Pakistan Journal of Pharmaceutical Sciences
Vol.8(1), January 1995, pp.17-27

ANTIFASCIOLIC EFFICACY OF INDIGENOUS PLANT DRUGS: *KALONJI SHAHTERAH* AND *KARANJWA* IN BUFFALOES

SHAFEEQ-UR-REHMAN KAILANI, MUHAMMAD SHOAIB AKHTAR[a] AND MUHAMMAD ASHRAF
Department of Medicine and Surgery, College of Veterinary Science, Lahore
'Department of Physiology and Pharmacology,
University of Agriculture, Faisalabad-38040, Pakistan

ABSTRACT

A study was carried out to evaluate antifasciolic effect of powdered plant drugs including Kalonji (*Nigelia sativa* seeds), Shahterah (*Fumaria parviflora* aerial parts) and Karanjwa (*Caesalpinia crista* seeds) in buffaloes. During the search for the infected animals, prevalence of fascioliasis in the random samples was 33.1 ± 0.7% but in symptomatically suspected cases it came to be 64 ± 3.2%. The drug trial results showed that all the three drugs possess significant efficacies against fascioliasis. Their highest doses tested produced highly significant ($P < 0.001$) decrease in EPG counts on 15th days. Maximum antifasciolic efficacy, judged on the basis of % EPG count reduction was shown by Shahterah (93.2 ± 05%) which was followed by Karanjwa (89.7 ± 1.7%) and Kalonji (88.2 ± 0.4%). No visible side effects were produced by any of these plant drugs. Single oral treatment with 25 mg/kg of Kalonji or 60 mg/kg of Shahterah and 40 mg/kg of Karanjwa exerts highly significant antifasciolic efficacies on the day 15 after treatment.

From these data, it is conceivable that at these dosages the drugs are sufficiently potent and safe to treat fasciola infection in buffaloes. However, further chemical and pharmacological studies would decide the exact mechanism(s) of action, the active principles contained and the real worth of these indigenous drugs for the treatment of fascioliasis in the ruminants.

Introduction

The incidence of fascioliasis has markedly increased due to the existence of large water reservoirs, network of rivers and canals, rapid irrigation-dam projects, inadequate veterinary education in the livestock owners and improper agricultural planning in this country. Michel et al. (1981) has recommended the regular use of anthelmintics to control the infection but Gilson (1969) has reported that most modern synthetic anthelminitics are ineffective against immature flukes and the resistant strains.

Moreover, Booth et al. (1988) have recorded that these drugs are neither completely effective against common flukes and other parasites nor they retain their efficacies on their continuous administration due to the development of resistance. In addition, al-most all of them adversely affect the milk and meat production of animals during the course of their treatment and even long after their use (Brander and Pugh, 1991).

Plants have been used from ancient times to cure diseases and it is not surprising, therefore, that several medicinal plants have been used to treat fascialiasis as well (Satyavati et at, 1987). Amongst such plant drugs Kalonji *Nigella sativa* seeds have been frequently prescribed as an anthelmintic in the traditional medicine (Said, 1%9) Similarly other plants including Shahterah, (*Fumaria parviflora* aerial parts) and Karanjwa (*Caesalplaia crista* seeds), have also been used to treat worm infestations (Nadkarni, 1954; Akhtar, 1988). As far as ascertained, no systematic research work has been conducted to prove their efficacy against fascioliasis in animals. The present research was, therefore, designed to study the antifasciolic activity of the above three indigenous plant drugs in local buffaloes (*Bos bubalis*) under field conditions.

Materials and Methods

Indigenous Plant Drugs Screened

Kalonji *(Nigelia sativa* seeds), Shahterah *(Fumaria par*v*ifl*o*ra* aerial parts) and Karanjwa (*Caesalpinia crista* seeds) were purchased from the local herbal market of Lahore in the dried form. Electric grinder was used to grind all the materials and their powders were stored in air-tight glass bottles at 4°C.

Animal Used:

Buffaloes of mixed breed, irrespective of sex and age were used from different privately owned flocks for initial sampling. Seventy two buffaloes found positive for fascioliasis through coprological examination were selected for the drug-trials. These animals were found to have been usually kept under poor managemental and hygienic conditions by their owners.

Preparation and Administration of Plant Drugs:

The herbal drugs were made dust-free and further dried under the shade for a few days. They were then finely powdered with an electric grinder. Gum tragaeanth also finely powedered, its 2% w/v aqueous solution was prepared and stored in refrigerator at 4°C. At the time of medication, calculated amounts of the powedered drug were weighed according to the dosage level. The weighed amount of drug suspended in 100 ml of 2% gum solution and was given orally by using a drenching gun. However, Shahterah (*Fumaria parviflora*) was suspended in 300 ml of the gum solution because of its low

solubility and higher dosage level. The dosages of drugs were selected on the basis of preliminary trials in buffaloes and keeping in mind the doses used in human beings as anthelmintic in traditional medicine.

Grouping of Animals:

The selected buffaloes were divided into three (3) main groups, K, S and Kr of 24 each. The three groups were further divided into four sub-groups of 6 animals each. The first three sub-groups received the test drug in 3 different doses while the fourth one secured as untreated control. The group-K was subdivided into K_1, K_2, K_3 and K_0. The first three of these were treated with graded doses of Kalonji while K_0 received only the gum solution. Similar, groupings were done in group-S and group-Kr animals which received Shahterah and Karanjwa, respectively. Eggs per gram (EPG) counts were made on zero (0) day before medication and then on 3rd, 10th and 15th days post-medication. Numberings of animals were marked on their backs with white paint for their proper identification.

Collection of Faecal Samples:

Faecal samples were collected during September, 1988 to February, 1988 and from September, 1989 to December, 1989 from Harbanspura and Gwala Colony, Lahore. Faecal sample were taken from the freshly passed faeces in plastic envelops and were properly labelled. Samples of the diarrhoeic buffaloes were also collected in separate containers and the percentages of the disease in normal and diarrhoeic cases were calculated.

Coprological Examination and EPG Counting

At first faeces were examined to detect samples positive for the eggs of genus Fasciola. A small quantity of faeces was taken soaked in tap water and its semi-suspension prepared which was spreaded over a microscope slide. Covered it under a cover slip and examined under low power of the microscope. Presence of operculated, thin-walled, bile tinged egg(s) confirmed the fascioiasis. All the -ve samples were re-examined after centrifugation a~ described in detail by Soulsby (1982). About 5-10 grams of faeces were mixed with water and the suspension was strained. The filterate was centrifuged (500 RPM) for 3-5 minutes. The supernatent was gently discarded off and sediment stirred homogenously. Few drops of the sediment were applied to a slide and added to it one drop of 1% aqueous of methylene blue mixed well and spreaded over the object glass, examined under low power lens (without cover slip). This stained the suspension blue, while the eggs, if present, remained yellowish brown and hence recognised easily. The antifasciolic efficacy of drugs was determined on the basis of percentage reductions in post-medication EPG counts by the zinc sulphate floatation technique using a McMaster slide (Soulsby, 1982).

Statistical Analysis:

Faecal EPG counts were expressed as Means ± SEM (Standard Error of Means). Percentage EPG reductions values on post-treatment days 3, 10 and 15 were calculated by the following formula:

$$\frac{\text{Reduction in EPG counts after treatment}}{\text{EPG counts before treatment}} \times 100 = \% \text{ age reduction}$$

Two way analysis of variances technique was used to ascertain significance of the data. LSD technique was used if F ratio was greater than 0.05, 3.9, 3.86 (Snedecor and Cochran, 1980).

Results

Prevalence of Fascioliasis:

The data showed that faecal samples of apparently normal buffaloes collect during the check period had 33.1 2 0.7 percent were positive. The data also showed that incidence of fascioliasis in buffaloes suffering from chronic diarrhoea was 64 ± 3.2 percent.

Efficacy of Kalonji (Nigella saliva):

As shown in Table 1, EPG count increased in untreated control group (K_0) on 10th and 15th days. However, treatment with 20 mg/kg body weight of Kalonji ca a significant ($P<0.05$) decrease in EPG counts on post-treatment day 3 and highly significant ($P<0.001$) decrease on days 10 and 15. At 25 mg/kg body weight, a highly significant ($P<0.001$) decrease in EPG counts occurred on all days checked. Their %age reductions were 52.6 ± 2.5, 70.7 ± 1.7 and 88.2 ± 0.4, respectively. The F ratios indicate that both the dosage levels and post-treatment days affect significantly the efficacy of the drug which was 6.15 for the dosage levels and 6.11 for past-treatment days.

Efficiacy of Shahterah (Fumaria parviflora):

Table 2 shows that Shahterah at 40 mg/kg dose has caused a significant (P c decrease in EPG counts on day 15 while the drug at SO mg/kg caused a sign decrease in EPG counts on day 3 and highly significant ($P<0.001$) on days 10 Its 60 mg/kg body weight produced 64.4 t 3.1, 835 ± 1.3 and 93.2 ± 0.5 % reductions on days 3, 10 and 15.

Efficiacy of Karanjwa (Caesalpinia crista)

The powedered Karanjwa seeds at 30 mg/kg body weight caused a highly significant ($P < 0.001$) decrease in EPG counts on post treatment days 10 and 15. At 35 mg/kg, the drug caused a significant decrease i.e. 33.7 ± 1.7% reduction on 3rd day while highly significant reductions i.e. 50.6 ± 2.5 and 75.9 ± 3.9% on days 10 and 15. The 40 mg/kg

dose caused 52.8 ± 1.5, 74 s 2 and 89.7 ± 1.7 percentage EPG reductions, respectively on the days checked (Table 3).

Relative efficacy of all the three drugs at their optimum levels $_{(L3)}$ on various days as compared with L_0 (Zero drug) shows that at L_3 all the drugs had highly significant ($P< 0.001$) antifasciolic effect and highest efficacy (on the basis of maximum %age EPG reductions) was shown by Shahterah i.e., 93.2 ± 0.5%, while efficacies of Karanjwa and Kalonji were 89.7 ± 1.7% and 88.2 ± 0.4%. Thus they are on second and third positions, respectively.

Discussion

The helminth parasites have been reported to cause loss in body weight, decreased feed consumption, reduced growth rate, delayed maturity, poor body condition, general unthriftiness and anaemia in animals (Soulsby, 1982). Moreover, their incidence has been found to be heaviest in Pakistan and other asian countries (Durrani and Hayat, 1964; Dhar et al., 1982). Modern fasciolicides are efficious but most of them possess several adverse effects (Levine, 1982). Hence development of newer, safe, curative and economical antifasciolic drugs has remained an active area of re-search. Thus anti-fasciolic activities of indigenous drugs including Kalonji *(Nigella sativa)* seeds, Shahterah *(Fumaria parviflora*) aerial parts and Karanjwa *(Caesalpinia crista*) seeds were evaluated.

Buffaloes suffering from chronic fascioliasis *were* purposely included in the study as eggs appear in their faecal samples only (Blood and Handerson, 1974). During search for positive cases, prevalence of the disease was 33.1 ± 0.7% in random samples and 64 ± 3.2% in buffaloes suffering from chronic diarrhoea. These findings are in agreement with Hayat et al. (1986), Maqbool and Irfan (1983) and Saleem et al. (1986). The immature flukes in the liver parenchyma become mature in 2-3 weeks (Saleem et al., 1986). In addition, as shown in Tables 1, 2 and 3, Mean EPG counts were increased in the untreated control on all the *days* checked due to maturation of flukes in the liver of animals. Hence, EPG counts were made uptill 15th day after treatment.

Kalonji seeds in 15 mg/kg body weight dose caused highly significant ($P<0.001$) %age EPG reductions on post-treatment days 10 and 15. However, its 20 mg/kg dose caused significant ($P < 0.05$) decrease of EPG counts on day 3 and highly significant on 10th and 15th days as compared to 0 day. The 25 mg/kg dose of this drug caused highly significant EPG reductions on all the days checked (Table 1). These findings are in agreement with Akhtar (1988) and Akhtar and laved (1991) against ovine paramphistomiasis and Monieziasis in sheep, respectively. Akhtar and Farah (1986) and Nath (1983) have reported that the drug contains nigelline, metarbin, melanthia,

melanthiginin and sapginin, etc. In addition, it contains fixed oils (37.5%) and volatile oils (1.5%) which delay its passage and ultimately their on-set of action (Ghaznavi, 1988). Therefore, maximum efficacy of the drug has been recorded on post-treatment day 15.

Shahterah aerial parts, as shown in Table 2, have also consistently decreased the EPG counts. Highly significant ($P<0.001$) EPG reductions were observed at its 50 mg/kg body weight dose on the days 10 and 15 when the %age reductions were 62.8 ± 2 1.9 and 75.4 ± 2 2.7%, respectively. However, its 60 mg/kg dose produced highly significant reductions on all post-treatment days checked. Akhtar and Farah (1986) have reported that this drug contains on dry matter basis 4% alkaloids which might be active principles. Similar, efficacies of Shahterah against the nematodes have reported previously by Akhtar and Javed (1985).

Karanjwa seeds, as given in Table 3, in 30 mg/kg dose achieved EPG reductions 25.3 3 1.1, 44.2 ± 1.9 and 56.5 ± 1.3 on days 10 and 15, respectively. At 35 mg/kg, caused a significant ($P < 0.05$) decrease in egg counts on day 3 and highly significant ($P < 0.001$) on days 10 and 15 after medication when the %age reductions were 33.7 ± 1.7, 50.6 ± 2.5 and 75.9 ± 3.9%, respectively. The 40 mg/kg dose caused 52.8 ± 1.5, 74.0 ± 2.1 and 89.7 ± 1.7% EPG reductions, respectively which were found to be highly significantly decreased on all the days checked. These data are in accordance with Akhtar et al. (1985) and Akhtar and Aslam (1989) who have also reported its efficacy against nematodes in buffaloes and sheep.

The results obtained have clearly indicated that the drug exert gradual effect upon the immature flukes and the peak effect is achieved on 15th day after treatment. This may be due to the fact that plant drugs generally take more time to be dissociated, emulsified, absorbed and excreted from the body. Also, the test plants as already cussed have been reported to possess high percentage of alkaloids which due to weak basic nature ionize in the acidic gastric juice. Thus in the investigation pro a small portion of these drugs must have dissociated in intestines on the days 3 a after medication. Therefore, less efficacies were obtained on these days as cons on 15th days. Virtually, all the test plants did not produce any visible signs of toxicity or side effects at the dosage levels checked. In addition, the cost of a single dose treatment of these plant drugs at the maximum effective dosage levels for an average buffaloe came to be Pak Re.1, indicating that the drugs are much more econ than the imported synthetic antifasciolic drugs. In conclusion, it is conceivable from the above discussion that Kalonji, Shahterah and Karanjwa are potent and safe to treat fasiohasis in the buffaloes. The study do seem to support the empirical use of these plants drugs as a deworming agent in traditional medicine (Nadkarni, 1954; Said 1969). However, further chemical and pharmacological studies will decide their exact mechanisms of action and real worth as anthelmintics in other ruminant species and infections.

Table 1

Mean ± SEM values, calculated regarding faecal eggs per gram (EPG) counts and their percentage reductions in buffaloes, suffering from natural fascioliasis, treated orally with powdered seeds of *Nigella sativa* (Kalonji) at the various dosage levels.

Treat-ments	Dosage (levels)	Pre-treatment E.P.G. Counts (Zero day)	Post-Treatment EPG Counts on			Percentage EPG Reductions on		
			3rd day	10th day	15th day	3rd day	10th day	15th day
Control	-	1600 ± 36.52	1600 ± 36.52	1650 ± 42.82	1700 ± 77.46	-	-	-
Nigella sativa	15 mg/kg body wt.	1500 ± 68.31	1100 ± 44.72	850 ± ** 92.2	650 ± ** 50.00	26.57 ± 0.77%	43.46 ± 5.1%	56.65 ± 2.71%
do	20 mg/kg body wt.	1600 ± 33.1	1000 ± 68.31	750 ± ** 95.74	400 ± ** 25.82	37.63 ± 1.54%	54.03 ± 3.37%	74.88 ± 1.36%
do	25 mg/kg body wt.	1700 ± 57.74	800 ± ** 25.82	500 ± ** 36.52	200 ± ** 0.00	52.56 ± 2.54%	70.65 ± 1.71%	88.17 ± 0.41%

Number of buffaloes in each subgroup = 6

*Significantly ($P < 0.05$) decrease in egg count value as compared to the decrease in control.

**Highly significant ($P < 0.001$) decrease in egg count value as compared to the decrease in control.

Table 2

Mean ± SEM values, calculated regarding faecal eggs per gram (EPG) counts and their percentage reductions in buffaloes, suffering from natural fascioliasis, treated orally with powdered aerial parts of *Fumaria paruiflora* at the various dosage levels.

Treat-ments	Dosage (levels)	Pre-treatment E.P.G. Counts (Zero day)	Post-Treatment EPG Counts on			Percentage EPG Reductions on		
			3rd day	10th day	15th day	3rd day	10th day	15th day
Control	-	1200 ± 73.03	1200 ± 60.55	1200 ± 57.73	1200 ± 60.55	-	-	-
Fumaria parviflora	15 mg/kg body wt.	1100 ± 45.81	950 ± 22.36	700 ± 89.44	600 ± * 89.44	13.33 ± 1.49%	36.94 ± 7.21%	46.11 ± 7.36%
do	50 mg/kg body wt.	1200 ± 77.46	750 ± * 61.91	450 ± ** 42.81	300 ± ** 44.72	37.8 ± 1.47%	62.76 ± 1.86%	75.43 ± 2.73%
do	60 mg/kg body wt.	1500 ± 109.54	550 ± ** 84.65	250 ± ** 34.15	100 ± ** 0.00	64.35 ± 3.08%	83.52 ± 1.33%	93.15 ± 0.51%

Number of buffaloes in each subgroup = 6

*Significantly ($P < 0.05$) decrease in egg count value as compared to the decrease in control.

**Highly significant ($P < 0.001$) decrease in egg count value as compared to the decrease in control.

Table 3

Mean ± SEM values, calculated regarding faecal eggs per gram (EPG) counts and their percentage reductions in buffaloes, suffering from natural fascioliasis, treated orally with powdered *Caesalpinia crista* seeds (Karanjwa) at various dosage levels.

Treat-ments	Dosage (levels)	Pre-treatment E.P.G. Counts (Zero day)	Post-Treatment EPG Counts on			Percentage EPG Reductions on		
			3rd day	10th day	15th day	3rd day	10th day	15th day
Control	-	1700 ± 93.1	1750 ± 92.19	1800 ± 73.02	1800 ± 63.24	-	-	-
Caesal-pinia crista	30 mg/kg body wt.	1600 ± 73.02	1200 ± 73.02	900 ± ** 73.02	700 ± ** 51.64	25.25 ± 1.09%	44.19 ± 1.91%	56.49 ± 1.32%
do	35 mg/kg body wt.	1800 ± 85.63	1200 ± * 85.63	900 ± ** 85.63	450 ± ** 80.62	33.74 ± 1.7%	50.6 ± 2.54%	75.89 ± 3.93%
do	40 mg/kg body wt.	1900 ± 51.64	900 ± ** 51.64	500 ± ** 51.64	200 ± ** 36.5	52.84 ± 1.5	73.97 ± 2.1%	89.67 ± 1.7%

Number of buffaloes in each subgroup = 6

*Significantly ($P < 0.05$) decrease in egg count value as compared to the decrease in control.

**Highly significant ($P < 0.001$) decrease in egg count value as compared to the decrease in control.

References

Akhtar M.S. and Javed I. (1985). Comparative efficacy of Humana *parvffiora* and morantel tartrate against mixed gastrointestinal nematode infection in sheep. *Pak. J. Pharmacol.* **2**: 31-35.

Akhtar M.S., Javed I., Hayat C.S. and Shah B.H. (1985). Efficacy and safety of *Caes-salpinia crista,* Linn. seeds, its extracts in water and methanol against natural *Neoascasis vindonun* infaction in buffalo-calves. *Pak. Vet. J.* **5**: 192-196.

Akhtar M.S. and Farah N. (1986). Phytochemical screening of *Morus alba* (stem bark *Nigella sativa* (seeds) and *Fumaria parviflora* (aerial parts). *J. Pharm. Univ. Kar*. **5** 9-14.

Akhtar M.S. (1988). Anthelmintic Evaluation of Indigenous Medicinal Plants for Veterinary usage. Final Research Report. pp.172. University of Agriculture, Faisalabad.

Akhtar M.S. and Aslam M. (1989). Field trial of glycosides of karanjwa seeds against nematodes in sheep. *Pak. J. Agri. Res*. **10**: 175-178.

Akhtar M.S. and Javed I. (1991). Efficacy of *Nigella sativa* Linn. seeds against Moniezia infection in sheep. *Ind. Vet. J.* **68**: 726-729.

Blood D.C. and Handerson J.A. (1974). Veterinary Medicine, 4th Ed. English Language Book Society, Bailliere Tindall, London.

Booth N.H. and McDonald L.E. (1988). Veterinary Pharmacology and Therapeutics, 6th Ed. Iowa State Univ. Press, Ames.

Brander G.C., Pugh D.M. and Bywater R.I. (1991). Veterinary Applied Pharmacology and Therapeutics, 4th Ed. Bailliere Tindall, London.

Dhar D.N., Sharma R.L. and Buccal G.C. (1982). Gastrointestinal parasitism in sheep in Kashmir. *Vet. Parasitol.* **11**: 271-277.

Durrani M.Z. and Hayat C.S. (1964). Gastrointestinal parasitism in sheep and goats in Lyallpur (Faisalabad) District. Proc. 17th Pak. Sci. Conf.

Ghaznavi K. (1988). *Tibb-i-nabvai (Peace Be Upon Him) our Jadded Science* (Urdu) Faisal Publishers Urdu Bazar, Lahore. **1**: 228-232.

Gilson T.E. (1969). Advances in Parasitology cited by Anderson, N. and Dobson R.J. (1975). Efficiency related to strategic control schemes for helminthiasis in sheep. *Aust. Vet. J.* **51**: 67-70.

Hayat C.S., Igbal Z., Hayat B. and Khan M.N. (1986). Studies on the season prevalence of Fascioliasis and lungworm disease in sheep at Faisalabad. *Pak Vet. Journal.* **6**: 131-134.

Maqbool M. and Irfan M. (1983). Effect of different fasciolicides against fascioliasis in buffaloes. *Pak. Vet.* **13**: 70-72.

Michel J.E., Latham J.O., Chureb B.M and Deech P.K. (1981). Use of anthelmintics for cattle in England and Wales during 1978. *Vet. Rec.* **108**: 252-258.

Nadkarni A.K. (1954). *Indian Materia Medica.* Vol.I. Popular Book Depot. Bombay.
Nath V.T. (1983). "Ayurvedic Jari Bootian" (Urdu) Matboaat-i-Sulemani, Urdu Bazar, Lahore. pp.523, 526, 529-30.
Said M. (1969). Hamdard's Pharmacopeia of Eastern Medicine. Times Press, Karachi.
Salem M.E., Mian M.S., Rabbani A. and Afzal M. (1986). A study on the incidence, chemotherapy and blood-picture in Ovine fascioliasis. *Pak. Vet. J.* **6**: 157.
Satyavati G.V., Gupta A.K. and Tandon N. (1987). Medicinal Plants of India, Indian Council of Medical Research, New Delhi, 201-206 pp.
Snedecor G.W. and Cochran W.G. (1980). Statistical Methods (6th ed.). Iowa State University Press, Ames. Iowa, U.S.A.
Soulsby E.T.I. (1982). Helminths, arthropods and protozoa of domesticated animals, English Language Book Society, London.