ORIGINAL ARTICLE



Anthelmintic efficacy of crude neem (*Azadirachta indica*) leaf powder against bovine strongylosis

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Abstract The present study was conducted to evaluate the anthelmintic efficacy of crude neem (Azadirachta indica) leaf powder against strongyle infections in cattle. Based on coproexamination, 30 cattle positive for strongyle infection with at least 250 [eggs per gram (EPG) of faeces] were selected and grouped as A, B and C (10 animals/group). Group A and B were treated respectively with fendendazole and neem leaf powder @ 5 and 500 mg/kg body weight, whereas Group C served as infected untreated control. Faecal sample from each animal of these groups was examined on day 0, 7, 14 and 28 post treatments and EPG was determined. The result showed significant decrease (p < 0.05) in EPG in Group A and B after day 7 post treatment but there was no significant variation in terms of EPG in control group. Thus it can be concluded that crude neem leaf powder has anthelmintic property and it can further be studied to isolate the active component to produce herbal anthelminthics.

Keywords Anthelminthic efficacy \cdot Cattle \cdot Neem leaf \cdot Strongyle

Introduction

Gastrointestinal (GI) parasitic infections are one of the major constraints for profitable dairy industry in tropical and

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subtropical countries including India. The economic impact of GI parasites in livestock industry includes both mortality and morbidity losses in terms of sub-optimum production of meat, milk, enhanced susceptibility to diseases, losses resulting from condemnation of carcasses, cost of drugs and veterinary aids (Rajakaruna and Warnakulasooriya 2011).

The control of GI nematodes is mainly based on the use of chemical anthelmintics (Waller 1987) but these are generally expensive and further not frequently available to farmers residing in rural areas. Furthermore, the development of resistance in helminths to various anthelmintic compounds (Waller and Prichard 1985) and chemical residue and toxicity problems (Kaemmerer and Butenkotter 1973) has worsen the condition. For these various reasons, interest in the screening of medicinal plants for their anthelmintic activity remains of great scientific interest (Akhtar et al. 2000).

In this regard, *Azadirachta indica*, commonly known as neem is well known for its medicinal use. It contains several biologically active constituents such as azadirachtin, meliantriol and salanin (Naganishi 1975; Lavie et al. 1967; Shin-Foon 1984). Of these constituents, azadirachtin has been demonstrated to have antimicrobial and antifungal properties (Kudom et al. 2011; Ramesh et al. 2011). Hence, the present study was designed to evaluate the anthelmintic property of crude neem leaf powder.

Materials and methods

Preparation of neem leaf powder

Mature neem leaves were collected from adult plant within the campus of College of Veterinary Science, Mhow (Madhya Pradesh). The leaves were collected in a polythene bag and brought to the laboratory for further

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processing. The neem leaves were dried under sun for 10 days at 8 h per day and grinded to make fine powder.

Selection and treatment of animals

A total of 30 strongyle positive cattle with eggs per gram (EPG) not less than 250 were selected from Dhar district of Madhya Pradesh. They were divided into three groups viz. A, B and C (10 animals/group). Group A was treated with fenbendazole (Panacur[®]) at a dose rate of 5 mg/kg body weight as a single oral dose. Group B was treated orally with a single dose of crude neem leaf powder @ 500 mg/kg body while the animals of Group C remained as infected untreated control. The approximate dose of neem leaf powder was calculated as per Ali (2005) who used similar dose rate and reported 53.6 % efficacy against *Gastrodiscus* in horse. The animals treated with neem leaf powder were kept under observation for 24 h. for any toxicity issue and no abnormal changes in their physiological parameters were detected.

Faecal sample examination

Faecal samples were collected directly from the rectum of cattle on day 0, 7, 14 and 28 post treatment. Faecal egg counts were determined by the modified McMaster technique using saturated sodium chloride solution as floating medium (Maff 1977) and EPG was determined by the following formula:

EPG = Total number of eggs in 2 chambers \times 50,

where, 50 was the dilution factor.

Data analysis

Statistical analysis was performed with the Statistical Programs for the Social Sciences (SPSS) version 16. Data satisfied the assumptions of the general linear model and were not transformed. Statistical significance of data was assessed by one-way analysis of variance (ANOVA). When ANOVA indicated significant effects (p < 0.05), the Tukey test was used to compare mean. Anthelmintic efficacy was

calculated by the faecal egg count reduction (FECR) test (Coles et al. 1992) according to the following formula:

$$= \frac{\text{Pre-treatment EPG} - \text{Post-treatment EPG}}{\text{Pre-treatment EPG}} \times 100$$

Results and discussion

The mean EPG of the Group A, B and C on day 0 was 400 ± 25.82 , 380 ± 19.44 and 395 ± 35.32 , respectively. After day 7 post treatment the mean EPG was recorded as 60 ± 19.44 , 85 ± 66.88 and 430 ± 37.42 , respectively. These results indicated a significant (p < 0.05) decrease in EPG in Group A and Group B at day 7 post treatment. On day 14 post treatment the EPG was recorded as 0 in Group A, 5 ± 5 in Group B and 420 ± 35.90 in Group C which revealed that on day 14 post treatment the animals of Group B showed significantly low (p < 0.05) EPG as compared to that of day 7 post treatment. No infection was detected in Group A and B at day 28 post treatment but Group C exhibited an EPG of 430 ± 24.94 . During the current study it was also observed that there was no significant (p > 0.05) changes in EPG of the animals of control group. Fenbendazole exhibited 85 and 100 % efficacy on day 7th and 14th of post treatment in Group A whereas crude neem leaf powder exhibited 78, 98 and 100 % efficacy on day 7th, 14th and 28th in Group B, respectively. The detailed findings of the present study have been shown in Table 1.

Anthelminthic efficacy of fenbendazole is well established and when it was compared with the crude neem leaf powder it has been shown that the leaves of *A. indica* have marked anthelminthic property though in crude form it is slow acting (Mahboob et al. 2008). The present study demonstrated the anthelminthic potential of *A. indica* leaves in control of GI parasites especially strongyle infection that has also been previously supported by other studies Neogi et al. 1964; Sharma et al. 1971; Kalesaraj 1974; Lal et al. 1976; Radhakrishnan et al. 2010. In one of the studies it was observed that cattle provided with feed blocks containing different levels of dried leaves of *A*.

Table 1 Anthelminthic efficacy of crude neem leaf powder against strongyle infection

Group	Anthelmintic	Dose	No. of	Route	Mean EPG)
		(mg/kg)	cattle treated		pre-treatment \pm SE (0 day)	7th day	14th day	28th day
А	Fenbendazole	5	10	Oral	$400^{a}\pm25.82$	$60^{\rm b} \pm 19.44 \; (85 \; \%)$	0 (100 %)	0
В	Neem leaf powder	500	10	Oral	$380^a\pm105.93$	$85^{\rm b}\pm 66.88~(78~\%)$	$5^{c} \pm 5 \; (98 \; \%)$	0 (100 %)
С	Control	-	10	_	$395^a\pm35.32$	$430^{a} \pm 37.42$	$420^a\pm35.90$	$430^{a}\pm24.94$

Means with same superscripts do not differ significantly (p < 0.05)

indica had significantly lower EPG as compared with the control cattle (Pietrosemdi et al. 1999). Further, studies in Philippines and Malaysia by various workers have also reported the efficacy of *A. indica* against nematodes of ruminants (Chandrawathani et al. 2002; Baldo 2001).

The antihelminthic property of neem probably due to the presence of an active alkaloid, azadirachtin, which interferes with the central nervous system of parasite via inhibition of excitatory cholinergic transmission and partly blocks the calcium channel resulting in expulsion parasites from host body (Qiao et al. 2013; Veerakumari and Priya 2006).

The anthelminthic activity of neem was associated with the alkaloids or other ingredients; hence, there is need for further studies in order to determine the active components, their lethal dose, appropriate route of administration as well as ascertain which particular parasite species or developmental stage are most susceptible to the effect of the extracts so as to further enhance their anthelmintic usefulness.

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References

- Akhtar MS, Iqbal Z, Khan MN, Lateef M (2000) Anthelmintic activity of medicinal plants with particular reference to their use in animals in the Indo Pakistan subcontinent. Small Rumin Res 38:99–107
- Ali M (2005) Prevalence and treatment trials against Gastrodiscus infestation and its effect on various blood components in horses. M.Sc. (Hons.) thesis, Deptt. Clin. Med. Surg., University of Veterinary and Animal Sciences, Lahore, Pakistan
- Baldo RC (2001) Comparative efficacy of pineapple (Ananas comosus) leaves bolus and albendazole against gastrointestinal nematodes of sheep. DVM. University of the Philippines, Department of Vet Parasitol, p 38
- Chandrawathani P, Brelin D, Nor FAS, Adnan M, Jamnah O, Rehana AS, Hoglund J, Waller PJ (2002) Evaluation of the neem tree (*Azadirachta indica*) as a herbal anthelmintic for nematode parasite control in small ruminants in Malaysia. Trop Biomed 19:41–48
- Coles GC, Bauer C, Borgsteede FHM, Geerts S, Walker PJ (1992) World Association for the Advancement of Veterinary Parasitology (WAAVP) methods for the detection of anthelmintic resistance. Vet Parasitol 44:35–44
- Kaemmerer K, Butenkotter S (1973) The problem of residues in meat of edible domestic animals after application or intake of organophosphatic esters. Residue Rev 46:1–240

- Kalesaraj R (1974) Screening of some indeginous plants for anthelmintic action against human Ascaris lumbricoides. Indian J Physiol Pharmacol 19:47–49
- Kudom AA, Mensah BA, Botchey MA (2011) Aqueous neem extract versus neem powder on *Culex quinquefasciatus*: implications for control in anthropogenic habitats. J Insect Sci 11:1–9
- Lal J, Chandra S, Sabir M (1976) In vitro anthelmintic of some indigenous medicinal plants on Ascaridia galli worms. Indian J Physiol Pharmacol 20:64–68
- Lavie D, Jain MK, Shpan-Gabrielith SR (1967) A locust phagorepellent from two Melia species. Chem Commun 18:910–911
- Maff (1977) Manual of Veterinary Parasitological Laboratory Techniques. Technical Bulletin No. 18, Ministry of Agriculture, Fisheries and Food, London, p 129
- Mahboob K, Khan JA, Khan MS (2008) Prevalence, chemotherapy and haematology of stronglyosis in horses of district layyah. J Anim Plant Sci 18:117–119
- Naganishi K (1975) Structure of the insect antifeedant azadirachtin. In: Runeckles VC (ed) Recent advances in phytochemistry, vol 5. Plenum, New York, pp 283–298
- Neogi NC, Baliga PAC, Shrivastava RK (1964) Anthelmintic activity of some indigenous drugs. Indian J Pharmacol 26:37–39
- Pietrosemdi S, Olavez R, Montilla T, Campos Z (1999) Empleo de hojas de Neem (*Azadirachta indica*; A. juss) en control de nematodes gastrointestinales de bovines a pastoreo. Revista de la Facultad de Agronomia 16:220–222
- Qiao J, Zou X, Lai D, Yan Y, Wang Q, Li W, Deng S, Xu H, Gu H (2013) Azadirachtin blocks the calcium channel and modulates the cholinergic miniature synaptic current in the central nervous system of Drosophila. Pest Manag Sci. doi:10.1002/ps.3644
- Radhakrishnan L, Gomathinayagam Balakrishnan V (2010) Evaluation of anthelmintic effect of Neem (*Azadirachta indica*) leaves on *Haemonchus contortus* in goats. Res J Parasitol 5:292–297
- Rajakaruna RS, Warnakulasooriya KN (2011) Gastrointestinal parasites in dairy cattle in Kandy district in Sri Lanka. Ann Res J SLSAJ 11:92–99
- Ramesh KS, Susan E, Devanand PF (2011) Bioactive constituents and antimicrobial activity of cell cultures of Azadirachta indica. Int J Pharm Bio Sci 2:617–628
- Sharma LD, Rhaga HS, Shristava PS (1971) In vitro anthelmintic screening of indigenous medicinal plants against *Haemonchus contortus* (Rudolphi, 1803) Cobbold., 1898 of sheep and goats. Indian J Anim Res 5:33–38
- Shin-Foon C (1984) The active principles and insecticidal properties of some Chinese plants, with special reference to Meliaceae. In Schmutterer and Asher, pp 255–261
- Veerakumari L, Priya P (2006) In vitro effect of azadirachtin on the motility and acetylcholinesterase activity of *Cotylophoron cotylophorum* (Fischoeder, 1901). J Vet Parasitol 20:1–5
- Waller PJ (1987) Anthelmintic resistance and future for roundworm control. Vet Parasitol 15:177–191
- Waller PJ, Prichard RK (1985) Drug resistance in nematodes. In: Campbell WC, Rcw RS (eds) Chemotherapy of parasitic infections. Phenum, New York, pp 339–362