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Distribution and historical biogeography of *Artemia* leach, 1819 (Crustacea: Anostraca) in Ukraine

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Abstract

The study of brine shrimp in the Ukraine, formerly part of the Russian Empire and then the USSR, has a fairly long history. In the territory of Ukraine we can identify the following areas of potential *Artemia* habitats: the Crimean peninsula; the coastal zone of the NW Black Sea; the north coast of the Sea of Azov; Donetsk Oblast; Zakarpattia Oblast. Now it can be recognized that at least two bisexual species (*A. salina* and *A. urmiana*) and the parthenogenetic populations of *Artemia* dwell in the Ukraine. In some lakes (Koyashskoye, Terecly-Konradskoye) we found *A. urmiana* and parthenogens both, but separated in time. In the waters of the Crimea and NW part of the Black Sea brine shrimp are found under salinities from 10-20 (if no other animals present) to 350 - 370 ppt. Larvae and adults of some Coleoptera, Hemiptera, ostracods *Eucypris inflata* (Sars, 1903) and probably other invertebrates may eat away *Artemia* completely in the Crimean lakes whose salinity is below 60-110 ppt. Hydromeliorative, mining, construction and other human activities can both create new habitat for brine shrimp, as well as destroy the existing natural ones. Despite the fact that the largest habitats of *Artemia* in the past (Siwash, Donuzlav, Kyzyl-Yar) ceased to exist, in the Crimea and the NW Black Sea (Kuyalnik) still have water with very promising opportunities for industrial harvesting of *Artemia* cysts. Authors hypothesized that the study of populations of brine shrimp in the Crimea is the key to understanding the phylogeny and biogeography of *Artemia.*

Key Words: *Artemia*, biogeography, ecology, Crimea, Ukraine

Introduction

The study of brine shrimp in the Ukraine, formerly part of the Russian Empire and then the USSR, has a fairly long history. For the first time knowledge of *Artemia* presence in the water bodies of modern Ukraine was realized as a result of the travels of P. Pallas during 1793-1794 around the Tauride province (Pallas, 1999). In the early 19th century, a number of scientists studied the brine shrimp in the waters of the NW part of Black Sea and Crimea (Fedchenko, 1870). In particular, Gasgagen in 1849 found the brine shrimp in the lagoons near Odessa, and Von Kreutzer and Dubitsky in the same year studied the brine shrimp in the lake Chokrak (Crimea) and concluded that brine shrimp present before the salt sedimentation in the lake occur. In the 1870s Odessa zoologist V. Schmankewitsch studied fauna in the hypersaline lagoons of NW part of the Black Sea. His classic experiments have shown that when brine shrimp were grown at different salinities they got dramatically different morphological forms (Schmankewitsch, 1876). Development of *Artemia* was studied by the famous zoologist N.V. Nasonov, who gave the first description of its ontogenesis (Nasonov, 1887). Then N. Gaevskaya conducted study on the effect of salinity on the variability of *Artemia* from Lake Chersonesus near Sevastopol (Gaevskaya, 1916). In the following years many researchers have paid attention to the study of various aspects of the *Artemia* biology on the territory of modern Ukraine (N. Khmeleva, L. Suschenya, P. Voronov, L. Radchenko, M. Ivanova, I. Rudneva, etc.). It should be noted that experimental *Artemia* study prevailed over its actual study in nature. Extensive investigations of *Artemia* in the lakes of the Crimea were conducted with the participation of the authors at the Institute of Biology of Southern Seas only since 2000.

Despite a long history of studies of shrimp in Ukraine the knowledge of its distribution in Ukraine are not sufficient. Many water bodies, in which we can assume the presence of *Artemia*, are still unstudied. This proposed work tries to somehow fill the gap. Results (published and not published) of our long-term studies (2000-2011), using other publications, and the Internet, personal communications of various people has made possible this summary compilation as set out in Table 1 below.

Distribution

In the territory of Ukraine we can identify the following areas of potential *Artemia* habitats (Fig. 1):

The Crimean peninsula
The coastal zone of the NW Black Sea
The north coast of the Sea of Azov
Donetsk Oblast
Zakarpattia Oblast

The Crimea: This area is situated in the northern Black Sea. The Crimea is the largest (nearly 26.5 thousands km^2) peninsula on the Black Sea. The Black Sea washes its shores in the south, west and northwest, the Sea of Azov in the northeast, and the Kerch Strait, which connects the two seas, in the east. In the north the narrow Perekop isthmus links the Crimean peninsula with the East European plain. There are 50 comparatively large and many small (from several to hundreds meters long) lakes in the Crimea. Academician P. Pallas had given the first description of saline lakes he found during his research trip round the Crimea in 1793-1794 (Pallas, 1999). In the mid-19th century G. Fedchenko (1870) made the first inspection of hypersaline lakes and had counted their number as 116. The exact count of small lakes is unknown for it is difficult to differentiate between a large puddle and a tiny lake. In the Crimea most of hypersaline lakes have marine origin and are seawater localities that have once separated from the main sea. Sasyk-Sivash that has the surface of 75.3 km^2 is the largest (Kurnakov *et al.*, 1936). At present all these lakes receive the infiltrated and storm inflows from the sea. For the majority of such lakes/enclosed lagoons the infiltration from the sea provides the basic supply in their water balance. The age (probably not greater than 3-5 thousands years), size and other characteristics of marine lakes vary broadly. This makes the lakes remarkably diverse in physico-chemical and biotic characteristics. For instance, the annual average salinity varies from 30 to 250‰; interannual differences are also considerable.

Tab. 1: *Artemia* sites in Ukraine

Region	Locality	Geographical coordinates	Reproductive mode	Species	Ref.
Krasnoperekopsk raion	Aygulskoe	45°59'N-34°35'E	P	Parthenogenetic population	17
	Kirleutskoe	45°56'N-34°1'E	?	?	17
	Kiyatskoe	45°58'N-33°56'E	P	Parthenogenetic population	17
Rozdolnye raion	Bakalskoye	45°45'N-33°10'E	B, P	Parthenogenetic Population, ?	?
Chornomorsky raion	Dzharylhatch	45°34'N-32°52'E	P	Parthenogenetic population	9, 17
	Liman	45°23'N-32°31'E	?	?	7
	Yarylhatch	45°33'N-32°51'E	P	Parthenogenetic population	7
Saky raion	Terekly-Konradskoye	45°11'N-33°13'E	B, P	*A. urmiana,* Parthenogenetic population	12, 10
	Oybursky	45°18'N-33°05'E	P	Parthenogenetic population	9, 17
	Shtormovoye	45°20'N-53°05'E	B	*A. salina*	9, 17
	Otar-Moinakskoye	45°12'N-33°30'E	P	Parthenogenetic population	9
	Sakskoye	45°10'N-33°30'E	P	Parthenogenetic population	5, 9
	Sasik-Sivash	44°50'N-33°25'E	B, P	*A. salina,* Parthenogenetic population	9
Kirovsk raion	Achi	45°9'N-35°25'E	P	Parthenogenetic population	17
	Arabatskaya strelka	45°42'N-35°09'E	P	Parthenogenetic population	9
	Sivash	46°00'N-34°50'E	P	Parthenogenetic population	9
Area of Feodosia	Adzhigol	45°05'N-35°25'E	P	Parthenogenetic population	17, 9
	Kuchuk-Adjigol	45°06'N-35°27'E	P	Parthenogenetic population	17
Lenin raion	Uzunlarskoye	45°5'N-36°7'E	P	Parthenogenetic population	17
	Koyashskoye	45°4'N-36°13'E	B, P	*A. urmiana*	1, 12, 10, 7
	Kirkoyashskoye	45°4'N-36°13'E	P	Parthenogenetic population	17, 7
	Tobechik	45°10'N-36°21'E	P	Parthenogenetic population	9, 17, 7
	Pond near West corner of Tobechik	45°18'N-36°30'E	B	*A. urmiana*	17
	Aktashskoye	45°22'N-35°49'E	P	Parthenogenetic population	17
	Gol	45°07'N-36°24'E	?	?	17
	Marfovskoye	45°12'N-36°6'E	P	Parthenogenetic population	17, 2, 7
	Shimakhanskoye	45°10'N-36°25'E	P	Parthenogenetic population	17, 7
	Chokrak	45°27'N-36°18'E	P	Parthenogenetic population	9, 17

Tab. 1: Continued

Region	Locality	Geographical coordinates	Reproductive mode	Species	Ref.
Area of Sevastopol	Chersonessus Lake	44°59'N-33°39'E	P, B	*A. salina,* Parthenogenetic population	4, 3, 17, 7
Kherson region	Odzhigol	46°25'N-32°13'E	?	?	9
	Small lakes of Kinburgski peninsula	46°25'N-31°40'E	B, P	*A. salina,* Parthenogenetic population	9, 15, 14
	lakes near Henhorka	46°15'N-34°40'E	P	Parthenogenetic population	9
	salt ponds at W. Syvash	46°15'N-34°40'E	P	Parthenogenetic population	9
	Dolgoye	46°30'N-35°35'E	P	Parthenogenetic population	9
	Sredneye	46°30'N-35°30'E	P	Parthenogenetic population	9
	Tchongar	46°00'N-34°35'E	P	Parthenogenetic population	9
Odessa region	Kuialnik liman	46°43'N-30°35'E	P	Parthenogenetic population	6, 9
Zaporizhzhya region	Solenoye	46°44'N-36°22'E	?	?	13
	Molochnui liman	46°32'N-35°19'E	?	?	18
Tiachiv Raion	lakes near Solotvyno	47°57'N-23°51'E	?	?	18

P: parthenogenetic; B: bisexual

References: 1. Abatzopoulos *et al*., 2009; 2. Balushkina *et al*., 2009; 3. Fedchenko, 1870; 4. Gaewvskaya, 1916; 5. Ivanova, 1990; 6. Kulagin, 1888; 7. Litvinchuk *et al*., 2006; 8. Makarov, 1984; 9. Mura & Nagorskaya, 2005; 10. Shadrin & Batogova, 2009; 11. Shadrin *et al*., 2001; 12. Shadrin *et al*., 2008; 13. Solonenko *et al*., 2010; 14. Vekhov, 1993; 15. Vekhov & Vekhova, 1992; 16. Zagorodnya *et al*., 2008; 17. Our date; 18. Personal communications of different persons

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All the lakes have a distinct annual salinity cycle; the variability range is different for each lake. With regard to chemical composition of the water the Crimean marine lakes divide into two types (Kurnakov *et al.,* 1936). One type includes lakes the brine of which contains calcium, magnesium or sodium sulphates; the major inflow into those lakes is from the sea. The other type includes lakes with the brine having the concentration of sulphuric acid ions sufficient (or less) to generate sulphuric calcium salt; those lakes receive water supply from terrestrial and underground sources. On the Crimean peninsula marine hypersaline lakes concentrate along the western, northern, eastern and southeastern coasts.

Notably less numerous is the group of continental hypersaline lakes, known in the local dialect as *koli*, that concentrate on the Kerch peninsula (Eastern Crimea), and locate in the calderas of the Prehistoric mud volcanoes (Shadrin, 2009). They are sulfate lakes (Shadrin, 2009). *Atremia* inhabits in both types of lakes.

Assuming that total annual land/air heat exchange performed through heat-conductivity is negligible and can be ignored, it can be deduced that the annual locality-specific evaporation would consume the amount of heat equal to the annual irradiation balance. Thereby the irradiation index of dryness for a year is determined as (Khromov & Petrosyan, 2001): $K_\delta = B/LP$

where B is annual irradiation balance (unit of energy), P – total annual precipitation (mm), and L – evaporation heat (unit of energy/mm). Irradiation index of dryness indicates the portion of irradiation balance used for rainfall evaporation. It was assumed that under the values greater than the conventional limit of climate dryness, $K_\delta = 3$, the climate in the locality is dry that favours salt accumulation and hypersaline lake formation (Efimov & Timofeyev, 1990). Thereby radiation dryness coefficients of the Crimean hypersaline lake distribution areas fluctuate around K_δ=*4-5*. According to (Efimov & Timofeev 1990), this estimate is greater than conventional dryness limit K_δ =*3,* i.e. the climate in the lake locality is actually dry. A more detailed description of some lakes is given in earlier published papers (Shadrin 2008, 2009).

The coastal zone of North-Western Black Sea (territories of Ukraine, Bulgaria, Romania): The area is located in the arid climate zone. A lot of different lagoons and lakes of marine origin are here. There are some hypersaline water bodies among them. *Artemia* were found in several hypersaline water bodies, located in Ukraine, as well as Romania and Bulgaria. Currently, several large closed lagoons, previously inhabited by *Artemia* (Kulagin, 1888), were connected to the sea, resulting its salinity became low.

Fig. 1: Areas with *Artemia* sites in Ukraine (more detailed in the Crimea)

The north coast of the Sea of Azov: This area has a dry climate: there are many different lakes and lagoons; some of them inhabited by *Artemia*. The fauna of many small lakes/ lagoons has not been investigated yet.

Donetsk Oblast: There are the salt lakes of karst origin (48 ° 52'N - 37 ° 37'E) within the territory of Slavyansk, Donetsk region. They are separated from one another by not high sand ramparts with varying widths of 150-200 m. The largest lakes are: Slepnoye (area of 0.29 km^2, depth to 2.2 m), Rapnoye (area of 0.22 km^2, depth to 7.5-8 m), Veysovo (area of 0.16 km^2, the depth to 19 m). There are a larger number of other lakes of smaller size and depth. Chemical composition of the brine consists of chloride-sodium, also sulfates. We could not find any information of *Artemia* presence in these lakes. We haven't found any paper on the fauna study in these lakes since the late 19th century.

Zakarpattia Oblast: Near the village of Solotvyno (Tyachiv district of Zakarpattia Oblast), near the Ukrainian-Romanian border, there are salt mines. On the edge of this village at the present time there are salt lakes of anthropogenic origin: they appeared in the XX century as a result of subsidence of the breed above the mines. The first, salt mine created the largest of the lakes - Kunigunda (formed in 1902, the area - 800-900 m2, depth 1.8 m, salinity - 140-150 ppt, temperature year-round above 17 °C), when the mine of same name suddenly sank immediately by 20 meters. *Artemia* was introduced in the lakes naturally (most likely by birds).

Factors determining the presence of *Artemia* in water bodies

In the waters of the Crimea and Kinburskaya Spit (NW part of the Black Sea), brine shrimp are sometimes found under very low salinities - 10-20 ppt (Vekhov, 1993; our data) if no other animals present. We found active adult *Artemia* under salinity 350 - 370 ppt. Larvae and adults of some Coleoptera (*Hygrotus enneagrammus* (Ahrens, 1833) and others), Hemiptera (Coricidae), ostracods *Eucypris inflata* (Sars, 1903) and probably other invertebrates may eat away *Artemia* completely in the Crimean lakes whose salinity is below 60-110 ppt. At the same time, the massive development of *Artemia* could occur simultaneously with the Chironomidae larvae (*Baeotendipes tauricus* Tshernovskij), Harpacticoida (mostly *Cletocamptus retrogressus* Shmankevich, 1875) and *Moina salina* Daday, 1888. Under highest salinities a limiting factor may be not high salinity itself, but the associated with it lack of oxygen. It is known that the solubility of oxygen in the water with increasing salinity is significantly reduced. In the Crimea *Artemia* may reach high density in thalassohaline as well as in athalassohaline (sulphate) lakes.

Impact of human activities on the distribution of *Artemia*

Hydromeliorative, mining, construction and other human activities can both create new habitat for brine shrimp, as well as destroy the existing natural ones. An example of the creation of new habitats may be of Solotvinskiy salt lakes that formed as a result of subsidence over the salt mines. But more often we meet with examples of the destruction of natural habitats of brine shrimp, of which there are numerous examples in the Crimea. Earlier in the vicinity of Sevastopol there were four hypersaline lakes (Khersoneskaya group of lakes), inhabited by *Artemia* (Fedchenko, 1870; Dagaeva, 1927), three of them had been covered and ceased to exist under the road construction in the second half of the 20th century. Severo-Krymskii Canal in the Crimea was built in 1961-1971 for irrigation in dry areas using the waters of the Dnipro River. It is one of the biggest canals in Europe. At the same time, the intensive cultivation of rice in northern Crimea was started, with discharge of fresh water from rice paddies in hypersaline Siwash, Gulf of the Azov Sea. Salinity in the Sivash resulted in decrease from 100-250 to 2-15 ppt. Before desalination Siwash was the largest habitat of *Artemia* in USSR (Voronov, 1973). At present, *Artemia* are found in only the small lagoons on the Arabatskaya strelka (the spit between Sivash and the Sea of Azov), as well as in small lagoons and large puddles near north and south parts of Sivash.

The Crimean largest closed hypersaline lagoon was Donuzlav in West Crimea (45°20' N - 33°00 E), and *Artemia* inhabited in it (Kulagin, 1888). In 1961 a canal that connected it to the sea was dug. Donuzlav desalinization began and now salinity in it varies around 15-17 ppt. Now there are small hypersaline lagoons and puddles only along its northern coast. In 1978 channel- fairway (20 meter width) in the lake Panskoe (45°32' N - 32°48'E) from Yarylgachskiy Bay was dug. On the north shore of the lake was built port Chernomorsk during 1979-1991. The Panskoe Lake was hypersaline, now its salinity is 17-19 ppt, and, of course, brine shrimp in it has disappeared. A large habitat of *Artemia* was hypersaline lake Kyzyl-Yar (45°03'N-33°55'E). Reservoir of fresh water from the North Crimean (Severo-Krymskii) canal has been built upper 20 kilometers in the gully, which flows into the lake. Fresh water seeped underground along the gully in the Kyzyl-Yar, and 20-25 years later the lake became completely freshwater. The same fate befell another hypersaline lake in the past – Bagaily (45°02'N- 33°38'E), just south of Kyzyl-Yar, the other girder flows into it from the same reservoir. Its salinity dropped from hypersaline to 10 ppt. As a result of human activities salinity in the lake Churbash (the area of Kerch) also decreased. It should be noted that, in consequence the arid climate of the Crimea is cause for a gradual salinization of a number of artificial small water reservoirs on the Kerch Peninsula. It is possible that over time they will become new *Artemia* habitats.

Species composition and some remarks on *Artemia* phylogeny

Now it can be recognized that at least two bisexual species (*A. salina* and *A. urmiana*) and the parthenogenetic populations of *Artemia* dwell in the Ukraine. In some lakes (Koyashskoye, Terecly-Konradskoye) we found *A. urmiana* and parthenogens both, but separated in time (Abatzopoulos *et al*., 2009`). The *A. salina* and parthenogenetic populations live in the Crimea as well as in the lakes and lagoons of NW part of the Black Sea, whereas *A. urmiana* have been found only in three lakes of Crimea. It should be noted that *A. urmiana* was found in the Crimea only recently (Shadrin *et al*., 2008; Abatzopoulos *et al*., 2009). Before us biologists have not studied the lakes, which are home to *A. urmiana* in the Crimea, so we can only wonder: is *A. urmiana* aboriginal of Crimea, or more recently brought by the birds?

The origin of species *A. urmiana* is also an intriguing question. Consider the known facts which indicate that Lake Urmia can not be the place of origin of this species (Manaffar *et al*., 2011): *A. urmiana* are considered to have branched ≈11million years ago, whereas the time frame for the formation of Lake Urmia is estimated to be 800–400 ka. Questions arise: Where did this species originally appear? When was it brought into Lake Urmia? The results of paleogenetic analysis of *Artemia* DNA recovered from Urmia sediment cores show that the cysts represent only a parthenogenetic type of *Artemia* from around 5,000 years ago (Manaffar *et al*., 2011). Consequently, *A. urmiana* were geologically recently introduced into Lake Urmia by birds.

Could the Crimea be an area of origin of *A. urmiana*? Consider the arguments in favor of this assumption. Certainly, it could not appear in modern hypersaline lakes of Crimea, where it is currently detected. Since the age of these lakes are only about 1.5-3 thousand years (Kurnakov *et al.*, 1936; Abatzopoulos *et al*., 2009). However near the modern hypersaline lake Koyashskoye, as well as near several other lakes, which we studied, there are deposits of more ancient salt lakes (Shnukov *et al*., 2009). Some of them were inspected by the well-known expert in sediments of salt lakes Prof. M. Zheng during our joint visit. Near the Koyashskoe Lake, as well as near several other hypersaline lakes in Crimea, we found stromatolites and oncolites, which indicate the presence of hypersaline lagoons/lakes here and in the Miocene (Gerasimenko *et al*., 2008). The Miocene (23.03 to 5.332 Ma) was the time of *A. urmiana* origin. Can you imagine a temporary relay race, in which cysts are passed like as baton from one hypersaline lake in the Crimea to another lake, which is younger. This leads to the conclusion that, as well as *A. salina*, *A. urmiana* is an aboriginal of the Crimea.

The Crimean peninsula is probably one of the centers of *Artemia* biodiversity formation; origin of *A. urmiana* - its separation from the common ancestor of *A. salina* and *A. urmiana/A. franciscana* - occurred in hypersaline paleolakes/lagoons of Parathetys in the Crimea or surrounding territories. The Crimea is a crossroad of the Afro-Eurasian waterbird flyway system. Lake Urmia and the Crimean Peninsula are in the same migration corridor of many bird species. It is shown that the most likely carriers of viable *Artemia* cysts between the Crimea and Lake Urmia are the following bird species Shelduck (*Tadorna tadorna* (L., 1758)), Redshank (*Tringa totanus* (L., 1758)) and Pied Avocet (*Recurvirostra avosetta* L., 1758) (Khomenko & Shadrin, 2009). The same or other species of birds have carried out the transportation of cysts *A. urmiana* in Lake Urmia, and then - in lakes of the Altai, where they also inhabit (Boyko & Muge, 2009). Migration of birds is the main vector of the spread and maintenance of the gene exchange flows between distant *Artemia* populations, which distribute spotty (Green *et al*., 2005; Khomenko & Shadrin, 2009).

Geological processes- plate tectonics, the growth of mountain systems, as well as climate change, led to significant changes in the distribution of areas with water bodies suitable for *Artemia* inhabitance. Also the bird migration routes as well as wind composition changed and that led to changes of the ways of brine shrimp gene flows, placed some populations in conditions of isolation, and led to *Artemia* speciation. In general, the diversification of *Artemia*, based on a data set of data of historical geology (Firstbrook *et al*., 1979; Goncharova & Scherba 1997; Koronovskii *et al*., 2006), biogeography (Triantaphyllidis *et al*., 1998; Munoz *et al*., 2008), and molecular biology (Remigio *et al*., 2001; Baxevanis *et al*., 2006; Hou *et al*., 2006; Munoz *et al*., 2008) can be represented by the following general scheme.

Distant ancestors of modern *Parartemia* and *Artemia*, probably lived in the salt lagoons of the ocean Panthalassa, the Tethys Sea and in lakes of the supercontinent Pangaea, 300-400 Ma. The Pangaea split into the Gondwana and Eurasia at about 150-220 Ma, it gave rise to the tectonic plate drift, which caused the most significant events in the evolution of protoartemies. The common ancestor for *Parartemia* (Parartemiidae), *Artemia* (Artemiidae) and *A. persimilis* existed about 85 million years ago, which coincided with the isolation of Australia in the late Mesozoic – a result of the Indo-Australian Plate drift (Coleman *et al*., 1998). The bird migration and wind routes between Australia and areas of the *Artemia* ancestor distribution disappeared and separate evolution histories of *Artemia* and *Parartemia* started. The lineage leading to *A. persimilis* diverged from the common ancestor of all *Artemia* species at the same time (between 80 and 90 MY ago) caused by separation of South America from Gondwana (Africa) as a result of plate drift (Baxevanis *et al*., 2006). These facts as well as logics raise the question: Is not *A*. *persimilis* representative of a particular genus, not *Artemia*? Considering all available data on historical geology, cytogenetics, and molecular genetics, we tend to the view that it should be isolated from the genus *Artemia*, as a new genus, for example, *ArgArtemia*. We can go further and ask: if the common ancestor gave rise to two families; Artemiidae and Parartemiidae, then is not *A. persimilis* representative of a separate family (Argartemiidae)?

In discussing the processes and rates of diversification of *Artemia* we must keep in mind not only plate tectonics, but the general trends of global climate change (Sorokhtin, 2007). Spatial heterogeneity of climate was few pronounced at the time of the existence of proto*Artemia*, the climate was much warmer and drier. Consequently, areas with hypersaline water bodies were more abundant; they had larger territories. This is did not help the diversification of the taxon. What were the forerunners of modern *Artemia*, which lived at that time we can only guess. The Cenozoic era was the start of the slow cooling of the climate that led to the decrease in the total number of hypersaline water bodies, increasing the distance between them, the northern border of the possible habitat of proto*Artemia* gradually shifted to the south. All this stimulated the processes of diversification of the taxon. On the border of the Mesozoic and Cenozoic one of the largest extinctions in the history of the Biosphere occurred. The Cretaceous–Paleogene extinction event occurred approximately 65.5 million years ago. It was a large-scale mass extinction of animal and plant species in a geologically short period of time. 16% families of marine animals (47% of the genera of marine animals) and 18% of the families of terrestrial vertebrates had vanished (Marshall & Ward 1996). How proto*Artemia*s taxon was affected by this mass extinction we do not know, but, as a rule, after mass extinctions the remaining species began an intense diversification (Benton, 1995; Sahney & Benton, 2008). During the Early and Middle Miocene (23 ~ 12 Ma) there was much warmer than in previous period, after that cooling went again (Sorokhtin, 2007).

Probably some the predecessor gave rise to the formation of genus *Artemia* in lagoons of the northwestern coast of the Tethys 50-60 Ma. *A salina* have diverged about 40 Ma (Baxevanis *et al*., 2006). The drift of the African plate to the north led to a reduction of the area of the Tethys Ocean, the convergence of Africa and Eurasia. At the time of the Late Miocene (15 Ma) Tethys broke up into the Mediterranean Sea and Paratethys. Reducing the distance between Africa and Eurasia led to the formation of a new system of migratory routes of birds, which created an opportunity for *Artemia* introduction into the North African lagoons and salt lakes. *A. salina* originated. When exactly was the ingress of *Artemia* into Africa difficult to say. The existence of a continuous exchange of *Artemia* genes between the northern shores of the Mediterranean and Africa by birds had not allowed an emergence of an independent species of *Artemia* in North Africa. The question of the existence of *Artemia* populations in South Africa requires separate consideration. A later diversification of *A. salina* associated with the evolution of the Mediterranean Basin, but the Asian *Artemia* bisexual line - more with Paratethys and its subdivisions later. The Messianian Salinity Crisis - a geological event during which the Mediterranean Sea went into a cycle of partly or nearly complete desiccation throughout the latter part of the Miocene, from 5.96 to 5.33 Ma - impacted on the final divergence of the *A. salina* line, but for the nomination of a hypotheses on this subject is not enough data at this time. The Mediterranean Sea had ceased to exist, disintegrated into a series of hypersaline lakes. Salinities in these lakes were slowly increased, probably thereby creating conditions for the increasing of the genetic diversity of *A. salina* and its diversification (Baxevanis *et al*., 2006). The Asian *Artemia* line - bisexuals (*A. urmiana, A. tibetiana, A. sinica* and *A. sp.* from Kazakhstan) and parthenogens - last shared a common ancestor with *A. franciscana* 32 Ma (Baxevanis *et al*., 2006). Most likely, the common ancestor of Asian bisexuals existed in Europe, to north from the area of *A. salina* distribution, possibly in the lagoon of the expanding Atlantic Ocean or Tethys, for example, at the place where the East European plain and the Baltic Sea now. In the then existing climatic conditions, hypersaline water bodies, suitable for *Artemia*, were distributed much more widely, probably in Greenland also. At that time the distance between Europe and North America was much smaller, and biological contacts were extensive (Mayr, 2001). Birds could fly freely between these continents, passing *Artemia* cysts. Further expansion of the Atlantic Ocean made the migration of birds between Europe and America difficult. Simultaneously there was a cooling of the climate during the period 65-24 Ma (Sorokhtin, 2007) that shifted sharply the boundaries of distribution of *A. urmiana* line to the south and east. Contact between populations that have evolved in the *A. franciscana* with the line of *A. urmiana* was interrupted.

The collision of Indian plate and Asia, which occurred approximately 45-50 Ma and which gave the beginning of the process of mountain formations, which led to the formation of the highest mountains and the Tibetan plateau, and as result changed the climate (Molnar, 2005). The delayed impact of the collision on the diversification of *Artemia* requires a separate detailed analysis. *A. sinica* branched off the *A. urmiana* groups 8 Ma, within a period for which geological and climatological evidence demarcate the elevation of the Tibetan plateau and subsequent separation of eastern and western landscapes (Baxevanis *et al*., 2006). A rapid increase in mean elevation of eastern Tibet began at between 9 and 13 Ma (Clark *et al*., 2005). Tibet had reached its maximum elevation at ~ 8 ±1 Ma (Molnar, 2005). Based upon molecular genetics data some authors made a conclusion that *A. urmiana* may have played an important role in the evolution of *A. sinica* (Hou *et al*., 2006). We agree with the conclusion.

Focusing our attention on *A. urmiana*, we take the Miocene, as a starting point ~ 11 Ma (Manaffar *et al*., 2011), when *A. urmiana* branched from a common ancestor with *A. franciscana* in paleolagoons of the Crimea and surrounding areas, which extended far to the north and east. The Crimea is located at the crossroad of the Mediterranean Basin and the different parts of the Paratethys. According to current geological data, Crimea is a remnant of the paleoisland arc in the Tethys that existed in the Mesozoic (Shnukov *et al*., 1997). It should be noted the Black Sea level rise (about 7 thousand years ago) - due to the waters from the Mediterranean Sea breaching a sill in the Bosporus Strait ≈ 5600 BC (Dimitrov and Dimitrov, 2004) - which flooded vast tracts of land, and today this land lays under Karkinitsky Bay (Black Sea) and the Sea of Azov. The Crimea is a remnant of the submerged land in the hypersaline water bodies of which, according to our hypothesis, there was the *A. urmiana* formation in the Miocene. Cyst of *A. urmiana* was transported by birds to the east and north and gave rise to the formation of all Asian *Artemia* species and populations.

Move to the Pleistocene. The Pleistocene is the epoch from 2,588,000 to 11,700 years BP that spans the world's recent period of repeated glaciations (Richmond and Fullerton, 1986). Repeated glaciations drastically narrowed the areas for the possible existence of *A. urmiana,* but not in the Crimea. The last Ice Age in the Altai region ended about 11-14 Ka (Okishev, 1983) that was due to warming and increased aridity of the climate, which led to the emergence of new regional distribution of salt lakes - the potential habitats of brine shrimp. A phase of intensive settlement of *A. urmiana* started (cyst dispersion by birds and wind). After the last ice age, most likely, from the region of Crimea and surrounding areas *A. urmiana* had been put in Lake Urmia (no more than 5 thousand years ago) and the Altai Lake.

A. urmiana and *A. tibetiana* are genetically very closely related species (Baxevanis *et al*., 2006; Hou *et al*., 2006), *A. tibetiana* branched from *A. urmiana* relatively recently, much later than *A. sinica* branched from *A. urmiana* as molecular genetic data show (Baxevanis *et al*., 2006). During the period 40 - 28 Ka on Tibetan Plateau, there was a huge freshwater lake, which gradually shrank and broke up into smaller ones, some of which gradually salinized (Zheng *et al*., 2000). Formation of hypersaline lakes on the Tibetan plateau (Zheng, 1997) has created the possibility of introduction of *A. urmiana* there. Birds brought *A. urmiana* cysts to the Tibetian salt lakes from the Crimea through Kazakhstan (or by ancient people with salt and later?). When? It is possible that *A. urmiana* was introduced here before its introducing to Lake Urmia and the lakes of Altai region. After some time, probably due to the continued growth of the mountains framing Tibet (Pamir, Tian Shan, Kunlun), and the climate change that disrupted migration routes of birds. The migratory routes of birds, capable of carrying *Artemia* cysts, have changed, so that the interrupted gene flow between the parental *Artemia* populations and populations which presented on the Tibetan plateau. It gave rise to the divergence of new species – *A. tebitiana,* its final branching from *A. urmiana.* But we cannot exclude other scenarios of what happened: it was transported from the Lake Urmia or from lakes of Altai region only about five thousand years ago. Such a rapid evolution in the independent species can not be ruled out, knowing that the high salinity, as well as UV, may contribute to a sharp acceleration of the evolution of crustaceans (Hebert *et al*., 2002). It should be noted that there is another well-reasoned point of view: bisexual *Artemia* in Tibet and Kazakstan belong to the species *A. urmiana* (Hou *et al*., 2006) that is consistent with the hypothesis about recently introducing of A. *urmiana* in Asian water bodies - after the last glaciation event.

Finding of *A. sinica* in the Tuva lakes (Eastern Siberia) (Boyko & Muge, 2009) gives us the possibility to suggest considering the paleogeographic data (Ganyushkin, 2001) two scenarios of its introducing into these lakes: *A. sinica* was brought from China into the Tuva lakes/paleolakes, 50 -12 Ka by birds or people. Likely birds transported *Artemia* cysts from North China lakes/lagoons. But we cannot completely exclude their transport by ancient people (with salt). More than three thousand years ago, there were contacts between the territory of contemporary Tuva, which was part of the Xiongnu Empire at that time, and North China. The Xiongnu, a powerful nomadic people based in modern Mongolia and Southern Siberia, ruled Xiongnu. Relations between early Chinese dynasties and the Xiongnu were complex, with repeated periods of military conflict and intrigue alternating with exchanges of tribute, trade, and marriage treaties (Gumilev, 1960). But we cannot fully exclude the possibility that *A. sinica* is Miocene relict here, it is may be older than *A. sinica* in China. Taking into account paleogeographic data and cyst viability after long rest such hypothesis does not seem quite incredible.

In the Baikal region and Transbaikalia the bisexual populations of *Artemia* were found (Shadrin Anufrieva, this volume), on which genetic analysis was not performed. Species identification of these populations remains unclear. Elucidation of their species affiliation may shed light on the phylogeny and historical biogegraphy, as well as on the routs and settling time of the Asian species of *Artemia*.

Thus, there are compelling reasons to believe that the Crimea, together with adjoining land now submerged, could be one of the main centers of formation of species diversity of *Artemia*. Only by conducting molecular genetic studies of *Artemia* in the Crimea with subsequent comparison of the genetic diversity and similarity of *Artemia* populations in Crimea, Lake Urmia, Altai, Tibet, East Siberia, Kazakhstan and China could confirm and refine this hypothesis or reject it as inappropriate and contrived.

Conclusion

Despite the fact that the largest habitats of *Artemia* in the past (Siwash, Donuzlav, Kyzyl-Yar) ceased to exist, in the Crimea and the NW Black Sea (Kuyalnik) still have water with very promising opportunities for industrial harvesting of *Artemia* cysts. But there are a lot of small-scale reservoirs promising cysts harvesting for local needs. When planning the billets of cysts in water bodies of Ukraine we should take into account the high variability of annual cycles in the development of *Artemia* population in different water bodies. We can say that the organization of *Artemia* cyst harvesting in the Crimea is a science-intensive industry in need of understanding the dynamics of hypersaline lake ecosystems. The theoretical basis for such a framework should be created based on the concept of multiplicity of alternative stable states of ecosystems (Shadrin, 2010). In developing a strategy for sustainable use of *Artemia* resources, in particular, in the Crimea should be aware that brine shrimp performs important natural functions and provides valuable socio-economic services, such as participation in the formation of therapeutic mud (Ivanova, 1994).

The study of *Artemia* in different water bodies of the Crimea with the use of molecular genetic approaches is one way to better understanding of the relationship of phylogeny and distribution of modern species as a whole and to test the hypothesis that the Crimea played the crucial importance in the formation of species diversity of *Artemia* in Eurasia -the study of populations of brine shrimp in the Crimea is the key to understanding the evolution and biogeography of *Artemia.*

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