

## The Significance of Subarachnoid Cerebrospinal Fluids (CSF) in the Development of Metacestode of *Coenurus Cerebralis* in Sheep with Reference to its Pathological Effect

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**Abstract:** Five one year-old Rahmani sheep showed nervous signs (circling and pushing their heads against objects). Necropsy showed congested meninges in the cerebellar of case (1). Fluid-filled cysts (1-2 cm diameter) were found in the cerebral subarachnoid space (cases 2& 3). No subarachnoid cysts were seen in 2 cases (4& 5). Microscopically, the affected brain (case 1) displayed oncosphere migrating in the cerebellar subarachnoid space. The oncosphere was partially surrounded by caseous necrosis followed by mononuclears. The underlying cerebellar folia showed necrotic Purkinje cells and disorganized neurons of the granular layer. The metacestode-wall (cases 2& 3) revealed invaginated elongated pouches (scolex-sacs) containing scolices at their bases. The inner lining of those scolex-sacs was corrugated. Each sac was connected via an opening to the subarachnoid cerebrospinal fluid (CSF). Small empty cavities (3-7 mm diameter) were detected in the right cerebral hemisphere of cases (4& 5). Such cavities were surrounded by a zone of caseous necrosis followed by mononuclears and giant cells. Others frequently contained caseated and calcified material. The surrounding nervous tissue showed encephalomalacia. It could be concluded that the CSF is required for the differentiation and growth of the metacestode and the scolices develop from the base of the invaginated outer surface of the metacestode-wall.

**Key words:** *Multiceps multiceps* • Pathology • Subarachnoid CSF • Metacestode

### INTRODUCTION

Coenurosis usually affects the brain and spinal cord of sheep, goats, cattle, horses and other ungulates besides man. *Taenia multiceps* is a taeniid cestode that its adult stage lives in the small intestine of dogs and other canids [1]. The larval stage (Metacestode or coenurus) of this cestode, reaches the central nervous system (CNS) via blood causing high losses among sheep because it usually affects the young animals [2- 6]. Coenurosis results from ingestion of contaminated pasture with eggs of *Multiceps multiceps*. The embryos penetrate the intestinal mucosa and migrate all-over the body particularly the CNS. Only those which reach the CNS develop to form metacestodes in 2-8 months and induce nervous symptoms and death. The rest, which reach other tissues, vanish. The affected lambs show pyrexia, listlessness, circling and a slight head aversion.

Encephalitis and convulsions may be followed by death within 4-5 days [7]. Similar signs were detected in the adult sheep in Ireland [8]. The prevalence was 9.8% of 7992 examined sheep in Pars Province, in Iran [3, 9] besides 2.3 to 4.5% in the Ethiopian highland sheep [10]. The bladderworm was found in the parietal and frontal lobes besides the lumbar region of the spinal cord [11]. Deep compartment was seen in the right cerebral hemisphere in ovine coenurosis [6]. Coenurosis induced hydrocephalus in calf besides perforation of frontal skull and eosinophilic aggregations around the metacestode. Moreover, spongiosis of the cerebral white matter and brain-stem were encountered [12]. Eosinophilic granulomatous reaction was found in coenurosis-infected ovine brain [13].

The present study was carried out to elucidate the lesions induced by the interaction between the coenurus larval stages and the CSF in naturally infected Rahmani sheep.

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## MATERIALS AND METHODS

Five Rahmani sheep (one year-old) showing nervous symptoms, belong to owners in Sharkia governorate, Egypt were included in this study. The signs were recorded and the brains were necropsied and fixed in 25% neutral buffered formalin at 5°C. Gross examination was done and specimens were collected from different parts of the brain. Five  $\mu\text{m}$ -thick paraffin-sections were prepared and stained with hematoxylin and eosin (HE) and microscopically examined.

## RESULTS

**Symptoms and Necropsy Findings:** The sheep developed pyrexia, listlessness and a slight head deviation. Circling and pushing heads against objects were observed. Macroscopically, the meninges were congested, particularly that covering the cerebellum in case (1). Moreover, the brain was edematous and congested. Cysts (1-2cm diameter) were seen in the cerebral subarachnoid space in cases (2& 3), meanwhile, the brain, in cases (4& 5), was congested and edematous.

**Histopathology:** The brain, in case (1), exhibited oncosphere of *Multiceps multiceps* in the cerebellar subarachnoid space, with thick eosinophilic outer basal layer, facing caseous necrosis followed by round cells (Fig. 1). The spaces, between the cerebellar folia, were plugged with fibrinous exudate and necrotic debris.

The wall of some oncospheres was partially degenerated and necrotic (Fig. 2). The molecular layer of the cerebellar folia, adjacent to the oncosphere, showed spongiosis (Fig. 3) and the Purkinje cell-layer completely disappeared. Moreover, the neurons of the cerebellar granular layer were widely separated by fibrillar eosinophilic material (Fig. 4). Meanwhile, the Purkinje-cells of the cerebellar folia, away from the oncosphere, displayed coagulative necrosis (Fig. 5), autophagia (Fig. 6) and vacuolated cytoplasm (Fig. 7). Necrosis and neurophagia were detected in the Purkinje cells, with eosinophilic infiltration, which seemed to share in neurophagia (Fig. 8). The choroid-plexus, of the 4th ventricle, was congested and highly infiltrated with eosinophils and round cells, associated with flattened covering epithelium (Fig. 9). The subarachnoid space, covering pons, showed partially occluded blood vessels with bluish-pink material besides leukocytic perivascular cuffing (Fig. 10). The ependymal lining of the 4th. ventricle, above the malacic pons, was flattened (Fig. 11).

The brain of cases (2& 3) revealed the importance of CSF for differentiation and development of metacestodes inside the cerebellar subarachnoid space. Each metacestode was represented by delicate wall with outer multicellular surface, towards the CSF, showing more nuclei and abundant cytoplasm than the inner surface, towards the larval cavity (Fig. 12). Such more nuclei and cytoplasm of the outer surface could potentiate the absorptive and excretory activities of the surface. The metacestode-wall showed numerous invaginated scolex-sacs with basally located scolices (Figs. 13& 14).

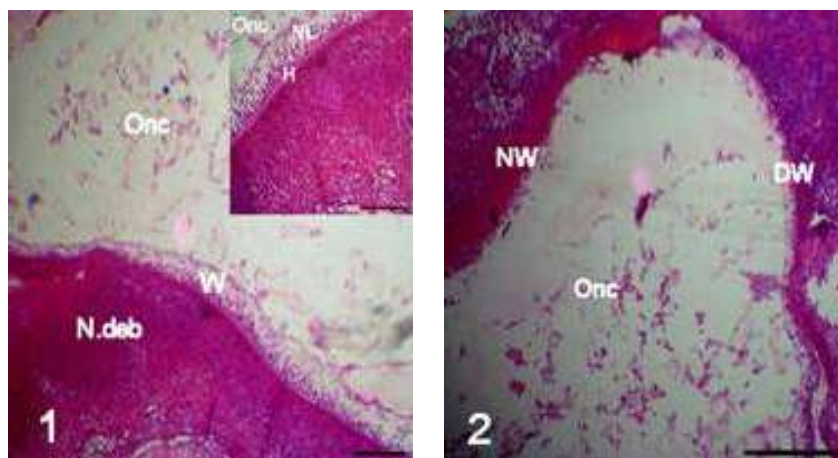


Fig. 1-2: Wall (W) of oncosphere (Onc), covering the cerebellar folia, surrounded by eosinophilic necrotic debris (N.deb) followed by mononuclears.. Insert detail of normal lining (NL) and hyalinized eosinophilic basal layer (H) ), HE. Bar, 50  $\mu\text{m}$  (1), Oncosphere (Onc) in the subarachnoid space displaying both degenerated (DW) and necrotic wall (NW) (2), HE. Bar, 50  $\mu\text{m}$

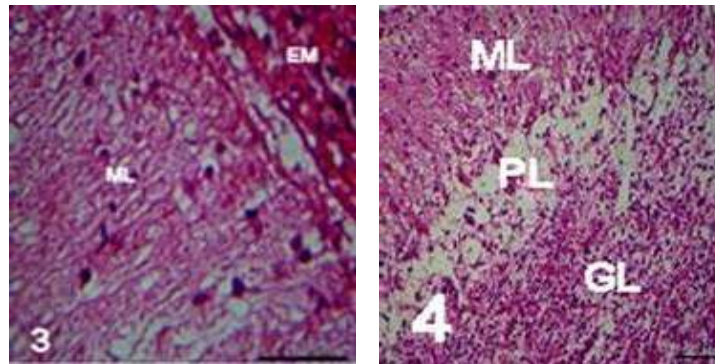


Fig. 3-4: The molecular layer (ML) of the folia, showing spongiosis with eosinophilic meningitis (EM) (3) HE. Bar 50  $\mu$ m, Complete disappearance of the Purkinje-cell-layer (PL), besides vacuolated molecular layer (ML) and indistinct granular layer (GL) (4). HE. Bar, 100  $\mu$ m.

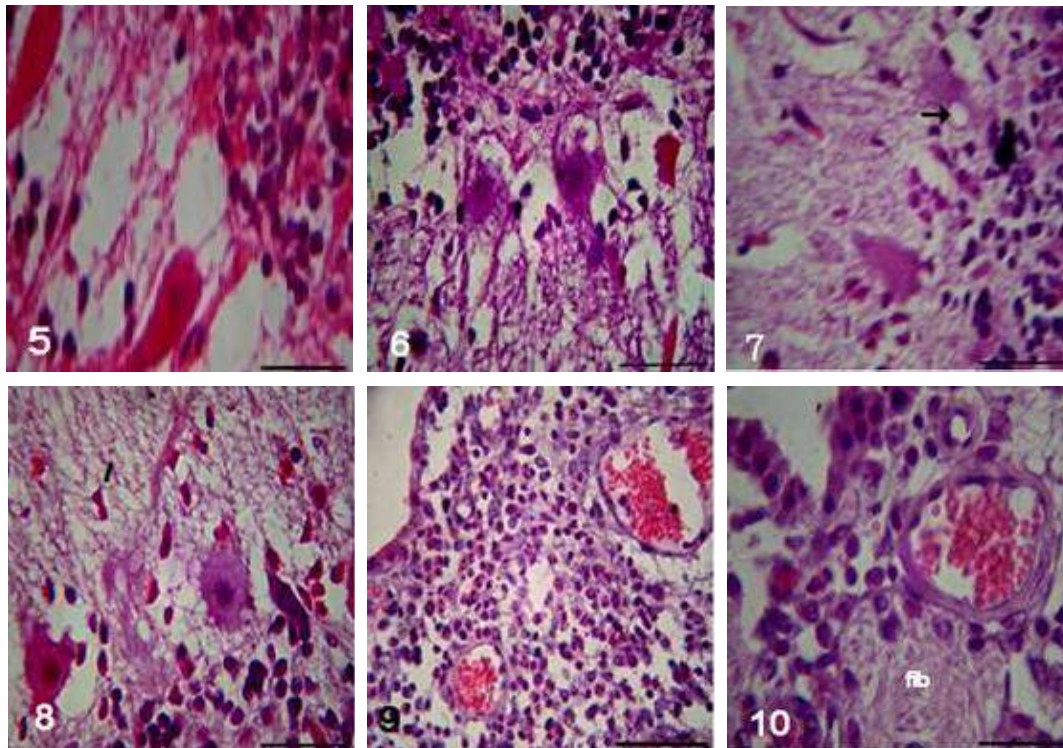


Fig. 5-8: The Purkinje cells, away from the oncosphere showed coagulative necrosis, with eosinophilic material among the neurons of the granular layer (5), Autophagia, (6), Vacuolation (7) and necrosis with neurophagia and eosinophilic infiltration (8), Choroid plexus of the 4th ventricle (4thV) showing congestion, extensive infiltration with eosinophils and round cells, besides developing flattened epithelium (9), occlusion of a blood-vessel with blue-pink fibrillar material (Fib) (10).HE. All Bars,

Most of those scolices displayed rostellum and suckers. The inner lining of the scolex-sac was an extension of the outer surface of the metacystode-wall. It was extensively corrugated (Fig. 15). The early invaginated scolices seem to be directed for differentiation by bluish coffee-bean-like cells (Fig. 16).

Different lesions were observed in cases (4& 5). Cerebral cavitations (3-7 mm diameter) were surrounded by a zone of caseous necrosis followed by round and giant cells, Langhans and foreign body (Fig. 17). They induced pressure atrophy of the surrounding nervous tissue. Some of the cerebral cavitations

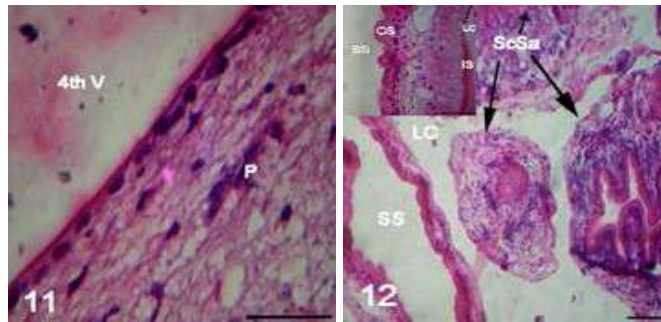


Fig. 11-12: Thin homogenous eosinophilic layer, covering flattened ependymal-lining of 4th ventricle (4thV) on malacic pons (P) (11). Scolex-sacs (ScSa), inside larval cavity (LC). HE. Bar, 100  $\mu$ m. Insert detail of the larval outer surface (OS), towards the subarachnoid space (SS), showing more nuclei and cytoplasm than the inner surface (IS), towards the larval cavity (LC) (12). HE. All Bars, 50  $\mu$ m.

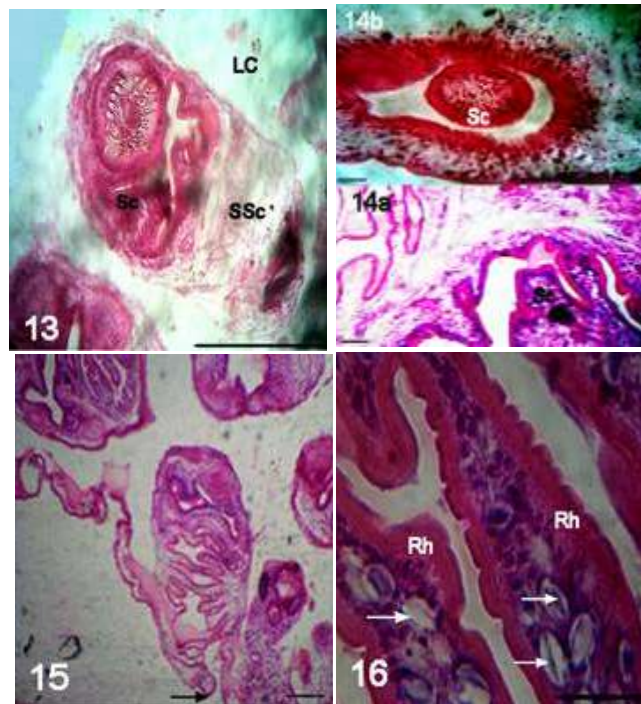


Fig. 13-16: Metacystode showing, (13) a scolex-sac (ScSa), with basally located scolex (Sc). HE Bar, 50  $\mu$ m., (14 a,b) Longitudinal (a) and cross (b) sections of scolex (Sc), at the base of parasitic sac. HE. All Bars, 100  $\mu$ m., (15) Corrugated inner surface of the scolex-sac, with a pore (arrow). HE. Bar, 100  $\mu$ m., (16) Bluish coffee-bean-like cells (arrows), apparently controlling the differentiation of the rostellum-hooks (Rh). HE. Bar, 50  $\mu$ m

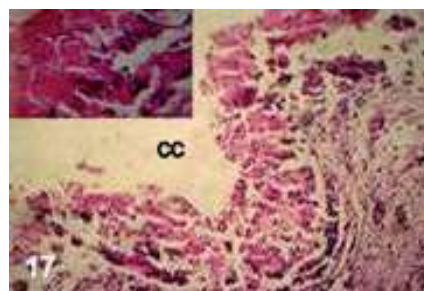


Fig. 17: Cerebral cavitation (CC) surrounded by a zone of caseous necrosis, followed by mononuclears and eosinophils and enclosed in a malacic brain tissue. HE. Bar, 100  $\mu$ m. Insert detail of mononuclears HE. Bar, 50  $\mu$ m

contained caseated and calcified material. The majority of the cerebral parenchyma showed encephalomalacia and demyelinated nerve tracts. Other fields showed focally replaced nervous tissue by bluish-pink homogenous oval to spherical masses which were stained violet with crystal violet, suggesting amyloid and caused pressure atrophy of the surrounding nervous tissue. Some cerebral blood vessels were partially or completely obliterated with perivascular encephalomalacia.

### **DISCUSSION**

This work seems to be the first to report the importance of the CSF for differentiation and development of the metacestodes. Moreover, it suggests the introduction of scolex-sac (developed by invagination of the metacestode-wall) for a developmental stage with the scolex located at its base. It developed by invagination of the metacestode-wall. The young scolex-sacs displayed deep-bluish bean-seed-like cells which seemed to play a role in the scolex-sac differentiation and growth, as it disappeared from the well-developed scolex-sacs. Meanwhile, Nourani and Pirali Kheirabadi [6] described large number of scolices which appeared as white clusters attached to the internal layer of the wall of the superficial cyst without mentioning invaginated pouches (scolex-sacs) for the scolices.

It is evident that the early developmental stage (oncosphere) may be destroyed during its migration in the subarachnoid space, inspite of the presence of CSF. Meanwhile, the metacestodes, in contact with the CSF in the cerebellar subarachnoid space, developed and produced numerous scolices inside scolex-sacs. The cavities of such scolex-sacs were in continuation with the cerebellar subarachnoid space to allow circulation of the CSF through the scolex-sacs for nourishment. Those parasites which entered the brain-parenchyma, where the CSF is scanty in the Virchow-Robin spaces, couldn't survive, as they were replaced by cavities surrounded with caseous necrosis, eosinophils, round cells and giant cells. Edwards and Herbert [14] found that the young metacestode, in the brain, could be destroyed by the immune response with complete recovery of the animal. The brain tissue, around such cavities, was malacic. Similar findings were described by Nourani and Pirali Kheirabadi [6].

According to our opinion, the caseous necrosis represented necrotic macrophages and eosinophils. Moreover, the presence of caseated and calcified

foci, surrounded with palisading macrophages, suggests that such cavities were previously occupied by metazoan parasites. Yoshino and Momotani [12] described eosinophilic infiltration with palisading histiocytes, caseation and calcification around metacestode-cyst in the left lateral ventricle of a calf.

The oncosphere, which reach the different tissues, vanish, probably because of body defense mechanisms and the absence of the CSF. It has been reported that only those parasites which reach the CNS develop to form metacestodes (coenurus) within 2-8 months [7]. Moreover, Nourani and Pirali Kheirabadi [6] did not find scolices in deep compartments, which they reported in the cerebral parenchyma of coenurosis affected sheep. Yoshino and Momotani [12] found one large cyst of *Coenurus cerebralis* (5x6 cm), containing fluid in dilated lateral ventricle of a calf showing internal hydrocephalus, where the metacestode found CSF to thrive. The current work, revealed that the scolices were derived from the invaginated outer surface of the metacestode-wall and not from its inner germinal layer, described by Yoshino and Momotani [12]. Moreover, the scolices developed from the basal pole of the scolex-sac. The inner lining of the scolex-sac (derived from the outer surface of the metacestode-wall) was extensively corrugated to increase its absorptive surface. It seems to form the outer absorptive surface of the adult cestode. The bluish bean seed-like-cells, associated with the early scolex-development, may direct the differentiation and growth of the scolex and its sac, as they disappeared from the well-differentiated scolices and its sacs.

The encountered eosinophilic fibrillar material, which widely separated the neurons of the cerebellar-granular layer, could be disintegrated axons of necrotic Purkinje cells. The necrosis and disappearance of the Purkinje-cell-layer with persistence of the cerebellar granular-layer-neurons could suggest that the Purkinje-cells are more vulnerable to destruction than the neurons of the granular layer. The encephalomalacia, all over brain, could be due to the partial or complete obstruction of blood vessels by the destruction-products of the parasite and inflammation, besides parasitic toxin. The intracytoplasmic vacuoles, in the Purkinje cells, besides the spongiosis and the deposition of the amyloid plaques resembled those of spongiform encephalopathy [15]. Ozmen [11] displayed atrophy in the central nervous system (CNS) organs due to pressure by the bladderworms, besides nonpurulent meningoencephalitis with perivascular cuffings. It could

be concluded that metacestodes require the CSF for nourishment, differentiation and development. Moreover, the scolices develop from the base of the invaginated outer surface of the metacestode-wall and the Purkinje cells are more vulnerable to destruction than the neurons of the granular layer.

#### REFERENCES

1. Scala, A. and A. Varcasia, 2006. Updates on morphology, epidemiology and molecular characterization of coenurosis in sheep. *Parasitologia*, 48: 61-63.
2. Razig, S.A. and M. Magzoub, 1973. Goat infected with *Coenurus cerebralis*: clinical manifestations. *Tropical Animal Health and Production*, 5: 278-80.
3. Gharagozlou, M.J., I. Mobedi, P. Akhavan and J.A. Helan, 2003. A pathological and parasitological study of *Coenurus Gaigeri* infestation of goats from Iran. *Indian J. Veterinary Pathology*, 27: 95-97.
4. Ghazaei, C., 2005. Evaluation of the effect of antihelminthic agents albendazole, fenbendazole and praziquantel in treatment of coenurosis disease in sheep. *J. Animal and Veterinary Advances*, 4: 852-854.
5. Welchman, D.D.E.B. and G. Bekh-Ochir, 2006. Spinal coenurosis causing posterior paralysis in a goat in Mongolia. *Veterinary Record*, 158: 238-239.
6. Nourani, H. and K. Pirali Kheirabadi, 2009. Cerebral coenurosis in a goat: pathological findings and literature review. *Comparative Clinical Pathology*, 18: 85-87.
7. Bussell, K.M., A.E. Kinder and P.R. Scot, 1997. Posterior paralysis in a lamb caused by *Coenurus cerebralis* cyst in the lumber spinal cord. *Veterinary Record*, 140: 560.
8. Doherty, M.L., H.F. Bassett, R. Breathnach, M.L. Monaghan and I. BA. McErlean, 1989. Outbreak of acute coenurosis in adult sheep in Ireland. *Veterinary Record*, 125: 185.
9. Oryan, A., N. Moghaddar and S.N. Gour, 1994. Metacestodes of sheep with special reference to their epidemiological status, pathogenesis and economic implication in Pars Province Iran. *Veterinary Parasitology*, 51: 231-40.
10. Achenef, M., T. Markos, G. Feseha, A. Hibret and S. Tembely, 1999. *Coenurus cerebralis* infection in Ethiopian highland sheep: incidence and observations on pathogenesis and clinical signs. *Tropical Animal Health and Production*, 31: 15-24.
11. Ozmen, O., S. Sahinduran, M. Haligur and K. Sezer, 2005. Clinicopathologic observations on *Coenurus cerebralis* in naturally infected sheep. *Schweiz Arch Tierheilkd*, 147: 129-34.
12. Yoshino, T. and E. Momotani, 1988. A case of bovine coenurosis (*Coenurus cerebralis*) in Japan. *Japanese J. Veterinary Sci.*, 50: 433-438.
13. Oruç, E. and U. Uslu, 2006. Comparative cytopathological and histopathological studies of sheep with suspected *Coenurus cerebralis* infection. *Türkiye Parazitoloji Dergisi*, 30: 285-288.
14. Edwards, G.T. and V. Herbert, 1982. Observations on the course of *Taenia multiceps* infections in sheep: clinical signs and postmortem findings. *British Veterinary J.*, 138: 489-500.
15. Davis, A.J., A.L. Jenny and L.D. Miller, 1991. Review article: diagnostic characteristics bovine spongiform encephalopathy. *The J. Veterinary Diagnostic Investigation*, 3: 266-271.