, the prevalence of gastrointestinal nematodes was 38.0% (Strongyles sp., Bunostomum sp., Trichuris sp., Strongyloides sp. and mixed infection were 20.0%, 1.0%, 3.0%, 4.0% and 10.0% respectively). Similarly Hossain (2010) studied Chapter 5 Discussion 🗷 226

nematodes infection in cattle in Bangladesh Agricultural University dairy farm from November 2009 to May 2010 by fecal sample examination. In the age groups, calves (33.33%) were mostly susceptible to nematode infection followed by young (30%) and adult (12.5%). Keyyu *et al.* (2003) determined the prevalence of gastro-intestinal nematode infections in indigenous Zebu cattle in the lower plain (lowland zone) of the southern highlands of Tanzania. The results indicated that immature cattle (<3 years) had significantly higher worm burdens than mature cattle. The present finding was also in agreement with the works of Gadre (2007) in cattle, Nuruzzaman (2010) in goat, Sangma (2010) in sheep.

The incidence of gastro-intestinal nematodes was more in female (62.0%) than male (44.2%). Hossain (2010) also studied nematodes infection in cattle in Bangladesh Agricultural University dairy farm from November 2009 to May 2010 by fecal sample examination and found the prevalence of nematode infection significantly higher in female (21.11%) than that of male (17.5%). On the other hand, Qureshi *et al.* (1997) observed that overall rate of infection was higher in male than in female buffaloes. The present finding was also in agreement with the works of Biswas (2012) in buffaloes, Islam (2012) in cattle.

This infestation rate of gastro-intestinal nematodes with their breed was higher in cross breed cattle 184 (58.4%) than in local breed cattle 35 (41.2%). The present finding was also in agreement with the works of Hossain (2010) who studied nematodes infection in cattle in Bangladesh Agricultural University dairy farm from November 2009 to May 2010 by fecal sample examination. In case of breed, cross breed (23.33%) were found more susceptible to nematode infection than that of local breed

(12.50%). Similar result also reported by Gadre (2007) in cattle, Nuruzzaman (2010) in goat

This infestation rate of gastro-intestinal parasites with their genotype was highest in L×F genotype of cattle 56 (60.9%) followed by L×F×SL genotype of cattle 46 (57.5%), L×SL genotype of cattle 82 (57.3%), and in local cattle 35 (41.2%). Due to lack of similar published literature of these findings could not be compared.

The prevalence of gastro-intestinal nematodes was 54.8% (rainy seasons-79.8%, autumn-57.7% summer-44.3% and winter seasons-34.8%). Highest incidence of gastro-intestinal nematodes was found in rainy seasons (June-August) followed by autumn (September-November) summer (March-May) and winter seasons (December-February). Lima (1998) observed tracer calves to assess the seasonality of infections of gastro-intestinal parasites in beef cattle extensively raised at a farm in the State of Minas Gerais, Brazil. Tracer calves acquired infections during all months of the year, however, highest worm burdens were observed in the rainy season. Katoch et al. (2000) observed seasonal incidence of gastrointestinal nematodes in goats. The highest egg was recorded in the rainy season and the lowest in the summer. Hossain (2010) also observed relatively higher prevalence of nematode infection in summer season (23.33%) than winter season (17.14%). Sahoo et al. (2002) recorded helminthes infection throughout the year with little seasonal variation, i.e., highest during the rainy (51.8%) followed by the winter (50.7%) and summer (47.6%) seasons. Waruiru et al (2001) observed the epidemiology of gastro-intestinal nematodes of dairy cattle in Central Kenya. The total worm burden in the animals was highest during the

rainy season (March-June and October-December) and lowest during the dry seasons (July-September and January-February).

Whereas Hirani *et al.* (2006a) observed prevalence of gastro-intestinal parasitism was highest during summer (51.83%) followed by monsoon (45.22%) and the lowest in winter (35.97%). Similar result also reported by Biswas (2012) in buffaloes, Hossain (2010) in cattle, Keyyu *et al.* (2003).

The prevalence of gastro-intestinal nematodiasis in cattle were 33.3%, 34.4%, 37.5%, 42.9%, 53.3%, 77.1%, 88.2%, 74.3%, 67.6%, 60.0%, 45.7% and 36.4% in January, February, March, April, May, June, July, August, September, October, November and December, respectively. The incidence of gastro-intestinal nematodes was highest in the month of June (77.1%) and July (88.2%). Whereas Hirani *et al.* (2006a) observed highest prevalence during September and the least in December and Ahmad and Ansari (1987) examined that the prevalence of *H. contortus* was highest in June to November and peaked in October at 92.0%. The prevalence of *B. trigonocephalum* peaked in March and April at 50.0-54.8%. Similar result also reported by Koroglu *et al.* (2001), Barry *et al.* (2002), Achi *et al.* (2003), Crespo *et al.* (2002).

The incidence of gastro-intestinal nematodes was highest in Rajshahi district (62.5) followed by Natore district (56.2) Naogaon district (42.9) and Chapai Nawabgonj district (38.0). Due to lack of similar published literature of these findings could not be compared.

## 5.2 Screening of Plants for in vitro Anthelmintic Activity

Water extracts of 10 indigenous medicinal plants (Neem, custard apple, papaya, garlic, bitter gourd, jute, chaste tree, pineapple, pomegranate and betel leaf) showed potential *in vitro* activities against adult parasites. The

Chapter 5 Discussion \( \notin \) 229

efficacy of water extract of these plants at the concentrations of 25 mg/ml and 50 mg/ml was much lower than that of concentration of 100 mg/ml. Custard apple, papaya, jute, chaste tree, pomegranate, (50 mg/ml) showed 30-40% and neem, garlic, bitter gourd, pineapple, betel leaf (50 mg/ml) showed 50-60% *in vitro* efficacy against gastro-intestinal nematodes of cattle.

In water extract, the plants (100 mg/ml), had highly significant activity (100%) against adult gastro-intestinal nematodes *in vitro* were: Neem (leaves), bitter gourd (fruit and seeds) and betel leaf (leaves), moderately significant activity (90%) against adult gastro-intestinal nematodes *in vitro* were: garlic (bulbs) and pineapple (leaves) and less activity (60-70%) against adult gastro-intestinal nematodes *in vitro* were: custard apple (leaves), papaya (leaves), jute (leaves), chaste tree (leaves) and pomegranate (leaves).

Ethanol extracts of 10 indigenous medicinal plants (Neem, custard apple, papaya, garlic, bitter gourd, jute, chaste tree, pineapple, pomegranate, betel leaf) showed much potential *in vitro* activities against adult parasites than that of the water extract. The efficacy of ethanol extract of these plants at the concentrations of 25 mg/ml was lower than that of concentration of 50 mg/ml and 100mg/ml. Custard apple, papaya, garlic, jute, chaste tree, pineapple and pomegranate, (25 mg/ml) showed 30-40% and neem, bitter gourd and betel leaf (25 mg/ml) showed 50% *in vitro* efficacy against gastro-intestinal nematodes of cattle. Custard apple, papaya, jute, chaste tree, pomegranate, (50 mg/ml) showed 60-70% and neem, garlic, bitter gourd, pineapple, betel leaf (50 mg/ml) showed 90-100% *in vitro* efficacy against gastro-intestinal nematodes of cattle.

Chapter 5 Discussion \( \noting 230 \)

The plants (100 mg/ml), had highly significant activity (100%) against adult gastro-intestinal nematodes *in vitro* were: Neem (leaves) garlic (bulbs), bitter gourd (fruit and seeds), pineapple (leaves) and) betel leaf (leaves). Custard apple, papaya, jute, chaste tree, pomegranate, (100 mg/ml) showed 80-90% *in vitro* efficacy against gastro-intestinal nematodes of cattle.

In vitro anthelmintic effects of several plants were also demonstrated against gastro-intestinal nematodes of animals in various parts of the world Iqbal et al. (2004 and 2006); Lateef et al. (2003); Rahman (2002); Shilaskar and Parashar (1989), Sujon et al. (2008), Mian (2011), Amin et al. (2008), Hoque et al. (2006), Mostofa and Amin (2005), Brahmachari (2004), Begum et al. (2010), Amin et al. (2009), Amin et al. (2010). The results presented alone were in agreement with those studies.

# 5.3 Efficacy of Two Indigenous Medicinal Plants and Two Patent Drugs against Natural Gastro-intestinal Nematodes in Cattle

Neem and bitter gourd and two patent drugs i.e. Ivertin® and Levavet®, significantly (p<0.05) reduced of EPG count in cattle of group B, C, D,E, respectively.

In group A: The mean EPG of control group on pre-treatment (day 0) was 660±47.33. The mean EPG on the 7<sup>th</sup>, 14<sup>th</sup> 21<sup>st</sup> and 28<sup>th</sup> day were 710±36.74, 796±42.61, 910±43.01and 1050±52.44, respectively in cattle. The mean EPG were increased 7.58%, 20.61%, 37.88% and 59.09% on 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>th</sup> and 28<sup>th</sup> day, respectively.

**In group B**: Mean EPG count before treatment was  $870\pm43.59$  and after treatment with neem mean EPG on  $7^{th}$ ,  $14^{th}$   $21^{st}$  and  $28^{th}$  day were

 $410\pm50.99$ ,  $330\pm51.48$ ,  $444\pm50.36$  and  $562\pm33.97$  respectively in cattle. Reduction of mean EPG on  $7^{th}$ ,  $14^{th}$   $21^{st}$  and  $28^{th}$  day after treatment were 52.87%, 62.07%, 48.97% and 35.40%, respectively.

In conformity to the present findings, Khalid *et al.* (2005) reported that neem (10% water extract of leaves) reduced significantly (p<0.01) EPG count 47.03%, 46.27%, 41.82% and 37.60% on 7th 14th 21st and 28th day, respectively in sheep. Rob *et al.* (2004) observed that water extracts of neem was 53.72% effective against haemonchosis in sheep. Brelin (2002) found that fresh neem leaves significantly reduced H. contortus in the abomasum of the treated sheep. Arunachal *et al.* (2002) noted that neem leaves, seeds and bark were 53%, 49% and 38% infective against gastro-intestinal helminths in sheep, respectively. Rahman (2002) found the effects of water extract of neem leaves was 62% in goat.

**In group C:** Mean EPG count before treatment was 860±64.03and after treatment with Karalla (bitter gourd) mean EPG on 7<sup>th</sup>, 14<sup>th</sup> 21<sup>st</sup> and 28<sup>th</sup> day were 490±57.88, 428±50.34, 560±65.50 and 714±66.15 respectively in cattle. Reduction of mean EPG on 7<sup>th</sup>, 14<sup>th</sup> 21<sup>st</sup> and 28<sup>th</sup> day after treatment were 43.02%, 50.23%, 34.88% and 16.98%, respectively.

In conformity to the present findings, Rahman (2002) reported that Karalla (bitter gourd) 100% water extract of fruits reduced significantly (p<0.01) EPG count 27%, 52%, 67% and 65% on 7th 14th 21st and 28th day, respectively and 50% ethanol extract reduced significantly (p<0.01) EPG count 35%, 65%, 79% and 41% on 7th 14th 21st and 28th day, respectively

Chapter 5 Discussion \( \noting 232 \)

In group **D**: Mean EPG count before treatment was  $730\pm47.85$  and after treatment with Ivertin® mean EPG on 7th 14th 21st and 28th day were  $0\pm0.00$ ,  $0\pm0.00$ ,  $0\pm0.00$  and  $0\pm0.00$ , respectively in cattle. Reduction of mean EPG on 7th 14th 21st and 28th day after treatment were 100%, 100%, 100% and 100%, respectively.

In conformity to the present findings, Abdalla (1989) reported 100% efficacy of ivermectin be faecal egg count, against gastro-intestinal nematodes (*Haemonchus SL Oesophagostomum V, Trichostrongylus W, strongyloides, Oestrus ovis Monizia, cooperia, chaberitain*) in sheep and cattle during the study of the effect of end parasites on growth rate. Tada *et al.* (1992) reported the 100% efficacy of ivermectin against gastro-intestinal nematodes in sheep and goat of Zambia. Fourteen goats and 5 ewes were treated with subcutaneous injection of ivermectin at a dose rate of 200 tg/kg body weight. The animals were naturally infected with. *Haemonchus contortur, Trichosrrongvlus % Oesopha Qostomwn* 5,P and Strongvloides V. The efficacy of ivermectin was 100% during faccal oval count on 7th and 14th day after treatment.

In group E: Mean EPG count before treatment was 662±33.38 and after treatment with Levavet® mean EPG on 7th 14th 21st and 28th day were 0±0.00, 34±18.87, 86±30.76 and 142±31.21, respectively in cattle. Reduction of mean EPG on 7th 14th 21st and 28th day after treatment were 100%, 94.86%, 87.01% and 78.55%, respectively.

In conformity to the present findings, Iqbal *et al.* (2006) observed levamisole (7.5 mg/kg) showed a 99.6% reduction in EPG against gastro-intestinal nematodes of sheep. In conformity to the present findings, Iqbal *et al.* (2006), Iqbal *et al.* (2004), Lateef *et al.* (2003), Keyyu *et al.* (2002),

Chapter 5 Discussion \( \otimes \) 233

Waruiru *et al.* (1998), Vazquez *et al.* (1984), Leguia *et al.* (1979) and Beck *et al.* (1971) reported similar findings in sheep. Gawor and Borecka (1999) observed levamisoic on the 4th and 28 days after dosing was 98.3% and 76.3%, respectively against gastro-intestinal nematodes in goats. Likewise, Mattos *et al.* (2000), Charles *et al.* (1989) and Guha and Banerjee (1987) observed similar results in goat. The present finding was also in agreement with the works of Shri-Kishan and Gupta (2004), Khan *et al.* (2003), Williams and Broussared (1995) and Beck *et al* (1971) in cattle. Similar results also stated by some researchers Olaho-Mulani and Kimani (1999) in camel, Qureshi *et al.* (1997) in buffalo, Quiroz-Romero. *et al.* (1992) in calves and Narasimhan (1988) in buffalo, Mostofa and Amin (2005), Sujon *et al.* (2008) Amin *et al.* (2008), Rahman *et al.* (2008).

# 5.4 Effects of Two Indigenous Medicinal Plants and Two Patent Drugs on Haematological Parameters in Cattle

#### 5.4.1 Total erythrocyte count (TEC, million/cu.mm.)

Two indigenous medicinal plants, i.e, Neem and bitter gourd and two patent drugs, i.e. Ivertin® and Levavet®, significantly (p<0.05) increased total erythrocyte count (TEC) in cattle of group, B, C, D, E, respectively. Mean TEC before treatment was 8.31±0.23 and after treatment with neem mean TEC on 7th 14th 21st and 28th day were 8.37±0.23, 8.66±0.20, 8.92±0.21 and 9.04±0.19 respectively in group B. Mean TEC before treatment was 8.62±0.35 and after treatment with Karalla (bitter gourd) mean TEC on 7th 14th 21st and 28th day were 8.66±0.37, 8.82±0.35, 8.99±0.35 and 9.09±0.35 respectively in group C. Mean TEC before treatment was 8.33±0.06 and after treatment with Ivertin® mean TEC on 7th 14th 21st and 28th day were 8.45±0.05, 9.23±0.06, 10.11±0.05 and 10.61±0.04 respectively in group D. Mean TEC before treatment was

 $8.88\pm0.23$  and after treatment with Levavet® mean TEC on 7th 14th 21st and 28th day were  $9.00\pm0.22$ ,  $9.65\pm0.17$ ,  $10.18\pm0.06$  and  $10.50\pm0.05$  respectively in group E. The mean TEC of control group- A on pretreatment (day 0) was  $9.11\pm0.24$ . Mean TEC on the 7th 14th 21st and 28th day were  $9.00\pm0.22$ ,  $8.82\pm0.23$ ,  $8.66\pm0.21$  and  $8.44\pm2.20$  respectively in group A.

Khalid *et al.* (2005) reported that neem and pineapple (10% water extract of leaves) increased TEC in sheep. Likewise, Amin *et al.* (2008), Sujon *et al.* (2008), Shahadat *et al.* (2008), Amin *et al.* (2010), Begum *et al.* (2010), Rahman (2002) observed water extract of neem and Karalla increased TEC on 21<sup>st</sup> day of post-treatment in goat. Rob *et al.* (2004) also stated water extracts of neem leaves increased Hb content in sheep on 28 day post-treatment. Similar response also reported by Khalid *et al.* (2005) due to Vermic in sheep. So the result of the present study similar to the reports of Amin *et al.* (2005) and Rob *et al.* (2004) in sheep. Khan *et al.* (2003) also observed levamisole increased significantly TEC in cattle. The improved level of TEC of blood in treated cattle might be due to elimination of blood sucking gastro-intestinal nematodes.

## 5.4.2 Haemoglobin content (Hb, g %).

Two indigenous medicinal plants, i.e, Neem and bitter gourd and two patent drugs, i.e. Ivertin® and Levavet®, significantly (p<0.05) increased haemoglobin (Hb) content in cattle of group in cattle of group, B, C, D, E, respectively. Mean Hb content before treatment was 8.58±0.37and. after treatment with neem mean Hb content on 7th 14th 21st and 28th day were 9.00±0.38 9.90±0.36, 10.70±0.24 and 11.48±0.21, respectively in group B. Mean Hb content before treatment was 9.08±0.57 and. after treatment with Karalla (bitter gourd) mean Hb content on 7th 14th 21st

and 28th day were 9.36±0.52, 9.82±0.47, 10.40±0.43 and 11.06±0.36, respectively in group C. Mean Hb content before treatment was 9.12±0.32 and. after treatment with Ivertin® mean Hb content on 7th 14th 21st and 28th day were 9.76±0.38, 10.72±0.32, 11.72±0.16 and 12.76±0.17, respectively in group D. Mean Hb content before treatment was 9.72±0.41 and. after treatment with Levavet® mean Hb content on 7th 14th 21st and 28th day were 10.04±0.38, 10.84±0.30, 11.68±0.23 and 12.40±0.17, respectively in group E. The mean Hb content of control group-A on pre-treatment (day 0) was 10.44±0.35. Mean Hb content on the 7th 14th 21st and 28th day were 9.72±0.41, 9.12±0.41, 8.28±0.28 and 7.80±0.16 respectively in group A.

Rahman (2002) reported that water extract of neem and Karalla increased haemoglobin (Hb) content on 21<sup>st</sup> day of post-treatment in goat. Amin *et al* (2010), Begum *et al*. (2010), Amin *et al* (2008), Khalid *et al*. (2005) also observed that neem and pineapple (10% water extract of leaves) increased Hb content in sheep. Similarly, Rob *et al*. (2004) noted that water extracts of neem leaves increased Hb content in sheep on 28 day post-treatment. The present finding was also in agreement with the work of neem leaves (@ 100mg/kg) by Hossain *et al*. (1996) in cattle. Similar responses reported by Khalid *et al*. (2005) due to Vermic® in sheep. Likewise, Islam *et al*. (2003) observed similar results in goat and Mukherjee (1992) in calves. Similar response also reported by Shri-Kishan and Gupta (2004) and Khan *el at*. (2003) due to levamisole in cattle. The increase in haemoglobin content in cattle might be due to the increase of total erythrocyte count (TEC).

#### **5.4.3 Packed cell volume (PCV, %)**

Two indigenous medicinal plants, i.e, Neem and bitter gourd and two patent drugs, i.e. Ivertin® and Levavet®, significantly (p<0.05) increased packed cell volume (PCV) in cattle of group, B, C, D, E, respectively. Mean PCV before treatment was 30.22±0.82 and after treatment with neem mean PCV on 7th 14th 21st and 28th day were 30.94±0.70, 31.64±0.60, 32.89±0.65 and 33.80±0.53, respectively in group B. Mean PCV before treatment was 31.01±1.05 and after treatment with Karalla (bitter gourd) mean PCV on 7th 14th 21st and 28th day were 31.74±0.87, 32.48±0.84, 33.51±0.71 and 33.91±0.68, respectively in group C. Mean PCV before treatment was 31.63±0.54and after treatment with Ivertin® mean PCV on 7th 14th 21st and 28th day were 32.30±0.63, 33.57±0.66, 34.43±0.62 and 35.10±0.58, respectively in group D. Mean PCV before treatment was 32.26±0.63 and after treatment with Levavet® mean PCV on 7th 14th 21st and 28th day were 32.87±0.47, 34.04±0.39, 34.73±0.42 and 35.34±0.42, respectively in group E. Mean PCV of control group on pre-treatment (day 0) was 31.94±0.54. Mean PCV on the 7th 14th 21st and 28th day were,  $31.24\pm0.48$ ,  $30.76\pm0.59$ ,  $29.74\pm0.38$  and  $28.67\pm0.59$ , respectively in group A.

This findings supported by the earlier reports of Khalid *et al.* (2005) and Rob *et al.* (2004) in sheep. Amin *et al* (2010), Begum *et al.* (2010), Amin *et al* (2008), Khalid *et al.* (2005) reported that neem and pineapple (10% water extract of leaves) increased PCV in sheep. Rob *et al.* (2004) also noted that water extracts of neem leaves increased PCV in sheep on 28 day post-treatment. Likewise, Rahman (2002) observed water extract of neem and Karalla increased PCV on 21<sup>st</sup> day of post-treatment in goat. Similar responses reported by Khalid *et al.* (2005) due to Vermic® in

Chapter 5 Discussion \( \otimes \) 237

sheep. Islam *et al.* (2003) also noted-similar findings in goat. Shri-Kishan and Gupta (2004) also found levarnisole increased PCV in cattle.

#### 5.4.4 Total leukocyte count (TLC, thousand/cu.mm.)

Two indigenous medicinal plants, i.e, Neem and bitter gourd and two patent drugs, i.e. Ivertin® and Levavet®, significantly (p<0.05) decreased total leukocyte count (TLC) in cattle of group, B, C, D, E, respectively. Mean TLC before treatment was 10.34±0.38 and after treatment with neem mean TLC on 7th 14th 21st and 28th day were 10.13±0.38, 9.87±0.38, 9.63±0.40 and 9.35±0.42, respectively in group B. Mean TLC before treatment was 9.99±0.59 and after treatment with Karalla (bitter gourd) mean TLC on 7th 14th 21st and 28th day were 9.78±0.58, 9.55±0.59, 9.39±0.58 and 9.18±0.54, respectively in group C. Mean TLC before treatment was 10.18±0.21 and after treatment with Ivertin® mean TLC on 7th 14th 21st and 28th day were  $9.97\pm0.21$ ,  $9.46\pm0.26$ ,  $9.21\pm0.22$ and 8.83±0.24, respectively in group D. Mean TLC before treatment was 9.20±0.41 and after treatment with Levavet® mean TLC on 7th 14th 21st and 28th day were  $9.16\pm0.27$ ,  $8.68\pm0.27$ ,  $8.19\pm0.32$  and  $8.06\pm0.23$ , respectively in group E. Mean TLC of control group on pre-treatment (day 0) was 9.30±0.52. Mean TLC on 7th 14th 21st and 28th day were  $9.75\pm0.46$ ,  $10.01\pm0.48$ ,  $10.57\pm0.40$  and  $11.13\pm0.35$ , respectively in group A.

Rahman (2002) observed water extract of neem and Karalla decreased TLC on 21st day of post-treatment in goat. The present finding was also agreement with the' work of Rob *et al* (2004) due to water extracts of neem leaves in sheep on 28 day post treatment. Likewise, Khalid *et al*. (2005) reported that neem and pineapple (10% water extract of leaves) decreased TLC in sheep.

Chapter 5 Discussion \( \noting 238 \)

#### 5.4.5 Differential leukocyte count (DLC, %)

#### 5.4.5.1 Eosinophil count (%)

Two indigenous medicinal plants, i.e, Neem and bitter gourd and two patent drugs, i.e. Ivertin® and Levavet®, decreased significantly (p<0.05) eosinophil count of differential leukocyte count in cattle of group B, C, D and E, respectively. Mean eosinophil count before treatment was 13.60±0.67 and after treatment with neem mean eosinophil count 7th 14th 21st and 28th day were  $13.00\pm0.44$ ,  $11.80\pm0.58$ ,  $10.80\pm0.58$  and 9.80±0.58, respectively in group B. Mean eosinophil count before treatment was 13.60±0.81 and after treatment with Karalla (bitter gourd) mean eosinophil count 7th 14th 21st and 28th day were 12.60±0.81, 11.60±0.81, 10.20±0.58 and 9.00±0.44, respectively in group C. Mean eosinophil count before treatment was 13.60±0.81 and after treatment with Ivertin® mean eosinophil count 7th 14th 21st and 28th day were  $12.60\pm0.81$ ,  $11.40\pm0.92$ ,  $9.80\pm0.80$  and  $7.60\pm0.50$ , respectively in group D. Mean eosinophil count before treatment was 12.40±0.50 and after treatment with Levavet® Mean eosinophil count 7th 14th 21st and 28th day were  $11.40\pm0.50$ ,  $10.20\pm0.48$ ,  $9.40\pm0.40$  and  $8.40\pm0.40$ , respectively in group E. Mean eosinophil count of control group on pre-treatment (day 0) was 11.80±0.86. Mean TLC on 7th 14th 21st and 28th day were  $12.20\pm0.73$ ,  $13.20\pm0.73$ ,  $13.80\pm0.58$  and  $14.80\pm0.58$ , respectively in group A.

The present finding was also agreement with the work of Khan *et al.* (2003) in cattle. The similar findings due to levamisole were also reported by Shri-Kishan and Gupta (2004) and Khan *et al.* (2003) in cattle. These findings could not be compared due to lack of similar published reports on neem and Karalla, on eosinophil count. The decrease in eosinophil

Chapter 5 Discussion \( \noting 239 \)

count of differential leukocyte count might be due to the decrease of parasitic infection in cattle.

#### 5.4.5.2 Neutrophil count (%)

Two indigenous medicinal plants, i.e, Neem and bitter gourd and two patent drugs, i.e. Ivertin® and Levavet®, increased significantly (p<0.05) neutrophil count of differential leukocyte count in cattle of group B, C, D and E, respectively.

Mean neutrophil count before treatment was 22.40±0.92 and after treatment with neem, mean neutrophil count on 7th 14th 21st and 28th were  $23.00\pm0.89$ ,  $23.80\pm0.73$ ,  $24.60\pm0.81$ and  $25.20\pm0.58$ , respectively in group B. Mean neutrophil count before treatment was 22.80±1.24 and after treatment with Karalla (bitter gourd), mean neutrophil count on 7th 14th 21st and 28th day were 23.20±1.06, 24.20±1.06, 24.80±0.96 and 25.40±0.92, respectively in group C. Mean neutrophil count before treatment was 23.00±0.70 and after treatment with Ivertin®, mean neutrophil count on 7th 14th 21st and 28th day were  $24.00\pm0.70$ ,  $25.00\pm0.63$ ,  $26.00\pm0.63$  and  $27.20\pm0.66$ , respectively in group D. Mean neutrophil count before treatment was 23.00±1.00 and after treatment with Levavet®, mean neutrophil count on 7th 14th 21st and 28th day were  $23.60\pm0.81$ ,  $24.40\pm0.81$ ,  $25.40\pm0.81$  and  $26.00\pm0.70$ , respectively in group E. Mean neutrophil count of control group on pretreatment (day 0) was 25.00±0.70. Mean neutrophil count on 7th 14th 21st and 28th day were  $23.80\pm0.48$ ,  $23.00\pm0.31$ ,  $22.20\pm0.37$  and 21.60±0.40, respectively in group A.

Chapter 5 Discussion \( \noting 240 \)

Due to lack of similar published literature of neem, korolla, ivermectin and levamisole on neutrophil count of differential leukocyte count, these findings could not be compared.

#### **5.4.5.3** *Monocyte count* (%)

Two indigenous medicinal plants, i.e, Neem and bitter gourd and two patent drugs, i.e. Ivertin® and Levavet®, caused very little changes in monocyte count of differential leukocyte count in cattle of group B, C, D and E, respectively. Mean monocyte count before treatment was 4.0±0.31 and after treatment with neem mean monocyte count on 7th 14th 21st and 28th day were  $3.8\pm0.37$ ,  $4.2\pm0.66$ ,  $4.2\pm0.48$  and  $5.0\pm0.00$ , respectively in group B. Mean monocyte count before treatment was 4.8±0.37 and after treatment with Karalla (bitter gourd), mean monocyte count on 7th 14th 21st and 28th day were  $4.4\pm0.40$ ,  $4.00\pm0.63$ ,  $4.20\pm0.58$  and  $4.40\pm0.24$ , respectively in group C. Mean monocyte count before treatment was 3.6±0.24 and after treatment with Ivertin® mean monocyte count on 7th 14th 21st and 28th day were  $3.8\pm0.58$ ,  $3.6\pm0.40$ ,  $3.8\pm0.48$  and  $3.6\pm0.40$ respectively in group D. Mean monocyte count before treatment was 4.60±0.50and after treatment with Levavet® mean monocyte count on 7th 14th 21st and 28th day were  $4.60\pm0.50$ ,  $4.40\pm0.67$ ,  $4.40\pm0.40$  and 3.00±0.31 respectively in group E. Mean monocyte count of control group on pre-treatment (day 0) was 4.20±0.37. Mean monocyte count on 7th 14th 21st and 28th day were  $3.80\pm0.66$ ,  $3.80\pm0.66$ ,  $3.4\pm0.50$  $3.8\pm0.48$ , respectively in group A.

Khan *et al.* (2003) stated that monocyte was not influenced appreciably by levamisole in cattle. These findings could not be compared due to lack of similar published reports of neem and bitter gourd on monocyte count.

Chapter 5 Discussion \( \noting 241 \)

#### 5.4.5.4 Lymphocyte count (%)

Two indigenous medicinal plants, i.e, Neem and bitter gourd and two patent drugs, i.e. Ivertin® and Levavet®, caused very little changes in lymphocyte count of differential leukocyte count in cattle of group B, C. D and E, respectively. Mean lymphocyte count before treatment was 61.20±0.80 and after treatment with neem mean lymphocyte count on 7th 14th 21st and 28th day were 60.20±0.86, 60.60±0.92, 61.00±0.54 and 59.80±0.66, respectively in group B. Mean lymphocyte count before treatment was 58.80±0.73 and after treatment with Karalla (bitter gourd), mean lymphocyte count on 7th 14th 21st and 28th day were 59.40±0.67, 59.80±0.66, 60.00±0.70 and 60.00±0.89, respectively in group C. Mean lymphocyte count before treatment was 60.20±0.37 and after treatment with Ivertin® mean lymphocyte count on 7th 14th 21st and 28th day were  $60.20\pm0.73$ ,  $61.20\pm0.58$ ,  $62.40\pm0.50$  and  $63.40\pm1.07$ , respectively in group D. Mean lymphocyte count before treatment was 58.60±0.24 and after treatment with Levavet® mean lymphocyte count on 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and  $28^{th}$  day were  $58.80\pm0.58$ ,  $60.00\pm1.18$ ,  $59.80\pm0.86$  and  $61.00\pm0.44$ , respectively in group E. Mean lymphocyte count of control group on pretreatment (day 0) was 59.40±0.40. Mean lymphocyte count on the 7th 14th 21st and 28th day was 60.20±0.37, 60.00±0.89, 59.00±0.83 and 59.80±0.73, respectively in group A.

Khan *et al.* (2003) stated that lymphocyte were not influenced appreciably by levamisole in cattle. These findings could not be compared due to lack of similar published reports of neem and bitter gourd on monocyte count.

Chapter 5 Discussion \( \noting 242 \)

#### **5.4.6** Mean corpuscular volume (cubic micron)

Two indigenous medicinal plants, i.e, Neem and bitter gourd and two patent drugs, i.e. Ivertin® and Levavet®, were also caused little changes mean corpuscular volume (MCV) in group B, C, D and E, respectively. Mean MCV before treatment was 36.35±0.16 and after treatment with neem mean MCV on 7th, 14th, 21st and 28th day were 36.97±0.34, 36.54±0.39, 36.87±0.35 and 37.38±0.46, respectively in group B. Mean MCV before treatment was 36.02±0.36 and after treatment with Karalla (bitter gourd), mean MCV on 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 36.97±0.63, 36.91±0.61, 37.37±0.73 and 37.39±0.76, respectively in group C. Mean MCV before treatment was 37.96±0.47 and after treatment with Ivertin®, mean MCV on 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 38.19±0.51, 36.35±0.72, 34.04±0.66 and 33.06±0.62, respectively in group D. Mean MCV before treatment was 36.37±0.52 and after treatment with Levavet®, mean MCV 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 36.56±0.52, 35.28±0.29, 34.10±0.22 and 33.63±0.31, respectively in group E. Mean MCV of control group on pretreatment (day 0) was 35.14±0.97. Mean MCV on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and  $28^{th}$  day was  $34.75\pm0.88$ ,  $34.91\pm0.90$ ,  $34.41\pm0.73$  and  $34.00\pm0.66$ , respectively in group A.

Due to lack of similar published literature of neem, bitter gourd, Ivertin® and Levavet® on mean corpuscular volume (MCV), these findings could not be compared.

### **5.4.7** Mean corpuscular haemogtobin concentration (%)

Two indigenous medicinal plants, i.e, Neem and bitter gourd and two patent drugs, i.e. Ivertin® and Levavet®, increased significantly (p<0.05) mean corpuscular haemoglobin concentration (MCHC) in cattle

of group B, C, D and E, respectively. Mean MCHC before treatment was 28.34±0.54 and after treatment with neem mean MCHC on the 7<sup>th</sup>, 14<sup>th</sup>,  $21^{\text{st}}$  and  $28^{\text{th}}$  day were  $29.36\pm0.71$ ,  $31.64\pm0.45$ ,  $33.07\pm0.22$  and 35.73±0.27, respectively in group B. Mean MCHC before treatment was 29.17±0.93 and after treatment with Karalla (bitter gourd) mean MCHC on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 29.53±0.91, 30.52±0.76, 31.71±0.77 and 33.15±0.67, respectively in group C. Mean MCHC before treatment was 28.81±0.73 and after treatment with Ivertin® mean MCHC on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 30.20±0.86, 31.91±0.51, 34.07±0.54 and 36.39±0.78, respectively in group D. Mean MCHC before treatment was 30.09±0.80 and after treatment with Levavet® mean MCHC on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 30.51±0.79, 31.83±0.68, 33.63±0.52 and 35.09±0.34, respectively in group E. Mean MCHC of control group on pre-treatment (day 0) was 32.70±1.07. Mean MCHC on the  $7^{th}$ ,  $14^{th}$ ,  $21^{st}$  and  $28^{th}$  day was  $31.11\pm1.23$ ,  $29.65\pm1.22$ ,  $27.83\pm0.84$ and 27.21±0.34, respectively in group A.

The effects of neem, bitter gourd, Ivertin® and Levavet® on mean corpuscular haemoglobin concentration (MCHC) have an increase level in this study. Arlian *et al.* (1988) reported that ivermectin treated rabbit have an increase level of mean corpuscular hacmoglobin concentration (MCHC) in comparison to those of untreated controlled rabbit. The increase in MCHC might be due to the increase of packed cell volume (PCV) and haemoglobin (Hb) content.

#### 5.4.8 Mean corpuscular haemoglobin (micro micro gram)

Two indigenous medicinal plants, i.e, Neem and bitter gourd and two patent drugs, i.e. Ivertin® and Levavet®, increased significantly (p<0.05) mean corpuscular haemoglobin (MCH) in cattle of group B, C, D and E, respectively. Mean MCH before treatment was 10.30±0.20 and

Chapter 5 Discussion \( \noting 244 \)

after treatment with neem mean MCH on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> were 10.85±0.25, 11.56±0.22, 12.19±0.10 and 13.36±0.24, respectively in group B. Mean MCH before treatment was 10.50±0.29 and after treatment with Karalla (bitter gourd) mean MCH on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> were 10.90±0.21, 11.26±0.29, 11.85±0.30 and 12.39±0.30, respectively in group C. Mean MCH before treatment was 10.94±0.37 and after treatment with Ivertin® mean MCH on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> were 11.54±0.41, 11.61±0.32, 11.59±0.14 and 12.02±0.13, respectively in group D. Mean MCH before treatment was 10.94±0.30 and after treatment with Levavet® mean MCH on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> were 11.14±0.25, 11.23±0.22, 11.47±0.19 and 11.80±0.15, respectively in group E. Mean MCH of control group on pre-treatment (day 0) was 11.46±0.26. Mean MCH on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day was 10.77±0.23, 10.31±0.22, 9.56±0.17 and 9.24±0.07, respectively in group A.

Due to lack of adequate literature of neem, bitter gourd, Ivertin® and Levavet® treatment on mean corpuscular haemoglobin (MCH), the present findings could not be compared with the findings of other workers. The increase in MCH content might be due to the increase of total. erythrocyte count (TEC) and haemoglobin (Hb) content.

# 5.5 Effects of Two Indigenous Medicinal Plants and Two Patent Drugs on some Serum Biochemical Parameters in Cattle

## 5.5.1 Alanine aminotransferase level of serum (U/I)

Two indigenous medicinal plants, i.e, Neem and bitter gourd and two patent drugs, i.e. Ivertin® and Levavet®, could not significantly changed alanine aminotransferase (ALT) level of serum in cattle of group B, C, D and E, respective1y. Mean ALT level before treatment was 20.22±0.74

Chapter 5 Discussion \( \notin \) 245

and after treatment with neem mean ALT level on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 20.40± 1.16, 19.58±0.67, 20.32±1.15 and 19.22±0.94, respectively in group B. Mean ALT level before treatment was 19.90±1.29 and after treatment with Karalla (bitter gourd) mean ALT level on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 19.26±1.14, 19.82±1.15, 19.52±1.00 and 19.70±0.82, respectively in group C. Mean ALT level before treatment was 20.14±0.56 and after treatment with Ivertin® mean ALT level on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 19.48±1.10, 19.40±0.80, 19.56±0.77 and 19.48±0.92, respectively in group D. Mean ALT level before treatment was 18.52±0.56 and after treatment with Levavet® mean ALT level on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 18.22±0.86, 18.32±0.81, 18.06±0.75 and 17.78±0.90, respectively in group E. Mean ALT of control group on pre-treatment (day 0) was 18.26±0.99. Mean ALT on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day was 18.40±0.75, 18.14±1.14, 18.00±1.22 and 19.24±1.00, respectively in group A.

Due to lack of adequate literature of neem, bitter gourd, Ivertin® and Levavet® treatment on alanine aminotransferase level of serum, the present findings could not be compared with the findings of other workers.

#### 5.5.2 Aspartate aminotransferase level of serum (U/I)

Neem, bitter gourd, Ivertin® and Levavet®, could not significantly changed aspartate aminotransferase (AST) level of serum in cattle of group B, C, D and E, respectively. Mean AST level before treatment was 32.84±1.05 and after treatment with neem mean AST level on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 31.26±0.93, 31.88±0.99, 32.72±1.46 and 31.96±0.85, respectively in group B. Mean AST level before treatment was 31.74±1.30 and after treatment with Karalla (bitter gourd) mean AST

level on the  $7^{th}$ ,  $14^{th}$ ,  $21^{st}$  and  $28^{th}$  day were  $33.14\pm1.10$ ,  $32.46\pm0.67$ ,  $31.02\pm1.25$  and  $31.82\pm0.81$ , respectively in group C. Mean AST level before treatment was  $32.22\pm0.51$  and after treatment with Ivertin® mean AST level on the  $7^{th}$ ,  $14^{th}$ ,  $21^{st}$  and  $28^{th}$  day were  $31.72\pm0.95$ ,  $32.36\pm0.78$ ,  $31.16\pm0.77$  and  $31.36\pm0.73$ , respectively in group D. Mean AST level before treatment was  $30.50\pm0.68$  and after treatment with Levavet® mean AST level on the  $7^{th}$ ,  $14^{th}$ ,  $21^{st}$  and  $28^{th}$  day were  $31.94\pm1.01$ ,  $31.60\pm0.64$ ,  $29.80\pm1.29$  and  $30.06\pm0.88$ , respectively in group E. Mean AST level of control group on pre-treatment (day 0) was  $30.28\pm1.07$ . Mean AST level on the  $7^{th}$ ,  $14^{th}$ ,  $21^{st}$  and  $28^{th}$  day were  $29.94\pm1.13$ ,  $31.10\pm0.91$ ,  $29.36\pm1.12$  and  $31.58\pm0.76$ , respectively in group A.

These findings could not be compared due to lack of similar published reports of neem, bitter gourd, Ivertin® and Levavet® treatment on aspartate aminotransferase (AST) level of serum.

## 5.6 Effects of Two Indigenous Medicinal Plants and Two Patent Drugs on Body Weight (Kg) in Cattle

Neem, bitter gourd, Ivertin® and Levavet®, significantly (p<0.05) increased body weight in group B, C, D and E, respectively. Mean body weight before treatment was 124.90±2.05 and after treatment with neem mean body weight on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 125.40±2.05, 126.00±2.06, 126.90±2.00 and 127.80±1.89, respectively in group B. Mean body weight before treatment was 124.50±1.10 and after treatment with Karalla (bitter gourd) mean body weight on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 124.70±1.06, 125.50±0.89, 126.70±0.94 and 127.60±0.87, respectively in group C. Mean body weight before treatment was 126.80±1.12 and after treatment with Ivertin® mean body weight on the 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day were 127.30±1.12, 128.20±1.07, 129.10±1.06 and 130.40±0.90, respectively in group D. Mean body weight before

treatment was  $123.90\pm1.93$  and after treatment with Levavet® mean body weight on the  $7^{th}$ ,  $14^{th}$ ,  $21^{st}$  and  $28^{th}$  day were  $124.50\pm2.02$ ,  $125.50\pm1.93$ ,  $126.40\pm1.85$  and  $127.40\pm1.81$ , respectively in group E. Mean body weight of control group on pre-treatment (day 0) was  $125.40\pm1.46$ . Mean body weight on the  $7^{th}$ ,  $14^{th}$ ,  $21^{st}$  and  $28^{th}$  day was  $125.00\pm1.38$ ,  $124.20\pm1.45$ ,  $123.60\pm1.42$  and  $122.90\pm1.52$ , respectively in group A.

Khalid *et al.* (2005) reported that body weight was increased significantly in neem and Karalla treated sheep. On the other hand, body weight was decreased in untreated sheep. Hossain et al. (1996) also observed neem leaves and neem seed kernels increased body weight of cattle. Bauck et al. (1989) observed that ivermectin treated calves gained an increase average body weight per day in comparison to the control animals. The average daily weight gained by ivermectin treated cattle was recorded as 0.08 kg for each individual animal. Kennedy (1990) studied the effect of Ivermectin on gaining body weight of calves in USA. Calves treated with Ivermectin indictable preparation (200µg/kg, S/C) significantly gained more weight than the control animals. Similar response also reported by Ahmed et al. (1994) in sheep, Amin et al. (2010), Amin et al. (2008), Begum et al. (2010), Mian (2011). Khan et al. (2003) observed that body weight was increased significantly due to levamisole in cattle. On the other hand, body weight was decreased in untreated cattle. Similar results also reported by Vassilev (1993) and Redl (1991) in cattle. Khan et al. (2003) also found that body weight was increased significantly due to levamisole in cattle. Similar effects reported by Beriajaya et al. (1986) due to levamisole in sheep and Redl (1991) in cattle. The parasitic infection might be responsible to arrest the growth. The body weight was increased might be due to removal of parasitic load which facilitate the weight regain through proper digestion, absorption and metabolism of feed nutrients in the parasite free gastro-intestinal tract.

# CHAPTER-6 SUMMARY AND CONCLUSIONS

#### **SUMMARY**

The experiment was performed in the Department of Animal Husbandry & Veterinary Science, Faculty of Agriculture, and University of Rajshahi in collaboration with the Department of Pharmacology, Physiology and Parasitology, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh, Bangladesh. The detailed research work was conducted to investigate the prevalence of gastro-intestinal nematodes in cattle during the period from June, 2011 to May, 2012 in greater Rajshahi in Bangladesh, to compare anthelmintic efficacy of 10 indigenous medicinal plants *in-vitro* and to evaluate the effects of 2 indigenous medicinal plants viz. Neem and bitter gourd and 2 patent drugs Ivermectin (Ivertin®) and levamisole (Levavet®) against natural gastro-intestinal nematodes in 25 cattle for 28 days during 1st June, 2012 to 28th June, 2012 at Binodpur, Rajshahi. The research works were conducted to observe the prevalence of gastro-intestinal nematodes in cattle.

The overall prevalence of *Ascaris* sp., *Strongyles* (*Haemonchus* sp., *Trichostrongylus* sp., *Oesophagostomum* sp. and *Mecistocirrus* sp.), *Bunostomum* sp., *Trichuris* sp. *Strongyloides* sp. and *mixed infection* were 6.5%, 25.5%, 2.25%, 5% 5% and 10.5%, respectively. The prevalence of gastro-intestinal nematodes was 36.7% in the age of 1 to < 6 months, 62.5% in the age of 6 to <12 months, 67.0% in the age of 12 to <24 months in the age of >24 months 38.0%. The incidence of gastro-

intestinal nematodes was highest in 12 to <24 months old cattle followed by 6 to <12 months old cattle, >24 months old cattle and 1 to < 6 months old cattle. In the age of 1to < 6 months the prevalence of gastro-intestinal nematodes was 36.7% (Ascaris sp. and Strongyles were 33.3% and 3.3% respectively), in the age of 6 to <12 months, the prevalence of gastrointestinal nematodes was 62.5% (Ascaris sp Strongyles Trichuris sp. Strongyloides sp. and mixed infection were 15.0%, 25.0%, 5.0%, 5.0% and 12.5% respectively), in the age of 12 to <24 months, the prevalence of gastro-intestinal nematodes was 67.0% (Strongyles sp., Bunostomum sp., Trichuris sp., Strongyloides sp. and mixed infection were 35.0%, 4.0%, 7.5%, 7.0% and 13.5% respectively), in the age of >24 months, the prevalence of gastro-intestinal nematodes was 38.0% (Strongyles sp., Bunostomum sp., Trichuris sp., Strongyloides sp. and mixed infection were 20.0%, 1.0%, 3.0%, 4.0% and 10.0% respectively). The seasonal prevalence of gastro-intestinal nematodes was significantly highest in rainy seasons(June-August) 79.8% followed by autumn (September-November) 57.7% summer (March-May) 44.3% and winter seasons (December-February) 34.8% and This infestation rate of gastro-intestinal nematodes with their breed higher in cross breed cattle 184 (58.4%) than in local breed cattle 35 (41.2%). This infestation rate of gastro-intestinal parasites with their genotype highest in L×F genotype of cattle 56 (60.9%) followed by L×F×SL genotype of cattle 46 (57.5%), L×SL genotype of cattle 82 (57.3%) and in local cattle 35 (41.2%). Highest incidence was also observed in female (62.0%) than male (44.2%). The incidence of gastro-intestinal nematodes was highest in the month of June and July. The incidence of gastro-intestinal nematodiasis in cattle were 33.34%, 34.37%, 37.5%, 42.85%, 53.33%, 82.86%, 88.23%, 74.29%, 67.64%, 60%, 45.72% and 36.37% in January, February, March, April,

May, June, July, August, September, October, November and December, respectively. The incidence of gastro-intestinal nematodes was highest in Rajshahi district (62.5) followed by Natore district (56.2) Naogaon district (42.9) and Chapai Nawabgonj district (38.0).

Screening of 10 indigenous medicinal plants (Neem, custard apple, papaya, garlic, bitter gourd, jute, chaste tree, pineapple, pomegranate and betel leaf) of Bangladesh having reported anthelmintic activity and to determine the comparative efficacy *in-vitro*. Freshly prepared water and ethanol extracts of 10 indigenous medicinal plants were screened *in vitro* in this study, at various concentration levels, using adult gastro-intestinal nematode parasites of cattle. In water extract, within these 10 plants, 3 plants (Neem, bitter gourd and betel leaf) showed *in vitro* 100% efficacy against adult worms, 2 plants (garlic and pineapple) showed 90% efficacy and others (custard apple, papaya, jute, chaste tree and pomegranate) showed below 80% efficacy at a concentration of 100 mg/ml. In ethanol extract, within these 10 plants, 5 plants (Neem, garlic, bitter gourd pineapple and betel leaf) showed *in vitro* 100% efficacy against adult worms, and 5 showed 80-90% efficacy at a concentration of 100 mg/ml.

Among 10 tested plants 2 medicinal plants (neem, and korolla) and 2 patent drugs (Ivertin and Levavet®) were administered against gastro-intestinal nematodes in cattle for *in vivo* trail. Twenty five (25) naturally parasitized female cattle of Binodpur, Rajshahi, were selected for efficacy study of 2 indigenous medicinal plants and 2 patent drugs *in-vivo*. The animals were divided into five (5) groups (A, B, C, D, and E), each group consisting of five (5) cattle. Ethanol extract of leaves of neem @100 mg/kg b. wt and bitter gourd @100 mg/kg b. wt were administered orally to the cattle of group B and C respectively. Cattle of group D and E were

treated with ivermectin (Ivertin®) @0.2 mg/kg b. wt pour on and levamisole (Levavet®) @ 7.5 mg/kg b. wt. respectively. Cattle of group A was kept as infected control group. Prior to trials, initial body weight, of gastro-intestinal nematodes, biochemical, total count egg hematological and clinical parameters were examined and recorded. During the experimental period, faecal samples were examined on 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day. Clinical (body weight), haematological (TEC, Hb, PCV, TLC, DLC, MCV, MCHC and MCH) and biochemical parameters (ALT and AST) were also observed on 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day for the determination of effects of neem, bitter gourd, Ivertin® and Levavet®. A significant (p<0.05) reduction of EPG count was found on 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day of neem (52.87%, 62.07%, 48.97% and 35.40%), bitter gourd (43.02%, 50.23%, 34.88% and 16.98%), Ivertin® (100%, 100%, 100% and 100%) and Levavet® (100%, 94.86%, 87.01% and 78.55%) treated cattle of group B, C, D, and E respectively. The EPG count of control group (A) were significantly (p<0.05) increased up to last day of experimental period. The EPG count of untreated control group (A) were significantly (p<0.05) increased about 7.58%, 20.61%, 37.88% and 59.09% on 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day, respectively.

After treatment with Need, bitter gourd, Ivertin® and Levavet®, total erythrocyte count (TEC), haemoglobin (Hb) content, packed cell volume (PCV), neutrophil count of differential leukocyte count (DLC), mean corpuscular haemoglobin concentration (MCHC) and mean corpuscular haemoglobin (MCH) were increased significantly (p<0.05) in cattle. On the other hand, total leukocyte count (TLC) and eosinophil count of differential leukocyte count (DLC) were decreased significantly (p<0.05) in treated cattle. Those indigenous medicinal plants and patent drugs were

caused very little changes monocyte count and lymphocyte count of differential leukocyte count (DLC) in cattle. The alanine aminotransferase (ALT) and aspartate aminotransferase (AST) level of serum was not significantly changed in cattle. In treated cattle body weight was increased significantly (p<0.05) with neem, korolla, bitter gourd, Ivertin® and Levavet®.

#### **CONCLUSIONS**

The prevalence of gastro-intestinal nematodiasis in cattle were studied in relation with season, region, months of a year, sex, age, breed genotype of the host in greater Rajshahi of Bangladesh.

#### In first study -

- The maximum prevalence of *Strongyles* was (46.6%) and the minimum was *Trichuris* (4.2%) in the study area.
- The highest incidence of gastro-intestinal parasitic infestation was found in 12 to <24 months (67.0%) old cattle and lowest in 1 to <6 months (36.7%).
- The maximum incidence of gastro-intestinal nematode infestation was found in female (62.1%) than male (44.2%) in the study area.
- The highest prevalence of gastro-intestinal nematode infestation was found in cross breed (58.4%) and lowest in local breed (41.2%).
- The highest prevalence of gastro-intestinal nematode infestation was found in L×F genotype (60.9%) and lowest in local breed (41.2%).
- The autumn season was fevourable for the gastro-intestinal nematode infestation (57.8%) among the other season.
- The lowest gastro-intestinal nematode infestation (33.4%) was found in the month of January and highest infestation (88.3%) was found in July.
- The highest gastro-intestinal parasitic infestation was found in Rajshahi district (62.5%) and lowest in Chapai Nawabgonj district (38.0%).

#### In second study –

- In water extract, within these 10 plants, 3 plants (Neem, bitter gourd and betel leaf) showed *in vitro* 100% efficacy against adult worms.
- In ethanol extract, within these 10 plants, 5 plants (Neem, garlic, bitter gourd pineapple and betel leaf) showed *in vitro* 100% efficacy against adult worms.

#### In third study -

- A significant (p<0.05) reduction of EPG count was found on 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> day of neem (52.87%, 62.07%, 48.97% and 35.40%), bitter gourd (43.02%, 50.23%, 34.88% and 16.98%), Ivertin® (100%, 100%, 100% and 100%) and Levavet® (100%, 94.86%, 87.01% and 78.55%) treated cattle of group B, C, D, and E, respectively.
- After treatment with Neem, bitter gourd, Ivertin® and Levavet®, total erythrocyte count (TEC), haemoglobin (Hb) content, packed cell volume (PCV), neutrophil count of differential leukocyte count (DLC), mean corpuscular haemoglobin concentration (MCHC) and mean corpuscular haemoglobin (MCH) were increased significantly (p<0.05) in cattle.
- Total leukocyte count (TLC) and eosinophil count of differential leukocyte count (DLC) were decreased significantly (p<0.05) in treated cattle.
- In treated cattle body weight was increased significantly (p<0.05) with neem, korolla, bitter gourd, Ivertin® and Levavet® at 21 days after treatment.

#### **CHAPTER-7**

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### **APPENDICES**

Appendix –I: Average incidence of gastro-intestinal nematodes among different age of cattle in greater Rajshahi.

age \* infected Crosstabulation

			infecte	ed	
			noninfected	Infected	Total
age	1 to <6 months	Count	38	22	60
		Expected Count	27.2	32.8	60.0
		% within age	63.3%	36.7%	100.0%
		% within infected	21.0%	10.0%	15.0%
		% of Total	9.5%	5.5%	15.0%
	7 to <12 months	Count	15	25	40
		Expected Count	18.1	21.9	40.0
		% within age	37.5%	62.5%	100.0%
		% within infected	8.3%	11.4%	10.0%
		% of Total	3.8%	6.2%	10.0%
	12 to <24 months	Count	66	134	200
		Expected Count	90.5	109.5	200.0
		% within age	33.0%	67.0%	100.0%
		% within infected	36.5%	61.2%	50.0%
		% of Total	16.5%	33.5%	50.0%
	> 24 months	Count	62	38	100
		Expected Count	45.2	54.8	100.0
		% within age	62.0%	38.0%	100.0%
		% within infected	34.3%	17.4%	25.0%
		% of Total	15.5%	9.5%	25.0%
Total		Count	181	219	400
		Expected Count	181.0	219.0	400.0
		% within age	45.2%	54.8%	100.0%
		% within infected	100.0%	100.0%	100.0%
		% of Total	45.2%	54.8%	100.0%

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	32.328 <sup>a</sup>	3	.000
Likelihood Ratio	32.634	3	.000
Linear-by-Linear Association	.037	1	.847
N of Valid Cases	400		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 18.10.

## Appendix –II: Incidence of GI nematodes species in different age of cattle in greater Rajshahi

Age \* parasites Cross tabulation

Age		parasites							
		Negative	Ascaris .sp	Strongylus	Bunostomum sp.	Trichuris sp	Strong- yloides	mixed infection	Total
1 to <6	Count	38	20	2	0	0	0	0	60
months	Expected Count	27.2	3.9	15.3	1.4	3.0	3.0	6.3	60.0
	% within Age	63.3%	33.3%	3.3%	.0%	.0%	.0%	.0%	100.0%
	% within parasites	21.0%	76.9%	2.0%	.0%	.0%	.0%	.0%	15.0%
	% of Total	9.5%	5.0%	.5%	.0%	.0%	.0%	.0%	15.0%
7 to <12	Count	15	6	10	0	2	2	5	40
months	Expected Count	18.1	2.6	10.2	.9	2.0	2.0	4.2	40.0
	% within Age	37.5%	15.0%	25.0%	.0%	5.0%	5.0%	12.5%	100.0%
	% within parasites	8.3%	23.1%	9.8%	.0%	10.0%	10.0%	11.9%	10.0%
	% of Total	3.8%	1.5%	2.5%	.0%	.5%	.5%	1.2%	10.0%
12 to <24	Count	66	0	70	8	15	14	27	200
months	Expected Count	90.5	13.0	51.0	4.5	10.0	10.0	21.0	200.0
	% within Age	33.0%	.0%	35.0%	4.0%	7.5%	7.0%	13.5%	100.0%
	% within parasites	36.5%	.0%	68.6%	88.9%	75.0%	70.0%	64.3%	50.0%
	% of Total	16.5%	.0%	17.5%	2.0%	3.8%	3.5%	6.8%	50.0%
<24	Count	62	0	20	1	3	4	10	100
months	Expected Count	45.2	6.5	25.5	2.2	5.0	5.0	10.5	100.0
	% within Age	62.0%	.0%	20.0%	1.0%	3.0%	4.0%	10.0%	100.0%
	% within parasites	34.3%	.0%	19.6%	11.1%	15.0%	20.0%	23.8%	25.0%
	% of Total	15.5%	.0%	5.0%	.2%	.8%	1.0%	2.5%	25.0%
Total	Count	181	26	102	9	20	20	42	400
	Expected Count	181.0	26.0	102.0	9.0	20.0	20.0	42.0	400.0
	% within Age	45.2%	6.5%	25.5%	2.2%	5.0%	5.0%	10.5%	100.0%
	% within parasites	100.0%	100.0%	100.0%	100.0%	100.0%	100.0%	100.0%	100.0%
	% of Total	45.2%	6.5%	25.5%	2.2%	5.0%	5.0%	10.5%	100.0%

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	1.529E2 <sup>a</sup>	18	.000
Likelihood Ratio	157.460	18	.000
Linear-by-Linear Association	8.769	1	.003
N of Valid Cases	400		

### Appendix –III: Incidence of gastro-intestinal nematodes in different sex of cattle

gender \* infected Cross tabulation

	-	gender * infected	infecto	ed	
			noninfected	infected	Total
gender	female	Count	90	147	237
		Expected Count	107.2	129.8	237.0
		% within gender	38.0%	62.0%	100.0%
		% within infected	49.7%	67.1%	59.2%
		% of Total	22.5%	36.8%	59.2%
	male	Count	91	72	163
		Expected Count	73.8	89.2	163.0
		% within gender	55.8%	44.2%	100.0%
		% within infected	50.3%	32.9%	40.8%
		% of Total	22.8%	18.0%	40.8%
Total		Count	181	219	400
		Expected Count	181.0	219.0	400.0
		% within gender	45.2%	54.8%	100.0%
		% within infected	100.0%	100.0%	100.0%
		% of Total	45.2%	54.8%	100.0%

	Value	df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	12.426 <sup>a</sup>	1	.000		
Continuity Correction <sup>b</sup>	11.716	1	.001		
Likelihood Ratio	12.448	1	.000		
Fisher's Exact Test				.001	.000
Linear-by-Linear Association	12.395	1	.000		
N of Valid Cases <sup>b</sup>	400				

# Appendix –IV: Incidence of gastro-intestinal nematodes in different breed of cattle

breed \* infestation Cross tabulation

			infest	ation	
			Negative	infected	Total
breed	local	Count	50	35	85
		Expected Count	38.5	46.5	85.0
		% within breed	58.8%	41.2%	100.0%
		% within infestation	27.6%	16.0%	21.2%
		% of Total	12.5%	8.8%	21.2%
	cross bred	Count	131	184	315
		Expected Count	142.5	172.5	315.0
		% within breed	41.6%	58.4%	100.0%
		% within infestation	72.4%	84.0%	78.8%
		% of Total	32.8%	46.0%	78.8%
Total		Count	181	219	400
		Expected Count	181.0	219.0	400.0
		% within breed	45.2%	54.8%	100.0%
		% within infestation	100.0%	100.0%	100.0%
		% of Total	45.2%	54.8%	100.0%

	Value	df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	8.027 <sup>a</sup>	1	.005		
Continuity Correction <sup>b</sup>	7.346	1	.007		
Likelihood Ratio	8.006	1	.005		
Fisher's Exact Test				.007	.003
Linear-by-Linear Association	8.007	1	.005		
N of Valid Cases <sup>b</sup>	400				

## Appendix –V: Incidence of gastro-intestinal nematodes in different genotype of cattle in greater Rajshahi

genotype \* infestation Cross tabulation

			infesta	ation	m . 1
			Negative infected		Total
genotype	local	Count	50	35	85
		Expected Count	38.5	46.5	85.0
		% within genotype	58.8%	41.2%	100.0%
		% within infestation	27.6%	16.0%	21.2%
		% of Total	12.5%	8.8%	21.2%
	L×SL	Count	61	82	143
		Expected Count	64.7	78.3	143.0
		% within genotype	42.7%	57.3%	100.0%
		% within infestation	33.7%	37.4%	35.8%
		% of Total	15.2%	20.5%	35.8%
	L×F	Count	36	56	92
		Expected Count	41.6	50.4	92.0
		% within genotype	39.1%	60.9%	100.0%
		% within infestation	19.9%	25.6%	23.0%
		% of Total	9.0%	14.0%	23.0%
	L×F×SL	Count	34	46	80
		Expected Count	36.2	43.8	80.0
		% within genotype	42.5%	57.5%	100.0%
		% within infestation	18.8%	21.0%	20.0%
		% of Total	8.5%	11.5%	20.0%
Total	-	Count	181	219	400
		Expected Count	181.0	219.0	400.0
		% within genotype	45.2%	54.8%	100.0%
		% within infestation	100.0%	100.0%	100.0%
		% of Total	45.2%	54.8%	100.0%

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	8.344 <sup>a</sup>	3	.039
Likelihood Ratio	8.330	3	.040
Linear-by-Linear Association	4.383	1	.036
N of Valid Cases	400		

## Appendix -VI: Incidence of gastro-intestinal nematodes of cattle in greater Rajshahi during different seasons

**Seasons \* Infection Cross tabulation** 

		-	Infectio	n	
			Non infected	Infected	Total
Seasons	Summer	Count	54	43	97
		Expected Count	43.9	53.1	97.0
		% within Seasons	55.7%	44.3%	100.0%
		% within Infection	29.8%	19.6%	24.2%
		% of Total	13.5%	10.8%	24.2%
	Rainy season	Count	21	83	104
		Expected Count	47.1	56.9	104.0
		% within Seasons	20.2%	79.8%	100.0%
		% within Infection	11.6%	37.9%	26.0%
		% of Total	5.2%	20.8%	26.0%
	Autumn	Count	44 60		104
		Expected Count	47.1	56.9	104.0
		% within Seasons	42.3%	57.7%	100.0%
		% within Infection	24.3%	27.4%	26.0%
		% of Total	11.0%	15.0%	26.0%
	Winter	Count	62	33	95
		Expected Count	43.0	52.0	95.0
		% within Seasons	65.3%	34.7%	100.0%
		% within Infection	34.3%	15.1%	23.8%
		% of Total	15.5%	8.2%	23.8%
Total	•	Count	181	219	400
		Expected Count	181.0	219.0	400.0
		% within Seasons	45.2%	54.8%	100.0%
		% within Infection	100.0%	100.0%	100.0%
		% of Total	45.2%	54.8%	100.0%

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	46.331ª	3	.000
Likelihood Ratio	48.640	3	.000
Linear-by-Linear Association	5.140	1	.023
N of Valid Cases	400		

Appendix -VII: Incidence of gastro-intestinal nematodes of cattle in greater Rajshahi among different months in a year.

**Months \* Infection Cross tabulation** 

			Infect	ion		
			Negative	Positive	Total	
Months	January	Count	20	10	30	
		Expected Count	13.6	16.4	30.0	
		% within Months	66.7%	33.3%	100.0%	
		% within Infection	11.0%	4.6%	7.5%	
		% of Total	5.0%	2.5%	7.5%	
	February	Count	21	11	32	
		Expected Count	14.5	17.5	32.0	
		% within Months	65.6%	34.4%	100.0%	
		% within Infection	11.6%	5.0%	8.0%	
		% of Total	5.2% 2.8%			
	March	Count	20	12	32	
		Expected Count	14.5	17.5	32.0	
		% within Months	62.5%	37.5%	100.0%	
		% within Infection	11.0%	5.5%	8.0%	
		% of Total	5.0%	3.0%	8.0%	
	April	Count	20	15	35	
		Expected Count	15.8	19.2	35.0	
		% within Months	57.1%	42.9%	100.0%	
		% within Infection	11.0%	6.8%	8.8%	
		% of Total	5.0%	3.8%	8.8%	
	May	Count	14	16	30	
		Expected Count	13.6	16.4	30.0	
		% within Months	46.7%	53.3%	100.0%	
		% within Infection	7.7%	7.3%	7.5%	
		% of Total	3.5%	4.0%	7.5%	
	June	Count	8	27	3:	
		Expected Count	15.8	19.2	35.0	
		% within Months	22.9%	77.1%	100.0%	
		% within Infection	4.4%	12.3%	8.8%	
		% of Total	2.0%	6.8%	8.8%	
	July	Count	4	30	34	
		Expected Count	15.4	18.6	34.0	
		% within Months	11.8%	88.2%	100.0%	
		% within Infection	2.2%	13.7%	8.5%	
		% of Total	1.0%	7.5%	8.5%	
	August	Count	9	26	3:	
		Expected Count	15.8	19.2	35.0	

			Infect	tion	
			Negative	Positive	Total
	-	% within Months	25.7%	74.3%	100.0%
		% within Infection	5.0%	11.9%	8.8%
		% of Total	2.2%	6.5%	8.8%
	September	Count	11	23	34
		Expected Count	15.4	18.6	34.0
		% within Months	32.4%	67.6%	100.0%
		% within Infection	6.1%	10.5%	8.5%
		% of Total	2.8%	5.8%	8.5%
	October	Count	14	21	35
		Expected Count	15.8	19.2	35.0
		% within Months	40.0%	60.0%	100.0%
		% within Infection	7.7%	9.6%	8.8%
		% of Total	3.5%	5.2%	8.8%
	November	Count	19	16	35
		Expected Count	15.8	19.2	35.0
		% within Months	54.3%	45.7%	100.0%
		% within Infection	10.5%	7.3%	8.8%
		% of Total	4.8%	4.0%	8.8%
	December	Count	21	12	33
		Expected Count	14.9	18.1	33.0
		% within Months	63.6%	36.4%	100.0%
		% within Infection	11.6%	5.5%	8.2%
		% of Total	5.2%	3.0%	8.2%
Total	·	Count	181	219	400
		Expected Count	181.0	219.0	400.0
		% within Months	45.2%	54.8%	100.0%
		% within Infection	100.0%	100.0%	100.0%
		% of Total	45.2%	54.8%	100.0%

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	52.975 <sup>a</sup>	11	.000
Likelihood Ratio	56.326	11	.000
Linear-by-Linear Association	5.090	1	.024
N of Valid Cases	400		

## Appendix –VIII: Incidence of Gastro-intestinal nematodes of cattle in different districts of grater Rajshahi

**District \* Parasite Cross tabulation** 

		District * Parasite Cross	s tabulation		
			Parasi	te	
			Non infected	Infected	Total
District	Rajshahi	Count	75	125	200
		Expected Count	90.5	109.5	200.0
		% within District	37.5%	62.5%	100.0%
		% within Parasite	41.4%	57.1%	50.0%
		% of Total	18.8%	31.2%	50.0%
	Chapai Nawabgonj	Count	31	19	50
		Expected Count	22.6	27.4	50.0
		% within District	62.0%	38.0%	100.0%
		% within Parasite	17.1%	8.7%	12.5%
		% of Total	7.8%	4.8%	12.5%
	Natore	Count	35	45	80
		Expected Count	36.2	43.8	80.0
		% within District	43.8%	56.2%	100.0%
		% within Parasite	19.3%	20.5%	20.0%
		% of Total	8.8%	11.2%	20.0%
	Nonagon	Count	40	30	70
		Expected Count	31.7	38.3	70.0
		% within District	57.1%	42.9%	100.0%
		% within Parasite	22.1%	13.7%	17.5%
		% of Total	10.0%	7.5%	17.5%
Total		Count	181	219	400
		<b>Expected Count</b>	181.0	219.0	400.0
		% within District	45.2%	54.8%	100.0%
		% within Parasite	100.0%	100.0%	100.0%
		% of Total	45.2%	54.8%	100.0%

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	14.580 <sup>a</sup>	3	.002
Likelihood Ratio	14.613	3	.002
Linear-by-Linear Association	6.900	1	.009
N of Valid Cases	400		

Appendix -IX: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) against G.I nematodes (no. of EPG) in Cattle.

Group	Treatment	Tag No.	Day 0	Day 7	Day 14	Day 21	Day 28
		101	700	750	800	900	1050
		102	620	650	700	800	950
A	No Drug (Control)	103	780	800	880	950	1000
	(Control)	104	700	750	900	1050	1250
		105	500	600	700	850	1000
		106	900	450	350	420	500
		107	800	350	250	400	480
В	Neem	108	1000	550	500	600	650
		109	750	250	200	300	550
		110	900	450	350	500	630
	Korolla	111	1050	650	550	680	800
		112	700	400	350	450	500
С		113	850	450	400	500	750
		114	950	600	540	750	880
		115	750	350	300	420	640
		116	720	0	0	0	0
	Ivermectin	117	750	0	0	0	0
D	200mg/kg	118	630	0	0	0	0
	pour-on	119	650	0	0	0	0
		120	900	0	0	0	0
		121	650	0	100	200	250
	Levavet 7.5mg (orally)	122	680	0	20	50	150
E		123	780	0	0	30	80
		124	610	0	50	100	150
		125	590	0	0	50	80

Appendix -X: The Effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on total erythrocyte count (million/cu.mm.) in cattle.

Group	Treatment	Tag No.	Day 0	Day 7	Day 14	Day 21	Day 28
		101	8.78	8.72	8.60	8.48	8.32
		102	9.02	8.93	8.72	8.56	8.32
A	No Drug (Control)	103	9.52	9.38	9.14	8.92	8.63
	(Control)	104	9.78	9.64	9.52	9.32	9.10
		105	8.46	8.37	8.16	8.02	7.84
		106	8.92	8.96	9.21	9.43	9.62
		107	7.76	7.82	8.22	8.50	8.70
В	Neem	108	8.70	8.76	9.00	9.30	9.00
		109	8.37	8.47	8.74	9.06	9.32
		110	7.82	7.86	8.16	8.34	8.60
	Korolla	111	8.46	8.49	8.70	8.92	9.01
		112	7.72	7.75	7.91	8.07	8.14
C		113	8.93	8.96	9.10	9.23	9.36
		114	8.18	8.0	8.38	8.56	8.71
		115	9.81	9.84	10.03	10.19	10.27
		116	8.32	8.38	9.16	10.11	10.67
	Ivermectin	117	8.28	8.37	9.06	10.14	10.64
D	200mg/kg	118	8.30	8.42	9.44	10.29	10.72
	pour-on	119	8.56	8.67	9.18	9.98	10.48
		120	8.20	8.44	9.34	10.06	10.58
		121	8.16	8.33	9.17	10.07	10.52
	Levavet	122	9.34	9.48	9.97	10.30	10.64
Е	7.5mg/kg (orally)	123	8.82	8.94	9.50	10.00	10.32
		124	9.44	9.54	10.14	10.34	10.57
		125	8.64	8.72	9.49	10.20	10.48

Appendix -XI: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on haemoglobin content (g %) in cattle.

Group	Treatment	Tag No.	Day 0	Day 7	<b>Day 14</b>	Day 21	Day 28
		101	10.0	9.4	8.8	8.2	7.8
		102	9.8	9.2	8.6	7.6	7.6
A	No Drug (Control)	103	10.4	10.0	9.4	8.6	7.8
	(Control)	104	11.8	11.2	10.6	9.2	8.4
		105	10.2	8.8	8.2	7.8	7.4
		106	9.4	9.8	10.6	11.6	12.4
		107	7.6	8.2	9.2	10.2	11.4
В	Neem	108	9.5	10.3	10.9	11.4	12.8
		109	8.4	8.8	9.6	10.8	12.2
		110	8.0	8.4	9.8	10.4	11.6
	Korolla	111	9.2	9.6	10.4	10.8	11.6
		112	7.6	8.0	8.8	9.6	10.4
С		113	10.00	10.2	10.8	11.8	12.0
		114	8.0	8.4	9.2	9.8	10.6
		115	10.6	10.8	10.4	11.2	11.6
		116	8.2	8.6	10.0	11.2	12.6
	Ivermectin	117	9.2	9.6	10.4	11.8	12.8
D	200mg/kg	118	10.0	10.6	11.6	12.2	13.4
	pour-on	119	9.6	10.6	11.4	11.8	12.4
		120	8.6	9.4	10.2	11.6	12.6
		121	9.2	9.6	10.6	11.2	12.0
	Levavet 7.5mg/kg (orally)	122	11.0	11.2	11.8	12.4	12.8
Е		123	9.6	9.8	10.6	11.6	12.4
		124	10.2	10.6	11.2	12.0	12.8
		125	8.6	9.0	10.0	11.2	12.0

Appendix -XII: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on packed cell volume (%) in cattle.

Group	Treatment	Tag No.	Day 0	Day 7	Day 14	Day 21	Day 28
		101	33.17	32.00	31.80	30.10	29.00
		102	32.20	32.00	31.35	29.70	28.15
A	No Drug (Control)	103	30.70	30.10	29.30	28.80	27.40
	(Control)	104	33.00	32.10	32.00	31.00	30.80
		105	30.65	30.00	29.35	29.10	28.0
		106	32.50	32.80	33.20	34.90	35.00
		107	28.00	29.10	30.20	31.25	32.38
В	Neem	108	31.50	32.30	32.95	33.70	34.85
		109	30.20	30.55	31.00	32.80	34.00
		110	28.90	29.95	30.85	31.80	32.78
	Korolla	111	30.45	31.65	31.80	32.90	33.10
		112	28.00	29.10	30.05	31.58	31.95
С		113	32.65	32.90	33.85	34.70	34.90
		114	29.95	30.85	31.88	32.85	33.70
		115	34.00	34.20	34.85	35.55	35.90
		116	30.30	31.50	32.60	33.50	34.80
	Ivermectin	117	31.10	31.30	32.55	33.00	33.50
D	200mg/kg	118	32.10	32.50	34.35	35.45	35.80
	pour-on	119	33.50	34.70	35.85	36.30	36.90
		120	31.15	31.50	32.50	33.90	34.50
		121	31.35	32.10	33.25	33.70	34.30
	Levavet	122	33.50	33.85	34.80	35.30	36.00
Е	7.5mg/kg (orally)	123	31.60	32.30	33.20	33.80	34.50
		124	34.05	34.20	35.15	35.90	36.50
		125	30.80	31.90	33.80	34.95	35.40

Appendix -XIII: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on total leukocyte count (thousand/cu.mm.) in cattle.

Group	Treatment	Tag No.	Day 0	Day 7	Day 14	Day 21	Day 28
		101	10.10	10.45	10.75	10.90	11.90
		102	9.85	10.15	10.45	10.90	11.10
A	No Drug (Control)	103	8.60	8.80	8.95	10.15	10.90
	(Control)	104	7.60	8.50	8.75	9.25	9.95
		105	10.35	10.85	11.15	11.65	11.80
		106	9.21	9.00	8.85	8.40	8.00
		107	11.25	11.00	10.80	10.50	10.25
В	Neem	108	9.70	9.50	9.10	8.95	8.70
		109	10.60	10.40	10.15	10.05	9.80
		110	10.95	10.75	10.45	10.25	10.00
	Korolla	111	10.45	10.25	10.00	9.90	9.70
		112	11.45	11.20	11.00	10.75	10.45
С		113	8.78	8.60	8.30	8.15	8.05
		114	10.88	10.65	10.45	10.30	10.00
		115	8.40	8.20	8.00	7.85	7.70
		116	10.55	10.40	10.00	9.80	9.30
	Ivermectin	117	10.40	10.15	9.80	9.50	9.20
D	200mg/kg	118	9.50	9.30	8.56	8.50	8.00
	pour-on	119	9.85	9.65	9.20	8.90	8.55
		120	10.60	10.35	9.75	9.35	9.10
		121	9.65	19.45	9.00	8.75	8.50
	Levavet 7.5mg/kg (orally)	122	878	8.55	8.10	7.10	7.50
E		123	9.50	9.40	8.75	8.45	8.20
		124	8.55	8.50	8.05	7.85	7.50
		125	10.30	9.90	9.50	8.80	8.60

Appendix -XIV: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on eosinophil count (%) of differential leukocyte count in cattle

Group	Treatment	Tag No.	Day 0	Day 7	Day 14	Day 21	Day 28
		101	11	12	13	13	14
		102	12	12	13	14	15
A	No Drug (Control)	103	10	11	12	13	14
	(Control)	104	11	11	12	13	14
		105	15	15	16	16	17
		106	12	12	10	9	8
		107	15	14	13	12	11
В	Neem	108	12	12	11	10	9
		109	14	13	12	11	10
		110	15	14	13	12	11
	Korolla	111	14	13	12	10	9
		112	16	15	14	12	10
С		113	13	12	11	9	8
		114	14	13	12	11	10
		115	11	10	9	9	8
		116	15	14	13	11	8
	Ivermectin	117	12	11	10	8	6
D	200mg/kg	118	13	12	11	10	8
	pour-on	119	12	11	9	8	7
		120	16	15	14	12	9
		121	12	11	10	9	8
	Levavet 7.5mg/kg (orally)	122	11	10	9	9	8
Е		123	13	12	10	9	8
		124	14	13	12	11	10
		125	12	11	10	9	8

Appendix -XV: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on neutrophil count (%) of differential leukocyte count in cattle

Group	Treatment	Tag No.	Day 0	Day 7	Day 14	Day 21	Day 28
		101	26	24	23	22	22
		102	24	24	23	22	22
A	No Drug (Control)	103	25	24	23	23	22
	(Control)	104	27	25	24	23	22
		105	23	22	22	21	20
		106	24	24	25	26	26
		107	20	21	22	23	24
В	Neem	108	22	22	23	24	25
		109	25	26	26	27	27
		110	21	22	23	23	24
	Korolla	111	22	22	23	24	24
		112	19	20	21	22	23
С		113	25	25	26	27	27
		114	26	26	27	27	28
		115	22	23	24	24	25
		116	21	22	23	24	25
	Ivermectin	117	24	25	25	26	27
D	200mg/kg	118	22	23	25	26	27
	pour-on	119	25	26	27	28	29
		120	23	24	25	26	28
		121	24	24	25	26	27
	Levavet 7.5mg/kg (orally)	122	23	24	24	25	25
Е		123	20	21	22	23	24
		124	22	23	24	25	26
		125	26	26	27	28	28

Appendix -XVI: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on monocyte count (%) of differential leukocyte count in cattle

Group	Treatment	Tag No.	Day 0	Day 7	<b>Day 14</b>	Day 21	<b>Day 28</b>
		101	5	4	3	4	2
		102	4	4	6	5	4
A	No Drug (Control)	103	3	2	2	3	4
	(Control)	104	4	3	4	2	5
		105	5	6	4	3	4
		106	4	3	4	3	5
		107	5	4	5	4	5
В	Neem	108	4	5	4	6	5
		109	4	3	2	4	5
		110	3	4	6	4	5
	Korolla	111	6	5	3	5	4
		112	4	5	6	5	5
C		113	5	5	3	2	4
		114	4	3	3	4	5
		115	5	4	5	5	4
		116	4	2	3	4	3
	Ivermectin	117	3	4	3	2	3
D	200mg/kg	118	4	5	4	5	4
	pour-on	119	4	3	5	4	3
		120	3	5	3	4	5
		121	5	4	2	3	4
	Levavet	122	5	6	5	5	2
Е	7.5mg/kg	123	6	5	4	5	3
	(orally)	124	4	3	5	4	3
		125	3	5	6	5	3

Appendix -XVII: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on lymphocyte count (%) of differential leukocyte count in cattle

Group	Treatment	Tag No.	Day 0	Day 7	Day 14	Day 21	Day 28
		101	58	60	62	61	61
		102	60	60	58	56	60
A	No Drug (Control)	103	60	59	58	59	57
	(Control)	104	59	61	62	60	61
		105	60	61	60	59	60
		106	60	59	61	60	58
		107	64	63	62	63	62
В	Neem	108	60	58	59	61	60
		109	62	60	58	61	60
		110	60	61	63	60	59
	Korolla	111	58	60	62	61	63
		112	61	60	60	62	61
C		113	57	57	59	60	59
		114	60	61	60	59	58
		115	58	59	58	58	59
		116	60	62	63	64	66
	Ivermectin	117	61	61	62	63	65
D	200mg/kg	118	59	58	60	61	62
	pour-on	119	60	61	60	62	64
		120	61	59	61	62	60
		121	59	60	64	63	61
	Levavet	122	58	58	59	58	62
Е	7.5mg/kg	123	59	60	59	60	60
	(orally)	124	59	57	57	59	60
		125	58	59	61	59	62

Appendix -XVIII: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular volume (cubic micron) in cattle

Group	Treatment	Tag No.	Day 0	Day 7	Day 14	Day 21	Day 28
		101	37.78	36.70	36.98	35.50	34.86
		102	35.70	35.83	35.95	34.70	33.83
A	No Drug (Control)	103	32.25	32.09	32.06	32.29	31.75
	(Control)	104	33.74	33.30	33.61	33.26	33.85
		105	36.23	35.84	35.97	36.28	35.71
		106	36.43	36.61	36.05	37.01	36.38
		107	36.08	37.21	36.74	36.76	37.22
В	Neem	108	36.21	36.87	36.61	36.24	38.72
		109	36.08	36.07	35.47	36.20	36.48
		110	36.96	38.10	37.81	38.13	38.12
	Korolla	111	35.99	37.28	36.55	36.88	36.74
		112	36.27	37.55	37.99	39.13	39.25
C		113	36.56	36.72	37.20	37.59	37.29
		114	36.61	38.56	38.04	38.38	38.69
		115	34.66	34.76	34.75	34.89	34.96
		116	36.42	37.59	35.59	33.14	32.61
	Ivermectin	117	37.56	37.40	35.93	32.54	31.48
D	200mg/kg	118	38.67	38.60	36.39	34.45	33.40
	pour-on	119	39.14	40.02	39.05	36.37	35.21
		120	37.99	37.32	34.80	33.70	32.61
		121	38.42	38.54	36.26	33.47	32.60
	Levavet	122	35.87	35.71	34.90	34.27	33.83
Е	7.5mg/kg	123	35.83	36.13	34.95	33.80	33.43
	(orally)	124	36.07	35.85	34.66	34.72	34.53
		125	35.65	36.58	35.62	34.26	33.78

Appendix -XIX: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular haemoglobin concentration (%) in cattle

Group	Treatment	Tag No.	Day 0	Day 7	Day 14	Day 21	<b>Day 28</b>
		101	30.15	29.38	27.67	27.24	26.90
		102	30.43	28.75	27.43	25.59	27.00
A	No Drug (Control)	103	33.88	33.22	32.08	29.86	28.47
	( = ==== = = )	104	35.76	34.89	33.13	29.68	27.27
		105	33.28	29.33	27.94	26.80	26.43
		106	28.92	29.88	31.93	33.24	35.43
		107	27.14	28.18	30.46	32.64	35.21
В	Neem	108	30.16	31.89	33.08	33.83	36.73
		109	27.81	28.81	30.97	32.93	35.88
		110	27.68	28.05	31.77	32.70	35.39
	Korolla	111	30.21	30.33	32.70	32.83	35.05
		112	27.14	27.49	29.28	30.40	32.55
C		113	30.63	31.00	31.91	34.01	34.38
		114	26.71	27.23	28.86	29.83	31.45
		115	31.18	31.58	29.84	31.50	32.31
		116	27.06	27.30	30.67	33.43	36.21
	Ivermectin	117	29.58	30.67	31.95	35.76	38.21
D	200mg/kg	118	31.15	32.62	33.77	34.41	37.43
	pour-on	119	28.66	30.55	31.80	32.51	33.60
		120	27.61	29.84	31.38	34.22	36.52
		121	29.35	29.91	31.88	33.23	34.99
	Levavet	122	32.84	33.09	33.91	35.13	35.56
Е	7.5mg/kg	123	30.38	30.34	31.93	34.32	35.94
	(orally)	124	29.96	30.99	31.86	33.43	35.07
		125	27.92	28.21	29.59	32.05	33.90

Appendix -XX: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on mean corpuscular hemoglobin (micro micro gram) in cattle

Group	Treatment	Tag No.	Day 0	Day 7	Day 14	Day 21	Day 28
		101	11.39	10.78	10.23	9.67	9.38
		102	10.86	10.30	9.86	8.88	9.13
A	No Drug (Control)	103	10.92	10.66	10.28	9.64	9.04
	(Control)	104	12.07	11.62	11.13	9.87	9.23
		105	12.06	10.51	10.05	9.73	9.44
		106	10.54	10.94	11.51	12.30	12.89
		107	9.79	10.49	11.19	12.00	13.10
В	Neem	108	10.92	11.76	12.11	12.26	14.22
		109	10.04	10.39	10.98	11.92	13.09
		110	10.23	10.69	12.01	12.47	13.49
	Korolla	111	10.87	11.31	11.95	12.11	12.87
		112	9.84	10.32	11.13	11.90	12.78
С		113	11.20	11.38	11.87	12.78	12.82
		114	9.78	10.50	10.98	11.45	12.17
		115	10.81	10.98	10.37	10.99	11.30
		116	9.86	10.26	10.92	11.08	11.81
	Ivermectin	117	11.11	11.47	11.48	11.64	12.03
D	200mg/kg	118	12.05	12.59	12.29	11.86	12.50
	pour-on	119	11.21	12.23	12.42	11.82	11.83
		120	10.49	11.14	10.92	11.53	11.91
		121	11.27	11.52	11.56	11.12	11.41
	Levavet	122	11.78	11.81	11.84	12.04	12.03
Е	7.5mg/kg	123	10.88	10.96	11.16	11.60	12.02
	(orally)	124	10.81	11.11	11.05	11.61	12.11
		125	9.95	10.32	10.54	10.98	11.45

Appendix -XXI: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on alanine aminotransferase level of serum (U/I) in cattle

Group	Treatment	Tag No.	Day 0	Day 7	Day 14	Day 21	Day 28
		101	18.2	19.5	19.1	18.5	20.5
		102	19.7	17.3	16.7	17.0	21.5
A	No Drug (Control)	103	17.1	18.4	16.4	15.5	20.3
	(Control)	104	15.3	16.3	16.3	16.5	16.0
		105	21.0	20.5	22.2	22.5	17.9
		106	18.1	16.2	17.7	17.6	16.8
		107	20.3	19.8	21.2	18.3	17.7
В	Neem	108	22.2	21.1	20.3	19.7	19.9
		109	19.1	22.8	20.4	23.2	22.2
		110	21.4	22.1	18.32	22.8	19.5
	Korolla	111	20.5	22.4	21.1	22.6	18.8
		112	23.1	19.8	20.1	19.8	22.6
С		113	17.7	16.2	17.5	18.5	19.3
		114	22.0	20.7	23.3	20.2	20.1
		115	16.2	17.2	17.1	16.5	17.7
		116	21.3	20.3	21.0	22.0	22.8
	Ivermectin	117	20.4	22.6	19.5	17.5	18.7
D	200mg/kg	118	18.2	16.1	17.7	19.5	17.8
	pour-on	119	19.7	18.1	21.3	18.5	18.0
		120	21.1	20.3	17.5	20.3	20.1
		121	19.2	20.5	16.8	18.4	20.6
	Levavet	122	17.3	16.6	20.4	16.5	15.8
Е	7.5mg/kg	123	18.8	19.7	19.5	20.7	16.3
	(orally)	124	17.2	16.0	16.1	16.7	19.1
		125	20.1	18.3	18.8	18.0	17.1

Appendix -XXII: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on aspartate aminotransferase level of serum (U/I) in cattle

Group	Treatment	Tag No.	Day 0	Day 7	Day 14	Day 21	Day 28
		101	30.4	29.5	32.0	32.5	34.1
		102	31.9	27.3	33.6	27.7	30.5
A	No Drug (Control)	103	29.0	30.3	30.2	28.4	31.4
	(Control)	104	27.0	28.6	28.2	26.7	29.7
		105	33.1	34.0	31.5	31.5	32.2
		106	30.6	32.0	29.5	27.7	29.2
		107	35.6	33.0	34.6	31.7	32.5
В	Neem	108	34.2	30.3	32.6	35.8	32.8
		109	30.2	32.9	29.7	33.1	34.2
		110	33.6	28.1	33.0	35.3	31.1
	Korolla	111	32.2	29.1	33.8	32.1	33.3
		112	34.7	33.4	34.0	35.0	33.0
С		113	29.6	33.1	31.2	28.5	31.6
		114	34.2	35.5	32.6	31.3	32.4
		115	28.0	34.6	30.7	28.2	28.8
		116	33.3	31.5	32.4	31.7	31.0
	Ivermectin	117	32.6	34.0	34.6	33.2	30.6
D	200mg/kg	118	30.5	32.6	29.8	31.6	32.4
	pour-on	119	31.7	28.3	33.0	28.5	33.5
		120	33.0	32.2	32.0	30.8	29.3
		121	31.2	34.4	30.5	28.8	29.8
	Levavet	122	29.0	32.5	32.4	30.1	33.5
Е	7.5mg/kg	123	31.0	29.1	33.6	34.6	28.8
	(orally)	124	28.9	30.1	30.1	27.2	29.5
		125	32.4	33.6	31.4	28.3	28.7

Appendix – XXIII: The effects of Neem leaves powder (Orally), Korolla (fruits), Ivermectin (Pour on formulation) & Levamisole (Orally) on body weight (Kg) in cattle.

Group	Treatment	Tag No.	Day 0	Day 7	<b>Day 14</b>	Day 21	<b>Day 28</b>
		101	126.0	125.5	124.0	123.5.0	121.0
		102	127.0	126.5	126.0	125.0	125.5
A	No Drug (Control)	103	123.5	123.0	122.5	122.0	121.0
	(Control)	104	129.5	129.0	128.5	128.0	127.5
		105	121.0	121.0	120.0	119.5	119.5
		106	120.0	120.5	121.0	122.0	123.5
		107	128.0	128.5	129.0	130.0	130.5
В	Neem	108	130.5	131.0	131.5	132.0	133.0
		109	125.5	126.0	127.0	128.0	128.5
		110	120.5	121.0	121.5	122.5	123.5
	Korolla	111	124.0	124.5	125.0	126.0	127.0
		112	121.5	121.5	123.0	124.0	125.0
С		113	127.5	127.5	128.0	129.5	130.0
		114	123.0	123.5	124.5	126.0	127.0
		115	126.5	126.5	127.0	128.0	129.0
		116	126.5	127.0	128.0	129.5	130.5
	Ivermectin	117	123.0	123.5	124.5	125.5	127.5
D	200mg/kg	118	130.0	130.5	131.0	132.0	132.5
	pour-on	119	127.0	128.0	129.5	130.0	132.0
		120	127.5	127.5	128.0	128.5	129.5
		121	120.0	120.5	121.0	122.0	123.5
	Levavet	122	121.0	121.5	123.0	124.0	125.0
Е	7.5mg/kg	123	122.0	122.5	124.0	125.0	125.5
	(orally)	124	126.0	126.5	127.5	128.5	129.5
		125	130.5	131.5	132.0	132.5	133.5