ANALYSIS OF ANTIGEN SHARING AMONGST GASTRO-INTESTINAL NEMATODES OF SHEEP AND GOATS USING COUNTER IMMUNO ELECTROPHORESIS

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ABSTRACT
Antigen sharing amongst gastro-intestinal nematodes viz., Haemonchus contortus, Oesophagostomum columbianum and Bunostomum trigonocephalum was evaluated by counter immuno electrophoresis. Two antigens namely soluble extract antigen (SEA) and gut integral membrane antigen (GIMA) were probed with rabbit hyper immune sera raised against SEA of three referral nematodes to discern out the image of identity. The immunoreactivity pattern of referral nematode antigens showed generally 2-3 precipitation lines with homologous and 1 precipitation line with heterologous sera. Evidently these results suggested the existence of cross-reacting antigenic determinants amongst the SEA and GIMA of referral nematodes.

KEYWORDS: Antigen sharing, nematodes, counter immuno electrophoresis

INTRODUCTION
Infections with metazoan parasites have been a continuous constraint to profitable livestock production since ages. Of these, parasitic nematodes belonging to the order Strongylidae represent diverse groups of parasites with variable economic significance in small ruminants. Parasitic gastro-enteritis caused by H. contortus, B. trigonocephalum and O. columbianum constitute important group of pathogenic nematode species of sheep and goats (Jasmer and McGuire, 1996 and Knox, 1999). They adversely affect both wool and milk production and growth in domestic animals. In heavy infections, mortality may arise as an important cause of economic loss, while moderate infections frequently cause stunted growth leading to premature culling of infected animals. Despite the increasing evidence of cross-reactivity among the number of helminth parasites, information on G.I. nematodes is, however, scanty (Cuquerella et al., 1994 and Molina et al., 1999). Therefore, the aim of the present study was to elucidate the extent of antigens shared amongst the three G.I. nematodes namely H. contortus, B. trigonocephalum and O. columbianum by counter immuno electrophoresis.

MATERIALS AND METHODS
B. trigonocephalum and O. columbianum, were collected from a local abattoir of sheep and goats. The parasites were recovered from their respective sites of predilection at necropsy following standard technique (Sahu and Misra, 1988). The collected worms were washed repeatedly with distilled water followed by physiological saline and phosphate buffered saline (pH 7.4). The worms were identified upto species level using standard keys (Sousby, 1982).

Soluble extract antigen for each species of the referral nematodes parasite was obtained by processing adult parasites of H. contortus, O. columbianum and B. trigonocephalum separately using standard technique (Keslue et al., 1986) as described by Arunkumar and Sharma (2010).

The gut integral membrane antigen for each referral nematode was obtained from dissected out worm intestines following the procedures described by Smith (1993).

About 10 worms of mixed sex were placed on a microscopic slide in a few drops of cold homogenizing buffer (0.1 M PBS, pH 7.4. 1mM Na-EDTA and 1mM PMSF) and were transected 2 or 3 times with a scalpel blade. By applying a gentle finger pressure after placing a second slide on top of the microscopic slide, the organs were allowed to exude out of the dissected worms. Under stereoscopic binocular microscope, the pieces of intestines were picked out of the debris manually into the homogenizing buffer and stored at -20°C. The worm intestines previously stored -20°C were thawed at room temperature and centrifuged at 10,000 g for 10 minutes in microcentrifuge and the resulting pellet was weighed. After adding sufficient homogenizing buffer to create a 10% (w/v) suspension, this preparation was subjected to homogenization.