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Some Responses Made by Ten Duck Hosts Inoculated with Phanerozoite Stages of *Plasmodium lophurae*

By DELMA E. HARDING

METHOD

Ten White Pekin ducks were inoculated with phanerozoite stages of the malaria parasite, *Plasmodium lophurae*, to determine what responses might be made by the host tissues. The ducks were brought to the laboratory when one day old, and placed on a regular feed starter. This was the only ration given to them during the course of the experiment.

Inoculum for the duck hosts was made from a saline emulsion of two turkey brains whose blood smears had indicated that no malarial-infected red blood cells were present in the general circulation. If such were present, they were not seen. An examination of the brain capillaries of the two turkeys showed a great number of exoerythrocytic forms (phanerozoites) of *P. lophurae* to be present.

In preparing the inoculum, the entire brain was removed from each of two turkeys, macerated, and diluted 1:20 with normal saline. Approximately 0.9 cc of the emulsion was injected into each of the five-day-old hosts, without regard to their weight. The average weight of the duck hosts on the day of their inoculation was 63.7 grams, although individual weights ranged from 57 grams to 70 grams.

Smears were made daily from blood of leg veins, from the day the ducks were inoculated until the last one was killed on the 21st day following the inoculation date, to determine whether *P. lophurae* could be demonstrated in the red blood cells of the duck hosts. These smears were fixed in methyl alcohol and stained with Giemsa stain. One duck was sacrificed on each of the following days after inoculation: 9, 12, 13, 15, 16, 17, 18, 19, 20 and 21.

Imprints of organs or bits of macerated organs from the sacrificed ducks were placed on slides, fixed in methyl alcohol and stained with Giemsa stain. Organs for sectioning were removed from the sacrificed ducks, fixed in Bouin's or formol-saline solution, and later cut to a thickness of 5 microns. These were stained with toluidine blue, phloxine and Orange G in a method according to Tomlinson and Grocott, (1944).

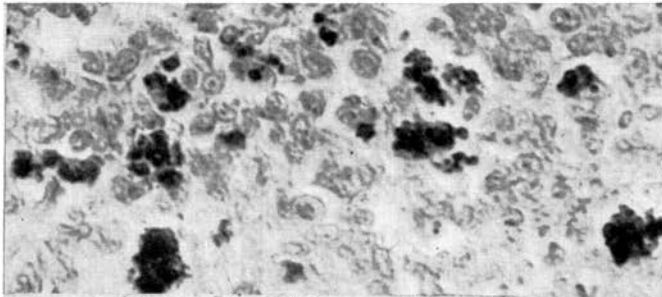


Fig. 1 Chick Spleen. Day 3 Infection

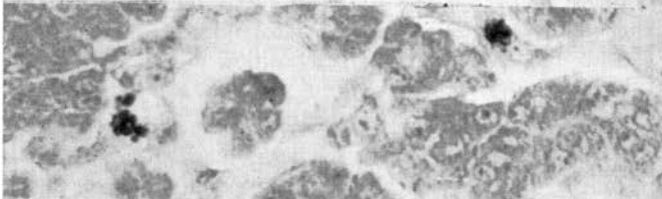


Fig. 2 Chick Liver. Day 3 Infection

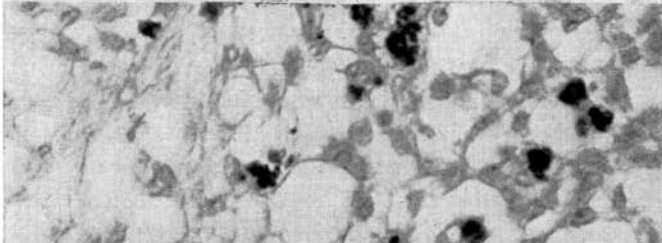


Fig. 3 Chick Lung. Day 3 Infection



Fig. 4 Chick Kidney. Day 3 Infection

DISCUSSION AND RESULTS

On the 13th day following inoculation of ducks with phanerozoite stages of *P. lophurae*, the presence of blood stages of the malaria parasites was demonstrated in blood smears of one duck. Infection was only 0.03 percent, however, and was found in only one of the eight ducks still alive at this time (See Table 2). Parasitized red blood cells were next observed on the 15th day. At this time a low grade of infection could be demonstrated in blood smears from all except one of the seven ducks that had not yet been sacrificed.

In sections of various organs (kidneys, liver and spleen) of the ducks which were sacrificed on days 9 and 12, some malarial pigment appeared, but infected red blood cells were not demonstrated in the blood stream. A slight increase in the number of heterophils was also observed.

On sections of organs of ducks sacrificed on the other days which have been indicated, the presence of malarial pigment could be demonstrated in the adrenals, brain, kidney, liver, lung, pancreas and spleen (See Table 1).

Seven of the ten ducks used as hosts eventually exhibited low grade infections of the red blood cells. Three never did show blood stages of the malarial parasite, *P. lophurae*.

Although all the organs listed in Table 1 were examined very carefully for the presence of exoerythrocytic stages of the parasite, none were observed.

The two following tables show the results of the examinations made of the organs and blood of the ten duck hosts.

Table 1.

Results of examinations of various organs observed in ducks inoculated with phanerozoite stages of *P. lophurae* from turkey brains

Organ examined	No. of ducks examined	Percent of organs showing pigment	No. of organs showing phagocytosis	No. of organs showing exoerythrocytes
Adrenals	6	16.66	0	0
Bone marrow	3	0	0	0
Brain	10	10	0	0
Gonads	10	0	0	0
Heart	10	0	0	0
Kidney	10	20	1	0
Liver	10	60	2	0
Lung	10	20	0	0
Pancreas	10	40	0	0
Spleen	10	90	3	0

Table 2.

Percentages of parasitized red blood cells percent in ducks which had been inoculated with phanerozoite stages of *P. lophurae* from turkey brains. Inoculations occurred when ducks were 5 days old.

Duck No.	Day of the infection									
	12	13	14	15	16	17	18	19	20	21
40	Killed									
41	.00	Killed								
42	.00	.03	Killed							
43	.00	.00	.00	.10	.10	Killed				
44	.00	.00	.00	.10	Killed					
45	.00	.00	.00	.27	.43	.73	Killed			
46	.00	.00	.00	.03	.23	.80	5.57	Killed		
47	.00	.00	.00	.07	.13	.27	.00	.00	Killed	
49	.00	.00	.00	.00	.00	.00	.00	.00	.00	Killed
50	.00	.00	.00	.20	.83	.73	.13	.00	.00	.00
Mean	.00	.008	.00	.11	.29	.51	1.45	.00	.00	.00

SUMMARY

1. Seven of ten White Pekin ducks inoculated with phanerozoite stages of the malarial parasite, *P. lophurae*, exhibited low degrees of infection of the blood stages of the parasite.
2. Phanerozoite stages were observed in none of the ducks hosts, up to the time the last one was sacrificed on the 21st day after inoculation.
3. Phagocytosis of pigment and of infected red blood cells was demonstrated in three different organs including the kidney, liver and spleen.
4. Malarial pigment in the form of tiny granules was observed in seven different organs including the adrenals, brain, kidney, liver, lung, pancreas and spleen.

References

Tomlinson, N. J. and Grocott, R. G. 1944. A Simple Method of Staining Malarial Protozoa and Their Parasites in Paraffin Sections. Amer. Jour. Clin. Path. 14:316-326.

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