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Abstract

Gastrointestinal parasitism is one of the diseases that has the highest economic impact on the Argentinian beef production system, making it inefficient. In the region of Pampa Húmeda, it has been estimated that annually 22 million dollars are lost because of the death of calves and 170 million dollars are lost in sub-clinics. A mathematical model with fuzzy parameters was made for the analysis of the free living stages of gastrointestinal parasites, whose objectives were to estimate the pool of L3 larvae available for migration to pasture and the levels of infection in pasture at any time of the year for different climatic conditions. The model is formulated in terms of a system of three difference equations. These equations describe the abundance of parasites in each of the successive stages of the population development. The model was calibrated and tested with climatic data and previous fieldwork carried out in Tandil *(*37° 19’ S, 59° 08’ 05’’ W*)*, Buenos Aires province (Argentine). The comparison with other fieldwork data achieved satisfactory results.

Keywords: Mathematical model; Parasitic gastroenteritis; Cattle-Nematoda.

Introduction

Gastrointestinal parasitism is one of the diseases that causes major economic impact on the beef production system in Argentina. It causes great weight loss, which can exceed 30kg per cow per year, and high mortality of calves (Entrocasso, P., 1981; Steffan, P. E., et al., 1982). From an economic perspective, in 1998 estimated costs added up to some 22 million dollar annual loss due to cattle mortality, and 170 million dollars owing to subclinic problems. Therefore, the production system becomes inefficient.

In Argentina, cattle feed on pasture during the whole year. Nematode parasites, in particular *Ostertagia spp*. and *Cooperia spp.Haemonchus spp.*, are predominant in the Pampa region (Fiel et al.,1994) Since they live in pastures for a given period of their life cycles, they can be easily ingested by cattle and thus reinfect them.

Since the 1960s, parasites' life cycles have been studied in a variety of environments. The results have helped understand how processes occur under different environmental conditions. A large number of authors obtained valuable results on the compound effects of environmental variables such as temperature (Lutzelschwab, C., et al., 2005; Michel, J. F., et al., 1975), humidity, and rainfall (Onyiah, L., et al., 2005; Stromberg, B. E., 1997), and microclimate of dung (Krecek, R. C., et al., 1990; Rossanigo, C., 1995), as well as biological factors such as hatching times (Young, R. R., et al., 1980a), length of hatching and length of infective larvae (Gibson, M., 1981; Pandey, V. S., 1972; Rose, J. H., 1969), time of development from egg to infective larva, mortality rates of pre-infective and infective larvae (Pandey, V. S., 1972), and conditions for migration and survival in pasture (Al Saqur, et al., 1982; Almería, S.,et al., 1999; Durie, P., 1961; Langrová, I., et al., 2003; Persson, L., 1974a; Persson, L., 1974b; Rose, J. H., 1970; Stromberg, B. E., 1997). It has become clear that the epidemiology of these parasite infections changes according to the climate zone. Experts believe that a minimum of five years of fieldwork is necessary in order to obtain a clear understanding of the epidemiology of any cattle area in Argentina.

There are numerous investigations on different epidemiological aspects in almost all cattle areas around the world (Steffan, P. E., et al., 1986; Williams, J. C., et al., 1987). This has made possible to obtain very interesting and important statistical results. Lately, cases of resistance to several antihelmintics drugs have been recorded so that the strategies that vets normally use are not always efficient whenever considering cost and benefits. Therefore, it is necessary to generate new strategies for the control of parasite populations in order to obtain a maximum benefit in productivity with a minimal drug use.

The variability of responses to different environmental factors make relevant the use of modelling tools to help understand the complexity of the dynamics of parasite life cycles and to find the appropriate control mechanisms. Some parasite models, (Louie, K., et al., 2007; Learmount, J. et al., 2006, Ward, C. J.; 2006a, Ward, C. J., 2006b; Smith, G., et al., 1987; Smith, G., et al, 1986; Young, R. R., et al., 1980b), have given very useful insights into the population dynamics of gastrointestinal parasites, becoming interesting complements to field and laboratory research. However, some aspects of the pre-infective cycle have not been taken into account. For example, the effect of rainfall on larval pre-infective mortality rate has not been considered so far. Recent models proposed by Ward

(Ward, C. J., 2006a and 2006b) and Louie (Louie, K., et al., 2007), based on the theory of dynamic systems, are being used as control tools. Nevertheless, it is worth mentioning that the hypotheses for those models are not totally adequate for Argentina, since climatic conditions and the way cattle is handled are not the same as in other countries.

Here we propose a mathematical model for the free-living stages of gastrointestinal parasites adapted to the environmental conditions in the Pampa region of Argentina, where cattle raising is extensive.

Mathematical modeling with fuzzy logic: an introduction

"*A wealth of empiric knowledge is very difficult to translate into mathematical functional relationships.*"

Mathematical modelling using fuzzy logic was introduced by L. A. Zadeh in 1965 (Zadeh, L. A., 1965). The modelling of non quantifiable concepts by means of fuzzy sets yields the possibility of giving mathematical meaning to natural language statements. Human thinking and behaviour are strictly connected with imprecision and uncertainty. The traditional Boolean algebra, characterized by categorical values of truth and falsehood, is not appropriate to cope with such problems.

If we have a proposition such as *"The rain is pouring down"*, then it is not always possible to assert whether it is true or false. When we learn that rainfall is *40* mm/hr, then the truth or, more appropriately, the compatibility of '40' with 'downpour' is a matter of definition. It depends on our understanding of the concept "*downpour"*. If the preposition is *"Rainfall is above 40 mm/hr"* and we know how much it rains, then we can give a ‘yes’ or ‘no’ answer to whether the proposition is true or false.

Zadeh gave the notion of a fuzzy subset. Clearly, both “*40 mm/hr*” and “*60 mm/hr”* are a lot, but there are some diferences: *60 mm/hr* is much more than *40 mm/hr*, and their effects could be very different. This suggests that membership in a fuzzy subset should not be on a 0 or 1 basis, but rather on a 0 to 1 scale. That is, the membership should be an element of the interval *[0, 1].*

In very formal terms:

A fuzzy set *A* defined in an *X* universe is a set of pairs $(x, \mu_A(x))$ where *x* belongs to *X* and $\mu_A(x)$ is a number in the interval *[0, 1]* representing the degree of membership of *x* in *A*.

Clearly any given point of *X* can be member of different fuzzy sets with different degrees of membership.

Two notations for fuzzy sets can be found. A fuzzy set can be defined as sets of pairs as above. In this case, *A* is called *the linguistic label*. Otherwise, it can be defined by the specific function $\mu_A(x): X \rightarrow [0, 1]$ representing this notion and would be denoted by μ_A. The notation *A* stands for the concept of “downpour” and μ_A gives the degree of *rainfall intensity* that has been assigned to each member of *X*.

Usually, the degree of membership is assigned through the application of a certain number of rules. Fuzzy rule-based systems (FRBS) have four components: an input processor, a collection of fuzzy rules called rule base, a fuzzy inference machine, and an output processor. These components process real-valued input to provide real-valued output as follows:

- Input processor (Fuzzification): Here, non quantifiable input is translated into fuzzy sets of their respective universes.
- Rule base: This is a key knowledge-encoding component of fuzzy rule-based systems. Essentially, fuzzy rules are fuzzy relations of the Cartesian product of the universes of the variables of interest. The base is composed by a collection of fuzzy conditional propositions in the form of *IF-THEN* rules. Usually, a fuzzy rule has the form

  IF x is A then y is B
- Fuzzy inference: The fuzzy inference machine performs approximate reasoning using the compositional rule of inference. In this work, the Mamdani method is used (Barros L. C., 2006).
- Defuzzification: In fuzzy rule-based systems, the output is usually a fuzzy set. Often, especially in system modelling, a real number is required as output. The output processor provides real-valued output through defuzzification, a process used to choose a real number that is representative of the corresponding fuzzy set.

The model

The life-cycle of nematodes is direct, without an intermediary host. There are two stages, one within the host or parasitic stage, and another in the environment or free-living stage. During the free-living period larvae grow successively from eggs through *L1, L2*, and *L3* larval stages.

The aims of this work are to estimate the pool of *L3* larvae available for migration to pasture and the levels of infection in pasture at any time of the year.

In order to carry out these objectives, a discrete population model with fuzzy parameters was built. This model describes the development of these gastrointestinal parasites through the free-living stages under the effect of environmental factors. It is formulated in terms of a system of three difference equations. These equations describe the abundance of parasites in each of the successive stages of the population development. The processes taken into account are (Figure 1):

1) Recruitment of larvae to the *L3* population pool available for migration to pasture. (Module 1)
2) Survival of the *L3* larval population in dung waiting for appropriate conditions for migration to pasture. (Module 2)
3) Emigration of *L3* larvae from dung to pastures. (Module 3)

The difference equations are solved with daily step. The fuzzy parameters were built based on data and information from publications related to the subject and the expertise provided by parasitologists[1].

Computation of development time from egg to *L3* larva

There is a direct relationship between development time of larvae and temperature over the range from 4°C to 40 °C.

Given that *L(a,t)* is the length of an individual larva at age *a* which was born on day *t*, then the development can be described by the differential equation:

$$\frac{\partial L(a,t)}{\partial a} = r(t)\,L(a,t) \qquad (1)$$

together with the initial condition

$L(0, t)=l_0(t)$ [μm]

where:

$r(t)=AT(t)+B$ is the function that regulates the development rate depending on the temperature $T(t)$;

$l_0(t)$= length of a larva at hatching on julian day t [μm]

When a larva reaches a length of 850μm, it is considered to be in L3 stage.

A and *B* were obtained by fitting laboratory data obtained by Pandey (Pandey, V. S., 1972).

From these equations it is possible to estimate the minimum time of development from egg to *L3* stage which is noted $\tau(t)$. When day *t* is fixed, the partial differential equation is changed into an ordinary differential equation that can be easily solved. The development time is the value of age a_t at which $L(a_t, t) \cong 850$μm and hence $\tau(t)=a_t$.

Recruitment of larvae to the *L3* population pool available for migration to pasture (Module 1).

This module describes the dynamics of eggs and larvae within dung. All the eggs contained in the dung which are hatched in one day by the herd are considered a cohort corresponding to that day of *"arrival"* as initial time. Each of the cohorts

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[1]The Parasitology Group of the Faculty of Veterinary Sciences of the Universidad Nacional del Centro de la

makes one and only one contribution, which is then considered as the initial condition for that cohort. It is worth noting that the development time of different cohorts may vary, because the environmental temperature affects development (Fiel, C. A. et al, 1994; Pandey, V. S., 1972). As explained above, the function $\tau(t)$ defines the time it takes for a cohort initiated on julian day t to reach the *L3* stage.

Precipitation produces high mortality among eggs and pre-infective larvae (*L1* and *L2*) due to "dung washing". Rainfall removes eggs and pre-infective larvae from the dung and takes them to the pasture where the conditions are not appropriate for continuing their development. For this reason, the only process that affects the cohort is mortality, and hence the cohort dynamics is described by the following equation:

$$H(t+a,\ a)=(1-\square_P)\ H((t+a-1,a-1) \quad \text{if } a\leq\tau(t)$$

together with the initial condition

$H(t,0)$= amount of eggs contributed to pasture on julian day t.

where:

$H(t+a,\ a)$ = amount of preinfective larvae (eggs, *L1* and *L2*) aged a, which "started" on julian day t.

$\square_P$ = rate of preinfective mortality;

$\tau(t)$= minimum development time for cohort initiated on julian day t.

Survival of the *L3* larvae population in dung awaiting to appropriate conditions for migration to pasture. (Module 2)

This module describes the population of L3 larvae in dung ready to migrate to

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Provincia de Buenos Aires. This research group has more than 20 years of expertise in the field.

pasture. The population pool grows as larvae from different cohorts complete their development and reach stage L3. Losses from the pool are due to mortality while waiting for the environmental conditions that allow migration, and to mortality during migration.

When rainfall occurs, migration takes place. The proportion of larvae in the pool that migrate to pasture on a given day depends on the time of the year and the amount of precipitation recorded on that day.

The dynamics of this population is described by the following difference equation:

$$L3D(t+1)=(1-\delta_{DP})(1-\mu_{ID})L3D(t)+L3ND(t+1)$$

where:

L3D(t) = amount of L3 larvae in the dung pool on day *t*

L3ND(t) = amount of L3 larvae in dung that have completed their development precisely on julian day *t*

μ_{ID} = rate of mortality inside dung;

δ_{DP} = rate of migration from dung to pasture.

Emigration of *L3* larvae from dung to pastures (Module 3).

The dynamics of the population of *L3* larvae in the pasture is described in this third module. *L3* larvae that survive and migrate due to rainfall are considered as a contribution to the larval population in the pasture. Losses are due to mortality in the pasture, which depends on the time of the year and the temperature:

$$L3P(t) = (1-\mu_{P})\ L3P(t-1)+ \delta_{DP}\,(1-\mu_{ID})\ L3ND(t)$$

where:

L3P(t) = amount of larvae in pasture on day *t*

$\square_{P}$ = rate of mortality in pasture;

$\square_{ID}$ = rate of mortality inside dung;

δ_{DP} = rate of migration from dung to pasture.

Computer simulations

The model was implemented using GNU Scilab 5.0. The inputs of the model are:

1) Weather data: the model includes data on temperature and precipitation.
   a) Temperature is measured in Celsius degrees (ºC). Maximum ($Tmax_n$) and minimum ($Tmin_n$) annual average temperature for year *"n"* (1994-1998) are taken for constructing a temperature function . The temperature on julian day *t* of year n is represented by:

$$T(t) = \frac{T\,max_n + T\,min_n}{2} + \frac{T\,max_n - T\,min_n}{2} cos\left(\frac{2\pi(t-45)}{365}\right)$$

   b) Precipitation is measured in *[mm]* and is given daily.
2) Initial and final day [day, month, year] of the period in which the animals are in the pen "scattering" eggs over the pasture. These days are called “the period of study”.
3) Egg per grams of dung *(EPG)* in the period of study.
4) Average weight of the animals in the pen.

The program runs the simulation with these data and creates two graphs which are useful to the vets or managers. One of the graphs describes the dynamics of L3 larval population available for migrating to pasture *(module 1)* and the precipitation recorded over the period of the simulation. The second graph describes the infection of pastures on a daily basis *(module 3)*.

Fuzzy parameters

Pre-infective mortality rate

As mentioned above, each cohort goes through three stages, from egg to *L1*, to *L2*, to *L3* within dung. Conditions in dung are sufficient for their development (Fiel, C. A. et al, 1994). The air temperature strictly determines the length of this period. The eggs never hatch if the temperature is below 4^{o}C or above 40^{o}C. The hatching increases as temperature rises from 10 ^{o} to 35 ^{o}C (Pandey, V. S., 1972; Williams, J. C., et al., 1971 and 1987). Mortality increases when temperature reaches values outside the above-mentioned range (Levine, N. D., 1978). There is an increase in the development time when the temperature decreases. Thus, during warm months only a few days will be necessary to reach the *L3* stage, whereas several weeks are necessary in cold months, specially during wet and cold winters (Catto, J. B., 1982; Durie, P., 1961).

Consequently, the linguistic variables Age, Season, and Rainfall (input) and Mortality (output) were selected. The membership functions and their parameters are detailed in Table 1.

In order to build the rule-based system of fuzzy parameters, the findings of researches carried out by the Parasitology Group as well as bibliographic information were taken into account. This rule-base system appropriately reflects several situations that the parasites face in this period. For example:

*IF(Age is **not L3**) and (Season is **Summer**) AND (Rainfall is **Drizzle2**) THEN (Mortality is **Moderate**)*

*IF(Age is **L3**) and (Season is **Winter**) AND (Rainfall is **Drizzle2**) THEN (Mortality is*

Moderate)

The complete list of rules is summarized in Table 2.

Migration from dung to pasture

Larvae need one film of water to be able to migrate out of dung. Thus, this process depends on rainfall. In summer, a monthly precipitation of more than 50 mm is sufficient for *Trichostrongylus* and *Ostertagia* to be able to migrate (Fiel, C. A. et al, 1994; Gordon, H., 1973; Toledo, H. O., 1980) whereas in winter 10 *mm* are enough.

Therefore the linguistic variables *Season* and *Rainfall* (input) and *Migration* (output) were selected. The membership functions and their parameters are detailed in Table 3. The rule-based system of fuzzy parameters was built similarly to that of the pre-infective mortality parameters. The complete list of rules is summarized in Table 4.

Mortality in pasture

The survival of an L3 larva in the pasture depends on the energy accumulated in its intestinal cells and its ability to be eaten by bovine once in the grass. Temperature and humidity are the most important factors for the life cycle of larvae (Levine, N. D., 1978). Larval activity is lesser in winter than in summer. In winter, its energy is slowly exhausted, while in summer the reserves are quickly depleted. Thus the infection in pastures is lower in summer than in winter (Steffan, P. E., et al., 1986). Low temperatures, high rainfall and a good coating fodder are associated with high periods of survival (Besier, R., et al., 1993).

The linguistic variables selected are *Season* and *Temperature* as input and

Migration as output. The membership functions and their parameters are detailed in Table 5. The rule-based system of fuzzy parameters was built similarly to that of the pre-infective mortality parameters. The complete set of rules is summarized in Table 6.

Study Case

Characteristics of the region

Tandil is located in the Southeast region of the province of Buenos Aires (37°19′08″S 59°08′05″W), exactly in the hilly system of Tandilia which runs in the direction Northwest-Southwest, in an elevated portion of the Humid Pampas. The climate in the region is mild and humid, with an annual average temperature of 13.7°C. Average seasonal temperature is 16.63°C in spring, 19.63°C in summer, 10.36°C in autumn, and 8.43°C in winter. Rainfall exhibits a regular distribution with an annual precipitation of 888.6 mm. It is very homogenous in spring (*230mm*), summer (*256mm*), and autumn (*248mm*), while in winter (*143mm*) the weather is somewhat drier (Source: National Meteorological Service 1911-1991).

Field Trial

The data used in the construction of this model were provided by field experiments that were carried out from late 1994 until the end of 1997. A 0.96 hectare paddock located on the University Campus (UNCPBA) in Tandil was used for the field work. The paddock was divided into 16 sub-paddocks. Two naturally infected calves contaminated the sub-paddocks with eggs of gastrointestinal parasites. Faecal samples for egg counts and coprocultures were taken weekly from the "contaminating" calves during the grazing period. Faecal egg counts were used to plot the contamination of the paddock. Coprocultures allowed the identification of which nematode species were present in the contamination.

On the 15th day of each month, faecal matter was collected from the paddock. Then weekly samples were taken in the lab from collected faecal matter in order to analyse the development from egg to *L3*. Simultaneously, grass samples were regularly taken from the paddock over the 16-month period to assess the infection of pastures as well as the survival of *L3* larvae in pasture. (Fiel, C.A., et al., 2008)

Initial conditions for computer simulations

Simulations were run taking as initial conditions two calves weighing 250 Kg and which had been grazing for 6 months in the same plot.

For the Tandil region, the values of *A* and *B* for equation 1 were adjusted to data to be: A=0.0509 and B=0.0922 and the length l_0 set to 350μm. Therefore this equation can be written as:

$$\frac{\partial L(a,t)}{\partial a} = \left(0.0509T(t) - 0.0922\right)L(a,t)$$
$$IC\ L(0,t) = 350\ \mathrm{m}\mu$$

For the summer-autumn simulations, the animals entered the paddock on January *1st* and were moved out on June 30th. For the winter-spring simulations they entered on July *1st* and left on December *31st*. While in the paddock, the animals were allowed to contaminate the pasture with eggs. Model populations of L3 larvae available for migration and in pasture were monitored for 550 days. The day of the year in which the animals enter the plot is recorded as *Day 1*. Simulations were run using real precipitation data as input. Precipitation data for the period 1994-1998, were provided by the National Meteorological Service. The temperatures in the simulations are detailed in Table 7. These conditions represent the average climatic conditions in the Tandil area in the period of study. Eggs per gram *(EPG)* of faecal matter were taken from data provided by Fiel and collaborators. With this input, the model scenario presents the same conditions as the experimental work carried out by Fiel and collaborators.

Numerical results

Development Time

The comparison between experimental field data and the output of this model is displayed in Figure 1.

From field trial, Fiel estimated development times from egg to *L3* within the ranges of 1-2 weeks in summer, 3-5 weeks in autumn, 4-6 weeks in winter and 1-4 weeks in spring, depending on meteorological conditions.

Computer simulations calculated development time in different months. The development times (in days) estimated by the model were compared with data from the field. As Table 7 shows, satisfactory results were obtained, as the ranges in the field and from simulations, are very similar.

Recruitment of L3 larvae to population pool available for migrating to pasture.

There are no field data about densities of larvae *L3* available for migration to pasture. The simulated results are presented in Figure 2.

As can be expected, computer simulation results differ depending on the time of the year in which the first day is set.

Results of computer simulations

Period January-June (summer-autumn)

The first *L3* larvae available for migration to pasture appeared from 5 to 10 days after the animals had entered the plot. This quick availability is not surprising because in summer the development time reaches its minimum. Dung plays a very important role, since it is a shelter for parasites. It ensures survival and availability of parasites for long periods of time. Period length depends on the season and the level of rainfall at the time of study. In the computer simulation, these periods lasted from 230 to 300 days, depending on particular conditions that vary from year to year.

Period July - December (winter-autumn)

The larvae appeared in the pasture between 35 and 40 days after the introduction of the animals in the paddock. When the first eggs were laid in the pasture the temperature was minimum. Consequently, the development times are longer as compared to the summer-autumn period. The dung "shelters" were emptied after 380 / 460 days.

Presence of L3 in pasture

Fiel reports that the pasture infectivity showed a clear seasonality during autumn-winter and spring field experiments, while a high mortality of larvae in pasture occurred in summer. *Ostertagia* and *Cooperia* were the predominant genera, with a general survival rate of more than one year.

Here again computer simulation results differ depending on the time of the year in which the first day is set.

Results of computer simulations

Period January-June (summer-autumn)

The first *L3* larvae in the pasture were recorded in moderate levels in summer, but survived only few days there. The peak of infection was recorded during the first fortnight of winter. The *L3* larvae survived in pasture until mid-spring.

Period July-December (winter-spring)

The first *L3* larvae in the pasture appeared 70 days after the animals entered the paddock. The maximum abundance of larvae took place in mid-spring, sustaining significant levels until mid-summer. Then, it decreased steadily until disappearing by the end of summer as can be seen in Figure 2. Model outputs appropriately reflect the long periods of infection detected during field trials.

Conclusions

Needless to say, epidemiological dynamics is very important when attempting an effective and efficient control of the disease that causes large economic losses. Accomplishing the characterization of the dynamics requires a lot of effort. A minimum of four years of data collections is required to develop a complete study of the disease. The relevant indicators needed to characterize the disease are:

1. The egg-to-*L3* development time inside the fecal paths. The knowledge of these development times permits to determine the minimum delays for the pasture to become infected.

2. The pasture infectivity levels over time. The determination of infectivity levels is a very useful tool in the diagnosis of the disease because it allows assessing the exposure risk of animals in the paddock. It also allows inferring infectivity patterns

and relating them to climate variations and cattle management (Fiel, C. A. et al, 1994).

3. The survival of L3 in pasture. The fact that the survival of L3 depends on climate permits to estimate how long the delay will be before cattle can be reintroduced in the paddock with a minimal risk of infection.

The construction of a model that can be used as a tool to understand better the dynamics of the infections, as well as to contribute to efficient cattle management, requires consistent quantitative and qualitative data. This model is built based on the data on egg-to-L3 development time, pasture infectivity levels over time, and L3 survival in pasture provided by the Parasitology Group. These findings have been fundamental for setting the linguistic variables and the inference rules for the different fuzzy parameters that are being used in the model. Consistent data sets have been used for calibration and corroboration of the model.

The model based on fuzzy logic produces very satisfactory results. The simulations adequately mimic the dynamics of the infection of pastures. This accurate representation obtained from the model is reflected in the similarity of the output with field experiment data in several key aspects:

- Estimation of egg-to-L3 development time under different climatic conditions and in different seasons.
- Estimation of time of the first L3 larvae appearing in pasture under different climatic conditions and in different seasons.
- Estimation of the Julian day in which the peak infection is expected.
- Estimation of the duration of the infection.

This is necessary information for a diagnosis of the disease.

Methods for counting eggs in dung seem to be quite reliable. However, the estimation of pasture infection is influenced by various factors that can lead to poor assessments. Among them, we can list weather, type of pasture, the time of the year in which samples are taken, the time of day (Fiel, C. A. et al, 1994), as well as the laboratory techniques used in processing the samples. It is worth noting that on warm summer days, larvae take refuge in very low, shaded, and hard to reach places. Not only can these factors cause variability in the estimates of L3 larval densities in pastures, but also the way in which the grass is collected or the larvae are recovered may reduce the counts to any figure between 20 to 60% of the larvae actually present (Ferreyra, D. A., et al., 2003). We believe that the difficulty in obtaining accurate density estimations of larvae actually present in pasture may be the cause of the difference in the density values obtained in the simulations as compared to those estimated from field experiments, which are always lower.

Regarding L3 larvae in dung ready to migrate to pasture, it is not yet possible to evaluate the results obtained from the simulations because no data on this matter has been recorded during field experiments. It would be interesting to have this information from future field assessments in order to fully corroborate the model and indirectly evaluate the reliability of the pasture infection estimates. However, model estimates of L3 larvae in dung seen reasonable and can be indirectly evaluated as reliable given that they are source to the infection levels.

The results of field experiments show that the dynamics of L3 larvae in pasture varies from one year to the other due to the strong dependence of the life cycle of the parasite on the weather. The model exhibits the same sensitivity to temperature and precipitation in the simulations as can be seen in Figure 2 and in Tables 7 to 10 that summarise the results.

One important feature of this model is its simplicity. It is expressed in terms of three difference equations and their fuzzy parameters. The simplicity is a consequence of having chosen fuzzy logic for the definition of the parameters. This permitted to handle a large number of variables and information that is not straightforwardly quantifiable and that could not have been possible to include if using classic mathematical tools. The inclusion of knowledge gained through experience along many years of research is a very valuable aspect of this modelling approach.

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Table captions:

Table: 1 Linguistic variables for pre-infective mortality rate (Input).

Table 2: The base of rule for fuzzy parameter pre-infective mortality (Input).

Table 3: Linguistic variables for migration rate (Input).

Table 4: The base of rule for fuzzy parameter migration (Input).

Table 5: Linguistic variables for rate of mortality in pasture (Input).

Table 6: The rule-based system of fuzzy parameter mortality in pasture (Input).

Table 7: Development time estimated by model and from field trials for Tandil (S=summer, W=winter, A=autumn, Sp=spring).

Table 8: Beginning of infection estimated by model and from field trials for Tandil's pastures.

Table 9: Peak of infection estimated by model and from field trials for Tandil's pastures.

Table 10: End of infection estimated by model and from field trials for Tandil's pastures.

Figure captions:

Figure 1: Diagram of model.

Figure 2: Result of simulations as compared to field data. Note that magnitudes do not necesarily correspond.

Table 1

	Age ([%])	Season([d])	Rainfall([mm])	Mortality([%])
1	L1 (0-0.6)	Summer (-45-136)	Drizzle1 (0-2)	Little(0-0.35)
2	L2 (0.3-0.9)	Winter (136-319)	Drizzle2 (2-10)	Moderate (0.2-0.6)
3	L3 (0.7-1)	Spring (228-365)	Rain (5-20)	Great (0.5-1)
4		Autumn (45-228)	Downpour1 (10-50)	
5			Downpour2 (50-600)	

Table 2

IF (Age is L3)

Season/Rainfall	1	2	3	4	5
1	1	1	2*0.5	2	2
2	1	2	2	3*0.5	2
3	1	1	1	1	1
4	1	1	1	1	1

IF (Age is not L3)

Season/Rainfall	1	2	3	4	5
1	2	2	2	3*0:8	3
2	1*0:1	1*0:5	1	2*0:5	2
3	1*0.5	1	2*0.5	2	3*0:5
4	1*0:5	1	2*0:5	2	3*0:5

Table 3

	Season ([days])	Rainfall ([mm])	Migration ([%])
1	Summer (-45-136)	Drizzle 1 (0-2)	Little (0-0.1)
2	Winter (136-319)	Drizzle 2 (2-10)	Moderate (0.05-0.5)
3	Spring (228-365)	Rain (5-20)	Great (0.35-0.6)
4	Autumn (45--228)	Downpour 1 (10-50)	
5		Downpour 2 (50-600)	

Table 4

Season/Rainfall	1	2	3	4	5
1	1*0.1	1*0.5	1	2	3
2	1	2	3*0.7	3*0.9	3
3	1*0.5	1	2	3*0.7	3
4	1*0.5	1	2	3*0.7	3

Table 5

	Season ([days])	Temperature ([^{o}C])	Mortality ([%])
1	Summer (-45-136)	Low (0-14)	Little ()
2	Winter (136-319)	Moderate (12-26)	Moderate ()
3	Spring (228-365)	High (23-35)	Great ()
4	Autumn (45—228)		

Table 6

Season/Temperature	1	2	3
1	1	2	3
2	1	2*0.5	3*0.5
3	1	1	2*0.5
4	1	2*0.5	3*0.5

Table 7

Semester : (Year)	**Temperature ([^{o}C])**		**Precipitation ([mm])**	**Development time ([days])**	
	Max ºC	Min ºC		Field	Simulation
1:(1995)	21.15	7.75	324.2	S=14; A=28-42	S=8-11; A=11-29
2:(1995)	19.7	4.78	279.6	W=28-42; Sp=7-14	W=31-45; Sp=13-30
1:(1996)	22.56	8.32	553.9	S=7-14; A=21-42	S=6-9; A=9-27
2:(1996)	19.5	5.4	484.1	W=21-42; Sp=14	W=30-42; Sp=13-29
1:(1997)	21.86	9.32	464.9	S=7-14; A=21-35	S=7-10; A=10-25
2:(1997)	18.18	6.22	516	W=21-35; Sp=14	W=29-39; Sp=14-29

Table 8

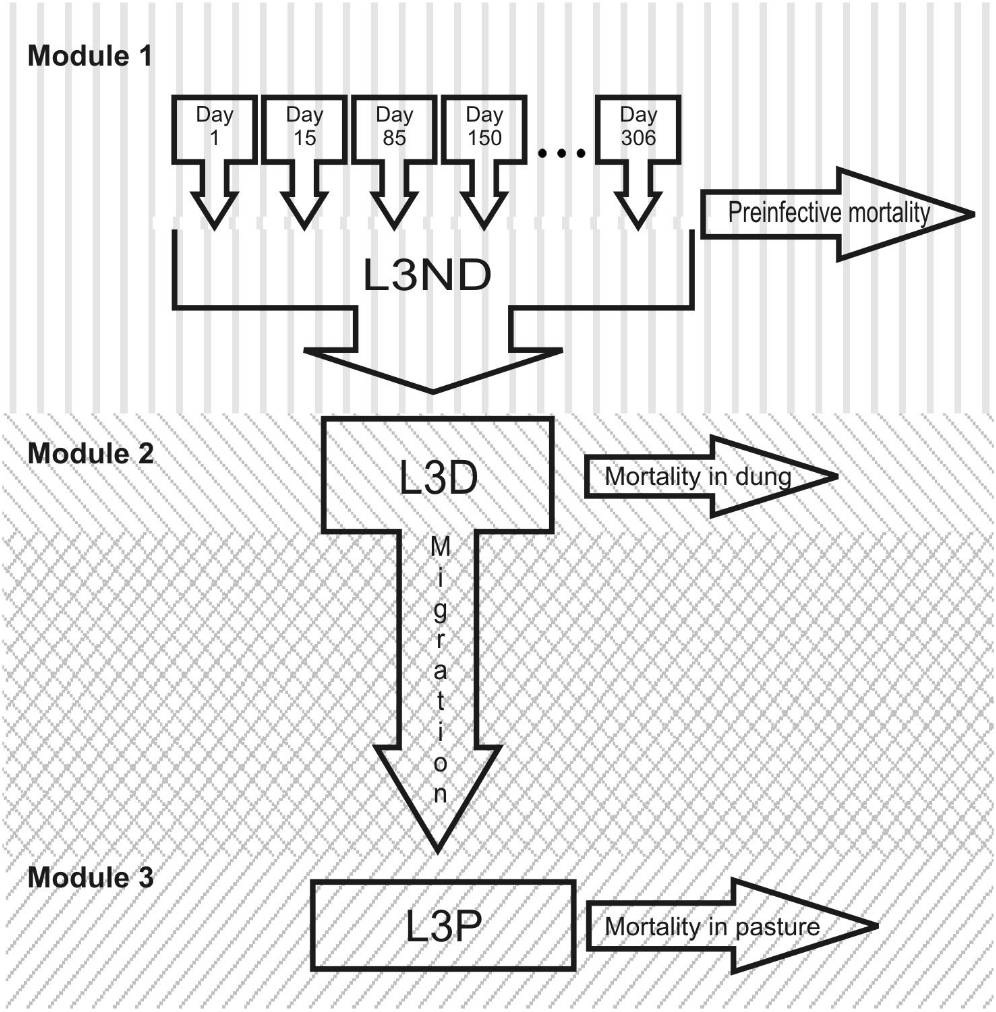
Semester: (Year)	**Beginning of infection**	
	Field	Simulation
1:(1995)	April	January
2:(1995)	October	October
1:(1996)	April	January
2:(1996)	October	October
1:(1997)	March	January
2:(1997)	October	October

Table 9

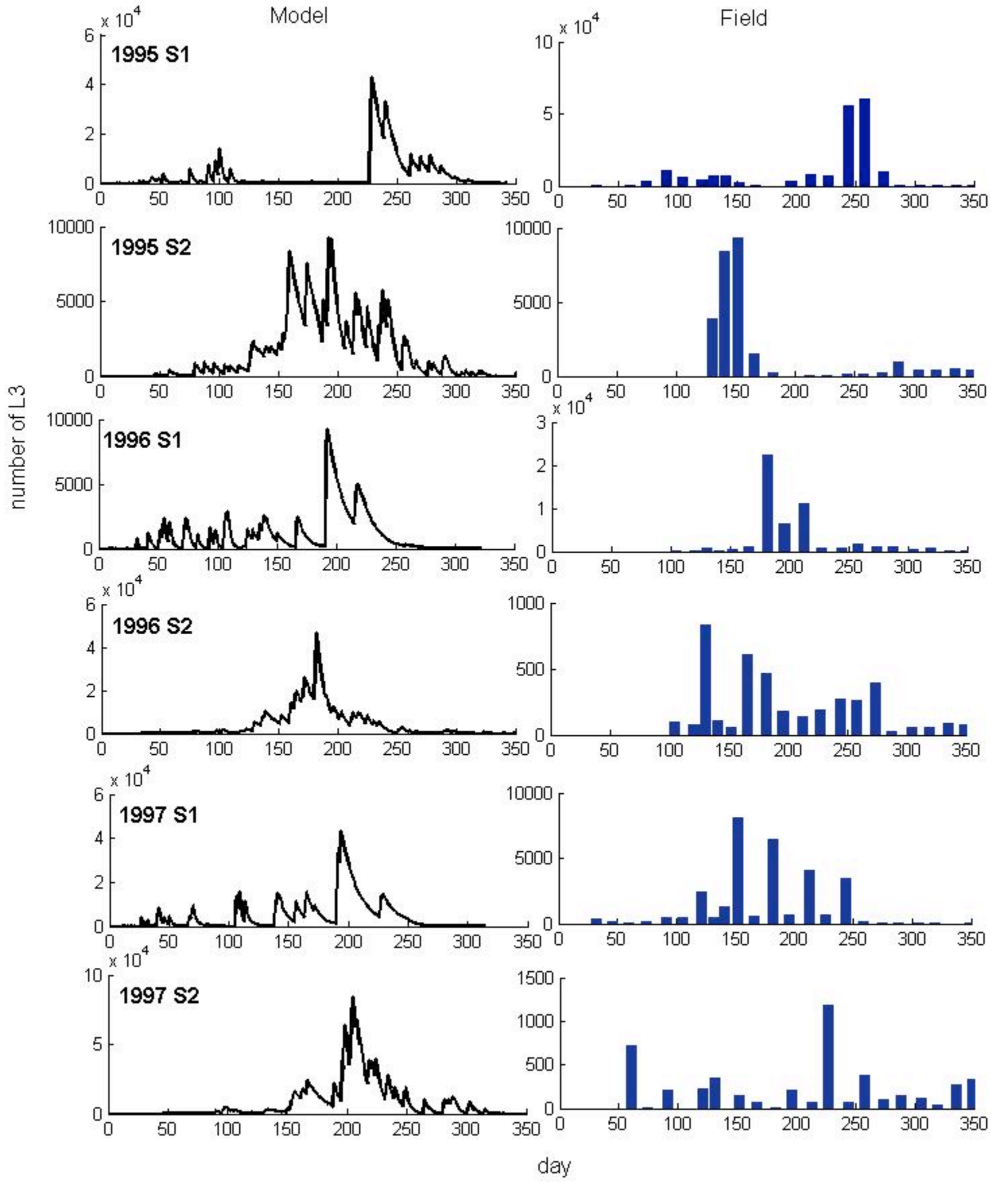
Semester: (Year)	**Peak of infection**	
	Field	Simulation
1:(1995)	November 1995	September 1995
2:(1995)	November-December 1995	November-December 1995
1:(1996)	July 1996	July 1996
2:(1996)	August 1997	November 1996
1: (1997)	July 1997	July 1997
2: (1997)	April 1998	December 1997-January 1998

Table 10

Semester: (Year)	End of infection	
	Field	Simulation
1:(1995)	End of December 1995	End of December 1995
2:(1995)	End of June 1996	End of June 1996
1:(1996)	End of November 1996	End of October 1996
2:(1996)	End of June 1997	End of June 1997
1:(1997)	End of June 1997	End of September 1997
2:(1997)	End of October 1998	End of October 1998



Module 1
Day 1
Day 15
Day 85
Day 150
Day 306
Preinfective mortality
L3ND
Module 2
L3D
Mortality in dung
Migration
Module 3
L3P
Mortality in pasture


Model
Field
1995 S1
1995 S2
1996 S1
1996 S2
1997 S1
1997 S2
number of L3
day