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Research Article

Seasonal Variation of Zebra Mussel (*Dreissena polymorpha* Pallas, 1771) Colonization on Turkish Narrow-Clawed Crayfish (*Astacus leptodactylus* Eschscholtz, 1823) in Lake Eğirdir, Turkey

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ABSTRACT

In this study, the hosting status of *Astacus leptodactylus,* which has one of the highest economic value of the inland waters fisheries of Turkey, dependent on zebra mussels (*Dreissena polymorpha*) was studied. Seasonal and sexual differences of zebra mussel prevalence, density situation on crayfish and some morphometric properties of zebra mussels retrieved from host were determined. Field works were carried out in January, April, July and October of 2015 with 8 fishing operations. 200 fyke net with different mesh sizes were used in sampling process. Sex of the crayfish and if they have mussels on them were determined by organoleptic observation. *Mann-Whitney U, Kruskal Wallis* and *Chi-Square* tests were used for evaluating of nonparametric data. A total of 4024 crayfish were caught and *D. polymorpha* was determined on 624 of them. The annual prevalence was found as 15.51% for both sex groups and it is higher in female (19.81%) than in males (9.82%). The highest mean abundance was found to be 1.93 in the spring season. The mean weights of the mussels were 0.2277 g and the average shell size was 12.3802 mm indicating a strong exponential relationship between weight and shell size.

Keywords: Invasive alien species, freshwater ecosystems, aquatic invasions

INTRODUCTION

The freshwater zebra mussels *D. polymorpha* have become one of the most dominant species in many lakes and rivers of Europe, since it started spreading from the Caspian area at the beginning of the 19th century (Stanczykowska, 1977). It is a relatively small mollusk (25-34 mm long) which is native to the Black Sea basin and can form dense colonies in the hard and soft substrates of waterbodies. Over the last decades, the zebra mussel has invaded and spread across Eurasia and North America (Higgins and Vander Zanden, 2010). Some agencies such as fishing equipment ballast water, boat hulls, aquarium dumping, and perhaps by waterfowl has mediated to introducing successfully to several continents of *Dreissena* sp. (Reynolds and Donohoe, 2001).

Zebra mussels have a short maturation time (1-2 years), as well asthe ability for high dispersion and a really high fecundity (large number of eggs produced) (Higgins and Vander Zanden, 2010). Zebra mussels have high filtering capability and their seasonal production of free-swimming veliger larvae which will attach to hard substrates with their byssus, a strong filamentous structure secreted by bivalves. This species has caused much disruption both economically and ecologically on unexploited freshwater system of the worldwide (Reynolds and Donohoe, 2001).

Monotypic colonization is the typical characteristic of zebra mussels. Once established, zebra mussels have proven nearly impossible to remove. Therefore, their control is of considerable interest (Reynolds and Donohoe, 2001; Johnson and Carlton, 1996). Zebra mussels are physiologically strong organisms. They live longer than many other bivalves. An environment like Great Lakes which has the greatest diversity of freshwater bivalves with 297 species (Ricciardi et. al., 1998) and constitutes an extremely suitable habitat for zebra mussels. *D. polymorpha* shares characteristics of many successful invasive species: rapid growth, prolific reproduction at an early age (0-1+) and a relatively short life span. This usually provides the potential for rapid exponential growth of zebra mussel populations within a waterbody (Frances, 2006). This suggests that juveniles are capable of making decisions of where to colonize. One factor in deciding where to colonize is the threat of predation. Interior substratum is not suitable for Dreissenids using of protection and live, but rocky substrata is a very good alternative for that intended use (Thorp et. al., 1998). There are many zebra mussels predators in North American waters, for instance crayfish by Stewart et. al., (1998) and sunfish by Molloy et al. (1997) and Magoulick and Lewis (2002). Predator and prey interactions are very strongly in the biological communities (Polis and Strong, 1996).

In the previous studies, which were carried out on crayfish and zebra mussels relationship, (*I*) the connection location of zebra mussel on the crayfish was researched by Berber et al. (2018) and (*II*) predation effect of crayfish on the zebra mussels was investigated (Martin and Corkum ,1994; Perry et al. 1997; Reynolds and Donohoe, 2001; Kutluyer et al., 2013). However, there are no studies connected with the hosting status of natural crayfish stock for invasive zebra mussels by evaluating of seasonally.

We aimed to determine the seasonal and sexual differences of Zebra Mussel prevalence and density on crayfish, and also investigated some biometric characteristics of zebra mussels which were extracted from *A. leptodactylus*.

MATERIAL AND METHOD

The fieldworks were conducted in January, April, July and October of 2015 with 8 fishing operations. 200 fyke net with 14, 26, 34 and 42 mm mesh size were used in the fishing trial. Fyke nets were harvested once every two days. The sex of the crayfish and whether or not they carried mussels on them were determined by organoleptic observations. Morphometric measurements of crayfish were determined with 0.01mm sensitivity using digital caliper according to Rhodes and Holdich (1984). In the evaluating of nonparametric data *Mann-Whitney U, Kruskal Wallis* and *Chi-Square* tests were used. In addition, Dunn's test was utilized for multiple comparisons. *Pearson Correlation* test was used to determine the relationship between the morphometric measurements of crayfish and the total number of Dreissena they carry. *R* (v3.3.3) based *RStudio* (v1.0136) program was used for all statistical calculations. Prevalence, mean intensity and mean abundance values have been modified from Bush et. al., (1997) as follows.

$$\textit{Prevalence}=\frac{\text{Number of crayfish with mussel}}{\text{Number of total caught crayfish}}\times 100$$

$$\textit{Mean Intensity}=\frac{\text{Number of total mussel}}{\text{Number of crayfish with mussel}}$$

$$\textit{Mean Abundance}=\frac{\text{Number of total mussel}}{\text{Number of total caught crayfish}}$$

RESULT AND DISCUSSION

Of the 4024 crayfish examined, Dreissena was seen at 624 (454 female and 170 male), but not at 3400 (1838 female and 1562 male). There is statistical difference between sex groups by hosting or not ($p<0,05$) (Table 1). Minimum, maximum and mean number of zebra mussel collected from crayfish are given Table 2. Statistical differences were found between male and female crayfish with regard to the carrying of zebra mussels ($p<0,05$); presence of Dreissena on females is higher than on males. The annual prevalence was found to be 15.51 but showed great seasonal differences. The prevalence was highest in female in the spring season (%44.99). The host situation of crayfish for Dreissena was of minimum level in the autumn and winter months (Table 4). Mean density has shown statistical differences between sampling seasons ($p<0,05$). Mean abundance was highest in the spring months. It is thought that the cause of this density is due

Table 1. Hosting status of crayfish by different sex groups

Sex Groups	Dreissena (+)	Dreissena (-)	X^2	*p*
♀♀	454	1838	75,185	0,000
♂♂	170	1562		

Table 2. Mean density of D. polymorpha which are hosted by A. leptodactylus

Sex Groups	Mean±SE	*Min. (n)*	*Max. (n)*	*Man.Whit (U)*	*Wilcoxon (W)*	*p*
♀♀	4.41±0,24	1	43	32934	47469	0,004
♂♂	3.56±0,33	1	28			

*Calculations were conducted only host crayfish

Table 3. *D. polymorpha* presence on *A.leptodactylus* by season and sex groups

Season	Sex Groups								
	♀♀			♂♂			♀♀+♂♂		
	+	-	Prv. (%)	+	-	Prv. (%)	+	-	Prv. (%)
Spring	368	450	44.99	123	199	38.20	491	649	43.07
Summer	86	772	10.02	45	516	8.02	131	1288	9.23
Autumn	-	81	-	2	76	2.56	2	157	1.26
Winter	-	535	-	0	771	0.00	-	1306	-
Annual	454	1838	19.81	170	1562	9.82	624	3400	15.51

(+: . *D. polymorpha* positive, -. *D. polymorpha* negative, *Prv:* Prevalence)

Table 4. Mean density and abundance of D. polymorpha which are hosted by A. leptodactylus

Season	Total Number of		Total Caught Crayfish	Mean Density	Mean Abundance	Number range of Dreissena	
	Host Crayfish	Dreissena				Min. (N)	Max. (N)
Spring	491	2203	1140	4.49 ± 0.23^{ab}	1.932	1	43
Summer	131	384	1419	2.93 ± 0.34^{a}	0.270	1	28
Autumn	2	20	159	10.0 ± 9.00^{b}	0.125	1	19
Winter	0	0	1306	0	0	0	0
Total	624	2607	4024	4.177	0.647	0	43

Table 5. Presence of mean density *D. polymorpha* on crayfish by length class

Length Groups	n	Dreissena (+)	Dreissena (-)	Mean±SE	Min.-Max.
3-6	21	21	140	2.10 ± 0.42^{a}	1-9
7-9	543	543	2380	4.25 ± 0.21^{b}	1-43
>10	60	60	879	4.25 ± 0.61^{ab}	1-19

to the fact that Dreissenan and crayfish life cycle are not in the period of shell change. Mean density was calculated as 10.0 for the autumn season as highest value. It is thought that, this result stemming from very poor data which gained for autumn season.

The presence of *D. polymorpha* on crayfish has been shown differences by length class (X^2: 81,325) (p<0,05). It is said that, small length class of crayfish were less sensitive then middle and big length class in regard to Dreissena invasion. The highest prevalence in the female was found in the 6 cm length group with 25.938% and in the males was found in 5 cm length group with 13.636%. The highest prevalence in the calculation for both sex groups was 21.023% in the 6 cm length group (Table 6). In general, the prevalence of the individuals above the 10 cm group was significantly lower than the other groups. In general, prevalence of female individuals was higher than male individuals.

Minimum and maximum number of *D. polymorpha* which were isolated from each crayfish and values of mean density and abundance are given in Table 7. It was determined that, while the number of *D. polymorpha* isolated from female crayfish was minimum 1 and maximum 43 (an individual in the 7 cm size group) in males, min 1 and maximal 28 (an individual in the 8 cm size group).

It is seen that the prevalence is very high in 6,7,8 cm length classes according to advanced length classes. The main reason for this situation is thought to be catching composition-oriented. Because the young crayfish constitute a significant portion of the prey in this group the prevalence is higher than others. In general, prevalence of female individuals was higher than male individuals. This situation can be explained by the fact that the males who tend to grow faster than females have changed the shell before the sampling period and thus they are free from the Dreissena on them.

The average density and abundance of *D. polymorpha* in female and male crayfish showed significant differences according to the length classes (p<0,05). In general, the average intensity in malesincreases in parallel to the length increase. *D. polymorpha* was not found in female crayfish above 10 cm and in male crayfish above 13 cm. Prevalence was observed more in small size

Table 6. *D. polymorpha* presence on *A. leptodactylus* by length class and sex groups

Length Class	Sex Groups								
	♀♀			♂♂			♀♀+♂♂		
	+	-	Prv (%)	+	-	Prv (%)	+	-	Prv (%)
3					1			1	
4	1	13	7.143		8		1	21	4.545
5	11	62	15.068	9	57	13.636	20	119	14.388
6	83	237	25.938	28	180	13.462	111	417	21.023
7	185	561	24.799	65	462	12.334	250	1023	19.639
8	139	479	22.492	43	461	8.532	182	940	16.221
9	25	255	8.929	16	167	8.743	41	422	8.855
10	10	100	9.091	4	87	4.396	14	187	6.965
11		64		4	64	5.882	4	128	3.030
12		37			42			79	
13		23		1	21	4.545	1	44	2.222
14		7			9			16	
15					3			3	

(+: . *D. polymorpha* positive, -. *D. polymorpha* negative, *Prv*: Prevalence)

Table 7. Mean density and abundance values by different length class and sex

Length Class	♀♀					♂♂				
	Min.	Max.	Sum	Mean Density	Mean Abundance	Min.	Max.	Sum	Mean Density	Mean Abundance
3										
4	9	9	9	9.000	0.643					
5	1	4	18	1.636	0.247	1	4	17	1.889	0.258
6	1	21	332	4.000	1.038	1	12	88	3.143	0.423
7	1	43	852	4.605	1.142	1	18	188	2.892	0.357
8	1	31	683	4.914	1.105	1	28	165	3.837	0.327
9	1	17	93	3.720	0.332	1	19	74	4.625	0.404
10	1	4	15	1.500	0.136	5	18	38	9.500	0.418
11						5	10	30	7.500	0.441
12										
13						5	5	5	5.000	0.227
14										
15										

groups as Dreissena was common in seasons when the crayfish were small in size (Table 7).

In female crayfish, there is an increase in both the number of host crayfish and the number of hosts starting from the 5 cm height class. A similar situation is observed in male crayfish. This increase has a tendency to decrease by making a 7 cm height class peak and after the 10 cm length class, both the number of host crayfish and the number of hosts decrease significantly. A similar situation is observed in male crayfish.

The correlation matrix of relations between some body measurements of crayfish and the *D. polymorpha* numbers which they carry is given in Table 8. In general, the relationship between the number of zebra mussels on the body and the different body lengths was found to be very weak.

The Dreissena keep on crayfish at the end of the winter months and spring months continue to grow on crayfish until July until beginning of shell change period. Figure 1(A) and (B) shows Dreissena colonization on crayfish and Figure 4(C) shows some examples of different sized Dreissena isolated from crayfish.

The spread and overgrowth of zebra mussels which are defined as an aquatic invasive species in its settling environment has become an important ecological problem for all countries around

Table 8. Correlation matrix between number of the *D. polymorpha* and measured body part of *A. leptodactylus*

Correlations	Total Number of *D. polymorpha*	Total Length	Carapace Length	Abdomen Length	Carapace Width	Abdomen Width
Total Number of ***D. polymorpha***	1					
Total Length	0.066	1				
Carapace Length	0.056	0.977	1			
Abdomen Length	0.069	0.974	0.903	1		
Carapace Width	0.063	0.957	0.922	0.946	1	
Abdomen Width	0.089	0.918	0.860	0.933	0.898	1

the world. According to Bobat et al. (2001), it is estimated that the spread of the zebra mussel to the other major continents from the natural spreading areas is through the bilge (ballast) waters of the ships. Access to other water sources at the places of transportation is due to their ability to attach themselves to boats. But it is not a question of intercontinental spread to Turkey. The zebra mussels naturally found in the Euphrates Basin (East-South Turkey) and breeding below the damage limit have grown extensively by finding a more suitable environment (temperature, pH, calcium, nutrients, adhesive surface, etc.) to breed due to the changing ecological conditions after dam construction. In addition, the pouring of domestic waste into the Atatürk Dam Lake increased the phosphate content in the aquatic environment, which also played a nurturing role for the mussels (Bobat et al., 2001). Another element of dissemination way may be commercial fishermen by means of carrying fishing gears from one fishing location to the another. For example, gillnets, trammel nets, crayfish fyke-nets and beach seine net etc. In addition, the sales of second-hand boats widely held in the inland waters of Turkey. Zebra mussels cause deterioration of the water quality and in the food chain in the lakes (Noonburg et al., 2011) where they are oriented towards eutrophication, which cause irreversible damage in the long run. For this reason, there is need for studies on the distribution and overproduction of mussels. Ardura et al. (2017) have developed a species-specific environmental *DNA* marker for early detection of a global invasion problem, zebra mussel, and consequently report that the method is consistent, reproducible, rapid, inexpensive and technically easy.

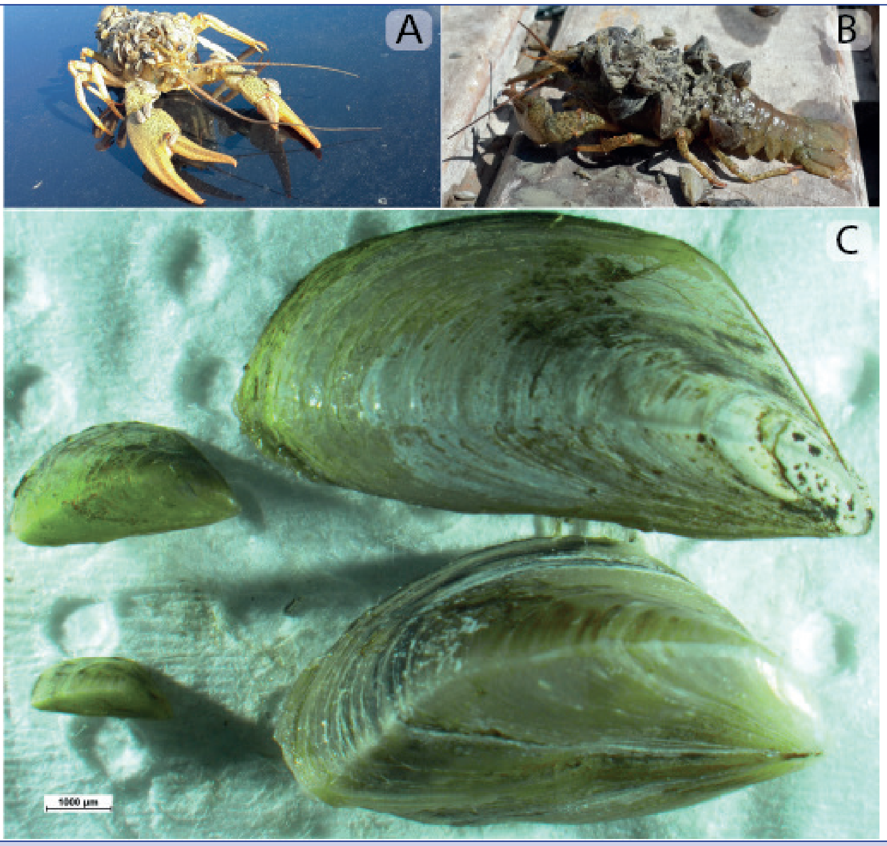

Figure 1. Different sized Dreissena samples isolated from crayfish.

It is thought that the prevalence of female individuals is higher than that of male individuals, which is generally due to the fact that male individuals with better growth performance are more likely to molt than females. Generally male crayfish are more frequently molting than females during the year are in the higher prevalence than females compared to males because they are removed from the Dreissena on them end of the moulting process.

According to Bolat (2001), male crayfish can molt 45-50 times a year and females 30-35 times (Groves, 1985; Alpbaz, 1993). In Turkey, it has been reported by some researchers that the crayfish species *Astacus leptodactylus* start to molt in June. It has been reported that immature individuals molt 8 times in the first year, 5 times in the second years and 2-3 times in the third year, after maturation males molt twice a year and females only once a year (Atay, 1984; Anonymous 1985; Erdem, 1993; Bolat, 2001).

Brazner and Jensen (2000), have reported that the number of zebra mussels per crayfish ranged from 16 to 431, attached zebra mussels ranged in size from 1.2 mm up to 12.0 mm, with a mean size of 3.6 mm. The number of colonized individuals, length range of Dressenia and number of zebra mussels for per crayfish infestation were a bit smaller than our result. This may be related to the relative motility and/or moulting characteristics of these different organisms. This kind of works can give some results to countries, understanding of seriousness of the situation and spend the budget for the struggle.

CONCLUSION

Zebra mussels cause many ecological and economical dangers in aquatic areas. Dressenia and crayfish interactions have been unexplained up until now except prey-predator relationship. In this context, recently increasing of the crayfish production in Eğirdir Lake may derive from Dreissenia based feeding regime of crayfish. Zebra mussels present crayfish a with very good camouflage ability to protect them from predators in addition to a potential food relationship. However, there are some observed dangers of zebra mussels on crayfish. Such as, (I) using extra energy for carrying of penetrated Dreissena during move in habitat and (II) deteriorating of the fyke net selectivity. While most crayfish with a normal body shape (below the minimum landing size) can easily escape the fyke net, carrying of the penetrated zebra mussel on the shell makes escaping the fyke net almost impossible. New studies are needed, especially focus on the escape of caught crayfish (carrying the zebra mussel on its shell) from the fyke net easily for sustainable stock management.

Conflicts of interest: The authors have no conflicts of interest to declare.

Ethics committee approval: This study was conducted in accordance with ethics committee procedures of animal experiments.

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