

REVIEW

AN OVERVIEW ON GENUS *ARTEMIA* LEACH, 1819 (CRUSTACEA: BRANCHIOPODA, ANOSTRACA) FROM ROMANIA

Alexia Berindean, Karina Paula Battes, Beatrice Kelemen, Mirela Cîmpean

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INTRODUCTION

The genus *Artemia* Leach, 1819, known as the brine shrimp, is distributed world-wide in inland and coastal hypersaline waters, characterized by variable chemical and hydrological conditions (Van Stappen, 2002; Maniatsi *et al.*, 2011). Besides its central role in lacustrine foodwebs, it is also of high economical importance, being heavily used in aquaculture (Van Stappen *et al.*, 2020). It was first described in 1758, by an unknown Iranian geographer (Asem, 2008), and the two sexes were only pictured clearly by Schläsler (1756). Linnaeus described it as *Cancer salinus* in 1758, but Leach transferred it to *Artemia salina* in 1819, this being the first *Artemia* species to be described (Asem *et al.*, 2010).

The genus is comprised of a few sibling species and superspecies, generally separated by reproductive isolation or characterized as clusters of populations (Browne and Bowen, 1991; Asem *et al.*, 2010). The current taxonomic classification of the genus is: Subphylum Crustacea Brönnich, 1772; Class Branchiopoda Latreille, 1817; Subclass Sarsostraca Tasch, 1969; Order Anostraca Sars, 1867; Family Artemiidae Grochowski, 1896; Genus *Artemia* Leach, 1819 (Browne and Bowen, 1991).

The species of this genus are distributed globally, apart from Antarctica (Vanhaecke *et al.*, 1987). *Artemia salina* (Linnaeus, 1758) is distributed in the Mediterranean basin and South Africa (Muñoz *et al.*, 2008; Asem *et al.*, 2010), along its asexual counterpart, *Artemia parthenogenetica* Bowen & Sterling, 1978 (Barigozzi, 1974).

The latter is distributed in Europe, Africa, Asia and Australia. The origin of *A. parthenogenetica* in Australia is discussed in McMaster *et al.* (2007) and Muñoz *et al.* (2010) with the spread being linked to migratory flyways and to the Asian populations. On the American side, *Artemia franciscana* Kellogg, 1906 is considered a superspecies (Browne and Bowen, 1991; Gajardo *et al.*, 2002), *Artemia persimilis* Piccinelli & Prosdocimi, 1968 is confined to South America, and *Artemia monica* Verrill, 1869 is particular to Mono

Lake, California (Asem *et al.*, 2010). *A. franciscana*, with the native range in America, Caribbean and Pacific islands, is responsible for massive invasions in Europe and Asia, due to the commercial use of cysts (Muñoz *et al.*, 2013; Muñoz *et al.*, 2014; Horváth *et al.*, 2018). The Asian populations are comprised of *Artemia sinica* Cai, 1989 in Central and Eastern Asia, *Artemia tibetiana* Abatzopoulos, Zhang & Sorgeloos, 1998 in China (Tibet), *Artemia urmiana* Günther, 1899, endemic to Urmia Lake in Iran (West Azerbaijan Province) (Asem *et al.*, 2010), and *Artemia* sp. Pilla & Beardmore, 1994 from Kazakhstan (Pilla and Beardmore, 1994).

The species of this genus are split between bisexual and asexual species. Based on genetic studies, Abreu-Grobois and Beardmore (1980, 1982) postulated that the ancestral bisexual form originated in the Mediterranean and in the Middle East. Old World and New World species are split accordingly: bisexual species: *A. salina*, *A. urmiana*, *A. tibetiana* and *A. sinica* in Eurasia; *A. franciscana* and *A. persimilis* in the Americas; and the asexual species *A. parthenogenetica*, found exclusively in the Old World and Australia (Maniatsi *et al.*, 2011; Hontoria *et al.*, 2012).

The bisexual species are diploid, with $2n=42$ (except for *A. persimilis* with 44), and the asexual species vary from $2n$ to $5n$ (Perez *et al.*, 1994; Maniatsi *et al.*, 2011). *A. parthenogenetica*, an obligate parthenogenetic form, has its origin, phylogeny and distribution still under analysis. (Muñoz *et al.* (2010) suggested that the origin of parthenogenesis in *Artemia* is in Central Asia, and that the maternal sexual ancestor of *A. parthenogenetica* is *A. urmiana* together with an undescribed species in Kazakhstan (*Artemia* sp.). It was also hypothesized that shortly after their origin, parthenogens underwent a rapid expansion in Central Asia and towards the Mediterranean Basin, reaching its current distribution.

This is thought to be caused by their adaptability to critical conditions (described in Gajardo and Beardmore, 2012) and the capacity of single females to establish an entire population. On

the issue of polyploidy in *A. parthenogenetica*, Maniatsi *et al.* (2011) analysed its origin and purpose in *Artemia*, and concluded that it occurred multiple times during its evolution and it gives evidence for the geographical distribution of parthenogens. Furthermore, they proposed that due to current inferences on multiple origins of parthenogenesis and polyploidy, as well as levels of diversity within parthenogens, the binomen *A. parthenogenetica*, suggested by Barigozzi (1974), should be abandoned, in favour of the concept of agamospecies (Mayr, 2001).

Since these closely related asexual lineages are identified on the basis of certain diagnostic or derived traits, the absence of sexual reproduction means that the biological species concept cannot be applied (Maniatsi *et al.*, 2011).

Another issue in *Artemia* taxonomy is the description of all individuals as *A. salina*, especially in older literature (Browne and Bowen, 1991; Asem *et al.*, 2010), thus making it difficult to rely on previous information on the species.

Due to the taxonomical issues described above, there is a lot of confusion regarding the real distribution of the genus in hypersaline lakes from Eastern Europe, Romania included. In this context, the aim of this paper is to review all the records of genus *Artemia* in Romania.

ARTEMIA DISTRIBUTION IN EUROPE

The distribution of *Artemia* in Romanian salt lakes is closely tied to that of Europe and the Mediterranean Basin. Many studies have described its biology (Barigozzi, 1974; Criel and Macrae, 2002; Gajardo and Beardmore, 2012; Zhang *et al.* 2013), morphology (Hontoria and Amat, 1992), biogeography (Vanhaecke *et al.*, 1987; Brtek and Thiéry, 1995; Triantaphyllidis *et al.*, 1998; Van Stappen, 2002), biodiversity (Amat *et al.*, 2007; Van Stappen, 2008; Eimanifar *et al.*, 2014), phylogeography ((Muñoz *et al.*, 2008; Muñoz *et al.*, 2010; Eimanifar *et al.*, 2014) and phylogeny (Baxevanis *et al.*, 2006; Maniatsi *et al.*, 2011).

Vanhaecke *et al.* (1987) describes over 350 *Artemia* sites, on 5 continents. Later, Triantaphyllidis *et al.* (1998), reviewed the distribution sites of *Artemia* worldwide, specifying the manner of reproduction and, where known, the species. For Europe and the Mediterranean, Triantaphyllidis *et al.* (1998) reported almost exclusively *A. parthenogenetica*, the Old-World asexual species, together with several records of *A. salina* in Cyprus, Italy and Spain. Subsequently, many studies described local populations of *Artemia*, from: Macaronesia (Hontoria *et al.*, 2012), Algeria (Selselet *et al.*, 2011); Portugal (Pinto *et al.*, 2013);

Crimea (Shadrinet *et al.*, 2012), some along other branchiopods.

In recent years, an increasing number of articles documented the dispersal of the invasive American species, *A. franciscana*. These invasions have been linked to both migratory flyways and to the commercial use of cysts (Muñoz *et al.*, 2013; 2014). Populations of *A. franciscana* have been described in Portugal, Spain, France, Italy (Scalone and Rabet, 2013; Muñoz *et al.*, 2014) and recently in the Balkans, in Zadar Peninsula, Croatia (Horvath *et al.*, 2018).

RECORDS OF ARTEMIA IN ROMANIA

First records of *Artemia* in Romania (Table 1) date back to the eighteenth century, and were made from the hypersaline lakes from Transylvania (Alexe *et al.*, 2018). Other early descriptions of *Artemia* sp. were also made from Transylvanian lakes (Maxim, 1931, 1937, 1943, from Ionescu *et al.*, 1998), but also from Lake Techirghiol (Bujor, 1900, 1928, 1930, from Ionescu *et al.*, 1998).

Botnariuc and Orghidan (1953) systematized the Phyllopod fauna from Romania, and cited *A. salina* as the only species present in all hypersaline lakes from the country (Table 1). Pora *et al.* (1960, 1962) and Onea *et al.* (1967, 1968) described *A. salina* in Lakes Techirghiol and Lacul Sărat Brăila, in the context of researches made on sapropels - sediments with therapeutic value. The following records, from 1978 to 1998, referred either to *A. salina* or to *Artemia* sp. (Table 1).

Triantaphyllidis *et al.* (1998) placed for the first time *A. parthenogenetica* in the Romanian hypersaline lakes outside Transylvania, based on personal communications and not published research. Van Stappen (2002) replaced *A. parthenogenetica* with "Artemia parthenogenetic strains" in the same lakes as Triantaphyllidis *et al.* (1998).

Demeter and Stoicescu (2008) mentioned *A. salina* once again, but did so based on the fact that it was the only species officially recorded in the Phyllopod fauna from Romania (Botnariuc and Orghidan, 1953).

Since no molecular studies were conducted on specimens from the Romanian salt lakes, as far as we know, the confusion remains: what species inhabit our saline lakes? *A. salina*, *A. parthenogenetica*, or the invasive *A. franciscana*? Due to the rapid expansion of this invasive American species (Muñoz *et al.*, 2014; Van Stappen *et al.*, 2020), and its arrival in the vicinity of Romania (Horvath *et al.*, 2018), we must be prepared to preserve our native lineages and species. It is therefore paramount to correctly identify and characterize our *Artemia* populations.

Table 1. Records of *Artemia* sp. from the Romanian salt lakes

Year	Taxa cited	References	Location and details
1794-1885	<i>A. salina</i> , <i>Artemia</i> sp.	J. Grossinger (1794) cited by J. Daday (1884) and G. Entz (1886); J. Hankó (1844) and V. Sill (1861), E. von Friedenfels (1880, 1885), from Alexe <i>et al.</i> , 2018	First observations, documented records and exhaustive description of <i>Artemia</i> sp. from Transylvanian lakes, part of the historical Hungary kingdom at that time (e.g. Lakes Turda and Ocna Sibiului).
1900-1930	<i>Artemia</i> sp.	Bujor, 1900, 1928, 1930 from Ionescu <i>et al.</i> , 1998	Records in Lake Techirghiol.
1931-1943	<i>Artemia</i> sp.	Maxim, 1931, 1937, 1943, from Ionescu <i>et al.</i> , 1998	Records in Lakes Ocna Sibiului, Turda, Sic.
1953	<i>A. salina</i>	Botnariuc and Orghidan, 1953	Record of <i>A. salina</i> as the only species in Romania, found in Lakes: Techirghiol, Tuzla (Constanța County), Telega (Ploiești County), Ocna Sibiului, Turda, Sovata, Cacica (Suceava County), and others.
1960-1962	<i>A. salina</i>	Pora <i>et al.</i> , 1960, 1962	Records in Lake Techirghiol.
1967-1968	<i>A. salina</i>	Onea <i>et al.</i> , 1967, 1968	Records in Lacul Sărat Brăila.
1978	<i>Artemia</i> sp.	Bulgăreanu <i>et al.</i> , 1978, from Ionescu <i>et al.</i> 1998	Records in Baia Miresii, Slănic Prahova.
1980	<i>Artemia</i> sp.	Trică, 1980, from Ionescu <i>et al.</i> 1998	Records in Lakes from Slănic Prahova: Baia Baciului, Baia Neagra, Baia Verde 1, Baia Verde 2 and Baia Verde 3.
1982	<i>Artemia</i> sp.	Ionescu <i>et al.</i> , 1982, from Ionescu <i>et al.</i> 1998	Record in Lake Ursu.
1987	<i>Artemia</i> sp.	Vanhaecke <i>et al.</i> 1987 (data source: Caspers, 1957; Pora <i>et al.</i> , 1960, 1962)	Record in Lake Techirghiol, parthenogenetic reproduction.
1996	<i>A. salina</i>	Bulgăreanu, 1996	Considered a frequent taxa with a major contribution to peloidogenesis (formation of muds with therapeutic value), in anthroposaline lakes (i.e. lakes formed by abandoned salt mine collapse or of mixed origin) from: Coștiui, Ocna Șugatag, Ocna Dej, Sic, Cojocna, Turda, Ocna Mureș, Sovata, Ocna Sibiului, Ocnele Mari, Slănic, Telega.
1998	<i>Artemia</i> sp.	Ionescu <i>et al.</i> , 1998	Review of previous Romanian records and original data: Lakes Ursu, Bătrân Ocna Șugatag; Cloșca Ocna Sibiului, Crișan Ocna Sibiului; Baia Baciului Slănic Prahova; Baia Neagră Slănic Prahova; Baia Verde 1 Slănic Prahova; Baia Verde 2 Slănic Prahova; Baia Verde 3 Slănic Prahova; Baia Miresii Slănic Prahova.
1998	<i>A. parthenogenetica</i>	Triantaphyllidis <i>et al.</i> , 1998 (data source: Liliana Pana, personal communication)	First mention of <i>A. parthenogenetica</i> : Lake Techirghiol, Lacul Sărat, Brăila; Baia Baciului Slănic Prahova; Baia Neagra, Slănic Prahova; Baia Verde I, Slănic Prahova; Baia Verde II, Slănic Prahova; Baia Verde III, Slănic Prahova; Baia Roșie, Slănic Prahova; Telega Băi; Telega II; Telega III.
2002	<i>Artemia</i> parthenogenetic strains	Van Stappen, 2002 (data source: Liliana Pana, personal communication)	<i>Artemia</i> parthenogenetic strains: Lake Techirghiol, Lacul Sărat, Brăila; Baia Baciului; Baia Neagra, SP; Baia Verde I, SP; Baia Verde II, SP; Baia Verde III, SP; Baia Roșie, SP; Telega Băi; Telega II; Telega III; species status unknown: Ocna Sibiului; Movila Miresii; Sovata.
2007-2008	<i>A. salina</i>	Demeter and Stoicescu, 2008	Review of previous records in lakes from: Sic, Turda, Techirghiol, Tuzla, Sângiorgiu Mureș, Sovata, Slănic Prahova, Telega, Ocna Sibiului, Cacica Suceava. Year of last record in Romania: 2007.
2018	<i>Artemia</i> sp.	Alexe <i>et al.</i> , 2018; personal data	Records of parthenogenetic populations in lakes from: Ocna Dej, Jucu, Cojocna, Turda, Ocna Mureș, Ocna Sibiului, Sovata, Techirghiol, Slănic Prahova, Lacul Sărat Brăila.

CONCLUSION

The present paper reviewed the previous records of genus *Artemia* in Romania, listing the references and the locations known in the literature. Future studies are strongly required in order to clarify the taxonomic issues associated with this genus in our region. *Artemia* species must be correctly identified and their ecology re-assessed, for better understanding the hypersaline lakes, extreme environments in desperate need for protection.

ABSTRACT

The genus *Artemia*, known as the brine shrimp, represents an important inhabitant of hypersaline lakes of Romania. Worldwide, the genus consists of six well-characterized bisexual species and several clonal lineages described with the binomen *Artemia parthenogenetica*. Despite its conspicuous appearance and numerous records in the literature, dating back to the eighteenth century, no positive identification can be provided for *Artemia* in Romania: the citations change from *Artemia salina*,

to *Artemia* sp, *Artemiaparthenogenetica*, or parthenogenetic strains of *Artemia*. The present paper represents a review of previous records of the genus in Romania. Further studies are mandatory to clarify all taxonomic issues associated with this genus.

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AUTHORS' ADDRESS

BERINDEAN ALEXIA, BATTES KARINA PAULA (correspondent), CÎMPEAN MIRELA - Babeş-Bolyai University, Faculty of Biology and Geology, Department of Taxonomy and Ecology, Cluj-Napoca, Romania; Center of Systemic Biology, Biodiversity and Bioresources "3B", Advanced Hydrobiology and Biomonitoring Laboratory (LabHAB), Babeş-Bolyai University, Cluj-Napoca, Romania; e-mails: alexia.berindean@stud.ubbcluj.ro, karina.battes@ubbcluj.ro, mirela.cimpean@ubbcluj.ro;

KELEMEN BEATRICE - Babeş-Bolyai University, Faculty of Biology and Geology, Department of Molecular Biology and Biotechnology, Cluj-Napoca, Romania; Molecular Biology Center, Interdisciplinary Research Institute on Bio-Nano-Sciences, Babeş-Bolyai University, Cluj-Napoca, Romania, e-mail: beatrice.kelemen@ubbcluj.ro.