

MODELS FOR THE DISTRIBUTION ON PASTURE OF INFECTIVE LARVAE OF THE GASTROINTESTINAL NEMATODE PARASITES OF SHEEP

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Summary

Two models are proposed for the distribution on pasture of infective larvae of the gastrointestinal nematode parasites of sheep. These models were developed to include as many as possible of the known biological components. Procedures for estimating the parameters of the models are outlined and advantages of these models over earlier attempts to describe the distribution of infective larvae on pasture are briefly discussed.

I. INTRODUCTION

In studies on the population dynamics of gastrointestinal nematode parasitism in sheep it is important to obtain some measure of the rate at which the infective larvae of these parasites are ingested by the grazing animal. It is likely that the rate of larval intake by the host depends largely on the grazing behaviour of the sheep, and the distribution and abundance of infective larvae on the pasture. In this paper, specific models are developed for the distribution of infective larvae on pasture incorporating as many of the known biological components as possible.

A brief description of the biological processes requiring mathematical treatment follows:

- (1) Eggs which are laid by the female parasites in the alimentary tract of the host are passed out in the faeces onto the pasture. Under favourable environmental conditions, the eggs undergo several stages of development, culminating in the appearance of third-stage larvae which are infective for the host. The infective third-stage larvae migrate only a small distance away from the faecal deposit to adjacent herbage (Dinaburg 1944; Furman 1944), where they may survive for a limited period. The host becomes infected either by penetration of the infective larvae through its skin, or, for the great majority of these parasite species, by ingestion of the infective larvae with the herbage.
- (2) The rate of development to the third larval stage and the rate of mortality, both during development and in the third larval stage, depend on micro-climatic factors, principally temperature and humidity.
- (3) Observations by Crofton (1954) on fields being grazed by sheep have shown that the distribution of faecal deposits is not random, i.e. is not described by a Poisson law. He has also shown that sheep, while grazing,

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move in a more or less well-integrated group, which at any point in time occupies an area rarely less than one-sixth and never more than one-third of the total pasture area. Assuming that the periods of grazing and defaecation are broadly coincident, it is likely that the distribution of faeces deposited per unit area of the total pasture area within a unit of time will be "overdispersed" statistically, i.e. the variance of the distribution will be appreciably greater than the mean (Bliss and Fisher 1953).

- (4) It has been shown by Hunter and Quenouille (1952) that replicate faecal worm egg counts (eggs per gram of faeces) from the same sheep followed a Poisson series, but that the distribution of egg counts between sheep fitted reasonably well to the negative binomial distribution with $k \approx 0.7$. It is likely, therefore, that the distribution of total egg numbers in faecal masses deposited by a flock of sheep in a unit of time will also be overdispersed.

In the following development these biological aspects are important, since we require mathematical models to describe the distribution of third-stage larvae on the pasture. Thus the distribution of faecal deposits on the pasture, the distribution of egg output of the flock at a given point in time, and the rate of mortality of the larval stages of the parasite must be considered during construction of the models. These points will be emphasized in the next section.

II. THE DISTRIBUTION

We consider the situation where a fixed number, S , of sheep are introduced onto a pasture of total area A at time $t = 0$ and are removed at $t = t_1$. The probability generating function (p.g.f.) for the distribution of faecal deposits for the i th sheep is assumed to be of the form

$$\{p/(1-qS)\}^{a_i t}, \quad (1)$$

where $q = 1-p$ and $i = 1, 2, \dots, S$. The expression (1) specifies a negative binomial distribution with parameters p and $a_i t$. Now, if the effects of sheep on the total distribution of faecal deposits are stochastically additive and independent, then the p.g.f. for the S sheep may be written as

$$\{p/(1-qS)\}^{at}, \quad (2)$$

with

$$a = \sum_{i=1}^S a_i.$$

Now let the time segment $[0, t]$ be partitioned into n intervals of equal length t/n and label the points of subdivision $t_0, t_1, t_2, \dots, t_n$. Then, because of the infinite divisibility property of (2), the p.g.f. of total faecal deposits for any of the subintervals is given by

$$\{p/(1-qS)\}^{at/n}. \quad (3)$$

Furthermore, consider the p.g.f. of the number, N , of eggs dropped onto the pasture during the i th time interval $[t_{i-1}, t_i]$. We denote the average number of eggs per faecal deposit at some time $\bar{t}_i \in [t_{i-1}, t_i]$ by $\lambda(\bar{t}_i)$ and assume that the distribution of the numbers of such eggs follows a Poisson law with parameter $\lambda(\bar{t}_i)$, which is a continuous function of time. (Initially, we assume, contrary to (4) of the Introduction, that $\lambda(\bar{t}_i)$ is the same for all sheep. Subsequently, this restriction will be removed.) Moreover, if the probability that a given egg is in the infective larval stage at time t after being dropped is denoted by the continuous function $f(t)$, then it is easily shown that at time t , the distribution of larvae developing from eggs deposited during the i th subinterval is again Poisson with parameter

$$\mu(t, \bar{t}_i) = \lambda(\bar{t}_i) f(t - \bar{t}_i).$$

Thus the p.g.f. for the number of larvae surviving at time t from the i th interval is given approximately by

$$\left\{ p/[1 - q \cdot \exp\{-\mu(t, \bar{t}_i)(1-s)\}] \right\}^{at/n}. \quad (4)$$

Therefore, for the p.g.f. of the total number of larvae on the pasture at time t we have also approximately,

$$g_n(t, s) = \prod_{i=1}^n \left\{ p/[1 - q \cdot \exp\{-\mu(t, \bar{t}_i)(1-s)\}] \right\}^{at/n} \quad (5)$$

The larger n becomes, the smaller is the interval width tn^{-1} and the closer does $g_n(t, s)$ approach to the conceptual p.g.f. with continuous time increments.

We therefore define the limiting p.g.f. as follows:

$$\begin{aligned} g(t, s) &= \lim_{n \rightarrow \infty} g_n(t, s) \\ &= \lim_{n \rightarrow \infty} \exp \left\{ atn^{-1} \sum_{i=1}^n \ln \left[p/[1 - q \cdot \exp\{-\mu(t, \bar{t}_i)(1-s)\}] \right] \right\} \\ &= \exp \left\{ at \ln p - a \int_0^t \ln [1 - q \cdot \exp\{-\mu(t, x)(1-s)\}] dx \right\} \end{aligned} \quad (6a)$$

by the continuity of e^v and the definition of the Riemann integral.

In the above derivation it was implicitly assumed that $t \leq t_1$. However, if $t > t_1$ we consider the interval $[0, t_1]$ and apply entirely analogous reasoning to that used above. Thus, for $t > t_1$

$$g(t_1, s) = \exp \left\{ at_1 \ln p - a \int_0^{t_1} \ln [1 - q \cdot \exp\{-\mu(t, x)(1-s)\}] dx \right\}. \quad (6b)$$

Since $\lambda(t)$ and $f(t)$ are continuous functions of t , the integral $\int_0^t [\lambda(x)f(t-x)]^a dx$, $a \geq 0$, exists, and if the expressions (6a) and (6b) are suitably differentiated with respect to s , it is found that

$$\begin{aligned} E_s(N) &= aqp^{-1} \int_0^t \lambda(x) f(t-x) dx & t \leq t_1 \\ &= aqp^{-1} \int_0^{t_1} \lambda(x) f(t-x) dx & t > t_1 \\ V_s(N) &= aqp^{-2} \int_0^t [\lambda(x) f(t-x)]^2 dx + E_s(N) & t \leq t_1 \\ &= aqp^{-2} \int_0^{t_1} [\lambda(x) f(t-x)]^2 dx + E_s(N) & t > t_1 \end{aligned} \quad (7)$$

where the subscript S is used to emphasize that these values refer to the application of S sheep to the area.

It is important to notice that (6a) and (6b) are not only sheep additive, but space additive as well. Although these formulae refer to a particular area of size A , they may be considered as the convolution of the effects of S sheep on L areas each of size A/L . In this case, each subplot has a negative binomial distribution with parameters p and at/L . These features emphasize the flexibility of the model.

It is clear from (7) that if the sheep are left on pasture indefinitely, then the following theoretical equilibrium is reached at $t = \infty$,

$$e(N) = aqp^{-1} \lim_{t \rightarrow \infty} \int_0^t \lambda(x) f(t-x) dx.$$

In order to obtain bounds for $e(N)$, notice that

$$aqp^{-1} \inf_x [\lambda(x)] \int_0^\infty f(x) dx \leq e(N) \leq aqp^{-1} \sup_x [\lambda(x)] \int_0^\infty f(x) dx,$$

and therefore

$$e(N) = \lambda(\bar{x}) \int_0^\infty f(x) dx, \quad \bar{x} \in [0, \infty].$$

However, when the sheep are removed at $t = t_1$, we obtain

$$\begin{aligned} e(N) &= aqp^{-1} \lim_{t \rightarrow \infty} \int_0^{t_1} \lambda(x) f(t-x) dx \\ &= aqp^{-1} \int_0^{t_1} \lambda(x) \left[\lim_{t \rightarrow \infty} f(t-x) \right] dx \\ &= 0, \end{aligned}$$

as required, since $f(\infty) = 0$.

If, in the above model, sheep additivity does not seem to be a valid assumption, then the model can be applied to varying flock sizes and different α values estimated for each flock size. This, in fact, would allow the null hypothesis of sheep additivity to be investigated, since under this hypothesis the α 's should be proportional to the flock sizes.

However, as mentioned in the Introduction, it may not be safe to assume a constant $\lambda(t)$ for each sheep as was implicitly done in the above derivations. If this is so, and sheep additivity can be assumed, then each sheep must be given its own $g(t, s)$ and the parameters α_j and $\lambda_j(t)$, $j = 1, 2, \dots, S$, must be separately estimated and the average number of larvae on the pasture would then be given by

$$E_s(N) = qp^{-1} \sum_{j=1}^S \alpha_j \int_0^t \lambda_j(x) f(t-x) dx. \quad (8)$$

Some investigation would be necessary in order to show whether or not expressions (7) are satisfactory approximations.

If the investigator is seriously concerned about both the assumptions of sheep additivity and constant $\lambda(t)$, then the whole model may have to be changed. One way of doing this is to assume that the numbers of larvae at time t which develop from eggs deposited on the pasture during the i th time interval has p.g.f.

$$\{u/1-v[1-f(t-\bar{t}_i)(1-s)]\}^{k(\bar{t}_i)}, \quad (9)$$

where $k(t)$ is an arbitrary function of time and $f(t-x)$ is as defined earlier. The p.g.f. (9) is the result of compounding a negative binomial distribution [parameters u and $k(\bar{t}_i)$] with a binomial distribution [parameter $f(t-\bar{t}_i)$]. Analogous reasoning to that used earlier shows that in this case, assuming the same distribution of faecal deposits,

$$g(t, s) = \exp \left\{ at \ln p - a \int_0^t \ln [1 - q\{u/(1-v[1-f(t-x)(1-s)])\}^{k(x)}] dx \right\}, \quad (10)$$

which is an extremely complicated distribution. However, the mean turns out to be

$$E_s(N) = atqp^{-1} v u^{-1} \int_0^t k(x) f(t-x) dx, \quad (11)$$

which is of the same form as (7). The increased generality is achieved by the introduction of the additional parameter u . Of course, the same type of modification for $t > \bar{t}_i$ applies to (10) as for (6a).

It is interesting to notice that (11) can be written down directly from other considerations. If $f(s)$, $g(s)$, and $k(s)$ are p.g.f.'s of random variables X , Y , Z , then the mean of the compound variate specified by $f(g(k(s)))$ is simply $E(X) \cdot E(Y) \cdot E(Z)$. Thus for any time subinterval i , the mean number of third-stage larvae is given by

$$atn^{-1}qp^{-1}vu^{-1}k(\bar{t}_i)f(t-\bar{t}_i),$$

and summing this expression over all intervals, the contribution from intervals being stochastically independent, and letting $n \rightarrow \infty$, gives the result (11). Uniqueness is guaranteed by the Continuity Theorem (Feller 1960, p. 262).

We now turn to problems of estimation and consider model (6a) in detail. Suitable procedures for the other models can be worked out in a similar way.

III. ESTIMATION

The quantity which is of major practical importance is the concentration of larvae per unit of area. In order to estimate this in an efficient manner from a given area A and a given number of sheep S , we subdivide A into L subunits of equal size a so that $La = A$. Moreover, the time interval $[0, t]$ is also subdivided into T equal intervals of length t/T . The p.g.f. for faecal deposits corresponding to any subinterval and for areas of size a , is given by

$$\{p/(1-qs)\}^{at/TL}.$$

Now S sheep are introduced at $t = 0$ and the distribution of faecal deposits in the L subareas recorded for each time interval. If r_{ij} is the number observed in the j th plot for time interval i , then, if we let $at = \beta$, $k' = \beta/LT$, and $m' = k'\gamma$, where $\gamma = (1-p)/p$, moment estimates of k' and m' are given by

$$\begin{aligned}\hat{m}'_i &= \bar{r}_i, \\ \hat{k}'_i &= \bar{r}_i^2 (S_i^2 - \bar{r}_i)^{-1},\end{aligned}$$

where

$$\bar{r}_i = \sum_{j=1}^L r_{ij}/L,$$

and

$$S_i^2 = \sum_{j=1}^L (r_{ij} - \bar{r}_i)^2 / (L-1).$$

From Anscombe (1950) the variances of $\hat{m}'_i$ and $\hat{k}'_i$ are given by

$$\begin{aligned}V(\hat{m}'_i) &= m'(1+\gamma)/L \\ &= \beta\gamma(1+\gamma)/L^2T, \\ V(\hat{k}'_i) &= 2k'(k'+1)(1+\gamma)^2/\gamma^2L \\ &= 2\beta(\beta+LT)(1+\gamma)^2/\gamma^2L^3T^2,\end{aligned}$$

and it can be shown that $O(\hat{m}'_i, \hat{k}'_i) = 0$.

Since we obtain T estimates of m' and k' , one set for each subinterval, and since $V(\hat{\theta}_i) = V(\hat{\theta}_j)$, all i, j , and $\theta = m', k'$, it follows that the best linear estimates for the combined data are given by

$$\bar{m}' = \sum_{i=1}^T \hat{m}'_i/T, \quad \bar{k}' = \sum_{i=1}^T \hat{k}'_i/T,$$

with variances

$$\begin{aligned}V(\bar{m}') &= \beta\gamma(1+\gamma)/L^2T^2, \\ V(\bar{k}') &= 2\beta(\beta+LT)(1+\gamma)^2/\gamma^2L^3T^3.\end{aligned}$$

However, since we are interested in the distribution of faecal deposits at time t and not for a subinterval, the parameters of interest are $k = Tk' = \beta/L$, and $m = Tm' = \beta\gamma/L$. Estimates of these are given by $\bar{m} = T\bar{m}'$ and $\bar{k} = T\bar{k}'$ and the variances are

$$V(\bar{m}) = \beta\gamma(1+\gamma)/L^2,$$

$$V(\bar{k}) = 2\beta(\beta + LT)(1+\gamma)^2/\gamma^2 L^3 T.$$

As stated earlier, it is the concentration of larvae which is of interest and we consider the quantity $C = E(N)L/A$ (remembering that $E(N)$ is now referred to areas of size $a = A/L$) which is estimated by

$$\hat{C}_S = L\widehat{E}_S(N)/A = L\bar{m}A^{-1}\int_0^t \widehat{\mu(t,x)}dx, \quad (12)$$

where

$$\int_0^t \widehat{\mu(t,x)}dx = \widehat{\int_0^t \cdot}.$$

is an estimate of the required convolution integral. We now find the variance of $\hat{C}_S$ as a function of L and T and determine for which values of these two parameters it is minimized. Straight forward calculations show that

$$\begin{aligned} V(\hat{C}_S) &= (L^2/A^2) \left\{ V(\bar{m}) \left(\widehat{\int_0^t \cdot} \right)^2 + m^2 V \left(\widehat{\int_0^t \cdot} \right) \right\} \\ &= (\beta\gamma/A^2) \left\{ (1+\gamma) \left(\widehat{\int_0^t \cdot} \right)^2 + \beta\gamma V \left(\widehat{\int_0^t \cdot} \right) \right\}. \end{aligned} \quad (13)$$

It is shown below that for one method of estimating $\int_0^t \mu(t,x)dx$, $V \left(\widehat{\int_0^t \cdot} \right) = O(T^{-1})$

and therefore the conclusion is that, in this case, $V(\hat{C}_S)$ is independent of L and decreases with increasing A and T . However, the estimate of k increases in precision with an increase in both L and T , while the variance of $\bar{m}$ only depends on L .

It is, of course, possible to estimate the parameters k and m more efficiently by maximum likelihood methods (see Anscombe, loc. cit.). In this case an expansion for $V(\hat{C}_S)$ is easily obtained in terms of estimated variances of $V(\hat{k})$ and $V(\hat{m})$ of the maximum likelihood estimates. However, the additional rather heavy computational work necessitated by the maximum likelihood procedure does not really seem warranted.

There remains the question of the estimation of $\int_0^t \mu(t,x)dx$. There are numerous ways in which successive values of $\mu(t,x)$ can be estimated to provide ordinates for numerical integration. For ease of illustration, we consider just one direct

approach and we concern ourselves with the case $t \leq t_1$. The analysis for other situations would be analogous.

Assume now that $[0, t]$ has been subdivided, as described above, into T sub-intervals and let t_i be the upper boundary for the i th interval. Then during each interval i ($i = 1, 2, \dots, T$) F fresh faecal deposits are marked, and at time t the average number of larvae emanating from these deposits, in each of the T groups, is determined. If n_{ij} represents the number of larvae in the j th faecal deposit from the i th subinterval which survive to time t , then $\mu(t, t_i)$ is estimated by

$$\sum_{j=1}^F n_{ij}/F = \hat{\mu}(t, t_i).$$

Suppose now, that in order to estimate $\int_0^t \mu(t, x) dx$, we use the trapezoidal rule for numerical integration, then

$$\int_0^t \widehat{\mu(t, x)} dx = \frac{t}{2T} \sum_{i=1}^T \left\{ \hat{\mu}(t, t_i) + \hat{\mu}(t, t_{i-1}) \right\}$$

and, neglecting errors of integration,

$$\begin{aligned} V\left(\widehat{\int_0^t \cdot}\right) &= \frac{t^2}{4T^2F} \sum_{i=1}^T \left\{ \mu(t, t_i) + \mu(t, t_{i-1}) \right\} \\ &\simeq \frac{t}{2TF} \int_0^t \mu(t, x) dx. \end{aligned} \tag{14a}$$

For the case of $t > t_1$, it is the interval $[0, t_1]$ which is subdivided and measurements of larvae numbers are made at time t . Thus, for this case

$$V\left(\widehat{\int_0^{t_1} \cdot}\right) \simeq \frac{t_1}{2TF} \int_0^{t_1} \mu(t, x) dx. \tag{14b}$$

If, finally, it is desired to bring the discussion down to a sheep per unit area basis, then since $a = S\bar{a}$, for an average sheep the expected concentration is $C = C_s/S$. Obviously, $\hat{C} = \hat{C}_s/S$ and $V(\hat{C}) = V(\hat{C}_s)/S^2$.

No detailed discussion of estimation procedures for model (10) will be presented here. Obviously, the faecal component can be estimated as for (6a) and the remaining expression,

$$vu^{-1} \int_0^t k(x) f(t-x) dx, \tag{15}$$

approximated in various ways. For instance, $f(t)$ can be obtained by a separate investigation, while $vu^{-1} k(t)$ can be calculated by establishing the egg output of

individual sheep in the flock at different points on the time scale. If T different time intervals are used, then the $(T+1)$ parameters u and $k(t_i)$, $i = 1, 2, \dots, T$, can be estimated by maximum likelihood and the final expression obtained by numerical integration. Alternatively, the number of third-stage larvae, at time t , associated with faecal deposits dropped during previous time intervals can be obtained and the whole expression (15) approximated by numerical integration as for (6a). However, attention would have to be given to the variances of these estimates since they would not be of the same form as (14a) and (14b).

IV. DISCUSSION

The distribution on pasture of the infective larvae of the gastrointestinal nematode parasites of sheep has been considered by Crofton (1952, 1954). He sampled the most evenly grazed portion of three pastures and showed that the observed frequencies of infective larval numbers agreed fairly closely with theoretical frequencies calculated according to Neyman's Contagious Distribution Type A (Neyman 1939). It is intrinsic in this distribution that the clumps of organisms are Poisson-distributed. Since Crofton (1954) has shown that this is unlikely to be true for the distribution of faecal deposits over a field being grazed by sheep, he has pointed out that this limits the usefulness of the Neyman model to small areas of pasture only.

Donald (unpublished data) has fitted the negative binomial to the distribution of infective larval numbers recovered from 50 4-in. quadrat samples of pasture collected from a $\frac{1}{4}$ -acre field being grazed by five sheep, and has found $k \approx 0.2$. While this is consistent with a contagious distribution, several quite different hypothetical situations will give rise to a negative binomial distribution (Anscombe, loc. cit.) Thus, obvious difficulties of interpretation arise when attempts are made to compare the distribution of infective larvae of different species and to follow movements of the distributions with time.

The main purpose of this paper is to show how to construct models describing the distribution on pasture of the infective stages of parasites of grazing animals. Of the two models developed here, (10) is slightly more general since it incorporates component (4) of the Introduction. However, this increased generality introduces an extra parameter u , and the problems of estimation are increased. The simple properties of the Poisson distribution are lost and the rather natural interpretation of $E_S(N)$ is somewhat destroyed.

However, for most purposes (7) should provide a sufficiently accurate description of the distribution of the larvae on pasture. Once the faecal component has been estimated, α and p , different theoretical curves for $\lambda(t)$ and $f(t)$ can be used in (7) in order to investigate the effects such changes would have on infective larval populations on pasture. This would provide information, say, on the comparative behaviour of two different species of parasite or on the behaviour of a single species under different environmental conditions. Furthermore, the effect of each component of the model (faecal distribution, egg numbers per faecal deposit, and the mortality rate of the free-living larval stages) can be isolated and its ultimate influence on infective larval populations determined.

The introduction of a time element into the models seems advantageous. The influence of time on the total distribution of infective larvae is now clearly specified, and this enables theoretical questions, such as equilibrium values, to be settled. This was not possible in earlier studies when less specific models were fitted to estimates of infective larval populations on pasture.

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