

*PODONEVADNE TRIGONA OVUM* (ZERNOV, 1901)  
(CLADOCERA), AN IMMIGRANT SUBSPECIES WITH CASPIAN  
ORIGIN, NEWLY PENETRATED IN ROMANIAN WATERS

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The analysis of zooplankton in the Danube-Black Sea Canal revealed the presence of a new species for the Romanian fauna: Caspian cladoceran *Podonevadne trigona ovum*. Its appearance occurred at the end of the first year of the ecosystem (1985). In this phase of the canal colonization, its presence was limited to the mixing zone of marine and freshwaters. Further, its adaptation at the ecological conditions of the new ecosystem had major difficulties, which led to its disappearance from the zooplankton community structure in the next year (1986). The renewal of the studies after two decades (2005) reveals that the immigrant overcomes these difficulties, populating the entire biotope, except the contact area with the river waters. The analysis of the frequency and abundance of the immigrant shows that it became one of the main components of the predator zooplankton. The invasive behavior of some species belonging to Onychopoda Infraorder, resulted in significant changes in the structure of some ecosystems, suggests the usefulness of monitoring the evolution of *Podonevadne trigona ovum* in the Danube-Black Sea Canal.

*Key words:* *Podonevadne trigona ovum*, immigrant species, zooplankton, predator, parthenogenetic, gamogenetic, resting eggs.

#### INTRODUCTION

The researches focused on the forming and development processes of the zooplankton community in the Danube-Black Sea Canal have revealed the presence of a new species for the Romanian waters: the cladoceran *Podonevadne trigona ovum* (Zernov, 1901).

The origin of the cladocerans fauna belonging to the Onychopoda Infraorder, which includes the mentioned species, is located in the Caspian basin. The particular ecological characteristics of this group reflect, to a significant extent, the complexity of the genesis of this basin.

This process starts in Miocene, 5 million years ago. The formation of a deep depression area with tectonic origin in the south-eastern extremity of the Sarmatian Lake, led to the primordial Caspian basin. Its location coincides with the southern end of the current pool.

The new tectonic events created conditions for further expansion of the primary basin in an area corresponding to the middle sector of the current basin.

The strong orogenetic movements registered at the end of the Miocene resulted in the fragmentation of the Sarmatian Lake in two separate pools: the Caspian and Pontic region.

The important hydrological contribution of some tributary waters determined, finally, the emergence of the northern sector. Unlike the middle and especially the southern ones, the northern sector is characterized by low depth and salinity (Dumont, 1998; Rivier, 1998).

Repeated transgressions and regressions recorded in the Pliocene, during 2 million years, have generated major changes (within 25–120 m) of the water levels.

The registered increases in the transgression phases have produced the considerable expansion of the Caspian basin. Therewith, there were created the conditions to establish some temporary communication channels with the Azov, Pontic and Aral basins.

In interglacial periods, the melting of glaciers also caused a considerable increase of water flow from the northern tributaries of the basin. A consequence of this fact was the establishment of a temporary communication route between the Caspian and Baltic basin.

The long geographical isolation of the Caspian basin created a preponderant endemic fauna and regional and temporal variations in salinity have been an important factor of its speciation.

The ability of species to live in waters with different concentrations of salinity could be improved during the temporarily emergence periods of the connection lines with other basins. It is remarkable that some of these species currently maintain the tendency to populate new ecosystems.

The Infraorder Onychopoda includes 33 species and subspecies. Currently, two prefer freshwaters, 7 marine waters and 24 are brackish species. The first two ecological categories no longer reflect the structure of the fauna of the Caspian basin. In the same situation, there are two brackish species. One of these is *Podonevadne trigona ovum*. As a result, currently only 46% of species remain endemic any longer, 21% inhabit both native basin and other basins, and 33% have disappeared from the structure of the cladoceran fauna of the mentioned basin.

In the first stage, the saltwater species left the Caspian basin and entered the Azovo-Pontic, Aralic or Baltic basin. In the next stage, some of them colonized more distant seas, such as the Mediterranean Sea or the North Sea, and eventually spread in the Atlantic, Pacific and Indian Oceans. Integrated in the biocenotic structure of the new ecosystems, this species became dominant, due its high prolificity and short generation time.

These are the species *Evadne nordmanni* from the Baltic Sea, or *Pseudoevadum tergestina*, *Evadne spinifera*, *Podom intermedius* and *Pleopis polyphemoides* that inhabit different areas of the planetary ocean (Zenkevich, 1963; Gieskes, 1970; Della Croce & Venugopal, 1972; Longhurst & Seibert, 1972; Brosch & Taylor, 1973; Onbe, 1974; Thiriot, 1974; Kos, 1977; Frontier, 1979).

The colonization of the remote areas has been achieved mainly through resting eggs transported by birds. A wide and circumpolar spread, favored by the existence of a short life cycle and a high prolificacy, exhibits also the freshwater species *Polyphemus pediculus* (Ekman, 1904; Keilchack, 1906; Butorina, 1963, 1971, 1993).

In a recent period, the anthropogenic factor also exerted an important role in the spread (active or passive) of some species, as a result of the construction of canals, reservoirs, intensive practice of shipping or the cultivation of alien species. A surprisingly, fast and wide spread was recorded for freshwater species *Bytotrephes longimanus*. It recently settled (during 1970-1980) the great lakes of North America, where it got the ballast water of ships. In Lake Michigan, even if it is not very abundant, this species has already produced changes in the structure of planktonic community and fisheries (Marlene, 1990). It also maintains the role of dominant species in the zooplankton of some large lakes in the manner recorded in Europe half a century ago (Gerd, 1947; Drako & Stetsenko, 1956; Fenyuk, 1960; Mordukhai-Boltovskaya & Mukhordova, 1970; Glamadza, 1971; Buschman, 1978).

Among the cladocerans of the Onychopoda Infraorder that currently continue to expand, there are listed some low brackish water species, also tolerant in freshwater, such as *Cercopagis pengo*, *Corniger maeoticus* or *Podonevadne trigona ovum*. In their case, the construction of canals and reservoirs represented an opportunity to expand their habitat in tributary waters of the Black, Azov and Caspian Seas (Rivier, 1998).

Researches carried out in the last half century evidenced the entering of some immigrant species belonging to Onychopoda in the Romanian Black Sea sector (*Podon leuckarti*, *Pleopis polyphemoides*, *Pseudoevadne tergestina*, *Evadne nordmanni*, *E. spinifera*, *Cercopagis pengoi* and *Cornigerius maeoticus*). The last two species were also found in some lakes with marine or Danube floodplain origin (Mordukhai-Boltovskoi & Negrea, 1965; Negrea, 1983). The extending process of some species in the Romanian spaces continues in the current period. *Podonevadne trigona ovum* is the cladoceran species that recently entered in the Danube-Black Sea Canal.

In the opinion of the specialists, the genesis of species occurred 77.000 years ago, during a major transgression (Zernov, 1901). It was adapted to environments with low salinity or freshwaters, and colonized the outskirts of huge flooded areas in the northern part of the Caspian basin. The flooding of the Manych depression area, which temporarily stopped the isolation phase of the Caspian basin, has allowed its migration in the Azov basin. As a result of the flooded area stint and the break of the contact with Azov basin during the following regression period, *Podonevadue trigona ovum* disappeared from the Caspian basin. Instead, it has maintained its presence in the Azov Sea and Chalkar Lake, located within the former flooded areas (Behning, 1928).

In the next phase, it came in the Black Sea, where successively populated the lagoons of Bug, Dnieper and Dniester rivers. The construction of the Volga-Don canal and some reservoirs of the Volga course, Don, Dnieper and the Dniester in the period 1930-1975 provided the species a new opportunity to populate other ecosystems (Tseeb, 1962; Mordukhai-Boltovskoi & Galinskii, 1974; Gusynskaya, 1989).

### MATERIAL AND METHODS

The researches were conducted during 1985–2005 in the Danube-Black Sea Canal. In the hierarchy of Earth's major channels it ranks third (after Panama and Suez) and is located at 44°11'46.78" N latitude, 28°39'46.62" E longitude. Its main branch (Danube km 299 – Agigea Seaport) has a length of 64 km, and the secondary one (Poarta Albă-Midia Năvodari Seaport) has 31 km (Fig. 1).

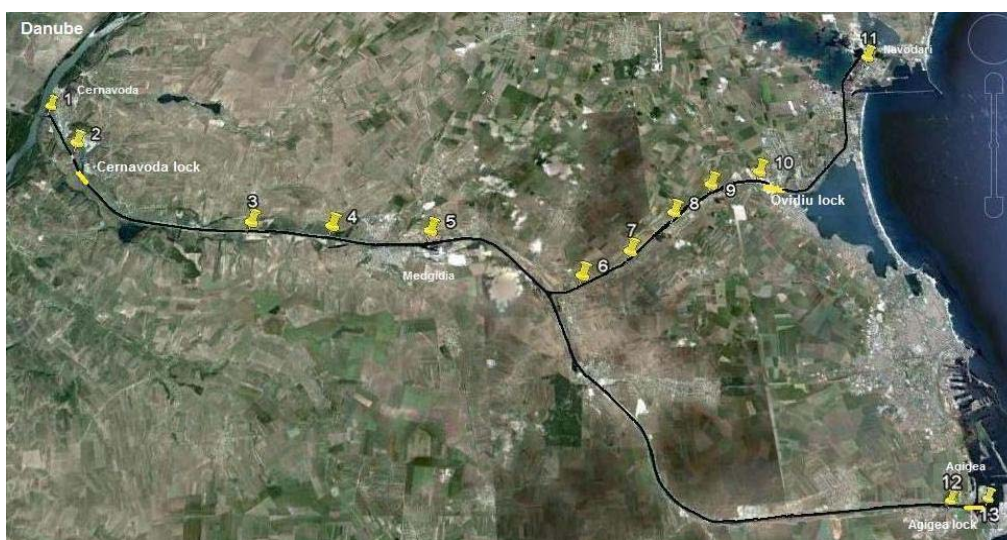


Fig. 1. The map of Danube – Black Sea Canal – the sampling sites of zooplankton (www.earthgoogle.com).

The minimum depth of two branches is 7 and 5.5 m, respectively, their width varies between 140 and 100 m, and the flow of water transit between 0.3 to 0.9, respectively from 0.13 to 0.23 m<sup>3</sup>/s.

The sampling of zooplankton in the canal was carried out, initially, in 4 stations in 1985 and 11 sampling points, in 2005 (Table 1) with seasonal periodicity. To this goal we used a Schindler Patalas device with 5-liters capacity. For each sample, there were taken 50 liters of entire water column. The samples were concentrated by filtering through a plankton net with a mesh of 65 mm. The preservation of samples was done with 4% formaldehyde solution.

Table 1

Sampling sites of zooplankton

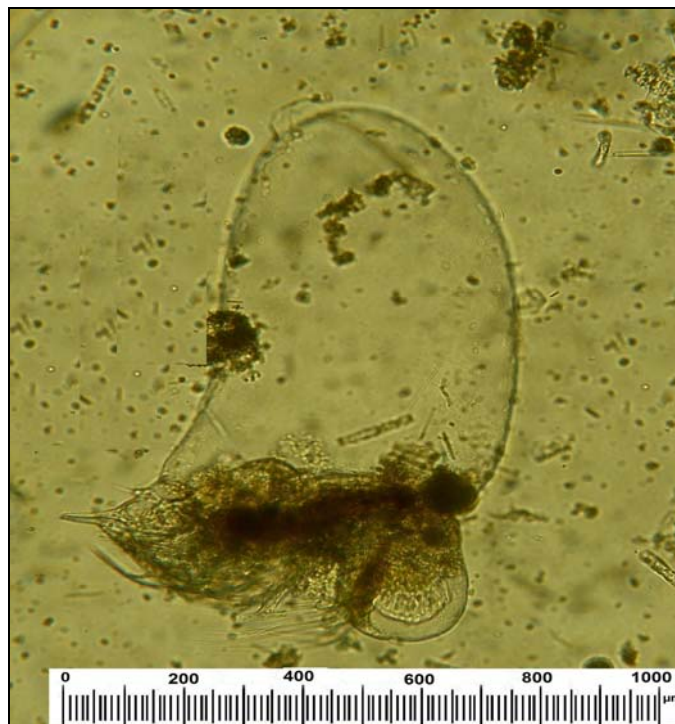
No st.	Canal km	Period	Geographic landmark	Canal section
1	64+000	2005	Cernavodă City	Danube – Black Sea
2	61+000	2005	Upstream Cernavodă lock	Danube – Black Sea
3	47+600	2005	Mircea Vodă	Danube – Black Sea

Table 1

(continued)

4	40+000	2005	Medgidia City	Danube – Black Sea
5	37+900	1985-1986, 2005	Medgidia City	Danube – Black Sea
6	28+700	2005	Poarta Albă City	Danube – Black Sea
7	23+000	2005	Basarabi City	Danube – Black Sea
8	20+700	2005	-	Poarta Albă – Năvodari
9	15+100	2005	-	Poarta Albă – Năvodari
10	10+000	2005	Downstream Ovidiu lock	Poarta Albă – Navodari
11	2+000	2005	Navodari City	Poarta Albă – Năvodari
12	0+000	1985 -1986	Upstream Agigea lock	Danube – Black Sea
13	-	1985 -1986	Black Sea	Agigea Zone

The material analysis revealed the presence of a new cladoceran species for the Romanian waters: *Podonevadne trigona ovum* (Fig. 2). Based on data obtained from the analysis of zooplankton samples (Tables 2-4), we could determine where and when entering the immigrant in canal, its abundance frequency (Table 5) and the prevalence in the numerical structure of the total (Table 6) and predator zooplankton (Table 7).

Fig. 2. *Podonevadne trigona ovum* (orig.).

### RESULTS AND DISCUSSION

The low velocity of water flow, the heterogeneous nature of the environment, caused by the presence of submerged and emerged macrophytes in coastal area, the river water intake and the influence of the marine terminal area of the canal, have created favorable conditions for the existence of zooplankton.

The low values of species richness (Fig. 3) and abundance (Tables 2-4), recorded in the first year of existence of the ecosystem (1985), showed significant increases over the next two decades.

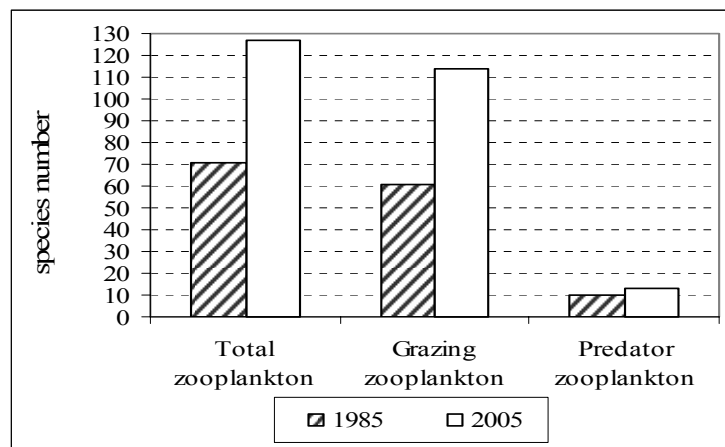


Fig. 3. The dynamics of zooplankton species richness on trophic levels (species number).

Table 2

The dynamics of total zooplankton abundance (ind./l)

Station	V	VI	X	XI	Xa V-XI
1985					
2	42.2	34.6	10.0	4.8	22.9
5	1067.0	102.1	203.2	37.3	352.40
12	260.2	34.4	60.6	3.0	89.55
13	29.2	52.8	34.7	2.1	29.70
Xa	349.65	55.98	77.13	11.80	123.64
2005					
Station	III	VIII	IX	XII	Xa III -XII
1	7.3	156.8	4.9	13.7	45.67
2	13.2	139.7	5.34	3.7	40.48
3	13.3	196.7	262.5	2.0	118.62
4	7.1	456.7	266.96	12.3	185.76
5	10.4	412.1	299.7	22.0	186.05
6	4.3	1131.6	372.2	0.7	377.20
7	1.9	1044.0	258.6	1.8	326.75
8	109.3	1825.4	462.7	3.1	600.12

Table 2  
(continued)

9	5.3	1538.5	227.9	5.43	444.28
10	213.5	1387.6	1108.8	655.8	841.43
11	533.7	710.8	369.0	248.6	465.52
Xa	83.57	818.17	330.78	88.10	330.16

Table 3

The dynamics of herbivore zooplankton abundance (ind./l)

Station	V	VI	X	XI	Xa V-XI
1985					
2	42.2	33.4	10.0	4.8	22.6
5	1067.0	91.32	203.2	34.0	348.88
12	258.1	31.9	54.8	2.2	86.75
13	26.1	50.5	23.0	1.0	25.15
X a	348.35	51.78	72.75	10.5	120.85
2005					
Station	III	VIII	IX	XII	Xa III-XII
1	7.3	154.7	3.8	13.7	44.87
2	13.0	135.7	5.0	3.5	39.30
3	13.3	180.9	256.6	2.0	113.2
4	7.1	446.2	264.7	12.3	182.57
5	10.4	386.5	287.3	22.0	176.55
6	4.3	1049.3	370.6	0.7	356.22
7	1.9	1001.8	245.5	1.8	312.75
8	109.3	1766.7	451.7	3.1	582.7
9	5.3	1482.0	225.7	4.93	429.48
10	213.5	1283.1	1099.2	654.4	812.55
11	533.7	671.7	357.0	248.6	452.75
Xa	83.55	778.05	324.28	87.91	318.45

Table 4

The dynamics of predator zooplankton abundance (ind./l)

Station	V	VI	X	XI	Xa V-XI
1985					
2	-	1.2	-	-	0.3
5	-	10.78	-	3.3	3.52
12	2.1	2.50	5.8	0.8	2.8
13	3.1	2.30	11.7	1.1	4.55
Xa	1.3	4.24	4.2	1.3	11.17
2005					
Station	III	VIII	IX	XII	Xa III -XII
1	-	2.1	1.1	-	0.80
2	0.2	4.0	0.34	0.2	1.18
3	-	15.8	5.9	0.2	5.47
4	-	10.5	2.26	-	3.19

Table 4  
(continued)

5	-	25.6	12.4	-	9.50
6	-	82.3	1.6	-	20.97
7	-	42.2	13.1	-	13.83
8	-	58.7	11.0	-	17.43
9	-	56.5	2.2	0.5	14.80
10	-	104.5	9.6	1.4	28.77
11	-	39.1	12.0	-	12.78
Xa	0.02	40.12	6.5	0.21	11.70

Table 5

The dynamics of the *Podonevadne trigona ovum* frequency (%)

Year	Months	%	Ecological significance
1985	V	-	-
	VI	-	-
	X	25.00	accidental species
	XI	-	-
	Xa V-XI	6.25	accidental species
2005	III	9.09	accidental species
	VIII	81.82	persistent species
	IX	54.54	persistent species
	XII	-	-
	Xa III-XII	36.36	accessories species

Table 6

The dynamics of the percentage of *Podonevadne trigona ovum* in the total zooplankton abundance (%)

Year	Station	Months			
		V	VI	X	XI
1985	2	-	-	-	-
	5	-	-	-	-
	12	-	-	1.81	-
	13	-	-	0.30	-
2005	Station	III	VIII	IX	XII
	1	-	-	-	-
	2	2.26	-	-	-
	3	-	0.71	-	-
	4	-	1.36	-	-
	5	-	2.69	3.20	-
	6	-	2.89	0.05	-
	7	-	2.02	2.74	-
	8	-	1.05	1.21	-
	9	-	3.01	0.96	-
	10	-	0.49	-	-
	11	-	0.12	0.54	-
	Xa 1-11	0.21	1.30	0.79	-



Table 7

The dynamics of the percentage of *Podonevadne trigona ovum* in the predator zooplankton abundance (%)

Year	Station	Months			
		V	VI	X	XI
1985	2	-	-	-	-
	5	-	-	-	-
	12	-	-	23.40	-
	13	-	-	0.86	-
2005	Station	III	VIII	IX	XII
	1	-	-	-	-
	2	100	-	-	-
	3	-	8.86	-	-
	4	-	59.04	-	-
	5	-	64.45	77.42	-
	6	-	30.62	12.50	-
	7	-	50.00	54.20	-
	8	-	32.71	50.91	-
	9	-	82.12	100	-
	10	-	6.51	-	-
	11	-	2.05	16.67	-
Xa 1-11	9.09	30.58	26.82	-	

The taxonomic analysis of the zooplankton recorded the emergence of *Podonevadne trigona ovum* in the Danube-Black Sea Canal at the end of the first year of ecosystem existence (1985), in the terminal sector of the canal (upstream, Agigea sluice). This is a subspecies of *Podonevadne trigona* species, belonging to the *Podonevadne* genus, *Podonidae* family, *Onychopoda* infraorder, *Cladocera* suborder (Boxshall, 2001; Martin & Davis, 2001). The concomitant presence of species in the adjacent marine coastal zone (Table 1) offered an explanation for the source of the immigrant in the canal. In the structure of zooplankton constituted in the first year of ecosystem existence the immigrant cladoceran presented the characteristics of accessories species (Table 5).

In this phase, the adaptation to the ecological conditions of the new ecosystem encountered major difficulties, which could not be overcome. As a result, the species could not be identified in the structure of zooplankton in the following year (1986).

The renewal of the studies in 2005 noted the recurrence of *Podonevadne trigona ovum*. Its presence in 10 of the 11 analyzed sampling sites in this year reflected the upturn in the process of adaptation of the species to the ecological conditions of the ecosystem. Its absence could be recorded only in the confluence area between the fluvial and canal waters. The avoiding of the fluvial environment is a feature also noted in other onychopod species regarded as typical planktonic organisms. With high capacity of buoyancy and diminished adaptations for active moving, they show positive selectivity only for the current free waters (Egloff *et al.*, 1996).

The dynamics of the frequency index of the species showed significant differences during the annual cycle of life. In the early spring the presence of the cladoceran in our samples was relatively low (9.09%), corresponding to the accidental species type. Instead, in early autumn and, especially in late summer, there were recorded high values (54.54% and 81.82%) (Table 6), reflecting the constant status of species and holding a significant role in the structure of the zooplankton in canal.

The populational structure of the immigrant cladoceran was formed, exclusively, by the parthenogenetic females (Fig. 2). The dominance of the parthenogenetic females is a common situation for the cladocerans living in permanent aquatic ecosystems characterized by favourable environmental conditions (Berg, 1936). The gamogenetic females and the male individuals are characterized by a much smaller presence, usually located in the terminal life cycle (from October to December) or during unfavourable ecological periods. Their proportion is significantly higher in the temporary aquatic ecosystems (Negrea, & Negrea, 1975; Negrea, 1983).

The comparative analysis of the parthenogenetic females from the Azov Sea (Mordukhai-Boltovskoi & Negrea, 1965) and the Danube-Black Sea Canal revealed the existence of similar dimensions of body length limits. In contrast, the height and volume of the brood poach were clearly higher for the canal specimens. They express the existence of some particular ecological conditions.

For the species belonging to *Podonevadne* genus, characterized by reduced possibilities of movement, the brood poach has an important hydrostatic role. In the dense environment of salt water the volume and height of brood poach is relatively low and clearly higher in freshwater, where water density is lower. For the species *P. camptonyx* and *P. angusta* (whose optimum salinity of water varies within 11 to 13.5 ‰) the brood poach is narrow and less bulky. For *P. trigona trigona* (optimal salinity 3–8 ‰) the brood poach is wider and slightly higher (Rivier, 1998). The population of *P. trigona ovum* (optimal salinity 3–4 ‰) from the Azov Sea have a globular shaped brood poach. It is much higher and the protective valves are provided with deformations that increase the floating ability (Mordukhai-Boltovskoi & Negrea, 1965). The population of the Danube-Black Sea Canal, which lives exclusively in freshwater, has a very tall and bulky brood poach (Fig. 2).

The fecundity of the parthenogenetic females is much higher than that of gamogenetic females. However, the gamogenesis plays an extremely important role in the survival of the species by resting eggs, able to remain viable in the most difficult environmental conditions. Transported by birds, they contribute to the spread of the species into new ecosystems (Rivier, 1998).

The absence of effective organs of movement, the small size of the individuals and the large volume of the brood poach, drastically restrict the speed of movement for the species belonging to *Podonevadne* genus. The speed of *P. trigona trigona* is  $1\text{ m h}^{-1}$  (Egloff *et al.*, 1996) and, probably *P. trigona ovum* has

a similar value. However, the researches performed in 2005 showed that it is present in almost the entire basin of the Danube-Black Sea Canal.

Reported at that speed, the colonization of 64 km of the canal by *P. trigona ovum* would have lasted 87 months. Due to the transportation of the resting eggs by birds, the colonization process of the Canal lasted, probably, a much shorter period. Also, presumably, the crossing distance between the origin basin of immigrant and the mouth of the Danube-Black Sea Canal was made in a similar manner.

The *Podonevadne* genus is a nocturnal predator filtered type. They feed mainly on ciliates, rotifers, nauplii, small cladocerans and, in addition, nanoplanktonic algae (Egloff *et al.*, 1996).

The percentage of the abundance of the mentioned cladoceran in the structure of total zooplankton stands at low level (Table 6). However, the emphasized values are comparable with those of other predatory species found in the zooplankton structure of different aquatic ecosystems (Zinevici & Parpală, 2007).

Instead, the analyzed species showed a much higher percentage as part of the predator zooplankton. It represented 30.58% of the abundance of the predator zooplankton, in August and 26.82%, in September (Table 7).

Very high values have emerged in the middle of the canal, located between 4-9 stations. The analysis of these data reveals that *P. trigona ovum* became a numerically dominant species in the structure of the predator zooplankton.

These data highlight the strength of *P. trigona ovum* reflected in zooplankton community trophic relationships in two decades of existence in the Danube-Black Sea Canal ecosystem.

## CONCLUSIONS

The cladoceran *Podonevadne trigona ovum* is a Caspian immigrant subspecies and entered the terminal area of the Danube-Black Sea Canal in the first year of the ecosystem (1985).

Adapting to the new ecological conditions, over the next 20 years it has spread to almost all parts of the ecosystem, except for the river confluence area.

The annual dynamics of the abundance presented the highest values during summer and slightly lower in early autumn.

The analysis of the frequency and percentage of the immigrant in the structure of the predator zooplankton emphasized an integrating process in the structure and trophic relationship of the new ecosystem.

In the next stage, there can be predicted that *Podonevadne trigona ovum* will populate a series of lakes, with lagoon origin, from the Romanian Black Sea sector.

Invasive behavior of some species belonging to the Onychopoda infraorder resulted in significant changes in the structure of some ecosystems, suggests the usefulness of monitoring the evolution of *Podonevadne trigona ovum* in the Danube-Black Sea Canal.

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