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Field based efficacy assessment of some ethno-herbal practice as an alternative anthelminthic approach in Black Bengal goat in Tribal area of Bengal

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ABSTRACT

Bengal goats reared in semi-intensive system by marginal tribal farmers of West Bengal, India are naturally infested with endoparasites due to different managemental and climatic factors. Easily available ethnoveterinary herbs like *Swertia chirata*, *Piper Nigrum* and *Nigella sativa* mixture were assessed as an alternative anthelminthic approach compared with market available dewormers in parasite infested goat in field level. The study based on 36 bengal goats revealed that the herbal mixture is as effective as chemical dewormer to reduce the parasitic load. Moreover, the total protein and AST value is better in herbal treated goats.

Keywords: Tribal, Goats, *Nigella sativa*, *Piper nigrum*, *Swertia chirata*, Anthelminthic, AST.

INTRODUCTION

Small ruminants like goat and sheep play an important social and economic role around all over the world particularly Asia, Africa and Oceania. The small and marginal tribal farmers of growing country like India, Bangladesh etc. are largely dependent on goat population for their livelihood and thus goat industry become an important contributor of national GDP. West Bengal is one of the largest goat populated state of India having mostly the Black Bengal goat population within it, which is famous for unique meat, hide and fecundity characteristics. Due to the backyard situation, most of the goat population in Bengal are unorganised and reared within open system. Due to the prevalence of hot and humid climate in region, goats are naturally exposed to various endoparasitic infection while grazing over open field. As most of the backyard tribal farmers are mainly involved in primary work like daily labour at agricultural field, usually they have neither enough money nor time to treat goats scientifically. Cumulatively natural endoparasitemia prevails in the caprine body system and subsequently exerts its inhibitory nature on sustainable development of livestock farming.

Severe endoparasitemia in goat is a slow process which exerts its action in a grave manner in rural area. The total body growth, productive and reproductive performance are highly affected by the extent of parasitic load within goat alimentary system. That ultimately compels the poor livestock farmer into more poor condition from the economic point of view.

Several ethnoveterinary practices are generally practiced in different region of world. Most of them comprises locally available herbs. It has been also found that daily used kitchen spices from herbs also possess different antibiotic, antihelminthic and other remedial functions as ethnoveterinary medicine. As these resources are easily available within home and cost-efective for the farmer, so these types of herbal components also may be tried to eliminate the endoparasitic load from goat as alternative therapy.

Nigella sativa seeds (commonly known as Kalazeera) are one of the daily used kitchen spices in Bengal. This famous historical herbal products has a large spectrum of medicinal activities like diuretic, antihypertensive, antidiabetic, anticancer and immunomodulatory, analgesic, antimicrobial, anthelmintics, analgesics and anti-inflammatory, spasmolytic, bronchodilator, gastroprotective, hepatoprotective, renal protective and antioxidant properties [1].

Swertia chirata (commonly known as Chirata) is another ethnomedicinal herb commonly available and used by common people to treat themselves for liver associated problems. It also exhibits diverse biological activities, such as antibacterial, antifungal, antiviral, anticancer, anti-inflammatory, antidiabetic and antioxidant activities [2]. The bitter taste of the herb is attributed by the presence of

different active components like amarogentin (most bitter compound isolated till date), swerchirin, swertiamarin etc. It is also used by rural farmers for their livestock in Bengal region [3].

Black pepper (*Piper nigram* linn seed) is generally used as a spice and seasoning in the region. Several research outcome indicates that the seeds possess antiparasitic efficacy [4] which coincides with the traditional knowledge of villagers. In most of the cases though it is wise known for its antibacterial property [5].

As the common spices or herbs are easily available in village and the exact efficacy level of the spices over parasitic infection is still not so clear, so the research was carried out to explore the potentiality of traditionally used these three spices against endoparasitemia in goat.

MATERIAL AND METHOD

1. Selection of area and time- The operational areas are situated at tribal dominated two adjuscent villages namely, Srimantapur and Daspara, Bagdah block at North 24 Pargana district of West Bengal. Most of the goat farmers of these area are from low economic strata and usually have no time and awareness for veterinary care of their livestock. Due to the prevalence of parasites, pre-monsoon season were selected for the study.

2. Collection of plant materials- The plant materials are taken as per the form the local people used to use those in normal life as a spice. Thus *Nigella sativa* seeds, black pepper seeds and *Swertia chirata* dried stem with dried leaves are procured from fixed local shop. The ingredients are thoroughly checked for any impurities and then grinded separately by mixer machine to make a pulverished form. All powders are mixed with equally and packaged at plastic pouch having 6 grams inside each packet.

3. Selection of animals- a total no. of 36 of clinically checked adult male bucks having symptoms of parasitemia and Egg per gram or epg>1000, body weight- average 12-14 kg were selected for the study. The owners are advised not to incorporate any kind of alteration to the normal lifestyle of the experimental goats except prescribed medication for 15 days. All the animals are closely observed during the period of experiment.

4. Design of experiment- All goats were randomly divided into 3 groups having 12 animals in each group. Group I animals were treated with a total of 6 gram of herbal mixed powder daily in empty stomach in the morning for 14 days. Due to the bitter taste and low solubility of Swertia powder into water, the owners are advised to mix little bagasse with the mixture. At the beginning of the experiment, Group II and Group III animals were dewormed with Fenbendazole @ 7.5 mg/Kg body weight and Albendazole with Praziquantel combination (Praziplus- Albendazole 300 mg, Praziquantel 50mg per tab) @ 10 mg/Kg body weight once p.o. in empty stomach respectively. Blood and faecal samples were collected for analysis at weekly interval in the early morning before administration.

5. Parasitic infection parameter- parasitic infection was monitored through the egg per gram or EPG count at day 0, day 7 and day 14 for all the groups by Mcmaster technique [6]. The reduction of EPG was evaluated to monitor efficacy of treatment.

6. Biological samples collection procedure- Blood samples were taken aseptically from jugular vein of selected goats both in serum vial (3ml) and EDTA coated vial (3ml) for biochemical and haematological assays.

7. Purchase of chemicals and reagent- Diagnostic kits to assess biochemical parameters like serum glutamic oxaloacetic transaminase (SGOT), serum glutamic pyruvic transaminase (SGPT), Blood urea Nitrogen (BUN) and Serum creatinine were purchased from Cogent, India. Other chemicals and reagents were purchased from E-Merck (India) and Rankem Pvt. Ltd.

8. Statistical analysis- The statistical comparison was performed using SPSS- version 21. The two way ANOVA followed by Bonferroni post hoc ($p<0.05$) test was computed to know the within group (at different days) and between group (at same day) comparison.

RESULTS

1. Clinical findings during the period

The clinical signs and symptoms of endoparasitemia in animals were monitored in all the group during the experimental period. Irrespective of group, all the selected animals were noticed with dullness, loss of normal alertness, inappetance and apathy to food in pretreatment period. The faeces were sticky and semi-solid in consistency as well. During the first two days post treatment, no major observable changes in symptoms were noticed. After 3rd day in case of group II and group III animals, the faecal consistency began to change into solid form. By the end of the study period it had been restored to normal pelleted form. Similar alteration were recorded after 7 days post treatment in Group-I animals. Appetance and alertness also improved with the course of treatment. Skin tent examinations revealed gradually improved rehydration status and skin tonicity by the end of observation period. The conjunctival mucous membrane started to look pinkish at the last phase of experimental period compared to the initial pale appearance for all groups. No marked difference had been observed in case of body temperature, respiration and heart rate.

Parasitic load in goats was measured through egg per gram estimation (Table1). The experimental animals from three different villages exhibited mixed nematodal infestation by microscopic fecal egg observation. However, eggs of Strongyle species were predominant among those nematode eggs. The initial mean EPG for group I, group II and group III were decreased upto 74.51%, 78.84 % and 80.84 % respectively after 14 days post treatment (Graph 1). Our observation recorded no significant variation of EPG among three different groups post treatment at 14th day.

The haematological status was assessed based on Haemoglobin percentage (Hb%), Total erythrocytic count (TEC) and Total leucocytic count (TLC) in different groups (Table 2). The haemoglobin status in all the treatment groups improved significantly ($p\leq0.05$) after first week of treatment and continued until 14th day post remedy. However, maximal improvement of 41.41% was recoreded in herbal mix group in comparison to 32.08%, 35.98% in group II and group III respectively (Graph 1). Similarly, the mean TEC as a valuable component of hemogram also enhanced significantly ($p\leq0.05$) post treatment in subsequent two observation periods in all the groups. Total leucocytic count also varied with the

progress of the experiment. The typical high TLC (seen in parasitemia) of pre-treatment animals of three groups returned to normal with significant reduction post treatment. There was no significant variation was observed in reduction rate between the three different treatment groups.

Serum AST and ALT level (table 1) were measured as a biomarker of liver function test. At beginning of experiment, the mean serum AST or SGOT levels were recoded elevated pertaining hepatic involvement. The herbal mix treatment was able to normalise the serum AST level more effectively after first week of treatment compared to two other commercial anthelmintic preparations. Moreover, at the end of the observation period the serum AST value in herbal mix treatment group was significantly lower compared to other two groups (Fig 1). Likewise, mean serum ALT or SGPT level also significantly altered from pre treatment value in all the three groups. But unlike the AST results at the end of study period, there was no significant difference observed between the serum ALT levels of three different treatment groups. Change in SGOT level were more prominent than change in SGPT level.

Serum BUN and creatinine level are considered as biomarkers of kidney function. The pre-treatment value of mean BUN for group I, II and III were returned to normal level after first week of treatment in all the three groups without much variations in reduction rate between groups (Graph 1). However, there was no significant difference observed in mean creatinine level in different observation periods within all the groups.

Table 1: Change in hemato-biochemical parameters and egg per gram count in parasite infested Bengal goats (n=12) by different treatment.

Days of experiment	Grp I treated with herbal mixture	Grp II treated with Fenbendazole	Grp III treated with Albendazole-Praziquantel
Hemoglobin % (gm/dl) change			
0^{th} day	7.92 ± 0.3^{a}	7.76 ± 0.18^{a}	7.67 ± 0.3^{a}
7^{th} day	9.08 ± 0.62^{b}	9.17 ± 0.25^{b}	9.25 ± 0.41^{bc}
14^{th} day	11.2 ± 0.22^{d}	10.25 ± 0.36^{cd}	10.43 ± 0.37^{d}
TEC (mil/mm^3)change			
0^{th} day	8.42 ± 0.32^{a}	8.32 ± 0.33^{a}	8.45 ± 0.29^{a}
7^{th} day	9.66 ± 0.57^{b}	9.83 ± 0.42^{b}	9.83 ± 0.42^{b}
14^{th} day	11.08 ± 0.25^{c}	10.58 ± 0.3^{bc}	11.08 ± 0.25^{c}
TLC (No./ mm^3)change			
0^{th} day	11400 ± 457.16^{c}	10966.67 ± 380.93^{c}	10633.33 ± 300.73^{c}
7^{th} day	9360 ± 417.85^{b}	9333.33 ± 338.29^{b}	8900 ± 428.95^{b}
14^{th} day	7860 ± 283.9^{a}	7833.33 ± 207.63^{a}	7650 ± 253.96^{a}
Total Protein (gm/dl) change			
0^{th} day	5.9 ± 0.36^{a}	5.37 ± 0.29^{a}	5.95 ± 0.25^{a}
7^{th} day	6.82 ± 0.11^{b}	6.58 ± 0.18^{b}	6.72 ± 0.15^{b}
14^{th} day	7.6 ± 0.14^{c}	6.9 ± 0.16^{b}	7.05 ± 0.11^{bc}
Serum AST (U/L) change			
0^{th} day	80.6 ± 1.29^{d}	78 ± 2.08^{cd}	81 ± 2.82^{d}
7^{th} day	55.6 ± 4.23^{a}	70.67 ± 2.76^{bc}	74.5 ± 2.14^{bcd}
14^{th} day	49.4 ± 1.44^{a}	68.67 ± 3.07^{b}	74.67 ± 1.28^{bcd}
Serum ALT (U/L) change			
0^{th} day	30 ± 0.84^{de}	31 ± 1.54^{e}	29.67 ± 1.72^{cde}
7^{th} day	26.2 ± 1.49^{bcd}	23.83 ± 0.79^{ab}	25.83 ± 1.62^{abc}
14^{th} day	21.8 ± 1.07^{a}	22.67 ± 0.67^{ab}	24.33 ± 1.31^{ab}
Serum BUN (mg/dl) change			
0^{th} day	16.2 ± 1.83^{b}	17.43 ± 0.97^{b}	17.88 ± 0.85^{b}
7^{th} day	11.76 ± 0.29^{a}	11.72 ± 0.31^{a}	11.82 ± 0.46^{a}
14^{th} day	11.82 ± 0.31^{a}	11.92 ± 0.3^{a}	11.65 ± 0.2^{a}
Serum Creatinine(mg/dl) change			
0^{th} day	1.29 ± 0.18	1.20 ± 0.15	1.22 ± 0.08
7^{th} day	1.15 ± 0.04	1.12 ± 0.06	1.11 ± 0.12
14^{th} day	1.16 ± 0.14	1.14 ± 0.07	1.12 ± 0.06
EPG change			
0^{th} day	1228 ± 37.02^{c}	1267 ± 50.17^{cd}	1275.67 ± 41.66^{cd}
7^{th} day	555.2 ± 98.07^{b}	303.67 ± 24.39^{a}	289.33 ± 22.15^{a}
14^{th} day	313.6 ± 46.11^{a}	270.33 ± 14.55^{a}	244.33 ± 18.15^{a}

Different small letters indicating significant ($p<0.05$) variation among treatment groups

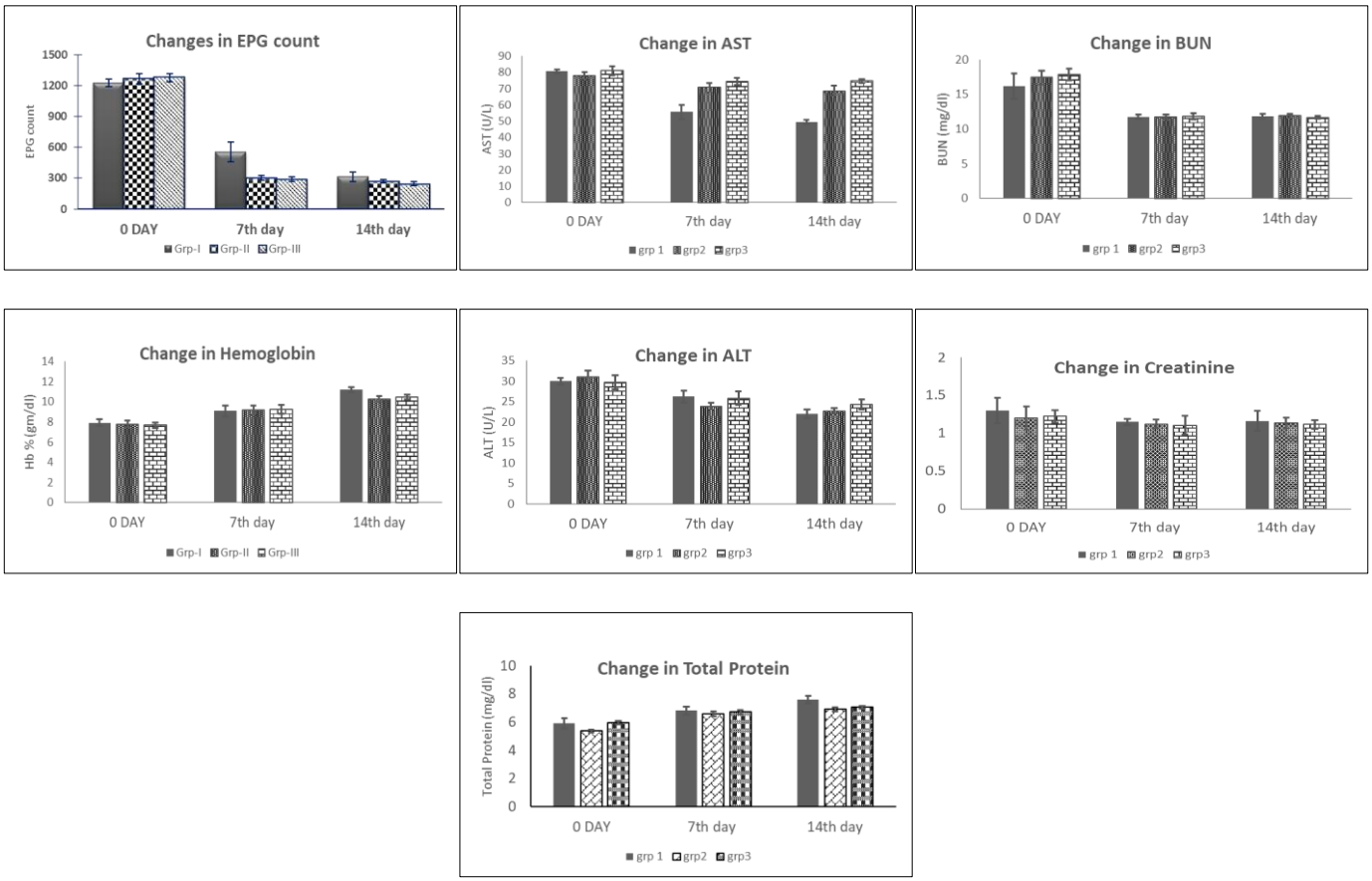

Figure 1: Graphical representation of change in parasite load (EPG), Hemoglobin (gm/dl), Hepatic biomarkers (AST and ALT, U/L), Urinary biomarkers (BUN and creatinine, mg/dl), and Total protein (mg/dl) of helminth infected goats after 14 days treatment with herbal mixture (Gr. I), Fenbendazole @7.5 mg/Kg body weight, single treatment (Gr.II) and Albendazole-Praziquantel combination @ 10mg/Kg body weight, single treatment (Gr.III)

DISCUSSION

Endoparasitemia in pre monsson period is a major health problem in grazing black Bengal goats of West Bengal [7]. The present study also recorded higher EPG count (>1000) and fulminating clinical signs of parasitemia in the selected experimental goats of the region. The hot-humid climatic condition along with low awareness about mitigation of endoparasitic problems among goat farmers might be the predisposing factors behind this observation. The herbal mix and commercial anthelmintic treatment effectively lowered the FEC (Fecal egg count) count in the goats at the end of the study period. It is also proved by the gross changing pattern of fecal solidity. It has been found that herbal mixture is capable to decrease the endoparasitic load as fast as compared to the market available drug establishing the ethnomedicinal practice in correct way. However, reduction of FEC was greater after first week of treatment in commercial anthelmintic group compared to herbal treatment group. Devi *et al.* [8] also showed that Piper nigrum based formulation showed better anthelminthic properties. Iqbal *et al.* [9] also showed that whole plant of Swertia can significantly reduce the hemonchus epg in ovine population. The result is also corroborated with the result of Ullah *et al.* [10] who opined for the anthelminthic property of Nigella sativa due to its active ingredient Thymoquinone.

The main detrimental effect of endoparasites is usually reflected through altered hemogram. The most prominent sign of this altered hemogram is anaemia and pale mucous membrane. Haemoglobin status and Total erythrocytic count are two important factor to judge the haematological status of any animal. The result shows that initially the Hb level and TEC was lower than average. Just after medication, as the parasites load were beginning to decrease, the Hb level started shifting towards normal range. It has been observed that up to one week the rise in the Hb level were almost in the same manner for all the three experimental group. The greater recovery rate of haemoglobin in the ethnomedicinal treatment group might be due to some other beneficial factor of the herb besides the decreasing parasitic load pattern. Total erythrocyic level also increased day by day simultaneously with the decreasing parasitic load but no specific alteration had been observed in that case like Hemoglobin. Hassan *et al.* [11] also reported that blood parameter like PCV, Hb and RBC were improved in black Bengal goat due to anthelminthic treatment.

White blood cells are protector of the body as well as the integral part of immune system. The difference in the white blood cell count is an important indicator for the invasion of pathogen in blood. Due to the chemokines secreted by the endoparasites initially the TLC value of all three groups were recorded elevated than normal. The revert of TLC count to normal from day 0 to day 14 in all the groups owe to the elimination of parasitic load in body system. Aktarujjaman *et al.* [12] also found similar pattern of change after treatment for endoparasites.

Hepatic enzymes are very much associated with the presence of pathogen, hemogram and amount of fluid present in body. Presence of

nematodes inside the body disturbs the normal metabolic process inside the body producing unwanted metabolic products. These malfunctions creates pressure on liver and thus alteration of hepatic enzymes may be observed [11, 13]. In ruminant, AST is more important than ALT as a hepatic enzyme. This study recorded the AST level of herbal dewormer treated group was significantly altered towards normal range than that of the synthetic commercial drug control groups. However the ALT levels between the all three different groups were recorded comparable at the end of treatment period. It has been found that incorporaton of black pepper (*Piper nigram*) can induce a positive effect on hepatic and immune status [14]. Moreover, the restoration of the hepatic enzyme level may be attributed by the hepatoprotective effect of *Swertia chirayata* due to the presence of its active components [15]. Thus the herbal dewormer may act as a bio immune enhancer as well as a dewormer to help a green goatery system.

Keeping the similarity with hepatic value, the Urea levels were also decreased in all three treatment group significantly at day 14 from pretreatment value. The metabolic disturbances due to the load of endoparasites may create the pressure on kidney having effect on this change initially. With the advancement of body condition, the BUN level came down to normal blood level, but creatinine had not shown any changes throughout the treatment period. The result is similar with previous report [16].

Most of the endoparasites including strongyles rapidly reduce the blood volume as well as protein reserve of the affected animal when present in a large number. Our results also showed that initially the total protein content of the serum were significantly lower level which gradually increases day by day with the advancement of decreasing parasitic load and increasing hemogram. Even at the end of day 14, the herbal treated group showed significantly superior protein status than other groups. It has been already reported that *Nigella* and *Swertia* can exhibit positive immunomodulatory effect [17, 18] while *Piper nigrum* L. as well as its active principle piperine, have a significant protective action against oxidative stress to cells [19]. Similar work done by another group of scientists [20] also proved that crude powder and cold extract of *Nigella*, *Swertia* and *Piper* significantly increased blood glucose, total protein and albumin in goats.

The selected villages are situated on the delta region of Ganges River. The soil are fertile for every spices and green vegetables. The spices and vegetables used in this study are traditionally cultivated and used by the villagers in a non-synchronous way. From the cost effective point of view almost zero cost is required to practice the old traditional herbal way to deworm the goats unlike the chemical dewormer. Moreover present findings suggest that herbal dewormer may be used as an alternative therapy which can potentially reduce the worm load within small ruminants like goat. It may enhance the immune status and body condition better than chemical as reflected by total protein and AST value of treated groups. Strong anthelminthic resistance in farm animals had been pointed out at different region of world due to indiscriminate use of chemical anthelminthics [21]. Potential targeted use of alternative herbal therapy may reduce the abrupt use of chemoprophylaxis. Moreover the tribal people who has natural apathy to visit the medicine store or veterinary dispensary for the ailment of livestock may easily practice this cost effective approach if the findings are properly disseminated and recommended among them by extension.

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