1.0 INTRODUCTION

Echinococcosis, a cyclozoontic helminthosis caused by the dwarf dog tapeworm *Echinococcus granulosus* is highly endemic and is considered to be one of the most important parasitic diseases. The socio-economic impact is considerably high since both man and livestock are involved as intermediate hosts. The economic losses in animal production and health hazard along with treatment costs of cystic echinococcosis in man are enormous. The losses accrued due to this parasite are mainly because of condemnation of infected organs, lowered meat, milk and wool production. Cystic echinococcosis is widely reported from all parts of the world but occurs most frequently in areas where transmission between sheep and dog takes place. Intermediate hosts for most *Echinococcus* species are large herbivores whereas definitive hosts for *E. granulosus* are canids.

This parasite is distributed worldwide but is more common in areas where hygienic conditions are poor and literacy is low. The role of uncontrolled stray dog population especially in underdeveloped countries need not be overstressed. There is enough evidence to show that the disease is spreading because of a lack of appropriate legislation on animal slaughter, dog management and sanitary facilities (Schwabe, 1986).

In India, a high prevalence of cystic echinococcosis has been reported in man as well as in livestock. The socio-economic, cultural and religious factors have frequently played an important role in the transmission of infection to
human beings. While much attention has been focused on the prevalence, diagnosis and epidemiology of cystic echinococcosis, less attention has been focused on *E.granulosus* infection in dogs.

The prevalence of echinococcosis in dogs have been reported by many workers in India from different regions Acharya, 1939; Maplestone and Bhaduri, 1940; Reddy et al., 1968; Sahai, 1969; Sahasrabudhe et al., 1969; Khuddus and Krishna Rao, 1971; Hedge et al., 1974; Sharma and Venkatataratnam, 1974; Reddy and Reddy, 1988; Singh and Dhar, 1988; Chowdhury and Tada, 2001 and Prathiush, 2007 with different prevalence rates.

*E.granulosus* is cosmopolitan in distribution. The parasite is adapted to definitive hosts of the family Canidae and a wide range of intermediate hosts including human beings, domestic and wild herbivores and omnivores (Thompson and McManus, 2001). It is wide spread in sheep-rearing areas (Torgerson and Health, 2003). The clinical, economic and zoonotic significance of this worm are almost completely confined to infection of intermediate hosts. *E. multilocularis*, which causes alveolar or multilocular echinococcosis, has principally a holarctic (Eurasia and North America) distribution (Lightowlers, 1990). While the other species such as *E. oligarthrus, E. vogeli* and a new species *E. shiquicus* recently reported by Xiao et al., (2006) have a limited geographical distribution. These species are morphologically distinct in both adult and larval stages and are restricted to the South American and Arctic regions in sylvatic areas, respectively.
The adult *E. granulosus* is only a few millimeters long, varying between 2-7 mm in length with 3-4 segments (Eckert and Deplazes, 2004). Anteriorly, the adult parasites possess a scolex, which has four muscular suckers and two rows of sickle shaped hooks, one large and one small, on the rostellum.

The gravid proglottids and/or eggs are shed in the faeces and the eggs are brown in colour and are morphologically indistinguishable from tapeworms of the genus *Taenia* (Eckert *et al.*, 2001). *Taenia hydatigena, Taenia ovis, Taenia multiceps* and *Taenia pisiformis* are the common *Taenia* species that occur in dogs. The size of the eggs ranges from 30-40 µm and they have a thick radially striated shell, a single hexacanth embryo and the oncosphere with three pairs of hooks (Thompson and McManus, 2001).

*E.granulosus* is an obligatory heterogeneous parasite with a complex life cycle. It requires two mammalian hosts to complete its life cycle. This involves the definitive hosts (domestic dogs and wild canids) and the intermediate hosts (domestic and wild ungulates and human beings) (OIE, 2004). The definitive host is infected by ingestion of offals containing fertile hydatid cysts (i.e. cysts with viable protoscolices). The protoscolices evaginate and attach to the intestinal mucosa and develop into adult stages (McManus *et al.*, 2003). The pre-patent period of *E. granulosus* in the definitive host ranges from 34-58 days (Thompson, 1995). The adult worm passes out gravid proglottids containing eggs, or free eggs are passed out with the faeces. These gravid proglottids, or eggs, are dispersed and contaminate the environment, feed,
grass or water, etc, which are sources of infection to many intermediate hosts, including humans, over a wide area (Thompson, 1995).

The infective eggs in grass, feed or in water are ingested by the intermediate hosts (Horton, 2003), and hatch into oncospheres (larvae) inside the stomach and intestines. The liberated larvae penetrate the small intestine and reach their final location passing through vascular and lymphatic systems to the liver and lungs. They rarely spread to other organs (Soulsby, 1982). Humans are normally accidental intermediate hosts because they are rarely involved in the transmission cycle. They can be considered as ecological aberrant hosts (Torgerson and Heath, 2003). Once the oncosphere has reached its final location (liver and lungs), it develops into a cyst (primary).

There are no pathogenic effects in definitive hosts even if the animals are heavily infected with *E. granulosus* (Eckert and Deplazes, 2004). Therefore, final hosts (mostly dogs), infected with *E. granulosus* show no clinical signs except itching on the back (sledge-like position) and in heavy infections they may have diarrhoea. The infected definitive host passes thousands of eggs daily through faeces. The survival of the infective eggs is influenced by environmental factors, such as humidity, temperature, vegetation cover and soil types (Soulsby, 1982). High temperatures and desiccation are the most important factors limiting the survival time of *Taenia* eggs in nature (Schantz, 1996). In contrast, the eggs are highly resistant and can remain infective for many months or up to about one year at lower ranges of temperatures about 4°C to 15°C (Eckert and Deplazes, 2004).
The development of sensitive and specific ante-mortem diagnostic methods for the detection of canine echinococcosis is important for epidemiological baseline data and for surveillance of hydatid control programmes. Screening of dogs for *E. granulosus* has traditionally been done by arecoline purgation followed by examination of the purge. Although the specificity of purgation can be 100 per cent, it is time-consuming, biohazardous, has variable sensitivity and requires trained personnel (WHO/OIE, 2004). Furthermore, the eggs of taenid cestodes are morphologically indistinguishable by light microscopy due to extreme morphologic similarity and identification by microscopic examination of the feces is risky and non-specific. Two major diagnostic methods have been extensively used in dogs, purgation with arecoline compounds and necropsy of the small intestine. Necropsy is the method of choice and is considered as the gold standard.

Detection of parasite antigens in faeces has become an important alternative laboratory based method for diagnosis of intestinal infections caused by *E. granulosus*. Coproantigen ELISA assays have been developed for diagnosis of canine echinococcosis incorporating polyclonal antibodies against somatic or excretory/secretory antigens of adult *E. granulosus* (Allan *et al.*, 1992). Coproantigen specificity was initially reported to be high (96 per cent) with good sensitivity (77–88 per cent) based on confirmation by arecoline purge (Lopera *et al.*, 2003). Recently, a meta-analysis of several published coproantigen ELISA studies indicated an overall sensitivity of 83 per cent.
versus necropsy and 76 per cent versus purgation, and a specificity range of 88–96 per cent (Allan and Craig, 2006).

An important recent advance has been the development of copro-polymerase chain reaction (copro-PCR) for the detection of canine echinococcosis by amplification of parasite derived DNA from faeces with reportedly high sensitivity and specificity (Abbasi et al., 2003).

In India, both immunological and molecular techniques have been standardized for diagnosis of cystic echinococcosis in intermediate hosts. But even though dogs are the main disseminators of infection as definitive hosts, very few studies have been undertaken to observe the prevalence of echinococcosis. The scanty reports are mainly based on post mortem observation. Therefore the present study was undertaken with the following objectives.

1. To study the protein profile of somatic and excretory/secretory antigens of *Echinococcus granulosus* by sodium do-decyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE).
2. To identify the immunodominant peptides by Enzyme immuno transfer blot (EITB).
3. Standardization of ELISA and EITB with excretory secretory antigens of *Echinococcus granulosus* worm in dogs.
4. To detect the copro-DNA of *Echinococcus granulosus* in dogs by Polymerase chain reaction (PCR).