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Monogenean parasites in Adriatic cage-reared fish

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Monogenean parasites are the most ubiquitous and abundant parasites in the aquatic environment. In confined and stressful rearing conditions, their population easily can overpass their usual balance, proliferate and induce serious and hard-to-eradicate diseases. During a 9-month period, seven facilities along the Adriatic coast were monitored for the presence, prevalence and abundance of these parasites. Only three monogenean species - Diplectanum aequans*,* Lamellodiscus elegans *and* Sparicotyle chrysophrii - *were isolated from only three cage-reared hosts* - Dicentrarchus labrax*,* Sparus aurata *and* Diplodus puntazzo - *but without clinically visible symptoms. Parasite population dynamics showed a strong relationship with environmental factors such as salinity and temperature and marked host specificity.*

Key words: Monogenean infections, cage-reared fish, Adriatic Sea

INTRODUCTION

Monogenean trematodes are the most numerous group of parasites. Because of their primitive life cycle, rapid reproduction and prevalence in the host and geographical distributions, they are very suitable for mathematical modeling in ecology. The highest number of monogenean species has been isolated from fish populations, while the remaining species were found on amphibians and reptiles (exceptionally cephalopods and isopods). Around 95% of the monogenean species are ectoparasites on gills and skin, an archaic type of life, but a smaller proportion has exchanged their external habitats for mezzo or endoparasitism in the nasal or mouth cavity, esophagus, stomach, urinary bladder, cloaca, coelom, rectal glands, ovaries, visceral wall or heart (EUZET & COMBES, 1998).

Among the members of the monogenean group there are generalist genera with an affinity for diverse hosts while, as in case of Adriatic cage-reared fish monogeneans, there are specialist genera (DESDEVISES *et al.,* 2002a). Generally, it is accepted that specific genera have an affinity for a particular group of fish, for example Monocotylidae are specific to chondroichthyes (CHISHOLM & WHITTINGTON, 1998).

The monogenean body consists of two parts: the anterior prohaptor and the posterior opisthaptor (Fig 1).

Species are identified mainly by the morphology and accessory adhesive organs of the opisthaptor. The prohaptor also has adhesive structures such as paired or unpaired suckers, paired pseudosuckers or sucking grooves and glandular structures. The mouth is (sub)terminal,

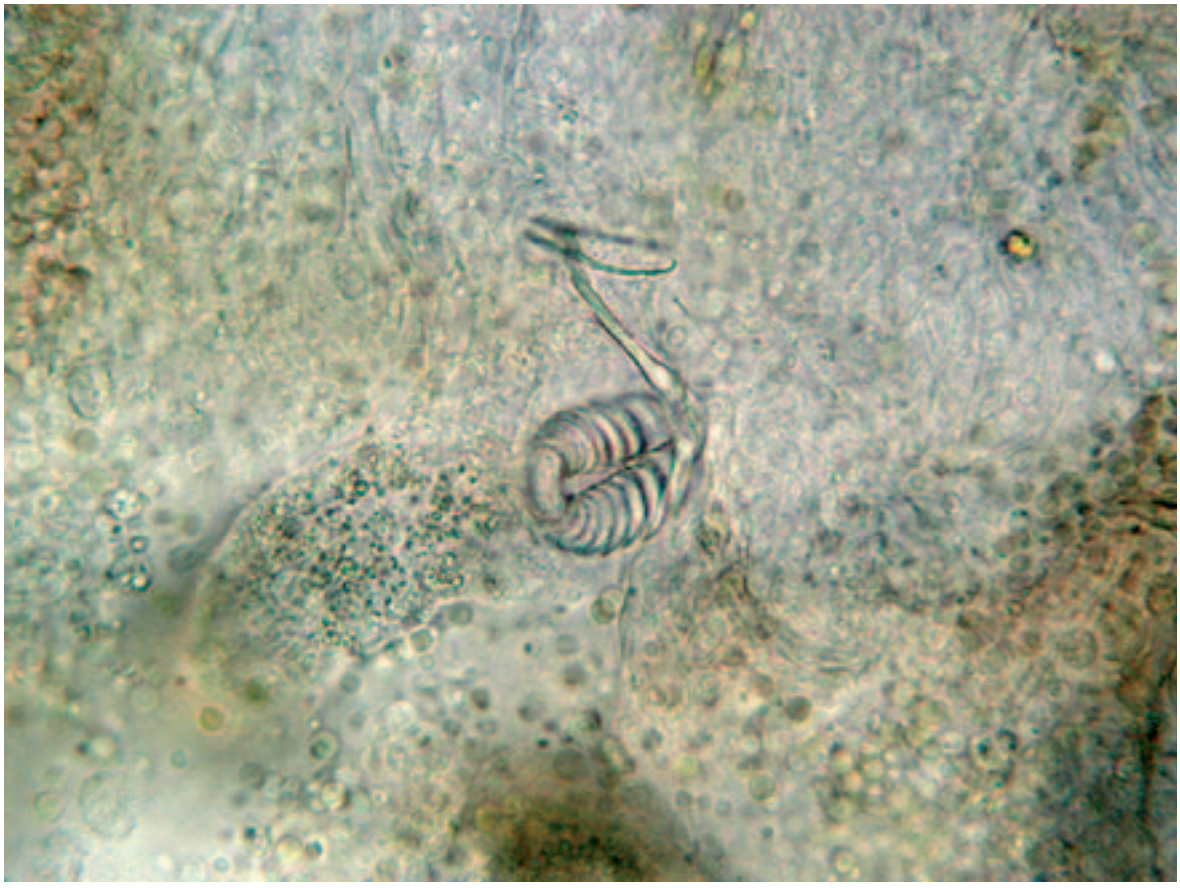

Fig. 1. A detail of lamelodisc of L. elegans *(400 x)*

an esophagus is usually present and the caeca are bifurcated.

The morphology of the reproductive system also helps identify species. Monogeneans are hermaphrodites with one or more testicles, enveloped by a curled vas deferens.

A copulatory male organ (the cirrus) can be present, simple or complex, with or without spiculi. The same is true for the genital porus. Ovaries are compact, connected to a short uterus. A receptaculum seminis is present, but there may not be a vagina. In species with a vagina, sclerotization with thorns is very usual (Fig 2).

The parasites are mostly oviparous with a direct life cycle (GELNAR, 2001).

This is the first study of the prevalence and abundance of monogeneans amongst Adriatic cage-reared fish.

MATERIAL AND METHODS

Seven fish farms in the Adriatic Sea were monitored from June 2001 to March 2002. The farms were located at: I - Kaldonta Bay (Cres Island), II - Vela Luka Bay (Šolta Island), III - Peleš Bay (near Primošten), IV - Žižanj Island, V - Žižanj Island, VI - Maslinovac Island (Pelješac peninsula) and VII - Tajan Island (Pelješac peninsula). Sampling in winter 2002 was conducted only in facilities II and III because of weather conditions.

The same fish population was followed for nine months. Fish were collected once every three months from offshore net pens, always from the same cages. Samples included 30 sea bass (*Dicentrarchus labrax*), sea bream (*Sparus aurata*), sharp-snout bream (*Diplodus puntazzo*) and red sea bream (*Pagellus bogaraveo*), all

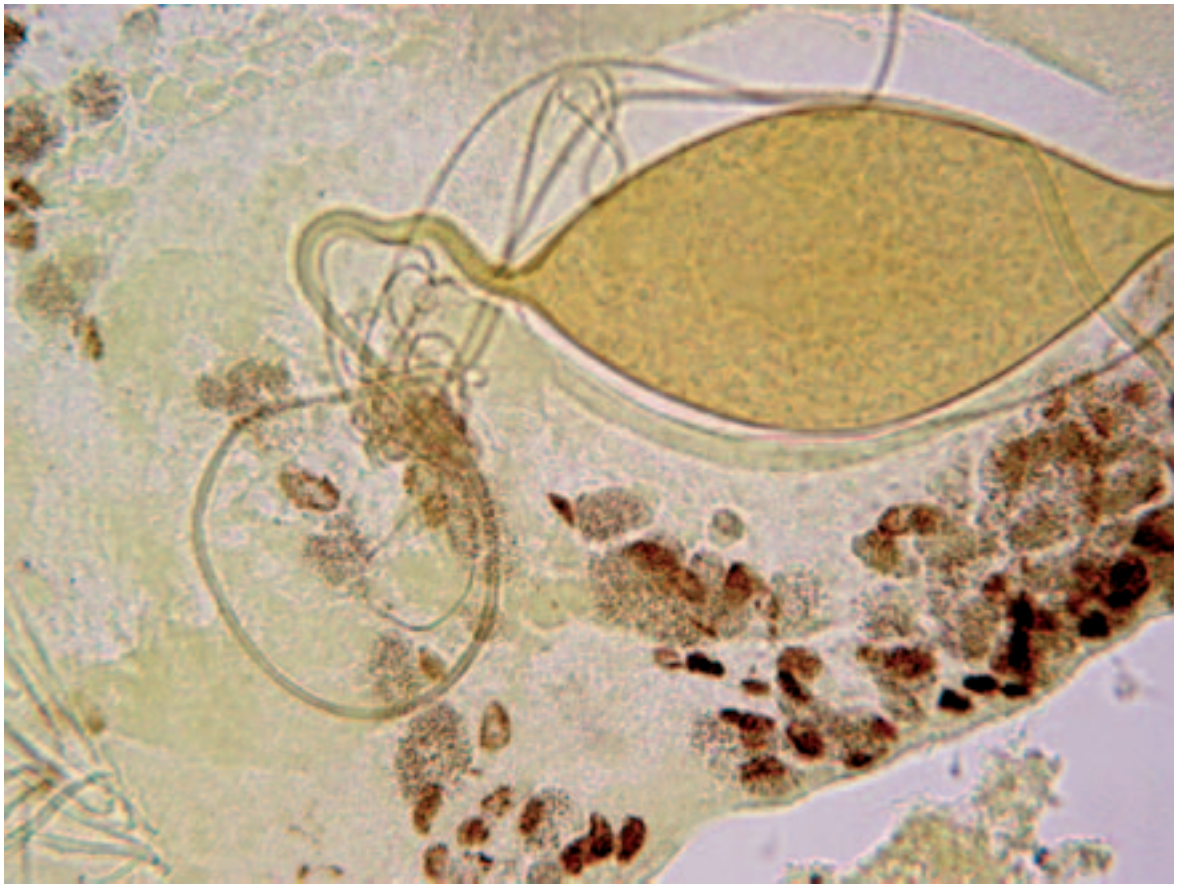

Fig. 2. An egg of S. chrysophrii *in uterus with a detail of vaginal armature (lower left corner) (400 x)*

aged over one year. During the samplings, temperature, salinity, nutrient salts and oxygen were measured.

Sampled fish were put on ice and brought to the laboratory within hours, where they were autopsied and measured. Fish gills and fins were placed in Petri dishes, while scrapings of skin and nasal cavities were mounted on slides for examination under a dissecting microscope at 20x magnification. Monogeneans were detached with dissecting needles, counted and collected in watch glasses. For fixation, the parasites were ruptured between the slide and cover slip by finger pressure and a mixture of 4% formaldehyde and glycerin (5:1) was placed on the edge of the cover slip. After evaporation of the excess fixative, the edges were sealed with DU-NOYER sealant.

For every fish and fish species from a particular facility, the prevalence and abundance were calculated according to BUSH *et al.* (1997).

RESULTS

Diplectanum aequans (Monogenea, Diplectanidae)

As one of the most ubiquitous monogeneans in cage-reared fish, *D. aequans* was found in all the monitored farms, but with marked differences in prevalence and abundance (Table 1). It showed a high specificity for sea bass and was located mostly on the first gill arch. Gross pathologies were easily noticed as hemorrhagic to yellowish necrotic areas, with subsequent depletion of changed lamellae. In fresh mounts, eggs were noticed throughout the year, being most abundant in summer. The mean prevalence

of *D. aequans* at all the facilities throughout the nine months was 62.03±23.63% with a mean abundance of 1.98±1.62.

Lamellodiscus elegans (Monogenea, Lamellodiscinae)

L. elegans showed specificity for members of the Sparidae family, especially sharp-snout bream. It was ubiquitous and present throughout the year, having the highest abundance among all the isolated monogeneans. It preferred the first gill arch and peripheral parts of the lamellae, however, in mass infections, it was distributed throughout the length of the lamella (Fig 3).

The mean prevalence of *L. elegans* was 64.95±33.13% with a mean abundance of 13.00±21.45.

Sparicotyle chrysophrii (Monogenea, Microcotylidae)

Although *S. chrysophrii* was the largest monogenean in the Adriatic cage-reared fish, it made no marked changes or gross pathologies in the host because of its low prevalence and abundance. It was ubiquitous, specific to sparids (especially the sharp-snout bream), and present throughout the year. Mixed infections with the more abundant *L. elegans* were frequent, mainly in the sharp-snout bream. *S. chrysophrii* was isolated mostly from the central part of the lamellae, with opisthaptor hooks attached to rows of neighboring lamellae (Figs. 4, 5).

The mean prevalence was 35.50±27.23% with a mean abundance of 2.44±6.86.

No monogenean species were isolated from red sea bream.

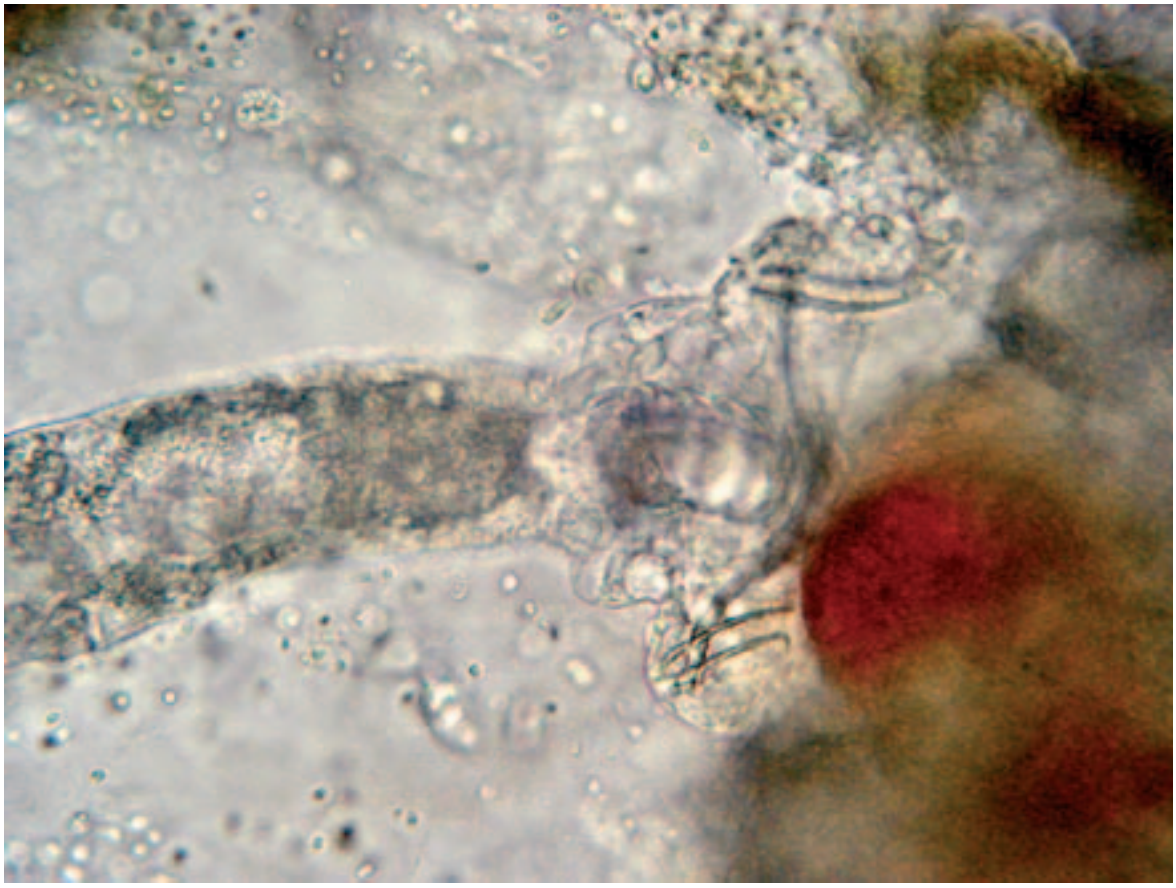

Fig. 3. Posterior part with attaching apparatus of L. elegans *(400 x) embedded in lamellar epithelia (400 x)*

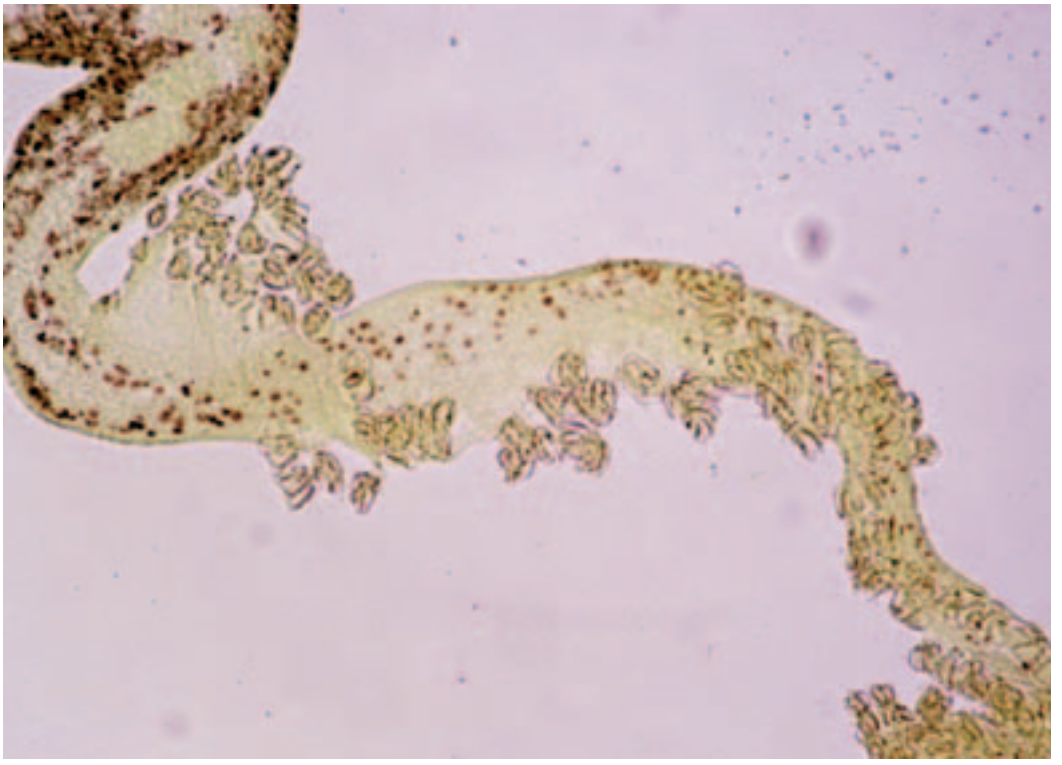

Fig. 4. Posterior part of S. chrysophrii *with attaching apparatus (100 x)*

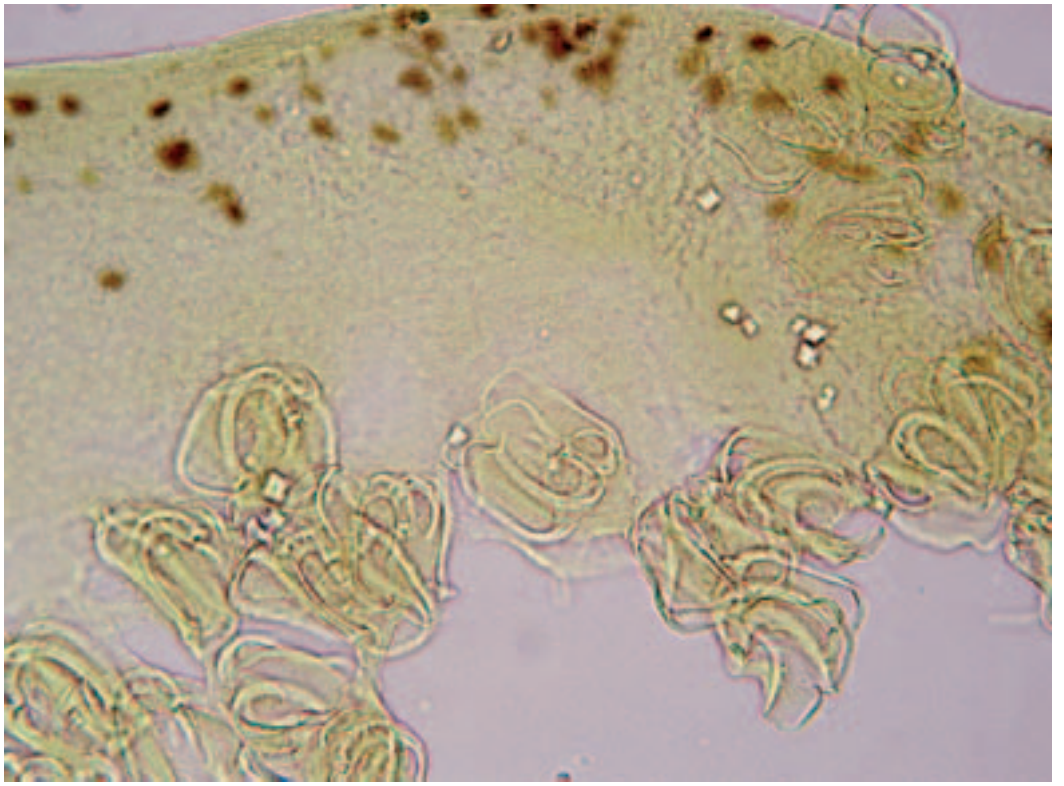

Fig. 5. Numerous hamuli of S. chrysophrii *(400 x)*

Table 1. Prevalence and abundance of monogeans in cage-raised fish in seven Adriatic facilities

Facility	Summer		Fall		Winter		Spring	
	Prevalence	Abundance	Prevalence	Abundance	Prevalence	Abundance	Prevalence	Abundance
Diplectanum aequans in *Dicentrarchus labrax*								
I	100	3.58	85.71	4.07	-	-	60	2.8
II	85.71	3.21	93.33	5.13	66.66	1	46.14	0.7
III	71.42	1.21	46.66	1.6	64.28	1	30	0.4
IV	100	4.21	78.57	4.78	-	-	66.66	1.22
V	57.14	0.86	92.85	4.29	-	-	60	1.9
VI	28.57	0.36	35.71	0.43	-	-	44.44	1.33
VII	50	0.71	42.85	0.58	-	-	20	0.2
Lamellodiscus elegans in *Sparus aurata(*)* and *Diplodus puntazzo*								
I	100	19.25	100	28.5	-	-	100; 60*	14.2; 1.2*
II	21.48*	1.71*	53.33*	1.73*	3.33*	0.3*	50*	0.9*
III	0	0	0	0	0	0	30*	0.5*
IV	100	80	100	20	-	-	85.71; 45.45*	12.14; 0.64*
V	0	0	0	0	-	-	60*	0.8*
VI	0	0	0	0	-	-	0	0
VII	0	0	0	0	-	-	0	0
Sparicotyle chrysophrii in *Sparus aurata(*)* and *Diplodus puntazzo*								
I	6.13; 16.66*	0.58; 0.25*	7.14; 42.85*	28.5; 0.5*	-	-	30	1.1
II	42.85*	0.64*	66.67*	1.33*	10*	0.1*	20*	0.2*
III	0	0	0	0	0	0	10*	0.1*
IV	100	1.67	71.42	3	-	-	14.28	0.14
V	0	0	57.14*	0.71*	-	-	30*	0.3*
VI	0	0	0	0	-	-	0	0
VII	0	0	0	0	-	--	0	0

DISCUSSION

Diplectanum aequans is an almost unavoidable parasite in sea bass cage populations, causing mortalities mainly in juvenile fish. During mass infections in summer months, mortalities complicated with *Photobacterium damselae* subsp. *piscicida* often occur (MLADINEO, 2002). In rearing conditions, it was isolated only from sea bass, while in the wild it parasitizes hosts from a wide range of fish families (HAYWARD, 1996; KRITSKY *et al.*, 2000). The only record of Diplectanidae among wild Adriatic Sea fish concerns *Sciena umbra* and *Umbrina cirrosa,* in which RADUJKOVIĆ & RAIBAUT (1989) found five Diplectanidae species.

D. aequans is greatly impacted by environmental factors, especially temperature. CECCHINI *et al.* (1998) noticed that developing oncomiracidia are preserved for quite some time at 5°C, allowing the proliferation of the parasite

in early spring when temperatures are still low and other parasites are absent (CECCHINI *et al.*, 2001). This pattern was noticed in our study, as *D. aequans* was present throughout the year. The peak season of infection was the end of summer/beginning of fall, when the temperature was around 25°C. This temperature allows the parasite to develop a numerous population in fall, which survives in winter at a lower abundance and prevalence. Salinity is another important environmental factor for *D. aequans* development. Less saline locations near river deltas (VI and VII) have considerably lower prevalence and abundance values. Although the prevalence of this monogenean was high, its abundance was not high enough (maximum 5.13) to threaten a serious disease outbreak in more or less controlled rearing conditions.

Lamellodiscus elegans has both seasonality and a strong specificity for sparid species. *L. elegans*, a member of the subfamily Lamellodiscinae, has been subjected to many studies of co-evolution trends between hosts and parasites (DESDEVISES *et al.*, 2002b). This monogenean is the most prevalent and abundant species in Adriatic cage-rearing systems while, in wild fish populations, RADUJKOVIĆ & RAIBAUT (1989) found moderate infection (22.2-50% prevalence; 4.65-19 abundance). Even when the abundance was higher than 80 parasites per host, *L. elegans* was restricted only to sparid species, especially the sharp-snout bream. DESDEVISES *et al.* (2002a) explain this as a preference of the parasite for larger hosts, in this case, the sharp-snout bream. On the other hand, CARO *et al.* (1997), although their study did not include Diplectanidae, did not find a strong correlation between trematodes and host size.

Lamellodiscus sp. does not show interspecific competency as mixed infections with monocotylid monogenean are frequent, although at a lower abundance and prevalence. Intraspecific competency also does not exist as, in extreme cases of infection, two to five individuals could be isolated at the same site. The same environmental factors that have an impact on *D. aequans* affect *L. elegans,* though *L. elegans* is more sensitive to low salinity. *L. elegans* followed the same seasonal pattern as *Diplectanum* sp.; the highest values were recorded during summer, when a major gross pathology occurred but without mortality.

Sparicotyle chrysophrii had the lowest prevalence and abundance of the three parasites but the same seasonal pattern. It was not found in facilities near freshwater in-flows and preferred sharp-snout bream. RADUJKOVIĆ & RAIBAUT (1989) isolated the parasite in the wild only from sea bream; CAFFARA *et al.* (1998) isolated it from sea bream in lagoon rearing systems.

Similar to other studies (COGNETTI VARRIALE *et al.*, 1993; OLIVA & LUQUE, 1998), all isolated species preferred the central part of the first gill arch lamellae.

Only the red sea bream had no monogenean infections even captured from the wild and kept for rearing, perhaps due to the very low density of hosts in the net pen.

CONCLUSIONS

Monogenean parasitofauna of Adriatic cage-reared fish did not show marked species richness; only three monogenean species were isolated from only three fish species. The most parasitized was the sharp-snout bream, where all sampled fish often harbored monogeneans. However, the sharp-snout bream is not widely reared in the Adriatic.

While the prevalence, abundance and host specificity differed, the three parasites had a similar and well-established pattern of population dynamics throughout the year.

Mixed infections and same microhabitat preferences were not unusual, proving intra- and interspecific tolerance of the monogeneans. Low salinity affected all three species. There have been no reports of mortality related to monogeneans, even in cases with obviously gross pathological changes on the gills. Thus, it can be concluded that the monogenean populations in this rearing system are still tolerable.

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Istraživanje jednorodnih metilja kavezno uzgojene ribe Jadranskoga mora

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SAŽETAK

Jednorodni metilji predstavljaju najrasprostranjeniju i najabundantniju skupinu nametnika u vodenom okolišu. U ograničenim i stresnim uvjetima uzgoja njihova populacija lako prelazi njihovu uobičajenu populacijsku ravnotežu, proliferirajući i inducirajući ozbiljnu bolest, tešku za iskorjenjivanje.

Tijekom devetomjesečnog razdoblja, sedam uzgajališta uz obalu Jadranskoga mora praćeno je s obzirom na prisutnost, prevalenciju i abundanciju ovih nametnika.
Samo tri vrste jednorodnih metilja - *Diplectanum aequans*, *Lamellodiscus elegans* i *Sparicotyle chrysophrii*, izolirane su s tri vrste uzgojenih ribljih domaćina - *Dicentrarchus labrax*, *Sparus aurata* i *Diplodus puntazzo*, ali bez indukcije klinički vidljivih znakova invazije.

Njihova populacijska dinamika ovisi jako o okolišnim čimbenicima kao što su slanost i temperatura te specifičnosti prema domaćinu.

Ključne riječi: Invazije jednorodnim metiljima, kavezno-uzgojena riba, Jadransko more