SUCCESSFUL CULTIVATION OF AVIAN PLASMODIA IN DUCK EMBRYOS

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So far as is known to the author, there is no record in the literature of the successful cultivation of any Plasmodium in bird embryos. Attempts with negative results have been reported on two occasions. Huff and Bloom (1935) attempted to inoculate canary embryos with P. elongatum "in order to determine whether the primitive blood cells and primitive erythroblasts would harbor these parasites." Gavriloff, Bobkoff and Laurencin (1938) attempted to inoculate chick embryos with P. gallinaeum.

In the present study three species of Plasmodium, P. cathemerium, P. elongatum and P. lophurae, were successfully cultivated in duck embryos. The inoculations of embryos were made under the allantoic membranes with the infected blood from adult ducks. The results can be summarized as follows.

P. cathemerium. The domestic duck (Anas buschas domestica) was first found to be susceptible to avian Plasmodium 2 years ago (Wolfson, 1938). Since then, the wood-thrush strain of P. cathemerium has been maintained in the duck, and its behavior in this host was reported (Wolfson, 1940).

Of eight embryos infected with P. cathemerium, which lived long enough to be examined, three gave positive results. The remaining five were examined 6 or fewer days after inoculation. In the first positive case (a 21-day-old embryo) the presence of parasites, 6 days after inoculation, was demonstrated by a subinoculation of its blood into a clean duck. The parasites appeared in the peripheral blood of the latter after a prepatent period of 5 days. In the second case (a 24-day-old embryo) the parasites were observed directly in the blood and tissues when this embryo was killed and examined 11 days after inoculation. The number of parasites in its heart blood at that time was about 540 per 10,000 erythrocytes. The third embryo, inoculated at the age of 14 days, was examined on four different occasions. A few parasites were observed in its blood on the eighth day after inoculation. It contained about 500 and 2,000 parasites per 10,000 erythrocytes on the tenth and eleventh days after inoculation, respectively. This embryo was found dead on the thirteenth day after inoculation when the number of parasites in its heart blood was about 1,500 per 10,000 erythrocytes.

P. elongatum. This parasite was inoculated into a duck from a latent canary, and was subinoculated ten times thereafter. Most of the eighteen birds studied either died or were killed for the experimental purposes during the first or second weeks after the inoculation. In all cases, the parasite number in the peripheral blood did not exceed 500 per 10,000 erythrocytes and was usually under 100. Examinations of bone marrow revealed a very large number of parasites even when their number in the peripheral blood was low. It is of interest to note that exoerythrocytic schizogony persisted in the ducks in-
infected with P. elongatum, whereas in the case of P. cathemerium it was observed only in canaries and not when the parasites were inoculated into the duck (Wolfson, 1940).

Three embryos inoculated with P. elongatum were examined 7 or more days after inoculation. One inoculated at the age of 14 days, was found dead 7 days later. When autopsied and examined less than 24 hours after its death, about 3,400 parasites per 10,000 erythrocytes were found in its heart blood. The other two embryos examined both on the eighth and the tenth days after inoculation, revealed no infection.

P. lohur. This organism was inoculated into a duck from a chicken and was subinoculated ten times thereafter. About one half of the twenty birds died, apparently from the infection, between 7 and 18 days after inoculation. The parasite number in the peripheral blood usually exceeded 5,000 per 10,000 erythrocytes and was as high as 12,000 in several cases. This large number was due to numerous multiple infections, while some red blood cells usually remained uninfected. The parasites preferred the mature erythrocytes to reticulocytes and no exoerythrocytic stages were found at autopsy.

Eight embryos inoculated with P. lophur. were examined 7 or more days later. All of them were examined 7, 8 or 9 days after inoculation and were found negative. Two of them were again examined 14 days after inoculation with negative results. The third was killed on the sixteenth day after inoculation as it was breaking out of the shell. Parasites were found in its blood and tissues. Its blood inoculated into adult ducks produced infections.

Increased synchronicity of segmentation

Some evidence was obtained from the above experiments indicating that the synchronicity in segmentation of Plasmodium increased considerably during the residence of the parasite in the embryo. It is generally believed that periodicity in the segmentation of Plasmodium depends upon the diurnal changes in the environment of the host, associated with the periodic intake of food, muscular activity, and changes in the internal and external temperature. Since all these factors can be more easily controlled for the embryo than for the adult bird, the cultivation of Plasmodium in embryos may be of interest in the study of the factors influencing the length of the asexual cycle and the degree of synchronicity in segmentation.

REFERENCES

Huff, C. G., and Bloom, W.
Gavriloff, W., Bobkoff, G., and Laurencin, S.
Wolfson, F.