NATURALIZATION OF *MELANOIDES TUBERCULATA* AND *TAREBIA GRANIFERA* (THIARIDAE, GASTROPODA) MOLLUSKS UNDER THE HYDROECOLOGICAL CONDITIONS OF ZAPORIZHZHYA NPP COOLING POND

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Fedonenko O., Marenkov O., Ananieva T. 2018. Naturalization of *Melanoides tuberculata* and *Tarebia granifera* (Thiaridae, Gastropoda) mollusks under the hydroecological conditions of Zaporizhzhya NPP cooling pond. *Acta Biol. Univ. Daugavp.*, 18 (1): 39 – 46.

The results of studies on the populations of *Melanoides tuberculata* and *Tarebia granifera* (Thiaridae, Gastropoda) in the cooling pond of the Zaporizhzhya NPP are presented. Primarily, mollusks have occurred in the cooling pond because of the casual release from the aquarium. Since 2015, the number of mollusks has rapidly increased, which in 2017 caused problems in the work of hydraulic structures of nuclear power plants. The number of mollusks per 1 m² of concrete walls of the outlet channel ranged from 420.5 to 619.8 exemplars. In clusters of gastropods, the ratio of representatives of *M. tuberculata* and *T. granifera* species was 1:12. The mollusks *M. tuberculata* and *T. granifera* are completely naturalized under conditions of the cooling pond of the Zaporizhzhya NPP and are actively increasing their numbers. It was found that the average chemical quality index value characterized the water of the Zaporizhzhya NPP cooling pond as 'good' and 'clean'. The main indicators of hydrochemical regime and salt composition of the water have completely satisfied to the requirements of the fishery standards and point to the developed favorable environment for the life of mollusks. To control their invaders, it is necessary effective biomelioration measures using hydrobionts, which are potential predators for the invasive mollusks.

Key words: alien species, the cooling pond, melania, Gastropoda, NPP, invasive species.

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INTRODUCTION

Recently created artificial ecosystems, such as the techno-ecosystems of cooling ponds of nuclear power plants, are most often amenable to biological invasion processes (Jeschke et al. 2014). A classic example of the spontaneous introduction of hydrobionts into the cooling ponds of nuclear power plants is the mass development of mollusks of the genus *Dreissena* (Sylayeva et al. 2012).

Cooling ponds are not only technical reservoirs, but also elements of ecological water complexes, the state of which should be no worse than the ecological state of the surrounding water bodies (Badr et al. 2012, Khilchevsky & Hrebin 2014, Novoselova & Protasov 2015). Since these water bodies are affected to thermal pollution, they are characterized by transformed hydrological, hydrochemical and hydrobiological regimes (Protasov et al., 2017). The constant elevated temperature of the cooling ponds water contributes to the development of warm-water flora and fauna, whose representatives can cause eutrophication, and also serve as biological disturbances in the water supply system (Protasov et al. 2015, Sri-aroon et al. 2015).

The influx of a significant amount of heated water in the cooling reservoirs can promote a shift in the carbonate-calcium equilibrium, which is their specific feature. As a rule, water masses of technogenic reservoirs, primarily coolers, are characterized by water supersaturation with calcium carbonate, which, in turn, is a prerequisite and reason for the formation of carbonate deposits on heat exchange surfaces. Enrichment of water with Ca^{2+} ions is beneficial for the development of mollusks, which actively use free calcium to form and grow their shells (Protasov et al. 2011).

In the last 20 years, the localized introduction of more than 10 species of freshwater and brackish water aquarium mollusks into natural and artificial water bodies has taken place in Ukraine (*Helisoma duryi* (Wetherby, 1879), *Physella spp., Ampullariidae spp., Melanoides tuberculata* (Müller, 1774), *Ferrissia fragilis, Tarebia granifera* (Lamarck, 1822), etc.). The spread of new species often occurs due to the draining of water from aquariums into open water bodies. Some thermophilic mollusks, in general, can take root in artificial reservoirs where the warmed waters are discharged (cooling ponds and channels of thermal power plants, nuclear power plants) (Alexandrov et al. 2007, Son 2007).

The aim of paper was to study of naturalization process of highly invasive Thiaridae family mollusks under the environmental conditions of the cooling-pond of Zaporizhzhya nuclear power plant in Ukraine.

MATERIALS AND METHODS

The research was conducted in August– September 2017 in water area of the cooling pond of the Zaporizhzhya NPP (47°50'42"N, 35°56'89"E), Ukraine. The cooling water body of the Zaporizhzhya NPP belongs to the lakepond type. A type of regulating the level regime is seasonal. The cooling pond was constructed by cutting off a part of the Kakhovka reservoir by an overburden sand dam. It has the following parameters: the water mirror area is 8.2 km², the volume is 47.05 million m³, the average depth is 5.87 m, the maximum depth is 13.5 m, and the coast line length is 11.2 km.

The average monthly temperature of chilled water in the pond in the hottest month is 28.7°C. During the winter months, the water temperature is kept at the level of 17-18°C. There is no ice cover on the cooling pond, thin ice can be found only in the coastal zone.

Selection and processing of hydrobiological samples were carried out according to conventional hydrobiological methods. Sampling of benthos was performed by the Ekman-Burge bottom grab at the stations of the heat sink with different types of soil (sand, muddy sand, black mud). Collection of mollusks at hydraulic structures was carried out using a hydrobiological scraper. The counting number of mollusks on concrete slabs in the nuclear power plant channels was done using 1 m² frame that was installed at the water's edge and was fixed on the slope of the slabs. Within the bounded confines of the space, the mollusks were manually selected, the material obtained was counted in situ, and some samples were fixed with 4 % formalin to refine the species composition.

Water samples for the hydrochemical analysis were taken monthly in the surface layer, the studies were carried out according to standard methods (Romanenko 2006, Sharamok et al. 2017). The metal contents in water were determined by atomic-adsorption spectrophotometry. The study uses average values of the main hydrochemical parameters from 2017. Statistical data processing was carried out by conventional methods using software packages for personal computers Microsoft Excel 2010. Parameters of water chemical composition were compared with normative criteria of water quality for fishery purposes (SOU 05.01-37-385:20, 2006, GSTU 2284:2010, 2013).

Environmental assessment of water quality in various categories was carried out in accordance with the 'Environmental Assessment Method of surface water for different categories' (Romanenko et al. 1998, Romanenko et al. 2001) and 'Common Implementation Strategy for the Water Framework Directive (2000/60/ EU), Guidance document № 10' (2000).

RESULTS AND DISCUSSION

Hydroecology

The hydrochemical regime of technogenic water bodies is similar in main features to the regime of natural reservoirs of a certain climatic zone (Table 1).

At the same time, the thermal regime characteristic of the heat sink contributes to the emergence of conditions under which the evaporation of water is observed to increase in comparison with natural water bodies, which in turn leads to an increase in the mineralization of water (Protasov et al. 2011). The water of the heat sink of Zaporizhzhya NPP is classified as fresh, hypogaline (Class I, Category 1) by the criterion of mineralization. The total content of the main ions in water, the concentration of chloride and sulfate ions also correspond to the 1 category of quality.

The regime and dynamics of sulfate ions SO_4^{2-} are directly dependent on the concentration of Ca^{2+} ions, since the formation of sparingly soluble $CaSO_4$ is possible in their presence. On the other hand, the content of Ca^{2+} ions is limited by its consumption by hydrobionts, especially during the formation and growth of shells of mollusks. The smoothed seasonal dynamics of

the concentrations of sulfate ions and calcium in the heat sink of the Zaporizhzhya NPP indicates their saturating equilibrium amounts.

In general, the salt composition of the water and the main indicators of the hydrochemical regime fully satisfy the requirements of the fishery standards and point to the developed favorable environment for the life of hydrobionts. Among the pollutants, the maximum permissible concentration was found for some heavy metals, such as manganese (2 times), zinc (4 times), copper (20 times).

For the integrated environmental assessment, averaged chemical water quality index $(I_{Xavg.})$ was calculated (Grytsenko et al. 2012, Sharamok et al. 2017):

$$I_{Xavg.} = (I_{Cavg.} + I_{TCavg.} + I_{Tav.}) / 3,$$

where the index of indicators of salt composition $I_{Cavg.} = 1$; the index of chemical trophosaprobiological (ecological and sanitary) indicators $I_{TCavg.} = 3.4$; index of specific indicators of toxic and radiation exposure $I_{Tavg.} = 2.9$. Thus, the value of the average chemical quality index $I_{Xavg.} = 2.4$, which characterizes the water of the heat sink at Zaporizhzhya NPP as 'good' and 'clean'.

Invasion of the mollusks

On the territory of Ukraine mollusk *M*. *tuberculata* was found in the cooling pond of the South Ukrainian NPP, where it settled down in the period from 2006 to 2011 and occurs at a depth of 6 m at a water temperature of $25-27^{\circ}$ C, completely dominating in benthic samples (Sylayeva & Stepanova 2016).

In 2013, two new species of freshwater mollusks of the Thiaridae family were recorded in the cooling pond of the Zaporizhzhya nuclear power plant: *Melanoides tuberculata* (Müller, 1774) and *Tarebia granifera* (Lamarck, 1822). Since 2015, the number of mollusks has increased rapidly, which in 2017 caused problems in the work of hydraulic structures of nuclear power plants. Fedonenko O., Marenkov O., Ananieva T.

Indicators	Value	Indicators	Value
Hydrophysical and general hydrochemical indicators		Contents of certain main ions	
Temperature, °C	25.3	Sodium, mg/dm ³	25.0
pH	7.9	Potassium, mg/dm ³	5.1
Transparence, cm	>31	Calcium, mg/dm ³	56.1
Suspended particles, mg/dm ³	<5.0	Magnesium, mg/dm ³	18.3
Dissolved oxygen, mg/dm ³	7.08	Chlorides, mg/dm ³	33.9
Mineralization by dry residue, mg/dm ³	403.0	Sulphates, mg/dm ³	60.0
Alkalinity total, mg-equiv/dm ³	3.8	Water hardness in total, mg-equiv/dm ³	4.2
		Hardness of carbon, mg-equiv/dm ³	3.3
Contents of biogenic elements		Organic matter content	
Ammonia nitrogen, mg/dm ³	0.30	Permanganate oxidability, mgO/ dm ³	4.32
\mathbf{N}	0.00	-	
Nitrite nitrogen, mg/dm ³	0.03	$BOD_{s}, mgO_{s}/dm^{3}$	2.68
Nitrite nitrogen, mg/dm ³ Nitrate nitrogen, mg/dm ³	2.37	BOD ₅ , mgO ₂ /dm ³ COD, mgO ₂ /dm ³	2.68 24.7
Nitrate nitrogen, mg/dm ³	2.37 0.32		
Nitrate nitrogen, mg/dm ³ Phosphorus of phosphate, mg/dm ³ The contents of specific toxic substances Ferum total, mg/dm ³	2.37 0.32	COD, mgO ₂ /dm ³	
Nitrate nitrogen, mg/dm ³ Phosphorus of phosphate, mg/dm ³ The contents of specific toxic substances	2.37 0.32 0.32 018 0.021	COD, mgO ₂ /dm ³ Nickel, mg/dm ³ Cadmium, mg/dm ³	24.7
Nitrate nitrogen, mg/dm ³ Phosphorus of phosphate, mg/dm ³ The contents of specific toxic substances Ferum total, mg/dm ³ Cuprum, mg/dm ³ Manganese, mg/dm ³	2.37 0.32 018 0.021 0.019	COD, mgO ₂ /dm ³ Nickel, mg/dm ³ Cadmium, mg/dm ³ Lead, mg/dm ³	24.7 0.0094 0.0002 0.002
Nitrate nitrogen, mg/dm ³ Phosphorus of phosphate, mg/dm ³ The contents of specific toxic substances Ferum total, mg/dm ³ Cuprum, mg/dm ³	2.37 0.32 0.32 018 0.021	COD, mgO ₂ /dm ³ Nickel, mg/dm ³ Cadmium, mg/dm ³	24.7 0.0094 0.0002

Table 1. Chemical composition of the water of the Zaporizhzhya NPP cooling pond (the average neans of data from 2017, S.E.M values do not exceed 5 %, n=9)

The spiral shell of these invasive mollusks poses a threat to the operation of thermal mechanical equipment, since the water purification system is not fully capable for removing small shells from the water flow. Also live mollusks accumulate on concrete structures and grids of mechanical water purification. The flow of shells of dead mollusks accumulates in dead-end sections, bends of pipes, filter elements, enter the concrete pools of the incubation department of the biological reclamation department of the reservoir, which negatively affects the operation of the equipment and can provoke a decrease in the flow ability of hydraulic structures (Fig. 1).

A direct calculation of the number of mollusks on an area of 1 m² on the concrete walls of the outflow channel showed that the number of mollusks *M. tuberculata* and *T. granifera* ranged from 420.5 to 619.8 exemplars (n=30) (Fig. 2). The bulk of mollusks were found near the surface of the water at a depth of up to 15 cm.

During the route investigations, the sections of the cooling pond were inspected with subsequent selection of hydrobiological samples. Inspection of pumping stations showed that the daily accumulation of mollusks in the chambers was 230-300 g. Sampling bottom sediments at different depths and at different types of bottom (sand, silted sand, black mud) of the heat sink and channels showed the absence of mollusks shells of the Thiaridae family. The maximum concentration of mollusks was observed in the coastal zone on the rocks, at the surface of concrete slabs, in crevices between concrete slabs of canals, in the shade of thickets of higher aquatic vegetation, in the roots of reeds of ordinary Phragmites australia and near clusters of algae of the genus Cladophora.



Fig. 1. *Melanoides tuberculata* and *Tarebia granifera* (Thiaridae, Gastropoda) in and its localities in Ukraine: 1 – the cooling pond of the Zaporizhzhya NPP ($47^{\circ}50'42''$ N; $35^{\circ}56'89'$ E); 2 – cluster of mollusks on concrete walls of the channel of the nuclear power plant; 3 – accumulation of *M. tuberculata* on a stony substrate in the region of the discharge channel of the nuclear power plant, September 15, 2017; 4 – accumulation of *M. tuberculata* and *T. granifera* on reed roots.

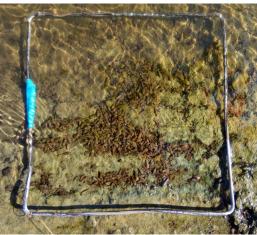


Fig. 2. Mollusks in the hydrobiological frame 25x25 cm.

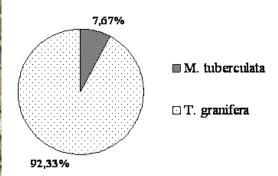


Fig. 3. Percentage ratio of Thiaridae mollusks on concrete walls of the outflow channels of Zaporizhzhya NPP.

Analysis of the species composition of samples collected from the concrete slabs of the outflow channel showed that in the clusters of gastropods the ratio of representatives of *M. tuberculata* and *T. granifera* species was 1:12 (Fig. 3).

The collected mollusks were divided into three size groups: I – less than 10 mm, II – from 10 to 15 mm, III – more than 15 mm. Analysis of the size series of mollusks of two species showed the following ratio: for mollusks *M. tuberculata* 1:2:4 (Σ n=142 specimens), for mollusks *T. granifera* 1:2:7 (Σ n=1710 specimens), respectively.

The developing eggs were found on concrete structures and shells of mollusks. Thus, it can be concluded that the mollusks *M. tuberculata* and *T. granifera* are completely naturalized under the conditions of the cooling pond of the Zaporizhzhya NPP and are actively increasing their numbers.

In order to minimize the negative impact of mollusks on the hydrostructures of nuclear power plants, it is necessary to develop a complex of biomeliorative measures to reduce the number of gastropod mollusks by introducing hydrobionts into the ecosystem of the heat sinks, which can control the number of mollusks by natural effect on its population. Among the promising biological methods for combating gastropod mollusks, the following trends can be singled out: complex stocking of the heat sinks by thermophilic fish eating mollusk, for example, the black carp Mylopharyngodon piceus (Richardson, 1846). It is also considered the possibility of introducing a predator mollusk Clea helena (von dem Busch, 1847).

CONCLUSIONS

Since 2013, the appearance of mollusks of the family Thiaridae has been noted in the cooling pond of the Zaporizhzhya nuclear power plant: *Melanoides tuberculata* (Müller, 1774) and *Tarebia granifera* (Lamarck, 1822), which were naturalized, and by 2017 they increased their numbers and spread throughout the reservoir. The

spiral-shaped shell of mollusks creates a threat to the operation of thermal mechanical equipment.

On the concrete walls of the outflow channel, the number of mollusks *M. tuberculata* and *T. granifera* ranged from 420.5 to 619.8 exemplars. The bulk of the mollusks were found near the surface of the water at a depth of up to 15 cm.

Inspection of pumping stations showed that the daily accumulation of mollusks in the receiving chambers was 230–300 g. Sampling bottom sediments at different depths and at different types of bottom (sand, silted sand, black mud) of the heat sink and channels showed the absence of mollusks shells of the family Thiaridae. The maximum concentration of mollusks was observed in the coastal zone on the rocks, at the surface of concrete slabs, in crevices between concrete slabs of canals, in the shade of thickets of higher aquatic vegetation, in the roots of reeds of ordinary *Phragmites australia* and near clusters of algae of the genus Cladophora.

In the groupings of gastropods collected from concrete slabs of the outlet channel of the Zaporizhzhya NPP, the quantitative ratio of *M. tuberculata* and *T. granifera* was 1:12.

Analysis of the dimensional rows of mollusks of two species showed the following ratio of size groups (I – less than 10 mm, II – from 10 to 15 mm, III – more than 15 mm): *M. tuberculata* 1:2:4 (Σ n=142 specimens), *T. granifera* 1:2:7 (Σ n=1710 specimens), respectively.

To control the mollusks invaders, it is necessary to develop a system of effective biomelioration using hydrobionts, which are potential predators of these mollusks.

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Received: 22.08.2018. Accepted: 01.09.2018.