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Parasitic infection by the nematode *Anguillicola crassus* (Kuwahara, Niimi et Itagaki, 1974) in the European eel *Anguilla anguilla* (Linnaeus, 1758) (Pisces Anguilliidae) of Lake Oubeïra (eastern Algeria)

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ABSTRACT In the course of our work on the European Eel *Anguilla anguilla* (Linnaeus, 1758) from the El Kala wetland (RAMSAR, 2000), we found that this species is of great biological and socio-economic importance. Unfortunately, this extraordinary global genetic heritage, long considered as a robust species with low vulnerability, is in constant decline throughout its range and made us aware of the fragility of the species. Therefore, we were interested in the impact of parasitism caused by the nematode ectoparasite *Anguillicola crassus* on the overweight of this species populating Lake Oubeïra (eastern Algeria), and those on a total of 724 eels sampled, with a total length between 10.5 and 89.9 cm, and a total weight between 17 and 1470 g. The epidemiological study reveals that eels suffer from several parasitic attacks including infection of the swim bladder by the nematode *Anguillicola crassus*. The presence of the eel in Lake Oubeïra throughout the year increases the rate of infection. Anguillicolosis is important in large individuals and in different sexes and stages of silvering.

KEY WORDS *Anguilla anguilla*; *Anguillicola crassus*; Overweight; Oubeïra; Wetland.

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INTRODUCTION

The European eel (*Anguilla anguilla* Linnaeus 1758) (Pisces Anguilliidae) has long appeared as a common species representing a major component of coastal environments, and continental waters close to the sea. It is of great socio-ecological interest, because it sometimes constitutes half of fish biomass; also it is considered to be an excellent bio-integrator of environmental quality (Robinet & Feunteun, 2002).

For the past 20 years, all specialists of the European eel have noted a worrying decline in the stock in the continental and coastal hydro-systems. This extraordinary global genetic heritage is undergoing a constant decline in its stocks throughout its entire range (ICES, 2006).

Among the factors that influenced the decline and deterioration of the stock, is suspected the presence of parasitism, in particular the nematode *Anguillicola crassus*, an allochthonous parasite responsible for a high mortality rate in eel populations, in different stages of development throughout Europe (Dekker et al., 2003; Kirk, 2003; Lefebvre et al., 2003; Eelrep, 2005). It has been reported in Algeria by (Meddour et al., 1999, Loucif et al., 2009). Its presence induces serious disorders in eel growth, anatomical structure and functionality, by reducing the migration and reproductive capacity of silver eels (El-Hillali, 2007), causing severe damage to the swim bladder, resulting in a high mortality rate.

MATERIAL AND METHODS

Fish sampling

The study was carried out on a fraction of the European *An. anguilla* eel population that inhabits Lake Oubeïra. Fishing was carried out using fixed traps or livewell traps on a total of 724 individuals, with a total length and weight between 10.5 and 89.9 cm, even 17 and 1470 g. The captures were treated *in situ* on eels anesthetized with acetone in doses ranging from 6–7 ml, diluted in 10 liters of water. The level according to developmental stages, sex and size classes to detect possible interactions with eel condition (Kc) and growth.

The examination of the gaseous bladders first allowed us to assess their state of degradation (opaque, hemorrhagic, pearly, translucent bladder). The netamodes were carefully collected and removed from their hemorrhagic liquid by rinsing with distilled water, then counted and stored.

Epidemiological indices

The classic epidemiological indices determined in this study are: Prevalence (P%), Average Intensity (I), and Average Abundance (A) without those described by Bush et al. (1997).

The different values of the parasite indices were compared statistically over time, between sexes and stages of development, using Minitab software 16.

Prevalences were compared using the Chi-square and Student's t tests, and for degree of infection and parasite abundance, we used the non-parametric tests of Kruskal-Walis and Man-Whitney (W). The Fulton (1911) Kc or fitness was calculated on the entire sample. In order to assess the impact of parasitism by *A. crassus* on the growth of *An. anguilla*, we also performed a comparison of this index between two fractions of the eel population (parasitized and non-parasitized), using the student t-test for matched data.

RESULTS

The nematode *Anguillicola crassus* infests over 45% of eels. It should be noted that yellow eels are the most affected. According to the Pearson correlation test there is a positive correlation between the distribution of the number of nematodes and the total length of eels parasitized Lt ($r = 0.667$).

Change in parasite indices

The rate of infection of Lake Oubeïra eels by *A. crassus* varies between 34.21% and 61.11%. The average parasite intensity values recorded vary from one month to the next, reaching maximum of 10.17. Depending on the season, winter and summer remain marked by high values of 7.51 and 6.89 respectively. This variation is statistically confirmed, and is highly significant according to the seasons (F=0**). The monthly evolution of the prevalence in the eel population of Lake Oubeïra shows considerable rates, the highest values being 61.11%, and a minimum of 34.21%; for the rest of the seasons the values are similar with an average prevalence of 48.25%.

According to the season, the prevalence is almost equally distributed, this seasonal homogeneity is statistically non-significant (H=3.46, P=0.392). Abundance values for *A. crassus* show little monthly variations in January (4.91). Depending on the season, the summer is largely dominated with an abundance of 24.36, which remains the most marked.

The correlation between the seasons is highly significant (F=0**). Variations in intensity, abundance and prevalence are statistically insignificant (H=3; P=0.39).

Sexual variation in infection rates in Lake Oubeïra eels by *A. crassus* is more marked in males with 49.30%, compared to females 44%. The intensity values are largely dominant in males with a value of 19.25, for 2.10 in females.
Concerning the abundance between the two sexes, the values remain close with a mean of 5.30 (Fig. 1). The prevalence is very high in males with a value of 84.13%. The same applies to abundance. Despite fluctuations in epidemiological indices between the two sexes, the statistical results show no significant variation (t=0.24, p=0.816 for abundance, and t=1.14; p=0.45 for mean intensity, t=1.48; p =0.35 for prevalence).

Parasite variation according to host size and developmental stages

Analysis of the evolution of different parasite indices within the eel population sampled in Lake Oubeïra, revealed that all the size classes were infected by *A. crassus*. The variation of parasitism according to the stages of development revealed that intermediate eels showed a prevalence of 46.23%, compared to silver eels with 33.33%, which is much lower than the yellow ones with 27.78%.

On the other hand, the greatest degree of infection is observed in silver eels and sometimes reaches up to (32 nematodes/ individual). The maximum reached is (42 nematode/ individual).

Prevalence is pre-dominantly high for silver with 59.46% as well as intensity and abundance 11.55 and 4.48. These ontogenic variations were statistically tested and showed that only abundance and intensity in yellow and silver eels express heterogeneity, that remains highly significant (F**).

Size class analysis of parasitism has shown that from a length greater than 57.2 cm, the infection rate increases (Fig. 2). In addition, size classes between 60–90 cm and 40–60 cm show a high prevalence with 46.86 and 41.67 respectively, which increases according to class centers. There is a significant correlation between the number of parasites and the size class of the eels examined using Chi² test (χ^2=73.26; P=0.000).

The KC Overweight

The monthly and seasonal values of the KC condition index for the total population of the Lake Oubeïra eel show little variation. They are generally between 0.11 and 0.19 (0.02 ± 0.09 standard deviation). The monthly evolution of the overweight varies and reflects heterogeneity. A winter peak is observed with a value of (Kc= 0.41 ± 0.02). In the warm season Kc = 0.16 and represents the lowest value.

Indeed, the analysis of the relationship between the Kc index and the temperature, expresses a positive correlation r = 0.99, ie Kc increases when the temperature rises. This coefficient does not seem to vary over time and is statistically insignificant (t=0.22; P=0.89).

In order to understand if parasitism influences the overweight status of our species, we made a simple comparison between parasitized eels and healthy ones using Student's t-test, which revealed a non-significant variation (t=0.94; p=0.44), which reflects that Kc evolves normally in healthy and infected individuals (Table 1).

DISCUSSION

Monitoring of variations in epidemiological indices of *anguillicolosis* in Lake Oubeïra shows that

Eel Groups	N	Average lengths(cm)	Average weights (g)	Average kc (g/cm³)
Healthy eels	249	82.8 ± 10.5	1044 ± 17	0.28 ± 0.214
Parasitized eels	171	89.9 ± 24.4	1418 ± 28	0.19 ± 0.07

Table 1. Comparison of condition coefficients in healthy and parasitized Eels.

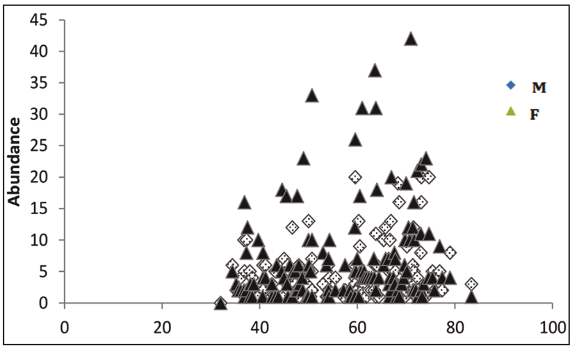

Figure 1. Variations in abundance of the nematode *Anguillicola crassus* according to the total length and sex.

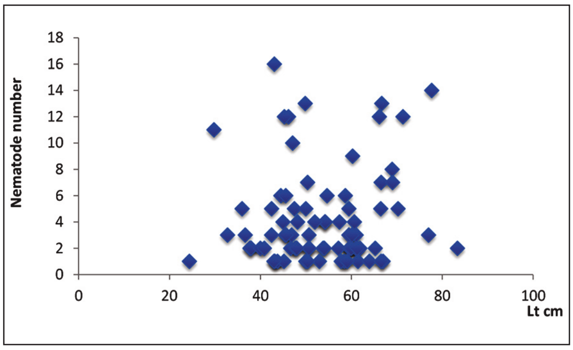

Figure 2. Distribution of parasites (*Anguillicola crassus*) according to size.

the parasite *A. crassus* is present in eels throughout the year. Paratenic host centres (of amphibians and aquatic insect larvae) are certainly present, favoring the progress of the life cycle of this parasite. The values of the lowest epidemiological indices are recorded in April and December as they oscillate to reach a maximum value in November and February 83%. The first prediction shows significant levels of prevalence (percentage of hosts in the population studied) marked in November P=61.11%, and of average intensity (average number of parasites per infected host) in January I=10.17 and of the abundance A=4.91.

Although it can be assumed that the presence of *A. crassus* was detected contemporaneously with its introduction, the parasite prevalence is often already high and above 50%. However, as prevalences increase rapidly, the average intensity level seems to reach a plateau of about 2–3 nematodes per eel, and persists over time (Ashworth & Blanc, 1997). Natural movements of infected fish as well as transfers of fish between neighboring tanks by eel fishermen probably facilitated this dispersion. This monthly variation is statistically confirmed.

Prevalence values recorded in Lake Oubeïra are high P= 61.11%, which however remain different from that reported in Morocco in the Moulouya estuary (Rahou et al., 2001), the Sebou estuary (Loukili & Belghyti, 2007) and in France, in the Rhone Delta (Levebvre et al., 2002).

Sauvaget et al. (2003) report, in southern Brittany, high prevalences in eels living in low salinity environments over 90%, compared to those living in estuarine conditions of barely 15%. The same trend is observed in 4 Tunisian lagoons by Gargouri Ben Abdallah & Maamouri (2006). Unlike the lake, the lagoons are brackish. According to some authors, salinity and calcium hardness have a negative effect on the hatching rate, nematode larval survival and infection power, but do not suppress them, even in seawater (Blanc, 1994; Kennedy & Fitch, 1990). Results obtained in the Camargue (Lefebvre et al., 2002 a, b) show high contamination in silver eels in brackish environments, but at lower salinities. There therefore seems to be a barrier to contamination by *A. crassus* for sites with high salinity. Monthly epidemiological variation may be related to thermal changes since hatching of *A. crassus* eggs occurs between 10 °C and 30 °C, with incubation time being inversely proportional to the temperature (Thomas & Ollevier, 1992).

Abstinence from feeding in the eel may have limited contagion by the parasite during these periods. Otherwise, the main active feeding phase was reported to be from October to April, with a maximum between December and February. This period represents the ideal phase for successful larval transmission available to intermediate or parenetic servers. Lagoon eels have a lower infection rate than Lake eels because the parasite life cycle takes place only in low salinity water. However, adult nematodes can survive in eels in the marine environment by "osmoconforming" with eel blood plasma (Kirk et al., 2002).

The study of the evolution of parasitism proves that monthly values reflected by abundance are stable, along the cycle with values of 2.57. Various studies have shown that larger eels are more infected than smaller ones (Audenaert et al. 2003; Schabuss et al., 2005). The first two size classes reflect a numerical equality with values of 1.20. Depending on the season, summer largely dominates over autumn, and the rest of the two seasons. Depending on the intensity, the entire population may be affected, whether large or small, as soon as it migrates to the lake.

According to Egusa (1979) the larvae *A. crassus*, ingested by the eel penetrate through digestive tract into the abdominal cavity, pass through the liver and invade the tissues of the gaseous bladder when moulting takes place, the larvae then appear in its light, for a week to a month. The same results were obtained by Haenen et al. (1989) in Europe. The lesional changes induced by *anguillicolosis* seem to be limited mainly to the gas bladder with the following consequences: haemorrage of the mucous membrane, hypertrophy and hyperhemia of the gas glands; thickening of the bladder walls, leading to a narrowing of its cavity (Molnar, 1993).

Repeated larval invasion of the bladder wall results in hemorrhage and connective tissue damage. In addition, the blood feeding the adults can cause mechanical damage to epithelium (Barse, 1999). The epithelial cells that line the swim bladder wall are subject to hypertrophy, and dysplasia. As a result, the inner surface of the bladder becomes inflamed and folded, reducing the volume of the lumen and causing loss of gas. The action of the parasite will alter the function of the swim bladder and thus reduce the eel's swimming ability at depth, which can seriously compromise their abilities for transatlantic migration at the time of the return to the Sargasso Sea (Kamstra, 1990; Boëtius I. & Boëtius J., 1980).

Concerning the analysis of our results of the overweight coefficient (K) Variations according to time, gender and silvering stage, low values were observed. Several authors have shown the existence of a positive correlation between high values of overweight and somatic growth (linear and weight) (Roche, 1983; Affandi, 1986; Berraho, 1990; El-Hilali, 1998). Low values are recorded in freshwater (Lecomte-finiger, 1983a; Mallawa, 1987), seasonally maximum values of (Kc) are noted in summer (Neveu, 1981a, b; El-Hillali, 1992), the degree of sexual maturity according to Rossi & Villani (1980), shows that the overweight is higher in females than in males, as well as development stages (El-Hillali, 2007). The same results were observed for eels from Lake Oubeïra.

The overweight of eels remains relatively low regarding to the elongated shape of the eel (Lecomte-Finiger, 1983a). In general, while the influence of *A. crassus* on growth is important in the case of massive infection (Sarti et al., 1985), it remains minor or undetectable in weakly parasitized eels (El-Hillali, 1992).

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