

**BRAZILIAN LIMNOTERRESTRIAL TARDIGRADES (BILATERIA,  
TARDIGRADA): NEW OCCURRENCES AND SPECIES CHECKLIST  
UPDATES.**

Clélia Márcia Cavalcanti DA ROCHA<sup>1\*</sup>, Edivaldo Lima GOMES JÚNIOR<sup>2</sup>, Érika  
Cavalcante Leite DOS SANTOS<sup>3,4</sup>

<sup>1</sup>UFRPE - Depto. de Biologia - Av. Manoel de Medeiros - Dois Irmãos - Recife – Pernambuco - Brasil. CEP 52171-090

\*Corresponding author. Email: cavalcanticlelia@yahoo.com.br.

<sup>2</sup>Universidade Federal de Pernambuco; CCB; Depto. de Zoologia/PPGBA. Av. Prof. Moraes Rêgo S/N, Cidade Universitária - – Recife - Pernambuco – Brasil. CEP 50670-420

<sup>3</sup>Faculdade de Ciências da Universidade do Porto – Departamento de Biologia – Rua do Campo Alegre S/N – 4169-007 – Porto – Portugal.

<sup>4</sup>Fundaçao CAPES, Ministério da Educação, Brasília – DF. CEP 70.040-020.

**RESUMO**

Inventários de tardígrados limnoterrestres brasileiros foram publicados até o início da década de 1950, mas sessenta anos se passaram desde o último registro. No estado de Pernambuco, foram recentemente identificados tardígrados pertencentes a seis espécies: *Pseudechiniscus novaezealandiae aeaspinosa*, *Milnesium tardigradum tardigradum*, *Doryphoribus flavus*, *Macrobiotus harmsworthi harmsworthi*, *Macrobiotus hufelandi hufelandi* e *Minibiotus aculeatus*. Estes resultados nos levaram a rever o checklist de espécies brasileiras. O presente artigo objetivou atualizar este checklist, para o que comparamos todas as publicações existentes até a década de 1950 com o checklist global. Esta revisão levou a algumas modificações para a lista original. Somando os resultados atuais à revisão das listas pré-existentes para o país elaboramos um checklist atualizado, com um total de 62 espécies de tardígrados de habitats limnoterrestres conhecidas para o Brasil.

**Palavras chave:** *Doryphoribus*, *Macrobiotus*, *Milnesium*, *Minibiotus*, *Pseudechiniscus*, Brasil.

**ABSTRACT**

Inventories of limnoterrestrial tardigrades in Brazil were published up to the early 1950s, but sixty years passed by since the last record. In the state of Pernambuco we identified tardigrades of six species: *Pseudechiniscus novaezealandi aeaspinosa*, *Milnesium tardigradum tardigradum*, *Doryphoribus flavus*, *Macrobiotus harmsworthi harmsworthi*, *Macrobiotus hufelandi hufelandi* and *Minibiotus aculeatus*. These results led us to review the checklist of Brazilian tardigrades. This study aimed to update this checklist, for what we compared all publications up to the 1950s to the global list. This revision led to some modifications to the original checklist. Summing the current results to those of previous publications we reached a total of 62 limno-terrestrial tardigrade species known for Brazil.

**Key words:** *Doryphoribus*, *Macrobiotus*, *Milnesium*, *Minibiotus*, *Pseudechiniscus*, Brazil.

## INTRODUCTION

Tardigrades are hygrophilous micrometazoans (0.05 to 1.20 mm) that inhabit a wide variety of environments in all continents and oceans. These habitats include marine, freshwater, and terrestrial environments such as soils, moss, and lichens. However, tardigrades are active only in the presence of water (Beasley *et al.* 2006). Due to this characteristic, the denomination 'limnoterrestrial' is more adequate to species that inhabit continental biomes.

Rahm (1932) began the taxonomic inventory of limnoterrestrial tardigrades in Brazil and was followed by Marcus (1936, 1937), Barros (1938, 1939a, b, 1942a, b, 1943) and Du Bois-Reymond Marcus (1944, 1952). Except for rare communications made in scientific events (Ozório *et al.* 2008; Da Rocha *et al.* 2012), more than 60 years have elapsed without any new record for the tardigrade fauna in Brazil. We identified in the state of Pernambuco individuals of 6 species, 5 genera and 4 families: *Pseudechiniscus novaezeelandi aspinosa* Iharos, 1963; *Milnesium tardigradum tardigradum* Doyère, 1840; *Doryphoribus flavus* Iharos, 1966; *Macrobiotus harmsworthi harmsworthi* Murray, 1907; *Macrobiotus hufelandi hufelandi* C.A.S. Schultze, 1833 and *Minibiotus aculeatus* Murray, 1910. Two species were the first records in Brazil: *Doryphoribus flavus* and *Minibiotus aculeatus*. So, the present study also contributes with

the update of the Brazilian species list.

## METHODS

Using metal scrapers, we collected mosses present on tree trunks at the campus of the Federal Rural University of Pernambuco (UFRPE) located in Dois Irmãos, Recife, northeastern Brazil (8°00' S - 34°08' W; Fig. 1). We stored samples in plastic bags and followed the procedures suggested by Christenberry (1979): to extract the tardigrades, the material was rehydrated and observed under a binocular stereomicroscope. The animals and their eggs were manually removed with stainless steel tweezers and mounted on permanent slides for microscopy, using Hoyer's medium and potassium iodide solution (Iodine tincture). We identified tardigrades under a microscope (Olympus CX 31), using pictorial keys by Nelson & MCinnes (2002), Pilato & Binda (2010), and Ramazzotti & Maucci (1983). We made microphotographs using a digital camera (Olympus C-5050 Zoom).

To update the Brazilian species checklist, we used the studies by Rahm (1932), Marcus (1936, 1937), Barros (1938, 1939a, b, 1942a, b, 1943), Du Bois-Reymond Marcus (1944, 1952), and Assunção (1999). We also compared the Brazilian species list with the up-to-date global list of tardigrades by Degma, Bertollani & Guidetti (2009-2012), and followed its order in Results.

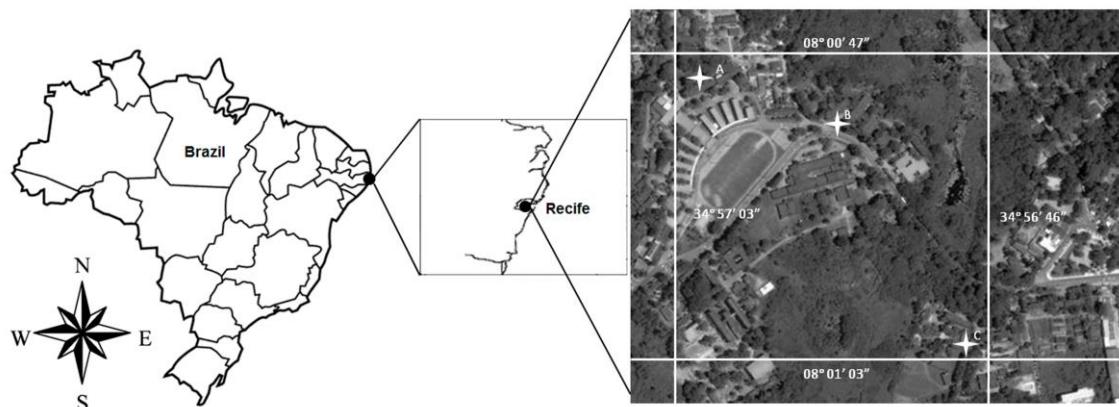


Figure 1. Location of the sampling site (A, B and C: sampling points).

## RESULTS

The original Brazilian species list needed some modifications: two species were invalidated (*Echiniscus fisheri* and *Macrobiotus sawaya*) and seven species were reclassified: *Diphascon scoticum* Murray, 1905 was placed in the genus *Adropion* Pilato, 1987 (*A. scoticum* Murray, 1905), *Isohypsibius augusti* Murray, 1907 was placed in the genus *Thulinius* Bertolani, 2003 (*T. augusti* Murray, 1907) and five species of the genus *Macrobiotus* were reclassified: *Macrobiotus acontistus* Barros, 1942, *Macrobiotus julietae* Barros, 1942 and *Macrobiotus marcus* Barros, 1942 were placed in the genus *Minibiotus* Schuster, 1980. Now these species are named *Minibiotus acontistus* Barros, 1942, *Minibiotus julietae* Barros, 1942 and *Minibiotus marcus* Barros, 1942. *Macrobiotus richtersii* Murray, 1911 was placed in the genus *Paramacrobiotus* Guidetti et al., 2009 (*P. richtersi* Murray, 1911). Finally, *Macrobiotus harmsworthi coronata* Barros, 1942 was reclassified to *M. coronatus* Barros, 1942.

In the samples collected in Pernambuco, we identified

individuals of 6 species, 5 genera, and 4 families, as reported below:

**Class:** HETEROTARDIGRADA Marcus, 1927  
**Order:** ECHINISCOIDEA Richters, 1926  
**Family:** Echiniscidae Thulin, 1928  
**Genus:** *Pseudechiniscus* Thulin, 1911  
**Species:** *Pseudechiniscus novaezeelandi aspinosa* Iharos, 1963

**Material examined:** 15 specimens collected in Dois Irmãos (Recife, state of Pernambuco, Brazil).

**Distribution:** Brazil (Du Bois-Reymond Marcus, 1944), Argentina, Colombia (Jerez Jaimes et al., 2001), and United States (Meyer, 2001).

**Ecological notes:** Found in mosses on trees.

**Class:** EUTARDIGRADA Richters 1926  
**Order:** APOCHELA Schuster, Nelson, Grigarick & Christenberry, 1980  
**Family:** Milnesiidae Ramazzotti, 1962  
**Genus:** *Milnesium* Doyère, 1840  
**Species:** *Milnesium tardigradum tardigradum* Doyère, 1840

**Material examined:** 4 specimens collected in Dois Irmãos (Recife, state of Pernambuco, Brazil).

**Distribution:** Cosmopolitan species (Meyer, 2001). This species had been previously recorded in Brazil (Assunção, 1999).

**Ecological notes:** Found in mosses on trees.

Order: PARACHELA Schuster, Nelson, Grigarick & Christenberry, 1980  
Family: Hypsibiidae Pilato, 1969  
Sub-family: Hypsibiinae Pilato, 1969  
Genus: *Doryphoribius* Pilato 1969  
Species: *Doryphoribius flavus* Iharos, 1966.

**Material examined:** 12 specimens collected in Dois Irmãos (Recife,

state of Pernambuco, Brazil) (Fig. 2).

**Distribution:** China (Beasley *et al.*, 2006), United States (Hinton & Meyer, 2007), Ecuador (Pilato *et al.* 2012) Costa Rica, Dominican Republic, Hungary, Italy, Turkey, Spain, South Africa and Cameroon (McInnes, 1994) This is the first record of this species in Brazil.

**Ecological notes:** Found in mosses under rocks and on trees.

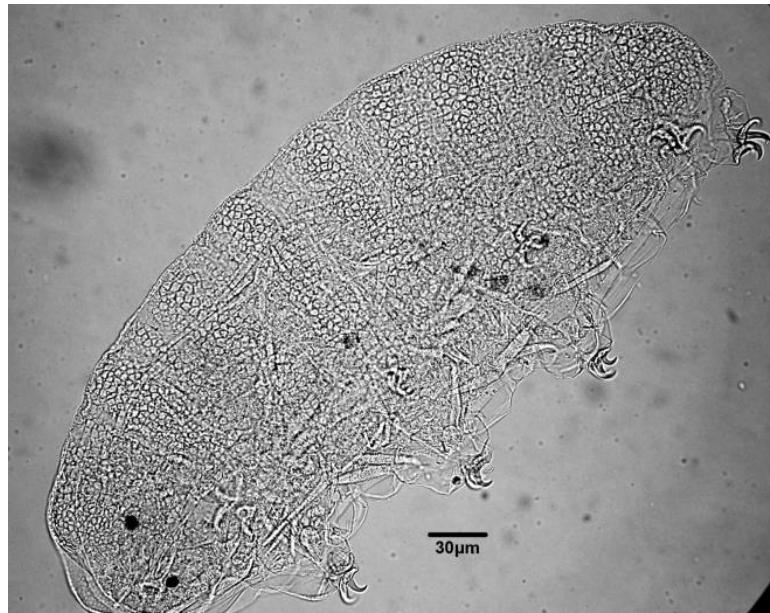


Figure 2. *Doryphoribius flavus* collected in Dois Irmãos (Recife, state of Pernambuco, Brazil).

Family: Macrobiotidae Thulin, 1928  
Genus: *Macrobiotus* C.A.S. Schultze, 1834  
Species: *Macrobiotus harmsworthi* harmsworthi Murray, 1907

**Material examined:** 8 specimens collected in Dois Irmãos (Recife, state of Pernambuco, Brazil).

**Distribution:** Probably a cosmopolitan species. Brazil, Tierra del Fuego, California (USA), Galapagos Islands, Croatia, Montenegro, Italy (Ramazzotti & Maucci, 1983), Antarctica (Utsugi & Ohyama, 1989), and Colombia

(Jerez Jaimes *et al.*, 2001; Lisi, 2014).

**Ecological notes:** Found in mosses in general.

Species: *Macrobiotus hufelandi hufelandi* C.A.S. Schultze, 1834

**Material examined:** 5 specimens collected in Dois Irmãos (Recife, state of Pernambuco, Brazil).

**Distribution:** Cosmopolitan species (Ramazzotti & Maucci, 1983). This

species had been previously recorded in Brazil (Assunção, 1999).  
**Ecological notes:** Found in soils, mosses, and even in freshwater environments.

Genus: *Minibiotus* R.O. Schuster, 1980  
Species: *Minibiotus aculeatus* Murray, 1910.

**Material examined:** 10 specimens collected in Dois Irmãos (Recife,

state of Pernambuco, Brazil); (Fig. 3).

**Distribution:** Australia and in the Carpathian region (Eastern Europe) (Ramazzotti & Maucci, 1983), Indonesia, Czech Republic and Slovakia (MCinnes, 1994). This is the first record of this species in Brazil.

**Ecological notes:** Found in mosses on trees.

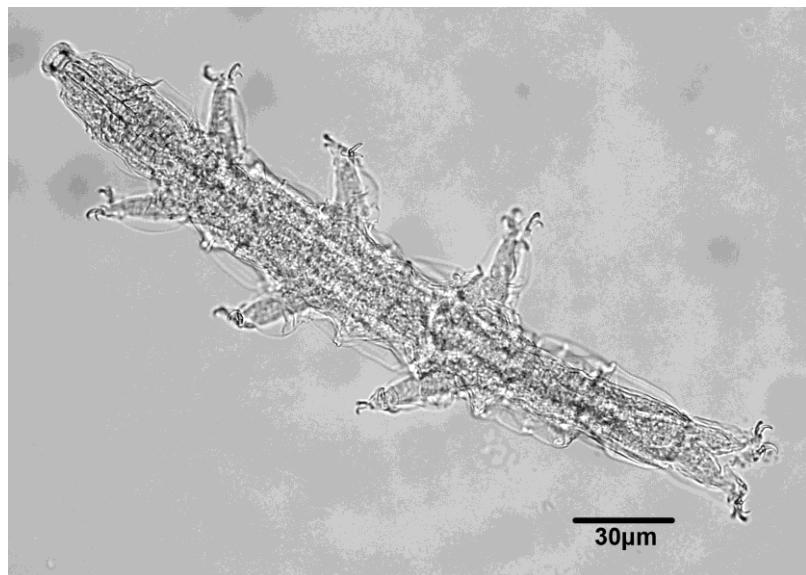


Figure 3. *Minibiotus aculeatus* collected in Dois Irmãos (Recife, state of Pernambuco, Brazil).

Our results increased to 62 the number of species of limnoterrestrial tardigrades in Brazil,

according to the up-to-date checklist presented in Table I.

**Table I.** Up-to-date checklist of limnoterrestrial tardigrades recorded in Brazil.

<i>Species</i>	<i>Distribution in Brazil</i>
<b>HETEROTARDIGRADA</b>	
<i>Brychoerus intermedius</i>	São Paulo
<i>Bryodelphax alzirae</i>	São Paulo
<i>Bryodelphax parvulus</i>	São Paulo
<i>Echiniscus blumi</i>	São Paulo
<i>Echiniscus crassispinosus fasciatus</i>	São Paulo and Paraná
<i>Echiniscus dreyfusi</i>	São Paulo
<i>Echiniscus duboisi</i>	São Paulo

<i>Echiniscus evelinae</i>	<b>São Paulo and Paraná</b>
<i>Echiniscus phocae</i>	<b>São Paulo</b>
<i>Echiniscus rufoviridis</i>	<b>São Paulo</b>
<i>Echiniscus spiniger</i>	<b>São Paulo</b>
<i>Echiniscus tenuis</i>	<b>São Paulo</b>
<i>Echiniscus testudo</i>	<b>São Paulo</b>
<i>Echiniscus viridis</i>	<b>São Paulo</b>
<i>Mopsechiniscus imberbis</i>	<b>São Paulo</b>
<i>Pseudechiniscus bispinosus</i>	<b>Pernambuco</b>
<i>Pseudechiniscus juanitae</i>	<b>São Paulo</b>
<i>Pseudechiniscus novaezeelandi aspinosa</i>	<b>São Paulo, Paraná and Pernambuco</b>
<i>Pseudechiniscus novaezeelandi marinae</i>	<b>São Paulo</b>
<i>Pseudechiniscus suillus</i>	<b>São Paulo and Paraná</b>
<i>Testechiniscus macronyx</i>	<b>São Paulo</b>
<b>EUTARDIGRADA</b>	
<i>Adropion scoticum</i>	<b>São Paulo</b>
<i>Calohypsibius verrucosus</i>	<b>São Paulo</b>
<i>Dactilobiotus ambiguus</i>	<b>São Paulo</b>
<i>Diphascon alpinus</i>	<b>São Paulo</b>
<i>Diphascon pingue</i>	<b>São Paulo</b>
<i>Doryphoribus evelinae</i>	<b>São Paulo and Minas Gerais</b>
<i>Doryphoribus flavus</i>	<b>Pernambuco</b>
<i>Hypsibius convergens</i>	<b>São Paulo</b>
<i>Hypsibius microps</i>	<b>São Paulo</b>
<i>Isohypsistius granulifer</i>	<b>São Paulo</b>
<i>Isohypsistius myrops</i>	<b>São Paulo and Rio Grande do Sul</b>
<i>Isohypsistius nodosus</i>	<b>São Paulo</b>
<i>Isohypsistius papillifer</i>	<b>São Paulo</b>
<i>Isohypsistius papillifer bulbosus</i>	<b>São Paulo</b>
<i>Itaquascon umbellinae</i>	<b>São Paulo</b>
<i>Macrobiotus coronatus</i>	<b>São Paulo</b>
<i>Macrobiotus echinogenitus</i>	<b>São Paulo</b>
<i>Macrobiotus evelinae</i>	<b>São Paulo</b>
<i>Macrobiotus furciger</i>	<b>São Paulo</b>
<i>Macrobiotus harmsworthi</i>	<b>São Paulo, Paraná and Pernambuco</b>
<i>Macrobiotus hibiscus</i>	<b>São Paulo</b>
<i>Macrobiotus hufelandii</i>	<b>São Paulo and Pernambuco</b>
<i>Macrobiotus occidentalis</i>	<b>Paraná</b>
<i>Macrobiotus orcadensis</i>	<b>São Paulo</b>
<i>Macrobiotus polyopus</i>	<b>São Paulo</b>
<i>Macrobiotus primitivae</i>	<b>São Paulo</b>
<i>Macrobiotus psephus</i>	<b>São Paulo</b>
<i>Macrobiotus stellaris</i>	<b>São Paulo</b>
<i>Milnesium tardigradum</i>	<b>São Paulo and Pernambuco</b>

<i>Milnesium tardigradum trispinosa</i>	<b>São Paulo</b>
<i>Minibiotus acontistus</i>	<b>São Paulo</b>
<i>Minibiotus aculeatus</i>	<b>Pernambuco</b>
<i>Minibiotus furcatus</i>	<b>São Paulo</b>
<i>Minibiotus intermedius</i>	<b>São Paulo and Paraná</b>
<i>Minibiotus julietae</i>	<b>São Paulo</b>
<i>Minibiotus marcusii</i>	<b>São Paulo</b>
<i>Murrayon pullari</i>	<b>São Paulo</b>
<i>Paramacrobiotus richtersii</i>	<b>São Paulo</b>
<i>Ramazzottius oberhaeuseri</i>	<b>São Paulo</b>
<i>Thulinus augusti</i>	<b>São Paulo</b>

## DISCUSSION

Rahm (1932), who began the studies about limno-terrestrial tardigrades in Brazil, recorded the presence of *Pseudechiniscus bispinosus* Murray, 1907 in the city of Olinda (state of Pernambuco, Brazil). Among the six species recorded here *Minibiotus aculeatus* and *Doryphoribus flavus* were recorded for the first time in Brazil.

It is important to highlight that *M. aculeatus* had been previously recorded in Australia and Eastern Europe, localities geographically distant from Brazil. Pilato & Binda (2001), based mainly on species of the genera *Macrobiotus* and *Minibiotus*, suggested that the distribution of tardigrades reflects main paleogeographic events (Beasley *et al.* 2006). This hypothesis could explain some biogeographic findings such as those reported here, as another hypothesis which considers tardigrades cosmopolitan due to their passive dispersal (Pilato & Binda, 2001). However, nowadays this hypothesis has been reconsidered, since 68% of the tardigrades are found in only one biogeographic region and only 6.8% are considered cosmopolitan. It is also important to consider that for

every six species recorded in the Palearctic region there is only one in the Neotropical region; much of the information about taxonomy and ecology of tardigrades is concentrated in Europe (MCinnes, 2007).

Pooling our results with previous studies, the total of limnoterrestrial tardigrades species known for Brazil reaches 62. In comparison with Italy, whose tardigrade fauna is well known (250 limnoterrestrial species) and where new species have still been often described (Pilato & Bertolani, 2005), the present species list might be a tiny part of what we suppose to be the biodiversity of tardigrades in Brazil, which still remains unknown.

The limnoterrestrial tardigrades have received much attention from the scientific community due to their unique biological characteristics that allow them to resist several situations of environmental stress, with a potential to be used in many branches of Science. Despite this recent global interest in the group, according to Guil & Cabrero-Sañudo (2007), 93% of the species recorded in South and Central America have been made by foreign scientists,

what indicates the lack of specialists in the region.

## CONCLUSION

Our results contribute to renew the interest of the Brazilian scientific community in this group historically disregarded in the country, and point to the need of further taxonomic efforts regarding the local fauna. Taking as an example our results, which may set the basis for very interesting biogeographic studies, we conclude that such efforts could result in invaluable findings to advance the knowledge about the group, and consequently, to effectively continue these studies in Brazil.

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