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[REVIEW]

Beautiful Claws of a Tiny Water Bear: a Review and Proposal for Claw Configuration

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Tardigrades in the genus *Milnesium* have complex double claws on each leg, which consist of two slender primary branches and two basal secondary branches. The latter usually have two or three hooks. For more than a century, taxonomists have had great difficulties evaluating the claw variation in species identification in *Milnesium*. This review explains the complicated story and proposes a new notation system for *Milnesium* claw configuration with a discussion about anterior-posterior axes on tardigrade legs.

Key words: tardigrades, *Milnesium*, claw configuration, anterior-posterior axis, ontogeny

Tiny claws of a tardigrade tell us about a piece of the animal's developmental history, as well as the great struggles of taxonomists to identify their favorite tardigrades. At the end of the 19th century, there were only two species in genus *Milnesium*: *Milnesium tardigradum* Doyère, 1840 and *Milnesium alpigenum* Ehrenberg, 1853, distinguished from each other by the number of hooks on the claws. However, the latter was abolished at the beginning of the 20th century and the genus *Milnesium* remained monospecific for several decades. At the turn of the century, taxonomists started to separate the cosmopolitan species into many distinct species, and then, the claws also received more attention. In this review I observed the hooks on the *Milnesium* claws and traced the flow of taxonomic thought regarding their significance.

Now the story begins. Once upon a time a French zoologist met a tiny bear ...

Early history of *Milnesium* taxonomy

One of the tardigrades described by Doyère (1840) was *Milnesium tardigradum*, which was abundantly found in roof mosses in Saint-Maur near Paris (Fig. 1). This tardigrade was used as one of his experimental animals to study desiccation tolerance (Doyère, 1842; Broca, 1860). In terms of morphology, it showed the characteristic complex claws on its legs. Each leg terminus was bilobed, with each lobe having a slender terminal claw accompanied by a short, robust claw at the base of the terminal claw (Fig. 1B). In this article I adopt "primary branches" for the slender pair, and "secondary branches" for the basal pair, although these claw structures have been called by several different terms as follows: unghia terminale/unghia basale or asta principale/asta secondaria (Ramazzotti, 1945), terminal claw/secondary claw (Morgan and King, 1976), ramo principale/ramo secondario

(Maucci, 1986), main branch/complex basal claw + secondary branch (Pilato and Binda, 1991), main branch/basal claw (Bertolani and Grimaldi, 2000), main claw branches/secondary branches (Meyer and Hinton, 2010), and primary branches/secondary branches (Michalczyk et al., 2012a). Doyère's text describing the secondary branches clearly stated, "deux situés en dessous et en dedans, l'antérieur divisé en trois crochets fortement courbés, le postérieur en deux" (Doyère, 1840, p. 283); i.e., two [secondary branches] situated below and within, the *anterior* one divided into three strongly curved hooks and the *posterior* in two. Although its detailed illustration (Doyère, 1840, Pl. 13-5) showed only the claws on one of the fourth pair of legs (Fig. 1b), specimens from Saint-Maur that I collected in 2006 certainly exhibit the same combination of claw hooks, or points, as Doyère (1840) described: three points on the anterior secondary branch and two points on the posterior secondary branch, on all legs from the 1st (legs I) to the 4th pair of legs (legs IV) (Fig. 1C).

The second species of this genus, *M. alpigenum* Ehrenberg, 1853, from Monte Rosa, Alps, was distinguished by "pedum uncinis omnibus tridentatis", i.e., foot hooks all triple-pointed (Ehrenberg, 1853, p. 530; 1854, plate 35B). Contrary to the clear description in his text, interestingly, its figure (Ehrenberg, 1854, Pl. 35B-1) might indicate a small fourth projection (Fig. 2). After a half century, Richters (1902, 1904) disagreed with the validity of *M. alpigenum* because the specimens he examined from several localities in Europe, Spitzbergen, and Java always showed three hooks. He hesitatingly expressed his suspicion of the incomplete description by the fine observer, Doyère. He wondered if Doyère (1840) observed specimens with weakly developed 3rd point on the double-hooked claw and his illustrator perhaps suggested it (Fig. 1B). Therefore, Richters (1902, 1904) preferred the amendment of the description of the claw of *M. tardigradum* rather than the acceptance of *M. alpigenum*.

Murray (1905) also described triple-hooked claws of *M.*

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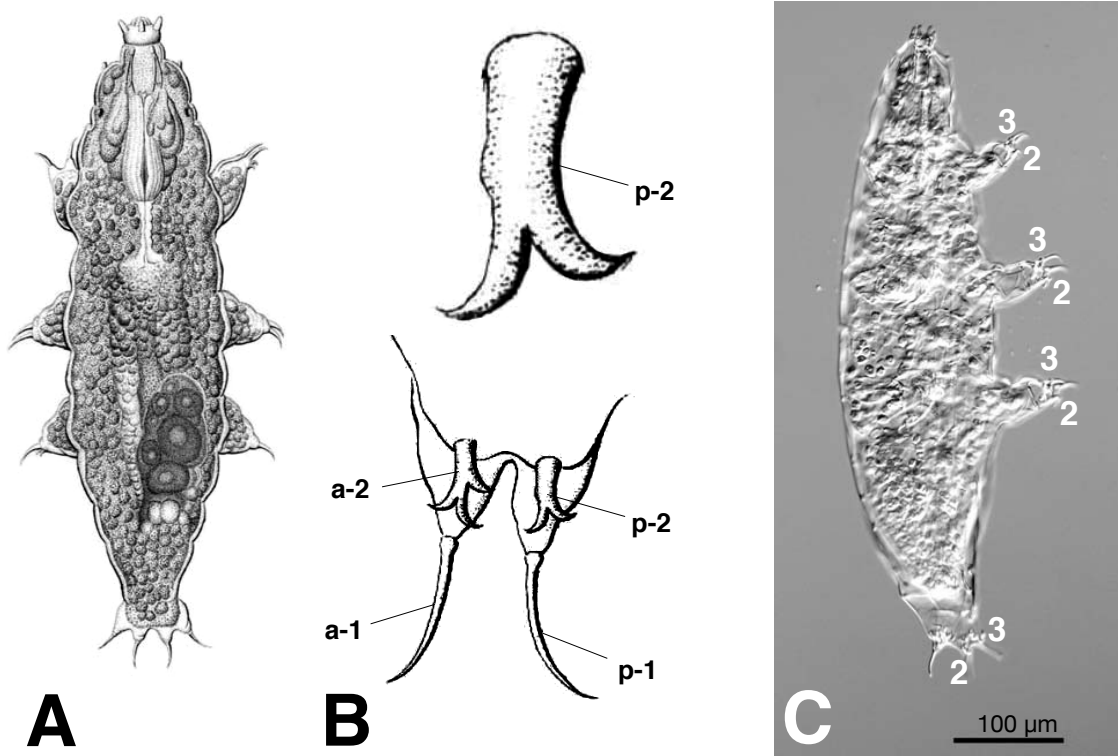


Fig. 1. *Milnesium tardigradum* Doyère, 1840. **(A)** Habitus and **(B)** right hind leg, ventral view, with an enlargement of the secondary branches (modified from Doyère, 1840, Pl. 13). a-1, anterior primary branch; a-2, anterior secondary branch; p-1, posterior primary branch; p-2, posterior secondary branch. **(C)** *Milnesium tardigradum* from type locality, St-Maur, indicating the number of hooks. Collected on 4 Feb 2006 by the author, mounted in glycerol.

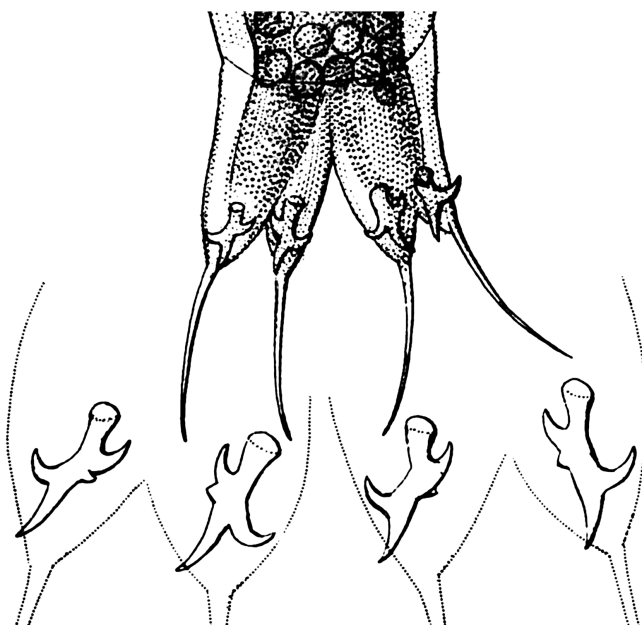


Fig. 2. *Milnesium alpigenum* Ehrenberg, 1853. Part of the original illustration (Ehrenberg, 1854, Pl. 35B-1), showing the 4th pair of legs and enlargement of the claw secondary branches.

tardigradum found from Loch Ness and stated, "according to Richters, *M. alpigenum* (Ehr.) is not a distinct species." Murray (1907) also said, "I agree with Professor RICHTERS in

uniting the two described species. [...] In some districts there appear to be constantly three points to these claws. In Scotland they are variable, [...] from one to three points, not only in different individuals from one district, but in one individual we may find all three conditions."

Later, Richters (1908) examined samples from the first German Expedition to Antarctica, the Gauss Expedition (1901–1903), and reported one specimen from Neu-Amsterdam [Île Amsterdam] that had exactly the same type of claws as Doyère described. He wrote, "genau der DOYÈRESCHEN Beschreibung: ein Steighaken mit zwei, der andre mit drei Krallen (2/3)." We can see here the first notation method to express *Milnesium* claw configuration, in which the number of points was shown in order of *posterior/ anterior*, or the opposite order compared to Doyère's description (*anterior/posterior*), despite his omission of showing definite anatomical positions. Then, another record on a Kerguelen specimen followed, described as a 3/4 claw on legs IV.

After these sub-Antarctic specimens, Richters (1908) continued further explanation about the issue of *Milnesium* 2/3 vs 3/3 claws. Although he repeated the previous statement that most specimens from numerous localities had 3/3 claws, he expressed his recent recognition of the existence of a certain European population with 2/3 claws in a communication with Botezat, the zoologist in Czernowitz University who had already published a short communication from Bukovina, where *Milnesium tardigradum* with 2/3 claws was found (Botezat, 1903). Richters received mosses sent from

Botezat and examined the specimens to confirm that the number of claw hooks was not constant even in Bukovina; from seven specimens obtained, four exhibited 3/3 claws (Richters, 1908).

Richters (1908) also pointed out another important observation that two specimens, one from Taunus and the other from Spitzbergen, had a very simple and robust claw on the first pair of legs. He also referred to a personal communication from Murray, who wrote about the same discovery in Scottish specimens, too. These observations are the first records of male *Milnesium* specimens in spite of no mention of sexual dimorphism, which was later described in detail by Thulin (1928) (Fig. 3). Richters (1908) wrote about several specimens with “zwei Krallen”; in this case he did not show the notation, but this could be the description of “2/2” claws. In spite of these findings, he still considered *M. tardigradum* was the only species in this genus, with individual variations of claw morphology.

Five years after the death of both Richters (1849–1914) and Murray (1865–1914), *Milnesium quadrifidum* Nederström, 1919 was described from Lapland as having 4-pointed secondary branches (Fig. 4), which was shown as 4/4 in Richters’s notation. Nevertheless, Marcus (1928, 1929), who agreed with Richters’s opinion that *M. alpigenum*

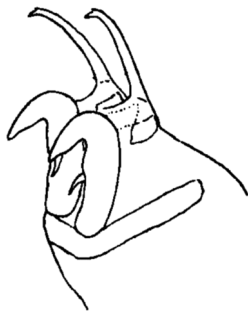


Fig. 3. The robust claws on leg I of male *Milnesium* (Fig. 14 in Thulin, 1928).



Fig. 4. *Milnesium quadrifidum* Nederström, 1919. Illustration in the original description, modified from Fig. 1 in Nederström, 1919.

was not valid, also did not consider *M. quadrifidum* as a distinct species but a form or a geographical race. In his later article, Marcus (1936) discarded his opinion of a geographical race, saying “Sie können nicht einmal als forma *quadrifida* bezeichnet werden und als geographische Rasse gelten”, citing Rahm (1928, probably an erroneous date) that recorded 3/4 and 4/4 *Milnesium* from Brazil as well as the Alps (Rahm, 1931).

Marcus (1936) explained Richters’s claw notation as follows: “Bei Milnesien mit 2/3 Haken trägt die Außenkralle 2, die Innenkralle 3 Haken”, i.e. in *Milnesium* with 2/3 hooks, the outer claw has two, the inner claw has three hooks. This was perhaps the first description in which the claw configuration of *Milnesium tardigradum* was defined as “outer/inner” in order, while Richters (1908) himself and Marcus (1928, 1929) wrote the formula vaguely, “ein [...] der andere [...]”, i.e., one and the other. Unfortunately, this definition by Marcus (1936) created confusion because it should not have been applied to the fourth pair of legs of *M. tardigradum*, in which claws IV have three points (hooks) on the outer (anterior) secondary branch, while two points on the inner (posterior) secondary branch (Fig. 1B).

Redescription of *Milnesium tardigradum*

The influence of the monographs (Marcus, 1929, 1936) was enormous and the monospecific genus *Milnesium* lasted until 1990, although several variations (Rahm, 1931; Ramazzotti, 1962) were recorded; furthermore, a new genus of Milnesiidae was established as *Limmenius porcellus* Horning, Schuster and Grigarick, 1978. Some specimens from Antarctica were shown to have 4–7 points on the secondary branches (Fig. 5), but yet they were identified as *M. tardigradum* (Sudzuki, 1964; Dastych, 1984) according to the cosmopolitan hypothesis. Finally, *Milnesium brachyungue* Binda and Pilato, 1990 sparked the new period of *Milnesium* taxonomy (Fig. 6). In 2005, as many as five new species were described at the same time (Tumanov, 2006), in which *M. tardigradum* from the European population was shown for comparison but illustrated there as having three points on all claws.

Continuing the revision of the genus, the redescription

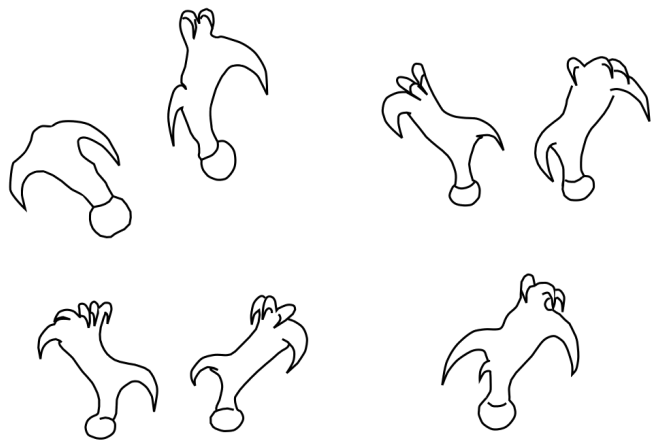


Fig. 5. Secondary branch examples of ‘*Milnesium tardigradum*’ from Evening Mountain, near Molodeznaya Station, Enderby Land, redrawn from Dastych, 1984, Fig. 33.

of *M. tardigradum* was published with establishment of its neotype (Michalczyk et al., 2012a, b). Contrary to the former opinions, this redescription placed particular emphasis on the claw configuration (CC), or the combination of the number of points on the secondary branches. The authors proposed a new notation system for CC shown as follows: [number of points on external claws I–III - number of points on internal claws I–III]-[number of points on anterior claws IV - number of points on posterior claws IV] (Michalczyk et al., 2012a, b). Based on this system, the claw configuration of *M. tardigradum* is expressed as [2-3]-[3-2], while *M. alpigenum* is [3-3]-[3-3]. This notation looks more complicated but was better than the former one because it avoids misunderstanding caused by applying the ambiguous terms, outer/inner or external/internal, to all claws I–IV. Instead, they replaced the terms external/internal on claws IV with the terms anterior/posterior. In spite of the complicated appearance of the new notation system, the unambiguity soon made its usage prevalent among tardigrade taxonomists.

Reconsideration of anterior-posterior axis on legs

Although this notation system has been the standard for the past decade, it still is problematic due to the terms *external/internal* for the movable legs. Imagine a schematic illustration of a hypothetical model of an animal with four pairs of legs, each of which forms a right angle with body axis (Fig. 7A), and it will clearly show the origin of the problem in the notation system. This hypothetical animal (Fig. 7A) has an obvious anterior-posterior distinction on all pairs of legs and there is no difference between legs I–III and legs IV. If legs I–III turn forward while legs IV turn backward, each pair of claws gets the outer/inner, or external/internal difference but it takes place in the opposite direction between claws I–III and IV (rotation model, Fig. 7B; Marchioro et al. (2013) also mentioned the rotation as the first scenario). Thus, the current notation system (Michalczyk et al., 2012), which is apparently derived from Richters-Marcus’s outer/inner system, was obliged to adopt different terms for claws IV in order to avoid confusion. As a result, the mixture of terms resulted in the complex notation as [2-3]-[3-2] for *M. tardigradum* (redescribed by Michalczyk et al., 2012a, b). However, if we go back to the description by Doyère (1840) and decide not to persist in using the terms *external/internal*, i.e. to adopt *anterior/posterior* on all legs, we will not encounter any confusion in describing the arrangement of claws I–IV.

Thus, I propose here a newer notation system. This is obviously originated from Doyère (1840), expressing claws in the *anterior/posterior* order on all pairs of legs. Now, the claws on leg I of *M. tardigradum* can be expressed as {3-2}^I in this system, in which the superscript Roman numerals show the leg position. Thus, the full notation of adult *M. tardigradum* could be shown as {3-2}^I{3-2}^{II}{3-2}^{III}{3-2}^{IV}, but in this case all legs have the same configuration and it can be expressed simply as {3-2}. The CC of *M. alpigenum* with three points on all legs is also simply expressed as {3-3}. If an individual has not only {3-2} but also {2-2} on Nth pair of

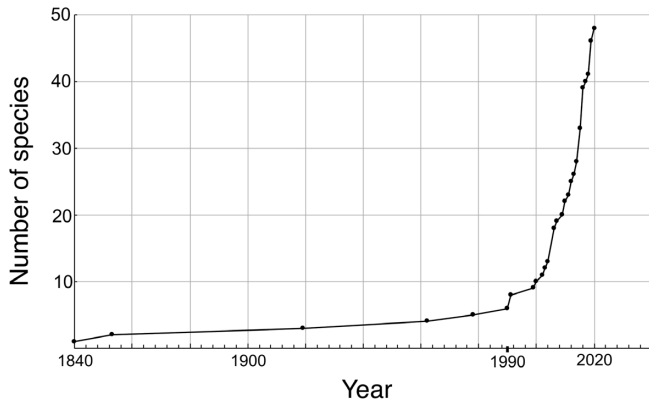


Fig. 6. Cumulative curve of new species descriptions in Family Milnesiidae from the establishment of genus *Milnesium* Doyère, 1840 to the present (summer of 2021).

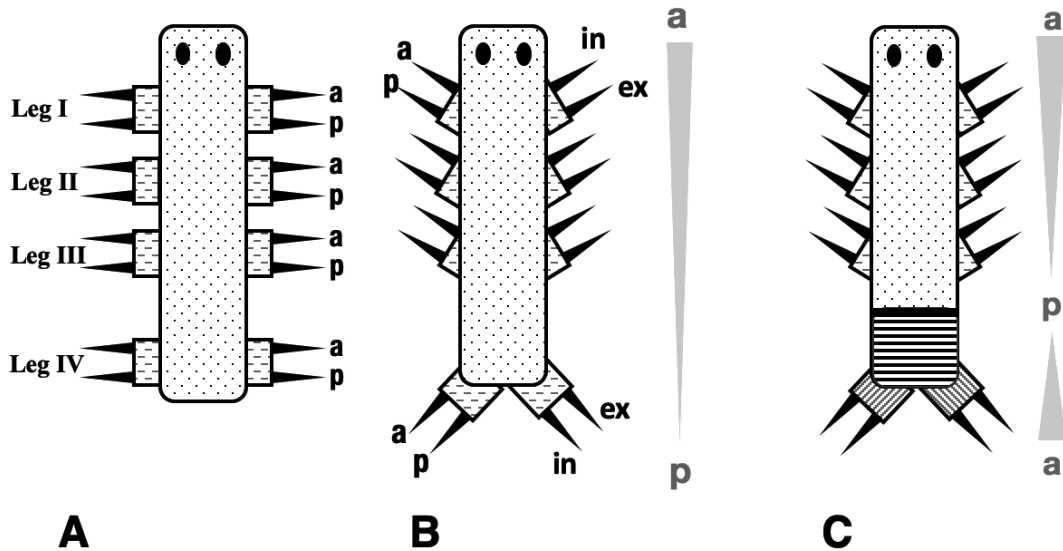


Fig. 7. Schematic models of the tardigrade body. (A) Hypothetical animal with four pairs of legs, each of which forms a right angle with body axis. (B) Rotation model in which front three pairs of legs (legs I–III) turn forward, while legs IV turn backward. (C) Reversal model, in which the last segment formed with the opposite A-P axis. a, anterior; p, posterior; in, internal; ex, external.

legs (claws N), {2-2}^N should be added after {3-2}. For example, the CC of *M. almatyense*, having {3-2} on claws I–III and {2-2} on claws IV, is {3-2}{2-2}^{IV}.

Advantages of the new notation are as follows: 1, it is easier to remember the definition than the current system; 2, its simple appearance shows the CC basic pattern, if any; and 3, it is easier to recognize which legs have the difference.

Table 1 exhibits all *Milnesium* species so far described with the adult CC, comparing the two notation systems. With the former notations, it looks as if there is a symmetrical relationship between claws I–III and IV, but it is, of course, an illusion formed by the notation system. The current form to describe claw configuration expresses the number of points in order as: [external-internal]^{I–III}-[anterior-posterior]^{IV}; in other words, it is [posterior-anterior]^{I–III}-[anterior-posterior]^{IV}. Therefore, it is obvious that the ‘reversed character’ is formed by the reversed notation system. The illusion is resolved by putting the current reversed system back to natural one as: {anterior-posterior}^{I–III}, i.e., the system that I proposed here, in which I use the curly bracket instead of the square bracket to distinguish it from the current one.

But is it possible for the terminal segment of a tardigrade to have the opposite morphogenesis that reverses the anterior-posterior axis (A-P axis) on legs IV (reversal model, Fig. 7C)? When the actual locomotive activity of a tardigrade is considered, legs IV apparently have quite different functions from legs I–III. Tardigrades walk mostly using legs I–III while legs IV appear to be dragged passively like an anchor, or the 4th legs are used to push the body. They climb mosses by reaching with the anterior legs while the hind legs hold onto the substrate (Schüttler and Greven, 2000/2001). Muscles of legs IV are indeed different from those of legs I–III due to the reduced movement of legs IV (Schmidt-Rhaesa and Kulesa, 2007; Marchioro et al., 2013). However, recent fine anatomical studies emphasized homology rather than differences between their morphology (Gross and Meyer, 2019). Thus, the leg rotation model (Fig. 7B) very likely explains the function and morphology of legs I–III and IV. On the other hand, the evolution of tardigrade bodies was also revealed by Hox gene expression analyses, which elucidated the loss of the intermediate region corresponding to the thorax and abdomen of arthropods by the lack of *Scr*, *Antp*, and *Ubx/abd-A* (Smith et al., 2016). These analyses suggested homology between arthropod mouth parts and tardigrade legs I–III (Smith and Goldstein, 2017), while legs IV on the last segment of tardigrades might have been formed under very different genetic circumstances.

Regardless, if A-P axis reversal occurred on the terminal segment, such animals must show some reversal in character states, e.g., {3-2}{2-3}^{IV}, which are different from the current state of *M. tardigradum*. Table 1 illustrates that no {3-2}{2-3}^{IV} type CC is actually extant. *Milnesium tetralamellatum* was once described as “[...] the complex basal claw + secondary branch on the hind legs has only 2 points on the outer claws and 3 points on inner claws” (Pilato and Binda, 1991), and later Morek et al. (2016) referred to this species as [2-3]-[2-3], i.e., {3-2}{2-3}^{IV}. However, recent re-examination revealed the mistake in the original description and *M. tetralamellatum* has turned out having [2-3]-[3-2] or {3-2} claws (Pilato, personal communication). Small indi-

viduals of *Milnesium barbadosense* showed [2-3]-[2-3]; however, the description was obscure with the application of inner/outer terms being unclear (Meyer and Hinton, 2012; Morek et al., 2016), but adult specimens from the type locality and all specimens from Mexico and Colombia expressed [3-3]-[3-3], or {3-3} (Meyer and Hinton, 2012; Moreno-Talamantes et al., 2019, 2020). The fact that there is an overwhelming majority of {3-2}, instead of {3-2}{2-3}^{IV}, suggests that the rotation model (Fig. 7B) is reasonable.

Developmental change in claw morphology

Many species in Heterotardigrada, especially Echiniscidae and marine arthrotardigrades, generally have two-clawed first-instar stage, which become four-clawed instars later (Murray, 1907; Bertolani et al., 1984; Hansen et al., 2016). “It is highly probable that all species of *Echiniscus* have only two claws in their earliest stage”, Murray (1907) wrote. As to several marine species of Styraconyxidae, each leg of the first instar has only two (internal) digits each with 3-pointed claws which consist of primary-, accessory- and secondary hook. In the 4-clawed juvenile stage, two internal claws are 3-pointed but external claws are simple without accessory hooks and secondary hooks, and adult specimens have 3-points on all claws (Kristensen and Higgins, 1984).

Some *Milnesium* species also show developmental changes in claw morphology. The modification of male claw I takes place at the second molt from juvenile to mature male (Rebecchi and Nelson, 1998). Moreover, many species have an ontogenetic CC change (Morek and Michalczyk, 2020; Morek et al., 2016, 2019a, 2020a; Sugiura et al., 2020). *Milnesium tardigradum* is one such species, in which a hatchling (1st instar) with {3-3} claws reduces the points to {3-2} after the first ecdysis (Morek et al., 2019a; Sugiura et al., 2020). This change is called an early negative CC change, notated as [3/2-3]-[3-3/2] (Morek et al., 2019a). By the proposed notation, it now can be shown as {3-3'²}, in which the superscript-lowercase Roman numerals express the timing of the change. In the same way, the early positive CC change of *M. pacificum*, in which 1st-instar {2-2} claws develop to be {3-2} after the first ecdysis (Sugiura et al., 2020), can be expressed as {2'³-2}. Late positive CC change takes place in *Milnesium variefidum* on claws I–III at the second ecdysis: from the 2nd (juvenile) into 3rd instar (adult), without any change on claw IV (Morek et al., 2016). This ontogenetic CC can be now expressed as {2ⁱⁱ-3-2}{2-2}^{IV}.

Aberrancy or heterochrony?

Some specimens of *Milnesium tardigradum* from Paris expressed {3-2}{3-3}^{IV}. As mentioned above, *M. tardigradum* has a negative CC change from {3-3} to {3-2} at the first ecdysis. Therefore, the adults with {3-2}{3-3}^{IV} could be explained as the result of incomplete CC change, keeping the vestige of the immature character on leg IV. Moreover, I have also encountered a specimen from Saint-Maur that has {4-2}^{II} on the right leg and {3-3}^{IV} (Fig. 8); it can now be notated as {3-2}{4-2}^{II}{3-3}^{IV}. Morek et al. (2019) reported the high frequency (24%) of aberrant claws, mostly additional spurs, in the Paris population of *M. tardigradum*. The slight swelling in Doyère’s illustration of the secondary branch (Fig. 1B) was probably only a small bump that is usu-

Table 1. Claw configuration in Milnesiidae.

Species name	Notation 2022		Notation 2012	Remark
	Adult	Ontogeny		
<i>Bergtrollus dzimbowski</i> Dastych, 2011	{3-3}		[3-3]-[3-3]	
<i>Limmenius porcellus</i> Horning et al., 1978	{3-3}		[3-3]-[3-3]	
<i>Milnesioides exsertum</i> Claxton, 1999	{3-3}		[3-3]-[3-3]	
<i>Milnesium alabamae</i> Wallendorf & Miller, 2009	{3-3}		[3-3]-[3-3]	
<i>M. alpigenum</i> Ehrenberg, 1853	{3-3}	{3-3}	[3-3]-[3-3]	
<i>M. antarcticum</i> Tmanov, 2006	{3-3}		[3-3]-[3-3]	
<i>M. argentinum</i> Roszkowska et al., 2015	{3-3}		[3-3]-[3-3]	
<i>M. asiaticum</i> Tumanov, 2006	{3-3}		[3-3]-[3-3]	
<i>M. barbadosense</i> Meyer & Hinton, 2012	{3-3}	{3-2 ⁱⁱ 3} ?	[3-3]-[3-3]	See text
<i>M. beatae</i> Roszkowska et al., 2015	{3-3}		[3-3]-[3-3]	
<i>M. bohleberi</i> Bartels et al., 2014	{3-3}		[3-3]-[3-3]	
<i>M. brachyungue</i> Binda & Pilato, 1990	{3-3}		[3-3]-[3-3]	
<i>M. burgessi</i> Schlabach et al., 2018	{3-3}	{3-3}	[3-3]-[3-3]	
<i>M. dornensis</i> Ciobanu et al., 2015	{3-3}	{3-3}	[3-3]-[3-3]	
<i>M. eurystomum</i> Maucci, 1991	{3-3}		[3-3]-[3-3]	
<i>M. fridae</i> Moreno-Talamantes et al., 2020	{3-3}		[3-3]-[3-3]	
<i>M. granulatum</i> Ramazzotti, 1962	{3-3}		[3-3]-[3-3]	
<i>M. cf. granulatum</i> (see Jacson & Meyer, 2019)	{3-3}	{3-3}	[3-3]-[3-3]	
<i>M. inceptum</i> Morek et al., 2019	{3-3}	{3-3}	[3-3]-[3-3]	
<i>M. longiungue</i> Tumanov, 2006	{3-3}		[3-3]-[3-3]	
<i>M. matheusi</i> Kaczmarek et al., 2019	{3-3}		[3-3]-[3-3]	
<i>M. minutum</i> Pilato & Lisi, 2016	{3-3}		[3-3]-[3-3]	small (ca. 300 μm)*
<i>M. pentapapillatum</i> Morek et al., 2020	{3-3}		[3-3]-[3-3]	
<i>M. sandrae</i> Pilato & Lisi, 2016	{3-3}		[3-3]-[3-3]	
<i>M. shilohae</i> Meyer, 2015	{3-3}		[3-3]-[3-3]	
<i>M. swansoni</i> Young et al., 2016	{3-3}		[3-3]-[3-3]	
<i>M. tumanovi</i> Pilato et al., 2016	{3-3}		[3-3]-[3-3]	
<i>M. validum</i> Pilato et al., 2017	{3-3}		[3-3]-[3-3]	
<i>M. zsalakoe</i> Meyer & Hinton, 2010	{3-3}		[3-3]-[3-3]	
<i>M. pseudotardigradum</i> Surmacz et al., 2019	{3-2}/{3-3}	{3-3 ⁱ 2 ⁱⁱⁱ 3}	[3/2-3]-[3-3/2]	See text
<i>M. beasleyi</i> Kaczmarek et al., 2012	{3-2}		[2-3]-[3-2]	
<i>M. cassandrae</i> Moreno-Talamantes et al., 2019	{3-2}	{2 ⁱ 3-2}	[2-2/3]-[2/3-2]	
<i>M. krzysztofi</i> Kaczmarek & Michalczyk, 2007	{3-2}		[2-3]-[3-2]	
<i>M. lagniappe</i> Meyer et al., 2013	{3-2}		[2-3]-[3-2]	
<i>M. pacificum</i> Sugiura et al., 2020	{3-2}	{2 ⁱ 3-2}	[2-2/3]-[2/3-2]	
<i>M. reductum</i> Tumanov, 2006	{3-2}	{2 ⁱ 3-2}	[2-2/3]-[2/3-2]	Morek et al., 2020a
<i>M. reticulatum</i> Pilato et al., 2002	{3-2}		[2-3]-[3-2]	
<i>M. tardigradum</i> Doyère, 1840	{3-2}	{3-3 ⁱ 2}	[3/2-3]-[3-3/2]	Morek et al., 2019; Sugiura et al., 2020
<i>M. tetralamellatum</i> Pilato & Binda, 1991	{3-2}	{2 ⁱ 3-2}	[2-3]-[3-2]	See text
<i>M. vorax</i> Pilato et al., 2016	{3-2}		[2-3]-[3-2]	
<i>M. berladnicorum</i> Ciobanu et al., 2014	{3-2}{2-2} ^{IV}	{2 ⁱ 3-2}{2-2} ^{IV}	[2-2/3]-[2-2]	
<i>M. variefidum</i> Morek et al., 2016	{3-2}{2-2} ^{IV}	{2 ⁱⁱ 3-2}{2-2} ^{IV}	[2-2/3]-[2-2]	
<i>M. almatyense</i> Tumanov, 2006	{3-2}{2-2} ^{IV}	{2 ⁱ 3-2}{2-2} ^{IV}	[2-2/3]-[2-2]	Morek et al., 2020a
<i>M. jacobi</i> Meyer & Hinton, 2010	{3-2}{3-3} ^{IV}		[2-3]-[3-3]	
<i>M. katarzynae</i> Kaczmarek et al., 2004	{2-2}		[2-2]-[2-2]	small (285–295 μm)*
<i>M. kogui</i> Londoño et al., 2015	{2-2}		[2-2]-[2-2]	small (312–336 μm)*
<i>M. dujiangensis</i> ** Yang, 2003	{2-2}{1-2} ^{IV}		[2-2]-[1-2]	small (314–363 μm)*
<i>M. wrightae</i> Kaczmarek et al., 2019	{3-3}{4-4} ^{IV}		[3-3]-[4-4]	
<i>M. quadrifidum</i> Nederström, 1919	{4-4}		[4-4]-[4-4]	
<i>M. swolenskyi</i> Bertolani & Grimaldi, 2000	?		?	amber fossil 90 mya

* Possibly described based on immature individuals.

** *nomen dubium*

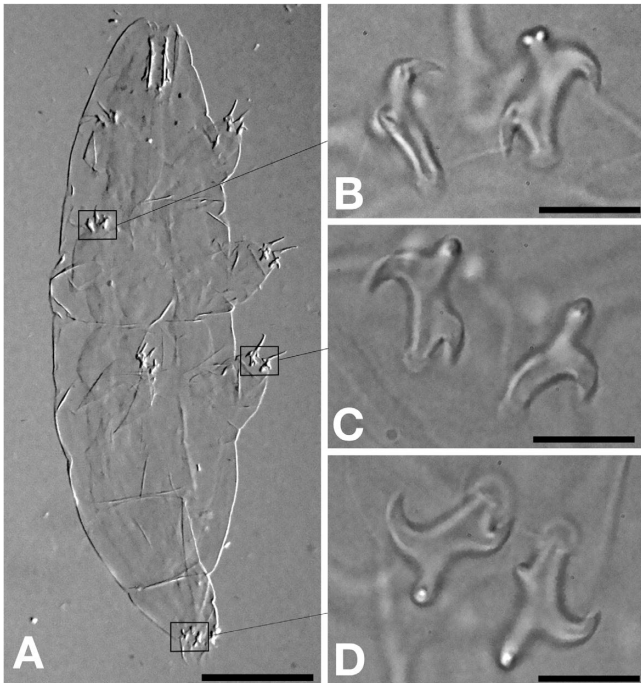


Fig. 8. A specimen of *Milnesium* cf. *tardigradum* from Saint-Maur, collected on 4 Feb 2006 by the author, mounted in Hoyer's solution. **(A)** Habitus. Scale, 100 μ m. **(B)** Secondary branches of right claws II showing {4-2} points. **(C)** Claws III with ordinary {3-2} points. **(D)** Claws IV with {3-3} points. Scales for **(B)–(D)**, 10 μ m.

ally observed, but Richters's suspicion about it as the third points has become a reality!

Irregular claws were also observed in *M. pacificum* (Sugiura et al., 2020). Such variation might not be abnormal but the result of developmental flexibility, instability, or a tendency toward heterochrony in this species. It is possible that the CC of {3-2}-{2-2}^{IV} (*M. almatyense*, *M. berladnicorum* and *M. variefidum*) might be formed by the incomplete positive CC change from {2-2} to {3-2}, absent on leg IV. On the contrary, *M. jacobi* with {3-2}{3-3}^{IV} might be formed by omitting the negative change on leg IV. Surmacz et al. (2019) reported a more complicated CC change in *M. pseudotardigradum* from Iceland, having a {3-3} hatchling, {3-2} 2nd-instar (juvenile) and 3rd-instar (young adult), and next-instar adults again having spurs on several posterior branches, and even complete {3-3} adults were sometimes found (Surmacz et al., 2019). Therefore, *Milnesium* spp. generally have great flexibility between {3-3} and {3-2}, and some individuals, or some species, e.g. *M. wrightae*, even have an extra fourth point or more. Specimens from an ice-free area along Lützw-Holm Bay, Antarctica, will be described soon, which have claws with a similar appearance to specimens from Enderby Land (Fig. 5).

The positive CC change of *Milnesium* increases the number of points, while the negative CC change decreases them. This developmental character is different from heterotardigrade ontogeny doubling the number of claws. Among the known 15 *Milnesium* species with {3-2} claws, at least 60% of the species show positive or negative change (Table 1). "All species with an adult CC different from [3-3]-[3-3] and for which ontogenetic tracking was conducted, a CC change

was observed", Surmacz et al. (2020) wrote. Perhaps most species with {3-2} claws have either positive or negative CC change. There is a great difference of ontogenetic background between the species with a positive CC change and those with a negative change, in spite of the same {3-2} CC in the adult. On the other hand, it has not yet been elucidated how many examples of CC change will be expected from 28 species including three other monospecific genera in Milnesiidae with {3-3}. At least four species proved to have a constant {3-3} (Table 1). However, the description of *M. barbadosense* suggested that it has a {3-2¹3} change (Meyer and Hinton, 2012), and an undescribed *Milnesium* PH.014 in Morek and Michalczyk (2020) has some ontogenetic variability (Morek et al., 2020b). Perhaps some other {3-3} members might have a similar CC change or fluctuation as in *M. pseudotardigradum*. Nevertheless, we can consider that {3-3} may be basic and {3-2} and other CC are the derived state based on our current knowledge (Table 1). Detailed phylogenetic analysis has also suggested the ancestral state as {3-3} CC, in spite of its tendency to fluctuate and the lack of support from fossils (Morek and Michalczyk, 2020).

In conclusion, *Milnesium* CC is generally quite flexible between {3-2} and {3-3}, and sometimes more spurs or hooks on the secondary branches are present, and claw morphology still has very important value in taxonomy as well as in developmental biology. *Milnesium* claws are beautiful, and beautiful things have thorns. We can now understand how Richters and Murray struggled with the claws of tiny bears. And yet, we still struggle with their claws even after taxonomic tools have changed dramatically.

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