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Key to the identification of the genera of bdelloid rotifers

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Abstract

We propose a simple, user-friendly key joined to a pictorial key to the bdelloid genera, in the attempt to make bdelloid identification more accessible to non-bdelloid-specialists. The key is mostly based on illustrations rather than on descriptions, and is accompanied by an introduction of the main features readily observable in active bdelloids.

Introduction

Bdelloids constitute a class of the phylum Rotifera, distinguished by two characteristics that are autoapomorphic to the whole class: (1) obligatory parthenogenetic reproduction (Gilbert, 1983), and (2) ability to withstand harsh periods through anhydrobiosis (Ricci, 1987, 1998). According to the more recent classification proposed for the taxon (Melone & Ricci, 1995), the class consists of three orders (Philodinavida, Philodinida and Adinetida), four families, and about 350 species assigned to 19 genera, recognized on morphology only (Donner, 1965). Bdelloids are widespread freshwater and soil inhabitants, able to use the thin water film surrounding soil particles, lichens or mosses. Most bdelloids are able to swim, but all tend to creep on surfaces; some bdelloid species (e.g. *Rotaria*) can be found occasionally in the plankton, but the typical habitat of the class is benthos or periphyton of lotic and lentic waters.

The size of the bdelloids ranges between 150 and 700 μm, but some species are exceptionally long (1600 μm, *Rotaria neptunia*). Although largely ignored, and mostly insignificant in terms of biomass like all small animals, bdelloid rotifers can be very abundant in the sediments of lotic and lentic waters (i.e. Schmid-Araya, 1993, 1995). Because of that, they can play a key role in food webs as most energy passes through small rather than large organisms (e.g. Ward et al., 1998). Most studies recording the taxonomic composition of freshwater microinvertebrate assemblages disregard the bdelloid communities and provide total counts of the bdelloid specimens only, without discriminating lower taxon categories (i.e. Kobayashi et al., 1998). The same is often done by rotifer students, too, who deal with the major class of Rotifera, the monogononts (i.e. De Smet, 1990). Thus, bdelloid identification is left to the few specialists, and the global information on taxa occurrence is scant and insufficient. The worldwide lack of interest in this neglected group of rotifers is mainly due to the difficulty of identifying the animals, for two reasons.

First, most bdelloids can be identified when alive only, because the different traits that are taxonomically significant can be observed only when the rotifer is active, feeds and crawls. This fact severely restricts the activity of the scientist, who has to collect the samples in the field and access a microscope as soon as possible in order to observe healthy and active specimens. Alternatively, dry samples of soil and moss can be collected and stored and, when convenient, hydrated: the bdelloids will be active in about two, six hours. As bdelloids often move restlessly and often rapidly contract and change their shape, diagnostic characters are not easily accessible, and bdelloid identification is mostly a matter of patience and prolonged observation.

Secondly, the available identification keys are quite

difficult to use. Two diagnostic keys to bdelloids are available at present, both somewhat dated, written by Bartoš (1951) and Donner (1965). The former is written in English, but is rather poor in details, often confusing and not very reliable. The latter is the most complete, well illustrated, but out of print and written in German, so that its access is restricted to the German-speaking scientific community. More recently, to illustrate a number of genera and species present in inland waters of Australia and New Zealand, Shiel (1995) produced a diagnostic key to the bdelloid species recorded.

Thus, identification of bdelloids requires a skilled observer, only few students are actually dealing with them, and most scientists record only that bdelloids are present in their samples, without further identification. While the difficulty with handling and recognizing bdelloid features is hard to be circumvented, we attempt to make bdelloid identification more accessible, by providing a simple, user-friendly key joined to a pictorial key to the bdelloid genera. The key is mostly based on illustrations rather than on descriptions, and we hope that it will encourage more scientists to approach this very common group of rotifers. The key is accompanied by an introduction of the main features readily observable in active bdelloids, without going into details, and referring to the most recent literature. The key does not reflect the phylogenetic relationships within the taxon, but provides only the diagnostic path that can easily be followed by non-specialists, leaving to the specialists the use of more comprehensive and detailed guides (i.e. Donner, 1965).

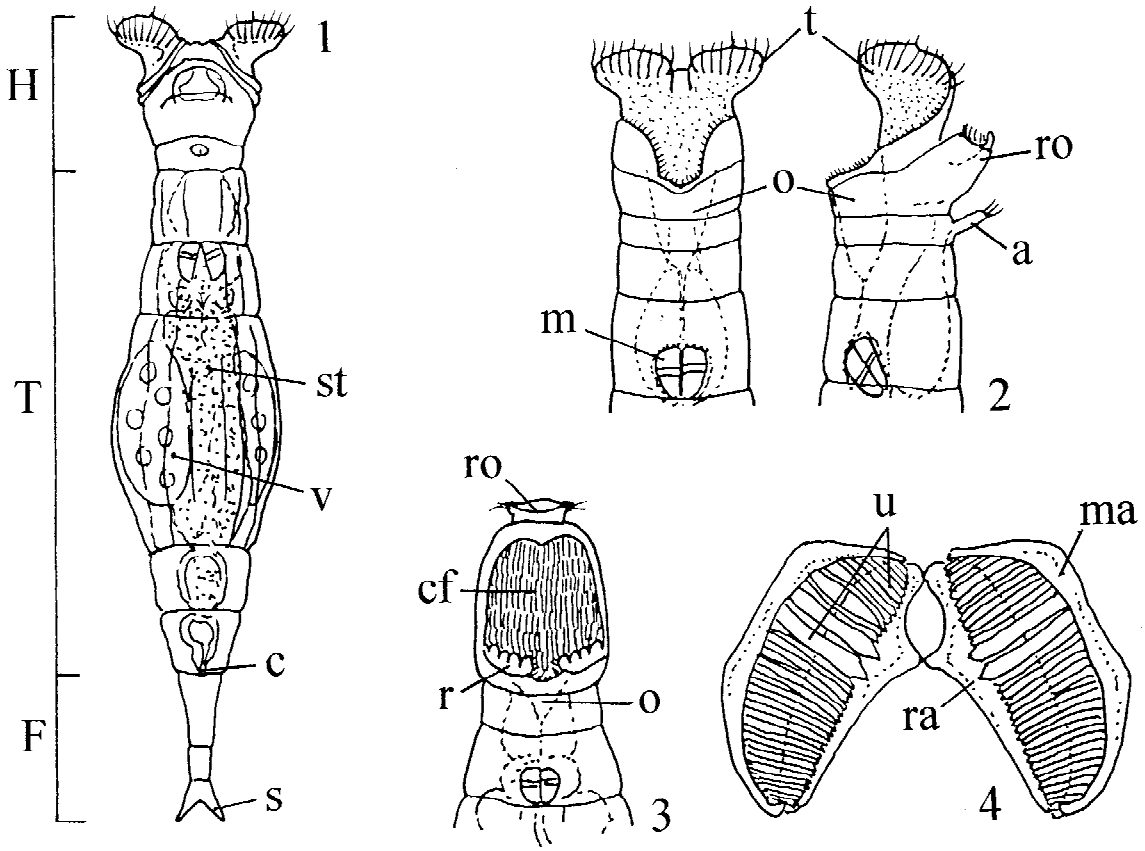
Morphology of bdelloids

The body of bdelloids consists of three main regions: head, trunk and foot (Figure 1). The head and the foot are telescopically retractable into the trunk and are superficially segmented. Cephalically, all bdelloids possess an apical rostrum, i.e. a retractable structure used for creeping, a dorsal antenna, and the ventral mouth opening which is surrounded by a ciliated structure (Figures 2, 3). This consists of a more or less complicated rotatory apparatus (corona), differently organized in the three orders (Melone & Ricci, 1995). The corona of the order Philodinida consists of two evident ciliated discs, the trochi (Figure 2), that can be more or less wide, and that are eventually covered with some membranous extensions (see *Scepanotrocha* and *Ceratotrocha*). The trochi are retractable into the head, and are used for filter-feeding and for swimming. The corona of the Adinetida consists of a ventral ciliated field, lined caudally by a stiff structure, the rake (Figure 3). The Philodinavida have an anomalous corona, that is reduced to a very small ciliated field as in *Philodinavus* and *Henoceros*, or organized into rudimentary trochi as in *Abrochtha*.

Eyes are present in some genera, visible as reddish spots, and are located either on the brain, i.e. dorsal to the mouth, or on the rostrum (see *Rotaria*).

The masticatory apparatus of rotifers, the mastax, occurs in the bdelloids at either of two different levels in the body. In Adinetida and Philodinida the mastax is far from the mouth (‘deep’) (Figures 2, 3) to which it is connected through a long oesophagus, so that the mastax is visible in the anterior part of the trunk. In Philodinavida the mastax is close to the mouth opening (‘superficial’) and can be partly protruded to grasp the food. The mastax is made of muscles and sclerotized articulated pieces, the trophi. The bdelloids have a unique trophi morphology, called ramate: a bilaterally symmetrical structure consisting of three paired parts, externally the manubria (not easily detected), the unci (with wide and narrow teeth) and medially the rami, which connect the two symmetrical halves (Melone et al., 1998) (Figure 4). Unlike that of monogononts, the shape of bdelloid trophi varies very little, and the number of teeth commonly reported refers to the strong, or major teeth on each uncus plate. The number of teeth is not diagnostically important, but can be helpful for identification when accompanied by other morphological traits. The only bdelloid group that can be identified from the trophi structure is the family Philodinavidae, for which other distinctive characters are also provided.

The trunk is the major region of bdelloid body (Figure 1). Most of it is occupied by digestive and reproductive apparatuses. The stomach is the most visible part of the digestive apparatus, but it can be transparent and empty if the rotifer is fasted or if it has recently recovered from dormancy. In well fed rotifers, three genera, *Habrotrocha*, *Otostephanos* and *Scepanotrocha*, can be discriminated by the appearance of the stomach content. In these species, the stomach is filled with distinct spherical pellets (food vacuoles) that are released as faeces. In the other genera, the faeces are produced as loose material, not packed into pellets, and a lumen is often visible inside the syncytial mass of the stomach. The reproductive apparatus is paired, and the glandular parts, the vitellaria, are evident lateral to the gut in mature, reproductive rotifers. The



Figures 1–4. Morphology of a bdelloid rotifer. Figure 1, general anatomy (dorsal view). Figure 2, head and anterior part of the trunk of a bdelloid (genus *Philodina*): ventral view (left) and lateral view (right). Figure 3, head and anterior part of the trunk of a bdelloid (genus *Adineta*): ventral view. Figure 4, ramate trophi. H, head; T, trunk; F, foot. a, antenna; c, cloaca; cf, ciliated field; m, mastax; ma, manubrium; o, oesophagus; r, rake; ra, ramus; ro, rostrum; s, spur; st, stomach; u, uncus; v, vitellarium.

vitellaria are syncytial masses with a fixed number of nuclei (commonly 8 in each one), that are readly visible because they contain a very large nucleolus. Embryos (in viviparous) or eggs (in oviparous species) can be seen in the trunk of adult animals. Oviparous bdelloids lay unsegmented eggs, in which the nucleus is usually distinguishable; thus the presence of segmented masses inside the rotifer body can suggest viviparity.

The foot is the body region posterior to the cloacal opening. The foot of bdelloids is frequently tapered and ends with two conical stiff appendages, the spurs (or a single one in *Henoceros*). Inside the foot are the pedal glands, whose ducts protrude as two, three or four ‘toes’, or form an undifferentiated adhesive surface. Length and shape of the foot vary between the genera, and these differences influence the overall appearance of bdelloids. *Rotaria* and *Embata* possess a long slender body because of their long foot, while *Habrotrocha* and *Bradyscela* have a short foot and appear stout. *Macrotrachela* and *Mniobia* appear to have an ‘extended belly’ because of their large trunk and short foot, and *Philodina* appears slim having a longish foot.

The foot is hardly visible when the animal feeds, because bdelloids retract the foot and ‘sit down’ to wheel the trochi. This is not true for *Adineta, Bradyscela, Henoceros* and *Philodinavus*, who lack trochi and commonly feed by scraping the biofilm. The toes are visible when the rotifer loops with leech-like movements, alternatively attaching to the substrate by rostrum and foot. The crawling of *Adineta* and *Bradyscela* differs from that of the other genera: these do not loop, but slide on the bottom either by using the ciliary beating of the ventral corona or by telescoping the foot for progression, and adhering with their ventral surface. Because of this behavior, the two genera can be easily recognized also under the stereomicroscope.

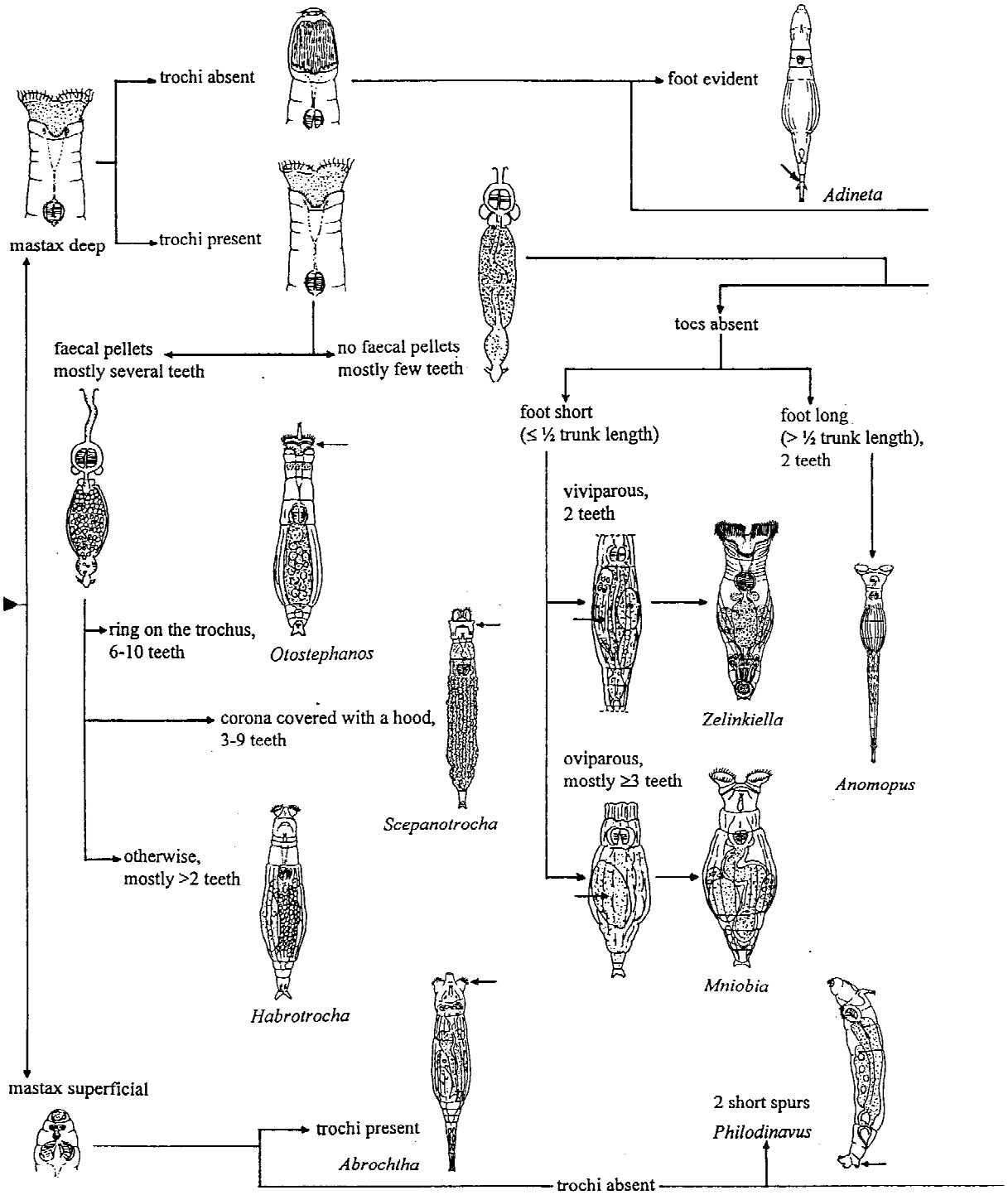


Figure 5a. Pictorial key to the bdelloid genera (redrawn after various sources).

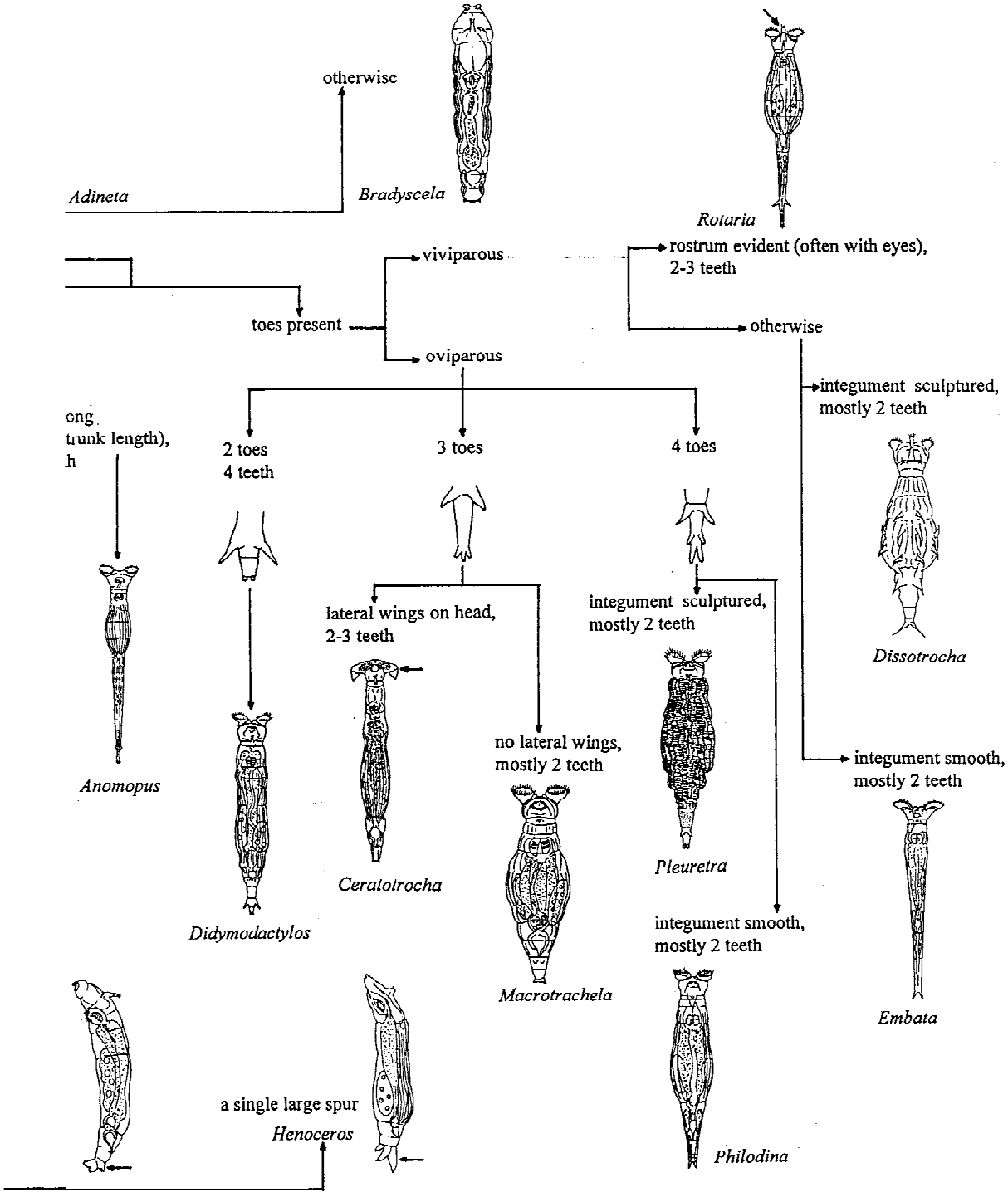


Figure 5b. Pictorial key to the bdelloid genera (redrawn after various sources).

Suggestions for observation

The extraction of bdelloids from the sample must be done with a pipette, taking care not to damage their tiny, soft bodies. The bdelloids must be quickly transferred to the slide to impede adhesion to the interior wall of the pipette, what would result in the loss of the specimen. When examining a bdelloid under a light microscope, it is advisable not to cover the slide with a coverslip, because the animals are easily disturbed and will contract into an indistinguishable ball. Bdelloids commonly move around in the water drop, thus showing their foot and eventually their toes. If suitable food is added to the water drop, they may wish to feed, or to swim if the attachment to the glass is lost. Good swimmers are *Rotaria, Embata* and *Philodina*. To examine the trophi structure, visible also when the animal is contracted, a coverslip must be added to the slide and a slight pressure exerted by subtracting water.

Bdelloid genera
Nineteen genera are listed for the class. The number of species attributed to each genus is based on Donner' s (1965) book, and no significant change has occurred since. A very short outline of each is given, describing their major features and the habitat they commonly occupy.

Abrochtha Bryce, 1910 (1 species)
About 500 μm in length. It has an underdeveloped corona, but is able to swim, and often does. The mastax is close to the mouth. The foot is easily visible. It lives in ephemeral rock pools and puddles.

Adineta Hudson and Gosse, 1886 (12 species)
The body is dorso-ventrally compressed, 200–700 μm, with a well-developed foot. It lacks trochi and has the ventral ciliated field with rake. It is unable to swim. The mastax is commonly very small in size and each uncus has 2 major teeth. Very active, its locomotion is easily noticed. Most species live in freshwater sediments, and are common also in soil, moss and lichens.

Anomopus Piovanelli, 1903 (2 species)
About 600 μm in length. The foot is very long and ends with short spurs and an adhesive disc. The trochi are wide and each uncus has 2 teeth. Both species are epizoic on freshwater crabs.

Bradyscela Bryce, 1910 (2 species)
About 200 μm long, it has a very short foot, with minute spurs, a number of papillae, and no toes. Unci plates with 2 teeth each. It lacks trochi, has a ciliated field ventrally and is unable to swim. The two species live in soil and moss.

Ceratotrocha Bryce, 1910 (4 species)
It is 150–350 μm long. The foot is quite short. The body appearance is rather slender. Each uncus has 2–3 teeth. The head morphology is peculiar because of membranous wings external to the trochi, visible when it feeds. All live in soil.

Didymodactylos Milne, 1916 (1 species)
Up to 500μm long. The foot is rather short and has two toes. The number of the uncus teeth is 4 or 5. It lives in moss and soil.

Dissotrocha Bryce, 1910 (7 species)
Viviparous, it is 300–500 μm long. Spines and stiff appendages are often present on the trunk. The foot is longish, commonly with long spurs. The trochi are seldom visible. Each uncus has 2 or 3 teeth. All species live in the sediments of lotic and lentic water, often among sphagnum.

Embata Bryce, 1910 (5 species)
Both oviparous and viviparous species, 250–650 μm long. The foot is very long and the trochi are wide, often extended. Two uncus teeth. Often epizoic on freshwater crustaceans.

Habrotrocha Bryce, 1910 (more than 100 species)
150–400 μm long, with a short foot and a quite long oesophagus. On each uncus there are usually several teeth (up to 10), of different width. Commonly it creeps and the trochi, seldom extended, are rather narrow. On careful inspection, the pellets inside the stomach are visible. Species of this genus occur in water sediments, soil and moss.

Henoceros Milne, 1916 (2 species)
Rotifers 130 to 200 μm long, with the strong rostrum always extended. The foot is peculiar, with a single spur and four strong toes, equally long. The trophi is close to the mouth. The trochi are absent, and the animals usually creep, but can swim by beating the cilia on the rostrum, and by using the single spur as a helm. Both species live attached to vegetation in running water.

Macrotrachela Milne, 1886 (about 100 species)
It is 200–500 μm long. Frequently each uncus has 2,

Key to identify bdelloid genera			
1		mastax deep	2
		mastax superficial	14
2	(1)	trochi present	3
		trochi absent	16
3	(2)	stomach with faecal pellets, usually several teeth	4
		stomach w/o faecal pellets, usually few teeth	5
4	(3)	ring present on the trochus, 6–10 teeth	*Otostephanos*
		trochi partially covered with a hood, 3-9 teeth	*Scepanotrocha*
		otherwise, mostly >2 teeth	*Habrotrocha*
5	(3)	toes absent	6
		toes present	8
6	(5)	foot short ($\leq {}^1/_2$ trunk length)	7
		foot long ($> {}^1/_2$ trunk length), 2 teeth	*Anomopus*
7	(6)	viviparous, 2 teeth	*Zelinkiella*
		oviparous, usually ≥ 3 teeth	*Mniobia*
8	(5)	viviparous	9
		oviparous	11
9	(8)	rostrum visible (often with eyes), 2–3 teeth	*Rotaria*
		otherwise	10
10	(9)	integument sculptured, 2–3 teeth	*Dissotrocha*
		integument smooth, 2 teeth	*Embata*
11	(8)	2 toes on the foot, 4 teeth	*Didymodactylos*
		3 toes on the foot	12
		4 toes on the foot	13
12	(11)	corona with lateral wings, 2-3 teeth	*Ceratotrocha*
		corona without lateral wings, usually 2 teeth	*Macrotrachela*
13	(11)	integument sculptured, usually 2 teeth	*Pleuretra*
		integument smooth, usually 2 teeth	*Philodina*
14	(1)	trochi present	*Abrochtha*
		trochi absent	15
15	(14)	2 short spurs	*Philodinavus*
		one spur and 4 big toes	*Henoceros*
16	(2)	foot and spurs evident	*Adineta*
		otherwise	*Bradyscela*

seldom 3, rarely up to 5 teeth. A few species have a sculptured cuticle on their trunk. The foot is short, with three short toes. The appearance of the body is solid. The trochi are wide, usually extended. The species occur in water sediments, moss, soil.

Mniobia Bryce, 1910 (about 50 species)

Similar to the previous genus in shape and size (300–600 μm), it can be distinguished by the number of teeth, seldom 2, mostly 3, up to 10. It is characterized by the absence of toes, but the adhesive disc is hardly visible. Most species live in soil and moss.

Otostephanos Milne, 1916 (9 species)

Between 200–500 μm long, it has a short foot. The stomach is filled with pellets. Distinguishable from *Habrotrocha* by the presence of a ring at the basis of the trochi pedicels. However, it seldom extends the trochi to feed. The number of teeth on each uncus is 6–10. Most species live in soil. Few occur in water, associated with aquatic plants.

Philodina Ehrenberg, 1830 (about 40 species)

100–800 μm long, the body is tapered, and the foot may be longish, with the four toes often visible. It commonly extends the trochi, that are wide and well

separated. Each uncus has usually 2 teeth, but 5 teeth also occur. Only one species is viviparous. The eyes are often present on the brain. Most species live in freshwater, but some dwell in moss and soil.

Philodinavus Harring, 1913 (1 species)
200–300 μm long. It lacks corona and has only a few cilia around the mouth. The trophi is close to the mouth. The rostrum is strong and the antenna is evident. The foot is short, with very small parallel spurs and four strong toes often visible. It lives among moss in creeks or waters rich in oxygen.

Pleuretra Bryce, 1910 (14 species)
From 200 to 400 μm long. The trunk integument is stiff, armour-like. The foot is short with four toes and the spurs are rather short. All species are oviparous, and usually have 2 teeth (only one with 5 teeth) on each uncus. The trochi are hardly ever extended. All species live in moss.

Rotaria Scopoli, 1777 (24 species)
400–1600 μm long, few species are 200 μm. All viviparous, they usually swim showing a wide corona. The rostrum, often with eyes, is commonly visible between the trochi. Each uncus has 2 teeth, rarely 3. The foot is long and slender, with long spurs and three toes that are long and easily visible. Most species live in water bodies, and few can be found in soil and moss.

Scepanotrocha Bryce, 1910 (9 species)
Mostly about 200 μm, the foot is short. It has pellets in the stomach. Each uncus has 3–9 teeth. The corona is covered with a dorsal membranous lobe that looks like a hood. This structure is visible when the animal feeds, which occurs infrequently. All species live in moss and soil.

Zelinkiella Harring, 1913 (1 species)
Its size is about 200 μm. It is viviparous. The foot is very short, with small spurs and adhesive disc. Each uncus has two teeth. It is marine and lives epizoic on *Synapta* (sea-cucumber), and occasionally on other marine invertebrates, like annelids and mollusks.

Conclusions

Although the key is limited to the discrimination among the genera only, we strongly hope that the use of English language and the indication of more accessible features of the animals will encourage scientists to dedicate more attention to this rotifer taxon that, till now, have received very scarce interest. Far from being exhaustive for the taxonomic knowledge, we hope that the easy pictorial key can be helpful for the people who need to know what animals are present in a given habitat, without particular requirements for list of species. The identification of bdelloid species is still left to the specialists.

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