

Eosinophilic Meningoencephalitis in Thailand

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**OBJECTIVE:** Continuation of epidemiologic, clinical, neuropathologic and experimental studies on eosinophilic meningoencephalitis.

**DESCRIPTION:** Epidemiologic study was terminated in May 69 and the data obtained from the previous years was analyzed. The clinical and pathologic studies were carried out only in Bangkok. Four experimental studies were performed to clarify the pathogenesis of eosinophilic myeloencephalitis caused by Gnathostoma spinigerum, based on some findings in the study of human cases.

**PROGRESS:** A. Clinical and Pathologic Study.

During the period 21 cases of eosinophilic meningitis were studied; 19 cases belonged to the typical form and two cases were myeloencephalitis. Of the 19 typical cases, 11 patients were male and 8 were female. In one case, a fifth stage A. cantonensis larva was seen in the posterior chamber of the eye and was subsequently surgically removed from the anterior chamber. This is the first case demonstrating the association between ocular angiostrongyliasis and eosinophilic meningitis. Both cases of eosinophilic myeloencephalitis were female and were admitted with cerebral hemorrhage. Eye swelling was noted in a case. One case died and the post-mortem findings were characteristic of Gnathostoma invasion of the central nervous system but no worm was recovered.

B. Laboratory Studies.

**Experiment 1:** To confirm the study that the early third stage larva of G. spinigerum in copepods are infective to mice, chicken and cats.

**Background:** Epidemiologic evidence on eosinophilic myeloencephalitis due to Gnathostoma indicated that patients may be infected by drinking contaminated water. Prof. Daengsvang has shown that mice may be infected by infected copepods. This study is designed to learn more about this and to study other animals including the definitive host.

**Materials and Methods:** Laboratory bred copepods were infected with first stage G. spinigerum larvae and were kept for 5, 10 and 16 days before being fed to 30 mice, 20 chickens, and 1 cat in each group. Each animal received 10 larvae from the copepods and were sacrificed at 6 hrs., 24 hrs., 5, 10, 15 and 30 days post-infection and examined for the third stage larva.

**RESULTS:** 1. In mice five-day-old infected copepods were non-infective but in 10-day and 15-day-old infected copepods the infection with third stage larvae was found in 40% and 80% respectively.

2. None of the 20 chickens infected with 15-day-old infected copepods yielded positive results.

3. Two cats infected with 20 and 50 larvae from 15-day-old copepods sacrificed on the 15th day post-infection were negative for gnathostomes. Another three cats infected with 20, 50 and 100 larvae each were still negative for Gnathostoma ova in stools 9-12 months post-infection.

4. Infected copepods can be kept alive in the laboratory for as long as 64 days. Gnathostome larvae had 2 molts in the copepods. The heaviest infection in one copepod was 22 larvae. In most instances a copepod was infected with 1-3 larvae.

Experiment 2: To study the morphological changes and development of the third stage infective G. spinigerum larva following repeated infections in transport hosts.

Background: It is suspected that the third stage G. spinigerum may develop to a more advanced stage after many passages in various transport hosts and these advanced stage larvae may cause the disease in man.

Materials and Methods: Third stage G. spinigerum larvae obtained from experimentally infected mice were divided into two groups, those from the liver and those from muscles; ten of each were separately infected into mice which were sacrificed on the 5th and 15th day post-infection. Sites of organs of recovery of larvae were recorded. The recovered larvae were used for reinfection until no larva was found. Tissues where larvae were found were sectioned and studied for pathological changes.

RESULTS: 1. After infection, third stage larvae may or may not enter the liver, no matter whether they have previously migrated to the liver or not. This finding is contrary to the other reports.

2. There were no morphological changes in these larvae even after as many as 15 reinfections. However, the non-encysted form of larvae can occasionally be seen in the musculature.

3. Frequency of organs involved where 400 larvae were recovered are as follows: muscle of back, 35%; muscle of hind legs, 22%; muscle of chest wall, 12%; muscle of abdominal wall, 10%; mesenteric fat, 8%; muscle of fore leg, 6%; muscle of neck, 2%; lungs, 2%; heart, 1%; diaphragm, 0.7%; kidney, 0.7%; stomach wall, 0.4%; and spleen, 0.2%. This may explain why the portal of entry of Gnathostome larvae into the central nervous system in man occurs mainly in the lower part of the spinal cord.

4. Sections from various tissues were made and will be studied.

Experiment 3: To prove experimentally that the chicken may serve as a transport host of G. spinigerum and to observe any morphological changes of the larvae.

Background: Chickens are noted to be a source of human infection and may be important in eosinophilic myeloencephalitis.

Materials and Methods: Third stage larvae were fed to chickens which were sacrificed on the 15th and 30th day post-infection.

RESULTS: Infection was complete in 25 per cent. Both encysted and non-encysted third stage larvae were found but without any morphological changes.

Experiment 4: To study the pathogenicity of the 4th and 5th stage Gnathostome larvae in mammals.

Background: All G. spinigerum recovered from man were fifth stage larvae or immature adult worms. We have shown in other studies that the spinal cord of the mouse is invaded by the 5th stage larva of G. spinigerum, recovered from man, following ingestion. This experiment was designed to confirm the finding.

Materials and Methods: 15 cats were infected with varying numbers of third stage larvae; 7 cats were sacrificed on the 5th, 9th, 12th, 20th, 34th, 60th, and 120th days after infection. The recovered larvae, all third stage, were injected into mice. The mice were subsequently sacrificed and third stage larvae were found in the muscle. In two instances, the larvae had 4 rows of hooklets and were longer than the ordinary third stage. None involved the CNS. This experiment is in progress.

#### PUBLICATIONS.

Epidemiologic Studies of 484 Typical Cases and the Etiologic Role of Angiostrongylus cantonensis. (Accepted for publication).