

ANNUAL PROGRESS REPORT

SEATO Medic Study No. 91: The Pathogenesis of Opisthorchis viverrini Infections

 I. Morbid Anatomic Changes in Naturally Infected Cats and Dogs in Udorn.

 II. The Pathogenesis of Laboratory Infected Cats

Project No. 3A 25601 A 811: Military Medical Research Program
 S. E. Asia

Task 01: Military Medical Research Program
 S. E. Asia

Subtask 01: Military Medical Research Program
 SEASIA (Thailand)

Reporting Installation: U.S. Army-SEATO Medical Research
 Laboratory, APO 146, San Francisco,
 California

 Division of Medical Research Laboratories
 Department of Geographic Pathology

Period Covered by Report: 1 April 1963 to 31 March 1964

Principal Investigator: Sylvanus W. Nye, Captain, USAF, MC

Associate Investigator: Dale E. Wykoff, Major, MSC

Reports Control Symbol: MEDDH-288

Security Classification: UNCLASSIFIED

ABSTRACT

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The objective of this study is to describe the histopathologic changes in animals, naturally infected and laboratory infected by *Opisthorchis viverrini*, in order to determine the pathogenesis of the disease in these animals. In the first

part of the study, cats were autopsied in Udorn and studied histologically. For the second part of the study, cats and rabbits infected with a standard dose of metacercariae are to be studied at serial intervals following infection. In the first part of the study it was found that the surfaces of the livers of larger and older heavily infected cats were uneven towards the edges and that flukes could be easily found on macroscopic examination on the cut surfaces. Where not distended by a fluke, large bile ducts were lined by folded mucosa with many outpouchings and crypts embedded in dense periductal tissue. Focal collections of lymphocytes were present around large ducts and there was heavy infiltration of eosinophils in some cases. In heavily infected cats small portal areas were also infiltrated by eosinophils. In a few animals there was an exudate of neutrophils and eosinophils in fluke containing ducts. Considerable scarring resembling biliary fibrosis was present in the peripheral parts of the liver lobes in some animals. No bile stasis was noted. In the study of the pathogenesis in laboratory infected animals 10 rabbits and 8 cats have been sacrificed to date. None of these animals have been studied histologically. Studies so far indicate that the lesions caused by the liver fluke of Opisthorchis viverrini closely resemble those caused by Clonorchis Sinensis.

BODY OF REPORT

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Objective: The objective of the first part of the study is to describe the morbid anatomic changes found in natural infection with Opisthorchis viverrini in cats. In the second part of the study, the objective is to study the host reaction

to known numbers of metacercariae with known durations of infection in order to describe the pathogenesis of the infection.

Description: For the first part of the study, 25 wild domestic cats were obtained from rural villages around Udorn in Northeastern Thailand. All stools were collected on each cat for three days and the number of eggs per gram of feces were determined. Five of the cats were sacrificed and the number of worms were counted in the livers of these cats. From this data the number of eggs produced per worm per day will be estimated, and from the data on the number of eggs per gram/feces, the number of worms in the livers of the 11 autopsied cats will be estimated. In addition to studying the 11 cats which were completely autopsied and in which estimates of a number of worms were made, 41 additional cat autopsies were performed by a technician in Udorn. These animals have not been completely studied. For the second part of the study, rabbits and cats have been infected with doses of approximately 200 metacercariae. These animals are being killed at intervals from 1 day to 1 year.

Progress: The tissue from the 11 completely autopsied cats has been studied histologically. Flukes were identified grossly in all but three of the 11 livers. In most of the heavily infected livers, flukes were easily identified, within scarred bile ducts on gross inspection. The livers of heavier and presumably older cats with large numbers of liver flukes showed mild irregularity and scarring of the surfaces of the livers near the edges. Flukes are found in moderately ectatic bile ducts in scarred portal areas. The mucosa of the duct near the flukes is folded and there are many outpouchings or crypts in the walls. These crypts are embedded in dense collagenous scar tissue which surrounds the ducts. Immediately beneath the epithelium of the duct, there are small capillaries many of which contain large numbers of eosinophils. The walls of these large scarred ducts in most cases do not contain heavy infiltrates of lymphocytes or other inflammatory cells, however, occasionally there is a large nodular collection of lymphocytes. In some cases there is a marked inflammatory reaction at the junction of the thickened and scarred portal area with the liver cells cords. In portal areas containing ducts which appear too small to contain flukes, there is also an increased amount of connective tissue around the ducts. In most instances there is a more marked inflammatory reaction in the walls of these ducts than there is in the large fluke containing ducts. The inflammatory infiltrate in these cases is composed of lymphocytes, small numbers of plasma cells and eosinophils. In a few cases there are heavy infiltrates of polymorphonuclear leukocytes and eosinophils in these smaller portal areas. In a few livers there are exudates of polymorphonuclear leukocytes and eosinophils into the ducts. The stimulus for this exudation is not apparent, however, quite frequently there is a pool of fibrinous looking material which appears to have been excreted by the fluke associated with these exudates. Three livers showed extensive fibrosis in the periphery of the lobules with a great increase of

fibrinous tissue connecting adjacent portal areas: However, in none of these were bile plugs present and there did not appear to be any retention of bile in liver cells. Necrosis of the liver cells is not present. Progress in the second part of the study with laboratory infected cats and rabbits is satisfactory. Some cats and rabbits have been sacrificed but no histological preparations have been made.

Summary and Conclusions: Infection with the liver flukes, Opisthorchis viverrini causes marked inflammatory reaction in the portal tissues of the liver which is associated with marked scarring and fibrosis of the portal areas. The ducts which contain flukes become ectatic and develop many outpouchings of the walls. It appears that the degree of scarring and inflammation in portal areas is related to the duration of infection and the severity of the infection.