

EVALUATION OF THE PARASITIC DEGREE AND THE EFFECTS CAUSED ON THE COMMERCIAL FISH STOCKS AT THE ROMANIAN BLACK SEA COAST

Aurelia Totoiu¹, Neculai Patriche²

¹National Institute for Marine Research and Development "Grigore Antipa" Constanta,
300 Mamaia Blvd, 900581, Constanta, Romania, e-mail: atotoiu@alpha.rmri.ro, atotoiu@yahoo.com

²Institute of Research and Development for Aquatic Ecology, Fishing and Aquaculture, Galati

Abstract

This paper presents the results of the parasitological research on parasite infestation at the main fish species of commercial interest on the Romanian Black Sea coast: *Sprattus sprattus* - sprat, *Engraulis encrasicolus* - anchovy, *Trachurus mediteraneus ponticus* - horse mackerel, *Psetta maxima maeotica* - turbot.

The investigations were carried out between 2016 and 2017, on fish collected from Sulina to Vama Veche. The following species of ectoparasites were identified: the ciliate - *Trichodina domerguei*, and the flat worm - *Mazocraes alose* and the endoparasites: *Contracaecum aduncum*, *Contracaecum* sp., *Anisakis* sp., *Porrocaecum* sp. and the flat worm *Botriocephalus scorpii* were found.

Ectoparasites, including those outside the body affecting fish species, especially the substrate of skin, fins, gills and eyes were rather low in number. *Trichodinina domerguei* affected fish outside the body, fins and gills and *Mazocraes alose* affected only the gills.

Specific analysis made to evaluate the effects of nematode worm parasites on the status of fish populations revealed high values of prevalence (percentage of infested fish), mean infestation intensity and abundance of parasites being higher in larger specimens compared to the smaller ones. Thus, for sprat, the larger specimens, 10-11 cm in length, reached 80% and anchovy 11-12 cm length reached 100%, while the specimens under these lengths were 40-70% infested.

The flat worm *Botriocephalus scorpii*, found in turbot stomach in very large numbers, caused severe lesions, especially inflammations, wounds, desquamation of the damaged epithelium. Toxic substances released by the worms are absorbed and produce whole body intoxication.

The presence of these parasites in natural fish populations may represent a real danger to the state of existing stocks, but also to their evolution.

Keywords: parasites, parasite intensity, prevalence, commercial species, Romanian Black Sea coast

1. INTRODUCTION

Fish, as well as other aquatic and terrestrial animals, may be affected by infectious diseases and parasite infestations. Parasitic diseases are caused by animal parasites - protozoans, worms and crustaceans.

Nematode worms are a large group of parasites, being mostly encountered in marine fish. Some nematode species infest fish as adults, while others as larvae; for the latter, the final hosts are various fish-eating vertebrates (predator fish, amphibians, reptiles, birds, mammals). Fish are either the intermediate host of nematodes, or the secondary and accumulation host [1].

Nematodes are spread by fish carrying such parasites through their feeding and spawning migrations, through intermediate hosts, water (in which parasites often survive and breed), as well as through direct contact [2].

The occurrence of nematode infestations is also determined by environmental conditions which may act both on the worms and the hosts, large agglomerations, food and fish migrations [3,4,5].

There is a tight connection between parasites and host, based mainly on the fact that parasites cause the increase of fatalities in hosts [6,7]. There are, however, cases in which a balance sets between parasites and hosts, but it depends on the intensity of parasite infestation, immunity of the hosts to parasites and the general living conditions of the hosts.

Parasites can affect the entire body surface, the gills, the nose cavities, the digestive tube, the urinary, blood and breeding system, as well as fish muscles. The influence of parasites on the host body is extremely varied, from mechanical lesions and the tonic influence given by the decrease of growth rate and lack of appetite to severe lesions, destruction of the affected organs and tissues and death of fish. Parasites are a significant limiting factor in fish growth and, even if parasitosis do not always cause fatalities, they infer deep morphological and physiological changes, mainly by decreasing the resilience of fish organisms to various environmental factors, by delayed growth or drop in prolificity [8,9,10,12]

Fish, like all living organisms, have a very close relationship with the environment they live in. The stock state of the main commercial fish species depends on the industrial fishing activity, but at the same time on the health status of the fish population. By the disease state we understand a complex of phenomena and organic manifestations in interrelation with a pathogen or more, from the moment of contact and until the consequences disappear.

2. EXPERIMENTAL

For the identification of parasitic diseases, the biological material consisted of fish collected along the Romanian coast, from Sulina to Vama Veche, during 2016 - 2017.

For assessing the parasitosis, macroscopic and microscopic examinations were carried out on the fish, with the aim of identifying the parasite and the reactions they can cause on the hosts (Fig. 1) [11].

The macroscopic examination was made by naked eye, on the oral cavity, gills and abdominal cavity, where each organ was studied to emphasize potential necrosed areas, cysts, parasites, color changes and other modifications visible by naked eye.

The parasites were viewed with a 10 lens and the 5 and 10 oculars. For the microscopic examination, full preparations were used (small portions of tissues and organs), as well as crushed preparations (squashes) between the blade and the lamella, making the film formed translucent and as thin as possible, allowing the sighting of potential parasites.

The fish parasite infestation degree was determined by the intensity values (number of parasites/host) and parasite infestation extension (number of infested fish), for each fish sample and the overall samples analyzed.

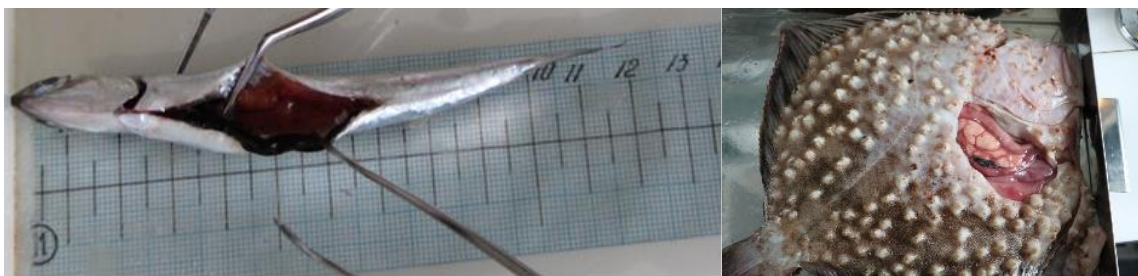


Fig. 1. Dissected fish

3. RESULTS AND DISCUSSION

Parasitological research on parasite infestation were made at the main fish species of commercial interest from the Romanian Black Sea coast: *Sprattus sprattus* - sprat, *Engraulis encrasicolus* - anchovy, *Trachurus mediteraneus ponticus* - horse mackerel, *Psetta maxima maeotica* - turbot, *Platichthys flesus luscus* - flounder.

Ectoparasites, including those outside the body, affecting fish species, especially the substrate of skin, fins, gills and eyes were rather low in number.

Trichodinina domerguei affected fish outside the body, fins and gills, and *Mazocraes alose* affected only the gills.

The ciliate *Trichodina domerguei*, found at turbot and horse mackerel, affected the fish outside the body, at fins and gills, causing skin erosion, punctures, ulcerations in the gills, inflammation, destruction of the gill respiratory epithelium and abundance of mucus (Fig. 2).

Mazocraes alose was found in low number in sprat, fixed on the gills, producing irritations, breathing disorders, fish reacting through a rich mucus secretion and epithelial hyperplasia. (Fig. 3). The highest parasitic degree was reported in turbot, followed by horse mackerel and sprat (Table 1).

Table 1. The degree of parasite of fish

Analyzed fish/No. of individuals	Parasite species	Invasion extension % infested fish			Invasion intensity No. of parasites/host		
		Min.	Max.	Max.	Min.	Max.	Med.
<i>Sprattus sprattus</i>	<i>Mazocraes alose</i>	0	20	10	2	10	6
<i>Psetta maxima</i>	<i>Trichodina domerguei</i>	0	70	25	12	100	40
<i>Trachurus mediterraneus ponticus</i>	<i>Trichodina domerguei</i>	0	40	20	0	Freq.	Rare

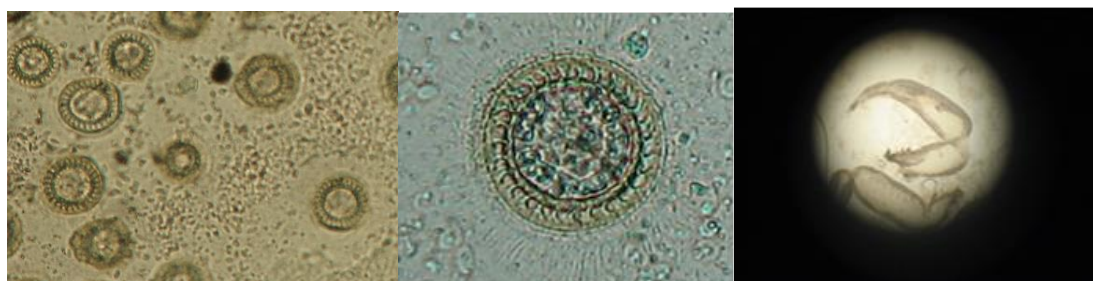


Fig. 2. *Trichodina domerguei*

Fig. 3. *Mazocraes alose*

The endoparasites encountered in the analyzed fish body were numerous, both in number of species and number of specimens/host.

Bothriocephalus scorpii is a flat worm, belonging to the Cestoda class, gray, slightly transparent, segmented in the form of a ribbon, 50-950 mm long and 1.3-6 mm wide. It has an elongated scolex body and a strobila with several protrusions that are narrowed and widened to the posterior. The worm was identified in turbot, the parasitosis being known as botriocephalosis (Fig. 4).

The parasite was reported in turbot from all the studied areas, most affected being the fish in the southern Romanian seaside, where the digestive tube, completely lacking food, was full of parasites. Being fixed in the intestinal mucosa of the fish, the parasites act mechanically causing damage and inflammation, degeneration, necrosis, conjunctive tissue proliferation. Toxic substances released by the worms are absorbed and produce whole body intoxication.

The prognosis is quite serious, with all large fish being affected by this parasite. Since the number of parasites is so high that it occupies the entire digestive tract of fish, as evidence being almost all the analyzed fish, there are serious nutritional disturbances, bloody lesions in the intestinal wall, intoxication and sometimes leading to mortalities in the fish population.



Fig. 4. *Botriocephalus scorpii* identified in turbot from the Romanian Black Sea area

Evaluation of parasites influence on natural fish stocks

Simulation studies have highlighted the technical difficulties that occur in establishing clear evidence of parasite-induced host mortality through ecological studies of hosts and parasites in their natural habitats.

By analyzing fish parasites and their parasitic extent, an assessment can be made to evaluate the influence of nematodes worms, the main parasites identified at investigated fish.

By fish dissections, nematodes, especially larvae were identified together with the effects they generate on affected organs and tissues (Table 2).

Contraecaecum aduncum parasites the viscera, being encapsulated or free in the abdominal cavity, intestine, pyloric appendage and fish liver. With the aging of the fish, there is an accumulation of larvae, which is why they can reach up to several hundred/fish (Fig. 5).

Porocaecum sp. larvae are located in the muscles near the abdominal cavity of the fish, the number of parasites increasing with the fish size (Fig. 6).

Anisakis sp. larvae, immobile nematode worms, parasite encapsulated in the liver, pyloric appendages and/or mobile, migrating into various organs of the body, muscles, gonads (Fig. 7).

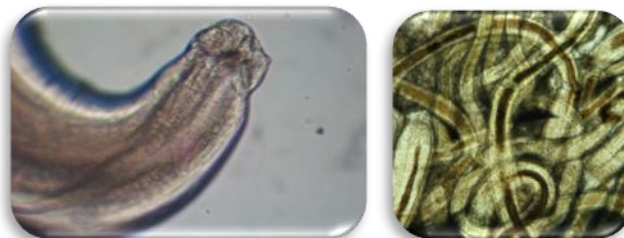


Fig.5. *Contraecaecum aduncum*

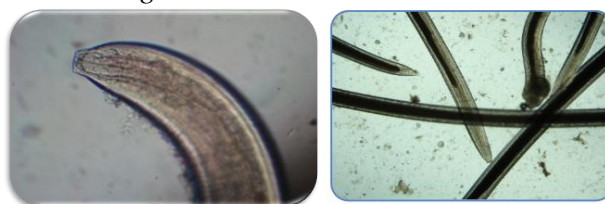


Fig.6. *Porocaecum* sp.

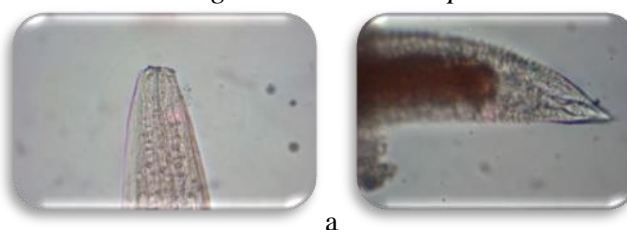


Fig.7. *Anisakis* sp. (a - front side; b - rear side)

Table 2 .The degree of infestation with endoparasites of fish in the Romanian marine area

Analyzed fish species /No. individuals	Parasite species	Invasion extension % infested fish			Invasion intensity No. of parasites/host		
		Min.	Max.	Med.	Min.	Max.	Med.
VAMA VECHE							
Sprat / 40	<i>Contracaecum sp.</i>	20	70	30	0	9	2
	<i>Porrocaecum sp.</i>	0	50	25	0	12	6
	<i>Anisakis sp.</i>	0	20	10	0	5	2
Anchovy / 30	<i>Contracaecum sp.</i>	50	70	60	3	12	5
	<i>Porrocaecum sp.</i>	40	70	55	7	20	8
	<i>Anisakis sp.</i>	0	60	25	0	5	1
Horse mackerel / 25	<i>Contracaecum sp.</i>	10	50	20	2	6	3
	<i>Porrocaecum sp.</i>	20	60	40	4	8	6
COSTINESTI							
Sprat / 40	<i>Contracaecum aduncum</i>	0	20	10	0	3	2
Anchovy / 30	<i>Contracaecum sp.</i>	0	70	15	0	7	3
	<i>Porrocaecum sp.</i>	20	60	30	5	15	10
	<i>Anisakis sp.</i>	0	30	5	0	5	2
Horse mackerel /30	<i>Contracaecum sp.</i>	0	20	10	0	5	2
	<i>Anisakis sp.</i>	0	50	20	0	6	1
EFORIE SOUTH							
Sprat / 40	<i>Contracaecum aduncum</i>	0	30	20	0	7	4
	<i>Porrocaecum sp.</i>	20	50	30	2	20	8
Anchovy / 40	<i>Contracaecum sp.</i>	40	70	55	2	14	5
	<i>Porrocaecum sp.</i>	40	70	55	4	12	4
	<i>Anisakis sp.</i>	0	70	20	0	6	1
Horse mackerel /32	<i>Contracaecum sp.</i>	0	50	20	0	5	2
	<i>Anisakis sp.</i>	0	50	30	0	8	2
AGIGEA							
Sprat / 40	<i>Contracaecum aduncum</i>	0	30	20	0	7	4
	<i>Porrocaecum sp.</i>	20	40	30	2	9	6
	<i>Anisakis sp.</i>	0	20	5	0	3	0
Anchovy / 35	<i>Contracaecum sp.</i>	20	50	30	2	14	5
	<i>Porrocaecum sp.</i>	40	30	10	4	12	4
	<i>Anisakis sp.</i>	0	20	5	0	6	1
Horse mackerel /27	<i>Contracaecum sp.</i>	0	50	20	0	5	2
	<i>Porrocaecum sp.</i>	0	50	30	0	8	4
NAVODARI							
Sprat / 40	<i>Contracaecum aduncum</i>	0	20	6	0	4	2
	<i>Contracaecum sp.</i>	20	60	40	2	12	6
	<i>Porrocaecum sp.</i>	10	60	30	2	8	4
	<i>Anisakis sp.</i>	0	10	4	1	4	2
Anchovy / 30	<i>Contracaecum sp.</i>	40	70	55	6	20	4
	<i>Porrocaecum sp.</i>	20	80	40	2	10	4
	<i>Anisakis sp.</i>	0	20	8	1	4	2
Horse mackerel /25	<i>Contracaecum sp.</i>	10	30	20	0	5	2
	<i>Porrocaecum sp.</i>	0	30	10	2	6	3
VADU							
Sprat / 40	<i>Contracaecum aduncum</i>	0	20	6	0	4	2
	<i>Contracaecum sp.</i>	10	60	40	2	12	6
	<i>Porrocaecum sp.</i>	0	60	30	2	8	4
	<i>Anisakis sp.</i>	0	10	4	1	4	2
Anchovy / 40	<i>Contracaecum sp.</i>	0	70	55	6	20	4
	<i>Porrocaecum sp.</i>	20	80	40	2	10	4
	<i>Anisakis sp.</i>	0	20	8	1	4	2
Horse mackerel /33	<i>Contracaecum sp.</i>	10	40	20	0	6	1
	<i>Porrocaecum sp.</i>	10	30	20	2	8	3

At Vama Veche, the main species caught (sprat, anchovies, horse-mackerel) were infested by nematodes of the genera *Contracaecum* and *Porrocaecum*, in intensities of up to 20 parasites/host and extensions of 60-70% infested fish. For sprat and anchovies, *Anisakis* sp. nematode was also reported in an invasion intensity that did not exceed 5 parasites/hosts.

In Costinesti, the most infested fish species was anchovy, which was parasited by three nematode species. For sprat and horse mackerel, the infestation degree was lower.

The infestation rate in the Eforie South station reached intensity levels of up to 20 parasites/hosts and extensions of 55-70% infested fish. Both anchovies and sprats have shown a high parasitic degree.

At Agigea, the three analyzed fish species showed different parasitic degrees, for sprat the maximum parasitic intensity was 9 parasites/host, anchovy reached the maximum parasitic intensity of 14 parasites/host, and in horse mackerel it was 8 parasites/host.

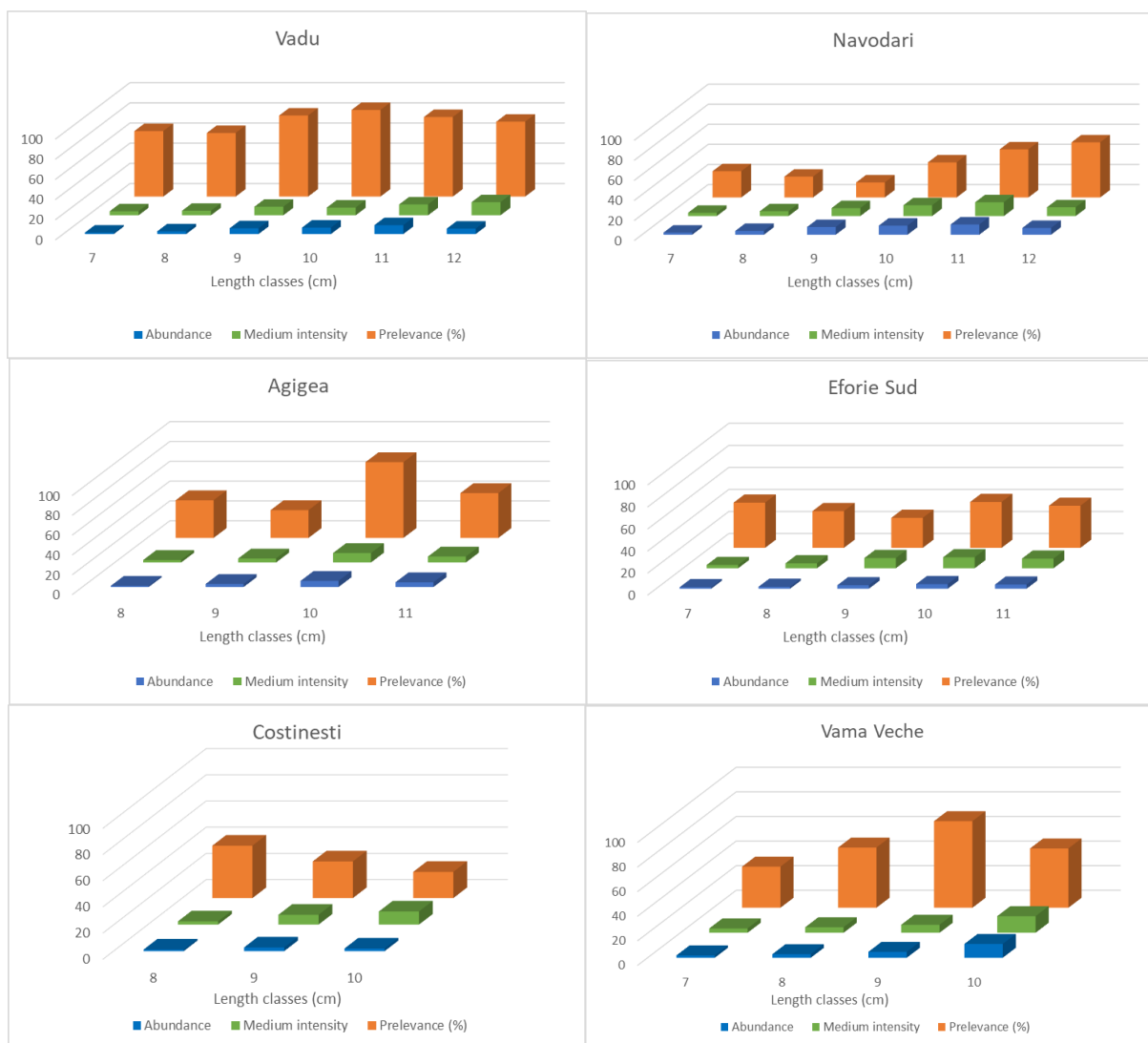


Fig. 8. Parasite infestation degree of sprat on length classes along the Romanian coast

The analyzed sprat samples from Navodari - Vadu marine area showed a high parasitic degree, four species of parasites (*Contracaecum aduncum*, *Contracaecum* sp., *Porrocaecum* sp., *Anisakis* sp.) being identified, with extensions of up to 60% infested fish. The anchovy analyzed from this area exhibited extensions of up to 80% infested fish. The horse mackerel in this area presented the extensions of 30-40%, infested fish (Table 2).

Therefore, the analyzed species were determined for at least 50 specimens for sprat and anchovies. The analyzed items included:

- prevalence - the percentage of infested fish;
- average intensity - average number of parasites/host infested;
- abundance - average number of parasites / total fish analyzed - infested and uninfested.

All investigations were made for each fish of the related length class in order to obtain the closest appreciation of the parasitic dominance and the effects of the infestations on the fish, as compared to their size. Sprat was one of the marine species mostly affected by nematodes, the intensity of infestation varied from one station to another. From the six analyzed areas, the stations where the fish had a larger size range (between 7 cm and 12 cm) were in Vadu and Navodari.

The most parasitic specimens were in the Vadu station, the prevalence having values, ranging from 63% -86%, the average intensity being of 10.8 parasites/host, and the abundance of 8.8 parasites / fish.

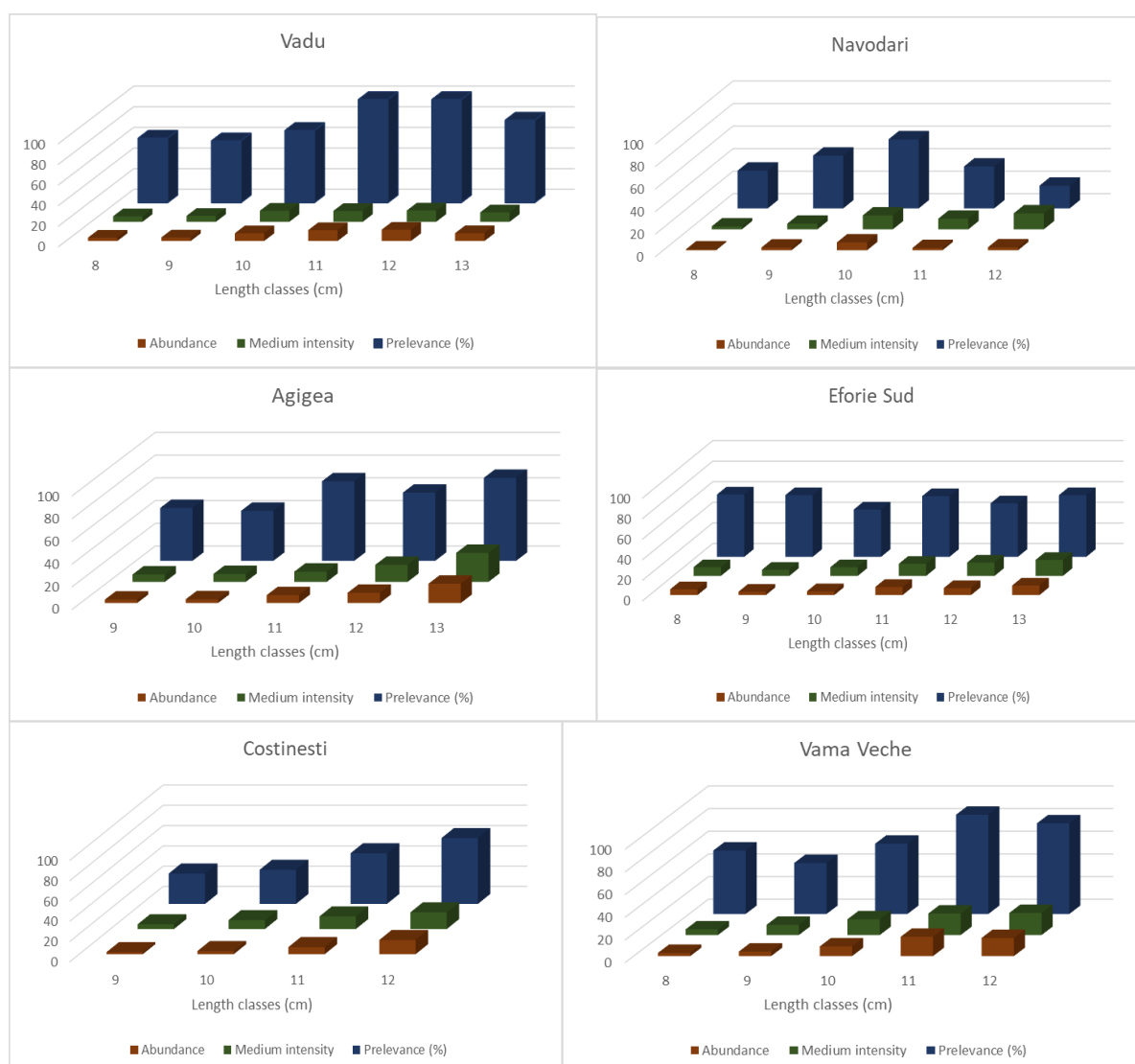


Fig. 9. Parasite infestation degree of anchovy on length classes along the Romanian coast

In other stations located in the southern coastal area, the level of parasites by size classes showed high values for the larger specimens, on average 13.5 parasites/host and 11.2 parasites/fish at

Vama Veche, 10 parasites/host and 2.8 parasites/fish in Costinesti, and in Eforie South 10 parasites / host and 4.2 parasites/fish (Fig. 8).

Anchovy presented a high level of parasites, expressed by the number of parasitic specimens from the total analyzed. The most parasitic specimens were identified in Vadu and Vama Veche stations. The anchovy specimens analyzed showed a higher degree of parasites, especially in the southern coastal area.

At Vama Veche, in small specimens of 8 - 9 cm length, the average invasion intensity was 5-9 parasites/host, while in 10 - 12 cm long specimens this parameter was 13.9 - 19.5 parasites/host. Anchovy specimens analyzed from the Costinesti station were 9 to 12 cm in length, with an infestation intensity of 3-14 parasites/host. At Eforie South, the larger anchovies reached the highest level of infestation. Thus, at a 13 cm length, the parasitic intensity was of maximum 40 nematodes/host (Fig. 9).

4. CONCLUSION

Of all five fish species studied, the most affected by parasites were turbot and anchovy, followed by sprat and horse mackerel. The analyzed fish species presented both ectoparasites and endoparasites.

The parasitic intensity was quite high in the case of the ectoparasite *Trichodina domerguei*, reaching 100 parasites/host in turbot. Also, the presence of the endoparasite *Botriocephalus scorpii*, with an infestation intensity of 100 parasites/host, was signaled.

Following parasitological analyzes, it can be concluded that sprat and anchovies are highly parasitized by species such as *Contracaecum*, *Porrocaecum* and *Anisakis*, but without endangering the natural populations and without causing significant mortality.

Sprat showed a high degree of parasite infestation in the northern part of the Romanian coast, with invasion extensions up to 86% and maximum intensities of 13 parasites/host.

Anchovy showed a higher degree of parasites than sprat in all the analyzed sampling stations, reaching 100% invasion extensions at Vadu and 88% at Vama Veche.

In the last years, researches in this domain are of big importance, parasites being utilized as biologic markers and as important indicators for fish circulation studies, migration and stock assessment, parasites being one of the causes for fish population reduction.

Assessing fish stocks from the health status point of view is of particular importance in the field of fisheries and human health.

References

1. Munteanu G., Bogatu D., *Tratat de Ihtiopatologie*, Timișoara, Excelsior art, 816, 2003.
2. Sinderman C., Effects of parasites on fish population: Practical consideration, *International Journal for Parasitology*, Vol.17, Issue 2, 371- 382, 1987.
3. Anderson R.M., Gordon D.M., Processes influencing the distribution of parasite numbers within host populations with special emphasis on parasite-induced host mortalities, *Parasitology*, 85 , 373 -398, 1982.
4. Bagge A.M., Poulin, R., Valtonen E.T., *Fish population size, and not density, as the determining factor of parasite infection: a case study*, 128 (Pt3), 305 -313, 2004.
5. MacKenzie K., Abaunza P., Parasites as biological tags for stock discrimination of marine fish: a guide to procedures and methods, *Fisheries Research*, Vol. 38, 45 – 56, 1998.
6. Lester R., J., G., A, Review of methods for estimating mortality due to parasites in wild fish populations, *Helgoland Marine research*, Vol. 37, No.1-4. 2010.
7. Radulescu I., Lustun L., Voican V., *Bolile pestilor*, Editura Ceres, Bucuresti, 1 – 261, 1976.
8. Gaevskaia A.V., Kovaleva A.A., *Bolezni promislavij rib Atlantiscego Okeana*, *Atlant NIRO* 1 – 125, 1975.

9. Zaharia T., Dumitrescu E., Maximov V., Cristea M., Nenciu M., Țoțoiu A., Observation on the parasite influence on the fish conservation status in Romanian marine Natura 2000 Sites, *Recherches Marines*, INCDM, Constanța, 42: 173-184, 2012.
10. Dumitrescu E., Zaharia T., Telembici A., Pathology of marine organisms of interest for the development of Mariculture on the Romanian Coastline - ACVAROM '98, Galati, 313-315, 1998.
11. Amlacher E., *Taschenbuch der Fishkrankheiten*, VEB Gustav Fisher Verlag Jena, 1 – 474, 1981.
12. ***, *Opređeliteli parazitov pozvociinah Cernovo i Azobskogo morei*, Kiev, "Naukova Dumka" 1 – 550, 1975.