

INVITRO AND IN-VIVO ANTHELMINTIC EFFECT OF SOME MEDICINAL PLANT PARTS AGAINST HAEMONCHUS CONTORTUS IN GOATS

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ABSTRACT

The results obtained in this study showed that Ethanolic extract of Neem leaves, Tulsi leaves and Garlic @10 mg/ml concentration (*in-vitro*) and @100 mg/kg (*in-vivo*) has significantly efficacious against *Haemonchus contortus*. *In-vivo* trial of Crude powder of Neem leaves, Tulsi leaves and Garlic bulb @ 150 mg/kg body weight was also found potent anthelmintic effect against naturally infected *Haemonchus contortus* in goats. From this experiment, it may concluded that cheap and easily available plant based dewormer helpful in poor farmers to reduce the losses in goat production, which occurs due to haemonchosis.

Key words: Anthelmintic effect, Medicinal plants, *Haemonchus contortus*, Goats.

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INTRODUCTION

Parasitic gastroenteritis caused predominately by *Haemonchus contortus* is a common health hazard in sheep and goats in India and is responsible for under productivity of these animals (Yadav *et al.*, 1992). Indiscriminate use of synthetic anthelmintics has contributed significantly to the development of resistance against *Haemonchus contortus* and other Gastro-intestinal helminths. Based on published literature, *Haemonchus contortus* seems to be the most involved species in case of anthelmintic resistance. Emergence of anthelmintic resistance along with toxic residual effect in animal products and exorbitant cost of synthetic anthelmintics need alternative approach i.e. plant based anthelmintics so as to overcome these hazardous effects and subsequently it may be sustainable, eco-friendly and biodegradable. Therefore, the present research plan was designed to evaluate the *in-vitro* anthelmintic activity of ethanolic extract and *invivo* anthelmintic efficacy of crude powder and ethanolic extract of Neem leaves, Tulsi leaves and Garlic bulb.

MATERIALS AND METHODS

Neem leaves, Tulsi leaves and Garlic bulbs plant parts were collected from their natural habitats and cleaned manually by removing the coarse impurity by hand and blowing the air to remove the dust and fine impurities. Then, these plant parts were shade dried in laboratory and further dried in incubator at 39°C for 6 hours to remove moisture. The dried leaves and bulbs were grinded in electric grinder machine at

room temperature to obtain coarse powder, which were used for extraction. Take 50 gm of coarse powder and soaked in 400 ml of the ethanol and stirred properly at every one hour interval in clean glass beaker covered with aluminium foil at room temperature. These were later filter through several layers of muslin cloth or Whatman filter paper and using separating funnels. Then after filtrated was concentrated by using Rotatory Vacuum Evaporator at 50-55°C. The extract residues were individually marked, kept in airtight glass petridishes in the cool and dark place at 4°C (refrigerator) for further use. For *invitro* trials, Adult *Haemonchus contortus* worms were separated from abomasums of goats which were collected from the local mutton shops. Abomasums were opened in a stainless steel tray and the abomasums contents were washed in tape water in laboratory several times until the sediment becoming transparent. Then, the adult *Haemonchus contortus* were collected with the help of a needle and placed in a petridish containing PBS (Phosphate Buffer Saline). Petri-dish containing the worms was kept in incubator at 38°C until required for experiment on the same day. Screening of Alcoholic extract of plant parts at various concentrations i.e. 25 mg/ml, 50 mg/ml and 100 mg/ml were performed in the 3 petridishes containing adult live *Haemonchus contortus* worms of goats in Phosphate Buffer Saline (PBS) whereas one petridish containing 30 adult worms in PBS kept as control to compare the exthanolic concentrations. Numbers of live & dead adult worms were counted at 1hr, 2hr and

3 hours interval and the percentage was calculated.

For *in-vivo* trial, 105 *Haemonchus contortus* naturally infected goats were selected and allocated into 7 groups each containing 15 goats.

Percent efficacy of was calculated as per the formula-

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

RESULTS AND DISCUSSION

The *in-vitro* efficacy of different concentration of Ethanolic extract of Neem leaves, Tulsi leaves and Garlic bulb viz. 25 mg/ml, 50 mg/ml and 100 mg/ml against 30 adult *Haemonchus contortus* were depicted in Table 1. The wormicidal effect of alcoholic extracts of these plants

at the concentrations of 25 mg/ml and 50 mg/ml was comparatively lower than that of concentration of 100 mg/ml. All these plant extracts were showed significantly higher wormicidal effects at 100 mg/ml concentration. Ethanolic extract of Neem leaves showed 100 % *in-vitro* efficacy during 3 hours observation. Sujon *et al.* (2008) also observed 100 percent *invitro* efficacy of ethanolic extract of Neem leaves against gastro-intestinal nematodes. Amin *et al.*(2009) has been observed 100, 100 and 84 percent *in-vitro* efficacy of aqueous extract of Neem leaves, Garlic bulb and Tulsi leaves, respectively against gastrointestinal nematodes in cattle. Anthelmintic effect of alcoholic extract of Tulsi leaves has been evaluated by Singh and Nagaich (2002) against nematodes in poultry. Iqbal *et al.* (2001a) have reported that extract of *Allium sativum* was found 100% *invitro* efficacy on 6 hour post application.

Table 1. Comparative *In-vitro* anthelmintic effect of Ethanolic extract of Neem leaf, Tulsi leaf and Garlic bulb against *Haemonchosis* in goats

Local name & Scientific name of Plant	Part used	Name of the worm	Concentration of alcoholic extract	Number of dead worms (%)		
				1hr	2hrs	3hrs
Neem (<i>Azadirachta indica</i>)	Leaf	<i>Haemonchus contortus</i>	25mg/ml	8	15	20 (67)
			50mg/ml	14	21	24 (80)
			100mg/ml	15	24	30 (100)
Control (PBS)	-	-do-	-	-	-	1
Tulsi (<i>Ocimum sanctum</i>)	Leaf	<i>Haemonchus contortus</i>	25mg/ml	6	11	17 (57)
			50mg/ml	8	14	20 (67)
			100mg/ml	9	16	24 (80)
Control (PBS)	-	-do-	-	-	-	2
Garlic (<i>Allium sativum</i>)	Bulb	<i>Haemonchus contortus</i>	25mg/ml	8	14	21 (70)
			50mg/ml	10	18	25 (83)
			100mg/ml	12	21	29 (97)
Control (PBS)	-	-do-	-	-	-	1

*Figure in parentheses indicate percentage

Crude powder of Neem was administered orally in naturally *Haemonchus contortus* infected goats and observed 67.41 percent efficacy on 15th Day Post-treatment (DPT) whereas feeding of Garlic bulb and Tulsi leaves powder revealed 56.47 and 54.02 percent efficacies, respectively on the basis of Egg Per Gram (EPG) reduction. The infected untreated control goats were continued to discharge the eggs of worms during the entire period of observation (Table 2). Jamar et al.(2014) also observed significant decrease in EPG in Strongyle infected

cattle after crude powder feeding of Neem leaves. Radhakrishnan et al. (2010) also observed significant decrease in Egg count as well as worm count in *Haemonchus contortus* infected goats. Worku et al. (2009) observed anthelmintic effect of Garlic juice. Oil of *Allium sativum* has also been reported to possess anthelmintic activity (Perry, 1980) and discards all injurious parasites in the intestine (Nadkarni, 1976). Kumar and Singh (2014) demonstrated that the dried clove powder of *Allium sativum* is potent source of helminthicide.

Table 2. Comparative *in-vivo* efficacy of Crude powder of Neem leaf, Tulsi leaf and Garlic bulb against *Haemonchus contortus* in goats

Local name & scientific name of Plant	Dose of crude powder	Name of the worm	Pre-treatment EPG (0-day)	Mean Post-treatment EPG (%)		
				5-day	10-day	15-day
Neem (<i>Azadirachta indica</i>)	@ 150 mg /kg body weight	<i>Haemonchus contortus</i>	593.33± 26.66	440 ±21.93	303.33 ±14.19	193.33 ±10.76 (67.41)
Tulsi (<i>Ocimum sanctum</i>)	-do	<i>Haemonchus contortus</i>	580 ± 23.29	473.33 ± 22.81	373.33 ±19.43	266.66 ±11.61 (54.02)
Garlic (<i>Allium sativum</i>)	-do-	<i>Haemonchus contortus</i>	566.66 ±21.63	466.67 ±22.18	373.33± 19.43	246.66± 18.55 (56.47)

*Figure in parentheses indicate percentage

On the basis of Egg Per Gram (EPG) count, naturally haemonchosis infected goats were treated with Ethanolic extract of Neem leaves @100 mg/kg body showed significantly 87.93 percent efficacy on 15 DPT whereas orally administration of ethanolic extract of Tulsi leaves and Garlic bulb at the dose rate of 100 mg/kg body weight were indicated 77.91 and 84.75 percent efficacies, respectively at end of observation. The infected untreated control goats continued to suffer from the infection with increased EPG counts during entire period of observation (Table 3).

Sujon *et al.* (2008) and Arora *et al.* (2010) have also been found significantly *in-vivo* efficacy of Ethanolic extract of Neem against gastrointestinal nematodes.

The results obtained indicate that these plant based anthelmintics may be used as strategic integrated control against *Haemonchus contortus* in goats to save the animals from the various disadvantages effects of chemical anthelmintics. The present study further need also research to estimate pharmacokinetics as well as its toxic effects.

Table 3. Comparative *in-vivo* efficacy of ethanolic extract of Neem leaf, Tulsi leaf and Garlic bulb against *Haemonchus contortus* in goats

Local name & scientific name of Plant	Dose of ethanolic extract	Name of the worm	Pre-treatment EPG (0-day)	Mean Post-treatment EPG (%)		
				5-day	10-day	15-day
Neem (<i>Azadirachta indica</i>)	@100mg/kg body weight	<i>Haemonchus contortus</i>	580 ±33.38	410 ±28.94	210 ±15.18	70 ±06.54 (87.93)
Tulsi (<i>Ocimum sanctum</i>)	-do-	<i>Haemonchus contortus</i>	543.33 ±23.33	366.66± 23.23	213.33± 15.01	120 ±09.51 (77.91)
Garlic (<i>Allium sativum</i>)	-do-	<i>Haemonchus contortus</i>	546.66 ±35.34	393.33± 34.45	230 ±25.72	83.33 ±07.97 (84.75)
Control goats	Infected untreated	<i>Haemonchus contortus</i>	593.33 ±20.62	703.33± 18.55	790 ±20.23	840 ±54.16

*Figure in parentheses indicates percentages.

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REFERENCES

Amin M R, Mostofa M, Hoque M E and Sayed, M A. 2009. *In vitro* anthelmintic efficacy of some indigenous medicinal plants against gastrointestinal nematodes of cattle. *J. Bangladesh Agril. Univ.* 7 (1): 57-61.

Arora N, Kumar A, Vihan V S and Sharma S D. 2010. Efficacy of indigenous plants against *Haemonchus contortus* infection in goats. *Indian Vet. J.* 87: 869-871.

Iqbal Z, Lateef M, Jabbar A, Ghayur M N, Gilani A H. 2006. *In-vitro* and *in-vivo* anthelmintic activity of *Nicotianan*

tabacum L. leaves against gastrointestinal nematodes of sheep. *Phytother Res.*, 20(1): 46-48.

Jamra N, Haque M and Das G. 2014. Anthelmintic efficacy of crude neem (*Azadirachta indica*) leaf powder against bovine strongylosis. *J. Parasit Dis.*

Kumar P, and Singh D K. 2014. *In-vitro* anthelmintic activity of *Allium sativum*, *Ferulaasa foetida*, *Syzygium aromaticum* and their active components against *Fasciola gigantica*. *J. Biol. Earth. Sci.* 4 (1): B57-B65.

Nadkarni K M. 1976. Indian Materia Medica, Vol. I. Pakistan. Popular Prakashan Pvt. Ltd, Bombay, India, pp.376- 377

Perry L M. 1980. Medicinal Plants of East and Southeast Asia. M.I.T. Press, London, pp.164-165.

Radhakrishnan L, Gomathinayagam S and Balakrishnan V. 2010. Evaluation of

anthelmintic effect of Neem leaves on *Haemonchus contortus* in goats. *Res. J. Parasitol.* 5 (4). 292-297.

Sujon M A, Mostofa M, Jahan M S, Das A R and Rob S. 2008. Studies on medicinal plants against gastrointestinal nematodes of goats. *Bangl.J. Vet.Med.* 6(2):179-183.

Worku M, Franco R and Baldwin K. 2009. Efficacy of Garlic as an anthelmintic in adult Boer goats. *Arch. Biol. Sci., Belgrade,* 61 (1), 135-140.

Yadav C L, Uppal, R P and Prerna. 1992. Clinical trail with closantel against *Haemonchus contortus* in kids. *Indian Vet.J.* 69:742-745.

