The three hosts of the *Ligula intestinalis* (Cestoda) life cycle in Lavernose-Lacasse gravel pit, France

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With 5 figures and 2 tables

Abstract: The life cycle of *Ligula intestinalis,* a pseudophyllidean cestode, was studied in a South-Western French gravel pit. Copepods, fish and piscivorous birds, constitute the hosts necessary for complete parasite development. In order to determine the actual species likely to be responsible for the very high level of parasitism observed, a two-year study was carried out. Our results show that the copepod *Eudiaptomus gracilis,* the roach (*Rutilus rutilus*) and the great crested grebe (*Podiceps cristatus*) were the most favourable hosts for continuation of the parasite life cycle. The study shows how alterations exerted by infective larval stages of *L. intestinalis* affect the population dynamics of roach.

Key words: Cestode, infection, parasitism, intermediate hosts.

Introduction

Ligula intestinalis is a common and widespread pseudophyllidean cestode which is strongly pathogenic in many natural freshwater fish populations throughout Europe (DUBININA 1980). The complex life cycle of the parasite requires three hosts: a copepod as the first intermediate host, a cyprinid fish as the second intermediate host and an avian predator as the final host. Free-swimming coracidia larval stages are eaten by planktonic copepods and transform in the gut of their first host into six-hooked oncospheres, which rapidly mature into procercoid forms on entering the haemocoel cavity. Infected copepods are ingested by planktivorous freshwater fish, essentially cyprinids (DU-

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BININA 1980) in which the procercoids develop into plerocercoid larvae in the host's abdominal cavity. The final hosts in which the parasites acquire their sexual maturity are ichthyophagous birds (DUBININA 1980).

Numerous reports have listed the pathogenic effects of such infections (DENCE 1958, ARME & OWEN 1968, 1970, WILSON 1971, SWEETING 1975, WYATT & KENNEDY 1989). The pathogen can have a detrimental effect on the host leading to high mortality rates in fish populations.

Infections are subject to numerous factors which vary with the individual host, its taxonomic position, the local environment in which it lives, and time. In Lavernose-Lacasse gravel pit most individuals of roach become infested with an over-dispersed population of *Ligula*, whereas at other sites in South-Western France, the prevalence of *Ligula* never exceeded 15 % (LOOT, unpubl. data).

The aims of the present study were i) to determine intermediate and final hosts which favour local parasite dynamics, ii) to quantify the magnitude of the pathogenic effects exerted by this tapeworm on fish populations and iii) to discuss its cost for intermediate hosts within the context of the adaptative "parasite increases trophic transmission" strategy (LAFFERTY 1999).

Material and methods

Study site

The Lavernose-Lacasse gravel pit is located near the city of Toulouse in South-West France (Fig. 1). This gravel pit belongs to the Garonne River alluvial plain and is 190 m above sea level. Its total surface area is about 23 ha with a maximum depth of 4 m and a mean depth of 2.7 ± 0.4 m. The average annual water temperature is about 14 °C (range 5–25 °C).

For the purpose of this study, we decided to survey the three host compartments necessary for the complete *L. intestinalis* parasitic life cycle, from April 1998 to April 2000. In addition, experimental infestations of copepods by coracidia larvae and of fish by infected copepods were investigated.

Zooplankton compartment

Natural population survey

Monthly samples were collected using a plankton net (mesh size 100 μm) vertically hauled from the bottom of the gravel pit to the surface of the water. The approximate volume of water which was filtered through the plankton nets each month is 120–130 litres. Zooplankton samples were immediately preserved in a final 4 % formaldehyde solution. In the laboratory, density and developmental stages (copepodites C1, C2, C3, C4 and adults) of each cyclopoid and diaptomid species present in the samples were investigated using a binocular microscope (8 × 50) (DUSSART 1967).
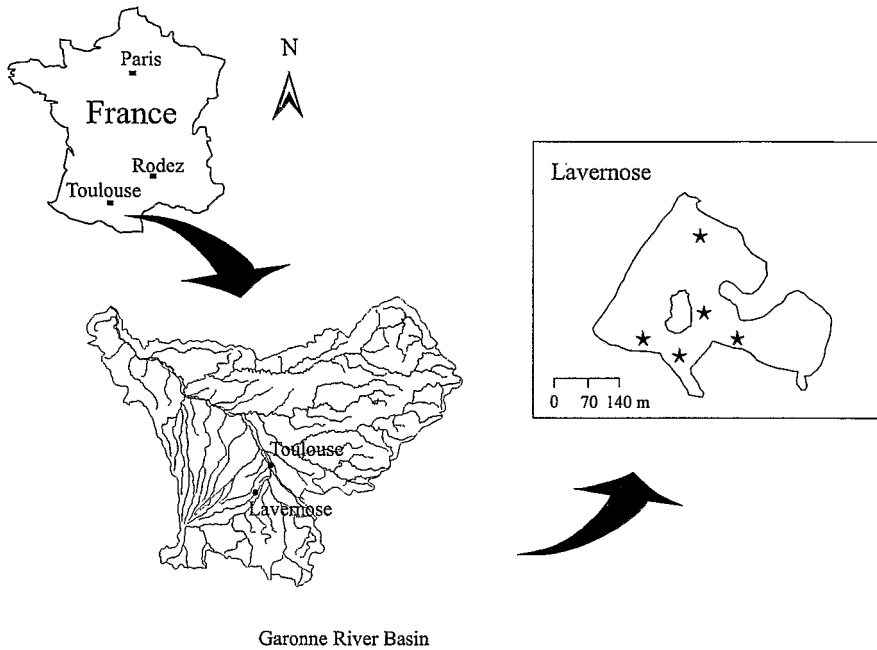
Garonne River Basin

Fig. 1. Geographical location of the Lavernose-Lacasse gravel pit. The stars on the gravel pit map represent the sampling stations of fish and copepods.

Experimental infestation of copepods

A grey heron was fed with an infected fish and parasite eggs were obtained after bird faeces sifting and centrifugation (see GERDEAUX 1986). Different species of living planktonic copepods were captured at Lavernose-Lacasse gravel pit in December 1999 using the plankton net. We considered the copepods to be parasite-free since during severe ligulosis of fish in Lavernose gravel pit we found only one crustacean infested with one procercoid by the examination of 600 individuals. Hence the risk of having used naturally infected copepods was negligible. All copepods were kept in an experimental tank ($100 \times 50 \times 50$ cm) in water from Lavernose at a temperature of 18 °C and natural photoperiod. They were fed each day with algae from Lavernose and *Paramecium* cultures. Experimental conditions were as similar to the environmental conditions as possible so as to avoid differences in the ability of the three copepod species to become adapted to the lab conditions. Mortality rates of *E. gracilis, Mesocyclops leuckarti* and *Acanthocyclops robustus* were not significantly different, e.g. 50 % when copepods were introduced into the tank and 5–6 % after 5 days of acclimation to the storage conditions. At this time the copepods were infected by introducing approximately 10 *Ligula* eggs per individual copepod present in the experimental tank. Coracidia are formed on the sixth day of development of eggs in water and the calculated hatching rate was about 60 %. Coracidia larvae which swim freely in the water are actively eaten by copepods. Twenty copepod specimens were taken each day after the date of

adding the eggs and for 30 days. Examination of living copepods anaesthetised with CO_2 solution proved to be the best method for the detection of procercoids (DUBININA 1980). We determined, under a microscope, the presence of parasitic larval forms in the haemocoel of each copepod species and stage of development.

Fish compartment

Natural population survey

Monthly overnight fish samples were taken with gill-nets. We used 30 m long and 1.7 m high clear nylon monofilament gill-nets of different mesh sizes (10, 12, 14, 17, 21 and 27 mm) in order to capture a wide range of fish specimens. The total number of fish investigated was 1651 over the two-year period. After their capture, the fish were dissected to determine the presence of L. intestinalis plerocercoids. In parallel, the fish specimens were aged using the scale measurements (HARTLEY 1947). The scales were cleaned in a 5 % KOH solution before the adhering tissues were rubbed off with a soft brush. Then, they were rinsed in water and mounted on microscopic slides for examining under a scale viewer. To estimate host fish mortality due to parasites, we used a method described by LESTER (1984) based on changes in parasite prevalence with host age. For parasites with low virulence, prevalence increases in older fish because older hosts have been exposed to infection for a longer period of time (ANDERSON & GORDON 1982). With a virulent parasite, prevalence decreases in older fish because of the death of the most heavily infected hosts which are selectively removed from the population. Maximum prevalence corresponds to the host age class (i.e. modal age class) subjected to the highest parasite impact (ROUSSET et al. 1996).

Experimental infestation of fish

Experimental infestations were performed on silver bream (Blicca bjoerkna (LINNAEUS, 1758)) and roach (Rutilus rutilus (LINNAEUS, 1758)). Silver bream were taken from the Lavernose-Lacasse gravel pit where tapeworm infestation had not been observed in this fish for a number of years, and roach were taken from the Garonne river where ligulosis has never been recorded (LOOT, unpubl. data). Individuals of the two fish species were placed singly in different aquaria. The experimental infestation started on 15 March 2000. In each aquarium, we placed 10 infected diaptomids per fish, each copepod harbouring a total of 4–5 procercoids. The experimental fish specimens were killed after infestation in October 2000. We observed the presence/absence of plerocercoid larval forms in the hosts' abdominal cavity and in parallel the fish specimens were aged.

Bird compartment

Natural population survey

Waterbirds were counted every week during two hours in the morning, from April 1998 to April 2000 using a telescope (20×60) and binoculars (8×30). The small area

and open character of the gravel pit permitted a complete census of the population (TA-MISIER & DEHORTER 1999).

Results

Zooplankton compartment

Observational data

In Lavernose-Lacasse gravel pit, the planktonic copepod community was relatively species-poor with four distinct species: *Eudiaptomus gracilis* (SARS, 1863), *Acanthocyclops robustus* (SARS, 1863), *Mesocyclops leuckarti* (CLAUS, 1857) and *Cyclops vicinus vicinus* (ULIANINE, 1875). The temporal variability during the two-year survey for these four copepod species is shown in Fig. 2. *C. vicinus vicinus* appears to be very rare at Lavernose-Lacasse with a peak in density (10 individuals/litre) in October 1998. The three other copepod species occurred more often during this survey. *M. leuckarti* showed a bimodal distribution of density indicating that it was relatively more frequent from April to the end of October. *A. robustus* was more frequent in our survey from March 1999 to April 2000, but this strongly contrasts with its relative scarcity from April 1998 to April 1999. Finally, *E. gracilis* was the most widespread copepod species present in almost all the samples during the two-year survey, and the most abundant species present within the planktonic community.

Experimental infestation

It was impossible to test the potential infectivity of *Cyclops vicinus vicinus* by coracidia larvae because of its rarity in natural conditions (Fig. 2). The three other copepod species showed different susceptibilities to infestation (Table 1). *E. gracilis* showed the greatest susceptibility to parasitic infection in terms of the total number of parasitized copepods obtained (prevalence 55 %). *M. leuckarti* was only slightly sensitive to infestation. We frequently observed remains of digested oncospheres (i.e. chitin hooklets) in its gut indicating that infestation had failed. In only one case did an oncosphere succeed in penetrating into the body cavity and then in developing into a procercoid. Infestation was never observed in the third copepod species, *A. robustus*.

The success of infestation by oncospheres strongly depended on the developmental stage of the host (Table 1). Although C4, C5 larval stages and adults of *E. gracilis* might have been infected when they were younger, no parasite was ever found in stages C1, C2 and C3. Only one adult *M. leuckarti* was found to be parasitized.
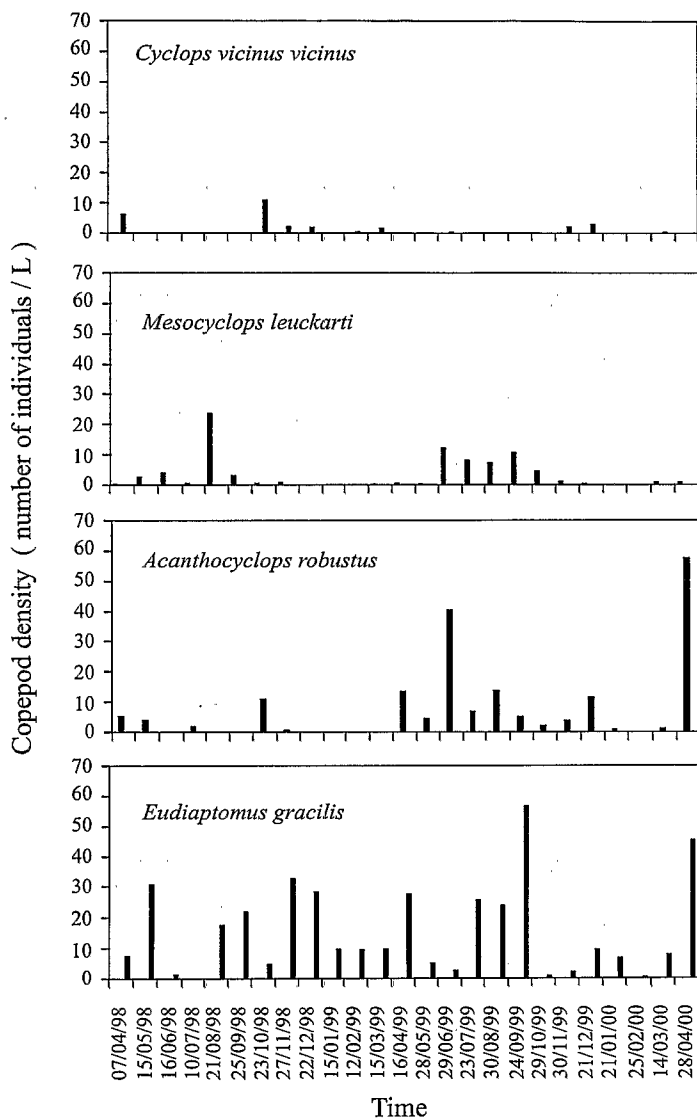
Fig. 2. Monthly changes (from April 1998 to April 2000) in copepod density (number of individuals per litre) of 4 species present within the Lavernose-Lacasse gravel pit. Only stages of development susceptible to parasitic infection by *L. intestinalis* (C4, C5 and adults, see Table 1) were considered in this analysis.

Fish compartment

Observational data

In Lavernose-Lacasse, the two most abundant cyprinid fish species were the roach, (65.7% of the total number of fish) and the silver bream (31.9%). The

Table 1. Experimental infestation of different copepod stages of three different species, *Eudiaptomus gracilis, Mesocyclops leuckarti* and *Acanthocyclops robustus,* by coracidian larval stages of the pseudophyllidean tapeworm *Ligula intestinalis.* The copepod stages were C1, C2, C3, C4, C5 and adults. Ratio of the number of infected hosts to the number of exposed hosts is indicated for each copepod stage.

Potential host	C1	C2	C3	C4	C5	Adults	Total
E. gracilis ·	0/10	0/30	0/28	25/44	68/90	113/174	206/376
M. leuckarti	0/3	0/3	0/9	0/15	0/15	1/20	1/65
A. robustus	0/1	0/2	0/2	0/4	0/5	0/12	0/26

common carp, *Cyprinus carpio* (LINNAEUS, 1758) (0.7 %), and the bleak, *Alburnus alburnus* (LINNAEUS, 1758) (0.5 %) were more sparcely represented. Despite the inspection of roach eyes, skin, abdominal cavity, digestive system and other organs *Ligula* was the only cestode parasite detected in this fish species. In natural conditions, roach was the only fish observed to be parasitized by plerocercoid larvae. The silver bream, which could have been a suitable host for *L. intestinalis* (BARUS & PROKES 1994), notably because this cyprinid species is abundant at Lavernose-Lacasse, was never found to be infected. The rare specimens of common carp and bleak caught were also never parasitized.

At first, parasitic prevalence increased rapidly with roach age (Fig. 3). Three-year old roach were strongly affected by plerocercoid forms (mean parasite intensity 5.98 ± 0.25, making up about 17.5 % of the total fish weight). They were the host age class subjected to the highest mortality (ANDERSON & GORDON 1982, ROUSSET et al. 1996). The oldest individuals were weakly infected. Patterns of prevalence changes with host age were similar for both years of the study. We observed seasonal differences in prevalence for both years (Fig. 4). The prevalence values increased in spring and decreased in winter.

Experimental infestation

Roach appears to be a suitable second intermediate host of *Ligula intestinalis* according to the total number of fish parasitized during the experiment (prevalence 18.2 %). Prevalence was maximum for two and three-year old fish. We failed to infest the oldest roach and the silver bream (Table 2).

Bird compartment

Observational data

Five species of aquatic birds were censused during the surveys: the great crested grebe, *Podiceps cristatus* (LINNAEUS, 1758), the cormorant, *Phalacro-*
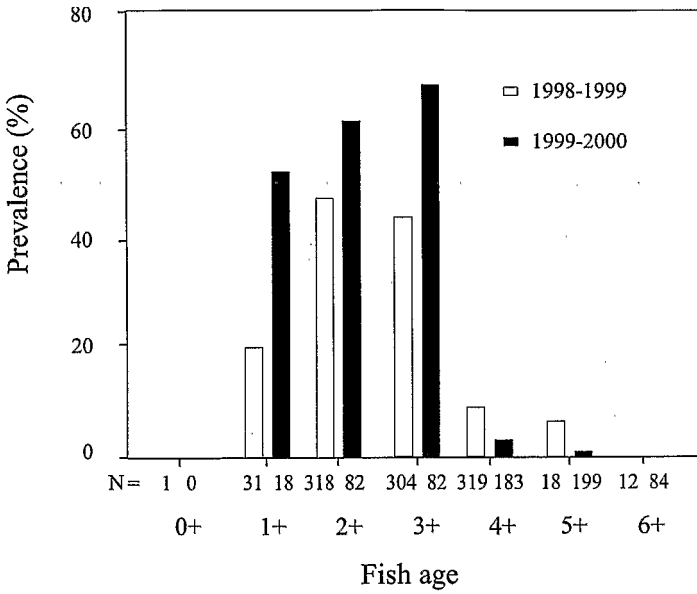
Fig. 3. Changes of prevalence in roach specimens infected by plerocercoid worms of *L. intestinalis* in relation to individual fish age. The age classes and the numbers of roach examined per class are given under each bar. Open bars correspond to the period April 1998–March 1999, and solid bars to the period April 1999–March 2000.
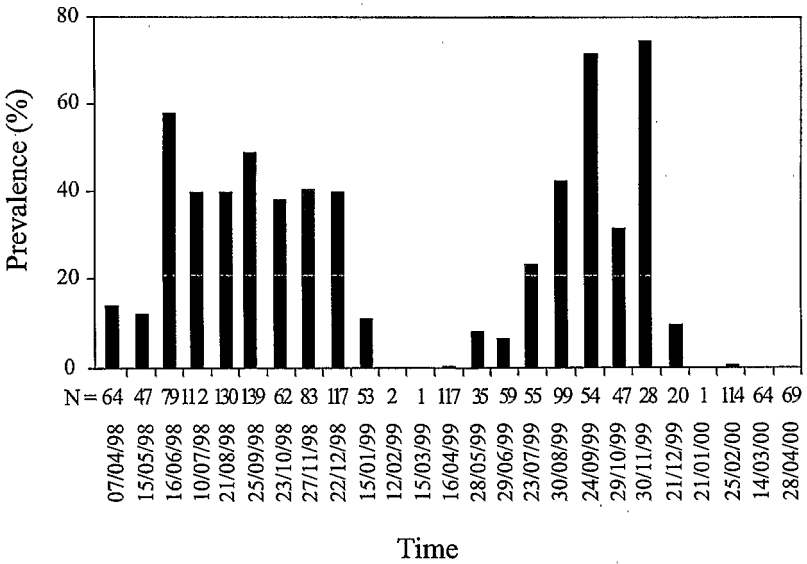

Fig. 4. Monthly changes (from April 1998 to April 2000) of prevalence in roach specimens infected by plerocercoid worms of *L. intestinalis*.

Table 2. Experimental infestation of different fish age groups of two cyprinid species, *Rutilus rutilus* and *Blicca bjoerkna* by infected copepods. Ratio of the number of infected hosts to the number of exposed hosts is indicated for each group.

Potential host	1+	2+	3+	4+	5+	6+
R. rutilus	1/10	9/22	7/27	1/12	0/13	0/15
B. bjoerkna	0/2	0/7	0/12	0/16	0/15	0/4

corax carbo (LINNAEUS, 1758), the grey heron, *Ardea cinerea* (LINNAEUS, 1758), the night heron, *Nycticorax nycticorax* (LINNAEUS, 1758) and the yellow herring gull, *Larus cachinnans* (PALLAS, 1811). All five are known to be potential final hosts for *L. intestinalis* (ROSEN 1920). In the Midi-Pyrénées region, the cormorant is known to be a wintering species (BOUSQUET 1992). It was present in the Lavernose-Lacasse gravel pit during the winter season from December 1998 to March 1999, whereas during the 1999/2000 winter cormorants were only observed in December (Fig. 5).

Both the grey heron and the night heron were very rarely observed at the study site and when so, in low numbers (Fig. 5). Finally, both the yellow herring gull and the great crested grebe were the two commonest birds over the two-year survey. The grebe is a sedentary bird species at Lavernose-Lacasse (SANTOUL & TOURENQ 2000), and it occurred in all counts with a fairly even number of specimens throughout the survey (3.60 ± 1.10).

Discussion

In this work, the question was posed as to how *Ligula intestinalis* can complete its life cycle in the Lavernose-Lacasse gravel pit and what are the community members which facilitate parasitic transmission and impart a high level of *Ligula* abundance at this site. Both field and experimental studies were conducted.

When looking for the first potential intermediate hosts of *L. intestinalis* at Lavernose-Lacasse, we found that not all the planktonic copepods present in the local community were potential agents of parasitic transmission. Firstly, in the experimental infestations of copepods only *E. gracilis* was infested. All experimental infestations of *M. leuckarti* except one failed although this copepod species eagerly swallowed a very large number of infective stages of the parasite. Interestingly, by feeding upon and consuming free-swimming coracidia, *M. leuckarti* presents a positive practical effect in controlling ligulosis, as previously hypothesised by DUBININA (1980). Under experimental conditions, GERDEAUX (1986) demonstrated that individuals of *C. vicinus vicinus* from Créteil Lake near Paris could be easily infected by *L. intestinalis* infective stages. However, it should be noted that even though the experiment demon-
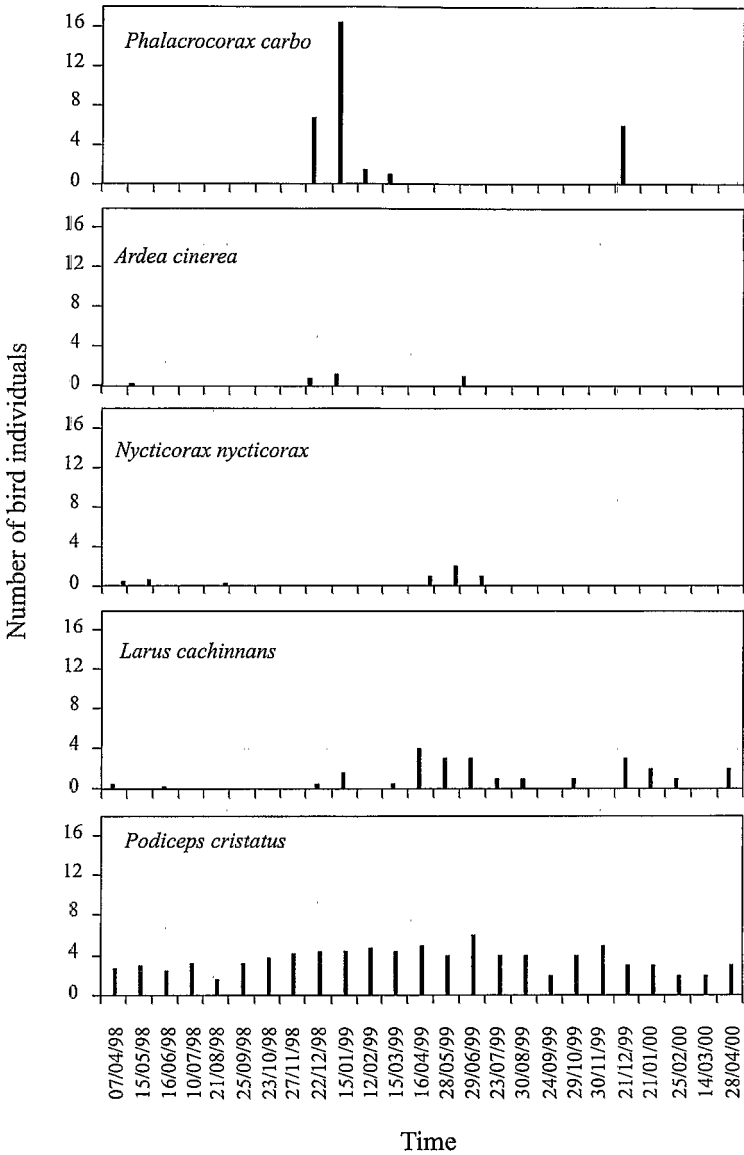
Fig. 5. Monthly changes (from April 1998 to April 2000) in numbers of individual birds of five species present within the natural community of Lavernose-Lacasse gravel pit. The bars represent mean values over 4 weeks.

strated that this copepod species is a suitable parasite host, it does not demonstrate its actual role in the zone investigated.

Secondly, infestation of various copepod species by the tapeworm larvae was shown by WATSON & PRICE (1960) to depend very closely on their mode

of life. Diaptomid copepods usually filter out the tiny particles of detritus and microplankton suspended in the water (RYLOV 1930 a, b, LOWNDES 1935, MALOVITSKAYA 1961). Interestingly, the spherical free swimming coracidia larvae roughly mimic, in body size and shape, common phytoplankton items such as flagellates and Protococcaceae which represent some of the favourite food items for diaptomids. On the contrary, most cyclopoids are predators (DZYUBAN 1937, RYLOV 1948, FRYER 1957, MONAKOV 1959) actively attacking their prey which are essentially zooplankton and other benthic animals like protozoa, rotifers, crustaceans or their own larvae. Thus, under natural conditions, diaptomid copepods are logically more prone than cyclopoid copepods to parasitic contamination by infective stages of *L. intestinalis* for behavioural reasons.

Thirdly, some copepod species are not so numerous in the local community and cannot, or could only very occasionally, contribute to the parasitic life cycle. For instance, *C. vicinus vicinus,* is certainly too scarce to be considered as an important agent of transmission. The widespread occurrence of *E. gracilis* in the Lavernose-Lacasse gravel pit throughout the two-year survey makes it a serious candidate as first intermediate host for the transmission of *L. intestinalis*.

Then, when considering the second intermediate host in the parasitic life cycle, i.e. fish, tests demonstrated that older roach were refractory to any parasitization by *L. intestinalis*. Older hosts may cease to acquire parasites as a consequence of immunity acquisition. Silver bream specimens were entirely refractory and this strongly contrasts with other studies demonstrating that the silver bream is a suitable host for infection by plerocercoids (DUBININA 1980, BARUS & PROKES 1994). It is therefore suggested that the inability for procercoids to develop into plerocercoid forms in silver bream from the Lavernose-Lacasse gravel pit is under physiological control, and that the host's immune system is involved in the inactivation of the parasite life cycle (ARME & OWEN 1968, TENORA et al. 1997). Given this, we must accept the hypotheses that other strains of *L. intestinalis* are able to colonize silver bream in other local communities or that *L. intestinalis* forms a complex of different species locally parasitizing different populations of fish.

The development of the plerocercoid phase occurs within the host fish's body cavity where it requires a long time, sometimes over a year, to achieve maturation. *L. intestinalis* greatly compresses the internal organs and simultaneously has a strong toxic influence, disrupting the normal activity of the host's individual organs and its general metabolism (ARME & OWEN 1968). In addition, the tapeworm modifies the normal swimming behaviour of the fish into a jerky movement and makes infected roach appear fat (LOOT et al., unpubl.) which seems to be an excellent way to attract predatory birds' attention. Moreover, in reducing the fish swimming and the probable escape ability the

parasite increases its present host's value as prey (LAFFERTY 1997). In the gravel pit studied, the three-year-old infected roach become easy prey for piscivorous birds. The decline of prevalence levels in winter may reflect death of more heavily infected roach and a lower transmission rate at this period. The high mortality of this strongly affected class of fish might be due to preferential predation exerted by some bird foragers, a scenario entirely compatible with the adaptative parasite-induced manipulation of the host that increases parasite trophic transmission (LAFFERTY 1997, LAFFERTY et al. 2000).

In nature, ichthyophagous birds serve as the final hosts for *L. intestinalis*. The pseudophyllidean plerocercoids found in fish and swallowed by piscivorous birds develop into mature worms in less than a week, and live only some days in the hosts's intestines. Mature pseudophyllidean cestodes are so ephemeral that infected piscivorous birds are scarcely observed in natural conditions. Consistent with this observation, there is scant evidence to demonstrate in nature, which bird species really serve as final hosts for *L. intestinalis*. Moreover, protected bird species cannot be used in experimental infestation tests. Rather, ecological surveys of the different bird species occupying a site as we did in the present work may be a convenient and simple way to evaluate the likelihood that a bird species is a final host for *L. intestinalis*. We observed that both grey and night herons occurred in very small numbers in the study site, where they only feed. These two species could not contribute to the persistence of the *Ligula intestinalis* life cycle.

The cormorant which occurs on the gravel pit during the winter season is an acceptable candidate for the final transmission of the parasite. However, the mode of life and occurrence of cormorants imply a limited dispersion of tapeworm eggs in the gravel pit. Indeed, the cormorant occurred irregularly throughout the study period. The birds occupy the open water for their foraging activities but have resting areas and a night refuge zone often far from the gravel pits (SANTOUL 2000). The yellow herring gull might also be a final host for *L. intestinalis*. However, it is an erratic winter visitor and its main activities are feeding in open water. If contamination did occur via this bird, it might only be due to chance events.

Finally, the great crested grebe showed stable numbers at the site throughout our two-year survey. Compared to that of other bird species, its behaviour including activities such as grooming, resting and feeding, is mainly restricted to water, implying the likelihood that more bird faeces containing parasite eggs may be released into the water than from other birds. Grebes usually consume 150–200 g of fish a day, with cyprinids such as roach, dace and bream forming the major part of their diet (SOBCZYK 1975). Roach specimens of 100–120 mm length (three years old) are preferentially eaten by these birds, thus making the great crested grebe the "ideal final host" responsible for *L. intestinalis* infestation in this locality.

The line of biological reasoning developed above may be supported by considerations on the physiological characteristics of bird species. DUBININA (1980) observed that in bird species with strong peristalsis, e.g. herons and cormorants, massive pseudophyllidean plerocercoid worms could not easily attach to the hosts' intestinal walls. In experiments, plerocercoids were excreted within one or two hours after infestation by parasitized prey (DUBININA 1980), rendering mating success with other sexually mature *Ligula* individuals highly improbable. In contrast, in gulls and grebes, weaker peristalsis may facilitate parasitic infection, maturation and reproduction of plerocercoid worms.

Conclusion

We showed both in natural and experimental conditions that the copepod species, *E. gracilis,* is the main and probably unique first intermediate host in the life cycle of *L. intestinalis* at Lavernose-Lacasse gravel pit. The involvement of roach as the second and also single intermediate host species of the tapeworm is also demonstrated by both observational data and experimental tests of infestation. We further demonstrated that three year old roach specimens were the most infected by plerocercoid larval forms. They are by far the specimens most probably preyed upon by the piscivorous birds that serve as final hosts. At the study site, the great crested grebe is the most likely target bird of *L. intestinalis*. However, we cannot discard the hypothesis that other bird species are also involved in the life cycle of *L. intestinalis*. Finally, we suggest that a review of regional host species involved in the parasite life cycle might provide answers to questions that remain open. Possible local adaptation, depending on the predictability of the three host compartments, should be further investigated using biological markers to detect possible local differences in strains of tapeworms.

References

ANDERSON, R. C. & GORDON, D. M. (1982): Processes influencing the distribution of parasite numbers within host populations with special emphasis on parasite-induced host mortalities. – Parasitology **85:** 373–398.
ARME, C. & OWEN, R. W. (1968): Occurrence and pathology of *Ligula intestinalis* infections in British fishes. – J. Parasitol **54:** 272–280.
– – (1970): Observations on a tissue response within the body cavity of fish infected with the plerocercoid of *Ligula intestinalis* (L.) (Cestoda). – J. Fish Biol. **2:** 35–37.

BARUS, V. & PROKES, M. (1994): Parasite load of *Ligula intestinalis* plerocercoids in adult silver bream, *Blicca bjoerkna*. – Helminthologia **31**: 91–94.

BOUSQUET, J. F. (1992): Statut du grand cormoran *Phalacrocorax carbo* en Midi-Pyrénées. – Le Pistrac **14**: 18–23.

DENCE, W. A. (1958): Studies on *Ligula*-infected common shiners (*Notropis cornutus frontalis* AGASSIZ) in the Adirondacks. – J. Parasitol. **44**: 334–338.

DUBININA, M. N. (1980): Tapeworms (Cestoda, Ligulidae) of the fauna of the USSR. – New Delhi: Amerind, 320 p.

DUSSART, B. (1967): Les copépodes des eaux continentales. – Boubée Paris, 500 p.

DZYUBAN, N. A. (1937): The nutrition of certain Cyclopidae (crustaceans). – Dokl. AN SSSR **17**(6): 315–318.

FRYER, G. (1957): The food of some freshwater cyclopoid copepods and its ecological significance. – J. Anim. Ecol. **26**: 263–286.

GERDEAUX, D. (1986): Ecologie du Gardon (*Rutilus rutilus* L.) et du Sandre (*Lucioperca lucioperca* L.) dans le lac de Créteil de 1977 à 1982. Etude de la ligulose du Gardon. – Thèse de doctorat d'état, 161 p.

HARTLEY, P. H. T. (1947): The natural history of some British freshwater fishes. – Proc. Zool. Soc. Lond. **117**: 129–206.

LAFFERTY, K. D. (1997): The ecology of parasites in a salt marsh ecosystem. – In: BECKAGE, N. E. (ed.): Parasites and Pathogens. Effects on Host Hormones and Behavior. – Chapman & Hall Ltd., London, UK, p. 316–332.

– (1999): The evolution of trophic transmission. – Parasitol. Today **15**: 111–115.

LAFFERTY, K. D., THOMAS, F. & POULIN, R. (2000): Evolution of host phenotype manipulation by parasites and its consequences. – In: POULIN, R., MORAND, S. & SKORPING, A. (eds.): Evolutionary Biology of Host-Parasite relationships: Theory Meets Reality. – Elsevier Science B.V., p. 117–127.

LESTER, R. J. G. (1984): A review of methods for estimating mortality due to parasites in wild fish population. – Helgoländer Meeresuntersuchungen **37**: 53–64.

LOWNDES, A. G. (1935): The swimming and feeding of certain calanoid copepods. – Proc. Zool. Soc. Lond. **2**: 687–715.

MALOVITSKAYA, L. M. (1961): The biology of the diaptomids – *Eudiaptomus gracilis* (SARS.) and *E. graciloides* (LILL.) (Copepoda, Calanoida) of Rybinsk reservoir. – Diss., Institute of Biology of Reservoirs of the USSR, pp. 1–132.

MONAKOV, A. V. (1959): Basic aspects of the biology of *Acanthocyclops viridis* (JUR.) and *Mesocyclops leuckarti* CLAUS (Copepoda, Cyclopoida). – Diss., Institute of Biology of Reservoirs of the USSR, pp. 1–128.

ROSEN, C. (1920): Recherches sur le développement des Cestodes. II. Le cycle évolutif de la Ligule et quelques questions générales sur le développement des Bothriocéphales. – Bull. Soc. Neuchâteloise Sci. Nat. **44**: 259–280.

ROUSSET, F., THOMAS, T., DE MEEÛS, T. & RENAUD, F. (1996): Inference of parasite induced host mortality from distributions of parasite loads. – Ecology **77**: 2203–2211.

RYLOV, V. M. (1930 a): Freshwater Calanoida of the USSR. Opredelitel organizmov presnykh vod SSSR (Freshwater organisms of the USSR), No. 1, Acad. Sci. USSR Press, pp. 1–288.

– (1930 b): Observation of the seston capture of *Diaptomus coeruleus* FISCH. – TR. Leningr. Obshch. Yestestvoisp. **60**: 149–176.

–´ (1948): Freshwater Cyclopoida. Fauna SSSR (Fauna of the USSR). – Crustaceans **3**(3): 1–318.

SANTOUL, F. (2000): L'avifaune aquatique des gravières de la plaine alluviale de la Garonne. Conditions d'accueil, modalités d'exploitation et de gestion des gravières de Saint Caprais et Lavernose-Lacasse. – Thesis, Univ. Toulouse, 180 p.

SANTOUL, F. & TOURENQ, J.-N. (2000): Capacité d'accueil des gravières de la plaine alluviale de la Garonne vis-à-vis du Grèbe huppé (*Podiceps cristatus* L.). – Annls. Limnol. **36**: 203–212.

SOBCZYK, R. (1975): Great crested grebes (*Podiceps cristatus* L.) on some of the lakes in the Mazurian lakeland. – Polskie Arch. Hydrobiol. **22**: 181–194.

SWEETING, R. A. (1975): Studies on *Ligula intestinalis*. Some aspects of the pathology in the second intermediate host. – J. Fish Biol. **10**: 43–50.

TAMISIER, A. & DEHORTER, O. (1999): Camargue canards et foulques. – Centre Ornithologique du Gard, Montpellier, 369 p.

TENORA, F., KRACMAR, S., BARUS, V. & DVOWACEK, J. (1997): A preliminary report on selected chemical elements in plerocercoids of the tapeworm *Ligula intestinalis* (Pseudophyllidea). – Helminthologia **34**: 180.

WATSON, N. H. F. & PRICE, J. L. (1960): Experimental infections of cyclopoid copepods with *Triaenophorus crassus* (FOREL) and *T. nodulosus* (PALLAS). – Can. J. Zool. **38**: 345–356.

WILSON, R. S. (1971): The decline of a roach *Rutilus rutilus* (L.) population in Chew Valley Lake. – J. Fish Biol. **3**: 129–137.

WYATT, R. J. & KENNEDY, C. R. (1989): Host-constrained epidemiology of the fish tapeworm *Ligula intestinalis* (L.). – J. Fish Biol. **35**: 215–227.