THE OCCURRENCE OF SECONDARY PARASITISM IN THE FROG.

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In a series of experiments on the life histories of certain Anuran parasites, especially those of *Rana pipiens*, conducted under the direction of Dr. H. E. Enders, several cases of secondary parasitism have been observed, two of which are described in this article.

By secondary parasitism is meant the occurrence of parasites in forms which themselves lead an essentially parasitic existence. To the uninitiated the mention of a parasite often brings to mind a few common forms such as the louse, the flea, the tick, or perhaps a tapeworm. Unless one is actively engaged in the study of parasitology, it is difficult to appreciate the tremendous number of parasitic forms that may be found at one's very elbow. Indeed, it may be conservatively stated that every animal, during its lifetime, is a host to one or more species of parasites which are dependent upon it for their existence.

This statement is prompted by a small, moribund, leopard frog, which the writer recently dissected and which contained the following astonishing list of parasites:

FLATWORMS.

Pneumonoeces coloradensis, in lungs,	5
Loxogenes arcanum, encysted in musculature of stomach and	in-
testine,	14
Clinostomes (sp.?), immature forms encysted generally,	137
Diplodiscus temperatus, in rectum,	5
Gorgoderina attenuata, in bladder,	3
ROUNDWORMS.	
Angiostoma nigrovenosum, in lungs,	7
Strongylus auricularis (?), in intestine,	4
Small, immature, active, free-living nematodes, (species?), in	n body
cavity,	17
PROTOZOA	

Opalina, and Nyctotherus, several hundred in intestine and rectum.

The infestation of parasitic forms by other parasites is a common phenomenon, and especially is this true in the case of Protozoa. Thus, the orders Flagellata, Gregarinida and Coccidiidea are replete with parasitic species that may be dependent upon other parasites, commonly insects, for their transfer to a final host. The Trypanosome of "sleepingsickness" and *Plasmodium vivax*, the causative agent of malaria, are two of a number of well known examples which may be cited. It is true that secondary parasitism among the higher metazoa is more difficult to find, but this is probably due to lack of research, rather than to a lack of material. It is now definitely known that fleas are intermediate hosts for larval stages of certain tapeworms. We have also good reason to believe that other helminth forms can be transferred by insects in this way. Doubtless this list could be further augmented, but

"Proc. 38th Meeting, 1922 (1923)."

the examples already cited will suffice to show some, of the parasite relationships that may be found. They indicate moreover, the economic value of studies in this field.

To the above list the writer would contribute a new form of parasite relationship, namely, the occurrence of Distome larvae in other immature parasites. The only analogous case of which we have any



Fig. 1. a, Anterior and posterior ends of larval cestode, showing Distome larva parasite in anterior portion, x 12; b, immature *Clinostomum marginatum* parasitized by three larval Distomes, x 12; e, *Agamodistomum marcianae* (?), the larval Distome infesting the above parasites, x 25. The small figures indicate one-half natural size.

record is one reported by Cort¹ in which a Trematode was parasitized by Gordius larvae. In the accompanying figure are shown two parasitic forms in which secondary infestation was found. The first (fig. 1a) is an immature Cestode, one of seven specimens taken from the same liver. These were free-living rather than encysted forms and were woven in and out throughout the parenchyma of the liver. They were

¹ Cort, W. W. Gordius Larvae Parasitic In a Trematode. Jour. Parasit., 1:198-199. 1915. teased out with the greatest difficulty, and when isolated, were glistening white in color, ranged in length from one to three inches, and posressed a well defined scolex with four suckers but no hooks. These Cestodes were also peculiar in having a cylindrical body. The available records of Anuran Cestodes list but three species from this country. These are, *Nematotaenia dispar* Goeze 1782, *Taenia pulchella* Leidy 1851, and *Cylindrotaenia americana* Jewell 1916. Jewell² considers the last two identical, and also questions the occurrence of *Nematotaenia dispar* in North America. If these contentions are valid then the Cestodes under discussion are either a new species or identical with *Cylindrotaenia americana*. They unquestionably belong to the Nematotaeniidae, even though from their immature condition the species could not be determined with certainty.

The second (fig. 1b) shows the larva of *Clinostomum marginatum*³ taken from a subcutaneous cyst. These forms always occur encysted singly, and are very abundant, it being nothing unusual to find several dozen in a single frog. From personal observation it would appear that two species are concerned. The first is a relatively large form with a smooth cuticle, while the other is a smaller, more slender form, with a spiny cuticle. It may be that these are different stages in the development of the same species. A fair degree of dexterity is needed to separate these forms from their tough membranous sheaths without injury. However, when once liberated they are active and extensile to a degree. In conducting the experiments already outlined, three of these Clinostomes and one Cestode were found that showed infestation, presumably with the same species of parasite. The illustrations for the figure were prepared from specimens mounted in toto, and stained with equal parts of acid carmine and alum cochineal. Attempts to measure living material were unsuccessful as these animals changed their shape too rapidly to yield accurate results.

The parasite found infesting the above forms was a diminutive larval Distome shown in figue 1c. In this case the specimen was teased out of one of the Clinostomes already mentioned and this illustration, like the others, was drawn to scale from a prepared mount. Careful search elicited the presence of these parasites in practically every frog examined, while in some individuals literally hundreds were found. These forms were active, free-living, and migrated extensively throughout the tissues of the host. The easiest way to obtain them in number was to scrape the inverted skin of a frog in normal salt solution. In the fine residue thus obtained numerous Distome and other larval Trematodes could usually be found under the microscope. Living specimens showed a well defined forked digestive tract and an emulsified structure of the protoplasm. They were unusually active and extensile, changing their shape continually in the process of locomotion. They were also more slender than the drawing would indicate, as the mounted

² Jewell, Minna E. Cylindrotaenia americana nov. spec. From the Cricket Frog. Jour. Parasit., 2:181-191. 1916.

³ Osborn, H. L. On the Distribution And Mode Of Occurrence in The United States And Canada Of Clinostomum Marginatum, A Trematode Parasite In Fish, Frogs And Birds. Biol. Bull. No. 5, Vol. XX, pp. 350-367. 1910.

specimen is much shrunken and fore-shortened. These forms occurred chiefly in the connective tissues of the host, but could be found generally throughout the body, as in the lungs, the digestive tract, the body cavity, and in the liver and spleen. In these last their presence was indicated by gray, granular, necrotic patches upon the surfaces. On several occasions these Distomes were observed in the web of a frog's foot, where they seemingly had no difficulty in making their way, moving rapidly across the field of the microscope.

The extent to which these Distomes are true parasites of the forms they infest is problematical. In all cases where this infestation occurred an excessive number of free-living Distomes were also found in the tissues of the frog. In neither the Cestode nor Clinostomes were these secondary parasites encysted, a fact which would suggest a fortuitous occurrence. It seems altogether probable that in their extensive migrations these small forms could accidentally force their way into other parasites, especially if the body substance of these offered less resistance than did the tissues of the primary host. These secondary parasites varied considerably in size, but were of the same general appearance. Because of their immature condition no exact identification is attempted. In their gross anatomy they agree closely with Agamodistomum marcianae,⁴ a cercaria described by Cort as follows: "The ventral surface is completely covered with spines which are very thickly set over the anterior tip and somewhat scattered in the post-acetabular region. The margin of the acetabulum is armed with two or three rows of closely set spines pointing in, which are so placed that they add greatly to the gripping power of the sucker. The host in which Agamodistomum marcianae complete its development is not known. Also its structure at this stage gives little clue to the systematic position of the adult. The character of its cephalic glands and excretory system, however indicates that it has developed from a forked-tailed cercaria."

⁴ Cort, W. W. The Exerctory System Of Agamodistomum Marcianae (La Rue), The Agamodistome Stage Of A Forked-Tailed Cercaria, Jour. Parasit., 4 130-134. 3 figs. 1918.