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Morphological ontogeny of *Amerus polonicus* (Acari: Oribatida: Ameridae), and comments on *Amerus* Berlese

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Abstract

The morphological ontogeny of *Amerus polonicus* Kulczynski, 1902 is described and illustrated, based on the individuals from laboratory culture. The juveniles of this species are unpigmented, with punctated integument. The larva has 11 pairs of gastronotal setae, including h_2 , the nymphs have 12 pairs. The nymphs are quadridentate and eupheredermous, i.e. they carry the exuvial scalps of previous instars on the gastronotum, using mainly the inward curving setae *la* and *lm* to protect the exuvial scalps against loss. In all juveniles, seta *d* is present on all femora, genua and tibiae, and setae *d* on femur I and *ft* on tarsus I are very long. All tarsi of juveniles are beaded, with long, thin distal parts. In the adult, seta *d* is lost on genua and tibiae I and II, but is retained on genua and tibiae III and IV.

Keywords: oribatid mites, laboratory culture, juveniles, development, leg setation

Introduction

The morphology of *Amerus* Berlese, 1896 is relatively well known due to the paper of Avanzati *et al.* (2003), who redescribed the adult of *A. troisi* (Berlese 1883), the type species of this genus, and described a new species, *A. cuspidatus* Avanzati *et al.*, 2003. According to Subías (2020), *A. polonicus* Kulczynski, 1902 is the third species of *Amerus sensu stricto*, whereas *Amerus lundbladi* Willmann, 1939 is included in subgenus *Neamerus* Willmann, 1939. *Amerus* belongs to Ameridae, which is rather poor in species. This family includes also six other genera (Subías 2004), of which *Hymenobelba* Balogh, 1962 has six nominal species, *Caenosamerus* Higgins & Woolley, 1969 has three species, and *Andesamerus* Hammer, 1962, *Haplamerus* J. & P. Balogh, 1992, *Ctenamerus* J. & P. Balogh, 1992 and *Pteramerus* Balogh, 1961 have two species each (Subías 2020).

The systematic status of *A. polonicus* is debated in literature. Some authors considered it as a synonym of *A. troisi* (Sellnick 1960; Bulanova-Zachvatkina 1975), but in figure of *A. troisi* by both authors seta *lp*, h_2 and h_3 are shorter and more similar to *A. polonicus* than to *A. troisi* adding to the confusion. Other authors treated *A. polonicus* as a separate species (Pérez-Íñigo 1976; Olszanowski *et al.* 1996; Mahunka & Mahunka-Papp 2000; Siepel *et al.* 2009; Miko 2016; Murvanidze & Mumladze 2016). Therefore, in this paper we aim to provide better diagnostic characters of *A. polonicus* to clarify its systematic status.

According to the catalogue of juvenile oribatid mites by Norton and Ermilov (2014) and further literature, the morphological ontogeny of *A. polonicus* or any species of *Amerus* is unknown. Grandjean (1965) provided data on hypertrichy of aggenital setae in the nymphs and adult of this genus, but it was not attributed to a specific species.

The aim of this paper is to describe and illustrate the morphological ontogeny of *A. polonicus*, and compare the morphology of the adult with congeners. As the description of the adult by Kulczyński (1902) is old and general, we also redescribe it.

Material and methods

The adults and juveniles of *A. polonicus* used in this study were collected on 15 November 2017 by J. Kowalski under oak (*Quercus robur* L.), at the edge of an old oak forest (Figure 1) near Sierpe (Mazovia Province, Poland, 52°48'32"N, 19°39'35"E). This forest was dominated by oak trees, 85–105 years old, with addition of hornbeam (*Carpinus betulus* L.), black alder (*Alnus glutinosa* Gaertn.), lime (*Tilia cordata* Mill.) birch (*Betula pendula* Roth), bird cherry (*Padus avium* Mill.) and Scots pine (*Pinus sylvestris* L.). Ten samples of volume 500 cm³ each were taken from the upper soil horizon (5 cm deep), including litter and were extracted in modified Tullgren funnels during 10 days into small chambers with water; 12 adults and two deutonymphs of *A. polonicus* were extracted. Five adults were then cultured successively in a small plastic box (7 cm² x 5 cm high), with the bottom filled with plaster of Paris and charcoal mixture and covered by a perforated lid. These mites were kept in controlled climate conditions (temperature 25°C, 90% air humidity), and fed with oak litter and fresh champignon mushrooms. To avoid intensive overgrowing of food by mycelium, adults of *A. polonicus* were cultured with *Hafenrefferia gilvipes* (C.L. Koch 1839), which feed on mycelium (the juveniles of this species showed extraordinary high feeding activity). Moreover, since the nymphs of *H. gilvipes* also bear exuvial scalps of previous instars on the gastronotum with many long setae (Seniczak *et al.* 2018), they easily rip up the mycelium, further aiding with mycelium growth control. While *A. polonicus* was cultured, we selected five larvae, five protonymphs, and four deutonymphs for the morphological study, and five tritonymphs left, from which one transformed in the adult. Four tritonymphs, six adults from culture, and two deutonymphs and seven adults from the extracted samples were also used in this morphological study. The time of development of *A. polonicus* was established on the base of the successive first stages that appeared in the culture, but fertility of the adults was difficult to determine because it concerned three females, cultured together with two males (determined in lactic acid after this experiment).

We measured the total length of mites (tip of rostrum to posterior edge of notogaster) in lateral aspect and width (widest part of notogaster) of mites in dorsal aspect, and length of anal and genital openings and setae perpendicularly to their size in µm. The illustrations of *A. polonicus* are limited to the body regions that show substantial differences between instars, including the dorsal and lateral aspect and some leg segments of the larva, tritonymph and adult, ventral regions of all instars, and the palp and chelicera of the adult. Illustrations were prepared from individuals mounted temporarily on slides in lactic acid, using the open-mount technique (Grandjean 1949). In the text and figures, we used the following abbreviations: rostral (*ro*), lamellar (*le*), interlamellar (*in*) and exobothridial (*ex*) setae, bothridium (*bo*), bothridial seta (*bs*), notogastral or gastronotal setae (*c*-, *d*-, *l*-, *h*-, *p*-series), cupules or lyrifissures (*ia*, *im*, *ip*, *ih*, *ips*, *iad*), humeral apophysis (*ha*), scalps of larva (L), protonymph (Pn) and deutonymph (Dn), subcapitular setae (*a*, *m*, *h*), cheliceral setae (*cha*, *chb*), palp setae (*sup*, *inf*, *l*, *d*, *vt*, *cm*, *acm*, *lt*, *ul*, *su*) and solenidion ω, pedotecta (*Pd*), discidium (*Dis*), epimeral setae (*1a*–*c*, *2a*, *3a*–*c*, *4a*–*c*), adanal and anal setae (*ad*-, *an*-series), aggenital seta (*ag*), leg solenidia (σ, φ, ω), famulus (ε) and setae (*bv*, *ev*, *d*, *l*, *ft*, *tc*, *it*, *p*, *u*, *a*, *s*, *pv*, *pl*, *v*). Terminology used follows that of Grandjean (1949, 1953, 1965) and Norton and Behan-Pelletier (2009). The species nomenclature follows Subías (2004, updated 2020).



FIGURE 1. General view of the edge of old oak forest, in which *Amerus polonicus* was found (enlarged view of litter in bottom-right corner).

For scanning electron microscopy (SEM), the mites were air-dried and coated with Au/Pd in a Polaron SC502, sputter coater and placed on Al-stubs with double-sticky carbontape. Observations and micrographs were made with a ZEISS Supra 55VP scanning electron microscope. The photos of the adult of *A. polonicus* were prepared using microscope Leica DM3000 and camera Leica DFC420.

***Amerus polonicus* Kulczynski, 1902**

Amerus troisi (Berlese 1883) *sensu*: Sellnick 1960; Bulanova-Zachvatkina 1975.

Amerus polonicus: Olszanowski *et al.* 1996; Mahunka and Mahunka-Papp 2000; Siepel *et al.* 2009, Miko 2016; Murvanidze and Mumladze 2016.

Diagnosis

Adults large (923–1021), oval, dark-brown, integument smooth, but some parts of body covered with thicker granular cerotegument. Rostrum with two deep incisions. Bothridial seta setiform, dorsosejugal furrow absent. Ten pairs of notogastral setae present, *lm* longest, *p*₃ shortest, all curved; setae *lp*, *h*₂ and *h*₃ clearly shorter than other dorsal setae. Epimeral seta *lb* longer than other ventral setae, genital setae (6 pairs), aggenital setae (12–13 pairs), adanal setae (3 pairs) and anal setae (2–3 pairs) short. Seta *l'* from palpal tibia absent, formula of palp setae (and solenidion): 0-2-1-2-9(1). Femora I and II with 5–6 setae. Seta *d* from all genua and tibiae I and II absent, but on genua and tibiae III and IV present.

Juveniles unpigmented, integument punctated, some parts of body covered with thicker granular cerotegument. In all juveniles, prodorsal setae *ro* and *le* of medium size, and *ex* short, whereas seta *in* long in larva and short in nymphs. In all juveniles, bothridium rounded, protruding above surface, and

bothridial seta setiform. Most gastrontal setae long and curved, with thin apical ends. Larva with 11 pairs of gastrontal setae, including h_2 , nymphs with 12 pairs. Nymphs quadridentate and eupherodermous and carry scalps of previous instars, using mainly curved inwards setae la and lm that protect exuvial scalps against loss. In all juveniles, seta d on all femora genua, tibiae present, and seta d on femur I and ft'' on tarsus I very long. In juveniles, all tarsi beaded, with long, thin distal parts.

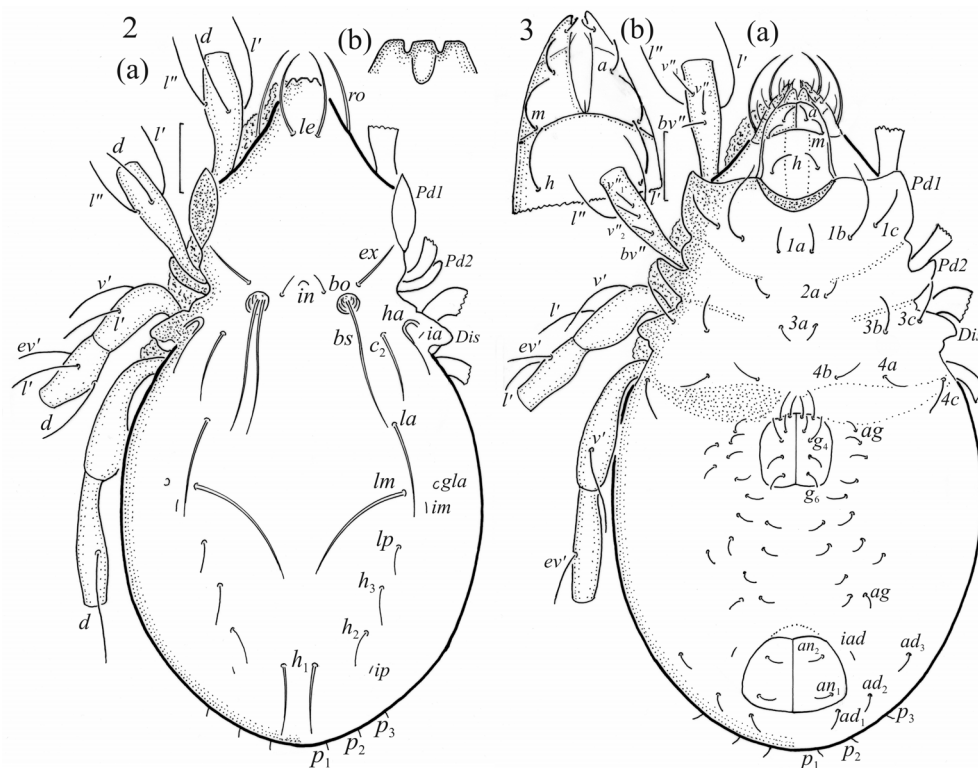
Redescription of morphology of adult

Measurements. Mean length (and range) of females – 966.7 ± 2.16 (923–1021, $N=6$) and males – 967.3 ± 1.38 (range 934–1004, $N=7$); mean width of females – 555.8 ± 7.06 (520–637) and males – 539.5 ± 2.31 (527–566).

Integument. Dark-brown, main body smooth, some parts of body covered with thick granular cerotegument, especially in dorso-sejugal depression, and on basal parts of legs.

Prodorsum. Subtriangular, rostrum with two deep depressions (Figs. 2a, 2b). Seta ro slightly shorter (140–144) than le (147–150), and inserted laterally (Figs. 2a, 3a, 4a), pair le inserted dorsally between pair ro . Seta in short (33), seta ex longer (105–107), all prodorsal setae smooth and slightly curved. Bothridium rounded, protruding above surface, bothridial seta long (193–197), setiform, barbed, distally thin and curved, in some individuals two bothridial setae present in one bothridium. Dorsal sejugal suture absent.

Notogaster. Anterior part of notogaster with deep depression, pair of humeral apophysis (ha) and 10 pairs of setae (including c_2) of different length (Figs. 2a, 4a, 5a, 5c, 5d, 6a, 6b, Table 1); setae lp , h_2 and h_3 clearly shorter than other dorsal setae. From dorsal setae, lm longest (192–195) and lp shortest (54–57), setae of p -series short (33–41). Lyrifissures ia , im , ip , ih , ips and opisthonotal gland opening in normal positions.



FIGURES 2–3. *Amerus polonicus*, male, legs partially drawn, scale bars 100 μm . 2. (a) Dorsal aspect, (b) rostrum (enlarged). 3. (a) Ventral aspect, (b) part of hypostome (enlarged).

TABLE 1. Measurements of some morphological characters of juvenile stages of *Amerus polonicus* (mean measurements of 4–6 juveniles and 10 adults in μm); Nd—not developed.

Morphological characters	Larva	Protonymph	Deutonymph	Tritonymph	Adult
Body length	390	475	722	864	967
Body width	218	238	371	475	552
Length of prodorsum	145	207	211	281	358 ¹
Length of: seta <i>ro</i>	56	69	77	105	142
seta <i>le</i>	99	102	107	141	148
seta <i>in</i>	69	2	3	3	33
seta <i>bs</i>	165	211	244	248	195
seta <i>c1</i>	112	116	141	168	Lost
seta <i>c2</i>	23	25	32	45	115
seta <i>c3</i>	241	291	363	422	Lost
seta <i>da</i>	231	Lost	Lost	Lost	Lost
seta <i>dp</i>	254	Lost	Lost	Lost	Lost
seta <i>la</i>	257	248	357	520	148
seta <i>lp</i>	165	215	429	468	56
seta <i>h1</i>	106	336	390	585	112
seta <i>h2</i>	48	56	183	330	59
seta <i>h3</i>	Nd	182	278	455	66
seta <i>p1</i>	Nd	43	78	165	40
seta <i>p3</i>	Nd	45	72	115	34
genital opening	Nd	41	80	118	99
anal opening	91	95	152	158	139

¹from rostrum to anterior edge of humeral apophysis.

Gnathosoma. Subcapitular seta *h* slightly longer (59) than *m* (56) and *a* (46), all smooth (Figs. 3a, 3b). Chelicera relatively thin (205 x 66) chelate, seta *cha* longer (25) than *chb* (19), both smooth (Fig. 4b). Palp relatively short (154–157), palpal setae on femur, genu and tibia relatively long and barbed, *l'* on tibia absent, palpal seta *cm* relatively long, other setae shorter, all smooth (Fig. 4c). Solenidion ω and eupathidia relatively long, seta *acm* short, separated from solenidion. Formula of palp setae [trochanter to tarsus (+ solenidion ω)]: 0-2-1-2-9(1). Axillary saccule of subcapitulum short (9).

Ventral and lateral regions. Epimeral seta *1b* long (127–130), *1c*, *3c* and *4c* shorter (72–105), other setae short (35–50, Fig. 3a). Six pairs of genital setae, g_{1-2} in anterior position; all short (28–50) and smooth. Aggenital setae (12–13 pairs), adanal setae (three pairs) and anal setae (2–3 pairs) short (32–37) and smooth. Among 13 adults investigated, two individuals with three pairs of anal setae, one individual with two pairs and one unpaired seta, and other individuals with two pairs. Lyrifissure *iad* short, located lateral to anterior part of anal plates. Seta ad_3 inserted laterally, relatively distant from *iad*, seta ad_1 inserted posterior to anal plate, and seta ad_2 between setae ad_1 and ad_3 . Pedotectum I large, oval (122 x 99), pedotectum II smaller (56 x 77), discidium well-formed, distally rounded (Figs 3a, 4a).

Legs. Trochanters III and IV and all femora oval in cross-section, dorsal parts with porose areas. Apical ends of trochanters III and IV with clear collars. All leg setae relatively long and finely barbed (Figs. 4a, 5a–c, 5e, 6a, 6b, 7), except for smooth ventral setae on all femora and setal pairs *p* on all

tarsi. All tarsi relatively thin and long, especially tarsus IV. Seta *d* on genua and tibiae I and II absent, but on genua and tibiae III and VI present; on latter segments, *d* inserted at some distance from proper solenidia (Fig. 7, Table 2). Formulae of leg setae (and solenidia, from trochanter to tarsus): I—1-(5-6)-3(1)-4(2)-20(2), II—1-(5-6)-3(1)-4(1)-15(2), III—2-3-3(1)-4(1)-15, IV—1-2-3-4(1)-12. Legs monodactylous.

TABLE 2. Ontogeny of leg setae (Roman letters), solenidia and famulus (Greek letters) of *Amerus polonicus*.

Leg	Trochanter	Femur	Genu	Tibia	Tarsus
Leg I					
Larva	—	<i>d, bv''</i>	(<i>l</i>), <i>d, σ</i>	(<i>l</i>), <i>v', d, φ₁</i>	(<i>fi</i>), (<i>tc</i>), (<i>p</i>), (<i>u</i>), (<i>a</i>), <i>s</i> , (<i>pv</i>), (<i>pl</i>) ε , ω_1
Protonymph	—	—	—	—	ω_2
Deutonymph	<i>v'</i>	(<i>l</i>)	—	<i>v'', φ₂</i>	—
Tritonymph	—	<i>v''₁</i>	<i>v'</i>	—	(<i>it</i>)
Adult	—	<i>v''₂¹</i>	<i>d</i> lost	<i>d</i> lost	<i>l'', v'</i>
Leg II					
Larva	—	<i>d, bv''</i>	(<i>l</i>), <i>d, σ</i>	<i>l', v', d, φ</i>	(<i>fi</i>), (<i>tc</i>), (<i>p</i>), (<i>u</i>), (<i>a</i>), <i>s</i> , (<i>pv</i>), ω_1
Protonymph	—	—	—	—	—
Deutonymph	<i>v'</i>	(<i>l</i>)	—	<i>l''</i>	ω_2
Tritonymph	—	—	<i>v'</i>	<i>v''</i>	(<i>it</i>)
Adult	—	<i>v''₁, v''₂¹</i>	<i>d</i> lost	<i>d</i> lost	—
Leg III					
Larva	—	<i>d, ev'</i>	<i>l', d, σ</i>	<i>v', d, φ</i>	(<i>fi</i>), (<i>tc</i>), (<i>p</i>), (<i>u</i>), (<i>a</i>), <i>s</i> , (<i>pv</i>)
Protonymph	<i>v'</i>	—	—	—	—
Deutonymph	<i>l'</i>	<i>l'</i>	—	<i>l'</i>	—
Tritonymph	—	—	<i>v'</i>	<i>v''</i>	(<i>it</i>)
Adult	—	—	—	—	—
Leg IV					
Protonymph	—	—	—	—	<i>fi'', (p), (u), (pv)</i>
Deutonymph	<i>v'</i>	<i>d, ev'</i>	<i>d, l'</i>	<i>v', d, φ</i>	(<i>a</i>), <i>s</i>
Tritonymph	—	—	<i>v'</i>	<i>l', v''</i>	(<i>tc</i>)
Adult	—	—	—	—	—

Note: structures are indicated where they are first added and are present through the rest of ontogeny; pairs of setae in parentheses, dash indicates no additions; ¹in some individuals absent.

Description of juvenile stages

Larva oval in dorsal aspect (Fig. 8a) and unpigmented, some parts of body covered with granular cerotegument. Prodorsum subtriangular, rostrum rounded, cerotegument irregularly punctated. Prodorsal seta *in* long, *ro* and *le* of medium size (Table 1), seta *ex* short; most with short barbs, except for smooth *ex*. Mutual distance between setal pair *in* almost as long as that between pair *ro*, and between setal pair *le* nearly half of that between pair *ro*. Seta *le* inserted closer to *ro* than to *in*. Bothridium large, rounded, bothridial seta long, setiform and barbed.

Gastronotum of larva with 11 pairs of setae, including *h₂* inserted laterally to posterior part of anal valves (Figs. 9a, 10a). Most gastronotal setae long and with short barbs, except for short and smooth *c₂*; all curved, distally thin and inserted on apophyses, apophyse at seta *dm* larger than at other setae. Anal valves (segment P) glabrous (Fig. 9a). Cupule *ih* lateral to anterior part of anal

valves, other cupules and gland opening not observed in punctated cerotegument. Seta *d* on femur I and *ft*'' on tarsus I very long, seta *d* on tibia I, *l*'' on genu III and most setae on tarsi long, other setae of medium size or short; most barbed, except for smooth *d* on tibia I and *p* on tarsi (Figs. 8a, 10a 11). Basal part of all tarsi short, beaded, with long and thin distal parts, especially of tarsus I, bearing setae *p* and *u*, other setae inserted on basal parts of tarsi. Solenidion ϕ_1 on tibia I long, adjoined to coupled seta *d*, other solenidia and famulus ϵ of medium size or short.

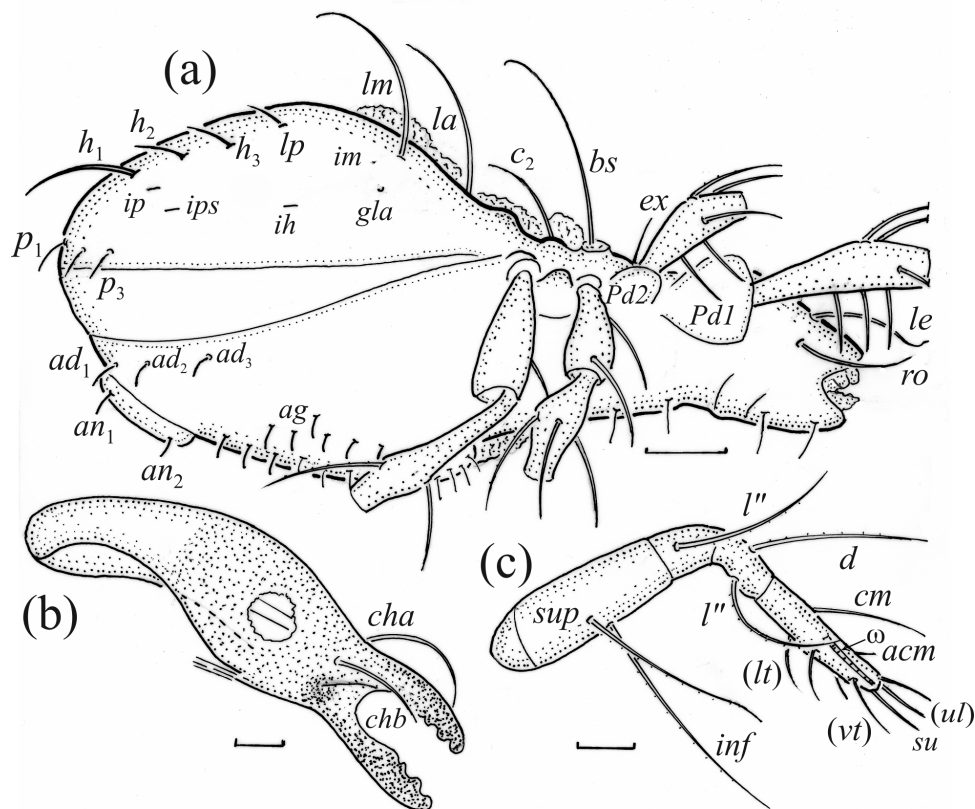


FIGURE 4. *Amerus polonicus*. (a) Adult, lateral aspect, legs partially drawn, scale bar 100 µm; mouthparts, right side, antiaxial view, scale bars 20 µm; (b) chelicera (Trägårdh organ indicated in 'transparent' area); (c) palp.

Nymphs oval in dorsal aspect, shape of prodorsum, prodorsal setae, bothridium and bothridial seta as in larva, but seta *in* short and smooth. Gastronotum of protonymph with 12 pairs of setae because setae *h*₃ and setae of *p*-series appear (Fig. 9b), remaining in deutonymph and tritonymph (Figs. 12a, 12b, 13), setae of *d*-series lost and remaining absent in all nymphs. In protonymph, seta *p*₁ long, other setae of *p*-series of medium size; all barbed (Fig. 9b), in deutonymph and tritonymph, all these setae long (Figs. 12a, 12b). In all nymphs, gastrontal setae on small apophyses. All nymphs carry exuvial scalps of previous instars, using mainly the inwards curving seta *la* and *lm* to protect the exuvial scalps against loss (Figs. 6c, 6d, 10b, 13, 14a, 14c, 15a). Prodorsum of tritonymph, exuvial scalps and legs with granular cerotegument. In protonymph, one pair of setae appears on genital valves, and three pairs in deutonymph and one pair in tritonymph added (Figs. 9b, 12a, 12b); all short and smooth. In deutonymph, two pairs of aggenital setae and three pairs of adanal setae appear, and in tritonymph five pairs of aggenital setae added; all short and smooth. Anal valves of protonymph (segment AD) and deutonymph (segment AN) glabrous, in tritonymph 2–3 pairs of anal

setae present (Fig. 12a, 12b), all short and smooth. Among four tritonymphs investigated, one individual with three pairs of anal setae, on individual with two pairs and one unpaired seta, and two individuals with two pairs. In protonymph, cupule *ips* located lateral to anterior part of anal valves, and cupule *ih* displaced posterolateral to *ips*, in deutonymph and tritonymph cupule *iad* located posterolateral to anterior part of anal valves, and cupule *ips* displaced posterolateral and cupule *ih* anterolateral to *iad*. Other cupules and *gla* opening not observed in punctated cerotegument. In tritonymph, seta *d* on femur I and *fl''* on tarsus I very long (Figs. 6c, 6d, 10b, 13, 14a, 14b, 15c, 15d, 16), seta *d* on femora II–IV and genu and tibia IV, *l'* on genu III and IV, and most setae on tarsi long, other setae of medium size or short; most barbed, except for smooth *d* on tibia I and *p* on tarsi (Fig. 16). Basal part of all tarsi short, beaded, with long and thin distal parts, especially of tarsus I, bearing setae *p* and *u*, other setae inserted on basal parts of tarsi. Solenidion ϕ_1 on tibia I long, adjoined to coupled seta *d*, other solenidia and famulus ϵ of medium size or short.



FIGURE 5. *Amerus polonicus*, male, photos, dorsal view. (a) Whole body, (b) anterior part of body, (c) notogaster, (d) central part of notogaster, (e) part of legs I and II, (f) male while and after moulting; scale bars: (a) 200 μm , (b–e) 100 μm , (f) 400 μm .



FIGURE 6. *Amerus polonicus*, adult, SEM micrographs. Adult, (a) dorsal view, (b) lateral view; tritonymph, (c) dorsal view, (d) lateral view.

Summary of ontogenetic transformations

In all instars of *A. polonicus*, the prodorsal setae *ro* and *le* are of medium size. In all juveniles, seta *ex* is short, and seta *in* is long in the larva and is short in the nymphs, whereas in the adult seta *in* is short and seta *ex* is of medium size. In all instars, the opening of bothridium is rounded and the bothridial seta is setiform. The larva has 11 pairs of gastronotal setae, including *h*₂, and the nymphs have 12 pairs (*h*₃ and *p*-series appear, *d*-series is lost in protonymph). The notogaster of adult loses setae *c*₁ and *c*₃, so that 10 pairs remain. The formulae of gastronotal setae of *A. polonicus* is 11-12-12-10 (from larva to adult). Formulae of epimeral setae is 3-1-2 (larva, including scaliform *lc*), 3-1-3-1 (protonymph), 3-1-3-2 (deutonymph) and 3-1-3-3 (tritonymph and adult). Formula of genital setae is 1-4-5-6 (protonymph to adult), aggenital setae is 2-7-(12-13) (deutonymph to adult), and segments PS-AN is 03333-0333-0(2-3)(2-3). Ontogeny of leg setae and solenidia is given in Table 2.

Distribution, ecology and biology

According to Subías (2004, 2020), *A. polonicus* has a Mediterranean distribution, but on the base of hitherto obtained data (Pérez-Íñigo 1976; Olszanowski *et al.* 1996; Mahunka & Mahunka-Papp 2000; Siepel *et al.* 2009; Miko 2016; Murvanidze & Mumladze 2016) and the type locality of Kulczynski (1902), this species occurs in Poland, Hungary, the Netherlands, Czech Republic and Georgia. *Amerus polonicus* inhabits Alpine meadows, forests and urban soils (Murvanidze & Mumladze 2016). In forests, this species prefers beech (*Fagus* sp.) litter (Beck & Woas 1991; Beck *et al.* 2007), and fermentation layer (Wunderle 1992). Probably therefore, in old beech forests this species was clearly more abundant in moder type of humus (plant association *Luzulo-Fagetum*) than in mull humus (plant association *Galio-Fagetum*, Beck *et al.* 2007).

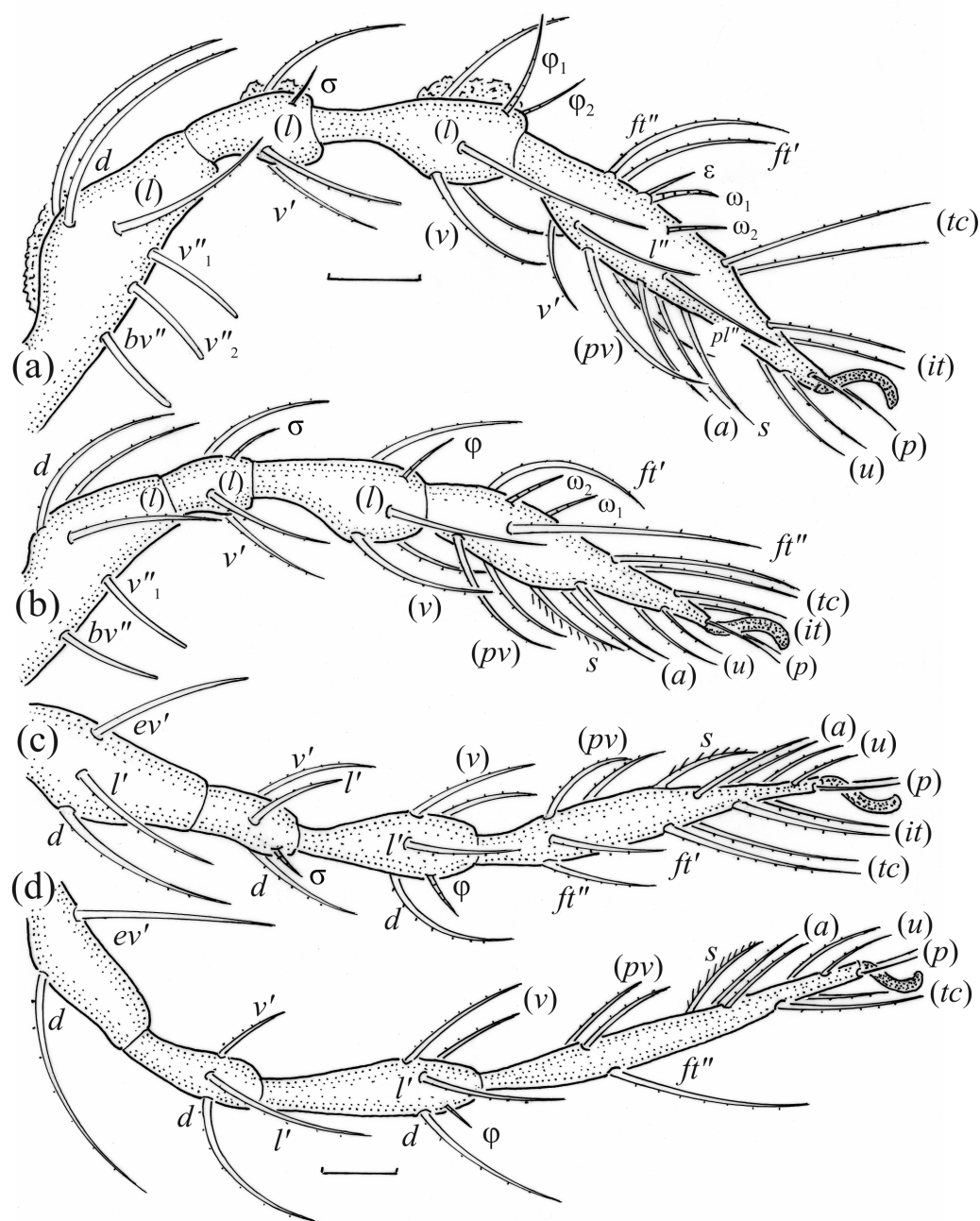
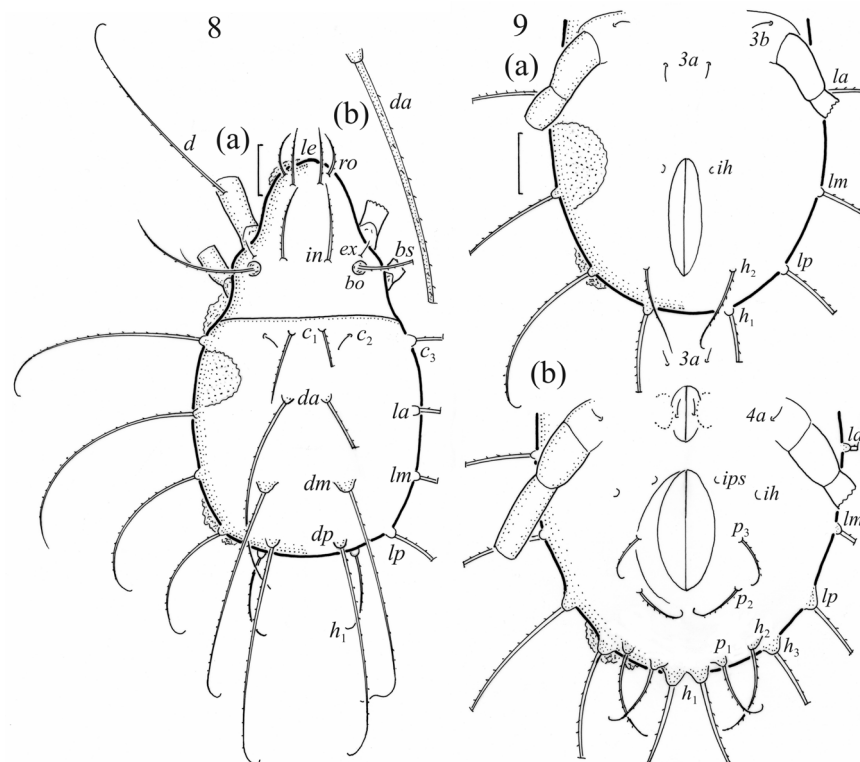


FIGURE 7. *Amerus polonicus*, adult, part of femur to tarsus, lateral aspect, seta on the opposite side not illustrated, but indicated in the legend, scale bars 50 μ m. (a) Leg I, tarsus (pl'); (b) leg II; (c) leg III; (d) leg IV.

In our investigation, *A. polonicus* was found in an old oak forest near Sierpc (Mazovia Province, Poland), but was not abundant. From 10 large samples (500 cm^3 each), we extracted 12 adults and two deutonymphs in total. During this extraction, the mites left the samples relatively quickly (in 1–3 days), and were present only in three of the total of 10 samples, so constancy of this species in these samples was 30% and mean density was 1.4 individuals per 500 cm^3 .



FIGURES 8–9. *Amerus polonicus*, legs partially drawn, scale bars 50 μm. 9. (a) Larva, dorsal aspect, (b) shape of seta *da* (enlarged). 10. Ventral side of hysterosoma, (a) larva, (b) protonymph.

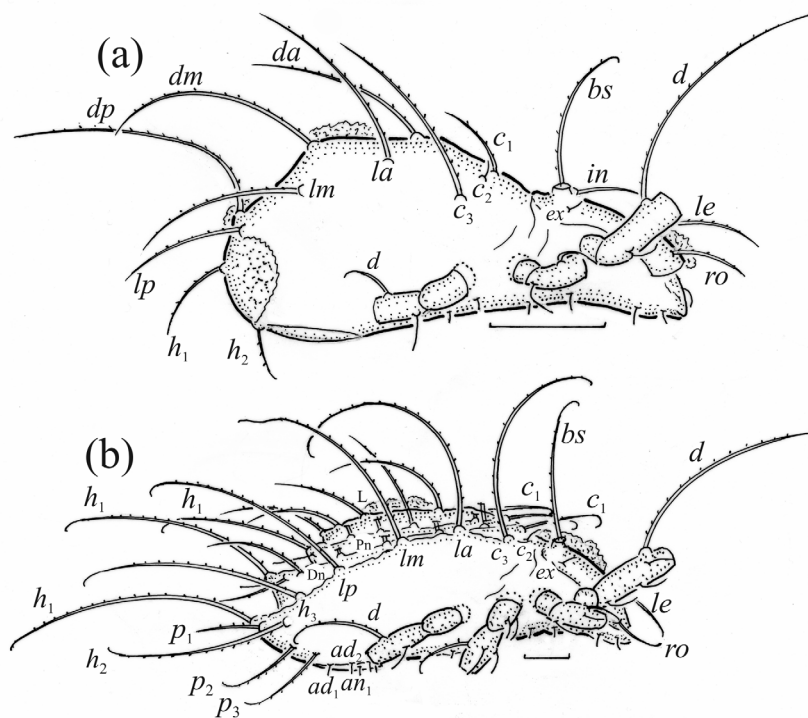


FIGURE 10. *Amerus polonicus*, lateral aspect, legs partially drawn, scale bars 50 μm. (a) Larva, (b) tritonymph.

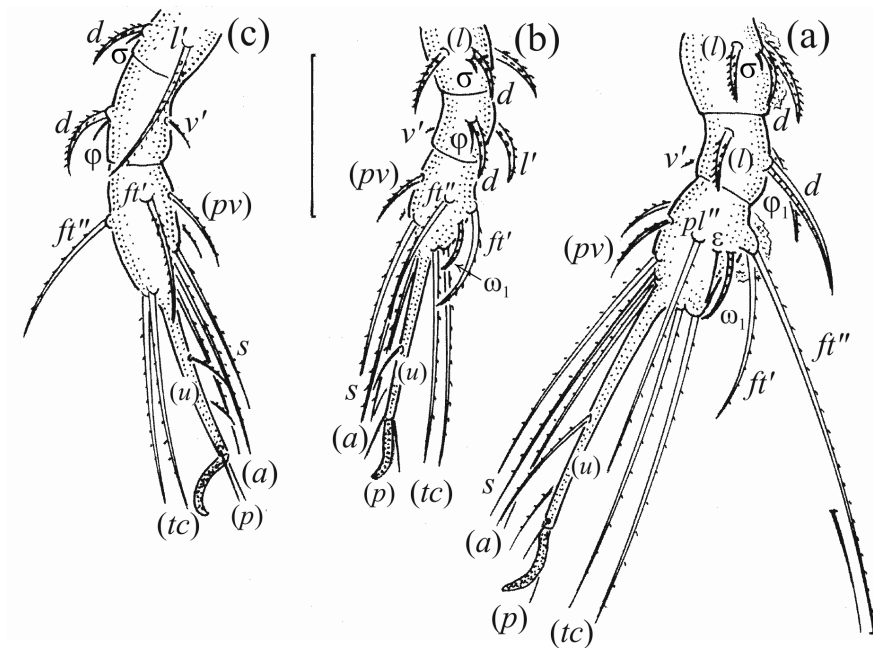
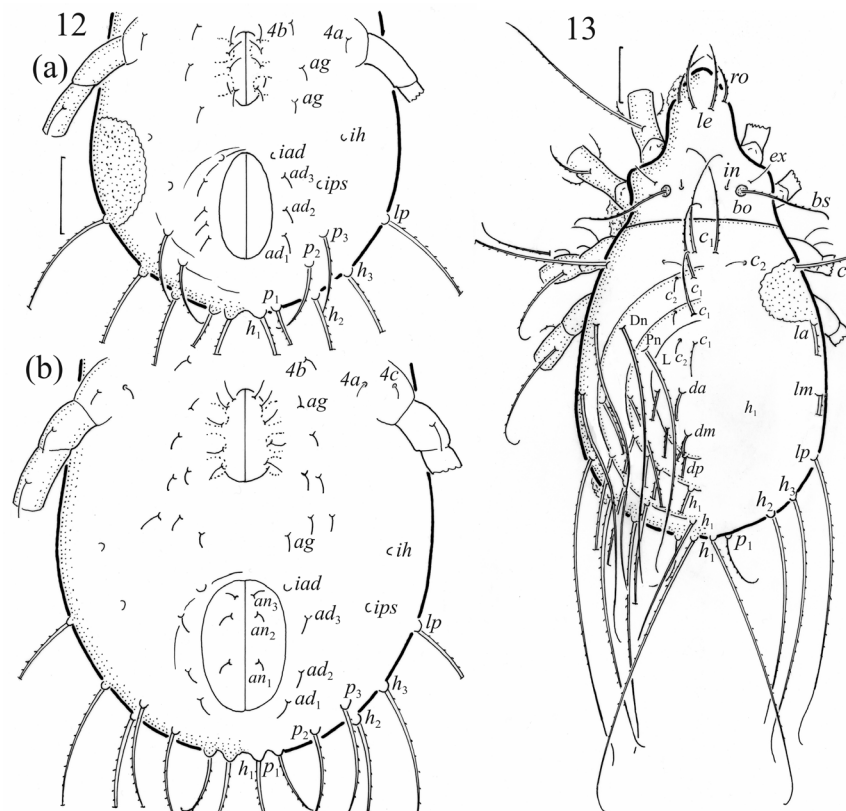


FIGURE 11. *Amerus polonicus*, leg segments of larva (part of genu to tarsus), right side, seta on the opposite side are not illustrated, but indicated in the legend, scale bar 20 μ m. (a) leg I, tarsus (pl'), (b) leg II, (c) leg III.



FIGURES 12–13. *Amerus polonicus*, legs partially drawn, scale bars 50 μ m. 12. Ventral side of hysterosoma, (a) deutonymph, (b) tritonymph. 13. Tritonymph, dorsal aspect.

In laboratory culture (temperature 25°C, 90% air humidity), the development of *A. polonicus* lasted 68 days, including the following stages: 13 days as egg, eight days as larva (+ one day as immovable pre-moulting form), seven days as protonymph (+ two days as pre-moulting form), 10 days as deutonymph (+ four days as pre-moulting form), 14 days as tritonymph (+ nine days as pre-moulting form), from which the adult appeared. While moulting, the tritonymphal hysterosoma broke around the opisthonotum posteriorly, and the adult slowly left the tritonymphal moult in the same direction (Fig. 5f). This action from breaking tritonymphal hysterosoma lasted about nine hours, and a new adult was light brown and moved slowly. From this culture, we obtained five larvae, five protonymphs, four deutonymphs, four tritonymphs and one adult in total. Among the adults obtained from field samples and culture, males were more abundant than females (sex ratio 1:1.2), and 50% of females were gravid. The gravid females usually carried three large eggs, each 337 x 182, which constituted about 35% of the total body length of females.

Comparison of morphology of *Amerus polonicus* with congeners and remarks

Avanzati *et al.* (2003) redescribed the adult of *A. troisi* and described *A. cuspidatus* Avanzati *et al.*, 2003, which differs from the former species mainly by the shape of rostrum, wider separated interlamellar seta in the latter species and on molecular level. These two species differ clearly from *A. polonicus* studied herein mainly by longer setae *lp*, *h*₂ and *h*₃ on the notogaster; in *A. polonicus*, these setae are clearly shorter than seta *h*₁, whereas in other species these setae are of similar length. The palp of *A. polonicus* has more setae on tarsus (formula 0-2-1-2-9) than those of other species (formula 0-2-1-2-7, Avanzati *et al.* 2003).

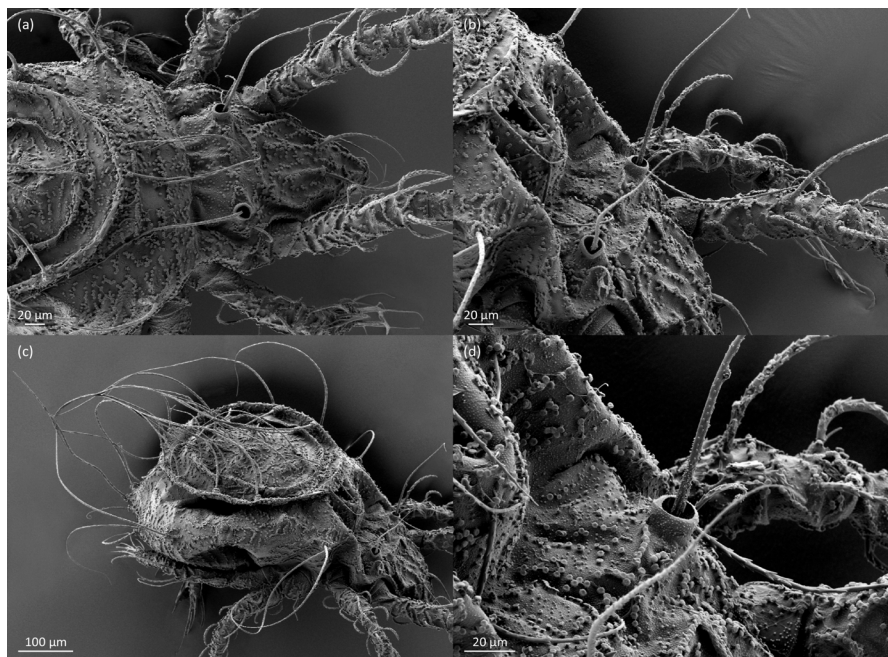


FIGURE 14. *Amerus polonicus*, tritonymph, SEM micrographs. (a) Anterior part of body, dorsal view, (b) anterior part of body, dorsolateral view, (c), exuviae of previous instars on the gastronomum, lateral view (d) bothridium and bothridial seta, dorsolateral view.

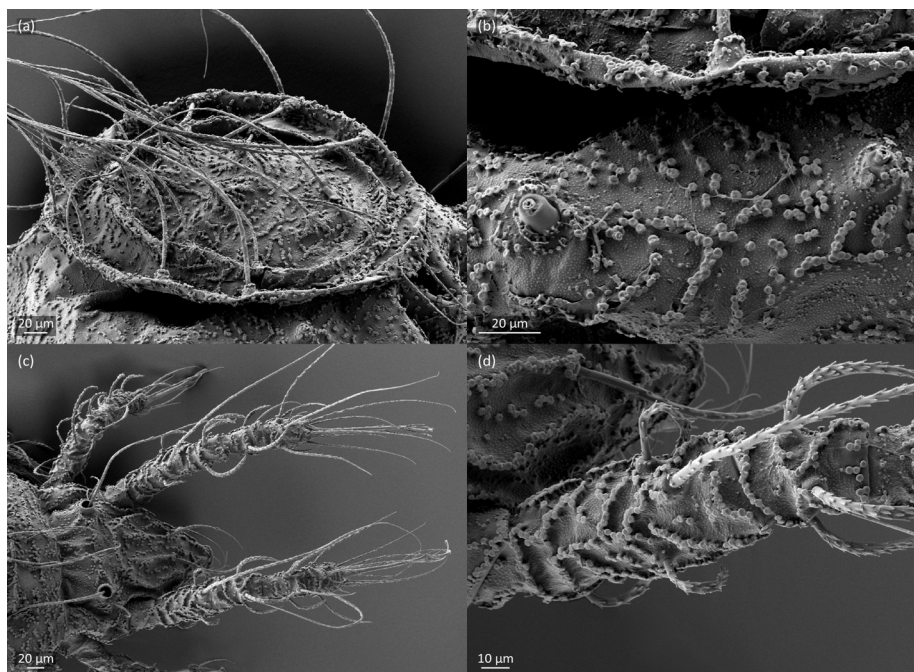


FIGURE 15. *Amerus polonicus*, tritonymph, SEM micrographs. (a), (b) Granular cerotegument on exuviae, (c) legs I and II, dorsal view, (d) fragment of leg I, dorsal view.

Willmann (1939) included *Amerus lundbladi* in *Neamerus* mainly based on some skeletal characters of one individual. However, the figures of this individual indicate that the body shape of *A. lundbladi* differs slightly from the other species, but the shape of rostrum and shape and distribution of prodorsal setae are similar in all species of *Amerus*. The notogastral seta c_2 is missing in the figure by Willmann (1939), but setae la and lm are long, as in other species of *Amerus*, and setae lp , h_2 and h_3 are slightly longer than in *A. polonicus*. In *A. lundbladi*, most ventral setae are missing, but the epimeral seta la is long and lc slightly shorter, as in *A. polonicus*. In all species, the bothridial seta is setiform, but in *A. lundbladi* it is barbed, whereas in other species is smooth. Willmann (1939) drew leg I of *A. lundbladi*, which has three relatively thick ventral setae on femur, as in *A. polonicus*. Therefore, more investigations are needed on *A. lundbladi*, including the juvenile stages, to contradict or support its membership in *Neamerus*.

The morphology of the adult *A. polonicus* varies in different geographical regions. For example, the individual from Iran (Akrami *et al.* 2008) has setae lp , h_2 and h_3 relatively longer and thicker than *A. polonicus* from Poland studied herein, especially seta lp . Also individuals from Turkey (Baran & Kilic 2013) and Hungary (Mahunka 1996) have these setae slightly longer than *A. polonicus* from Poland. In *A. polonicus* by Kunst (1971), seta lp is lacking, seta h_3 is longer, but seta h_2 is as short as in *A. polonicus* studied here. The individuals from Hungary have 13 pairs of aggenital setae (Mahunka 1996), whereas those from Poland have 12–13 pairs of these setae.

The shape of leg segments and setae of adult of *A. polonicus* is similar to that studied by Mahunka (1996). The leg setae are relatively strong, three smooth setae are present on the ventral part of femur I, and seta d is present on genu and tibia IV. The juveniles of *A. polonicus* have beaded tarsi with long and thin distal parts, similar tarsi occur in species of Damaeidae (Norton 1978, 1980; Seniczak & Seniczak 2011, 2013; Seniczak *et al.* 2013, 2016; Miko 2015). However, the amerid and damaeid species represent systematically distant groups, which suggest that the shape of leg segments is convergent.

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