OBSERVATIONS ON AN ACQUIRED IMMUNITY TO A METAZOAN PARASITE[1]

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A few years ago Reuling[2] first demonstrated that an animal may acquire immunity to a metazoan parasite. This was an important contribution to the literature of immunity and is of especial interest since this condition had been so long and variously sought without success. His results were gained upon fish acting as the host, and glochidia, the larval form of fresh-water mussels, as the parasite. It should be stated by way of explanation that the almost microscopic glochidium passes a parasitic period embedded in the superficial gill- or fin-tissues of an appropriate fish. Only under these conditions can metamorphosis to the free-living juvenile occur.

Reuling showed that at the first and second infections of fish a normal metamorphosis is completed in the average summer time of about two weeks. On the third and subsequent infections unusually heavy cysts were formed, but the glochidium sloughed off by the second or third day; this was accompanied by necrosis of the epithelium and a certain amount of glochidial disintegration. Hanging-drop preparations proved the blood of immune fish to exert a cytolytic effect on glochidia; tests indicated the probability of a weak precipitin in the antisera.

Having observed the general course of these experiments, I can testify to the accuracy of their broad conclusion. Nevertheless, incidental tests pursued during the last two summers bring out some additional information. My original objects

[1] Contribution no. 105. Published by permission of the United States Commissioner of Fisheries.

[2] Reuling, F. H. 1919. Acquired immunity to an animal parasite. Jour. Infect. Dis., vol. 24, pp. 337-346.

were two: first, to determine if more light infections are necessary to produce immunity than the heavy infections usually employed; that is, if there is an easy demonstrable quantitative factor; and secondly, to discover if an immunity once established remains effective through the following seasons. Both points have an important practical application in mussel propagation. Infections as they occur in nature are light in comparison with the heavy dosages given in propagative work. Should light infections prove as effective as heavy ones these feral fish might become immune and soon be useless as carriers. If an immunity builds cumulatively and holds from year to year it would further eliminate older fish from the host-function. These possibilities are especially significant in view of the extensive propagative program carried on by the Bureau of Fisheries. In restricted areas, rescued fish might thus be infected repeatedly without any certainty as to whether the glochidia metamorphose or slough.

In the experiments now to be described the host used was the large-mouth black bass, Micropterus salmoides;[3] the glochidia were from the Lake Pepin mucket, Lampsilis luteola. These were the forms used largely by Reuling. A good artificial infection places about 2000 glochidia upon the gills of a fish.

First, my observations give some additional information on the number of infections necessary to produce immunity. Reuling found two infections sufficient for medium-sized bass, whereas large individuals may become immune after one period of parasitism and the small ones require three. This difference he explained on the basis of varied past experiences as hosts in nature—the older fish having had more opportunity to acquire partial immunity.

A lot of nearly three dozen black bass was infected repeatedly during the summer of 1922. The first two infections were uneventful. The third probably produced more or less immunity, although there is only the positive record that the fish were free

[3] For those fish I am indebted to Dr. A. D. Howard and Mr. Barry J. Anson, who also conducted the first infection both in 1921 and 1922.

of the glochidia on the tenth day, whereas a parallel infection on wild bass in another experiment required a week longer. At the fourth infection, the fish became practically clean after four days; all but a few were entirely free at six days and all on the seventh. There was no rigid correlation between large size and early immunity.

The results of the summer of 1921 are even more interesting. The first two infections were without especial feature. Two or three days after the third treatment the number attached gradually decreased. Yet the last fish was not entirely clean until the thirteenth day. At the fourth infection there was the same gradual reduction, some fish retaining glochidia until the third, fifth, seventh, ninth, and fifteenth days. At the fifth and last dosage conditions were similar, the fish becoming clean on the eleventh, nineteenth, and twenty-fifth days. This infection was early in September, but on the eight days following the water was as warm as the average for August. Two other fish were given a fifth infection on September 29th;[4] by this time the water temperature had lowered considerably and the fish retained the parasites, the sole survivor shedding metamorphosed larvae in the middle of the following May, when the water had again become warm. It is important to note that this fish lost many glochidia within the first four days, and still later the original number was reduced to one-half, yet despite this apparent partial immunity the glochidia remaining eventually metamorphosed.

These experiments indicate that immunity is not always acquired in a sharp, clean-cut fashion after the second or third infection, but that four, five, or more infections may be necessary, with a gradual building up of a semi-immunity. This also records for the first time as many as five infections on fish not yet immune.

Furthermore in these cases the shedding of encysted individuals may be progressive. Reuling found the glochidia were

[4] This was performed by Mr. Barry J. Anson, who kindly followed and recorded the later progress of both fifth infections.

SUMMARY

Immunity to glochidial parasites (e.g., Lampsilis luteola) may be acquired by fish (e.g., black bass) in two to five or more infections.

Fish that become thoroughly immune at the second or third infection slough the attached glochidia rather promptly within forty-eight to seventy-two hours.

Fish that require four or more infections acquire an ill-defined immunity, and glochidia are lost progressively over several to many days.

Light infections are practically as effective as heavy dosages in producing immunity, although there is apparently a quantitative difference when the spread is extreme. The number of infections seems to be more important than the degree.

The permanency of acquired immunity remains to be proved, yet there are miscellaneous records which indicate that it lasts at least one year.