

Natural infection of *Lymnaea truncatula* by the liver fluke *Fasciola hepatica* in the Porma Basin, León, NW Spain

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ABSTRACT

For two years fortnightly malacological samplings were carried out to collect *Lymnaea truncatula* (Mollusca; Basommatophora) at five points in the upper and middle Porma river basin, León, Spain. The highest numbers of snails were collected in September, May and November. Of the 5486 molluscs examined, 11.41% harboured *F. hepatica* (Trematoda; Digenea) with an average intensity of 20.14. In general, the values of both infection prevalence and intensity increased with the size of the snails. It was in October when the highest figures for each parameter mentioned above were detected (18.73% and 28.48, respectively). The chi-square test showed statistically significant differences in relation to the infection prevalence among the groups of molluscs established according to: their length; the months in which they were collected; the sampling localities; monthly average ambient temperature; precipitation during the collection. Similarly, statistically significant differences were detected in the intensity of the infection among the groups of molluscs previously established, except for that based on the values of precipitation. Generally, parasites were found in the same snail at different stages of development. It seems that most mollusc infections occur in February-March and at the end of summer-beginning of autumn periods. The highest rate of rediae with mature cercariae ready to be shed were detected between September and December. Metacercariae in the grass samples were also observed at the end of autumn. For this reason, this period could be considered as the most suitable for infection of the definitive hosts to take place.

KEY WORDS: *Fasciola hepatica*, *Lymnaea truncatula*, Spain, prevalence

INTRODUCTION

Fascioliasis is a serious problem because of the large losses it causes among sheep and cattle, as well as its zoonotic repercussions.

According to OVER (1967) the life cycle characteristics of *Fasciola hepatica* Linnaeus, 1758 and the epidemiological nature of the disease it produces is greatly influenced by local conditions e.g., stockbreeding model, animal handling, presence and ethology of the intermediate host molluscs, pasture type and meteorological factors, amongst others. Thus, the conclusions reached after research in other countries (KENDALL, 1960; OLLERENSHAW, 1967; RONDELAUD, 1974; LEIMBACHER, 1981; SMITH, 1981, 1984; OVER, 1982; WILSON *et al.*, 1982) cannot be applied to Spain.

In Spain, although there is information available about the incidence of *F. hepatica* in the definitive hosts, field studies related to the ecological aspects of parasite transmission are scarce. Very little research on the life cycle of this trematode in the Pyrenees (PERAITA, 1976a, 1976b) and Salamanca (SIMON, 1968; SIMON & GARMENDIA, 1974) has been performed.

In view of this, we felt it appropriate to carry out an in-depth study of the *F. hepatica* cycle in our area, concentrating our research on the Porma basin (León province). The results obtained on the dynamics of parasite egg elimination by cattle and sheep in this area were published recently (GONZALEZ-LANZA *et al.*, 1989

and MANGA *et al.*, 1990, respectively). This paper records the data obtained from monthly tracking of: the *Lymnaea truncatula* (MULLER, 1774) populations, the prevalence and intensity of its infection by *F. hepatica* and the development of the larval stages found in the molluscs.

MATERIALS AND METHODS

Between March 1985 and March 1987, fortnightly malacological samplings were made at 5 places in the Porma basin (NW Spain) to collect the *L. truncatula* species, intermediate host of *F. hepatica*. Likewise, at the same intervals, herbage samples were collected throughout one year to detect metacercariae at the edges of one of the checked irrigation ditches.

The locations (Fig. 1), in which the study was carried out, were: Vegaquemada (1); Primajas (2); Orones (3); Redipollos (4); Cofiñal (5). These localities are situated in the mountains of León at altitudes ranging from 936 to 1200 m and have a continental climate within the Mediterranean–Atlantic transition. According to the meteorological data from two stations in the study area (period 1951–1980), supplied by the staff of the Duero Basin Meteorological Service (Valladolid, Spain), the maximum temperature oscillates between 14.8°C (February) and 32.6°C (July), the minimum between –14.8°C (January) and 0.5°C (August) and the average between 1.05°C (December) and 15.05°C (July). Average precipitation also varies between 36.5 mm (August) and 163.1 mm (December). Although, in previous surveys carried out along all of the Porma basin *L. truncatula* was found in 31 places, the 5 sampling localities mentioned above were chosen because molluscs infected by larval stages of *F. hepatica* were detected there.

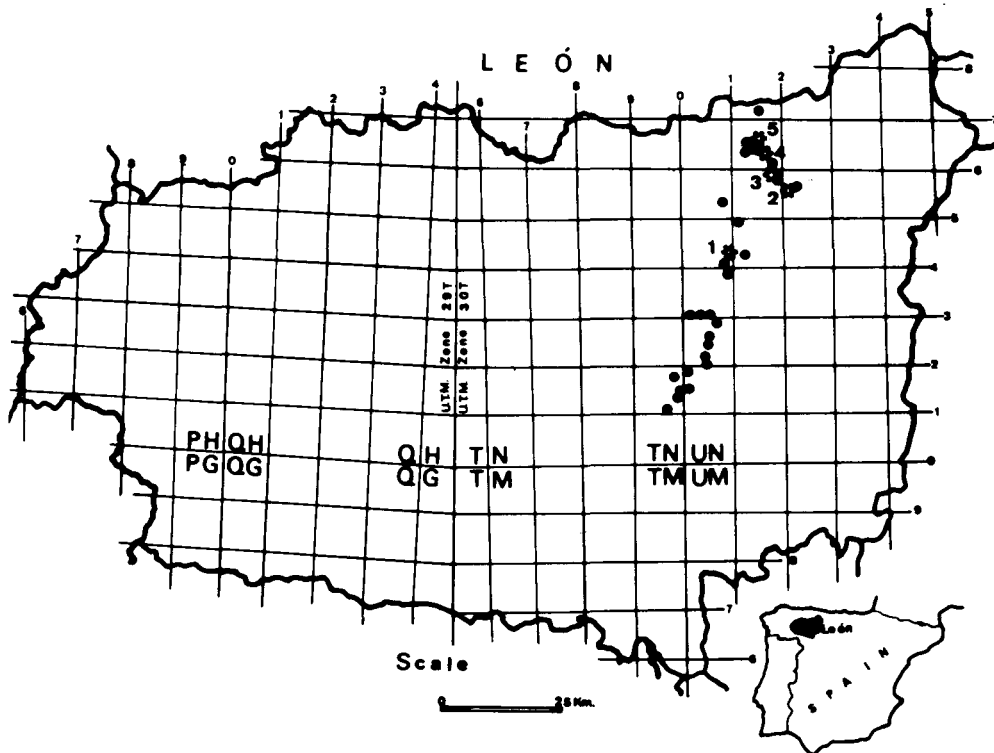


FIG. 1. Universal Transverse Mercator map showing the distribution of initial sampling points (●) and of sampling localities in which this study was carried out (★): 1) Vegaquemada, 2) Primajas, 3) Orones, 4) Redipollos and 5) Cofiñal.

The molluscs were collected from irrigation ditches, spring-swamps and small hollows waterlogged to a greater or lesser extent, in hay-meadows of the Molino-Arrhenatheretea R. TX., 1937 communities. Nevertheless, the habitat conditions of the molluscs varied according to the collection periods. The following categories were established visually: 1) wet ground, but no visible water; 2) stagnant or slow-moving water; 3) plentiful water moving more rapidly and sometimes flooding the nearest area; 4) water with snow or ice surrounding it.

When the molluscs were collected, water temperature and other physico-chemical parameters were recorded. Data concerning average monthly precipitation and temperature came from the meteorological stations nearest to the study area.

In order to obtain data on the abundance of *L. truncatula* in the study area, two sampling methods were used: 1) timed quantity (number of snails collected by one person in ten minutes) and 2) volume quantity (number of snails found in 1500 cm³ of mud collected at random in each point). However, abundance data is based on the joint results of both sampling methods.

After measuring the height and width of the shell, 5486 molluscs were checked for helminth presence. In general, between 30 and 70 *L. truncatula* specimens were studied (depending on abundance) for each collection point and date.

A stereomicroscope was used to dissect snails *in vivo* in order to detect, count and extract the different larval stages of the helminths found in their various organs. The parasites were studied in detail under the microscope using specimens of fresh material representative of the stages found in each mollusc or shed by them.

In order to confirm the identity of the *F. hepatica* cercariae shed by the molluscs, three lambs were infected with 50, 50 or 100 metacercariae. After this, the lambs were kept under controlled conditions in the Parasitology Laboratory animal enclosure.

Considering all the sample collection points, as a whole and individually, infection prevalence and intensity (maximum, minimum, average and standard error) were recorded: 1) for all the molluscs examined; 2) for the different groups of snails according to shell length (from mm to mm); 3) for the snails collected and examined at monthly intervals; 4) for the different groups of molluscs according to average ambient temperature during the collection month (from °C to °C); 5) for the groups according to average monthly precipitation (in 20 mm stages).

To find out whether there were differences in *F. hepatica* infection prevalence among the mollusc groups mentioned above (2-5), the contingency tables of the chi-square test (χ^2) were used. Likewise, one-way analysis of variance and lowest significant difference (LSD) between mean values were used to determine whether there were differences in the number of parasites per mollusc, with regard to the groups and divisions previously mentioned.

RESULTS

The physico-chemical parameter values for the water (T^a: 0°-15°C; pH: 6-8.5; conductivity: 85-850 mhos/cm; hardness: 20-250 mg/l; alkalinity: 5-200 mg/l; calcium: 15-190 mg/l; magnesium: 4-140 mg/l; chloride: 2.5-17.5 mg/l; sulphate: 1-70 mg/l; nitrogen, nitrate: 0.3-1.6 mg/l; iron: 0.01-0.09 mg/l; phosphate: 0.05-0.65 mg/l) and for the soil (pH: 5.4-7.4; total nitrogen: 0.24-0.62%; organic matter: 7.02-17.92%) varied greatly among the sample points and within each of them, according to the time of year when the samples were collected.

Live molluscs were collected in all the samplings carried out. The average abundance of *L. truncatula* was quite similar for the 5 sample localities and varied

between 40.28 ± 3.38 and 47.73 ± 5.98 . However, when individual values were considered, large variations, even within each point, were observed. The lowest values were recorded from Primajas (2) and the highest from Vegaquemada (171). The abundance of *L. truncatula* did not follow any clear seasonal pattern. In general, the highest numbers of snails were recorded in September, May and November whilst July and August appeared to be the least suitable for mollusc collection. In the samplings which showed greatest abundance, little water movement appeared to dominate (category 2 of those mentioned in 'Materials and Methods') and water temperature was between 8°C and 11°C. The minimum number of molluscs was recorded when samples were taken from wet earth with no visible water.

When the distribution of the size of molluscs examined monthly was considered, in general, the younger ones predominated. There was a predominance of those measuring 3–4 mm in the winter months and those measuring 4–5 mm during the rest of the year, except in June and July when the 5–6 mm size predominated. The highest percentage of examined snails, taking the total number into account, was the 4–5 mm size group (31.13%), followed by the 3–4 mm (24.63%) and the 5–6 mm (19.30%), while the lowest were the 9–10 mm (0.12%) and the 8–9 mm size group (0.43%). (As a point of reference, adult height varies between 8–12 mm according to JANUS, 1968).

Although, *L. truncatula* was found harbouring other trematodes (MANGA & GONZALEZ-LANZA, 1989), in this paper only data concerning its natural infection by *F. hepatica* are recorded. Nevertheless, due to the extent of the above mentioned *F. hepatica* study, the morphometric data on the stages studied and their variations with time will be published at a later date.

The infection prevalence for the 5486 *Lymnaea truncatula* specimens studied from the places of sampling varied between 0.63% and 33.85%, average 11.41 ± 6.11 . Similarly the maximum number of the parasites (sporocysts + rediae) per snail was 151, the minimum 1 and the average 20.14 ± 0.78 .

Prevalence and intensity values (Table I) increased with the size of the mollusc. As can be seen in Table II molluscs with *F. hepatica* were found every month. Maximum for infection prevalence (18.73%) and average intensity (28.48) were recorded in October, the minimum for prevalence in June (2.45%) and for average intensity (11) in May.

The infection intensity and prevalence values for the molluscs divided into categories according to temperature and precipitation are shown in tables III and IV, respectively.

TABLE I. Infection prevalence and intensity according to length of mollusc.

Length groups (mm)	Molluscs		Infection intensity	
	Exam.	Infec. (%)	Range	$\bar{x} \pm s.e.$
1–2	110	1.81	6–9	7.50 ± 1.50
2–3	588	3.74	1–24	10.40 ± 1.38
3–4	1349	6.00	1–37	11.35 ± 0.98
4–5	1705	8.68	1–70	16.28 ± 1.09
5–6	1057	16.27	1–77	19.76 ± 1.17
6–7	463	23.32	1–95	22.13 ± 1.99
7–8	173	32.36	3–151	38.93 ± 4.26
8–9	24	45.83	3–116	39.64 ± 9.89
9–10	7	85.71	18–51	39.33 ± 4.96

TABLE II. Monthly variations in *L. truncatula* infection by *F. hepatica*.

Months	Molluscs		Infection intensity	
	Exam.	Infec. (%)	Range	$\bar{x} \pm s.e.$
January	170	3.53	4-53	21.33±7.16
February	293	13.31	1-116	16.92±3.21
March	611	11.62	1-64	17.37±1.67
April	426	6.10	3-56	18.54±2.71
May	606	3.30	1-72	11.00±3.45
June	367	2.45	2-46	22.10±4.57
July	322	14.29	1-50	15.17±1.86
August	231	6.93	1-60	18.13±3.43
September	748	13.90	1-90	20.35±1.87
October	614	18.73	2-151	28.48±2.23
November	487	17.86	1-127	22.09±2.50
December	601	11.15	1-61	14.83±1.44

TABLE III. Prevalence and intensity of infection by *F. hepatica* for different groups of molluscs according to average ambient temperature.

Temperature (°C)	Molluscs		Infection intensity	
	Exam.	Infec. (%)	Range	$\bar{x} \pm s.e.$
(-1)-0	555	11.89	1-116	15.92±2.11
1-2	666	11.41	1-61	14.03±1.33
2-3	308	3.24	3-49	20.80±4.21
3-4	501	17.96	1-70	19.12±1.58
4-5	268	14.55	1-127	27.07±4.82
5-6	238	6.72	3-56	19.43±3.73
6-7	350	2.57	2-14	7.11±1.39
10-11	436	26.14	2-151	28.30±2.21
11-12	359	3.34	2-72	15.91±5.86
12-14	440	2.04	2-46	22.77±5.05
14-15	339	18.28	1-86	20.74±2.48
15-16	576	9.54	1-90	19.96±2.58
16-17	163	11.65	3-29	13.80±1.70
17-20	287	10.10	1-50	15.58±2.40

TABLE IV. Prevalence and intensity of infection by *F. hepatica* for different groups of molluscs according to average precipitation.

Precipitation (mm)	Molluscs		Infection intensity	
	Exam.	Infec. (%)	Range	$\bar{x} \pm s.e.$
0-20	947	11.19	1-75	21.03±1.53
20-40	838	8.71	1-151	24.22±2.91
40-60	800	10.12	1-116	21.00±2.32
60-100	562	8.54	2-49	16.12±1.78
100-120	462	16.01	1-70	18.29±1.86
120-140	553	11.75	1-127	21.21±3.10
140-160	214	10.28	1-36	14.72±1.99
160-180	130	17.69	1-116	21.30±5.35
180-200	306	0.65	5-16	10.50±5.50
200-220	408	18.87	1-86	20.67±2.12
220-240	266	13.15	1-61	16.08±2.06

TABLE V. Infection prevalence and intensity of the molluscs according to sampling localities.

Localities	Molluscs		Infection intensity	
	Exam.	Infec. (%)	Range	$\bar{x} \pm s.e.$
Redipollos	1016	33.85	1-127	22.41 \pm 1.12
Primajas	1113	13.38	1-87	15.00 \pm 0.97
Orones	1154	8.57	1-151	20.70 \pm 2.17
Cofiñal	1103	0.63	5-30	11.50 \pm 3.15
Vegaquemada	1090	0.64	3-37	18.42 \pm 5.76

The values for infection intensity and prevalence for the molluscs from the 5 sampling points are shown in table V. Due to the low *L. truncatula* infection figures recorded from Cofiñal and Vegaquemada (possibly because there was a lot of fast-moving water), only the results obtained from Redipollos, Orones and Primajas are discussed. At these 3 points the tendency towards increased values for both prevalence and intensity (Fig. 2) as the size of the snails increased was noted.

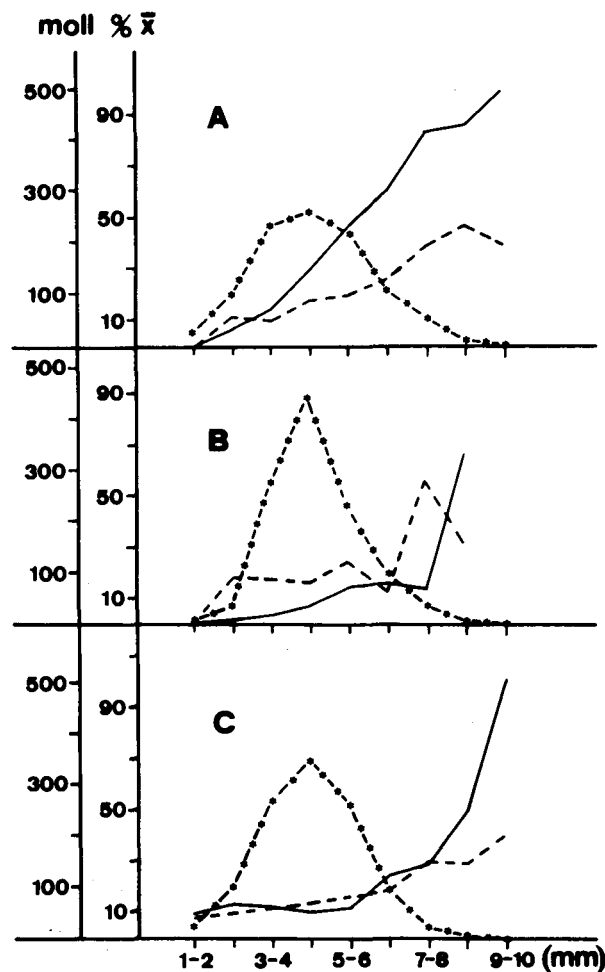


FIG. 2. Variation of *F. hepatica* infection according to the length of the molluscs. *—* No molluscs examined. — Prevalence (%). - - Intensity (\bar{x}). A) Redipollos, B) Orones, C) Primajas.

Similarly, the monthly prevalence patterns (Fig. 3) show certain similarities, with a pronounced maxima in October–November, February and July–August. As far as intensity is concerned, the results are more varied. The maximum value was recorded in Redipollos in October, in Orones in June, and in Primajas in April (Fig. 3).

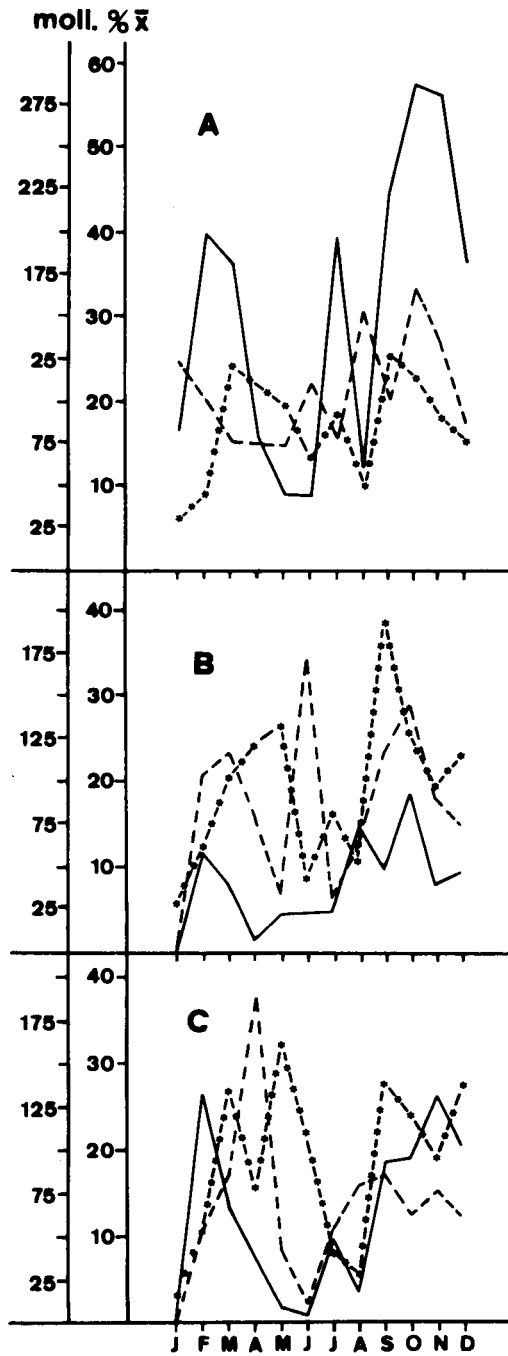


FIG. 3. Monthly variation of *L. truncatula* infection by *F. hepatica*. *—* No molluscs examined. — Prevalence (%). - - - Intensity (\bar{x}). A) Redipollos, B) Orones, C) Primajas.

When the chi-square test was applied, statistically significant differences were noticed ($p \leq 0.005$), in relation to the infection prevalence among the groups of molluscs established according to: 1) their length ($\chi^2 = 312.71$), 2) the months in which they were collected ($\chi^2 = 159.79$), 3) temperature values ($\chi^2 = 256.29$), 4) precipitation values ($\chi^2 = 60.14$) and 5) the sampling localities ($\chi^2 = 791.91$).

On applying 2×2 contingency tables, significant differences were detected as regards prevalence, between all the length groups ($p \leq 0.05$) and all the sampling localities ($p \leq 0.005$), except between Cofiñal and Vegaquemada.

Using 2×2 contingency tables, statistically significant differences were recorded for prevalence between several months. Nevertheless, it must be pointed out that the highest values of χ^2 for $p \leq 0.005$ were obtained between: October and May, October and June, November and June, November and May. According to our results October (the month in which the prevalence value was the highest) shows statistically significant differences from all the months except November and December. When the same type of analysis was used statistically significant differences were obtained, for prevalence, between various groups of molluscs according to temperature and precipitation.

By means of one-way analysis of variance statistically significant differences ($p \leq 0.005$) were observed concerning the intensity of the infection, among the groups of molluscs established according to: 1) their length ($F = 17.263$; D.F. = 7 and 598), 2) the months in which they were collected ($F = 3.624$; D.F. = 11 and 594), 3) temperature values ($F = 3.119$; D.F. = 16 and 589) and 4) the sampling localities ($F = 4.379$; D.F. = 4 and 601). Nevertheless, no statistically significant differences were found among the groups of molluscs constituted according to the values of precipitation.

When pairs of mean values of intensity were compared, taking into account the LSD test, significant differences for $p \leq 0.05$ were observed between: 1) May and September, May and November, November and July, November and December, October and the rest of the months (except January and June); 2) Primajas and both Orones and Redipollos.

Although statistically significant differences were observed when pairs of mean values of intensity for both length and temperature were compared, it cannot be deduced from our results that these parameters had any clear influence on infection intensity.

In general, *F. hepatica* rediae at different stages of development and, at times, sporocysts were found in the same snail. Six hundred and six *L. truncatula* specimens were detected with infection. Of these 40.26% were infected solely with undeveloped rediae or rediae with only germinal masses. Furthermore, these types of rediae were accompanied by rediae with immature cercariae (in 16.00% of the infected molluscs), by rediae with mature cercariae (in 13.53%) and by rediae with immature and rediae with mature cercariae (11.88%). Less frequently, snails harbouring rediae with daughter rediae together with undeveloped rediae or rediae containing cercariae in different stages of development were found. Only 1.15% of the infected snails contained sporocysts exclusively whilst 1.48% harboured them together with rediae.

The degree of larval stage development found in the molluscs did not follow any clear pattern throughout the year (Fig. 4). In our opinion, mollusc infection could occur at almost any time, since sporocysts were found in February, March, July, August, September and October and rediae with germinal masses every month. However, it seems that most infections occurred in both February–March and at the end of summer–beginning of autumn, since, as well as observing sporocysts at

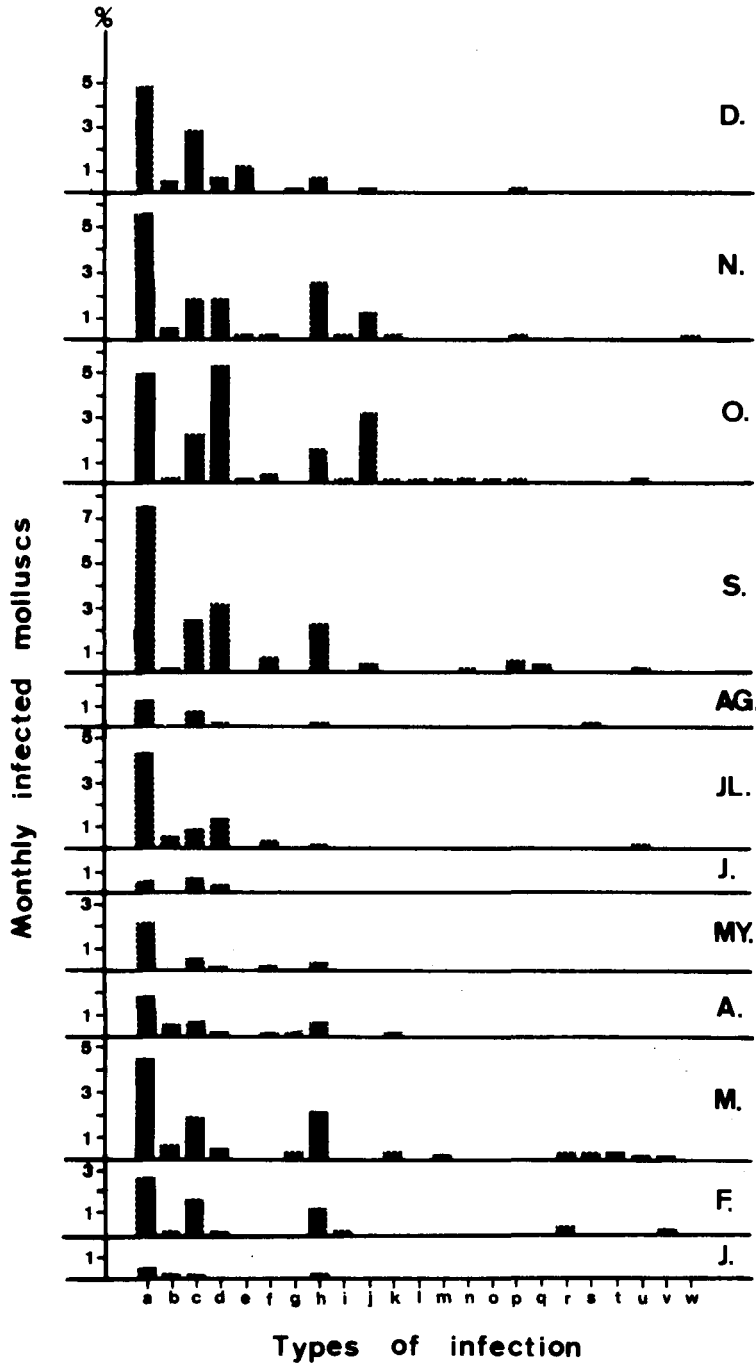


FIG. 4. Monthly percentage of infected molluscs (on the total of them) according to different stages of development that they harboured. a=Non-evolved rediae or those with germinal mass (Rgm); b=Rgm+Rediae with rediae inside (Rr); c=Rgm+Rediae with immature cercariae (Ric); d=Rgm+Rediae with completely mature cercariae (Rmc); e=Rgm+Cercariae recently shed by the molluscs (Cs); f=Rgm+Rr+Ric; g=Rgm+Rr+Ric+Rmc; h=Rgm+Ric+Rmc; i=Rgm+Ric+Rmc+Cs; j=Rgm+Rmc+Cs; k=Rgm+Rr+Rmc; l=Rgm+Rr+Rmc+Cs; m=Ric; n=Ric+Rmc; o=Ric+Rmc+Cs; p=Rmc; q=Rmc+Cs; r=Non-evolved sporocysts or those with germinal mass (Sgm); s=Sporocyst containing rediae (Sr); t=Rgm+Sgm; u=Rgm+Sr; v=Rgm+Sgm+Sr; w=Rgm+Ric+Sr.

this time, high percentages of molluscs harbouring rediae with germinal masses were found.

Rediae with mature cercariae ready to be shed were detected every month of the year (Fig. 4). Nevertheless, the highest rate of these rediae was recorded between September and December. Likewise, cercariae shedding by recently-collected snails and metacercariae in the grass samples were also observed at the end of autumn. For this reason, this period of time must be considered as the most suitable for the infection of the definitive hosts by the ingestion of infective metacercariae. Moreover, the great abundance of molluscs at that time of year could favourably influence the number of cercariae shed.

The three experimentally-infected lambs started eliminating eggs of *F. hepatica* between 8 and 10 weeks post-infection.

DISCUSSION

According to our results, it seems that *L. truncatula* can live in habitats with a wide range of chemical characteristics, as has been pointed out by GOLD (1980).

We agree with OVER & DAMEN-VAN HAPERT (1967) when they say that the correlation between the percentages of molluscs infected by *F. hepatica* and the infection danger to cattle and sheep is arbitrary, if the number of snails per unit of grazing land has not been estimated beforehand. However, it should be pointed out that it is difficult to come to any valid conclusions because of the peculiarities of the habitat of *L. truncatula*.

The great variation observed in the abundance of *L. truncatula* of the different biotopes, as well as within each of them, coincides with what STYCZYNSKA-JUREWICZ (1965) reported.

Infection prevalence found in our studies (11.41%) was higher than that recorded by OVER & DAMEN-VAN HAPERT (1967) (less than 1%) in Holland, SMITH (1984) (4.6%) in Great Britain, SIMON (1968) (5.36%) in Spain but lower than that given by LOMBARDEO *et al.* (1979) (30%) in *L. viator* from Argentina. SAVIN *et al.* (1978) recorded an infection percentage in *L. truncatula* which varied between 1% and 11% in Yugoslavia and CVETKOVIC & LEPOJEV (1978) recorded it as being between 0.25% and 50% in SR Serbia. Our intensity values agree, in general, with those obtained by RONDELAUD (1974).

In all the length groups, and for both prevalence and intensity, the figures we recorded were always higher (except for the average intensity of those measuring 7–10 mm) than those obtained by SMITH (1984). However, the tendency, observed by us, for the prevalence to increase with the size of molluscs coincides with that pointed out by KENDALL & OLLERENSHAW (1963), SIMON (1968), OVER (1971) and SMITH (1984).

As previously stated, statistically significant differences in the mollusc infection rate in relation to temperature and precipitation were noticed. To a certain extent this agrees with that found by CHOWANIEC (1961) for temperature.

The monthly pattern of *L. truncatula* infection prevalence observed in this study coincides with that found by OLLERENSHAW (1967) and SMITH (1981) in Wales, although a little different as far as time is concerned. They recorded the highest percentages between August and September and OLLERENSHAW (1967) found the lowest values in June (sometimes in May). This may be due to the fact that summer temperatures in Great Britain are lower and winter temperatures fall earlier than in the area of this study. SIMON (1968) found infected snails only between June and August (maximum in July) in Salamanca, with the development of larval stages beginning in May.

Mollusc infection in the Porma basin could logically occur at any time throughout the year, as *F. hepatica* egg elimination with the faeces of the definitive hosts takes place without any interruption (GONZALEZ-LANZA *et al.*, 1989; MANGA *et al.*, 1990). Moreover, in the study area the maximum annual temperatures (from 14.8°C in February to 32.6°C in July) are within the thermal limits between which the miracidium can mature and leave the egg, according to various authors (e.g. DIEZ & ROJO, 1976; LEIMBACHER, 1981). In addition, active *L. truncatula* specimens were found throughout the year. Nevertheless, it seems that most infections occur in both the February–March and the end of summer–beginning of autumn periods, since, as well as observing sporocysts at this time, high percentages of molluscs harbouring rediae with germinal mass were found.

The increase of the infection prevalence in February–March may be due to the fact that the contamination of grazing lands by *F. hepatica* eggs is great, since the highest levels of egg elimination by cattle (GONZALEZ-LANZA *et al.*, 1989) and sheep (MANGA *et al.*, 1990) in the same area occurred in winter. However, it is also possible that some of the parasites detected in the molluscs at the beginning of the year came from molluscs infected in autumn and which survived the winter, as stated by DROZDOWSKI (1958) and OLLERENSHAW (1971).

On the other hand, the increase in infection prevalence detected in October and July could be related to the increase observed in the number of snails in September and May. Moreover, temperatures at that time of year are more suitable for miracidium hatching and development. Contact between the miracidium and its intermediate host would be facilitated as spring and summer progress.

F. hepatica transmission to the definitive hosts may take place all the year round, because rediae with mature cercariae were detected every month and maximum annual temperatures were always over 9°C, the minimum value stated by KENDALL & McCULLOUGH (1951) for cercarial shedding. However, we considered the end of autumn the period most suitable for this event, since cercarial shedding by molluscs and metacercariae in grass samples were primarily observed between September and December. This fact could explain why the main egg elimination period by the definitive hosts was observed in winter (GONZALEZ-LANZA *et al.*, 1989; MANGA *et al.*, 1990).

Our interpretation of the periods when infection of both intermediate and definitive hosts occur only partly coincides with that of some other authors. KENDALL (1960) referred to autumn as the period when acute fascioliasis is usually found in the north of Europe. OLLERENSHAW (1967) found that in Wales the snails, infected from the end of May until the beginning of August, start to shed cercariae towards the end of July and continue until the end of October. RONDELAUD (1974) stated that cercarial shedding in summer fascioliasis in France took place between the end of August and the middle of October. OVER (1982) observed shedding of *F. hepatica* cercariae principally between April and October in The Netherlands. In Sweden, NILSSON (1974) noticed the existence of metacercariae on grazing land in May–June and September–October. ROSS (1967) pointed out that in Northern Ireland the greater part of the metacercarial infection occurred between August and February, while infections from February to June were negligible. SMITH & WILSON (1980) recorded metacercariae from October onwards and SMITH (1981) during most of the winter.

The small delay observed in our region in cercarial shedding may be due to the fact that low temperatures appear later than in the northern countries and this allows shedding to continue until the end of autumn.

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