

**COMPARATIVE EFFICACY OF TOBACCO LEAVES
OINTMENT AND NEEM LEAVES EXTRACT AGAINST
STEPHANOFILARIASIS IN CATTLE**

A Thesis

By

MD. ABDULLA AL MASUD

Registration No. 1305107

Semester: July- December, 2014

Session: 2012-13



MASTER OF SCIENCE (M.S.)

IN

PHARMACOLOGY

**DEPARTMENT OF PHYSIOLOGY AND PHARMACOLOGY
HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY
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*Submitted to the
Department of Physiology and Pharmacology
Hajee Mohammad Danesh Science and Technology University, Dinajpur
In partial fulfillment of the requirements
For the degree of*

**MASTER OF SCIENCE (M.S.)
IN
PHARMACOLOGY**

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**IN VIVO EFFECTS OF NEEM LEAVES EXTRACT AND
IVERMECTIN AGAINST NATURAL TICK INFESTATION IN
CALVES**

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DEDICATED

**TO
MY**

PARENTS

ACKNOWLEDGEMENT

All praises are to the Almighty Allah, who kindly enables the author to complete the present research work successfully and to submit the thesis leading to Master of Science (MS) degree in Pharmacology.

*The author express his profound indebtedness and sincere gratitude to his respected teacher and research supervisor, **Dr. Fahima Binthe Aziz**, Assistant Professor, Department of Physiology and Pharmacology, Hajee Mohammad Danesh Science and Technology University (HSTU) Dinajpur, for her scholastic direction, valuable suggestions and constructive criticism, encouragement and kind cooperation in carrying out this research work and writing up of the thesis.*

*The author is also highly obliged and expressing his gratification and sincere appreciation to his respective co-supervisor **Dr. Md. Mahmudul Hasan**, Lecturer, Department of Physiology and Pharmacology, Hajee Mohammad Danesh Science and Technology University (HSTU) Dinajpur -for his valuable suggestions, encouragement and kind cooperation during entire period of study and preparing the thesis.*

*The author owes arrears of gratitude to **Dr. Md. Bazlar Rashid** Assistant Professor, Department of Physiology and Pharmacology, Hajee Mohammad Danesh Science and Technology University (HSTU) Dinajpur, for his kind collaboration, encouragement and valuable suggestions to complete the thesis work.*

The author would also like to thank his co-workers for their encouraging attitude, kind help and all-out support in the entire period of the research work.

I thank to Dr. Md. Wasim Akram, Dr. Md. Akter Hossain Dr. Most. Nima Islam, Dr. Md Atikur Rahman for helping during this work. I would also like to offer my thanks to other staffs of Department of Physiology and Pharmacology, Hajee Mohammad Danesh Science and Technology University (HSTU) Dinajpur, for their co-operation.

The author is ever indebted to his beloved parent, brothers, sister's friends and well wishers for their endless sacrifices, heartiest blessings and moral support throughout his entire life. The author is also grateful to uncles, aunts, cousins, and other relatives and neighbors for their heartiest blessings, sacrifice and encouragement throughout the entire period of his academic life. The author also likes to

extend his thanks to his year mates, roommates (Md.Mehedi Hasan, Md.Rubel, Md.Noman and hall mates for their encouragement and co-operations.

And lastly I would like to express my thanks and gratitude to all those, whose names could not be mentioned, but have extended their co-operation and help during my research work.

December 2014

The Author

ABSTRACT

Stephanofilariasis one of the most harmful parasitic diseases caused by *Stephanofilaria assamensis* is endemic to Bangladesh. The disease could neither be fully controlled nor eradicated from these regions in spite of various attempts. The present study was conducted to find out the prevalence of the disease in the endemic areas, its economic impact and suitable measures for its eradication. The incidence rate was more in older animals. The direct and indirect economic losses to farmers by way of loss of draught days, delayed puberty, lower lactation yields and prolonged inter-calving period were enormous. The control and eradication of Stephanofilariasis in bovines could be successfully achieved especially by targeting the life cycle of the biological vector (*Musca conducens*), the intermediate host. Improvement in general cleanliness and hygiene to reduce the fly population. An experiment was carried out in nearby Villages and also in the Laboratory in the Department of Physiology and Pharmacology, Hajee Mohammad Danesh Science and Technology University, Dinajpur during the period from June-November/ 2013. The tobacco powder and the extract of neem leaves were screened for assessment their comparative efficacy against humpsore in cattle. Among these, neem leaves extract were used topically to cover the affected area and tobacco powder was applied topically twice daily to the cattle. 15gm tobacco powder was mixed with 85gm Vaseline for 28 days. Efficacy was recorded as compared to control. Extract of neem leaves showed gradual increase of efficacy up to day 30th of treatment. Among two drugs tobacco show better performance than neem leaves extract.

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LIST OF ABBREVIATION AND SYMBOLS

TEC	=	Total Erythrocyte Count
Hb	=	Hemoglobin
PCV	=	Packed Cell Volume
ESR	=	Erythrocyte Sedimentation Rate
Bwt	=	Body weight
cm	=	Centimeter
Kg	=	Kilogram
gm	=	Gram
RBC	=	Red Blood Cell
Fig	=	Figure
No.	=	Number
mm	=	Mili mètre
hrs	=	Hours
DVHR	=	District veterinary hospital
%	=	Percentage
DNA	=	Deoxy Ribo Nucleic Acid
GR	=	Glutathion Reductage
GPX	=	Glutathion peroxidase
SOD	=	Superoxidase dismutase
CAT	=	Catalase
GST	=	Glutathion S-transferase
DTD	=	DT-diaphorase
HSTU	=	Hajee mohammad danesh science and technology university
DMBA	=	7,12,-dimethyl benz anthracene
BAU	=	Bangladesh Agriculture Universit



CHAPTER I

INTRODUCTION

CHAPTER I

INTRODUCTION

Livestock reduces poverty and build up our national economy. Among Livestock plays an important role as the backbone of agriculture. It provides milk, meat, egg, skin, fuel, fertilizer and draft power. In Bangladesh cattle is the main source of draft power, urban transport and harvesting crops. About 50 % of the people depend on their income source from livestock.

According to our socio-economic situation, the knowledge of our farmer is very little about livestock rearing, management, farming and infectious-noninfectious diseases like hump sore, rabies, anthrax, black quarter, foot and mouth disease, PPR etc. As a result they are suffering from various problems in daily life. So it is important to provide knowledge about those diseases.

Stephanofilariasis is a parasitic disease caused by *Stephanofilaria assamensis* which is characterized by intensive itching, rubbing of the injured area with herd object, presence of lesion in hump region, surrounding the lesion skin become dry, rough, and alopecia may develop. Dermatitis of cattle caused by a filaroid worm, (Pande, 1335). Stephanofilariasis commonly known as humpsore, because, it is usually located on or near the hump and neck regions of cattle. This disease is transmitted by the bite of intermediate host *Musca conducens*. The disease is common in Bangladesh and India except in hilly region causing severe economic losses by decreasing productivity and cost of the animals and their skin quality as well. The disease also occurring in Buffalos, Goats, Elephants, Black rhinoceros and Nilgai at different locations of the body.

Stephanofilariasis in livestock are very common in Bangladesh. The *Stephanofilaria assamensis* entirely live on blood and lymph of all kinds of vertebrate animal. The damages caused by maggots are irritation, inflammation, exudation of lymph which coagulates to form crusts. *Stephanofilaria assamensis* cause dermatitis, alopecia and sometimes animals are infected secondarily by bacteria causing toxemia, septicemia and ultimately dead of the animals. (Chungsamarnyart, and Jansawan, 1991).

The present research is being conducted to study the efficacy of medicinal plant in veterinary practice. A large number of anthelmintics are now available in the market.

The indiscriminate use of anthelmintics made the parasites to be resistant against the drug, which have been reported by experts throughout the world including Bangladesh (Hannan, *et al.*, 2001). So, we should have to back the traditional uses of me declines.

The contribution of livestock sub-sector is remarkable in Bangladesh. The contribution of this sub-sector to Gross Domestic Product (GDP) is approximately 6.5 percent (DLS1998) when the value of milk, meat, egg skin, fuel, draft, fertilizer etc. is considered. This sub-sector meets about 28 (Brammer *et al.*, 1996) of Gross Domestic Product (GDP). It provides full time employment to about 20% and generates 13% of total foreign currency and partial employment to about 50% of the rural population (Alam, 1993).

Besides this, Stephanofilariasis cause lather defects (Noouddin and Day, 1993) which is probably with an annual economic loss of US\$ 220.95 Million in Bangladesh. In addition to this as the affected animal remains in constant agony, irritation and itching, there is a gradual loss of health, loss of performance and impaired milk production. Patnaik and Khan (1972) reported that although the disease in cattle did not kill the animal, it certainly caused poor performance in breeding bulls, milk cows and bullocks in India.

Different countries like Indonesia, India, and Thailand manufactured a variety of medical preparations from indigenous plants and herbs by some pharmaceutical industries. Such as anthelmintic, anti diarrhoeal preparation, astringents, skin ointments and clearing agents. In India, in ayurveda and unani system of medicine, the Neem seeds, fruits, leaves, oil, roots and bark has been used. Thus, over thousands of years, millions of Asians have used Neem medically.

Neem products act against various types of nematodes. Certain limnoid fractions extracted from Neem kernels are providing active against root knot nematodes, the type most devastating to plant. It is also a common remedy against maggots. In Bangladesh and India, villages use Neem oil to the hair to kill lice, reportedly with great success. Neem contains several active ingredients and they act in different way under many circumstances. Neem is used by German company as the active ingredient in tooth pastes and other oral hygiene preparations. It classes that its tests is High effective a both preventing and healing gum inflammation and peridental disease. On the other hand, the Neem has been studying since 1972 by researchers of the U.S Department of

Agriculture. In laboratory experiment they found that the plants ingredients foil even some of Americans most voracious garden pest. For instance, in one trial each half of several soybean leaves was sprayed with Neem extract s and placed in a container with Japanese beetles. The treated halves remained untouched but within 48 hours the other halves were consumed right down to their woody Veins. Shin-Foon Chiu (1948), found a component from Neem named deacetylazadirachtinol which appears to be as effective as azadirachtin in assays against the tobacco budworm, but it has not yet been widely tested in field practice.

Fenitrothion is a organophosphorus compound and insecticide used in horticulture and agriculture. It has an LD50 in rates of 250-500 mg/kg in fowls of 445 mg/kg. It is available in the local market by two trade names Sumithion (R) and Sovathion (R). One liter Sumithion (R) or Sovathion contains 500 gram active fenitrothion. In the present study Sovathion was used. Tobacco leaves (*Nicotiana tabacum*) contain an alkaloid, Nicotine which is water soluble and it has an anti parasitic action.

Most of the framers in Bangladesh cannot afford to buy modern medicine for the treatment of the livestock because of high price of the drug and poor economic condition of the farmers. So if we can develop the traditional system of medicine in Bangladesh, it will be highly beneficial for the farmers and for the overall improvement of the livestock. -Considering all these facts in the present study tobacco leaves ointment and neem leaves extract were selected with a view:

1. To study the comparative efficacy of tobacco leaves ointment and neem leaves extract against hump sore in cattle.
2. To determine the effect of tobacco leaves ointment and extract of neem leaves on some hematological parameters (TEC, Hb, PCV) in cattle.
3. To determine the effect of tobacco leaves ointment and extract of neem leaves on some clinical parameters (severity of infestation, body weight) in cattle.



CHAPTER II

REVIEW OF LITERATURE

CHAPTER II

REVIEW OF LITERATURE

Neem, Tobacco and Stephanofilariasis:

The purpose of this chapter is to provide a selective review of the research works accomplished in relation to the present study. Literatures on comparative efficacy of tobacco leave powder and neem leaves extract against stephanofilariasis in cattle which is related to this study has been reviewed under the following heading.

2.1 Introduction of Neem:

Azadirachta indica (Neem) is a tree in the mahogany belonging to the family Meliaceae. It is one of two species in the genus *Azadirachta*, and is native to India, Pakistan, and Bangladesh growing in tropical and semi-tropical regions. Neem is a fast-growing tree that can reach a height of 15–20 metres (49–66 ft), rarely to 35–40 metres (115–130 ft). It is evergreen, but in severe drought it may shed most or nearly all of its leaves. The branches are wide spread. **Zillur et al (1993)**

2.2 Toxonomic position of Neem:

Kingdom: Planate

Division: Magnoliophyta

Order: Sapindales

Family: Meliaceae

Genus: *Azadirachta*

Species: *A. indica*

Binomial name *Azadirachta indica*

2.3 Chemistry

The seed kernels of neem yield about 10% of a fixed oil, comprised primarily of glycosides. The yellow, bitter oil has a garlic-like odor and contains approximately 2% of bitter principles including azadirachtin, azadiradione, azadirone, gedunin, nimbidin, nimbin, nimbolide, nimbinin, nimbidol, margolene, mahmoodin, salanin, meldenin, vepinin, and other related limonoid triterpenes. Azadirachtin is the most active insecticidal component of Neem, with a yield of about 5 g from 2 kg of seeds. All parts of the tree yield beta-sitosterol. The leaves also contain quercetin, gallic acid, catechin, carotenes, and ascorbic acid. 2 Low concentrations of aflatoxin have been reported. Subapriya R, Nagini S. Medicinal properties of Neem leaves: a review. *Curr Med Chem Anticancer Agents* (2005).

2.4 Chemical compounds

Siddiqui was the first scientist to bring the anthelmintics, antifungal, antibacterial, and antiviral constituents of the neem tree to the attention of natural products chemists. In 1942, he extracted three bitter compounds from neem oil, which he named as nimbin, nimbinin, and nimbidin respectively. The process involved extracting the water insoluble components with ether, petrol ether, and ethyl acetate and dilute alcohol. The provisional naming was nimbin (sulphur-free crystalline product with melting point at 205 °C, empirical composition $C_7H_{10}O_2$), nimbinin (with similar principle, melting at 192 °C), and nimbidin (cream-colored containing amorphous sulphur, melting at 90–100 °C). Siddiqui identified nimbidin as the main active antibacterial ingredient, and the highest yielding bitter component in the Neem oil. These compounds are stable and found in substantial quantities in the Neem. They also serve as natural insecticides. Razzaghi-Abyaneh *et al* (2005).

2.5 Medicinal use

Neem products are believed to be anthelmintics, antifungal, antidiabetic, antibacterial, antiviral, contraceptive and sedative. It is considered a major component in Ayurvedic and Unani medicine and is particularly prescribed for skin disease. Ozoneem Aza is our brand for Azadirachtin Technical. It is yellowish powder that contains 15% to 25% Azadirachtin. Sinniah D, Baskaran G. *Margosa* (1981)

2.6 Bioactivity

Neem acts as a broad spectrum biopesticide controlling a large number of insect including caterpillars, beetles, whiteflies, leafhoppers, aphids, mites, trips, borer, mealy bug, leaf folders and many more. Sinniah D, Baskaran G. Margosa (1981).

Bovine stephanophilial dermatitis has been first reported from Indonesia (Ihle and The Landenberg, 1933) popularly called "Cascado". Similar condition has been reported as verminous dermatitis in United States (Chit wood, 1934) and humpsore in India (Pande, 1936). It has been also reported from Japan, Africa, Denmark, Russia and Bangladesh. In the present study, available literature on stephanofilariasis has been reviewed.

A skin disease caused by a filarial worm was first recorded by Dikmans (1934) in cattle in United States (North America) which was closely related to cascado of Indonesia. The lesions were found on the scrotum of two bulls.

Kraneveld and Djaenoidin (1937) started that stephanophilial lesions were treated with a number of drugs. Before using the drug they brushed the sore daily. Successfully result was obtained by using aqueous solution of cooper's essential dip in different concentration for different size of the seasons.

Mohan (1945) reported that humpsore was treated by intravenous injection of 1.5 grains of tartar emetic in water for every 100/lb .Body weight or 2.5 grains on alternate days to a total 6 injections, local application of 4 % tartar emetic ointment and surgical removal of the sore was also helpful.

Menon (1952) treated successfully 350 cases of *Stephanofilaria sis* with 1: 1000 euflavin solution and distilled oil of pinetar.

Ahmed (1961) treated the ear sores buffaloes successfully by intravenous injection of tartar emetic ointments solution and applying tartar emetic ointment (1:25) externally on the sore.

Schultz and Schafer (1965) stated that summer sore in cattle in East Germany, the cause of which suspected as *Stephanofilaria* although pathological examination proved no evidence of filarial infection were healed by local application of 2.3% Trichlorophon.

Patnaik and Roy (1968) treated successfully the lesions of *stephanophilaria assamensis* in goats and buffaloes in Orissa, India with 6% neguvon emulsion in 7 % linseed.

Mishra treated the ear sores of bullock caused by *stephanophilaria*, with potassium permanganate lotion (1:1000) or worm hypertonic magnesium sulphate solution followed by 5-10% formalin and 1.5-2% aqua mercurochrome.

Ivashkin.et al. (1971) treated *Stephanofilariasis* with 40% chlorphos ointment and found 89% effectiveness in cattle.

Pal and Sinha 1971 treated effectively 24-30 cases of *Stephanofilariasis* IN cattle by applying 6% Neguvon ointment.

Fadzil (1977) stated that neguvon at 6 or 10 % in Vaseline, coconut oil or linseed oil when applied to *stephanophilarial* lesions in cattle on alternate days proved effective. At varying intervals after withdrawal of treatment replace of sores occurred but those were thought to be reinfection.

Dadaev (1982) recommended treating humpsore with chlorphos cream applied to the affected skin areas 3 or 4 times during the grazing period and Phenothiazine at 0.03 g/kg body weight daily with salt or feed, to kill the fly larvae in cattle faeces.

Karin (1984) reported that *stephanophilarial* dermatitis around the eyes of cattle had been treated with curaminth brand of tetramisole hydrochloride in the form of ointment which was applied topically twice daily for a period of 3-5 days after removing the superficial layers of skin. After 7-10 days post treatment, the lesions were completely healed up and hairs were observed to be reinfected which were again treated with the same ointment.

Baki and Hossain 1986 used different formulation of Neguvon ointment (10%, 15% and 20% along with 5 % sulfanilamide and Vaseline in each formulation) to treat one thousand cattle affected with *Stephanofilariasis* of which 20% Neguvon ointment was found excellent. No side effect was noticed during the course of treatment.

Jonson, (1987) stated that various treatment had been evaluated and the most effective were based on the topical application of organophosphate chemicals.

Roy and Misra (1991) reported 35 exotic, cross-bred or indigenous cattle, aged 3-5 years, with typical lesions of hump sore on various parts of the body, 5 animals were left as

untreated controls and the others, in groups of 10, were treated by topical application of 1% ivermectin ointment, 0.5% Ivermectin ointment and 7.5% levamisole ointment daily for 4 weeks. The efficacy of the treatments, assessed after 6 months was 80, 50 and 50%, respectively.

Mostofa (1993) used an ointment made of 1% neguvon, 10% neem leaves and barks, 89% vaseline to treat humpsore of twenty five cattle. All the cattle were cured within 30 days by topical application of the ointment twice daily.

Baki (1995) recorded that 10% neguvon ointment mixed with 5% sulfanilamide powder and 5% zinc oxide was the most effective drug against humpsore as all animals had-both smaller and larger lesions were cured within 10-30 days. Their higher effectiveness may be due to the use of zinc oxide which has the astringent action and promoting healing of the lesions. A maximum of 30 gm of this ointment was required to cure the larger lesions.

Alam (1995) observed the therapeutic efficacy of fenitrothion (neguvon) and neem seeds against stephanofilariasis in cattle. Neem seed kernel ointment and 10% fenitrothion ointment were almost equally effective (100%) against stephanofilariasis in cattle. However, the recovery was faster (within 18-34 day) with 10% fenitrothion ointment followed by neem seed kernel ointment (27-42 days) and fresh neem seed ointment (30-39 days). On the other hand, 5% fenitrothion ointment was found almost ineffective against stephanofilariasis in cattle. From this result, neguvon and asuntol were suggested as the most effective drug against humpsore. Encouraging results were also obtained after surgical removal of humpsore lesions.

Rahim (1998) observed that subcutaneous preparation of ivermectin at the dose rate of 200 mg/ kg body weight against stephanophilial dermatitis in both single and double dose of frequency was found effective without any side effect.

The edematous skin disease in buffaloes and cattle in Egypt caused probably by stephanofilaria had been treated with furadan and neguvon (R). Furadan was given at the dose rate of 50 ml/ animal intra muscularly in which 6-12 animals were improved. Five of 14 cutaneously at the dose rate of 12 ml/ 40 kg body weight and other 8 animals were cured 2nd and 3rd doses (Fouad *et al.*, 1974).

Gilmour *et al.* (1999) reported that fenitrothion is an organophosphorus insecticide widely used for the control of locusts in Australia and overseas. It is sprayed on swarms and bands of locusts and on the pastures on which they feed. However, there is little Australian data on depletion rates of fenitrothion when, used this not be able to, guarantee that their cattle meet legal requirements with respect to residue levels following grazing on sprayed pasture. This paper reports on a study conducted in February 1988 in central western New South Wales on the rate of depletion of fenitrothion in cattle, pasture and soil after fenitrothion spraying. The cattle were 18 month old heifers in forward store condition. Fenitrothion was detected in the fat of 7 of the 66 exposed cattle. Residues in 2 of these exceeded the Maximum Residue Limit of 0.05 mg/ kg. No fenitrothion was detected in the fat of the 26 cattle slaughtered 14 days or later after treatment or in muscle and liver samples. The half life of fenitrothion was 2-3 days in the soil and 1-2 days the pasture. It is concluded that the 14 days slaughter with holding period currently approved by the National Registration Authority is appropriate.

Humpsore due to *Stephanofilaria assamensis* in Bangladesh recovered completely with Neguvon as 6% aqueous solution or 6% liniment in castor oil. In this trial one hundred and thirty six of 40 cattle were cured. The liniment proved superior to aqueous solution. More than 40 days were required to cure the severe lesions. The drug was recommended as safe economic and highly effective. (Rahman and Khaleque, (1974).

Dasgupta *et al.* (2004) numerous laboratory studies reveal that various naturally occurring dietary substances can modify the patho-physiological process of various metabolic disorders and can be an effective preventive strategy for various diseases, including cancer. Indian Neem tree, *Azadirachta indica* A. Juss.(family: Meliaceae), contains at least 35 biologically active principles and is widely grown all over the tropics. The effect of two different doses (250 and 500 mg per kilogram body weight) of 80% ethanolic extract of the leaves of *Azadirachta indica* were examined on drug metabolizing Phase-I and Phase-II enzymes, antioxidant enzymes, glutathione content, lactate dehydrogenase, and lipid peroxidation in the liver of 7-week-old Swiss albino mice. Also ant carcinogenic potential of *Azadirachta indica* leaf extract was studied adopting protocol of benzo(a) pyrene-induced fore-stomach and 7,12-dimethyl benz(a) anthracene (DMBA)-induced skin papillomagenesis. Our primary findings reveal its potential to induce only the Phase-II enzyme activity associated mainly with carcinogen detoxification in liver of mice. The hepatic glutathione S-transferase ($P < 0.005$) and DT-

diaphorase specific activities ($P < 0.01$) were elevated above basal level. With reference to antioxidant enzymes the investigated doses were effective in increasing the hepatic glutathione reductase (GR), glutathione peroxidase (GPX), superoxide dismutase (SOD) and catalase (CAT) activities significantly (from $P < 0.005$ to $P < 0.001$). Reduced glutathione measured as non-protein sulphhydryl was found to be significantly elevated in liver ($P < 0.005$) and in extrahepatic organs (from $P < 0.005$ to $P < 0.001$) examined in our study. Glutathione S-transferase (GST) and DT-diaphorase (DTD) showed a dose-dependent increase in extrahepatic organs. Chemopreventive response was measured by the average number of papillomas per mouse, as well as percentage of tumor-bearing animals. There was a significant inhibition of tumor burden, in both the tumor model system studied (from $P < 0.005$ to $P < 0.001$). Tumor incidence was also reduced by both the doses of *Azadirachta indica* extract.

Phukan *et al.* (2005) An epidemiological study on humpsore in cattle in Assam caused by *Stephanofilaria assamensis* Pande, 1936 conducted from March 1999 to February 2000 revealed that out of 1189 cattle, 523 (43.98%) were positive for humpsore. Highest infection was observed in the monsoon season (53.43%) followed by post-monsoon (43.16%), pre-monsoon (33.54%) and lowest in winter (26.67%). Males (46.36%) and crossbred animals (50.41%) had a higher infection rate than the females (38.35%) and native cattle (41.14%). Animals below one year of age were free from infection, while animals aged between three to < six years had highest infection rate (57.08%). The infection rate was inversely proportional to the age of the animal. The study revealed that out of the 622 flies collected from the lesions, 305 were *Musca pattoni*, 199 *M. domestica* and 118 belonged to the subfamily Limnophaginae. Out of these three, only *M. pattoni* was found to harbour the larval stage of *S. assamensis*. The highest (45%) incidence of *S. assamensis* larvae in *M. pattoni* was recorded in August and the lowest (16%) in December. No larvae could be found in January and February.

Odeyemi (2005) Petroleum ether extracts of leaves and kernels of the neem plant, *Azadirachta indica* A. Juss, mixed at varied dosages of 0, 50, 150, 250, 350 and 500 mg/ml with decorticated groundnut seeds (50 g), were tested for their potential to control the Khapra beetle, *Trogoderma granarium* Everts. The kernel extract was more potent than the leaf extract. The number of emerged adults decreased with increasing extract concentrations for both leaves and kernels. The number of emerged adults in the control (72.2 ± 4.83) was significantly higher ($P < 0.05$) than all neem extract variants. The

mortality of adults and larvae increased with increasing extract concentrations from 36.42 ± 4.62 % in the control to 60 ± 2.24 % at 500 mg/ml for leaf extract and to 55.08 ± 3.62 % at 500 mg/ml for kernel extract. Seed damage expressed as the number of holed seeds and weight loss of infested groundnut was significantly reduced ($P < 0.05$) in each treatment as dosages increased. There was no significant reduction in the viability of groundnut seeds treated with neem extracts when compared with the control. Regression analysis showed positive correlations between dosages, adult and larval mortality and seed germination in each treatment. A negative relationship was observed between the dosages and emergence of offspring, seed damage and weight loss for each treatment.

Mostofa *et al.* (2007) He examined *Catharanthus roseus* (Nyantara), *Azadirachta indica* (Neem), *Allium sativum* (Garlic) are medicinal plants, used in Ayurveda for treating various diseases, one of which is diabetes mellitus. In the present study of 12 months period from January to December 2007, aqueous extract of this plants were prepared and blood glucose lowering effect and improvement of body weight gain in Streptozotocin (50 mg/kg bwt i.p.) induced diabetic rats were measured and compared with that of a patent drug gimepride in the Department of Pharmacology, Bangladesh Agricultural University, Mymensingh. Rats were administered *Catharanthus roseus*, *Azadirachta indica*, *Allium sativum* extracts at the dose rate of 1g/kg, 500 mg/kg and 1g/kg body weight orally for 14 days, respectively. Blood glucose level and body weight was measured by Glucotrend kit and Electronic balance and that compared with a patent drug Glimepride at a dose rate of 100 mg/kg body weight. The data were compared statistically by using student's unpaired t-test. The herbal preparations of these plants significantly increased body weight gain and decreased blood glucose as compared with the patent drug. The present study clearly indicated the significant antidiabetic activity of *Catharanthus Roseus*, *Azadirachta indica* and *Allium sativum* and supports the traditional usage of the herbal preparations by Ayurvedic physicians for the therapy of diabetics.

Rahman *et al.* (2009) the experiment was carried out for 28 days from August to September 2007 to investigate the efficacy of Ivermectin and Neem plants against ectoparasites in calves. Effect of Ivermectin and Neem plants on certain blood parameters hemoglobin (Hb), packed cell volume (PCV), total leukocyte count (TEC) and erythrocyte sedimentation rate (ESR) were studied in calves. A total of 15 calves were examined for the presence of ticks by physical examination and were divided into

three equal groups as, Group A (infected control group), Group B (treated with Neem) and Group C (treated with Ivermectin). The therapeutic efficacy of Ivermectin was 100% against ectoparasites in calves on day 7, 14, 21 and 28 after the treatment of Ivermectin. Ivermectin and Neem showed significant effectiveness at 7th, 14th, 21st and 28th day of post treatment. The effects on TEC, Hb level was increased significantly at 7th, 14th and 28th day of treatment and PCV level was increased significantly on 28th day of treatment. On the other hand indigenous medicinal plant Neem leaves shown the efficacy of 68% at day 28 and body weight of Ivermectin and Neem treated calves also increased. All the calves after Ivermectin injection and Neem spray remained healthy, no adverse effect and calves appetite increased, growth and coat color improved rapidly.

Rai *et al.* (2010) examined Stephanofilariasis, one of the most problematic parasitic diseases caused by *Stephanofilaria assamensis*, is endemic to Andaman and Nicobar Islands and many other states of India as well as neighboring countries. The present study was conducted to find out the prevalence of the disease in the endemic areas of the islands, its economic impact and suitable measures for its eradication. The prevalence of the disease was varied from 10.2 to 70.5%. The incidence rate was more in older animals. The control and eradication of stephanofilariasis in bovines could be successfully achieved especially by targeting the life cycle of the biological vector (*Musca conducens*), the intermediate host. N.S. Randhawa *et al.* (1996)

2.7 Introduction of tobacco:

Nicotiana tabacum, or cultivated tobacco, is a perennial herbaceous plant. The origin of *Nicotiana tabacum* is an Argentina and *Nicotiana rustica* is in Peru of South America. It is found only in cultivation, where it is the most commonly grown of all plants in the *Nicotiana* genus, and its leaves are commercially grown in many countries to be processed into tobacco. It grows to heights between 1 to 2 metres. Research is ongoing into its ancestry among wild *Nicotiana* species. The tobacco got much popularity during the First World War.

2.8 Taxonomic position of Tobacco:

Kingdom: Plantae

Order: Solanales

Family: Solanaceae

Genus: *Nicotina*

Species: *N. tabacum*

Binomial name: *Nicotina tabacum*

2.9 Habitat and ecology

N. tabacum is sensitive to temperature, air, ground humidity and the type of land. Temperatures of 20–30 °C (68–86 °F) are best for adequate growth; an atmospheric humidity of 80 to 85% and soil without a high level of nitrogen are also optimal.

2.10 Botanical description

Nicotiana tabacum Linné is a robust annual little branched herb up to 2.5 m (8.2 ft) high with large green leaves and long trumpet shaped white-pinkish flowers. All parts are sticky, covered with short viscid-glandular hairs, which exude a yellow secretion containing nicotine.

2.11 Leaves

Leaves varied in size, the lower leaves are the largest at up to 60 cm (24 in) long, shortly stalked or unstalked, oblonged-elliptic, shortly acuminate at the apex, decurrent at the base, the following leaves decrease in size, the upper one sessile and smallest, oblong-lanceolate or elliptic.

2.12 Part used

Every part of the plant except the seed contains nicotine, but the concentration is related to different factors such as species, type of land, culture and weather conditions. The concentration of nicotine increases with the age of the plant. Tobacco leaves contain 2 to 8% nicotine combined as malate or citrate. The distribution of the nicotine in the mature

plant is widely variable such as 64% of the total nicotine exists in the leaves, 18% in the stem, 13% in the root, and 5% in the flowers.

Dutta and Hazasika (1976) stated that 79 cattle affected with stephanophilariasis were subjected to a variety of treatment. Sumithion as a 6% emulsion in liquid paraffin applied twice daily that cured 7 of 12 cattle in 15 days, malathion powder as a 6% ointment applied twice daily cured 7 of 12 cattle in 15 days, malathion powder as a 6% ointment applied twice daily cured 6 of 10 in 16 days. Phenothiazine as a 4,6 or 8% ointment applied twice daily cured 2 of 10 in 20 days, 9 of 14 in 14 days and 9 of 12 in 14 days, respectively, powder containing 100 parts plaster of paris, 20 parts of ahen and 10 each of naphthalene and of quinine, applied daily, cured one of 8 in 7 days, an ointment containing 8% tobacco leaf decoction, 2% oil of terebinth and 1% creosote, applied daily, cured 8 of 10 in 12 days and diethyl-carbamazine acid citrate at 5 µl/100 kg bodyweight given as 3 injections at 3 day intervals cured none of 3 cattle.



CHAPTER III

MATERIALS AND METHODS

CHAPTER III

MATERIALS AND METHODS

3.1 Study area

The experiments were conducted for a period of 6 months from June/2013 to November/2013 at Veterinary Hospital, Hajee Mohammad Danesh Science and Technology University, Dinajpur and also at villages of Subra, Baser hat, Ranigong, Kornai, and Gopalgong of Dinajpur Upazilla under Dinajpur district

3.2 Selection of cattle

A total of 75 cattle of both sexes of different ages were examined during the period of June to November/2013 to study the prevalence of hump sore. The site of lesion as well as age, sex and breed were recorded in all the animals showing evidence of Stephanofilariasis. The lesions were confirmed by clinical findings. Out of 75 cattle, 35 were examined in the Veterinary Hospital, Hajee Mohammad Danesh Science and Technology University, Dinajpur and also villages of Subra, Baser hat, Ranigong, Kornai, and Gopalganj of Dinajpur Upazilla under Dinajpur district

3.3 Experimental layout

A total of 15 clinical cases of stephanophilariasis having typical hump sore lesion on hump and neck region were selected for study purpose. The lesions of hump sore were 3.5-10.5 cm in diameter. Smaller lesions were considered less than 5 cm and larger lesions were considered greater than 5 cm. All the cattle were mostly cross-breed progeny of Holstein Friesian, Shahiwal and some local breed aging from 4 to 12 years. The draft purpose cattle were affected more. These 15 cattle were divided into 3 groups each consisting, of 05 cattle. One group (group C) was kept as untreated control group and rest two were treated with Tobacco leaves ointment and neem leaves extract respectively. All the cattle were maintained with balanced feed and water *ad libitum*.

Experimental layout

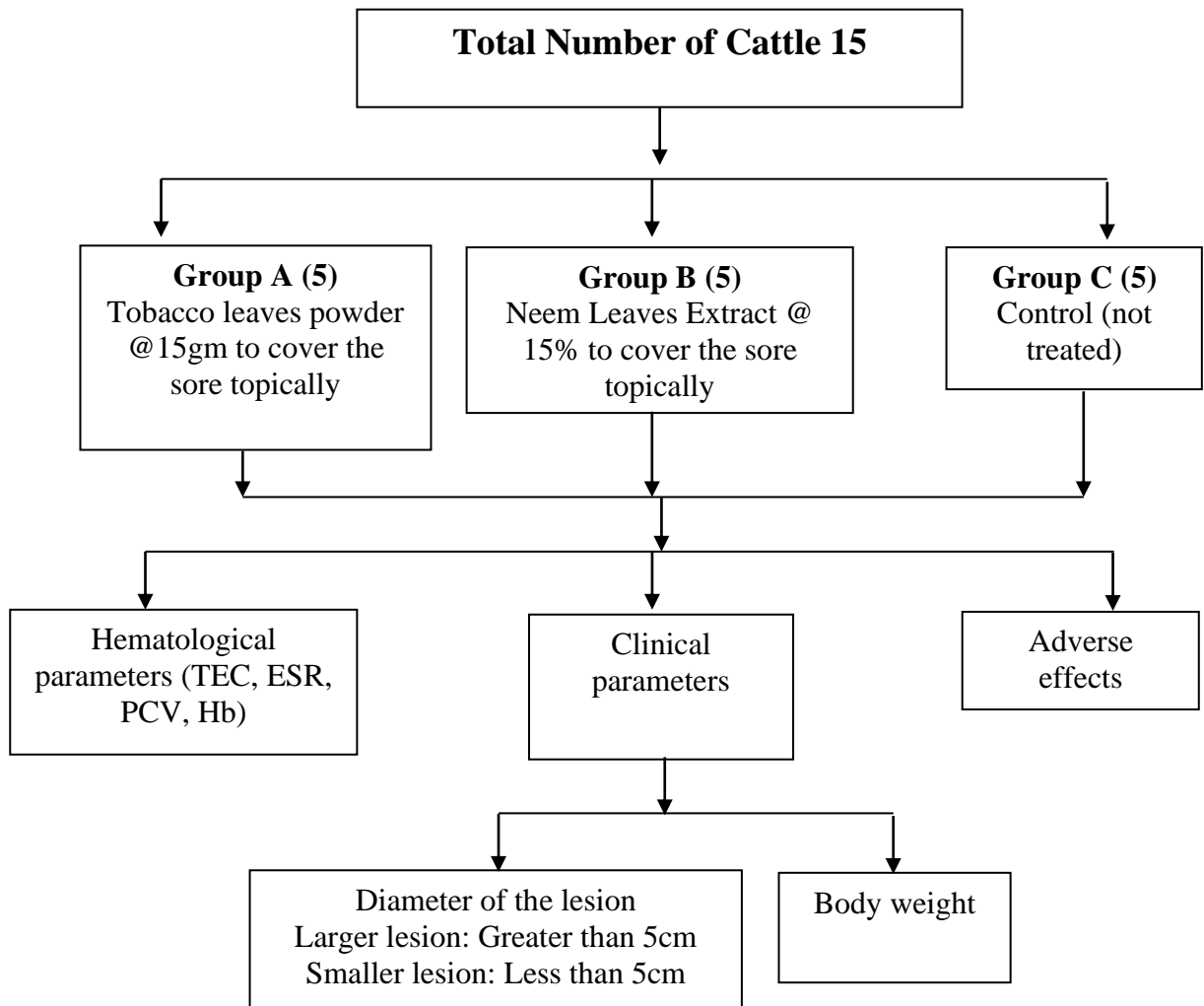


Fig.1. Experimental layout

3.4 Materials

3.4.1 Tobacco leaves

Dried tobacco leaves were collected from a tobacco shop in the Dinajpur town.

3.4.2 Neem leaves

Apparently healthy green leaves were collected from neem trees in the botanical garden of the Hajee Mohammad Danesh Science and Technology University, Dinajpur.

3.4.3 Others:

- Balance
- Slab and spatula
- Vaseline
- Knife
- Mortar and pastels
- Scale to measure the sore
- Digital camera
- Blander
- filter paper
- Hot air oven

3.4.4 Preparation of Neem leaves extract

Neem leaves (*Azadirachtin indicia*) were collected from the campus of Hajee Mohammad Danesh Science and Technology University, Dinajpur. To obtain 15% aqueous extract, 15g of neem leaves were weighted from electric balance then thoroughly washed in tap water. The leaves were cut into small pieces with the help of knife, there after the fleshy parts were mashed with the help of mortar and pastels. The extract was made up to 100 ml by adding distilled water and filtered it through the filter paper with the help of Beaker and Funnel. It was kept overnight at 40 degree centigrade and the supernatant was collected.



Figure I: Neem Leaves



Figure II: Weighted by electric balance



Figure III: Maceration



Figure IV: Added water to Neem

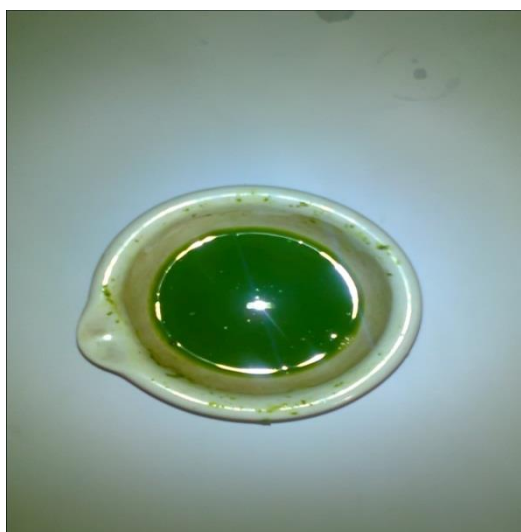


Figure V: Neem leaves extract

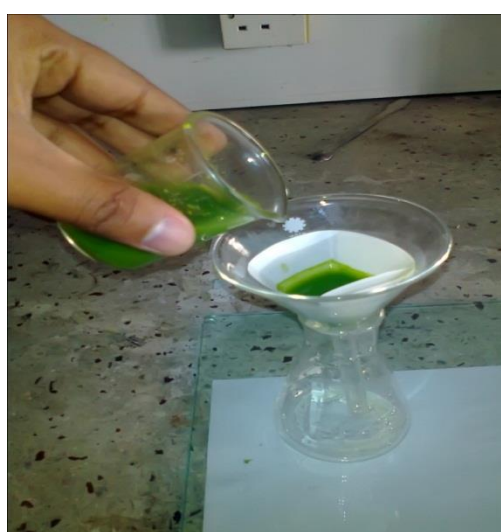


Figure VI: Filtration



Figure VII: Collect Neem leaves extract

3.4.5 Preparation of tobacco powder

Dried tobacco leaves were collected from a tobacco shop in the Dinajpur town. To obtain powder 15g of dry tobacco leaves were weighted from electric balance. Then the dry leaves turn into powder form with the help of pastels and mortar.



Figure VIII: Dry tobacco leaves



Figure IX: Weighted by electric balance



Figure X: Tobacco leaves powder

3.5 Application of drugs

Tobacco leaves ointment was applied topically twice daily to the cattle for 28 days of the cattle of Group-A. 15 gm tobacco powder was mixed with 85 gm Vaseline. And 15% neem extract were used in affected area topically. The fate of the lesions was inspected in two groups every alternative day for one month from the first application of the drugs. Efficacy of Tobacco and Extract of Neem leaves was assessed by observing clinical healing of lesions, and period of healing of the lesions.

3.6 Effect on body weight

The body weight of all treated and control groups of affected cattle were recorded before application of tobacco powder and neem leaves extract and on day 30 after application of drugs.

3.7 Effect on blood

Blood samples were collected from Jugular vein of the cattle of treated and control group in vials containing anticoagulant (Sodium Citrate 3.8%) at 1st, 7th, 14th and 28th day of treatment period to determine the effects of tobacco and neem on the following hematological parameters such as Total Erythrocyte Count (TEC), Hemoglobin (Hb) Content, Packed Cell Volume (PCV) and Erythrocyte Sedimentation Rate (ESR).

3.7.1 Total Erythrocyte Count (TEC)

Thomas red blood cell pipette was used for Total erythrocyte count (TEC). The pipette was filled up to 0.5 marks with blood and diluting fluid (Hayem's solution) was then drawn till it reached the 101 mark. The contents were thoroughly mixed for 2 minutes. The dilution of the contents was 1:200. The counting chamber and cover slip were cleaned and dried properly. The cover slip was placed on the chamber and cover slip were cleaned and dried properly. The cover slip was placed on the chamber in proper position. The content of the pipette was again shaken and 2 or 3 drops of the fluid were expelled. The chamber was then filled with the contents so that the fluid was flown under the cover slip by capillary action. The ruled area at the chamber was filled completely, taking care that excess did not run into the troughs and no air bubble appeared under the cover slip. The counting chamber was then placed under microscope and examined first with low power objectives (10 x) to ensure that there was an even distribution of the cells. The cells were then counted with the aid of high power objectives (100 x). The central squares of the counting chamber were counted in the far corner and one central square of the chamber. The counting and calculation of red blood cells were performed as per methods indicated by Coffin (1955). The number of RBC was calculated as follows: Number of RBC = No. of cell counted \times 10,000 and the result was expressed in million/cu.mm. (Coffin 1955).

3.7.2 Determination of Hemoglobin (gm %)

The hemoglobin estimation was performed by the acid Haematin method with the Hellige Hemometer (Coffin, 1955). Hydrochloric acid (N/10) solution was taken in the special graduated tube upto its 20 mark. The special Sahli pipette was filled with well mixed oxalated blood upto 20 mark and blood on the side of the pipette was wiped out by cotton. The content of the Sahli pipette was expelled into the special graduated diluting tube and thoroughly mixed. The tube was then allowed to stand for 10 minutes for development of acid haematin. Distilled water was then added drop by drop and each time mixed with the help of stirring rod until the colour of the solution matched with the standard colour of the comparator. The result was read as per method described by (Coffin, 1955).

3.7.3 Packed cell volume (PCV)

The PCV was determined as per method described by (Coffin, 1955). The Win Trobe hematocrit tube was filled up with well-mixed blood by special loading pipette up to 10 marks. Then the tube was centrifuged at 3,000 rpm for half an hour and reading was taken.

3.7.4 Determination of Erythrocyte Sedimentation Rate (ESR)

The collected blood sample was filled with the special loading pipette slightly above the '0' mm mark or '10' mark. The tip of the pipette was inserted to the bottom of the hematocrit tube and the blood was expelled by the pressure on the rubber bulb, withdrawn the pipette as the tube filled. Each tube was then placed in the special rack being certain that they were vertical. The tube was allowed to remain in the support without disturbance for an hour. The ESR value was determined as per methods describe by (Coffin, 1955).

A decorative graphic consisting of several overlapping squares in blue, red, and orange, and two intersecting lines, one horizontal and one vertical, in a light teal color. The text is centered within the intersection of these lines.

CHAPTER IV

RESULTS

CHAPTER IV

RESULTS

A research work was conducted -

1. To study the comparative efficacy of tobacco leave powder and neem leaves extract against hump sore in cattle.
2. To determine the effect of tobacco leave powder and extract of neem leaves on some clinical parameters (severity of infestation, body weight) in cattle.
3. To determine the effect of tobacco leave powder and extract of neem leaves on some hematological parameters (TEC, ESR, PCV and Hb) in cattle.

4.1 The prevalence of stephanofilariasis (Hump sore) in cattle of Veterinary Hospital at Hajee Mohammad Danesh Science and Technology University and nearby five villages of Dinajpur district.

Humpsore is prevalene in Dinajpur district. Out of 75 cattle of Veterinary Hospital at Hajee Mohammad Danesh Science and Technology University and nearby four villages - 15 were found affected with stephanofilariasis. Out of 15 positive cases the site of the disease was distributed over the neck region (28.12%) hump region (8.5%) and other (3.48%). The prevalence was recorded (40.1.5%). high case of male animals the lesions were mainly in the neck region. It was also observed that draft purpose cross-breed cattle were more susceptible to the disease than local breed.

4.2 To study the comparative efficacy of tobacco leaves powder and neem leaves extract against hump sore in cattle- tobacco is more effective than neem.

4.2.1 Tobacco powder

Cattle of group A (Figure XI & XII) treated with tobacco powder topically twice daily showed gradual improvement of the lesions within 7th days post treatment and marked improvement of the lesions within 25st days of treatment which was assessed by the gradual reducing of irritation and size of the lesions and formation of crust to the sore. At

the end of the 28th days of post treatment days the lesions were completely healed up with the formation of scar tissue and then new hair started to grow over the entire affected area (Table 1, Figure XI and XII).



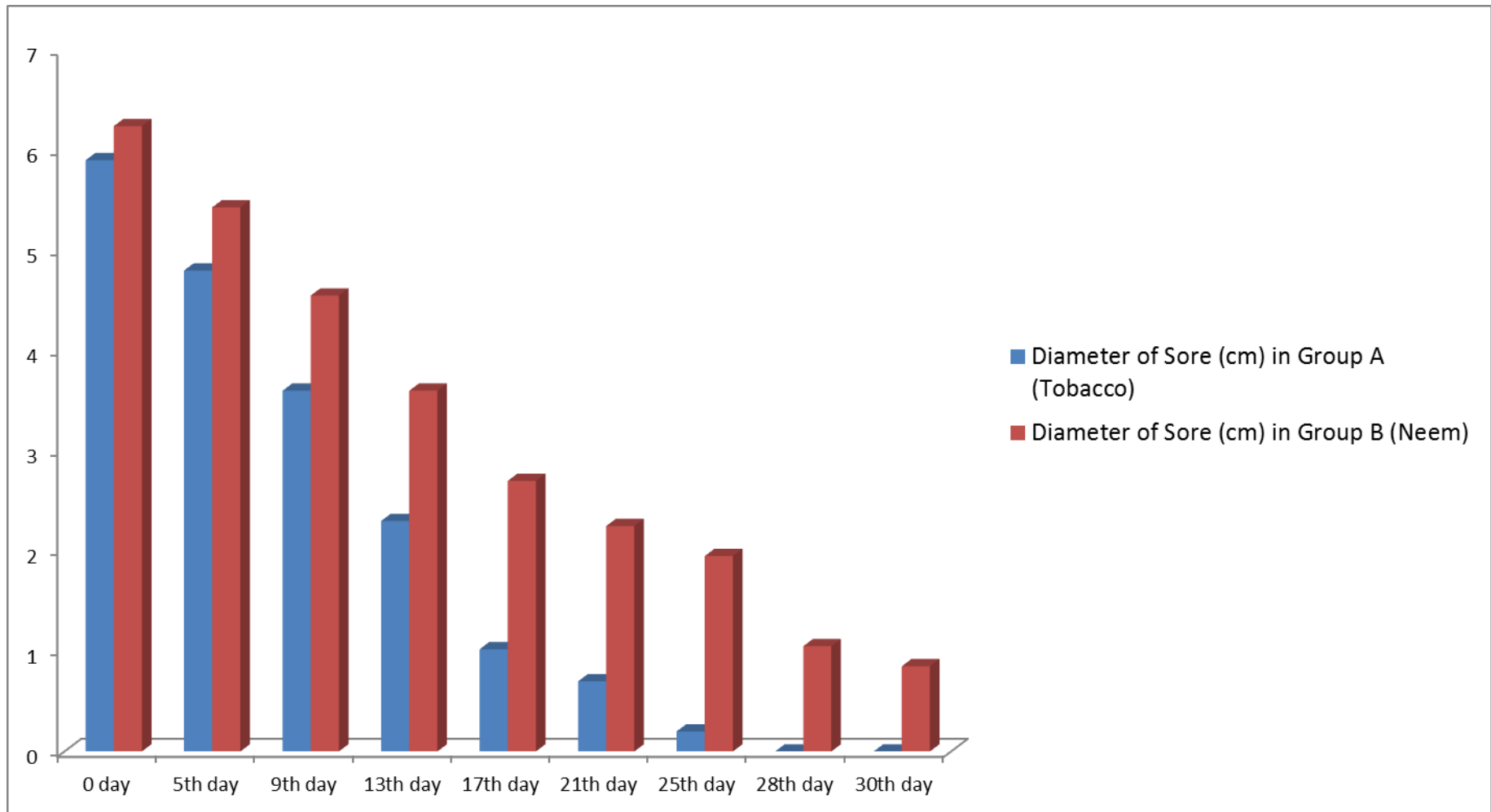
Figure XI: Hump sore lesion before treatment with tobacco ointment.



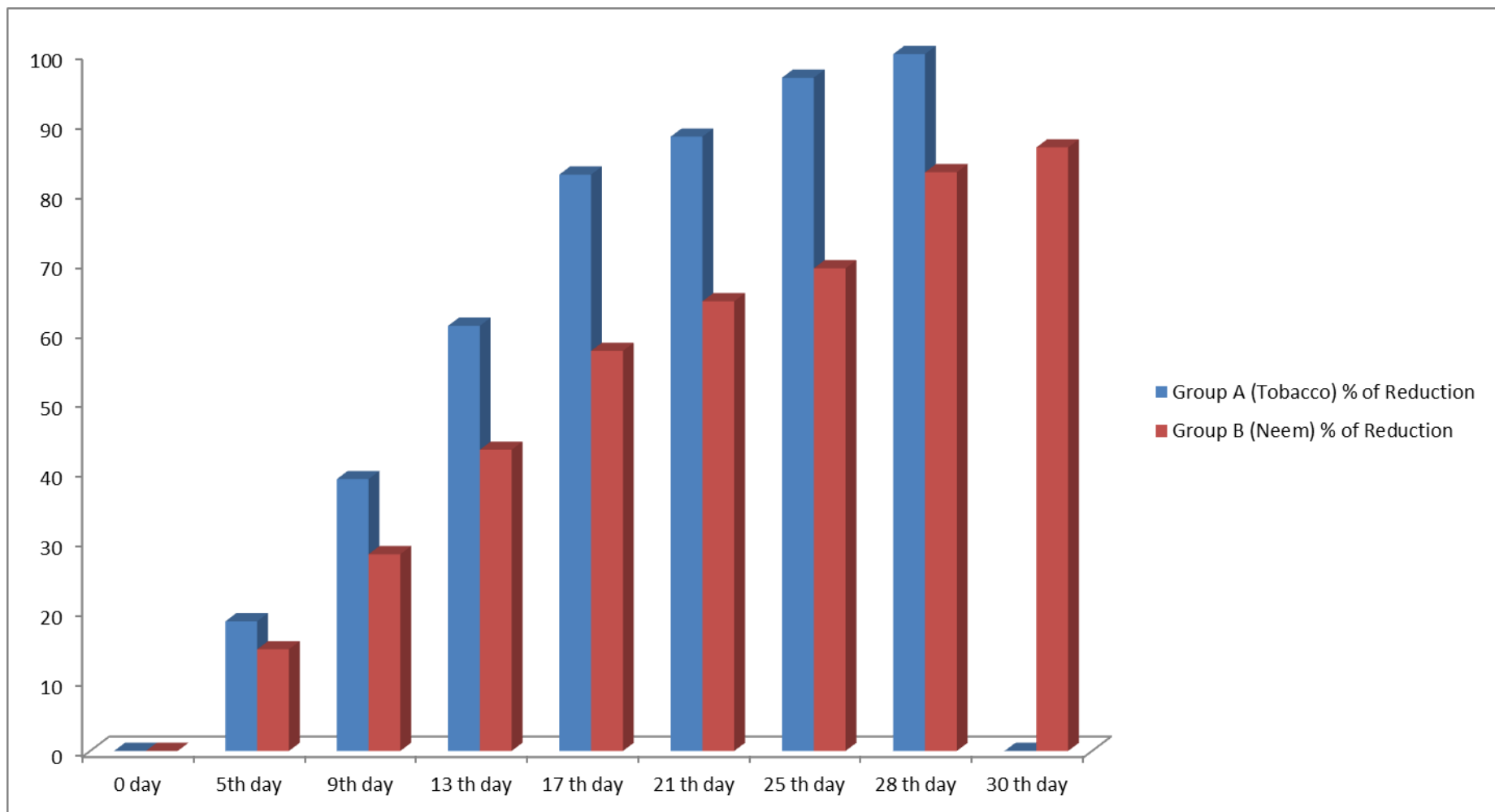
Figure XII: Hump sore lesion after treatment with tobacco ointment.

Table 1. Effect of tobacco powder and neem leaves extract against hump sore in cattle

Treatment Days	Diameter of sore (cm) in Group A after application of Tobacco (mean±SE)	Reduction (%)	Diameter of sore (cm) in Group B after application of Neem leaves extract (mean±SE)	Reduction (%)	Diameter of sore (cm) in Group C (Control group) (mean±SE)	Reduction (%)
0	5.9±0.14	0	6.24±0.16	0	6.4±.31	0
5 th	4.8±0.17	18.6	5.43±0.13	14.58	6.4±.34	0
9 th	3.6±0.13	39	4.55±0.10	28.23	6.5±.35	0
13 th	2.3±0.11	61	3.60±0.25	43.21	6.6±.36	0
17 th	1.02±0.12	82.7	2.70±0.13	57.41	6.7±.37	0
21 st	0.7±0.01	88.14	2.25±0.07	64.52	6.8±.39	0
25 th	0.2±0.03	96.6	1.95±0.02	69.25	6.9±.40	0
28 th	0	100	1.05±0.05	83.03	7.0±.41	0
30 th	0	0	0.85±0.06	86.60	7.1±.42	0



Graph 02. Effect of Tobacco powder and Neem leaves extracts against hump sore in cattle



Graph 03. Effect of Tobacco powder and Neem leaves extracts against reduction % of hump sore in cattle

4.2.2 Neem leaves extract

Stephanofilariasis of group B cattle (Figure XIII & XIV) treated with Neem leaves were used to covers the sore area topically around the sore showed slight improvement after the application which was assessed by reduction of itching, irritation and progressive drying. After application of 2-7th day there was significant improvement with reduction of the size of the lesions. After 30th days post treatment lesions were completely healed up by formation of smooth black scar tissue within 29th days. It was also observed that smaller lesions improved more rapidly than the larger lesions. (Table 1, Figure XIII & XIV).



Figure XIII: Hump sore lesion before treatment with neem leaves extract.



Figure XIV: Hump sore lesion after treatment with neem leaves extract

4.2.3 Comparative efficacy of tobacco leaves powder ointment and neem leaves extract

It was also observed that treatment with neem leaves extract required above 30 days to complete recovery of the sore. On the other hand topical application of tobacco leaves powder showed 100% effectiveness at the end of the 28 days. The larger lesions required longer time to heal up than that of smaller lesion when treated with neem leaves extract but the recovery time was shorter when treated with tobacco leaves powder.

No side effect or Adverse reaction was produced by any of the formulation (neem leaves extract and Tobacco powder) used in this experiment. The results of therapeutic trials are represented in Table I. The lesions (hump sore) of animals of the untreated control group (group C) did not show any improvement during the period of the experiment (Table 1).

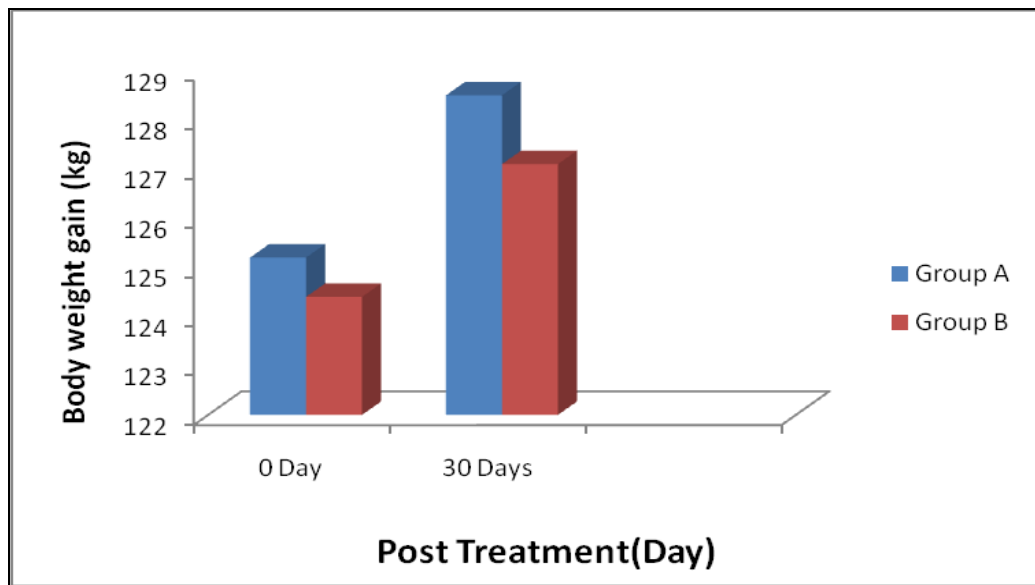
4.3 Effect of tobacco and neem leaves extract on body weight and hair coat

The body weight of treated cattle (Group A & B) was increased to the extent of 3.3% and 2.7% on 30th day, following tobacco and neem leaves extract application respectively. On the other hand, the body wt. of control group decreased gradually (Table 2).

Table 2. Effect of tobacco leaves powder and neem leaves extract on body weight in cattle

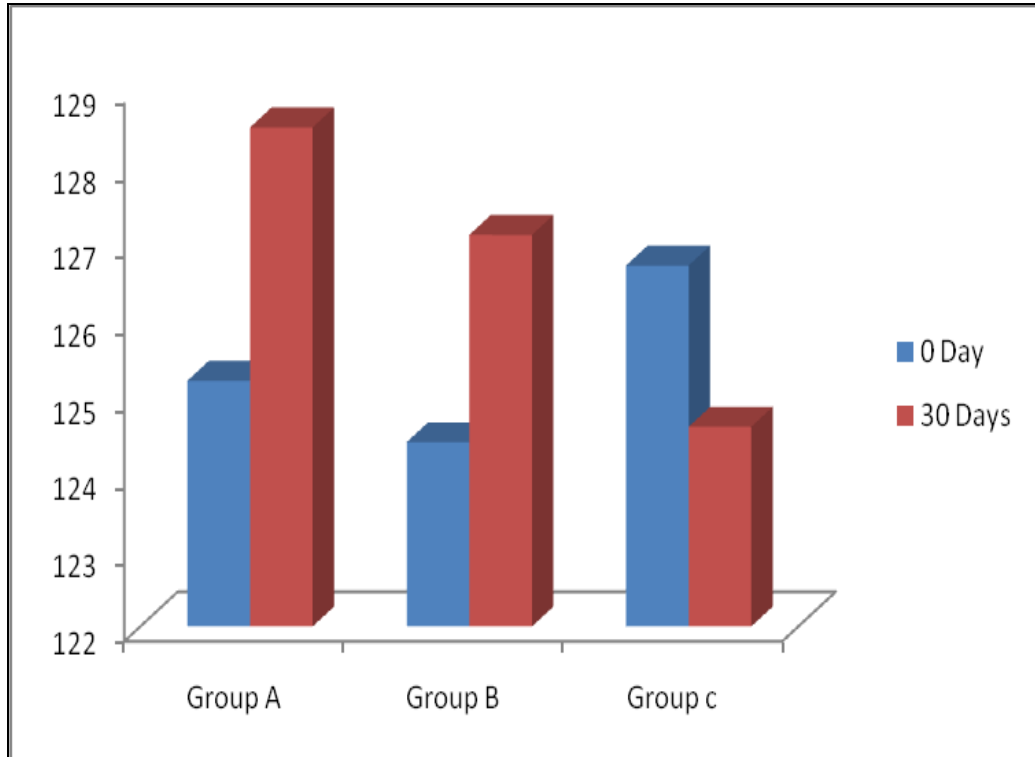
Group	Length of trial	Drug with dose	Pretreatment Day (0)	Post treatment (30 th day)	Live weight gain/loss (kg)	Improvement Percentage (%)
			Body weight (kg) (mean+SD)	Body weight (mean+SD)		
A (Tobacco)	30 days	Tobacco powder twice daily	125.2±7.5	128.5±9.5	3.3	2.6
B (Neem)	30 days	Neem leaves extract at the dose rate of 1gm/kg body weight	124.4.±9.30	127.1±3.4	2.7	2.17
C (Control group)	30 days	Untreated	126.7±5.5	124.6±2.6	-2.1	0

4.3.1 Effect of tobacco ointment and neem leaves extract on body weight in cattle



Graph 04. Effect of Tobacco powder and Neem leaves extract on body weight in cattle

4.3.2 Effect of tobacco and neem leaves extract compeering with control group on body weight in cattle



Graph 05. Effect of tobacco powder and neem leaves extract compeering with control group on body weight in cattle

4.3.3 Effect of tobacco and neem leaves extract on hair coat

To evaluate the anthelmintic activity and certain blood parameters of neem leaves extract and tobacco in comparison with the control group. In Group B, Neem at 1gm or 15% spray was found to be 68% effective against humpsore. Humpsore area are decreased within the selected area on 7th, 14th, 21st and 28th day after the treatment on the other hand in control group C, the number of ticks increased gradually on 7th, 14th, 21st and 28th day of treatment..

The hair coat of the infected cattle (group A and B) was rough with discolored wool on 1st day. In group A and B, after treatment with neem leaves extract and tobacco, the hair coat started to become smooth and shiny gradually and on 28th day of treatment the hair coat of the treated cattle were almost normal. The hair coat of the infected control group became more rough and discolored. The mean initial body weight of group A and B treated with neem leaves extract 15% solution or to covers the sore area and tobacco or 15gm mixed with 85 gm Vaseline were 125.2 kg and 128.5 kg on day 0 and on the 28th day of post treatment the mean values of body weight were 128.5kg and 127.1 kg, respectively. So body weight of cattle increased significantly in treated groups on 28th day of treatment. In control group (C), body weight of cattle was decreased upto 2.1 % within 30 days. On the other hand, the body weight was increased in groups (A and B) to the extent of 2.6%, 2.17%. So body weight of cattle increased significantly in treated groups on 28th day of treatment (Table A).

4.4 To determine the effect of tobacco powder and extract of neem leaves on some hematological parameters (TEC, ESR, PCV, Hb) in cattle.

Table 3. Effects of tobacco and neem leaves on TEC (million/cu. Mm) in cattle

Group	Treatment	Pre-unit treatment	Unit Post-treatment			
		0 day	7th day	14th day	21 st day	28th day
A	Tobacco	6.7±0.083	8.22 ±0.127** (18.25)	9.043 ±0.071** (8.13)	10.562 ±0.160* (12.72)	11.32 ±0.168* (9.25)
B	Neem	6.51±0.148	7.513 ±0.063 (13.43)	8.82 ±0.190** (16.19)	10.068 ±0.078 (11.50)	10.62 ±0.186** (5.20)
C	Control	7.14±0.112	6.82 ±0.112 (6.56)	6.52 ±0.147 (4.12)	6.48 ±0.17 (1.26)	6.57 ±0.155 (2.47)

Values given above are mean±SE of 05 cattle * significantly increased (P<0.05), ** significantly increased (P<0.01)

In control group (C), Hb content decreased upto 4.54% on 28th day of treatment. On the other hand, the Hb contents increased in treated groups A and B to the extent of 3.12%, 1.12%, respectively. So Hb content increased significantly in treated groups on 28th day of treatment (Table 4). In control group (C), PCV (%) values decreased upto 14.06% on 28th day of treatment. On the other hand, the PCV (%) values were increased in all treated groups (A and B) to the extent of 1.13%, 1.48% (Table 5). In control group (C), ESR (mm/1st hr) values increased up to 5.16 on 28th day of treatment. On the other hand, the ESR (mm/1st hr) values were decreased in all treated groups of A and B significantly ($P < 0.001$) (Table 6).

Table 4. Effects of tobacco and neem leaves on Hemoglobin Hb content (gm %) in cattle

Group	Treatment	Pre-unit treatment	Post-treatment			
		0 day	7th day	14 th day	21st day	28th day
A	Tobacco	6.40±0.140	7.28 ±0.097** (11.71)	7.51 ±0.150** (3.17)	8.31 ± 0.128* (9.34)	8.7 ± 0.194* (3.12)
B	Neem	7.12±0.115	7.36 ±0.115** (3.25)	7.66 ±0.108** (3.66)	8.60 ± 0.145** (10.97)	8.6** ± 0.112 (1.12)
C	Control	6.50±0.425	7.102 ±0.187 (6.94)	6.62 ±0.180 (6.87)	6.88 ± 0.193 (3.80)	6.59 ±0.166 (4.54)

Values given above are mean±SE of 05 cattle, * significantly increased (P<0.05), ** Significantly increased (P<0.01).

Table 5. Effects of tobacco leave ointment and neem leaves on packed cell volume (PCV) (%) values in cattle

Group	Treatment	Pre-treatment	Post-treatment			
		1 st day	7 th day	14th day	21st day	28th day
A	Tobacco	29.70±0.309	29.52±0.228** (0.57)	29.47 ±0.158** (0.32)	30.64 ±0.430** (3.97)	30.41 ±0.468** (1.13)
B	Neem	28.51±0.340	28.45 ±0.317** (0.35)	28.895 ± 0.170** (1.32)	29.85 ±0.882 (3.10)	29.38 ±0.382 (1.48)
C	Control	28.89±0.905	29.68 ±0.259 (2.12)	29.0 ±0.254(1.43)	26.47 ±0.264 (10.40)	23.22 ±0.294 (14.06)

Values given above are mean±SE of 5 cattle, * significantly increased (P<0.05), ** Significantly increased (P<0.01)

Table 6. Effects of tobacco powder and neem leaves extract on Erythrocyte sedimentation rate (ESR mm/1st hr)

Group	Treatment	Pre-treatment	Post-treatment			
		1 st day	7 th day	14th day	21st day	28th day
A	Tobacco	0.143±0.011	0.12 ±0.008 (30.95)	0.0916±0.021** (20.09)	0.083±0.013* (10.36)	0.030±0.006**(76.87)
B	Neem	0.117±0.010	0.0854 ±0.017 (41.72)	0.064±0.005** (42.34)	0.055±0.008 (21.00)	0.035±0.013**(38.90)
C	Control	0.947±0.021	1.044 ±0.014 (9.21)	1.405±0.017 (26.25)	1.827±0.025 (23.17)	1.926±0.032 (5.16)

Values given above are mean±SE of 05 cattle, * Significantly increased (P<0.05), ** Significantly increased (P<0.01)



CHAPTER V

DISCUSSION

CHAPTER V

DISCUSSION

The present research work was performed at the Veterinary Hospital at Hajee Mohammad Danesh Science and Technology University, Dinajpur and also examined in the nearby villages of Subra, Baser hat, Raniganj, Kornai, and Gopalganj of Dinajpur Upazila under Dinajpur district

Prevalence of stephanofilariasis in Veterinary Teaching Hospital of Hajee Mohammad Danesh Science and Technology University And nearby five villages-

High prevalence of humpsore in cattle in Dinajpur district and lesions were found to be distributed mainly on the neck region (29.12%), hump region (7.5%). The prevalence was recorded as 39.12%. In case of male animals the lesions were mainly in the neck region. It was also observed that draft purposes cross breed cattle were more susceptible to the disease than local breed. The present findings were similarly as reported by Alam (1995).

5.1 Effect of tobacco powder and neem leaves extract against and hump sore in cattle:

5.1.1 Effect of treatment with tobacco ointment

The topically applied tobacco powder was 100% effective against stephanofilariasis in cattle without showing any side effect and the complete recovery was observed within 28th day of 1st day topical use. The present findings have close correlation with the findings of Dutta and Hazasika (1976) .They topically applied 8% tobacco leaf decoction, 2% oil of terebinth and 1% creosote. was very effective (100%) against humpsore. In the present research study, tobacco powder was found more effective.

5.1.2 Neem leaves extract

Topical application of Neem leaves extract were used to covers the sore area topically .It was found 86.60% effective without any side effect. Animals with both larger and smaller lesions were cured within 30th days post treatment. After the 1st day treatment, the percentage of healing of the sore was less. In subsequent treatment on the 1st to 17th day the healing was remarkable with scar tissue formation and growth of new hair around the scar tissue. This may be due to the antiparasitic action of the Neem leaves extract.



CHAPTER VI

SUMMARY AND CONCLUSION

CHAPTER VI

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Topical application of tobacco powder was found to be 100% effective against stephanofilariasis in cattle. Animals with both smaller and larger lesions were cured within 28th days of post treatment days.

Similarly, topical application of Neem leaves extract was also found 86.60% effective against stephanofilariasis. All the cattle treated with neem leaves extract were found fully cured within 36th days of post treatment.

The mean body weight of the treated cattle increased on 28th days of post treatment with topical application of tobacco. The increase on body wt was 2.17% and 2.60% following subcutaneous application of neem leaves extract and topically application of tobacco respectively.

- ❖ Neem leaves extract were used @ covers the sore area topically against humpsore were (86.60%) effective within 30th days without any side effects. In subsequent treatment on the 1st to 29th day the healing was remarkable with growth of new hair around the scar tissue.
- ❖ Tobacco was 100% effective. The complete recovery was observed within 28th day of 1st day topical use
- ❖ Body weight of cattle treated with tobacco and neem leaves extract increased (2.6% and 2.17% on 30th days post treatment) appreciably. Among tobacco and Neem leaves extract, the tobacco found to be superior against hump sore in cattle. But Neem leaves extract has no side effects.

On the basis of above findings the following conclusion may be drawn

Further research works is to be needed to explore the adverse effects, bio-chemical analysis of neem leaves.



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