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Settlement patterns of molluscs, with particular reference on Great Mediterranean scallop (*Pecten jacobaeus*, Linnaeus) and biofouling organisms on different type of collectors and locations in Boka Kotorska Bay

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The settlement of molluscs' larvae of and other biofouling organisms on experimental collectors was studied, with particular reference Great Mediterranean scallop, Pecten jacobaeus. *Three types of experimental collector made from vegetable sacks, modelled based on the collectors used in Japan, were placed in four existing fish and shellfish farms located in the Boka Kotorska Bay. The experimental collectors were placed in the periods June–October and June–December 2017 and the period August 2017–February 2018 and monitored after immersion of four to six months, respectively.*

In total, 18 species of molluscs and also 28 species of biofouling organisms were determined. The most abundant ones found on the collectors were the following shellfish species (68 %) Talohlamys multistriata, Mytilus galloprovincialis, Modiolarca sp., Anomia ephippium and Limaria hians. *The most abundant group of biofouling organisms were crustaceans (18 percent) with a species of porcelain crab,* Pisidia longicornis, *and tunicates (5 percent) with the species* Phallusia mammillata *and* Ascidia mentula.

The main target species, Great Mediterranean scallop, was most abundant on collector Type II, with 28 individuals.

From three types of experimental collectors used in the process of collecting larvae of shellfish and biofouling species, collector type-I and type-II proved to be a more suitable basis for receiving larvae of shellfish due to the unimpeded flow of sea water that allows their successful growth and development. Species Talochlamys multistriata*, were found in great abundance and it could be good candidate to diversify the aquaculture production, thus potentially offering farmers an additional source of income.*

Key words: shellfish cultivation, diversification, experimental collectors, *Pecten jacobaeus*, Boka Kotorska Bay

INTRODUCTION

In the area of the Montenegrin coast, more precisely in the Boka Kotorska Bay, until now only two types of molluscs have been commercially cultivated – the Mediterranean mussel (*Mytilus galloprovincialis*) and the European flat oyster (*Ostrea edulis*). The cultivation of other edible species, for example the Great Mediterranean scallop, which represents a species of shellfish of commercial interest, would lead to the diversification of the production of molluscs in the area of the Boka Kotorska Bay.

This species of mollusc has been caught to a large extent in recent years for its gastronomic value, leading to the need to restock the natural populations, which requires the beginning of controlled cultivation in experimental conditions (PRANOVI *et al*., 2001; MANTHEY-KARL *et al.,* 2015). Japan was the cradle of controlled cultivation of species from the family Pectinidae during the 1920s (IMAI, 1977). At the end of the 1960s in Mutsu Bay technology for cultivating the shellfish *Patinopecten yessoensis* was established, variants of which are used today all around the world for cultivating species from this family (VENTILLA, 1982; PRATO, 2015; MARGUŠ & TESKEREDŽIĆ, 2005). Because of the impressive results achieved in Japan, other countries made an effort to develop scallop culture, as in the U.K. on the west coast of Scotland in the early 1970's; in France about 1977, in Russia in the 1970's, in Chile in the early 1980's, in Mexico in the early 1980's, in the U.S.A. in the 1960's with bay scallops and more recently with sea scallops, in the early 1980's in Australia and New Zealand, in 1981 in British Columbia and in eastern Canada (BOURNE, 2004). The main commercially exploited scallops occurring throughout a wide geographical range on the European continental shelf are: *Pecten maximus* (great scallop or king scallop), *P. jacobaeus* (scallop or Great Mediterranean scallop), *Aequipecten opercularis* (queen scallop), *F. glaber* (white or smooth scallop) and *Mimachlamys varia* (black scallop) (MATTEI & PELLIZZATO 1996; BRAND 2006). In Mediterranean coastal waters, *P. jacobaeus, F. glaber, A. opercularis* and *M. varia* occur in the northern Adriatic although fishing activity has severely reduced their distribution and abundance (MATTEI & PELLIZZATO 1996). *A. opercularis* is farmed in Spain, France, Ireland, United Kingdom and in the North Sea (NORMAN, *et al.* 2006) and *M. varia* is currently successfully farmed in France, Ireland and Spain (ROMAN 1991; LOURO, *et al*., 2003). In Adriatic Sea (Croatia), one of scallop species, *Chlamys varia*, showed a good potential for commercial aquaculture in Mali Ston Bay (RATHMAN *et al*., 2017).

The technique for cultivating scallops is a very simple process, consisting of collecting scallop larvae, cultivating juvenile specimens and their final cultivation up to a commercial size of specimens (VENTILLA, 1982; MARGUŠ, 1988; MARGUŠ, 1989; MARGUŠ, 1989a). Earlier, natural and artificial types of collectors were used for collecting scallop larvae, given that the specimens did not show any particular selectivity towards materials (EVANS *et al.,* 1973). Second hand rope placed horizontally between two of the vertical posts of a fish farm or between buoys was used as one type of collector, used for collecting mussel larvae. For species of the scallop family (Pectinidae), polyethylene sacks filled with used nylon netting were mainly used (PEHARDA & ONFORI, 2000). For many pectinidae species, artificial collectors for obtaining wild spat is commonly used in several countries (NARVARTE 2001; SLATER 2005; AVENDANO *et al.* 2006; GUO & LUO 2006; KOSAKA & ITO 2006; ROMAN 2007; SORIA *et al*., 2014). Similar collectors, as mentioned above, made from a mesh onion bag filled with monofilament nylon mesh netting was used for cultivation of scallop, *Pecten maximus*, in UK. A series of these can be attached to a rope with a weight on the end and suspended in the water from a float (LAING, 2002). Beside the type of collector used for the settlement of mollusc larvae, it is necessary to pay attention also to the location, season and method of placing the collectors.

The aim of this research was to estimate the quantity of shellfish species as well as the number of target species –- the Great Mediterranean scallop on different types of experimental collectors in order to show which types of collectors is most suitable for settlement of the molluscus larvae.

MATERIAL AND METHODS

Research area

Boka Kotorska Bay is located in the south-eastern part of the Adriatic Sea, and is made up of four smaller bays – the Bays of Kotor, Risan, Tivat and Herceg Novi. The surface area of the territorial waters of the Boka Kotorska Bay is 87.3 km^2 (STJEPČEVIĆ & ŽUNJIĆ, 1964). The bay is characterised by unique hydrography and dynamics, given that it is a semi-closed basin connected to the rest of the Adriatic Sea between Oštro Cape to the west and Mirište Cape to the east. The dynamics of the temperature regime of the sea water in Boka Kotorska Bay are highly influenced by atmospheric precipitation, which is particularly characteristic of the interior of the bay – the Bays of Kotor and Risan. There are also very plentiful streams, and land-based and underwater springs, especially in the interior of the bay (STJEPČEVIĆ & PARENZAN, 1980).

Boka Kotorska Bay is characterised by irregular flows of masses of water, which mainly depend on the sea state and free oscillations, also known as seiches (MANDIĆ *et al.*, 2001). Many factors other than the sea state also influence the direction and strength of sea currents, including winds, changes in pressure, as well as the mixing of fresh and salt water. The currents are characterised by stronger intensities during the autumn, winter and spring, while the current intensity during the summer is weaker.

The fish farms which are the subject of this paper – Site 1, Site 2 and Site 3 are located in the Bay of Kotor, while Site 4 is located in the Bay of Tivat (Fig. 1). The depths of the areas where the fish farms are located are different. The maximum depth of Site 3 is 30 metres, of Site 4 is 25 metres, and of Site 2 is 20 metres, while the water is shallowest at Site 1, which is 9 metres deep.

Fig. 1. Areas of the fish farms where experimental collectors were placed (red dots)

Spat collectors

For the collecting of the larvae of molluscs and biofouling organisms, three types of collector were used, which were placed at different time periods in four existing fish and shellfish farms in the area of Boka Kotorska Bay.

The type-I experimental collector was made from a vegetable sack that is made from plastic material, with dimensions 45 cm × 45 cm, with a mesh size of 5 mm (HRS-BRENKO, 1990; MARGUŠ, 1990,1994). The collector was filled with one more similar sack, fishing line, nylon netting and a tubular net which is otherwise used for cultivating mussels (longlines). This type of collector was placed in June 2017 at Site 1, Site 3 and Site 4. At each fish farm six collectors were placed, the depths varying from 2 metres below the water surface to 2 metres above the sea bed. The spacing between the collectors was 1.5 metres.

The processing of the experimental collector at Site 4 was carried out after four months of immersion (from June to October 2017), while processing of the collectors at Site 1 and Site 3 was carried out after six months of immersion (from June to December 2017).

The type-II collector was also made from a vegetable sack of dimensions 45 cm × 45 cm with a mesh size of 5 mm. It was filled with nylon netting and a tubular net which is otherwise used for cultivating mussels (longlines). It was placed in June 2017 at Site 1, Site 3 and Site 4. At each fish farm seven collectors were placed, with a spacing of 1.5 metres between them. The collectors were placed starting at 2 metres below the water surface and the last was located no less than 2 metres above the sea bed. Processing of the collectors was carried out in October 2017 at Site 4 and in December 2017 at Site 1 and Site 3.

Type-I and type-II collectors were placed alternately at Site 1, Site 3 and Site 4, so that both types of collector were to be found on the same rope. The processing of type-I and type-II collectors in the fish farms at Site 1 and Site 3 was carried out two months later than at Site 4 because of the small dimensions of the marine organisms that were noticed during the processing of the collectors at the aforementioned Site 4.

The type-III collector was made of a vegetable sack of dimensions 70 cm × 30 cm which had a mesh size of 2 mm and was filled with nylon netting and a tubular net which is otherwise used for cultivating mussels (longline). The collectors were placed in August 2017 at Site 2, Site 3 and Site 4, and were processed after six months of immersion, i.e. in February 2018.

At each of Site 2, Site 3 and Site 4, in the period from August 2017 to February 2018, 12 type-III collectors were placed with a spacing between the collectors of 1 metre, beginning at 2 metres below the water surface down to 2 metres above the sea bed.

Unlike the localities at which the experimental collectors' types I and II were placed in June, Site 1 was substituted by Site 2 in August, because of the much more suitable environmental conditions and to reduce anthropogenic influences.

After immersion of the collectors in the mentioned periods, their processing was carried out and the species and families of the specimens were determined. The collectors were stored in containers and submerged in 70 percent alcohol until they were processed. During the entire period of research, analysis of the basic physiochemical parameters of the sea water was carried out on a monthly basis, particularly of the temperature and salinity which were measured by a CRISON CM 35+ sensor at the fish farms where the collectors were placed.

The diversity of the molluscs and biofouling organisms was calculated with the help of diversity indices – Margalef's (1965) and Simpson's diversity indices and the Shannon–Wiener index (H) in order to show the diversity and richness of species. The determinations of the specimens of molluscs and biofouling organisms was carried out with the help of literature by POPPE & GOTŌ, (1993; 2000) and RIEDL (2002). The dimensions of the target species, *Pecten jacobaeus*, were measured by means of digital callipers with an accuracy of 0.1 mm.

PRIMER 6.0 software package (CLARKE & GORLEY, 2006), Bray-Curtis similarity and nMDS technique were used to determine similarity of shellfish community.

A one-way analysis of variance (ANOVA) was used to compare variances of the abundance of shellfish and of Great Mediterranean scallop on collectors with type of collector and site.

RESULTS

Eighteen different species of molluscs and 28 species of biofouling organisms were determined on the experimental collectors. Of the total 6,683 specimens collected, the most represented were: bivalves (Bivalvia) – 68 percent; crustaceans (Crustacea) – 18 percent; tunicates (Tunicata) – 5 percent; bristle worms (Polychaeta) – 4 percent; and sponges (Porifera) – 2 percent.

Site 1

At Site 1 (Fig. 1), only four of the seven type-I collectors placed were retrieved, and the view is that the other three became detached as a result of bad weather. On them were counted 361 specimens (12 species); not one Mediterranean scallop was among them, and the most represented species were identified as *Mytilus galloprovincialis* (42.1 percent), *Modiolarca sp.*

(20.22 percent) and *Talochlamys multistriata* (15.8 percent).

At Site 1, 16 biofouling species and a total of 123 specimens were counted. The most represented biofouling species was *Pisidia longicornis* (17.07 percent).

Of the seven type-II collectors placed at Site 1, not a single one was found and the reason for this is presumed to be that they became detached as a result of bad weather.

After the situation with the detachment of the collectors, it was decided to continue the research by relocating to the fish farm at Site 2 (Fig. 1).

On the type-III collectors at Site 2, 1,043 specimens (18 species) were recorded. At this fish farm all 12 collectors placed were processed, in which 12 specimens of the Great Mediterranean scallop were recorded at the following depths: one specimen each at 6 metres, 8 metres and 13 metres; two specimens each at 9 metres, 10 metres and 14 metres; and three specimens at 5 metres. The most represented species were *Modiolarca sp* (61.36 percent), *Limaria hians* (12.94 percent) and *Anomia ephippium* (8.25 percent). The largest number of molluscs (205) was recorded at a depth of 11 metres.

As far as biofouling organisms at Site 2 are concerned, 13 species with a total of 1,157 specimens were determined. The most represented biofouling organism was the porcelain crab *Pisidia longicornis* (63.35 percent).

Site 3

At Site 3 (Fig. 1) the type-I collectors had 425 specimens (15 species). Of the six collectors placed, five were processed, and the view is that one collector became detached as a result of bad weather. On this type of collector 25 specimens of the Great Mediterranean scallop were present – five specimens each at depths of 6 metres, 7.5 metres and 15 metres; seven at a depth of 10.5 metres; and three specimens at a depth of 16.5 m. The most abundant species were *Mytilus galloprovincialis* (31.06 percent), *Modiolarca sp.* (17.65 percent) and *Talochlamys multistriata* (9.88 percent).

The largest number of molluscs (93) was recorded at depths of 9 and 10.5 metres.

On the same type of collector at Site 3, 12 species of biofouling organisms with a total of 135 specimens were recorded, the most represented of which was *Pisidia longicornis* (40.74 percent).

On the type-II experimental collector at Site 3, 704 specimens (15 species) were found. In this fish farm six of the seven collectors placed were processed, as one collector became detached as a result of bad weather. Eighteen specimens of the Great Mediterranean scallop were recorded – one specimen at 12 metres; two each at 13.5 metres, 18 metres and 22.5 metres; four specimens at 21 metres; and six at 19.5 metres. The most abundant species were *Mytilus galloprovincialis* (39.49 percent) and *Talochlamys multistriata* (23.30 percent).

The largest number of molluscs, in total 107, was recorded at a depth of 19.5 metres.

On the type-II collectors at Site 3, 16 species of biofouling organisms were recorded with a total of 201 specimens, among which the most represented was *Pisidia longicornis* (44.78 percent).

On the type-III collector at Site 2, 364 specimens (16 species) were recorded. Two specimens of the Great Mediterranean scallop were recorded at a depth of 15 metres. The most abundant species were *Modiolarca sp.* (52 percent) and *Mytilus galloprovincialis* (18 percent). The largest number of molluscs (63) was recorded at a depth of 10.5 metres.

On the type-III collectors at Site 3, 11 species of biofouling organisms were determined with a total of 158 specimens, among which the most represented was the species *Protula sp.* (39 percent).

Site 4

At Site 4 (Figure 1) 350 specimens of molluscs (14 species) were collected on the type-I experimental collectors. Of the six placed collectors, five were processed. The view is that due to bad weather one collector became detached from the rope. Two specimens of the target species, the Great Mediterranean scallop, were found at a depth of 9 metres. The largest number of specimens of molluscs (121) was recorded at a depth of 3.5 metres.

In total, 135 specimens of biofouling species (15 species) were determined, the most abundant of which was the species of porcelain crab *Pisidia longicornis* – 16.92 percent.

The total number of specimens on the type-II collectors was 616 (16 species). Of the seven collectors placed, during the processing it was noticed that one collector had disappeared and the view is that it became detached due to bad weather. Nine specimens of the Great Mediterranean scallop were recorded at a depth of 13 metres, three specimens at 15 metres, two specimens at 17 metres and one specimen at 23 metres. The largest number of mollusc specimens was recorded at depth of 13 metres. The total number of specimens on type-II collectors was 616 (16 species). Of the seven collectors placed it was noticed during processing that one collector was missing, and the view is that it became detached as a result of bad weather. Nine specimens of the Great Mediterranean scallop were recorded – three specimens at a depth of 13 metres, one specimen at a depth of 15 metres, two at a depth of 17 metres, and one at a depth of 23 metres. The largest number of molluscs (179) was recorded at a depth of 13 metres. There were 150 specimens of biofouling species (16 species) recorded, with the most abundant being *Pisidia longicornis* – 29.33 percent.

On the type-III experimental collectors 663 molluscs (17 species) were collected. Five of the 12 collectors placed became detached as a result of bad weather. Two specimens of the Great Mediterranean scallop were collected at a depth of 8 metres. The most abundant species on this type of collector were *Modiolarca sp.* and *Limaria hians*. The largest number of molluscs (133) was recorded at a depth of 9 metres. There were 157 specimens of biofouling species (15 species), with the most represented being *Pisidia longicornis* (22 percent) and *Protula sp.* (19 percent).

The total diversity of the researched molluscs and biofouling organisms at the fish farms is given in Table 1. In total, 18 species of Bivalvia, six species of Tunicata, five species of Bryozoa, two species of Echinodermata, one species of Porifera, nine species of Crustacea, two species of Rodophyta, one species of Gastropoda, two species of Polychaeta and one species of Cnidaria were determined.

The vertical distribution of mollusc larvae determined on experimental collectors at fish farms are given in Table 2. The mollusc larvae were mostly collected at a depth of 11 metres, with a total of 391 specimens, with the most abundant species being *Modiolarca sp*.

The value of the temperature and salinity of the sea water in the period from June to September 2017 was measured at depths of: 0 metres, 10 metres and 30 metres at Site 3; 0 metres, 5 metres and 10 metres at Site 4; and 0 metres, 5 metres and 9 metres at Site 1 (Graph 1). For the period from October to February, the values of temperature and salinity at the aforementioned localities and at Site 4 were measured at a depth of 0.5 metres.

With the processing of 59 experimental collectors which were located at four fish farms, in total 71 specimens of the Great Mediterranean scallop (1.3 percent) were collected. The largest number of specimens was recorded at depths of 10 metres and 15 metres at Site 3 (Graph 2), where the temperature varied from 17.5 to 22°C and the salinity from 36.2 to 37.7‰ (Graph 1). At Site 4, whose maximum depth was roughly 25 metres, the largest number of scallop specimens was recorded at a depth of roughly 10 metres, where the temperature varied from 18.3 to 22.8°C and the salinity from 34.2 to 37.9‰ (Graph 1). In the area of Site 2, specimens of scallops were recorded at the middle of the water column (depending on the depth of the area), at a depth of between 5 and 14 metres (Graph 2).

The selected specimens of the Great Mediterranean scallop retrieved from the collectors were measured, and their mean length values are shown in Table 3.

On the basis of the mean length values of Great Mediterranean scallops in the fish farms, a variation in the dimensions was observed at different fish farms. At Site 3 the largest number of specimens of scallops was recorded at a depth of between 10.5 metres and 20 metres, and the

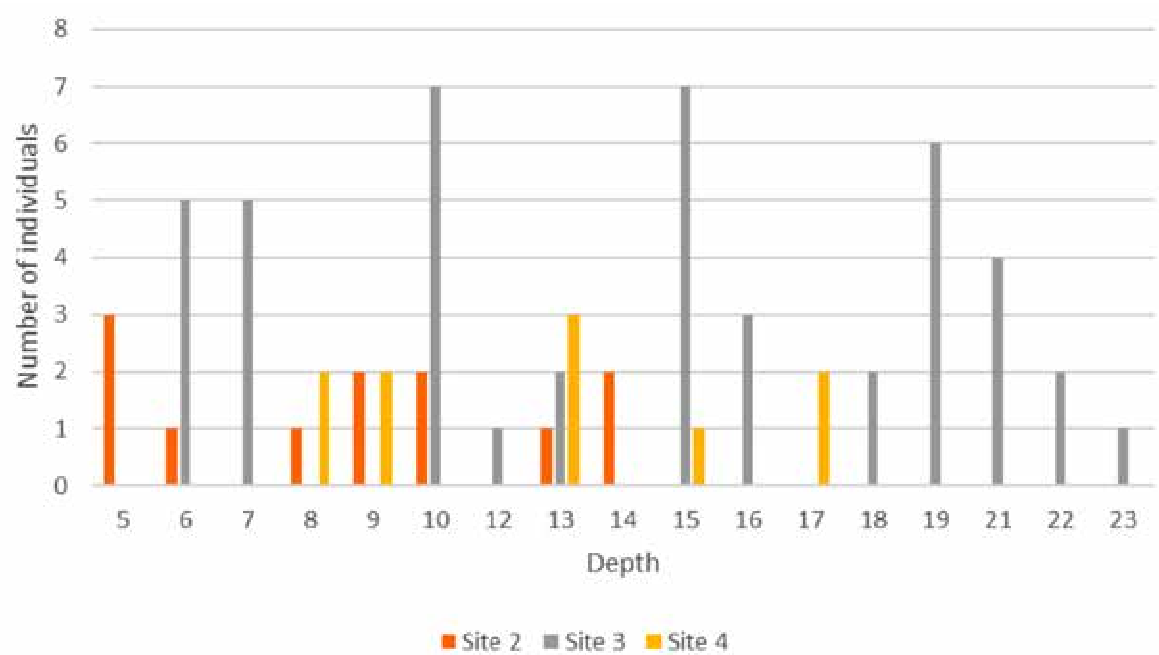
Table 1. Numerical and percentage participations of molluscs and biofouling organisms on experimental collectors at fish farms

Type of collector	**Type I**	**Type II**	**Type III**	**Total number of individuals**	**Percentage**
Bivalvia					
Musculus subpictus (Cantraine, 1835)	186	227	1253	1666	25%
Mytilus galloprovincialis (Lamarck, 1819)	363	298	103	764	12%
Talochlamys multistriata (Poli, 1795)	190	378	94	662	10%
Limaria hians (Gmelin, 1791)	50	115	234	399	6%
Anomia ephippium (Linnaeus, 1758)	71	108	115	294	4%
Flexopecten glaber (Linnaeus, 1758)	42	37	55	134	2%
Pecten jacobaeus (Linnaeus, 1758)	27	28	16	71	1%
Hiatella rugosa (Linnaeus, 1767)	32	19	5	56	1%
Ostrea edulis (Linnaeus, 1758)	36	36	11	83	1%
Fleopecten flexuosus (Poli, 1795)	8		26	34	1%
Aequipecten opercularis (Linnaeus, 1758)	12	16	40	68	1%
Pinna nobilis (Linnaeus, 1758)	68	8	12	88	1977/%
Pinctada radiata (Leach, 1814)	21	10	15	46	1%
Chlamys varia (Linnaeus, 1758)	12	12	25	49	1%
Acanthocardia sp. (Linnaeus, 1758)	14	13	47	74	1%
Polititapes aureus (Gmelin, 1791)		1	11	12	0.2%
Tapes decussatus (Linnaeus, 1758)	4	3	4	11	0.2%
Modiolus barbatus (Linnaeus, 1758)			4	4	0.1%
Tunicata					
Phallusia mammillata (Cuvier, 1815)	38	30	72	140	2.1%
Ascidia mentula (Müller, 1776)	8	27	46	81	1.5%
Styela plicata (Lesueur, 1823)	19	4	73	96	1.4%
Styela clava (Herdman, 1881)	4			4	0.06%
Trididemnum cereum (Giard, 1872)	4			4	0.06%
Diplosoma spongiforme (Giard, 1872)	2	2		4	0.06%
Bryozoa					
Bugula neritina (Linnaeus, 1758)	31	25	69	125	2%
Savignyella lafontii (Audouin, 1826)	2	5	1	8	0.1%
Schizobrachiella sanguinea (Norman, 1868)	2	2		4	0.1%
Crisia eburnea (Linnaeus, 1758)			4	4	0.1%
Fenestrulina malusii (Audouin, 1826)	2	1		3	0.05%
Echinodermata	L2				
Ophiothrix fragilis (Abildgaard in O.F. Müller, 1789)	13	5	3	21	0.3%
Psammechinus microtuberculatus (Blainville, 1825)	3	1	6	10	0.2%
Porifera					
Paraleucilla magna Klautau, Monteiro & Borojevic, 2004	28	15	100	143	2%
Crustacea					
Pisidia longicornis (Linnaeus, 1767)	87	134	798	1019	15%
Eriphia verrucosa (Forskål, 1775)	29	23	36	88	1%
Athanas nitescens (Leach, 1813)	5	11	45	61	1%
Eualus cranchii (Leach, 1817)	4	4		8	0.1%
Parapenaeus longirostris (Lucas, 1846)		1		1	0.02%
Macropodia rostrata (Linnaeus, 1761)	1	0		1	0.02%
Balanus Costa, 1778			1	1	0.02%
Perforatus perforatus (Bruguière, 1789)	1			1	0.02%
Rhodophyta					
Laurencia (J.V.Lamouroux, 1813)	4			4	0.1%
Wundermannia mimata	2	2		4	0.1%
Gastropoda					
Nassarius (Duméril, 1805)		1		1	0.02%
Polychaeta					
Protula (Risso, 1826)	30	36	182	248	4%
Polychaeta (Grube,1850)	1	19	15	35	1%
Cnidaria					
Sertularella (Gray, 1848)	3	1		4	0.1%

Table 2. Vertical distribution of species of molluscs on collectors during the research period.

Species / Depth (m)	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	21	22	23
Ruditapes decussatus			1		2			2	1	1		1		2	1	1					
Hiatella rugosa	6	1	4	1	5	8		4	3	3	4	1		6	1	3	2	1	3	2	1
Modiolus barbatus					2		1			1	5						11				
Ostrea edulis	4	11	6	3	5	10		1	2	10	2	4		5		8	2	1	5	1	1
Pinna nobilis	11	11	3	3	19	13	1	4	7	3	11	4		4							
Pinctada radiata	3	5	15	8	1	2	2			3	3			1	1						
Flexopecten flexuosus		4	3	1	2		1	2		2	1			3			1				
Acanthocardia sp.	1	6	8	14	7	6	6	2	5	7	3	4		2		1		2	1		1
Tapes aureus							1	1				3	5	5							
Talohlamys varia	1	1	5	5	2	2		2		1	5		1			3	3	1	1		
Pecten jacobaeus				3	6	9		7	9		1	6	2	6	3	4	2	6	4	2	1
Flexopecten glaber	6	5	11	9	13	10	8	11	8	9	7	12	1	8	2	1	1	2	10	1	1
Aequipecten opercularis		2	4	2	11	5	3	1	3	5	4	14	1	3	2	1	1	3	9	5	1
Mytilus galloprovincialis	66	56	18	26	48	41	2	58	37	9	27	87		36	40	39	40	37	18	33	1
Chlamys multistriata	27	24	55	50	13	38	3	56	13	25	17	25	1	54	27	66	27	34	24	11	14
Anomia ephippium	3	29	13	17	10	35	14	16	12	40	19	28		15	1	36	3	3	6		
Limaria hians	2	20	30	47	21	20	5	29	12	91	38	7	1	14	2	13	2	7	4	3	1
Modiolarca sp.	23	26	67	49	174	158	120	189	146	181	66	71	24	52	10	1	8	20	14	14	

Graph 1. Temperature and salinity at the Site 1, Site 3 and Site 4



Graph 2. Abundance of specimens of the Great Mediterranean scallop on collectors at fish farms

Table 3. Mean length values of specimens of the Great Mediterranean scallop in the researched areas

Site 2			**Site 3**			**Site 4**		
Depth (m)	***Pecten jacobaeus* (n)**	**Mean length values (mm) *Pecten jacobaeus***	**Depth (m)**	***Pecten jacobaeus* (n)**	**Mean length values (mm) *Pecten jacobaeus***	**Depth (m)**	***Pecten jacobaeus* (n)**	**Mean length values (mm) *Pecten jacobaeus***
5	3	23	**6**	5	19.5	**8**	2	24.5
6	1	/	**7.5**	5	22.55	**9**	2	16.24
8	1	12.83	**10.5**	7	23.93	**13**	3	13.54
9	2	8.63	**12**	1	15.54	**15**	1	9.30
10	2	17	**13.5**	2	24.13	**17**	2	11.05
13	1	/	**15**	7	15.8	**23**	1	8.06
14	2	9,87	**16.5**	3	18.48			
			18	2	18.63			
			19.5	6	18,37			
			21	4	18.77			
			22.5	2	16.71			

largest mean length value of the specimens was 24.13 mm, recorded at a depth of 13 metres. The richness and diversity of species on experimental collectors at Site 1, Site 2, Site 3 and Site 4 in the period from June to October/December 2017 and from August 2017 to February 2018 are given in Table 4.

The maximum value of the Shannon–Wiener diversity index (H’) on experimental collectors was 2.22 at Site 4 on a type-II collector, and the lowest was 1.42 at the same site but on a type-III collector. According to this, the greatest diversity of species was at Site 4 (type-II) and the lowest was at the same site on a type-III collector.

Table 4. Diversity indices at fish farms

Collector Type I						
Sites /Indices	Margalef diversity index		Simpson's diversity index		Shannon–Wiener diversity index	
	Shellfish	Biofouling species	Shellfish	Biofouling species	Shellfish	Biofouling species
Site 1	11.83	15.79	4	8.82	1,76	2.39
Site 3	14.83	11.80	6,34	4.58	2,21	1.91
Site 4	13.83	14.76	6,83	9.99	2,11	2.46
Collector Type II						
Sites/Indices	Margalef diversity index		Simpson's diversity index		Shannon–Wiener diversity index	
	Shellfish	Biofouling species	Shellfish	Biofouling species	Shellfish	Biofouling species
Site 1	/	/	/	/	/	/
Site 3	13,85	14.81	4,05	4.14	1,75	1.90
Site 4	15,84	15.80	1,68	6.58	2,22	2.24
Collector Type III						
Localities/Indices	Margalef diversity index		Simpson's diversity index		Shannon–Wiener diversity index	
	Shellfish	Biofouling species	Shellfish	Biofouling species	Shellfish	Biofouling species
Site 2	17.86	13.86	2.47	2.38	1.47	0.63
Site 3	15.83	10.80	3.17	4.49	1.70	1.83

The highest value of Margalef's index, 17.86, was recorded at Site 2 (type-III collector) and the lowest, 6.83, was recorded at Site 1 (type-I collector). The highest value of Simpson's diversity index, 6.83, was recorded at Site 4 (type-I collector), and the lowest, 1.68, was at the same site but on a type-II collector. On the basis of these indices, we conclude that the homogeneity is lowest at Site 4 on type-I collectors, and is highest at the same site on type-II collectors. As far as biofouling species are concerned, the maximum value of the Shannon–Wiener diversity index (H') on type-I collectors was 2.46 at Site 4, and the lowest, 0.63, was at Site 2 on type-III collectors. The values of Margalef's index were highest at Site 4 (15.80) and lowest at Site 3 (10.80) on type-III collectors.

The highest values of Simpson's diversity index were at Site 4 (9.99), and the lowest at Site 2 (2.38) on type-III collectors.

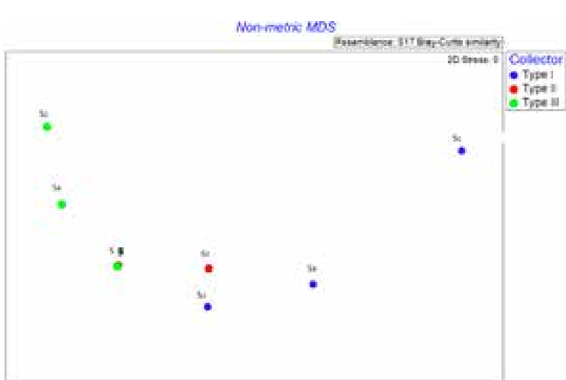


Fig. 3. MDS grouping of sites by shellfish species on collectors

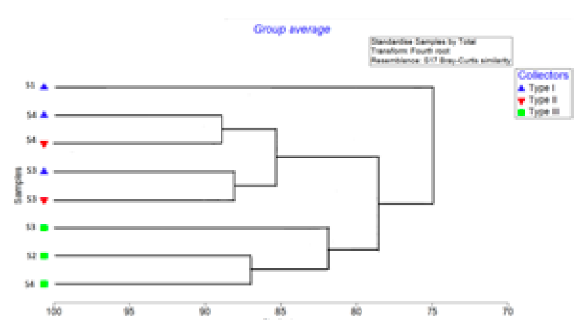


Fig. 4. Similarity of sites by shellfish species on collectors (Bray-Curtis method)

Significant differences were not observed in spat abundance on three types of collectors (p>0,005), as on different depth. On Type I and Type II (set in June) was collected highest number of Great Mediterranean scallop spat. In addition, highest number of shellfishes was on collector Type III on site S2. The statistical method of multidimensional scaling shows a difference of site S1 according to the types of shellfish compared to other sites. Sites S2, S3 and S4 with type-III collectors show correct grouping (Figure 3). Bray-Curtis similarity analysis for shellfish species showed the lowest similarity of the site S1 with the type-I collector compared to other sites, with a similarity percentage of 75%. It is noticed that the type-I collectors of site 4 show the greatest similarity with the type-II collectors at the same locality, 89%. The same situation is observed at the site 3 with a similarity percentage of type-I and type-II collectors of 88%. Type-III collectors, at sites S2, S4 show the greatest similarity of 87% (Figure 4).

DISCUSSION

PENA *et al.* (1996) successfully collected specimens on pine leaves, on the shells of used scallops, on palm bark and on polyethylene film up until 1970, when onions sacks (or vegetable sacks) filled with fishing line or similar plastic material began to be used as collectors. Collectors made from similar materials are also used in this paper. KNUCKEY (1995), PENA *et al.* (1996), MARGUŠ *et al.* (1990) and BRAND *et al.* (1980) describe collectors made from vegetable sacks as being the most suitable for settling larvae as well for retaining them when the molluscs' byssus separates from the substrate. One of the most important characteristics in these collectors, to which special attention should be paid, is the size of the openings in the mesh from which the collector is made. On the basis of the obtained results and the number of settled specimens, mesh size is one of the causes of the specimens remaining in the collectors.

LATROUITE (1978) highlights that mesh dimensions of 1.33 mm × 1.05 mm and 1.7 mm × 2.0 mm settle a larger quantity of specimens of Pectinidae than a mesh of size 3.5 mm × 3.5 mm, while in the same area of the Eastern Adriatic (Croatia) a larger settlement of specimens of the Mediterranean scallop was recorded on collectors with a larger mesh size (MARGUŠ *et al.,*1993; MARGUŠ & TESKEREDŽIĆ, 2005). The data obtained in this paper regarding the quantity of collected specimens of scallops and similar species of molluscs on type-I and type-II experimental collectors at Site 3 and Site 4 point to the fact that collectors with a larger mesh size are a good substrate for settling larvae. As both type of collectors were set on same rope and at same period of time it was not noted significant differences on abundance of organisms, mesh size of 5mm showed to be better material for collectors, BRAND *et al.* (1980), used collectors with larger mesh size, that resulted in good settlement of scallops. They also pointed that size of outer bag, should be small to reduce the numbers of spat escaping but not too small to cause siltation and reduced water flow. During the processing of type-III collectors which had a smaller mesh size in the aforementioned sites, only two specimens of the Great Mediterranean scallop were recorded, as well as a lower number of the other mollusc and biofouling species compared to sum of both collectors installed in June. Reduced number of spat in collectors with 2 mm mesh size was also noted in study BRAND *et. al* (1980). These collectors became badly clogged with silt which probably decreased water flow through the collector. Since the results of ANOVA do not show significant differences between collector types, based on mesh size and amount of collected organisms, we noted that target species, Great Mediterranean scallop was most abundant on the Type I and Type II collector which was better substrate for settlement. Besides the mesh size, the spawning intensity of various species for the particular period of the year also had an influence on the settlement of specimens on the collectors. TSOTSIOS *et al.* (2016) draws attention to the fact that the settlement of specimens on various different types of collectors largely depends also on the period when the collectors are in the water. It happens that different quantities of specimens are found in the same collector, that is, if a particular type of collector is well-settled the spring, this does not mean that it will also be well-settled during the summer or autumn. In MARGUŠ *et al.* (1993) and MARGUŠ & TESKEREDŽIĆ (2005), the period March to June or July in location of the mouth of the river Krka, is mentioned as being the most suitable part of the year for collecting scallop larvae, however some data about histological analysis of the gonad tissue of the Mediterranean scallop in literature has established that some molluscs spawn in January and February, while in some specimens the spawning is extended and is at its height in the summer and autumn (VALLI, 1979). Result of abundance of Great Mediterranean scallopon Site 3 and Site 4 on collectors Type I and II that were installed in june showed larger quantity of species (54) then on collector Type III (14). It could be linked with results aforementioned studies, according to spawning season of main target species. Earlier installed collectors, would be well settled because of spawning season that is at its height in summer (MARGUŠ *et al.*, 1993; MARGUŠ & TESKEREDŽIĆ, 2005; PRATO, 2021).

At Site 2 on a type-III experimental collector which was placed in August, 14 specimens of Mediterranean scallop were recorded, and more molluscs and biofouling species than in the other two fish farms on the same type of collector. On the basis of the results of the settled specimens of Great Mediterranean scallops on the collectors in August we can link the appearance of specimens in the late summer at the mentioned fish farms and of the data in the literature regarding the later, or rather the extended spawning during the summer and autumn (VALLI, 1979). Of course, suitable environmental conditions in the area of the fish farms also contribute to this, which at Site 2 could be the River Ljuta, as well as small springs which bring nutritional matter. Sea currents also influence the settlement of specimens – in the bay during the summer period these currents flow outwards. We can link this phenomenon with the very small number of scallop specimens recorded on the collectors placed in August (type-III). Also, during the autumn, due to an inflow of fresh water, there are more intensive dynamics in the surface layer (STJEPČEVIĆ, 1974). In the colder period, the area of Verige, which is located close to Site 4, is characterised by an outflowing current, which can lead to a decrease in the number of specimens on the collectors, because they are caried by the sea currents towards the open sea. Diversity indices indicate that the greatest diversity of shellfish was on Site 3 on Type I and Site 4 on Type II, but there was not significant difference because it differs in only one species.

The largest settlement of specimens during the research at Site 2, Site 3 and Site 4 was recorded at depths of 8 to 20 metres, but with no statistical significance.

Previous research on the settlement of specimens of the Great Mediterranean scallop in areas with a depth of 20 or more metres recommend a depth of 10 to 15 metres as being the most suitable for placing collectors. Also, they state that the maximum settlement of specimens is achieved on collectors which are located 6 to 8 metres above the sea bed (in area 21 metres deep) (MARGUŠ *et al.*, 1993; MARGUŠ & TESKEREDZIĆ, 2005). The influence of salinity on the settlement of specimens as one of two essential limiting factors in the growth of scallops also affects the settlement of larvae. The achieved results show the greatest settlement on collectors where there is a salinity of 37–38‰ at Site 4 and 34–38‰ at Site 3. MARGUŠ & TESKEREDZIĆ (2005) recorded the most intensive settlement of larvae at salinities of more than 30‰, which we can link with the achieved results of settlement on collectors, where a larger number of specimens is found at depths with a higher salinity. Scallop larvae spend roughly 3 to 4 weeks as plankton, and the most suitable temperature for their development is roughly 20˚C (MASON, 1983). The temperature at the localities varied from 8.4 to 27.1˚C. In terms of the appropriate temperature for the settlement of larvae at the middle of the water column (10 to 20 metres), this varied between 17 and 23˚C. According to CHAUVAUD *et al.* (1998) the temperature and salinity of the water have a greater influence on the normal development of specimens than do sea currents and food, but they also assert that the blooming of toxic algae significantly reduces the growth of specimens.

There are also very plentiful streams, and land-based and underwater springs, especially in the interior of the bay (STJEPČEVIĆ & PARENZAN, 1980), that could be one of the reasons for greater quantity of organisms on colletors installed in middle of water column.

On the experimental collectors, beside the specimens of the Great Mediterranean scallop, 18 species of molluscs and 28 species of biofouling organisms were determined (tunicates, algae and porcelain crabs). MDS analysis showed the correct grouping of positions based on the number of species on type II and type III collectors, and only site S1 stood out by number of species. There is no such big influence of the open sea that would cause a greater diversity of organisms. In terms of similarity of positions according to the Bray Curtis method, we again see the smallest similarity of site S1 of 75 percent compared to others, which again indicates a difference due to the position of shellfish farm in the inner part of Bay.

To compare with our results, KNUCKLEY (1995) collected as many as 47 species of mollusc on a similar type of collector in a study carried out in the area of the Timor Sea. The most represented species of mollusc recorded on experimental collectors in the fish farms were the species *Modiolarca sp*. (36.9 percent), *Mytilus galloprovincialis* (16.9 percent) and *Talochlamys multistriata* (14.6 percent). In the area of Croatia (the estuary of the River Krka) MARGUŠ *et al.* (1993) collected on a similar type of collector four species of scallop: the variegated scallop *Chlamys varia* being the most numerous species; then the Great Mediterranean scallop *Pecten jacobeus*; the scallop *Flexopecten flexuosus*; and the Queen scallop *Aequipecten opercularis*. PENA *et al.* (1996) recorded on collectors a diversity of nine different species of shellfish, of which five species belong to the family Pectinidae: *Flexopecten flexuosus*, *Chlamys varia, Aequipecten opercularis, Pecten jacobaeus* and *Palliolum incomparabile*, then the species *Mytilus galloprovincialis*, *Hiatella artica*, *Plagiocardium papillosum* and *Musculus costulatus*.

The biofouling organism which appears in experimental collectors in the largest numbers is the porcelain crab *Pisidia longicornis*. The appearance of the mentioned species in large numbers was also mentioned in the paper by PRATO *et al.* (2015), but they do not have any data about the possible influence of this species on specimens of molluscs and of the target species, the Great Mediterranean scallop. However, they assert that tunicates, Bryozoa and the other species of porcelain crabs can influence the survival of specimens on collectors which remain immersed for up to seven months. Apart from porcelain crabs, also tunicates (*Ascidia mentula, Phalusia mammillata*) and bristle worms (*Protula sp., Polychaeta sp.*) have appeared on experimental collectors as abundant organisms. We can also link the obtained data with the results of the dissertation by STJEPČEVIĆ (1974), which displayed the most represented species in the area of the Boka Kotorska Bay. In first place are tunicates, such as ascidians (*Ascidia mentula, A. aspersa, A. cristata*), and then various species of Bivalvia (small scallops – *Chlamys varia; Anomia sp.*), bristle worms (Polychaeta) are very numerous, then the cirripede crustaceans Balanus, which have been proven to feed abundantly on the larvae of oysters and mussels (STJEPČEVIĆ, 1974).

CONCLUSIONS

The data obtained by the analysis of experimental collectors in the area of the Boka Kotorska Bay points to the most suitable positioning of cultivation installations is in the middle of water column (10-20 m according to site depth), in which exist the most suitable conditions for the growth and development of specimens. The target species Great Mediterranean scallop, was represented on collectors by a total of 71 specimens, and these were most abundant at depths of between 10 and 15 metres. One more potential commercially significant species which appeared on the collectors in larger numbers was the dwarf fan shell, *Talochlamys multistriata*. The possibility of cultivating the mentioned species, as well as its introduction into the area of the Boka Kotorska Bay, should certainly be researched in more detail. The study undertaken showed that type-I and type-II collectors showed the highest settlement of larvae and that for successful settlement it is necessary to pay attention to the link between the type of collector used for settling mollusc larvae, the spawning period of the main specimens and the locality in which it is planned to place experimental collectors.

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Naseljavanje mlađi školjkaša sa posebnim osvrtom na vrstu Jakobska kapica (*Pecten jacobaeus*, Linnaeus), i obraštajnih organizama na tri vrste eksperimentalnih kolektora u Bokokotorskom zaljevu

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SAŽETAK

U radu je prikazana analiza naseljavanja mlađi školjkaša i obraštajnih vrsta na eksperimentalnim kolektorima, sa posebnim osvrtom na vrstu jakobova kapica, *Pecten jacobaeus, L.* Tri tipa eksperimentalnih kolektora napravljenih od vreća za povrće po uzoru na kolektore korištene u Japanu bili su postavljeni na četiri postojeća uzgajališta ribe i školjaka na području Bokokotorskog zaljeva. Eksperimentalni kolektori su postavljeni u periodu od lipnja do listopada i lipnja-prosinca 2017. i u periodu od kolovoza 2017. do veljače 2018. godine, i obrađeni nakon 4 odnosno 6 mjeseci.

Na ukupnom broju postavljenih eksperimentalnih kolektora sakupljeno je 18 vrsta školjki i 28 vrsta obraštajnih organizama. Na kolektorima su najzastupljenije bile školjke (68%) sa vrstama: *Talohlamys multistriata, Mytilus galloprovincialis, Modiolarca sp., Anomia ephippium* i *Limaria hians*. Od obraštaja najzastupljenija grupa organizama su bili rakovi (18%) sa vrstom račića, *Pisidia longicornis* i plaštenjaci (5%) sa vrstama *Phallusia mammilata* i *Ascidia mentula*. Ciljana vrsta, jakobska kapica, bila je najzastupljenija na kolektoru Tip II, s 28 jedinki.

Od tri vrste eksperimentalnoh kolektora korištenih u procesu prikupljanja ličinki školjkaša i obraštajućih vrsta, kolektor tip-I i tip-II pokazao se prikladnijom osnovom za prihvat ličinki školjaka zbog nesmetanog protoka morske vode koji omogućuje njihov uspješan rast i razvoj. Vrste *Talochlamys multistriata*, pronađena su u velikom obilju i mogla bi biti dobar kandidat za diverzifikaciju proizvodnje akvakulture, čime bi uzgajivačima potencijalno ponudili dodatni izvor prihoda.

Ključne riječi: uzgoj školjki; diverzifikacija; eksperimentalni kolektori; jakobova kapica; *Pecten jacobaeus,* Bokokotorski zaljev