AN ANALYSIS OF CYSTIC FLUID OF COENURUS GAIGERI FROM GOAT ORIGIN USING SODIUM DODECYL SULPHATE POLYACRYLAMIDE GEL ELECTROPHORESIS

*Arunkumar S and Prakash Krupakaran R¹
¹Department of Veterinary Parasitology, Veterinary College and Research Institute, Orathanadu – 614625, Thanjavur (District), Tamil Nadu
²Department of Veterinary Physiology and Biochemistry, Veterinary College and Research Institute, Tirunelveli-627358, Tamil Nadu Veterinary and Animal Sciences University, Chennai-600051

ABSTRACT
In the present study, Coenurus gaigeri cysts (a larval stage of Taenia gaigeri) were collected from goats slaughtered at the abattoir in Orathanadu, Thanjavur district, Tamil Nadu. The cysts collected were confirmed to be Coenurus gaigeri using their predilection sites, size and morphology. The cysts were washed thoroughly with PBS (pH 7.4) and after careful preservation in PBS, they were used for further processing (Skeurman and Hillard, 1966). The fluid of each cyst was aspirated with the help of a sterilized syringe and needle and collected directly in centrifuge tube. Then, it was centrifuged at 10,000 rpm for 15 minutes at 4°C and the supernatants were collected and stored at -20 °C till further use. The total protein content of the samples was estimated by Lowry method (1951). Each gel well was loaded with 80 µL of cystic fluid sample in 10 % SDS-PAGE under non - reducing conditions at 100 V for 8 hours. The gel was

Keywords: Coenurus gaigeri, Cystic Fluid, Protein Profile, SDS-PAGE

INTRODUCTION
Coenurus gaigeri, is a larval stage of Taenia gaigeri and affects thigh, neck muscle, diaphragm, heart, kidney, uterus, rectum and urinary bladder of domestic goats (Varma and Malviya, 1989; Patro et al., 1997). Coenurus gaigeri in goats may reach maturity in organs other than brain and spinal cord. The cysts are mostly seen in intramuscular and subcutaneous tissues of these animals and have worldwide distribution (Soulbsy, 1982; Sharma et al., 1995). The occurrence of this metacestode in goats has been reported by different workers in different parts of India (Dey et al., 1988; Sharma and Chauhan, 2006; Madhu et al., 2013). However, the characterization of cystic fluid will help to identify protein profile and further characterization by western blotting, which will be useful to know about the immunogenic proteins in C. gaigeri. Hence, the present study was aimed to identify the polypeptide profiles in cystic fluid of C. gaigeri in goats.

MATERIALS AND METHODS
The cysts were collected from goats slaughtered at local abattoir in Orathanadu and Thanjavur areas. The cysts collected were confirmed to be Coenurus gaigeri using their predilection sites, size and morphology. Mature C. gaigeri has a smooth inner surface and contains many scolecis. The cysts were washed thoroughly with PBS (pH 7.4) and after careful preservation in PBS, they were used for further processing (Skeurman and Hillard, 1966). The fluid of each cyst was aspirated with the help of a sterilized syringe and needle and collected directly in centrifuge tube. Then, it was centrifuged at 10,000 rpm for 15 minutes at 4°C and the supernatants were collected and stored at -20 °C till further use. The total protein content of the samples was estimated by Lowry method (1951). Each gel well was loaded with 80 µL of cystic fluid sample in 10 % SDS-PAGE under non - reducing conditions at 100 V for 8 hours. The gel was
silver stained by the method of Merril et al., (1981). The electro- phoretogram was studied using the protein marker (low molecular weight Genei, Bangalore) in the range of 3.5 to 205 kDa.

RESULTS AND DISCUSSION
In the present study, the total protein concentration of the cystic fluid was 1.54 mg/ mL and SDS-PAGE analysis was carried out in cystic fluid of C. gaigeri (Figure 1). On Sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE), two prominent bands at 66 kDa and 43 kDa and three minor bands at 25 kDa, 20 kDa and 14 kDa were observed in the cystic fluid of Coenurus gaigeri. Arunkumar et al., (2014) reported that three prominent bands ranging from 60 kDa to 10 kDa (60, 20 and 10 kDa) and three minor bands at 200 kDa, 47 kDa and 35 kDa in Cysticercus tenuicollis fluid. Among the prominent bands, the intensity was stronger in 60 kDa followed by 10 and 20 kDa and the lower bands were not sharper and found diffused. A band at 200 kDa was very faint among all the other bands. The intensities of low molecular weight prominent bands were weaker and the separation was not very discrete suggesting the presence of non-proteinaceous complex nature of these proteins. Protein bands with the molecular weights ranging from 13 to 120 kDa were observed in cystic fluid of C. gaigeri of goat. A sharp band at 66 kDa level was also reported in the fluid (Koradafshari et al., 2010). Similar results were observed in the present study. However, the slight variations in the relative molecular weights of the polypeptides may be due to the influx of season on the reproductive cycle of parasite and the geographical location of the parasite. Further studies are warranted to identify the immunogenic proteins in cystic fluid, which will be useful for serodiagnosis of C. gaigeri infection in goats.

Figure 1: SDS-PAGE Analysis of E/S Antigens of Coenurus gaigeri (Silver Staining)
Lane 1: E/S Antigens of C.gaigeri (10 microliters)
Lane 2: E/S Antigens of C. gaigeri (5 microliters)
Lane 3: Mid Range Protein Marker
REFERENCES