

## Parasites of Adriatic cage reared fish

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*With the rapid development of aquaculture in the Mediterranean, a number of parasitic diseases have emerged in cage-reared fish. In confined rearing conditions, the diseases can induce mortality and economic losses related to suppressed growth. With diversification of aquaculture products and the introduction of new fish species into the rearing system, new parasitic pathogens have found their way into new environments, resulting in adaptation of the parasite, new colonization on resident aquaculture species (primarily sea bass and sea bream), or increased parasite prevalence and abundance on the newly cultivated fish species. The parasitofauna of reared fish is impoverished in terms of species richness and has greater population values than in the wild fish population. While the parasitofauna of fish reared in the Mediterranean is discussed in numerous publications, only occasional findings specifically refer to the Adriatic Sea. Wild fish populations in the Adriatic Sea have been sampled for parasite isolation and identification but an overview of reared fish parasitofauna has never been reported. This was the main goal of this study.*

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**Key words:** sea bass, sea bream, parasitofauna, Adriatic Sea

### INTRODUCTION

There are four well-established species in Adriatic aquaculture: sea bass (*Dicentrarchus labrax*), sea bream (*Sparus aurata*), sharpsnout bream (*Diplodus puntazzo*), and red sea bream (*Pagellus bogaraveo*) with the last two comprising a minor part of the total production. Sharpsnout bream culture was devastated by severe mortalities caused by *Enteromyxum leei* (ex. *Myxidium leei*; Myxozoa: Myxosporidia) and its market appeal and value never reached those of sea bass and sea bream. Therefore, the reared population is decreasing. Red sea bream is reared only after capture from the wild and, despite having a good conversion

index, the mesenterial accumulation of adipose tissue makes the fish unsuitable for much of the market.

The most recently introduced and commercially valuable fish in Adriatic aquaculture is the northern bluefin tuna (also called the Atlantic bluefin tuna; *Thunnus thynnus*) whose pathology is still being revealed. Only recently, an excellent review of the pathology of diverse tuna species was published, with an abundant number of parasitological findings (MUNDAY *et al.*, 2003), but most of the information deals with wild fish populations from (sub)tropical waters and, therefore, does not apply to the Adriatic.

Parasites that have a low prevalence and abundance and minor pathological effects on their hosts in the wild can easily spread in populations confined to rearing systems, causing serious outbreaks of epizootic diseases (ATHANASSOPOULOU *et al.*, 1999; COMPANY *et al.*, 1999). Epidemics are optimized by the high stocking density of the host and its usually compromised immunity resulting from daily stress.

Most parasites are very hard to eradicate, especially if the therapy must be applied in semi or offshore net pens. The spectrum of chemotherapeutics is very narrow and most useful compounds are not licensed for aquaculture. The key solution lies in prevention based on optimal husbandry and zootechniques where appropriate stocking density, frequent changes of nets, and diet regulation play the biggest roles.

The scope of this report was to collect data from previous findings, add new information gathered by the author, and combine them in an overview that can be used as basic information in future studies.

## MATERIAL AND METHODS

Seven fish farms in the eastern part of the Adriatic were monitored from June 2001 to March 2002 (Fig. 1): Kaldonta Bay (Cres Island; F1), Vela Luka Bay (Šolta Island; F2), Peleš Bay (near Primošten; F3), Žižanj Island (F4), Žižanj Island (F5), Maslinovac Island (Pelješac Peninsula; F6), and Tajan Island (Pelješac Peninsula; F7).

Fish were collected every three months from offshore net pens at each facility, always from the same cage, for nine months. Samples comprised 9-15 sea bass (*Dicentrarchus labrax*), sea bream (*Sparus aurata*), sharpsnout bream (*Diplodus puntazzo*), and red sea bream (*Pagellus bogaraveo*) of at least one year. In total, 673 individuals were examined.

Collected fish were put on ice and brought to the laboratory within hours where they were autopsied and biometrical measures were recorded. Fresh smears were taken from the gills, skin, and fins, from three parts of the alimentary



Fig. 1. Map of the Adriatic Sea with locations of sampled facilities

duct (pyloric area, middle intestine, and rectal part), and from the spleen, liver, gonads, and kidney. If positive, smears were stained by May-Grünwald Giemsa. Myxosporeans were measured and identified according to LOM & ARTHUR (1989).

Gill monogeneans were counted on the middle third of the first gill arch. Cut 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 with 20x magnification. Monogeneans were detached with dissecting needles, counted, and collected in a watch glass. For fixation, parasites were ruptured between the slide and coverslip by finger pressure and a mixture of 4% formal-dehyde and glycerin (5:1) was added to the edge of a coverslip. After evaporation of the remaining fixative, edges were sealed with DU-NOYER sealant.

In addition, 62 northern bluefin tuna were sampled at an eighth facility on the northwestern

part of Brač Island during harvest in January 2003 and when daily mortality occurred in July 2003. Prior to rearing, these tuna had been caught in May 2002 in the waters near Jabuka Island and transported to the farm where they were fed mixed fish and frozen imported herrings from small boats.

Prior to washing the fish trunks, the body surface was inspected for the presence of any changes. Because of the value of the carcasses, no incisions on the fins, skin, or eyes were made. Visceral organs and gill arches were collected. Viscera samples were individually collected in plastic bags and transported to the laboratory. Blood samples were collected before evisceration from the incision made beneath the pectoral fins and through the heart. Sodium citrate was used as an anticoagulant.

Upon arrival in the laboratory, fresh smears of gills, kidney, spleen, liver, gall bladder, intestinal and stomach mucosa, and endocardial tissue from the ventriculus were examined under a light microscope. Digenean cysts from gills, cartilaginous parts of gill arches, stomach layers, pyloric ceca, skin, and intestine were collected, measured, and excysted with fine needles under a stereomicroscope. Individuals were stained in Borax carmine, mounted in Canada balsam, and fixed under a coverslip. Remaining cysts were collected and fixed in 70% alcohol. Myxosporidians and helminthes were collected as in the other cage reared fish.

## RESULTS

Parasites isolated from fish sampled at eight cage facilities are shown in Table 1.

Table 1. Parasites in cage reared fish in the Adriatic Sea

Parasite	Host	Parasitic form	Host status	Infection site	Facility	Mean prevalence (%)	Mean abundance
<b>MASTIGOPHORA</b>							
<i>Amyloodinium ocellatum</i>	sea bass	trophont	no signs	gill	1, 2, 4-7	30.84	0.38
	sea bream	trophont	no signs	gill	1, 2, 4-6	74.67	1.05
	sharpnose bream	trophont	no signs	gill	1, 4	73.57	1.13
	red sea bream	trophont	no signs	gill	3	20	0.2
<b>CILIOPHORA</b>							
<i>Cryptocaryon irritans</i>	sea bass	teront	no signs	gill	2	13.33	0.13
	sea bream	teront	no signs	gill	2	60	1.8
<i>Trichodina</i> sp.	sea bream	adult	no signs	gill	5	10	0.1
<b>MYXOZOA</b>							
<i>Ceratomyxa sparusaurati</i>	sea bass	spore, disporoblast	no signs	gall bladder	2	6.66	0.33
	sea bream	spore, disporoblast	no signs	gall bladder	1, 4	14.55	0.48
	sharpnose bream	spore, disporoblast	no signs	gall bladder	1	30	0.9
	red sea bream	spore, disporoblast	no signs	gall bladder	3	21.55	0.49
<i>C. thunni</i>	tuna	spore, disporoblast	no signs	gall bladder	8	23.33	1.9
<i>Sphaerospora dicentrarchi</i>	sea bass	spore, disporoblast	no signs	intestine, gall bladder	1-7	54.13	1.04
<i>Polysporoplasma sparisi</i>	sea bass	spore, disporoblast	no signs	kidney	6, 7	33.93	0.68
	sea bream	spore, disporoblast	no signs	kidney	1, 2, 4, 5	42.02	0.76
	sharpnose bream	spore, disporoblast	no signs	kidney	1	20	0.6
<i>Myxobolus</i> sp.	sharpnose bream	spore, disporoblast	no signs	intestine	1	10	0.1

Table 1. Cont'd

Parasite	Host	Parasitic form	Host status	Infection site	Facility	Mean prevalence (%)	Mean abundance
<b>MONOGENEA</b>							
<i>Diplectanum aequans</i>	sea bass	adult, eggs	gill necrosis	gill	1-7	62.03	2.07
<i>Lamellodiscus elegans</i>	sea bream	adult, eggs	no signs	gill	1-5	40.06	0.97
	sharpshout bream	adult, eggs	no signs	gill	1, 3, 4	97.62	29.01
<i>Sparicotyle chrisophrii</i>	sea bream	adult, eggs	no signs	gill	1-3, 5	33.9	0.46
	sharpshout bream	adult, eggs	no signs	gill	1, 4, 5	32.21	1.09
<b>DIGENEA</b>							
<i>Cardicola forsteri</i>	tuna	eggs	inflammation	gill, heart, kidney	8	63.34	14
<i>Coeliodidymocystis abdominalis</i>	tuna	encysted adult	no signs	among pyloric ceca	8	63.16	3.79
<i>Didymocystis wedli</i>	tuna	encysted adult	no signs	gill	8	73.68	13.26
<i>Platocystis alalongae</i>	tuna	encysted adult	no signs	skin	8	21.05	0.95
<i>Koellikerioides internogastricus</i>	tuna	encysted adult	no signs	stomach layers	8	21.05	0.32
<i>K. apicalis</i>	tuna	encysted adult	no signs	cartilage	8	73.68	6.79
<i>K. intestinalis</i>	tuna	encysted adult	no signs	intestinal mucosa	8	57.89	12.47
<b>NEMATODA</b>							
<i>Hysterothylacium aduncum</i>	red sea bream	third stage larvae	no signs	submesentery	1	12.5	0.11
<i>Anisakis simplex</i>	tuna	third stage larvae	no signs	submesentery	8	11.5	0.15
<i>Oncophora melanocephala</i>	tuna	adult	hemorrhages	pyloric ceca mucosa	8	57.89	1.74
<b>CESTODA</b>							
<i>Hepatoxylon trichiuri</i>	tuna	plerocercoid	hemorrhages	stomach mucosa	8	12.4	0.12
<b>COPEPODA</b>							
<i>Caligus minimus</i>	tuna	adult	no signs	gill	1	8.57	0.09
<b>ISOPODA</b>							
<i>Ceratothoa oestroides</i>	sea bream	adult	emaciation	buccal cavity	2.4	8.89	0.15
		adult	emaciation	buccal cavity	2	30.24	0.41

## DISCUSSION AND CONCLUSIONS

There are a number of excellent and detailed reviews of parasites in cultured and feral fish (CHRISTOFILOGIANNIS, 1992; RODGERS & FURONES, 1998; LE BRETON, 1999; SCHOLZ, 1999; MUNDAY *et al.*, 2003). The parasitofauna of reared fish differs in different regions and a complete review of data from the Adriatic are lacking.

In our study, in sea bass, the monogenean *Diplectanum aequans* was the most prevalent and most abundant parasite; *Sphaerospora dicentrarchii* was second. The parasite with the lowest prevalence was *Ceratomyxa* sp. In the first review of diseases of sea bass

reared in the Adriatic Sea, ŠARUŠIĆ (1990) reports only four pathogens: *Trichodina* sp., *Amyloodinium ocellatum*, *D. aequans*, and *Caligus minimus*, noting that *A. ocellatum* is the most pathogenic, especially for sea bass larvae in which mortality can reach 90%. PAPERNA & BAUDIN LAURENCIN (1979) reported a similar composition of parasitofauna in French facilities with the addition of *Colponema* sp. in sea bass gills that was not reported in the Adriatic review. Their report of *Myxidium* sp. in sea bass is interesting as it, together with the occurrence of this species in the Adriatic sharpshout bream, is very rare. Wild populations have a much greater

parasitofaunal richness than reared sea bass. The most common species are the same as in rearing systems: *D. aequans* and *S. dicentrarchii* (SANTOS, 1993).

In the Adriatic sea bream, the most prevalent species was *A. ocellatum* and the most abundant was *Cryptocaryon irritans*. The species with the lowest prevalence was *Trichodina* sp. PAPERNA & BAUDIN LAURENCIN (1979) reported finding only *Trichodina*, *A. ocellatum*, and *Colponema* sp. in specimens from France, roughly corresponding to the gill parasitofauna of Adriatic fish. A much greater number of parasite species was isolated from sea bream in a range of culture systems in Spain (ALVAREZ-PELLITERO *et al.*, 1995), and the report differs from the Adriatic only by the presence of coccidian (*Eimeria* sp.) and myxosporidian species (*Leptotheca* sp.).

In sharpsnout bream, the monogenean *Lamellodiscus elegans* was the most prevalent and abundant parasite and *Caligus minimus* was the least. After increases in sharpsnout bream production in the Adriatic during the past decade, there are now only two farms that maintain sharpsnout bream rearing systems. The main reason for this decline was the devastation of most stocks by an *E. leei* (Myxozoa) infection, but a second reason was the low appreciation and value of sharpsnout bream on the Croatian market. Most sharpsnout bream are produced in Greece and reports list only two myxosporidia (*Myxidium leei*, *Ceratomyxa* sp.), a coccidian (*Eimeria* sp.), and two monogeneans (*Lamellodiscus* sp., *Sparicotyle* sp.) as regular parasitofauna (COMPANY *et al.*, 1999). However, the total parasite load is greater in Adriatic sharpsnout bream than in the other sampled species, except for tuna.

Red sea bream is reared at only two of the monitored facilities. Its parasitofauna comprised the major parasites found in the other two sampled sparids, i.e., *A. ocellatum* and *C. sparusaurati*, and *Hysterothylacium aduncum* which is ubiquitous in wild populations.

In Adriatic conditions there is evidence of host switching and exchange of parasites between sparid hosts (sea bream, sharpsnout bream, and red sea bream; MLADINEO & MARŠIĆ-LUČIĆ, 2006). The majority of parasites present in sea bream can be found in sharpsnout bream with different prevalence or abundance values. These species are never maintained in monoculture so the passage of the parasites is eased by high stock density of mixed species.

Sampled tuna were host to a group of *Didymocystis* parasites, reported for the first time in the Adriatic Sea, and a newly isolated myxosporidian (MLADINEO & BOČINA, 2006). Although *Didymocystis* has high prevalence and abundance values, no gross pathological changes were noticed. The only potential threat is infection with *Oncophora melanocephala* sp.

In conclusion, sea bass parasitofauna in the Adriatic is similar to earlier reports while sparid parasitofauna differs in the diversity of myxosporidians and the presence of coccidian species. Tuna parasitofauna is unique in the diversity of digenean species, characteristic of populations where the food web makes possible the accumulation of a large number of intermediate hosts, a situation that does not exist amongst hatchery-raised fish. Research into tuna diseases is young and developing. Therefore, there are no earlier and appropriate studies carried out in the Mediterranean with which the results of the current study can be compared.

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## Nametnici na kavezno uzgojenoj ribi u Jadranu

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### SAŽETAK

Brzim razvojem marikulture na Mediteranu, pojavile su se brojne bolesti uzrokovane nametnicima na kavezno uzgojenoj ribi. U ograničenim uzgojnim uvjetima, bolesti mogu prouzročiti mortalitet i ekonomske gubitke zbog usporenog rasta. Uvođenjem novih proizvoda u marikulturi, te novih ribljih vrsta u uzgojnom sistemu, novi nametnički patogeni su pronašli svoj put u novim okruženjima adaptirajući se i kolonizirajući tako novo uzgojene vrste (pretežito lubina i arbuna) ili su povećali rasprostranjenost nametnika na novo uvedenim vrstama u uzgoju. Parazitofauna uzgojene ribe je osiromašena sa stanovišta bogatstva vrsta i ima veće populacijske vrijednosti nego parazitofauna divlje ribe. Iako je parazitofauna uzgojene ribe u Mediteranu istraživana u brojnim publikacijama, tek povremeni nalazi se odnose na Jadransko more.

Populacija divlje ribe u Jadanskom moru je bila uzorkovana za izolaciju i identifikaciju nametnika, ali pregled parazitofaune uzgojene ribe do sada nije objavljen. Ovo je bio glavni cilj istraživanja.

**Ključne riječi:** lubin, komarča, parazitofauna, Jadransko more