

with the 2-methyl-d-glucose of Brigl and Schinle has been definitely established.

P. A. LEVENE  
G. M. MEYER  
A. L. RAYMOND

THE ROCKEFELLER INSTITUTE  
FOR MEDICAL RESEARCH,  
NEW YORK

# THE EFFECT OF PHYSICAL AND CHEMICAL AGENTS ON THE OOCYSTS OF *EIMERIA TENELLA*

Coccidiosis in the domestic hen, *Gallus gallus*, represents a typical parasitological picture. The reservoir is the adult bird acting as a chronic carrier and disseminating a few oocysts. These oocysts under favorable conditions sporulate, and become infective to new hosts. Once the infection is established in younger birds, acute coccidiosis usually results and can only be checked in two basic ways: first, by removal of the infected birds, either by death, isolation or therapeutic measures; and secondly, by preventing the access of uninfected birds to the infective oocysts by sanitation or the creation of conditions unfavorable to the extra-corporal stages of the parasite.

Isolation or death of the infected birds is a costly and generally impractical method of control and, to date, no effective therapeutic agent has been found.

Unless sanitation is rigidly and thoroughly employed, an undercurrent of acute coccidiosis results, which may at any time break out into a devastating epidemic. In large poultry establishments, strict sanitation represents a large economic factor which depletes the net profits to the concern.

The exact conditions necessary for the development of the freshly passed, unsegmented oocyst into the mature, infective stage are but vaguely known, and practically nothing has been reported regarding the lethal limits of the oocysts. With this in mind, the writer has been directing his work toward a possible weak link in the parasitological cycle which will be of economic significance in the control of coccidiosis in poultry.

The results reported in this paper represent a progress report of the work now being undertaken in this laboratory. All work has been done on *Eimeria tenella*, the pathogenic species of coccidium in hens, isolated and described by Tyzzer<sup>1</sup> in 1929.

The prepatent period of coccidiosis produced by *Eimeria tenella* is approximately 165 hours regardless of the number of infective oocysts ingested by the host. There seems to be no correlation between the size of the infecting dose and the height and duration of the patent period. This is not surprising

<sup>1</sup> E. E. Tyzzer, "Coccidiosis in Gallinaceous Birds," *Am. Journ. Hygiene*, X, No. 2, 1, 1929.

since, as Tyzzer has shown, many factors may enter into the situation before oocysts are produced in the host.

There appears little, if any, difference in the susceptibility of the segmented and unsegmented oocysts to heat as shown in the following table:

TABLE I

Temperature	Segmented oocysts, infections produced				Unsegmented oocysts, mortality percentages
51° C.	+	+	+	+	23.5
53° C.	-	+	+	o	100.0
54° C.	+	-	-	-	100.0
55° C.	-	-	-	-	100.0
Controls uninfected	-	-	-	-	
Controls unheated	+	+	+	+	0.0
Time of exposure, 10 minutes					

The criterion used for viability of segmented oocysts was their ability to produce infections when fed in large numbers to chicks known to have been coccidia-free since hatching. The criterion used for viability of unsegmented oocysts in all experiments was their ability to segment when placed in a 2½ per cent. solution of potassium dichromate at 20° C. for 72 hours. All figures, in this and succeeding experiments, are exclusive of natural death and hence represent the mortality due to experimental conditions only.

The time required to kill washed, unsegmented oocysts is inversely proportional to the degree of heat used. Tabulated, the time required for 100 per cent. mortality of unsegmented oocysts is:

TABLE II

Temperature	Time required
45° C.	24 hours
50° C.	1½ hours
55° C.	3 minutes
60° C.	15 seconds
70° C.	15 seconds
80° C.	5 seconds
90° C.	5 seconds

Unsegmented oocysts do not show high resistance to ultra-violet rays. Washed oocysts, exposed to rays produced by a mercury vapor lamp, succumbed as shown in Table III.

The unit of ultra-violet rays used was the zinc sulfide unit of Clark.<sup>2</sup>

Certain reagents were also used in attempts to kill washed, unsegmented oocysts. Briefly, the technique

<sup>2</sup> J. H. Clark, "The Zinc Sulfide Method of Measuring Ultra-violet Radiation and the Results of a Year's Observations on Baltimore Sunshine," *Am. Journ. Hygiene*, IX, No. 3, p. 646, 1929.

TABLE III

Material	Units received	Mortality percentages
Control 1 { Covered with }	0	0
Control 2 { glass slide }	0	0
Slide 1 .....	1/4	8.22
Slide 2 .....	1/4	*
Slide 3 .....	1/2	53.43
Slide 4 .....	1/2	54.75
Slide 5 .....	3/4	98.83
Slide 6 .....	3/4	100.0
Slide 7 .....	1	100.0
Slide 8 .....	1	100.0

\* Slide 2 was accidentally destroyed.

employed was to suspend the oocysts in the reagent for the desired time, wash thoroughly, resuspend in a 2½ per cent. solution of potassium dichromate and incubate at 20° C. At the end of 72 hours of incubation, the oocysts were examined and counted. Those failing to develop were considered dead. The results obtained are presented in Table IV:

TABLE IV

Reagent	Strength	Mortality percentages
HgCl <sub>2</sub>	1 per cent.	100.0
HgCl <sub>2</sub>	0.1 " "	18.4
Iodine suspensoid		
Merck	5 " "	100.0
NaOH	0.5N	0.5
NaOH	2N	1.1
HCl	0.5N	1.3
HCl	2N	4.5
Chlorazene	4 per cent.	0.0
Formol	2 " "	31.4
Formol	5 " "	40.0
Cresol	2 " "	100.0
Cresol	5 " "	100.0
Phenol	2 " "	99.4
Phenol	5 " "	100.0
Controls	2.5 " "	0.0
Potassium dichromate		
Time of exposure, 48 hours.		

The comparative killing power of efficacious reagents are listed in Table V.

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TABLE V

Reagent	Strength	Time required for 100 per cent. mortality
Iodine suspensoid		
Merck	5 per cent.	1 hour or less
Cresol	2 " "	4 to 8 hours
Cresol	5 " "	4 to 8 hours
Phenol	2 " "	48 hours

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FREDERIC FISH

SCHOOL OF HYGIENE AND PUBLIC HEALTH,  
THE JOHNS HOPKINS UNIVERSITY

### GONADECTOMY IN THE GOLDFISH CARASSIUS AURATUS

GONADECTOMY upon fish has not been practiced to any great extent until within the last few years, when the first truly successful operations have been performed. Other work has largely been done to determine the relation between the development of certain secondary sex characteristics, especially nuptial coloration, and the gonads. That relationship has been quite definitely shown to exist. Removal of gonads in the goldfish was undertaken for a different reason, namely, to determine the effect, if any, upon the color change of the young common goldfish from its youthful brown to the orange of the adult. Although there is no difference in the color expression and behavior of the sexes of the goldfish, it was hoped that an upset of a hormonal balance might prove to be of value in a better understanding of the phenomenon, perhaps by changing the rate of time of depigmentation, or even in the total inhibition of the degenerating influence. This work was done as a part of a program of the study of pigment development and pattern formation now in progress at Wesleyan University.<sup>1</sup>

During the autumn of 1928 and 1929 some thirty-nine gonadectomies and twelve operative controls were performed upon young goldfish about five months old. Such fish, hatched in May, were over four centimeters long in October and early November, and no fish under four centimeters was used. Of the complete gonadectomies, twenty-one were upon males and eighteen upon females.

The gonads are paired organs, relatively large and decidedly soft in the goldfish, so that complete removal demands large incisions and careful manipulation. Unfortunately, they do not permit of tearing,

<sup>1</sup> H. B. Goodrich and I. B. Hansen, "The Post-embryonic Development of Mendelian Characters in the Goldfish *Carassius auratus*," in press.