Short communication

PREPARATION OF Toxoplasma gondii ANTIGEN*

Toxoplasma gondii infects all warmblooded animals and parasitises any nucleated cell. In view of economic and zoonotic importance of toxoplasmosis in man and animals, its early detection by serology is more useful. Antigens from parasites are complex There are various methods of structures. preparation of parasitic antigens to obtain maximum quantity of antigenic material with low level of host cell contaminants. Woodward (1982) made use of formalinised intact Toxoplasma gondii tachyzoites harvested from infected mice in contrary to a frozen sonicated sample, to detect IgM antibodies against Toxoplasma gondii in human samples by ELISA. A comparison of eight established methods for purification of peritoneal exudates of mice was undertaken by Dempster (1984). Although all of them removed more than 90.0 per cent of mouse leucocytes, the haemolysin digestion procedure was adopted as the best, because of its high zoite recovery. A simple method of antigen preparation was followed by Dubey et al. (1993), in which tachyzoites were filtered through nylon wool, sonicated and centrifuged at 10,000 x g for one hour at 4°C.

The present paper describes a simple method for preparation of antigen of *T. gondii* for use in ELISA based on the lines of Dubey *et al.* (1993).

Toxoplasma gondii tachyzoites were propagated in mice weighing approximately

20 to 25 gms. The peritoneal fluid containing tachyzoites was aspirated under aseptic conditions from 10 to 15 infected mice, pooled and incubated at room temperature with antibiotics. This pooled tachyzoite material was filtered through glass wool plugged in the barrel of a 10 ml glass syringe and sonicated at 50 volts for five minutes over ice. No organisms were visible at the end of sonication. The sonicated material was then centrifuged at 4°C in a refrigerated centrifuge (Remi C 24) at 17,000 rpm for one hour. The supernatant which formed the antigen was stored at -70°C in different aliquots and the pellet at the bottom was discarded.

Three batches of antigen were prepared and the protein content in each batch, estimated by uv-160A spectrophotometer was found to range from 15.0 to 17.6 mg/ml.

Dempster (1984) who described eight methods of purifying peritoneal exudates from mice, found that filtration through glass wool along with the other methods outlined, removed more than 90.0 per cent mouse leucocytes. In the present trial, filtration of peritoneal exudates through glass wool helped to remove most of the leucocytes and cell debris from the peritoneal fluid and yielded a good end-product. Satisfactory results were obtained by employing this antigen in an ELISA for the detection of *T. gondii* antibodies in chicken sera.

Summary

A simple method of preparation of antigen of *Toxoplasma gondii* from the tachyzoites in the peritoneal fluid of infected mice is described.

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K. Devada* and R. Anandan**Department of ParasitologyMadras Veterinary College, Chennai-7

References

Dempster, R.P. (1984). *Toxoplasma gondii*: purification of zoites from peritoneal exudates by eight methods. *Expl. Parasit*. **57**: 195-207

Dubey, J.P., Ruff, M.D., Carmago, M.E., Shen, S.K., Wilkins, G.L., Kwok, O.C.H. and Thulliez, P. (1993). Serologic and parasitologic responses of domestic chickens after oral inoculation with *Toxoplasma gondii* oocysts. *Am. J. Vet. Res.* **54:** 1668-1672

Woodward, R.C. (1982). Evaluation of an ELISA specific antigen for human IgG as a screening test for detecting anti Toxoplasma antibodies. *J. Clin. Microbio.* **16**: 367-372

Assistant Professor, Dept. of Parasitology College of Veterinary and Animal Sciences, Mannuthy, Kerala. 680 651

^{*} Present address:

^{**} Professor and Head (Retd.)