

On the occurrence and density of some tardigrade taxa in the city area of Plovdiv, Bulgaria

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Abstract. There is insufficient data on tardigrades' distribution, diversity and density. Moss and lichen samples were collected from four different substrates – ground, walls, rocks and trees. Specimens from genus *Echiniscus* Schultze, 1840, genus *Hypsibius* Ehrenberg, 1848, genus *Milnesium* Doyère, 1840, genus *Paramacrobitous* Guidetti, Schill, Bertolani, Dandekar & Wolf, 2009, genus *Ramazzottius* Binda & Pilato, 1986 and family Macrobiotidae Thulin, 1928 were found.

Key words: Tardigrada, Plovdiv city, habitats.

Introduction

Tardigrades, also called water bears or moss piglets, are small metazoans within the size range of 0.1 – 1 mm. They are found on every continent, from marine, freshwater and terrestrial habitats. Tardigrades are best known for their ability to undergo cryptobiosis and survive unfavourable condition, such as desiccation up to 10 years (Jönsson & Bertolani 2001), -272.95°C for 20 hours or -200°C for 20 months (Miller 1997), 151°C for a few minutes (Lindahl & Balsler 1999), pressures of 6000 atmospheres (Seki & Toyoshima 1998) and the vacuum and UV radiation of space (Jönsson *et al.* 2008).

Despite their ability to survive extreme environmental conditions, little is known about their ecology, behaviour, densities and the structure of their populations, especially at human settlements. Different factors, such as temperature, moisture, pH of the substrate, altitude, food availability, air pollution, inter and intraspecific competition, combined or separately, may play role in tardigrades densities and distribution. Random dispersal can also be considered as factor affecting water bears' populations.

Quantitative and qualitative research on tardigrades' populations are generally focused on the relation between abundance and altitude, different microhabitats, urban and rural areas. Some studies on substrate specificity and spatial distribution of terrestrial tardigrades established that there is weak substrate specificity and that the variation in tardigrades' diversity and abundance is high (Meyer 2006a, 2006b).

The aim of this study is to provide information on moss and lichen dwelling limno-terrestrial tardigrades in a city environment, and the importance of the base substrate on their occurrence and specimen-densities.

Material and Methods

A total number of 87 moss and lichens samples were collected from Plovdiv city and Plovdiv region. The sampling was mainly focused on urban habitats (55.2%) with fewer samples collected from suburban (22.9%) and from habitats in Plovdiv vicinity (21.8%). The samples were collected from four main substrate types – ground (29.9% of all samples), walls (24.1%), rocks (11.5%) and trees (32.2%).

At the laboratory the samples were soaked in tap water for 6 to 24 hours. After this period, water containing tardigrades, their eggs and sample particles was decanted and further examined under stereomicroscope and light microscope. Specimens were mounted on microscope slides with glycerol or Hoyer's medium. A total of 252 tardigrade specimens from 6 genera and 1 family were registered.

All species were identified to genus level using original descriptions and modern keys mainly by Ramazzotti & Maucci (1983), Guidetti & Bertolani (2005); Michalczyk *et al.* (2012a, b); Kaczmarek *et al.* (2011); Morek *et al.* (2016), Kaczmarek & Michalczyk (2017), Kaczmarek *et al.* (2017). Systematic follows Degma *et al.* (2018).

Results and Discussion

Tardigrades were found in 46% of all collected samples, with individuals from genus *Milnesium* found in 17.8% of the samples. Water bears were discovered in 50% of the urban samples, in 36.8% of the suburban and in 50% of the vicinities' samples. Individuals belonging to genus *Echiniscus*, *Hypsibius*, *Paramacrobiotus*, *Ramazzottius*, family Macrobiotidae and unidentified eutardigrades were also found in the samples.

Genus *Echiniscus* had average density of 3.5 ind./cm³ for the urban samples and was registered in one suburban locality with density of 6 ind./cm³. The average density of *Milnesium* spp. from the urban habitats was 6.9 ind./cm³, for the suburban – 2.7 ind./cm³ and for the samples collected from Plovdiv vicinity – 1.8 ind./cm³. The average density of family Macrobiotidae was 12.8 ind./cm³ for urban habitats and 6 ind./cm³ for Plovdiv vicinity, with no individuals identified from the suburban habitats. Specimens from genus *Hypsibius* and *Paramacrobiotus* were registered with densities of 1 individuals/cm³ and were found in few localities. The specimens from genus *Ramazzottius* had average of 5.1 ind./cm³ from urban samples, 5 ind./cm³ from suburban and 2 ind./cm³ from samples from Plovdiv vicinity.

Echiniscus specimens had average density of 4 ind./cm³ for rocks and were established in one sample from ground substrate with density of 5 ind./cm³, with no individuals found from walls and trees. In the present study highest average density of *Milnesium* spp. was established in samples collected from rocks (8.5 ind./cm³) and lowest – from trees (3 ind./cm³). The individuals of family Macrobiotidae had highest average density for samples from ground substrate (16 ind./cm³) and lowest - from rock substrate (3 ind./cm³). The higher density for ground substrates is due to sample containing more than 40 ind./cm³. Individuals from genus *Hypsibius* were established in two tree samples with average density of 1 ind./cm³ and specimens from genus *Paramacrobiotus* were present in one tree sample with density of 1 ind./cm³. *Ramazzottius* spp. had higher density for samples from walls (13 ind./cm³) and lower for rock formations (3 ind./cm³), trees (2.8 ind./cm³), with no individuals found from ground substrates (Fig. 1).

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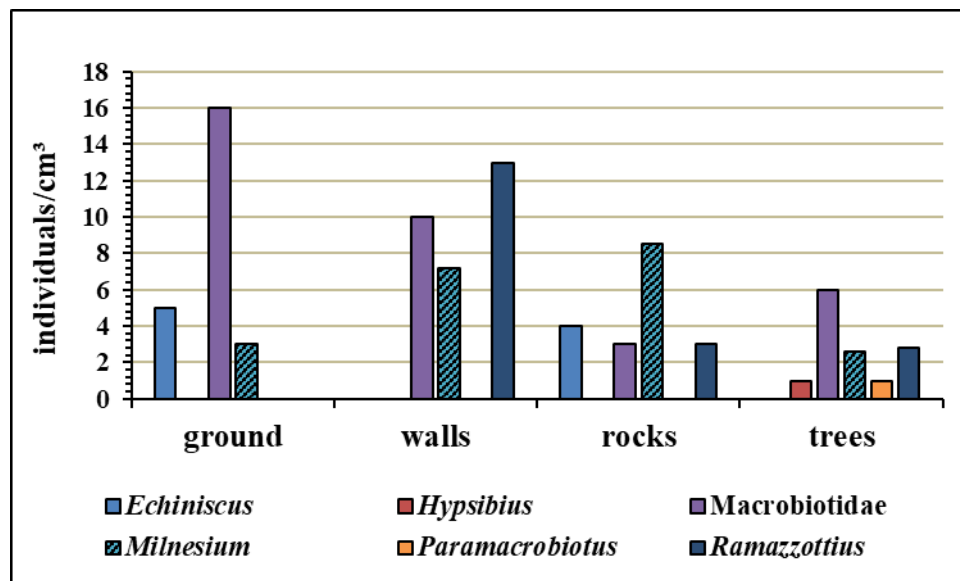


Fig. 1. Individuals per 1cm³ moss or lichens samples from different substrates.

References

- Degma, P., Bertolani, R. & Guidetti, R. (2009 – 2018) Actual checklist of Tardigrada species. Available at: <http://www.tardigrada.modena.unimo.it/miscellanea/Actual%20checklist%20of%20Tardigrada.pdf> (Accessed on 30 June 2018).
- Guidetti, R. & Bertolani R. (2005) Tardigrade taxonomy: an updated checklist of the taxa and a list of characters for their identification. *Zootaxa*, 845 (1):1-46.
- Jönsson, K. I. & Bertolani, R. (2001) Facts and fiction about long-term survival in tardigrades. *Journal of Zoology*, 255 (1): 121-124.
- Jönsson, K. I., Rabbow, E., Schill, R. O., Harms-Ringdahl, M. & Petra Rettberg, P. (2008) Tardigrades survive exposure to space in low Earth orbit. *Current Biology*, 18 (7): R729-R731.
- Kaczmarek, Ł. & Michalczyk, Ł. (2017) The *Macrobiotus hufelandi* group (Tardigrada) revisited. *Zootaxa*, 4363 (1): 101-123.
- Kaczmarek, Ł., Gawlak, M., Bartels, P., Nelson, D. & Roszkowska, M. (2017) Revision of the genus *Paramacrobotus* Guidetti et al., 2009 with the description of a new species, re-descriptions and a key. *Annales Zoologici*, 67 (4):627-656.
- Kaczmarek, Ł., Gołdyn, B., Prokop, Z. & Michalczyk, Ł. (2011) New records of Tardigrada from Bulgaria with the description of *Macrobiotus binieki* sp. nov. (Eutardigrada: Macrobiotidae) and a key to the species of the harmsworthi group. *Zootaxa*, 2781 (2): 29-39.
- Lindahl, K. & Balser, S. (1999) Tardigrade Facts. Illinois Wesleyan University. Available at: http://www.iwu.edu/~tardisp/tardigrade_facts.html. (Accessed on 29 April 2009).
- Meyer, H. A. (2006a) Interspecific association and substrate specificity in tardigrades from Florida, southeastern United States. *Hydrobiologia*, 558 (1): 129-132.
- Meyer, H. A. (2006b) Small-scale spatial distribution variability in terrestrial tardigrade populations. *Hydrobiologia*, 558 (1): 133-139.
- Michalczyk, Ł., Welnicz, W, Frohme, M. & Kaczmarek, Ł. (2012a) Redescriptions of three *Milnesium* Doyère, 1840 taxa (Tardigrada: Eutardigrada: Milnesiidae), including the nominal species for the genus. *Zootaxa*, 3154 (1): 1-20.
- Michalczyk, Ł., Welnicz, W, Frohme, M. & Kaczmarek, Ł. (2012b) Corrigenda of Zootaxa, 3154: 1-20: Redescriptions of three *Milnesium* Doyère, 1840 taxa (Tardigrada:

- Eutardigrada: Milnesiidae), including the nominal species for the genus. *Zootaxa*, 3393 (1): 66-68.
- Miller, W. R. (1997) Tardigrades: Bears of the moss. *Kansas School Naturalist*, 43 (3): 3-15.
- Morek, W., Gąsiorek, P., Stec, D., Blagden, B. & Michalczyk, Ł. (2016) Experimental taxonomy exposes ontogenetic variability and elucidates the taxonomic value of claw configuration in *Milnesium* Doyère, 1840 (Tardigrada: Eutardigrada: Aporhina). *Contributions to Zoology*, 85 (2), 173-200.
- Nelson, D. R. 2002. Current status of the Tardigrada: Evolution and ecology. *Integrative and Comparative Biology*, 42 (3): 652-659.
- Ramazzotti, G. & Maucci, W. (1983) Il Phylum Tardigrada. *III edizione riveduta e aggiornata. Memorie dell'Istituto Italiano di Idrobiologia*, 41 (1):1-1012.
- Seki, K. & Toyoshima, M. (1998) Preserving tardigrades under pressure. *Nature*, 395 (1): 853-854.
- Wright, J. C. (1991) The significance of four xeric parameters in the ecology of terrestrial Tardigrada. *Journal of Zoology*, 224 (1): 59-77.