Research and Reviews in Parasitology, 53 (1-2): 27-31 (1993)
© 1993 Asociación de Parasitólogos Españoles
Editorial Fontalba, S.A.
Printed in Spain

STUDIES ON THE EGG OUTPUT VARIATIONS AND DETERMINISM OF *METASTRONGYLUS* SPP., LUNGWORMS OF THE WILD BOAR (*SUS SCROFA* L.)

J.F. HUMBERT[1] & J. DROUET[2]
[1]*INRA Tours, Station P.A.P., Laboratoire d'Ecologie des Parasites, 37380 Nouzilly, France*
[2]*Laboratoire de Biologie Animale et Parasitologie, Université d'Orléans, B.P. 6759, 45067 Orléans Cedex 02, France*

Received 28 October 1991; accepted 9 June 1992

REFERENCE: HUMBERT (J.F.) & DROUET (J.), 1993.— Studies on the egg output variations and determinism of *Metastrongylus* spp., lungworms of the wild boar (*Sus scrofa* L.). *Research and Reviews in Parasitology*, 53 (1-2): 27-31.

ABSTRACT: The variations in the production of Metastrongyle eggs in the faeces of a wild boar population were studied over two years. Four parasite species were involved: *Metastrongylus salmi, M. confusus, M. pudendotectus, M. asymmetricus.* The first year sampling was characterized in winter by a strong increase in egg production. During the second winter, no egg-laying peak was observed. The seasonal variations in the egg output are not directly linked with climatic factors. They depend on the dynamics and the intensity of the first infestations of the young wild boars. The peak of egg production was also determined by the very high infestations of the young animals in the studied population. These massive infestations are observed when the animals eat at feeding-centers which are places of high parasitic risk. The frequency of the feeding-center visits depends on trophic factors in the forest.

KEY WORDS: Egg output, lung nematodes, *Metastrongylus* spp., wild boar, *Sus scrofa*, transmission.

INTRODUCTION

Metastrongyles are parasitic nematodes of the wild boar and domestic hogs. These lungworms have an indirect life cycle which was first demonstrated by HOBMAIER & HOBMAIER (1929). The intermediate hosts are earthworms, particularly the anecic species.

These earthworms are infected after ingesting eggs of Metastrongyles excreted in the dung of wild boar. Thus, egg output constitutes the first step in the parasite transmission. This phase of the life cycle has been the focus of much experimental work (KATES, 1941; DUNN, 1956; MACKENZIE, 1959; USTINOV, 1963; JAGGERS & HERBERT, 1968; ROSE, 1973).

However, no study describes this phenomenon in a population of wild animals in their natural habitat. The aims of the present paper are therefore, first, to determine seasonal variations in the egg production of Metastrongyles in a population of wild boar in the Chambord game reserve (France) and, second, to look for the controlling factors of these seasonal fluctuations.

MATERIAL AND METHODS

Collection area

This study was carried out in the Chambord game reserve (01° 30' E, 47° 35' N; altitude, 80 m) which occupies approximately 5400 ha of deciduous forest, pine woods, coppice and scrubs. Food is distributed to the animals at feeding centers during years in which wild fruit production (acorns, etc.) in autumn is low.

The wild boar population is estimated between 800 and 1500, depending on the year. Parasitism by Metastrongyles is high in this population. Four species are found in the lungs: *Metastrongylus pudendotectus* Vostokov, 1905; *M. salmi* Gedoelst, 1923; *M. confusus* Jansen, 1964; and *M. asymmetricus* Noda, 1973 (Nematoda: Metastrongylidae) (HUMBERT & FERTE, 1986; HUMBERT, 1988; HUMBERT & HENRY, 1989). The prevalence of the parasites is close to 92% in the boar population. The mean intensity exceeds 140 strongyles per animal (HUMBERT & DROUET, 1990).

Sampling method

From September 1985 to August 1986 and from June 1987 to August 1987, 32 to 35 dung samples were taken at the end of each month at feeding centers. Between these two periods, sampling was difficult due to the high acorn production in Autumn 1986. The animals thus no longer fed in specific areas, making sampling of fresh faecal matter impossible in a short time. However, samples of faecal matter were taken from rectum of boars killed during the hunting season between December 1986 and February 1987. To determine the number of eggs in faeces, eggs were recovered following flotation in a high density solution of $MgSO_4$ (d = 1,27) and counted in Mac Master slides (RAYNAUD *et al.*, 1979). Results are expressed in the number of eggs per gram (e.p.g.) of faecal matter.

Data analysis

Comparing means was done using the Student test for large samplings (n > 30) and the Mann-Whitney U-Test for small samplings. The index value "I" ($I = S^2/X$ or S^2 = variance, X = mean) and "d" allow to determine the sampling distribution model [$d = \sqrt{\chi^2} - \sqrt{2v-1}$, $v = n-1$ and $\chi^2 = I(n-1)$]. A Chi-square test (goodness-of-fit) was used for agreement with the negative binomial.

RESULTS

Two phases could be distinguished during the egg production cycle (Table 1) for the first year sampling (September 1985-August 1986):

— the first phase (October 1985 to January 1986) was characterized by a strong increase in the number of eggs per gram of faecal matter (>50 in January);
— the second phase (February 1986 to August 1986) was characterized by a marked decrease in egg production, attaining a zero value in July.

Differences between monthly means were statistically

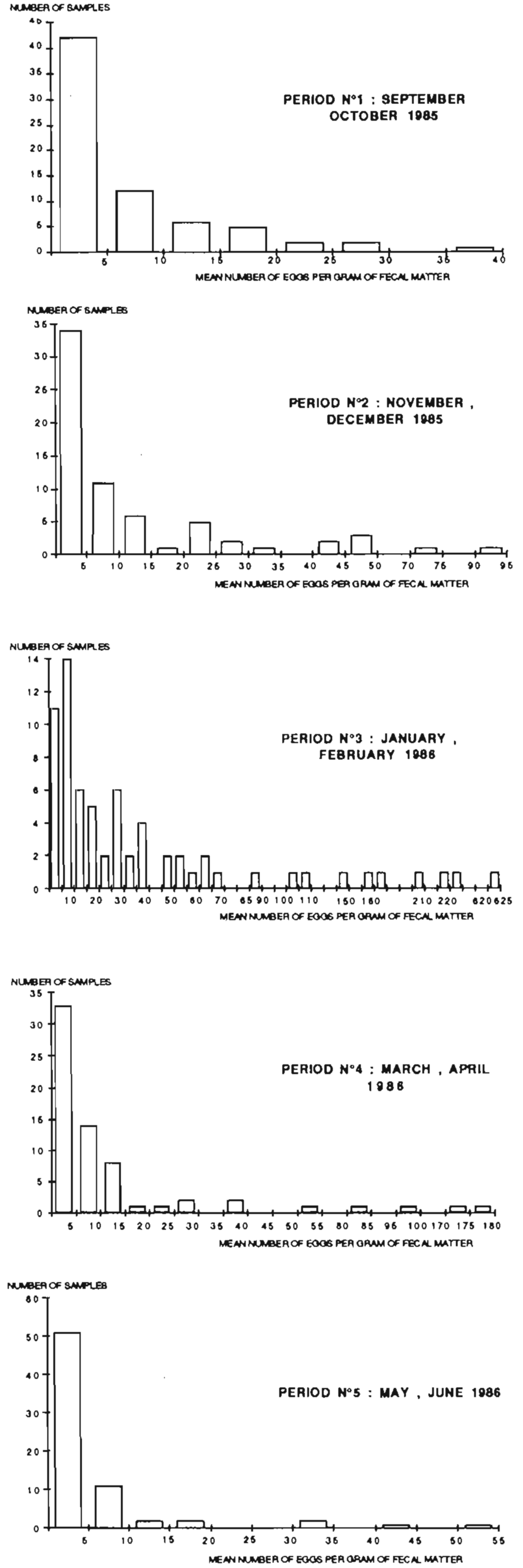

Fig. 1.— Distribution of droppings as a function of the mean number of eggs contained per gram of faecal matter.

significan for only 1 out of 2 months ($P > 95\%$) which allowed the regrouping of results over 2-month periods at the end of the study (first period: Sept.-Oct. 1985; second period: Nov.-Dec. ...).

The mean number of eggs in the contaminated samples was negatively correlated to the number of non-contaminated droppings (Table 1). The proportion of the latter was thus 7,7% in January and February, indicating that during this period more than 92% of the boar were parasitized. These findings agree with results reported in another study (HUMBERT & DROUET, 1990) for the estimation of the prevalence of adult parasites after necropsy of wild boar lungs.

The study of the distribution of the number of eggs per dropping (Fig. 1) revealed their highly aggregated character and showed that they fit the negative binomial distribution reasonably well ($P > 95\%$). Thus, a large number of droppings contained very few eggs and conversely a very low proportion was highly contaminated. The egg-laying peak (January, February 1986) was characterized by the appearance of samples containing a very large number of eggs and a decrease in samples containing few or no eggs. An analysis of the variations in the number of values found in three classes defined around the yearly egg output mean ($X = 14$) supported this observation (Table 2): the increase in egg output was partly due to a decrease in the number of samples containing none or very few eggs ($X < 5$) and partly due to an increase in the number of samples containing many ($X > 20$). The number of values found around the mean ($5 \leq X \leq 20$) did not change greatly.

During the second year, no complete egg-cycle could be established because of the absence of feeding-center visits by boars, thus preventing a regular harvest of faecal matter. Nevertheless, results were obtained that were similar to the two characteristic periods determined the previous year, maximum egg-laying in winter and absence of eggs in summer. No egg excretion peak was observed during the second winter (Table 3) and egg release followed in summer.

DISCUSSION

Two years of sampling showed the existence of monthly variations in egg output in faecal matter. These variations were characterized in the year 1985-86 by an egg-laying peak in the winter and a total absence of eggs in the summer. The results obtained the following year revealed that these fluctuations were not constant because during the same periods, the previous two observations were not noted. The emergence of the egg-laying peak was not directly linked with climatic factors. A negative correlation existed between the monthly mean in temperature and the level of egg-laying (Fig. 2) in the first year. Despite comparable temperatures and rainfall in winter of the second year, no egg-laying peak was observed. Low temperature does not appear to be a factor inducing massive egg output.

On the other hand, new infestations appear to be a determining factor. Studies by JAGGERS & HERBERT (1968) showed experimentally that the first infestation of domestic hogs with Metastrongyle larvae was always followed one month later by an increase in the number of eggs in the faeces of these animals. They determined that the ratio between the number of larvae ingested by the swine to the number of eggs per gram of faecal matter excreted one month later by these animals was equal to 10 at a maximum egg-laying peak. They also showed that re-infestations do not cause new increases in the number of eggs produced. These results can be used to compare conditions needed in a natural environment for an egg-laying peak to be observed. Young animals (age <1 year) must be present in populations because they are the only ones not likely to be parasitized (HUMBERT & DROUET, 1990) and thus the only ones able to be infested for the first time. To be significant, first infestations of these young boars must occur at the same time during the increase in the number of eggs produced by the population. If the number of newly-infested animals is too low, it will not appear in the final mean. The number of larvae ingested by the young wild boar must be sufficiently numerous to cause a large increase in the monthly mean egg-output (recall that the ratio of the number of larvae ingested to the number of eggs in the droppings is 10).

Such conditions were met in this study since there were young animals in the wild boar population. The births in 1985 were spread out from April to September. These animals were non-infested because the autumn of that year (Fig. 2) was characterized by an abnormal drought which resulted in an absence of surface activity of anecic worms (HUMBERT, 1988). It was thus impossible for

	Number of samples	Mean number of eggs per gram of faecal matter	Confidence limits*	Number of samples without egg
September 1985	35	9,2	± 3,2	12 (34%)
October	35	5,3	± 2,1	08 (23%)
November	35	9,3	± 5,9	15 (43%)
December	34	15,0	± 6,5	07 (21%)
January 1986	35	54,8	± 37,6	03 (9%)
February	33	39,4	± 19,9	02 (6%)
March	32	16,1	± 12,4	08 (25%)
April	34	14,1	± 11,0	01 (3%)
May	35	4,9	± 3,4	15 (43%)
June	35	5	± 3,1	12 (34%)
July	35	0	—	34 (97%)
August	35	0	—	35 (100%)

Table 1.— Results of the first year sampling. * = 0,95 confidence limits (± 1,96 S.E.).

	Sept.-Oct. 85	Nov.-Dec.	Jan.-Feb. 86	Mar.-Apr.	May-Jun.
$X \leq 5$	60,0*	49,3	16,2	50,0	72,9
$5 < X < 20$	32,8	29,0	36,8	34,9	21,5
$X \geq 20$	7,2	21,7	47,0	15,1	5,6

Table 2.— Distribution of samples as a function of lungworm eggs contained per gram of faecal matter (X). * = expressed as %.

	Dec. 86	Jan. 87	February	June	July	August
Mean number of eggs per gram of faecal matter (X)	5,6	4,6	16,9	14,3	7,3	9,8
Confidence limits (0,95)	$0,1 < X < 25$	$2 < X < 30$	$0,1 < X < 30$	$5 < X < 24$	$0,5 < X < 15$	$1 < X < 19$

Table 3.— Results of the second year sampling.

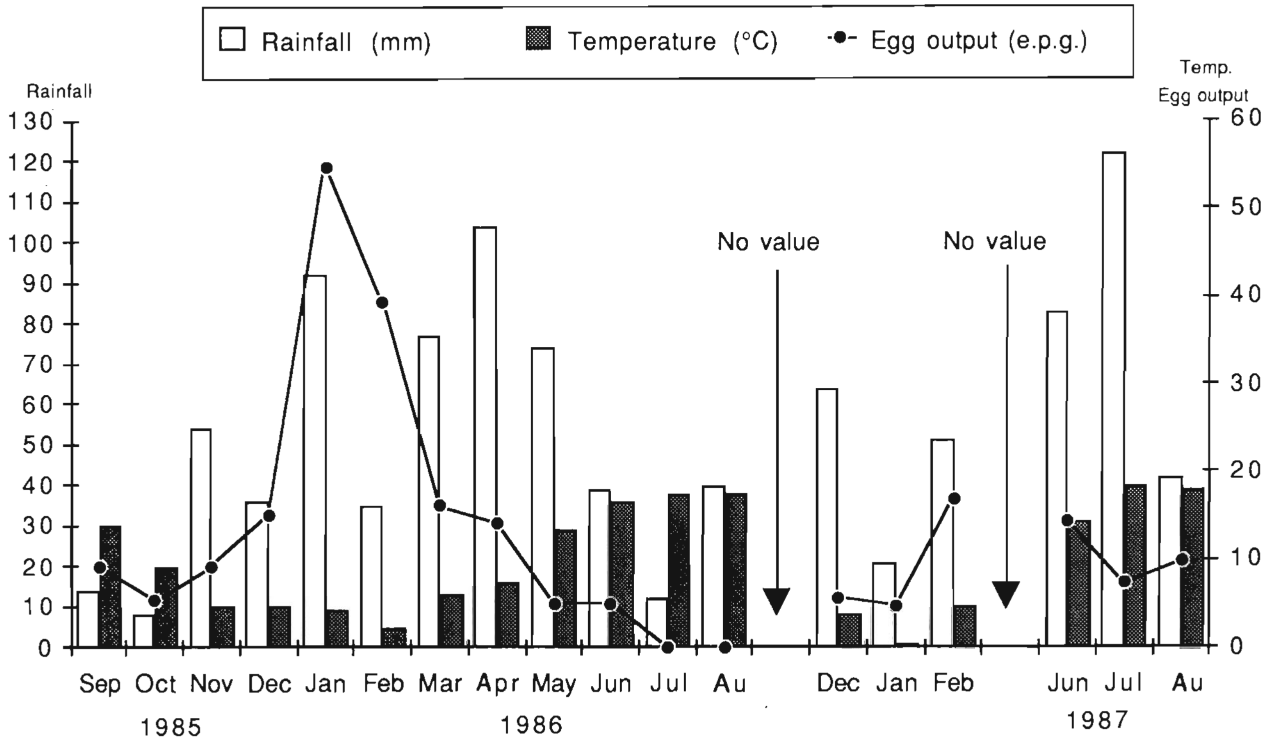

Fig. 2.— Relationships between climatic factors and monthly egg-laying means.

young animals to be infested before the rainfalls of November. Despite the disparity in births and thus in weanings, the young wild boars were not able to be infested, by eating earthworms, until the end of November, at which time the surface activity of earthworms started up again and the first infestations occured. The intensity of the first infestations was high. Autumn 1985 was marked by an absence of wild fruits in the forest which resulted in the wild boars feeding at the centers. In a preceeding study (HUMBERT & HENRY, 1989), these feeding centers were shown to be places of high risk for massive infestation. Over these zones more than 90% of the earthworms were parasitized, containing on the average 20 metastrongyle larvae per worm.

An additional observation confirmed that the increase in egg production was caused by the first infestations of young boars in the population. The sampling distribution of the number of eggs contained per gram of faecal matter were all aggregated, regardless of the period of study. In the boar population, a large number of pigs left droppings containing very few eggs and conversely, a small number of boars left highly contaminated droppings. It was thus interesting to correlate this observation to the results obtained from the study of boar parasitism (HUMBERT & DROUET, 1990). In this study, it was observed that the distribution of adult Metastrongyles in the boar population was contagious and that it agreed with the negative binomial. The analysis of this distribution showed that the most parasitized wild boars were very young animals (age < 1 year). The low number of highly contaminated dung was therefore probably left by these highly parasitized animals.

Because of the environmental factors during the second year, no egg-laying peak was observed. The rainfall in autumn 1986 was normal, the earthworms were thus accessible to predation, which resulted in a longer duration of the first infestations during the weanings of the young boars. Moreover, the abundant acorn production in the autumn of 1986 was followed by a cessation in visits by the wild boar to the feeding-centers, thereby reducing the probability of massive infestation.

CONCLUSION

This study showed seasonal variations in egg output of Metastrongyles. The variations depended mainly on the dynamics and the intensity of first infestations which were themselves determined by the following two factors:

— trophic factors: when the forest food supply (acorns) is abundant, the wild boar will randomly search for food in the park, thereby reducing the risk of massive infestation linked to feeding-center visits;

— climatic factors: accessibility to earthworm predation is determined by climatic factors; a lack of soil mosture or temperatures below zero (°C) result in an absence of surface activity of anecic species.

The results of this study showed that an analysis by coprology must extend over a sufficient period of time if sanitary conditions are to be evaluated in a given population. It is possible to underestimate the importance of an helminthosis following the period of the year in which the sampling is taken. On the other hand, in the case of monthly coprology follow-ups, the appearance of an egg-laying peak might mean that the young animals are highly infested and thus have a high risk of mortality.

ACKNOWLEDGEMENTS

We would like to thank C. Mary for permission to work at the game reserve of Chambord and F. Forget for help with the field work.

REFERENCES

DUNN (D.R.), 1956.— Studies on the pig lungworm (*Metastrongylus* spp.). Experimental infection of pigs with *M. apri*. *British Veterinary Journal*, 112: 327-337.

HOBMAIER (A.) & HOBMAIER (M.), 1929.— Die Entwicklung der Larve des Lungenwurmes *Metastrongylus elongatus* des Schweines und ihr Invasionweg, sowie vorläufige Mitteilung über die Entwicklung von *Choerostrongylus brevivaginatus*. *München Tierärztliche Wochenschrift*, 80: 365-369.

HUMBERT (J.F.), 1988.— *Les Métastrongles, nématodes parasites pulmonaires du sanglier (Sus scrofa L.). Biologie, épidémiologie et dynamique de la transmission*. Thèse de Doctorat, U.F.R. Faculté des Sciences, Université d'Orléans, Orléans, 161 pp.

HUMBERT (J.F.) & DROUET (J.), 1990.— Enquète épidémiologique sur la Métastrongylose du sanglier (*Sus scrofa* L.) en France. *Gibier-Faune Sauvage*, 7: 67-84.

HUMBERT (J.F.) & FERTE (H.), 1986.— Le parasitisme helminthique du sanglier (*Sus scrofa* L.) en France. *Symbioses*, 18: 75-84.

HUMBERT (J.F.) & HENRY (C.), 1989.— Studies on the incidence and transmission of lungworms and stomach worms of the wild boar (*Sus scrofa* L.) in France. *Journal of Wildlife Diseases*, 25: 335-341.

JAGGERS (S.E.) & HERBERT (I.V.), 1968.— Studies on the resistance of pigs to the lungworms *Metastrongylus* sp. Infections in minimal disease pigs from eight weeks of age. *Journal of Comparative Pathology*, 78: 161-172.

KATES (K.C.), 1941.— Observations on the viability of eggs of lungworms of swine. *Journal of Parasitology*, 27: 265-272.

MACKENZIE (A.), 1959.— Studies on lungworm infection of pigs. III. The progressive pathology of experimental infections. *Veterinary Record*, 71: 209-214.

RAYNAUD (J.P.), LEROY (J.C.), VIRAT (M.) & NICOLAS (J.A.), 1979.— Une technique de coproscopie quantitative polyvalente par dilution et sédimentation en eau, flottaison en solution dense (D.S.F.) et numération en lame de Mac Master. *Revue de Medecine Vétérinaire*, 130: 377-404.

ROSE (J.H.), 1973.— Lungworms of the domestic pigs and sheep. In: *Advances in Parasitology* (B. Dawes edit.), Academic Press, London and New-York, 2: 559-599.

USTINOV (D.), 1963.— Length of life of metastrongyle eggs in the external environment and resistance to various temperatures. *Trudy Vsesoyuznogo Instituta Gel'mintologii*, Moskva, 10: 56-63.