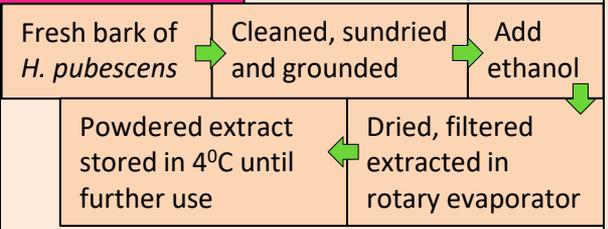


INTRODUCTION

Local Rajbanshi and Koch communities of Cooch Behar, West Bengal have been using plant materials as curative for various infections. In current study the bark of *Holarrhena pubescens* (Buch.Ham), Family- Apocynaceae, has been used to evaluate its anthelmintic effect through ultrastructural studies and histochemical localization of tegumental enzyme on model tapeworm, *Raillietina* sp.

MATERIALS

Plant extract Preparation:



Parasite Collection: Live model parasites were collected in 0.9% Phosphate Buffer Saline (PBS) from the intestine of freshly slaughtered host (*Gallus gallus domesticus*) at local abattoirs.



Fig 1. Host



Fig 2. *Raillietina* sp.

METHODS & RESULTS

Treatment:

The worms were incubated at $37 \pm 1^\circ\text{C}$ in PBS containing various dosages of the plant crude extract in triplicate. Praziquantel (PZQ), was used as the reference drug. One set of control parasite was maintained only in PBS for each concentration.

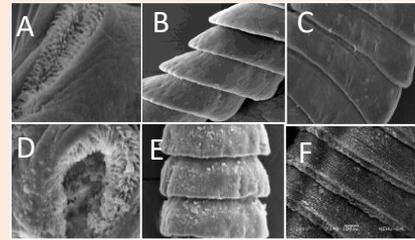
Results: Table 1. Observations of efficacy testing

	Dose	Time(h) of Paralysis	Time(h) of Death
Control	PBS		70 \pm 0.8
Praziquantel			0.04 \pm 0.4
Plant Extract	10 mg/ml	1.16 \pm 0.10h	2.75 \pm 0.6h
	5 mg/ml	2.33 \pm 0.5h	3.83 \pm 0.4h
	2 mg/ml	5.83 \pm 0.5h	13.75 \pm 0.5h
	1 mg/ml	11.66 \pm 0.6h	27.5 \pm 0.4h

Scanning Electron Microscopy (SEM): Control and treated parasites were fixed in 10% Neutral Buffer Formalin (NBF), dehydrated with ascending grades of acetone (30%-100%), coated with gold and examined through JSM-6360, Jeol scanning electron microscope.

Result: Fig3: Tegumental alteration observed through SEM

Control Parasite (A-Head, B & C- Proglottids)
 Plant extract treated Parasite (D-Head, E & F- Proglottid)



Histochemical Analysis: Control and treated worms were fixed and sectioned in a cryomicrotome and stained following standard protocols for each enzyme.

Result: Tegumental enzymes showed a marked reduction in their activities after treatment.

Fig 4-AcPase

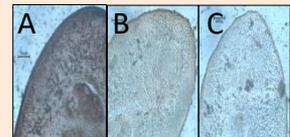


Fig 5-AlkPase

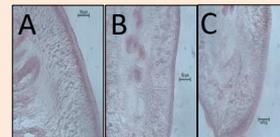


Fig 6-ATPase

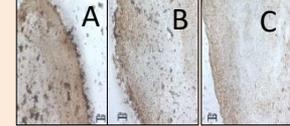


Fig 7- 5'Nu



Fig 4,5,6,7: Sections A-Control, B- Plant extract treated, C-PZQ treated parasite's proglottids.

CONCLUSION

Efficacy tests, SEM analysis and histochemical studies show that the stem bark crude extract of *H.pubescens* has anthelmintic potential.

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