

Molecular characterization of veterinary important trematode and cestode species in the mithun *Bos frontalis* from north-east India

J.K. Chamuah², O.K. Raina^{1*}, H. Lalrinkima¹, S.S. Jacob¹,
M. Sankar¹, A. Sakhrie², S. Lama² and P.S. Banerjee¹

¹Division of Parasitology, Indian Veterinary Research Institute, Izatnagar, India; ²National Research Centre on Mithun, Jarnapani, Nagaland, India

(Received 2 March 2015; Accepted 3 August 2015)

Abstract

Helminth infections in the mithun *Bos frontalis*, including the liver fluke *Fasciola gigantica*, hepato-gastric amphistomes *Explanatum explanatum*, *Paramphistomum epiclitum* and *Calicophoron calicophorum*, and the cestodes *Echinococcus granulosus* and *E. ortleppi* were studied in north-east India over a 2-year period from 2012 to 2014. Cystic echinococcosis caused by *E. granulosus* and *E. ortleppi* was found to be highly prevalent in the mithun, with *E. ortleppi* being reported for the first time. Molecular markers, including the internal transcribed spacer 2 (ITS-2), 28S rDNA and mitochondrial NADH dehydrogenase sub-unit1 (*nad1*) were used to confirm the identification of the trematode and cestode species.

Introduction

The rare bovine species known as the mithun *Bos frontalis* is part of a rich biodiversity in the forests of north-east India and in parts of Bhutan, Myanmar, China and Bangladesh (fig. 1). The mithun is generally found at an altitude of 1000–3000 metres above mean sea level and is well adapted to this environment (Chaurasia *et al.*, 2010). It plays an important role in the socio-economic and cultural life of the tribal population of this region, being used primarily as a beef animal. The mithun is raised under free-range conditions in its natural habitat and, as a result, no systematic methodology is adopted for the field diagnosis of the sub-clinical parasitic infections in the animal. Furthermore, the aetiological agents of the parasitic diseases have not been adequately recorded. No systematic studies on the epidemiology, genetic characterization and diagnosis of the helminths affecting the mithun have been conducted. Sporadic studies done on the prevalence of different parasites could not lead to the formulation of strategic control

measures on parasitic diseases of this rare species. Also, identification of the helminths has been based on conventional methods of morphometry, and characterization based on molecular tools like ribosomal DNA, internal transcribed spacers and mitochondrial genes, providing reliable genetic markers for differentiation of the parasite species/strains, has not been done. The present study focuses on the occurrence and identification of amphistomes, liver fluke and cystic echinococcosis prevalent in the mithun, using 28S ribosomal DNA, internal transcribed spacer 2 (ITS-2) and mitochondrial *nad1* as molecular markers.

Materials and methods

Collection and examination of helminths

Post-mortem examinations were conducted on 90 mithun slaughtered at various tribal rituals and ceremonies in Arunachal Pradesh and Nagaland, where infections with the trematodes *Fasciola gigantica*, *Paramphistomum epiclitum*, *Calicophoron calicophorum* and *Explanatum explanatum* and cestodes *Echinococcus granulosus* and *E. ortleppi* were recorded. The majority of these

*E-mail: rainaok@rediffmail.com