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Effects of severity and duration of infections on oocyst size of Eimeria necatrix, a coccidium of chicken

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MAJARO (O. M.). Effet de gravité et de durée des infections sur la taille des oocystes d'Eimeria necatrix, une coccidie du poulet. Revue Elev. Méd. vét. Pays trop., 1988, 41 (2): 167-170.

La taille des oocystes d'Eimeria necatrix dépend de la gravité et de la durée de l'infection. Plus l'infection est élevée, plus la taille de l'oocyste est réduite. La taille des oocystes trouvés au début de la période patente était significativement plus petite (P < 0,001) que celle des oocystes trouvés plus tard dans le cours de l'infection. Les résultats expérimentaux suggèrent que la taille des oocystes utilisée comme instrument majeur pour l'identification des coccidies n'est pas un élément fiable. Mots clés: Poulet - Eimeria necatrix - Oocyste -Coccidiose - Nigeria.

The oocyst is the most easily accessible stage of any coccidium parasitizing domestic animals including mammals. The size and shape of coccidia oocysts are most often described and employed in the identification of species of coccidia. Although, the size of oocysts is one of the principal characters by which species of coccidia are identified, surprisingly, few studies of size variations have been made. Most of the researchers who employed and relied on oocyst size as a tool of identification of coccidia species did so without considering the influence of severity and duration of infection on the oocyst size of coccidia measured.

Therefore, in view of the importance and the common use of oocyst size in the identification of coccidia species both on the field and research laboratories, this paper examines in details, the influence of severity and duration of Eimeria necatrix infection on the size of oocysts produced, and the reliability of oocyst size as a tool of identification of coccidia species.

Animals: Peterson \times Arbor Acre (P \times AA) broiler strain chickens of 3 weeks of age were employed in this study. The chickens were housed on metal-wirefloored cages which were electrically heated.

Inoculum: Sporulated Eimeria necatrix oocysts for infection were prepared from this culture maintained in this laboratory from birds kept in strict isolation using methods already described by EDGAR and SIEBOLD (4). Estimates of number of infective oocysts fed per os to the chickens were made in Fuschs-Rosenthal counting chambers.

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Experiment: The experiment consisted of 4 treatments with five 3-week-old broiler chickens per pen with 4 replicates per treatment.

Eighty 3-week-old coccidia free broiler chickens were inoculated orally with four levels of infection, 5,000, 15,000, 30,000 or 45,000 oocysts per bird. Faecal droppings were collected 24-hourly from each pen starting from day 7 through 56 days post inoculation; and each daily faecal collection processed immediatly for oocysts. The length and width of at least 20 oocysts of *E. necatrix* recovered from ten different infections under each treatment were measured under light microscopy (×10 ocular × 40 objective) using whipple disc. The readings obtained were multiplied by a predetermined standard factor of the microscope used. Significance tests were carried out by the Student « t »-test as described by SNEDECOR and COCHRAN (9).

The average dimension of *E. necatrix* oocysts in the ten experimentally produced infections are presented in tables I and II.

The results in table I showed the effect of different levels of infection on the size of *Eimeria necatrix* oocysts. Experimental results in table I suggested significant decrease (P < 0.001) in the length and width of *E. necatrix* oocysts as the levels of infection increase. The length and width in microns of oocysts recovered from chickens infected with 45,000 sporulated oocysts was significantly smaller (P < 0.001) than those of the chickens infected with 5,000 or 15,000 oocysts. Table II showed the mean length and mean width in microns of 1,000 unsporulated oocysts of *E. necatrix* from ten different infections observed from day 7 through day 56 post inoculation.

The results obtained showed significant increase (P < 0.001) in length and width as the infection progresses. In each of the ten experimentally produced infections, the mean length and width of the oocysts were significantly different (P < 0.001) from the overall size of the oocysts measured. The size of oocysts of *E. necatrix* recovered early in the patent period was significantly smaller (P < 0.001) than those collected late in the patent period.

TABLE I Size of oocysts with different levels of infections in microns (µ).

Level of infections*		No. of oocysts measured	Mean Length (μ) ± S.D.	Р	Mean width (μ)	
1	5,000	100	21.5 ± 0.8		18.0 ± 0.2	_
2	15,000	100	17.8 ± 0.3	P < 0.001	14.9 ± 0.4	P < 0.001
3	30,000	100	15.2 ± 0.2	P < 0.001	13.1 ± 0.4	P < 0.001
4	45,000	100	13.7 ± 0.4	P < 0.001	12.4 ± 0.4	P < 0.001

^{*} Number of oocysts inoculated per chicken.

TABLE II Size of unsporulated oocysts of Eimeria necatrix passed in faeces on days 7 through 56 post inoculation in microns (µ).

Period of infections	No. of oocysts measured	Mean Length (μ) ± S.D.	Р	Mean width (μ) ± S.D.	Р
D + 7 D + 8 D + 9 D + 21 D + 56	200 200 200 200 200 200	18.1 ± 0.4 18.2 ± 0.2 18.4 ± 0.4 21.7 ± 0.9 21.7 ± 1.1	P < 0.001 P < 0.001 P < 0.001 P < 0.001	14.4 ± 0.3 15.1 ± 0.6 15.0 ± 0.6 18.1 ± 0.3 18.2 ± 0.2	P < 0.001 P < 0.001 P < 0.001 P < 0.001

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Many researchers have isolated, described and identified many *Eimeria* species of domestic animals. These identifications were based commonly on the morphology and the size of coccidia oocysts. Surprisingly, most of these researchers did not relate the size of the oocysts measured to the degree of the severity of infection or duration of infection. However, few workers reported the effects of severity or duration of infection on oocyst size of different *Eimeria* species of poultry (1, 2, 3, 5, 6).

In the present study, the experimental results obtained suggest that oocysts became significantly (P < 0.001) smaller, the higher the levels of infection. This observation differs from that of JONES (6) who reported that oocysts size of *Eimeria acervulina* was independent of the severity of infection, whereas, DUNCAN (3) working with *Eimeria labbeana* agrees with the present findings, DUNCAN (3) observed that oocysts were markedly larger with an infection produced by a single oocyst than a massive dose of infection. It appears therefore, that those chickens that received mild to moderate level of infection had better potential to develop than those that received massive dose of infection, hence the marked differences in the sizes of oocysts produced at different levels of infection.

Experimental findings in the study also revealed that the size of oocysts of E. necatrix was dependent on the duration of infection. Smaller oocysts were recovered early in the patent period and later succeeded by bigger oocysts as the infection progressed. In a similar study, FISH (5) working with E. tenella observed a progressive increase in the length and width of oocysts as the infection progressed which agrees with the present study. JONES (7) observed no change in the dimensions of the oocysts of E. acervulina contrary to the present findings. BECKER et al. (2) working with E. brunetti in chicken observed that smaller oocysts were produced early in infection and later succeeded by larger oocysts thus confirming the present findings despite differences in the Eimeria species employed. BECKER et al. (1) studying E. necatrix observed a slight though significant increase in oocyst size after the first 3 days of patency followed by a reduction in length towards the end of the experiment. There was significant increase (P < 0.001) in the length and width of E. necatrix oocysts a day after patency up to 56 days when the experiment was terminated. There was no reduction neither in the length nor the width of the oocysts throughout the duration of the experiment contrary to the findings of BECKER et al. (1). BECKER et al. (1) and other early researchers did not mention the duration of their experiment as well as the levels of

infection. This might be responsible for the variations in the size of oocysts of Eimeria species reported by most of the researchers. The total number of oocysts measured by individual researcher, the non-standardised level of infection used and the strains of Eimeria species under investigations might also contribute to the oocyst size variations. Therefore, it is evident from the present investigation that size of oocysts decreases the higher the level of infection and progressively increases as the infection progresses. Since there is no prescribed standard levels of infection and particular period during the course of coccidial infection when oocysts could be collected and measured, the variations of oocyst size would remain and the application of oocyst size as a diagnostic tool remains meaningless and unreliable.

The effects of different levels of infection and duration of infection on the size of *Eimeria necatrix* oocysts were investigated. Experimental findings revealed that oocyst size was not only dependent on the levels of infection but also on the duration of infection. Researchers are therefore, advised not to rely heavily on the oocyst size as a valid criterion of diagnosis of coccidia. Shape Index (length/width) might be suggested instead of oocyst size, and if oocyst size is employed as a tool of diagnosis, it should be considered along with other criteria suggested by TYZZER (10) and KREIER (8).

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The oocyst size of Eimeria necatrix is dependent on severity and duration of infection. The higher the severity of infection the smaller the oocyst size. The size of the oocysts recovered early in the patent period was significantly smaller (P < 0.001) than those recovered late in the course of infection. Experimental results suggested that size of oocysts as a major tool of coccidia identification is not reliable. Key words: Chicken - Eimeria necatrix - Oocyst - Coccidiosis - Nigeria.

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