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Eriophyid mites in the genus *Aculodes* Keifer (Prostigmata: Eriophyidae) from grasses – the first taxon from South America and world species information

Vanina Alemandri^a, Graciela Truol^a, Renata S. de Mendonça^b and Denise Navia^c

^aInstituto de Patología Vegetal, Centro de Investigaciones Agropecuarias, Instituto Nacional de Tecnología Agropecuaria (IPAVE-CIAP-INTA), Córdoba, Argentina (emails: alemandri.vanina@inta.gob.ar, truol.graciela@inta.gob.ar); ^bFundação de Apoio à Pesquisa (FUNAPE)/Embrapa Recursos Genéticos e Biotecnologia, Parque Estação Biológica, Brasília, Brazil (email: mendonca.rsm@gmail.com); ^cEmbrapa Recursos Genéticos e Biotecnologia, Parque Estação Biológica, Brasília, Brazil (email: denise.navia@embrapa.br)

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Aculodes Keifer (Eriophyidae) mites are often associated with grasses (Poaceae) and so far all species in this genus were described from the Northern hemisphere. During surveys of eriophyid mites from grasses conducted in Argentina aiming to determine the host range of the wheat curl mite, a new *Aculodes* species was collected from the feather grass, *Stipa* sp. The first *Aculodes* species is described from the Southern hemisphere. In addition to the traditional morphological description, DNA sequences of two genomic regions – the ITS rDNA and the 16S region mtDNA – were obtained and deposited in public databases; genetic distances of the new taxon with eriophyid species/genera associated with grasses are presented. In order to contribute to *Aculodes* taxonomy, a list for world grasses-associated species is provided and information on its morphological traits is summarized.

Keywords: Eriophyoidea; Poaceae; Argentina; DNA data; taxonomy

Introduction

The genus *Aculodes* Keifer, 1966b (Eriophyidae, Phyllocoptinae, Anthocoptini) presently includes 26 species, most of them (21 species) associated with grasses (Poaceae) (Nalepa 1891; Keifer 1944, 1952, 1960, 1966a, 1966b; Sukhareva 1972, 1981, 1985, 1986, 1994; Huang 1992, 2001; Kuang 1997; Kuang and Pang 1997; Shi and Boczek 2000; Skoracka et al. 2001, 2009; Skoracka 2003, 2004, 2005; Kuang et al. 2005; Skoracka and Pacyna 2005; Xue et al. 2010). Until now, all *Aculodes* species were described from the Northern hemisphere, in Europe, Asia and North America. Only one species in this genus, *Aculodes mckenziei* (Keifer, 1944), had been reported from the Southern hemisphere, in Australia and New Zealand (Frost et al. 1990; Guy and Gould 1996). No *Aculodes* species had been described or reported from South America.

Knowledge on eriophyid mites on grasses in South America is scarce; only eight species belonging to the genera *Aceria*, *Abacarus*, *Catarhinus*, and *Schizaceae* have been described or reported from these plants in the continent (Keifer 1959, 1976, 1977, 1978; Flechtmann and Aranda 1970; Flechtmann 2000; Navia et al. 2006; Pereira et al. 2009; Castiglioni and Navia 2010). In Argentina, only the wheat curl mite, *Aceria tosichella* Keifer, 1969, had been reported (Navia et al. 2006).

In early 2000s, the wheat curl mite was collected for the first time in South America, in Argentina, from wheat plants infected with *Wheat streak mosaic virus* (Navia et al., 2006). Since then, surveys of eriophyid mites from grasses in that country have been conducted to determine the host range of the wheat curl mite. From these surveys a

new *Aculodes* species was found from the feather grass *Stipa* sp. This is the first *Aculodes* associated with grasses to be described from South America and even from the Southern hemisphere.

Five eriophyid mites have been described or reported from *Stipa* grasses – four Eriophyidae species, three in the genus *Aceria* – *A. stipaespinnulata* Skoracka, 2004 from *S. joannis* Celak and *S. capillata* L. from Poland; *A. stipaensis* Mitrofanov & Sharonov, 1988 from *Stipa* sp. from Ukraine (Mitrofanov et al. 1988); and *A. stipacea* Sukhareva, 1983 described from *S. lessingiana* Trin. et Rupr. from Ukraine and also reported from *S. capillata* in Kazakhstan – and one in the genus *Aculodes* – *A. fulleri* (Keifer, 1966a) from *S. californica* Merr. & Burt-Davy ex Hall. (presently referred as a synonym of *S. occidentalis* Thurb. ex S. Watson (The Plant List 2013)) from California, USA; and one Phytoptidae species, *Novophytoptus stipae* Keifer, 1962 (host plant *Achnatherum speciosum* (Trin. & Rupr.) Barkworth, presently referred as a synonym of *Stipa speciosa* Trin. & Rupr. (The Plant List 2013).

DNA-based resources have started to be used in eriophyid mite systematics about 15 years ago. Important advances have been done using molecular techniques, which had contributed to explore some questions that were difficult to answer some years ago (Navajas and Navia 2010). Cryptic species have been uncovered among eriophyid mites associated with grasses through an integrative approach, including DNA data (Skoracka and Dabert 2010; Skoracka et al. 2012; Miller et al. 2013). Molecular information can help to distinguish species and provide DNA data for new taxa can help to build a

consistent taxonomy (Dabert et al. 2008). Among molecular markers successfully used for eriophyid mite systematics, we can list the nuclear ribosomal Internal Transcribed Spacer (ITS) and the 16S mitochondrial (Navia et al. 2005; Navajas and Navia 2010; Skoracka et al. 2012; Miller et al. 2013).

In this paper, the description of a new *Aculodes* mite associated with *Stipa* grass in Argentina is presented. In addition to the traditional morphological description, including measures and drawings, DNA sequences of two genomic regions – the ITS rDNA and the 16S region mtDNA – were obtained and are presented. Genetic distances of the new taxon with eriophyid species/genera were estimated.

Taxonomic literature on *Aculodes* species associated with grasses is sparse and description of some species is not in English making difficult its access. In a way to contribute to *Aculodes* taxonomy and make easier new studies, a list for grasses-associated valid species is presented as well as a table with summarized information on its main morphological traits.

Material and methods

Morphological study

Mites were collected from leaf samples by direct examination under a dissecting stereomicroscope and directly mounted in modified Berlese medium (Amrine and Manson 1996). Slide-mounted specimens were studied using a research phase and differential interference contrast microscope (Eclipse 80i Nikon, Tokyo, Japan). Relevant structures for taxonomic purposes were measured using a graded eyepiece and illustrated using a camera lucida attached to the microscope.

Terminology follows that of Lindquist (1996) and classification is based on Amrine et al. (2003). Measurements are given in micrometers (μm) and, unless stated otherwise, refer to the length of the structure. In the description of the female, each measurement of the holotype precedes the corresponding range for the paratypes. Some measurements of paratypes could not be taken because of the position in which the specimens were mounted. The specimen that was drawing in lateral view stayed mounted in a position slightly, since then the ventral seta *e* was not represented in its drawing. The count of ventral opisthosomal annuli starts from the first full annulus behind the genitalia. Dorsal opisthosomal annuli were counted from the first full annulus behind the middle of the prodorsal shield rear margin. When the length of cheliceral stylets is not given, it means that it was not possible to measure this character, since they were in a bundle that was immersed with other gnathosomal structures. Measurements were conducted according to de Lillo et al. (2010) except for the following: (1) the body length, which was measured from the tip of the frontal lobe to the rear end of the anal lobe, not considering pedipalps; (2) the *sc* tubercles space measurement

(distance between the tubercles), not the *sc* setae distance; (3) empodium length, which includes its basal portion inserted into the tarsus.

No immature stages were found and only one male was found and studied. Female internal genitalia was not visible in the studied specimens.

Micrographs were obtained using a digital system consisting of the phase and differential interference contrast microscope (Nikon Eclipse 80i, Tokyo, Japan) connected to a digital camera (Nikon DS-Ri, 12.7 mega pixels, Tokyo, Japan) which was in turn connected to a computer with NIS Elements software (Nikon).

Type specimens are deposited as slide-mounted specimens in mite collections at Embrapa Recursos Genéticos e Biotecnologia, Brasília, Brazil and at Departamento de Entomologia, Fitopatologia e Zoologia Agrícola, Escola Superior de Agricultura “Luiz de Queiroz” (ESALQ), Universidade de São Paulo, Piracicaba, São Paulo, Brazil.

Molecular characterization

Specimens were preserved in absolute ethyl alcohol and then individualized in an eppendorf tube for DNA extraction.

DNA extraction

A Chelex method was used to extract DNA from a single mite following Carew et al. (2004). Microcentrifuge tubes containing mites were centrifuged at 20800 g for 5 min to ensure that mites were at or near the top of the tube. Three microlitres of Proteinase K (Roche) was added to each tube and mites were crushed using a plastic pestle moulded from a pipette tip (fresh pestle used for each mite extraction). One hundred microlitres of 5% Chelex (Bio Rad) solution was added per tube, before the tubes were gently vortexed, and incubated, initially for 1–1.5 h at 55°C, and then for 8 min at 90°C. Mite extractions were cooled on ice and stored at –20°C (Carew et al. 2009).

Polymerase chain reaction amplification (PCR)

The ITS region (a fragment of about 900 bp) was amplified using the forward and reverse primers 18S and 28SC as described by Navia et al. (2005). PCR reactions were carried out in a final volume of 25 μl containing 1 U *Taq* DNA polymerase (Invitrogen), 2.5 mM MgCl_2 , 0.25 mM dNTP, 0.5 μM of each primer, and 6 μl of DNA. The reactions were performed with a thermal cycler programmed for one cycle of 4 min at 94°C, followed by 35 cycles (30 s at 94°C, 30 s at 50°C, and 1 min at 72°C) and a final 5-min extension at 72°C. The 16S region (a fragment of about 400 bp) was amplified using the forward and reverse primers LR-J-12887 and WCM16S as described by Carew et al. (2009). PCR reactions were carried out in a final volume of 25 μl containing 0.75 U *Taq* DNA polymerase (Invitrogen), 2 mM MgCl_2 , 0.20 mM dNTP, 0.5 μM of each primer, and 6 μl of

DNA. The reactions were performed with a thermal cycler programmed for one cycle of 7 min at 95°C, followed by 40 cycles (20 s at 95°C, 45 s at 53°C, and 30 s at 72°C) and a final 5-min extension at 72°C. Amplification products were analysed by 1.5% agarose gel electrophoresis. The amplified products were purified using QIAquick PCR Purification Kit (QIAGEN, Germany) and sequenced in both directions with the amplification primers using an ABI 3130XL (Applied Biosystems) automated sequencer.

Sequence data

The genome regions were aligned using progressive multiple-sequence alignment: ClustalX® version 1.81 software (Thompson et al. 1997). All sequences generated in this work were recorded in GenBank (KF648353-KF648356). The ITS and 16S sequences generated here and the corresponding to predominant haplotype *A. tosichella* detected in Argentina published by Skoracka et al. (2012) and Miller et al. (2013) were used to calculate the distances. MEGA6 (Tamura et al. 2013) was used to choose the best substitution model for our data, and for pairwise comparison of genetic distances. Kimura-2-parameter (K2P) and Tamura 3-parameter (T92) were chosen as the best model for the ITS and 16S data set, respectively.

Results and discussion

Taxonomy

Family **Eriophyidae** Nalepa, 1898
 Subfamily **Phyllocoptinae** Nalepa, 1892
 Tribe **Anthoceptini** Amrine and Stasny, 1994
 Genus ***Aculodes*** Keifer, 1966b
Aculodes stipacolus Alemandri and Navia **sp. nov.**
 (Figures 1–3)

Differential diagnosis

The new species was compared to all *Aculodes* species associated with grasses. *Aculodes stipacolus* **sp. nov.** is most similar to *A. koeleriae* Sukhareva, 1985 and to *A. ponticus* Sukhareva, 1986 based on the prodorsal shield ornamentation pattern, with admedian lines complete and subparallel, submedian lines I absent, and submedian lines II following lateral margins of shield. However, it differs from both species in the presence of a prodorsal shield short median line (absent in *A. koeleriae* and *A. ponticus*); and in the number of empodium rays (9 symmetrical rays in *A. stipacolus*, 6–7 in *A. ponticus*, and 6–7 in *A. koeleriae*). The new species is also similar to *A. calamaabditus* Skoracka, 2003 based on the general aspect of the prodorsal shield – pointed, acuminate slightly curved, and broad-based frontal lobe; complete, subparallel and slightly curved admedian lines; and antero lateral area finely granulated; on the scapular seta (*sc*) length – 22 (21–29) in *A. stipacolus* and 21 (12–28) in

A. calamaabditus – in the overlapped number of dorsal annuli – 71 (64–74) in *A. stipacolus* and 62 (59–79) in *A. calamaabditus*. However, *A. stipacolus* differs from *A. calamaabditus* in the absence of submedian lines I (present in *A. calamaabditus*) and in the number of empodium rays (9 rays in *A. stipacolus*, 7–8 in *A. calamaabditus*). The new species share the number of empodium rays (9) with *Aculodes bambusae* Kuang, 1997 (8–9), *Aculodes dubius* (Nalepa, 1891), and *Aculodes multiricavus* Skoracka, 2004 (9).

Description

Female (*n* = 10). Body wormlike, 191 (191–223), 41 (37–46) wide, whitish.

Gnathosoma: 15 (14–17), projecting slightly downwards; pedipalp coxal seta (*ep*) 2 (2–2), dorsal pedipalp genual seta (*d*) simple, 8 (7–9), cheliceral stylets 18 (14–19), oral stylets 14 (12–14).

Prodorsal shield 37 (36–38), 28 (23–29) wide, subtriangular; frontal lobe pronounced, acuminate, slightly sinuous, relatively broad-based, 6 (6–8), 8 (8–9) wide; smooth. Line pattern of a short median line, faint, on rear 2/3 or 1/4 (longer when seen laterally); admedian line complete, subparallel, slightly curved on rear; submedian lines I absent; submedian lines II following lateral margins of shield; external antero lateral area finely granulated, some granules designing concave or diagonal lines. Scapular tubercles on rear shield margin, 16 (13–16) apart, scapular seta (*sc*) 22 (21–29), directed backward.

Legs: with all usual segments and setae present. **Leg I** 29 (28–32); femur 10 (9–10), ventral basifemoral seta (*bv*) 8 (7–10); genu 6 (6–7), antaxial genual seta (*l''*) 15 (15–19); tibia 7 (7–7), paraxial tibial seta (*l'*) 8 (7–10); tarsus 8 (7–8), antaxial fastigial tarsal seta (*ft''*) 20 (20–24), paraxial fastigial tarsal seta (*ft'*) 12 (12–16), paraxial unguinal tarsal seta (*u'*) 5 (5–6), tarsal empodium (*em*) 7 (7–8), simple, bilaterally symmetrical, 9 rays, tarsal solenidion (ω) 9 (8–10), curved, blunt. **Leg II** 26 (26–29); femur 10 (9–10), ventral basifemoral seta (*bv*) 10 (10–11); genu 5 (5–6), antaxial genual seta (*l''*) 8 (7–10); tibia 6 (5–6); tarsus 7 (7–8), antaxial fastigial tarsal seta (*ft''*) 18 (18–23), paraxial fastigial tarsal seta (*ft'*) 8 (8–9), paraxial unguinal tarsal seta 5 (5–7), tarsal empodium (*em*) 7 (6–7), simple, bilaterally symmetrical, 8 rays, tarsal solenidion (ω) 10 (9–10), curved, blunt.

Coxigenital region with 5 (5–5) microtuberculated annuli. **Coxisternal plates:** sternal line (internal coxisternal apodeme) 7 (6–7); coxisternum I and II densely ornamented with numerous curved short lines or dashes. Anterior seta on coxisternum I (*Ib*) 7 (7–10), 10 (9–10) apart; proximal seta on coxisternum I (*Ia*) 13 (12–14), 6 (5–7) apart; proximal seta on coxisternum II (*2a*) 28 (27–31), 20 (15–20) apart; **Female genitalia** 13 (11–13), 18 (16–19)

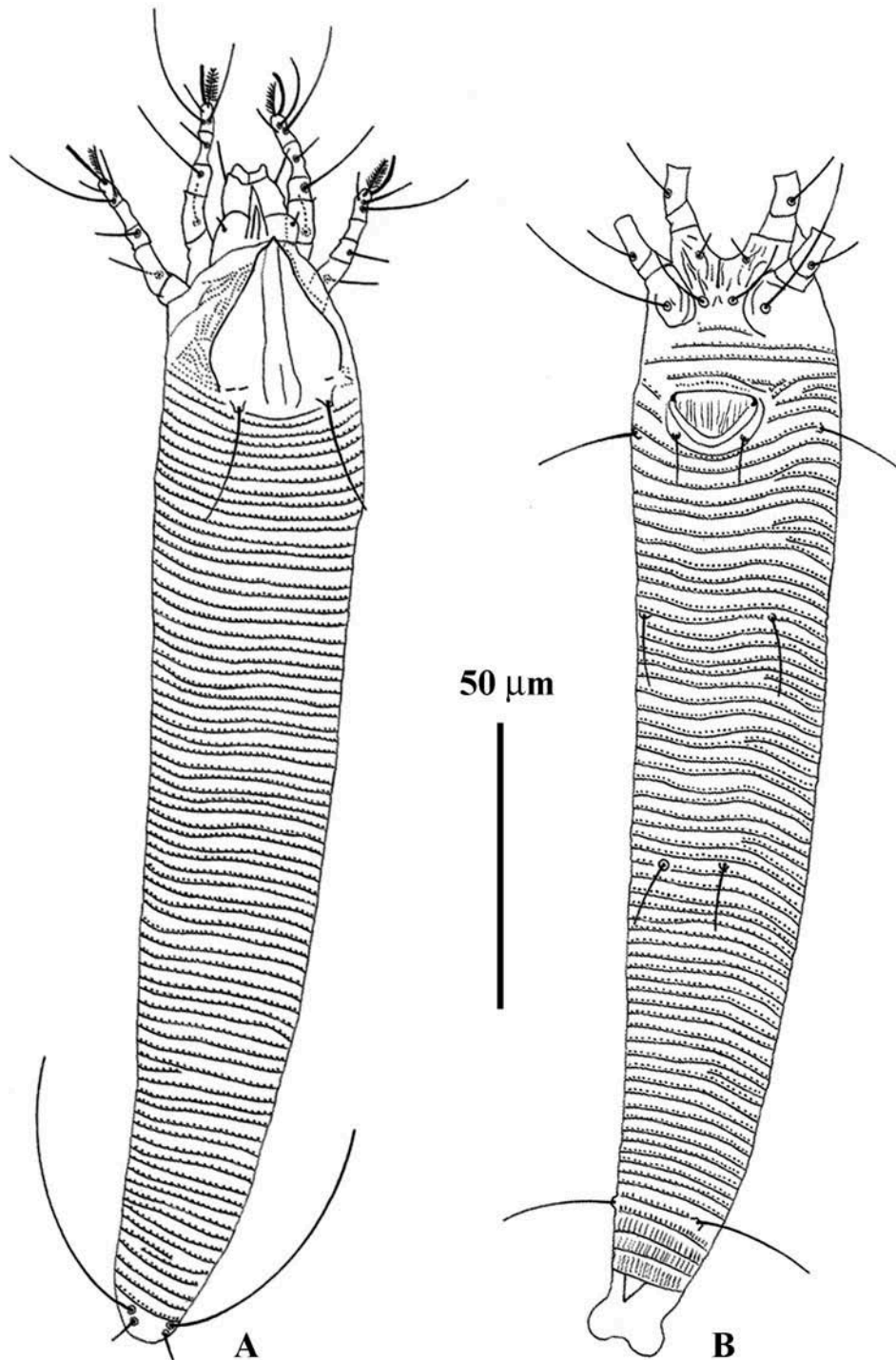


Figure 1. *Aculodes stipacolus* Alemandri and *Navia* sp. nov.: (A) dorsal habitus, female; (B) ventral habitus, female.

wide, coverflap with one transverse row of 10 (10–12) longitudinal ridges, genital seta *3a* 12 (10–19).

Opisthosoma evenly rounded, 71 (64–74) dorsal annuli, 62 (59–67) ventral annuli. Dorsal annuli with minute rounded microtubercles situated on or near rear margin of each annulus; ventral annuli with bead-like microtubercles situated on or near rear margin of each annulus; microtubercles more elongate on the last 5–7 ventral

annuli (posteriorly ventral seta *f*). Seta *c2* 21 (20–27), on ventral annulus 2 (2–2); seta *d* 29 (29–43), on ventral annulus 12 (12–16), 27 (19–27) apart, 20 (18–22) microtubercles apart; seta *e* 10 (10–14), on ventral annulus 31 (28–35), 12 (10–12) apart, 9 (7–10) microtubercles apart; seta *f* 18 (18–26), on ventral annulus 58 (55–63), 16 (8–16) apart, 14 (13–15) microtubercles apart. Caudal seta *h2* 56 (56–85), accessory seta *h1* 5 (4–6).

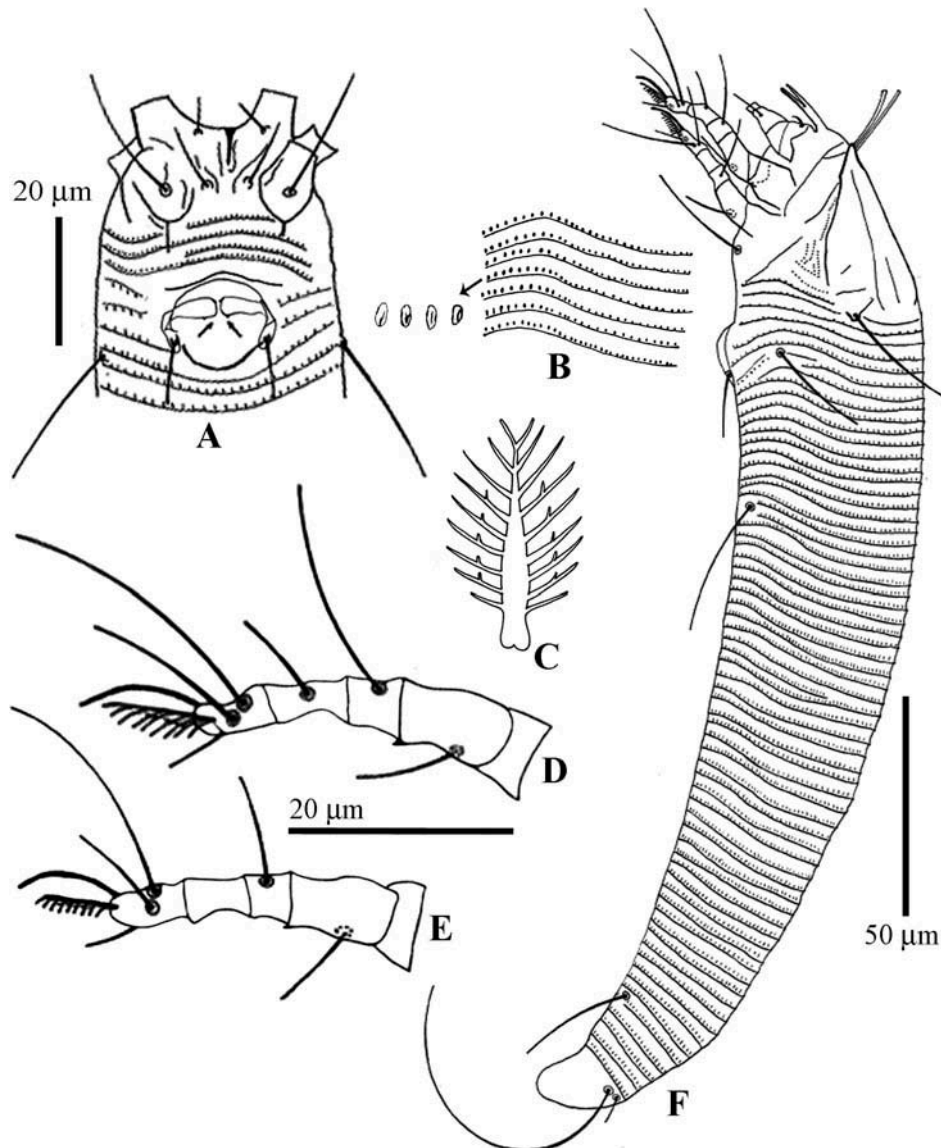


Figure 2. *Aculodes stipaculus* Alemandri and *Navia* sp. nov.: (A) coxigenital region, male; (B) detail of microtubercles in lateral view; (C) empodium, female (enlarged); (D) Leg I; (E) Leg 2; (F) Lateral view, female.

Male ($n = 1$): Smaller than female, 170, 33 wide. **Gnathosoma** 14; dorsal pedipalp genual seta (d) simple 7, other setae and structures not clearly distinguishable.

Prodorsal shield 32, 26 wide; frontal lobe and shield design similar to that of the female. Frontal lobe 5, 8 wide. Scapular tubercles on rear margin, 14 apart, scapular seta (sc) 19, directed backward.

Legs with usual series of setae. **Leg I** 24; femur 9, ventral basifemoral seta (bv) 6; genu 7, antaxial genual seta (l'') 12; tibia 6, paraxial tibial seta (l') 6; tarsus 7, antaxial fastigial tarsal seta (em) (ft'') 18, paraxial fastigial tarsal seta (ft') 11, paraxial unguinal tarsal seta (u') 5; tarsal empodium 6, simple, bilaterally symmetrical, 8 rays, tarsal

solenidion (ω) 8, slightly curved, blunt. **Leg II** 23; femur 9, ventral basifemoral seta (bv) 8; genu 4, antaxial genual seta (l'') 8; tibia 6; tarsus 6, antaxial fastigial tarsal seta (ft'') 20, paraxial fastigial tarsal seta (ft') 7, paraxial unguinal tarsal seta (u') 5, tarsal empodium (em) 6, simple, bilaterally symmetrical, 8 rays, tarsal solenidion (ω) 9, slightly curved, blunt.

Coxigenital region with 4 annuli, finely microtuberculated. **Coxisternal plates**: sternal line (internal coxisternal apodeme) 6; coxisternum I and II with sparse curved short lines or dashes. Anterior seta on coxisternum I ($1b$) 5, 9 apart; proximal seta on coxisternum I ($1a$) 10, 5 apart; proximal seta on coxisternum II ($2a$) 20, 15 apart; male genitalia 12, 16 wide, eugenital seta minute, genital seta $3a$ 10.

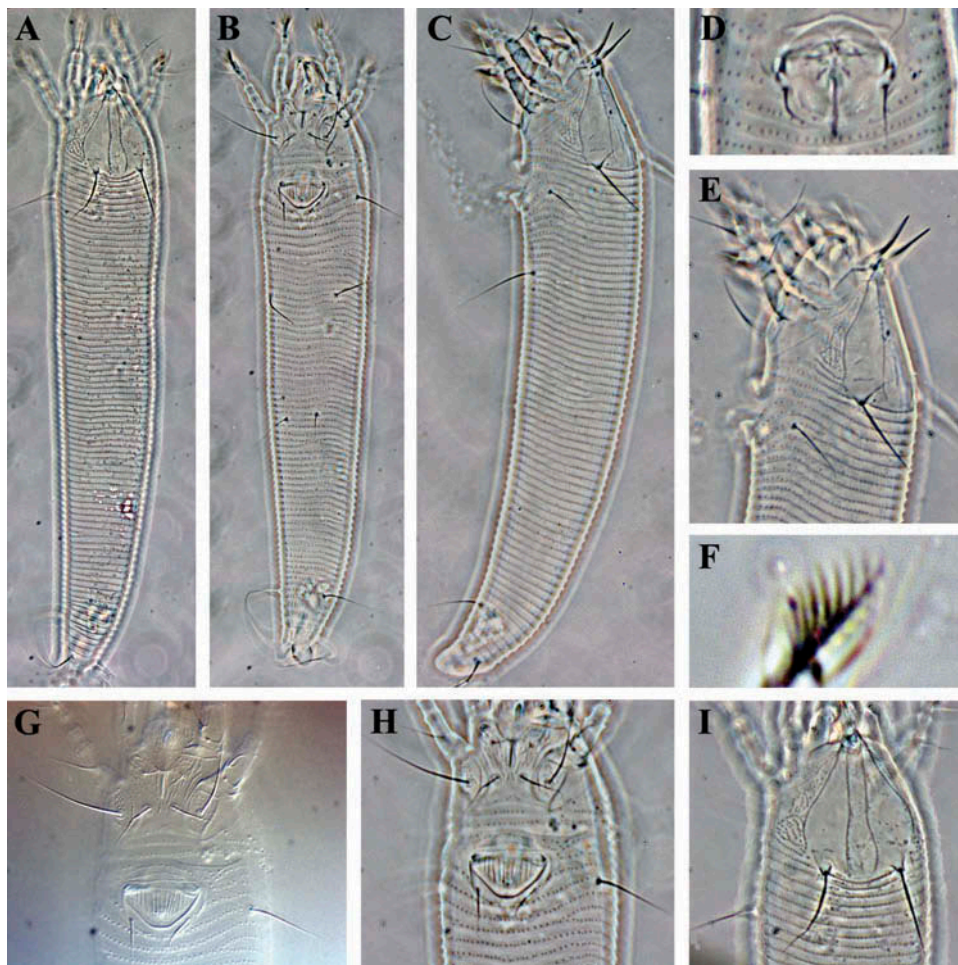


Figure 3. *Aculodes stipacolus* Alemandri and *Navia* sp. nov. micrographs. (A) dorsal view, female; (B) ventral view, female; (C) lateral view, female; (D) coxigenital region, male; (E) anterolateral view, female; (F) empodium; (G) coxigenital region, female, under DIC microscope; (H) coxigenital region, female, under phase contrast microscope; (I) anterodorsal view with prodorsal shield ornamentation details.

Opisthosoma evenly rounded, 55 dorsal annuli, 47 ventral annuli. Dorsal and ventral microtubercles similar to that of the female. Seta *c2* 20, on ventral annulus 2; seta *d* 33, on ventral annulus 10, 21 apart, 15 microtubercles apart; seta *e* 6, on ventral annulus 22, 9 apart, 5 microtubercles apart; seta *f* 14, on ventral annulus 43, 13 apart, 13 microtubercles apart. Caudal seta *h2* broken, accessory seta *h1* 3.

Type material. Holotype female (slide/position 1/1 indicated by a red circle) and fourteen females and one male paratypes, from *Stipa* sp. (Poaceae), Necochea, Province of Buenos Aires, Argentina (38° 45.10' S, 58° 45.25' W), 13 January 2012, collected by Mauro Polizzi, on five microscope slides. Holotype and 10 female paratypes on four slides deposited in the mite collection at “Embrapa Recursos Genéticos e Biotecnologia”, Brasília, DF, Brazil. One male and four female paratypes on one slide deposited at “Departamento de Entomologia, Fitopatologia e Zoologia Agrícola, Escola Superior de Agricultura ‘Luiz de Queiroz’ (ESALQ), Universidade de São Paulo”, Piracicaba, São Paulo, Brazil.

Relation to host. All specimens were collected on inner leaf blades, along midrib entire length.

Etymology. The specific designation *stipacolus* was formed as a composition between *stipa* that refers to the genus of the host plant, and the New Latin *-colus*, meaning dwelling in, inhabiting.

Molecular characterization

Three 16S and three ITS sequences of the *Aculodes stipacolus* sp. nov. were obtained. One haplotype was identified from 16S sequences (GenBank deposit No. KF648353) and three variants were obtained from the ITS sequences (GenBank deposit No. KF648354, KF648355, KF648356). The 16S sequence KF134860 of *Abacarus hystrix* (Nalepa) was incorporated in the distance analysis since it is the unique 16S sequence of an eriophyid mite in grasses different to *Aceria* available in the database. It was not possible to estimate the genetic distance with other mites of the same genus since no

sequences were available in GenBank. The distance between 16S haplotype of *A. stipacolus* (KF648353) and the predominant *A. tosichella* haplotype detected in Argentina (JQ512769) was 0.2280; and with *A. hystrix* (KF134860) it was 0.2539. The distance between the three ITS variants of *A. stipacolus* (KF648354, KF648355, KF648356) and the predominant *A. tosichella* haplotype detected in Argentina (JF960144) were respectively 0.2606, 0.2569, and 0.2587. These values can be considered as intergeneric distances in the Eriophyidae family.

***Aculodes* Keifer world species associated with grasses (Poaceae)**

A list of the 21 *Aculodes* species associated with grasses in the world is presented below. It includes information on type host, type locality, and habitus. Main morphological traits of the grass-associated *Aculodes* species are presented in Table 1.

***Aculodes agropyronis* (Keifer, 1960)**

Type host – *Agropyron smithi* Rydb., presently referred as a synonym of *Elymus smithii* (Rydb.) Gould (The Plant List 2013).

Type locality – Texas, USA.

Habitus – In young rolled leaves and on leaf plate near sheath.

***Aculodes bambusae* Kuang, 1997**

Type host – *Bambusa* sp.

Type locality – Hangzhou City, Zhejiang Province, China.

Habitus – Vagrant.

***Aculodes calamaabditus* Skoracka, 2003**

Type host – *Calamagrostis epigeios* (L.) Roth., presently the accepted name is *Calamagrostis epigejos* (L.) Roth (The Plant List 2013).

Type locality – Path in Forest, Lesna Dolina (16° 12'E, 51° 44'N), Glogow, Poland.

Habitus – Vagrants on upper leaf surface, often hiding in furrows.

***Aculodes capillaris* Skoracka, 2003**

Type host – *Agrostis capillaris* L.

Type locality – Forest path, Biedrusko near Poznan (16° 55'E, 52° 29'N), Poland.

Habitus – Vagrants on upper leaf surface, mostly near the top.

***Aculodes deschampsiae* (Sukhareva, 1972)**

Type host – *Deschampsia cespitosa* (L.) P. Beauv.

Type locality – Park of the Biological Institute, Leningrad University in old Petergof, Leningrad Region, Russia.

Habitus – Vagrant in grooves on upper leaf surface.

References – Drawings and taxonomic characterization also in Skoracka (2004).

***Aculodes dubius* (Nalepa, 1891)**

Type host – *Helictotrichon pratense* (L.) Besser ex Pilg.

Type locality – Austria? (in Amrine and Stasny 1994).

Habitus – In grooves between veins on surface of leaves; greening of flowers.

Reference – Drawings and taxonomic characterization also in Skoracka (2004).

***Aculodes festucae* Skoracka, Labrzycka and Rector, 2009**

Type host – *Festuca arundinaceae* Schreb.

Type locality – Gorski Kotar mountains, around 30 Km NE of Rijeka (14° 35.047'E, 45° 28.848'N), Croatia.

Habitus – Vagrants on upper leaf surface.

***Aculodes fulleri* (Keifer, 1966a)**

Type host – *Stipa californica* Merr. & Burtt-Davy ex Hall., presently referred as a synonym of *Stipa occidentalis* Thurb. ex S. Watson (The Plant List 2013).

Type locality – Upper King's Creek, Lassen Nat. Pk., Shasta Co., California, USA.

Habitus – The mites live in rib grooves on upper leaf surfaces.

***Aculodes holcusi* Skoracka, 2004**

Type host – *Holcus mollis* L.

Type locality – Mountain meadow with southern exposure, Przechyba, Beskid Sadecki Mts., Poland.

Habitus – Vagrant on upper leaf surfaces.

***Aculodes janboczeki* Skoracka, 2005**

Original description -Type host – *Bromus inermis* Leyss.

Type locality – Meadow near Oak Forest (48° 02.730' N, 20° 28.814' E), 580 m above sea level, Bukki Mts., Hungary.

Habitus – Vagrants on upper leaf surfaces.

***Aculodes koeleriae* Sukhareva, 1985**

Type host – *Koeleria cristata* (L.), presently referred as a synonym of *Koeleria pyramidata* (Lam.) P. Beauv. (The Plant List 2013).

Type locality – In steppe-like meadow, vicinity of Putsilovska, Pogradichnyy Distr., Maritime Territory, Primorskiy Krai, Russia.

Habitus – Vagrant on upper leaf surface; on strips between veins on upper surfaces of leaves.

***Aculodes kransnovi* Sukhareva, 1994**

Type host – *Sasa palmata* E. G. Camus.

Type locality – Batumskoy Botanical Garden, Georgia.

Habitus – Mites were found on the surface of the leaves, in the gaps between the ridges.

***Aculodes levis* Huang, 2001**

Type host – *Phyllostachys makinoi* Hayata.

Type locality – Kaohsiung, Tengchih, Taiwan, China.

Table 1. Morphological traits of species in the genus *Aculodes* Keifer associated with grasses.

Species	Number of empodium rays	Frontal lobe	Sc seta length	Procoxal shield ornamentation	Epigynum longitudinal lines; 3a seta length	Number of dorsal annuli; microtubercles shape	Number of ventral annuli; Microtubercles shape	Coxi genital region – sternal line; ornamentation; Number of coxigenital annuli
<i>A. agropyronis</i> (Keifer, 1960)	8	Short acuminate (text Keifer 1960); subtriangular, slightly rounded apically (drawing)	46	Design of clear lines: median line present on rear 1/3; admedian lines from anterior lobe base, somewhat sinuate, diverging to rear margin; submedian line I from anterior 1/4, running towards dorsal tubercle and bending laterally on approaching tubercle; submedian II from anterior lobe base, running back below tubercle; a lateral line and a few granules above coxae	11–12 longitudinal lines; 3a 27	55–60 annuli; microtubercles small, pointed, dorsally bead-like	No. not informed; microtubercles set ahead of annuli margin	Strong sternal line (subparallel line on each side of sternal line); coxal region with strong lines; 8 coxigenital annuli
<i>A. bambusae</i> Kuang, 1997	8–9	Subcircular, apically rounded (drawing)	Reaching the fourth annulus (drawing)	Median line complete (text); admedian lines complete, but interrupted in the 3/4 posterior; submedian lines complete; bifurcated and interrupted lines (discontinuous) in the rear half (drawing)	12–13 longitudinal lines (drawing)	45–48 annuli; absence of microtubercles	60–65 annuli; rounded microtubercles	Sternal line distinct; coxal region with short curved lines; 5 coxigenital annuli
<i>A. calamaabditus</i> Skoracka, 2003	7 (7–8)	Subtriangular, pointed	21 (12–28)	Median line on rear half; admedian from anterior lobe base diverging to rear margin, slightly concave in the middle; submedian lines from anterior 1/4 running towards outer sc tubercles, almost subparallel to lateral margin	13 (9–14) longitudinal lines; 3a 25 (23–25)	62 (59–79) annuli; numerous minute microtubercles along annuli margins	63 (67–76) annuli; microtubercles not as numerous as dorsal, conical, blunted top, along annuli margins	Sternal line slender; coxal region with numerous long lines and minute conical microtubercles; 6 (4–7) coxigenital annuli
<i>A. capillarisi</i> Skoracka, 2003	8 (7–8)	Subtriangular, distinct, pointed	45 (35–42)	Median line absent; admedian lines complete, from anterior lobe base, diverging to rear margin; submedian lines with conical microtubercles on rear half of shield, almost parallel to lateral margin	12 (11–13) longitudinal ridges; 3a 28 (17–33)	61 (54–60) annuli; minute, conical but with blunt top microtubercles	68 (61–71) annuli; conical, pointed microtubercles	Sternal line distinct; coxal region with numerous lines (most of them short) and numerous conical microtubercles; 5–6 coxigenital annuli
<i>A. descampsiae</i> (Sukhareva, 1972)	7–8 (in Skoracka 2004)	Pronounced, apically rounded (not acuminate) (in Skoracka 2004)	29–34 (in Skoracka 2004)	Median line on rear half, divided into two lines, at least one of them splitting anteriorly; admedian lines complete; submedian lines on rear 3/4, parallel to lateral margins; short transverse lines forming obtuse angle over tubercles bearing sc setae (in Skoracka 2004)	10–11 longitudinal lines; 3a 24–30 (in Skoracka 2004)	59–64 annuli; conical, pointed microtubercles (in Skoracka 2004)	66–75 annuli; conical, pointed microtubercles (in Skoracka 2004)	Sternal line distinct; coxal region with short, slender lines; 6 coxigenital annuli (in Skoracka 2004)

<i>A. ditubus</i> (Nalepa, 1891)	9 (in Skoracka 2004)	Triangular, pointed (in Skoracka 2004)	64–90 (in Skoracka 2004)	Median line absent; admedian lines complete, diverging from base of anterior lobe to rear margin, near shield; submedian lines I arched, with minute and conical microtubercles, on rear 2/3 of shield, connecting with admedian lines in its 1/3, with posterior fragment outside tubercles bearing <i>sc</i> seta; transverse, arched lines with minute and conical microtubercles over tubercles bearing <i>sc</i> seta (in Skoracka 2004)	9–13 longitudinal lines; 3α 31–45 (in Skoracka 2004)	54–64 annuli; conical, minute microtubercles (in Skoracka 2004)	74–83 annuli; conical, pointed microtubercles (in Skoracka 2004)	Sternal line slender; coxal region with subrounded microtubercles and numerous lines with minute microtubercles; 5–6 coxigenital annuli (in Skoracka 2004)
<i>A. festucae</i> Skoracka et al., 2009	7	Subtriangular, acuminate /slightly curved, enlarged base	21 (18–24)	Median line on the rear 2/3; admedian lines complete, sinuous, diverging to rear margin; submedian lines begin on the rear 2/3	10 (10–13) longitudinal lines; 3α 26 (20–28)	57 (51–57) annuli; small bead-like microtubercles	63 (58–63) annuli; small bead-like microtubercles	Sternal line distinct; coxal region with dashes, hipostomio base densely granulated; 7 coxigenital annuli
<i>A. fuller</i> (Keifer, 1966a)	7	Somewhat narrow and acute, a thin anterior projection visible in side view	32	Median line present on rear 1/3; admedian line complete, gentle sinuous, gradually diverging; submedian lines I a short line, subparallel to admedian, in shield centre; submedian lines II beginning at about 1/2 and arching back to rear margin below dorsal tubercle, somewhat granular; lateral line from side of anterior lobe to partial rings below dorsal tubercle	About 14 longitudinal lines, some transverse lines of granules at base; 3α 33	44 annuli; small, bead like, close together, hardly pointed, or rounded microtubercles	48–50 annuli; microtubercles tending to be ahead of ring margins, more pointed than dorsal	Strong sternal line; coxal region with lines of dashes and granules; 7 coxigenital annuli
<i>A. holcusi</i> Skoracka, 2004	8	Triangular, pointed	59 (58–65)	Median line on rear half, below median lines short lines forming a V-shaped figure; admedian lines complete, diverging to lateral margin of shield; submedian lines I on rear half, parallel to admedian; submedian lines II arched, connecting with admedian in its 1/4; subrounded and conical microtubercles and dashes	12 (11–12) longitudinal lines; 3α 40 (38–40)	50 (49–51), dorsal microtubercles irregularly distributed, large, subrounded, set along annuli margins	63 (60–63); ventral microtubercles minute, conical, slightly pointed, ahead from annuli margins	Sternal line slender; coxal region with wavy lines and dashes; 5 (5–6) coxigenital annuli
<i>A. koelerviae</i> Sukhareva, 1985	6–7	Subtriangular, apically acuminate (drawing)	43 (38–45)	Admedian lines very weak; submedian lines II following lateral margins	8–10 longitudinal lines; 3α 17 (15–20)	60 (57–66) annuli; absence of microtubercles, but presence of unevenly scalloped margin	55 (53–58) annuli; small microtubercles	Sternal line distinct; coxal region smooth (drawing); 3–4 coxigenital annuli (text), 8 (drawing)
<i>A. janboczebi</i> , Skoracka, 2005	7 (7–8)	Pointed (text), pronounced, triangular (drawing)	20 (18–23)	Median line absent; admedian lines complete and parallel; submedian lines I short, on rear half, parallel to admedian; submedian lines II as short as submedian I, sinuous; ocellar fields in the lateral area	13 (11–15) longitudinal lines; 3α 15 (14–19)	57 (55–64) annuli; conical and bead-like microtubercles	72 (63–75) annuli; conical and bead-like microtubercles	Sternal line slender; coxal region with numerous, minute microtubercles and dashes (set round setae tubercles); 5 (5–7) coxigenital annuli

(Continued)

Table 1. (Continued).

Species	Number of empodium rays	Frontal lobe	Sc. seta length	Prodorsal shield ornamentation	Epigynum longitudinal lines; 3a seta length	Number of dorsal annuli; microtubercles shape	Number of ventral annuli; Microtubercles shape	Coxi genital region – sternal line; ornamentation; Number of coxigenital annuli
<i>A. krasnovi</i> Sukhareva, 1994	5–6	Subtriangular, slightly acuminate apically (drawing)	Reaching ninth annulus (drawing)	Median line absent; admedian lines complete; submedian lines on 4/5 rear; sublateral lines complete (drawing)	6 short, interrupted longitudinal lines	46 (44–48) annuli; absence of microtubercles (drawing)	48–50 annuli; absence of microtubercles (drawing)	Sternal line slender; coxal region smooth; 8–9 coxigenital annuli (drawing)
<i>A. levis</i> Huang, 2001	5	Subcircular, apically rounded (drawing)	19	Dashed lines; median line complete; median and admedian lines subparallel; admedian and submedian lines not visible on anterior 1/4 shield	Longitudinal lines absent, coverflap smooth; 3a 10	56 annuli; minute, elongated microtubercles (drawing)	81 annuli; rounded microtubercles, larger than dorsal (drawing)	Sternal line indistinct; coxal region with small granules; 2 coxigenital annuli (drawing)
<i>A. mckenziei</i> (Keifer, 1944)	7 (Keifer 1944); 9 (in Skoracka 2004)	Narrow and pointed (text Keifer 1944), subtriangular (drawing); large, pointed (in Skoracka 2004)	43 (Keifer 1944); 47–64 (in Skoracka 2004)	Longitudinal lines curved to the rear, granules to the rear and side (Keifer 1944); median line on rear half; admedian lines complete, diverging from the base of frontal lobe to rear margin; submedian lines I subparallel to admedian lines, running laterally in front to sc tubercles; submedian lines II on rear 3/4, subparallel to lateral margin; large, conical pointed microtubercles on rear half (in Skoracka 2004)	12 longitudinal lines; 3a 25 long (in Keifer 1944). 11–15 longitudinal lines; 3a 38–50 (in Skoracka 2004)	60–71 annuli; acuminate microtubercles (in Keifer 1944); numerous, conical and pointed microtubercles (in Skoracka 2004)	65 annuli (in Keifer 1944); 69–81 annuli (in Skoracka 2004), conical and pointed microtubercles smaller than dorsal (in Skoracka 2004)	Anterior coxae broadly contiguous (in Keifer 1944), sternal line slender (in Skoracka 2004); coxal region with numerous lines and conical microtubercles; 5–8 coxigenital annuli
<i>A. mongolicus</i> Skoracka & Shi, 2001	8 (7–8)	Pronounced, elongate and pointed (text), triangular (drawing)	45 (42–45) (text), reaching sixteenth annulus (drawing)	Median line present in the posterior half; admedian lines complete, parallel to each other on the anterior region, diverging to lateral margins in the posterior half; submedian lines II subparallel to admedian, running laterad in front of sc tubercles; submedian lines II beginning from 1/3 of the submedian lines I and forming bows reaching posterior margin (text); sparse conical granules on posterior half, along some lines, and also on the antero-lateral margin (drawing)	10 (10–11) longitudinal lines; 3a 38 (38–48)	64 (56–62) annuli; triangular, minute, close to each other microtubercles	74 (61–70) annuli; triangular, but more pointed and larger than dorsal microtubercles	Sternal line strong; coxal region with numerous, short lines, or triangular microtubercles along lines; 8 (5–7) coxigenital annuli
<i>A. multiricavus</i> Skoracka, 2004	9	Triangular, pointed	27 (26–32), reaching tenth annulus	Median line on rear half; admedian lines complete, from anterior lobe diverging to lateral margin; submedian lines I short, on rear half, parallel to admedians; submedian lines II beginning on 4/5 rear, parallel to lateral margins; numerous triangular cavities on shield	13 (12–13) longitudinal lines; 3a 34 (32–35)	60 (58–60) annuli; minute, conical, and pointed microtubercles	66 (62–67) annuli; minute, conical, and pointed microtubercles	Sternal line distinct; coxal region with lines (coxae I) and dashes and conical microtubercles (coxae II); 6 (5–6) coxigenital annuli

<i>A. neglectivagrans</i> Skoracka, 2005	8 (7–8)	Triangular, pointed	42 (40–49)	Median line on rear half; admedian lines complete, parallel to each other; submedian lines I on rear 1/3, parallel to admedians; submedian lines II in rear 3/4, parallel to lateral margin; dashes on rear half, and between submedians II and lateral margins	15 (11–15) longitudinal lines, coverflap densely micropunctate; 3 <i>a</i> 24 (21–29)	69 (54–70) annuli; minute, conical and pointed microtubercles	75 (57–81) annuli; bead-like microtubercles	Sternal line distinct; coxal region with longitudinal lines and dashes; 4 (4–6) coxigential annuli
<i>A. ponticus</i> Sukhareva, 1986	6–7	Subtriangular, heavy dorsal projection about 8 long	15 (14–16)	Admedian lines distinct, extended from <i>sc</i> tubercles to anterior margin	11–12 longitudinal lines; 3 <i>a</i> 25 (18–30)	60 (58–68) annuli; minute microtubercles	55–58 annuli; minute microtubercles	Sternal line distinct (drawing); coxal region smooth (drawing); 5–6 coxigential annuli
<i>A. stipacolis</i> sp. n. Alemandri & Navia	9	Subtriangular, acuminate /slightly curved, enlarged base	22 (21–29)	Median line short, on the posterior area; admedian lines complete, subparallel; lateral setae complete, delimiting prodorsal shield, laterally finely granulated	10 (10–12) longitudinal lines; 3 <i>a</i> 12 (10–19)	71 (64–74) annuli; rounded, small microtubercles	62 (59–67) annuli; bead-like, small microtubercles	Sternal line distinct; coxal region with short irregular lines; 5 coxigential annuli
<i>A. stoloniferae</i> Skoracka, 2005	8	Triangular, small, pointed	50 (50–62)	Short submedian lines on posterior 1/4, laterad of <i>sc</i>	4 (3–6) longitudinal lines; 3 <i>a</i> 26 (24–29)	75 (64–75) annuli; subrounded microtubercles	72 (55–72) annuli; conical microtubercles	Sternal line distinct; coxal region with few dashes; 5 (4–5) coxigential annuli
<i>A. sybarici</i> Skoracka et al., 2009	7	Elongate, acuminate	30 (26–30)	Median lines absent; admedian lines complete, sinuous, from anterior lobe diverging to rear margin; submedian lines I short on rear part of shield, parallel to admedians, reaching short transversal arched lines over <i>sc</i> tubercles; submedian lines II begin in front of fifth and subparallel to laterad margin of shield; microtubercles present on rear surface of the shield, between admedian lines present up to rear half	12 (9–12) longitudinal lines; 3 <i>a</i> 22 (19–26)	58 (58–61) annuli; small, bead-like microtubercles	67 (60–67) annuli; small, bead-like microtubercles	Sternal line distinct; coxal region with regular dashes; 5–6 coxigential annuli
<i>A. tsukushiensis</i> Xue, Song & Hong, 2010	7	Triangular, acuminate (drawing)	18 (16–20)	Median lines discontinuous, admedian and submedian lines complete and subparallel, granules on lateral area	10 longitudinal lines; 3 <i>a</i> 20 (20–21)	61 (60–62) annuli; spiny microtubercles	64 (63–65) annuli; spiny microtubercles	Sternal line distinct; coxal region with short lines; 8 coxigential annuli (drawing)

Habitus – Vagrant on lower leaf surface. No damage observed.

***Aculodes mckenziei* (Keifer, 1944)**

Type host – *Leymus triticoides* (Buckl.) Pilg.
Type locality – Sacramento, California, USA.
Habitus – The mites occur in upper surface, leaf furrows. They cause some browning.
Reference – Drawings and taxonomic characterization also in Skoracka (2004).
Obs – Sukhareva (1981) considered two subspecies for this taxon, *A. mckenziei* subsp. *brevisetus* and *A. mckenziei* subsp. *trivialis*.

***Aculodes mongolicus* Skoracka & Shi, 2001**

Type host – *Hordeum brevisubulatum* (Trin.) Link.
Type locality – Arkhangy Aymag, Horgo Terhiyn Tsagaan Nuur National Park, Central Mongolia.
Habitus – Mites are vagrants on upper leaf surface.

***Aculodes multiricavus* Skoracka, 2004**

Type host – *Bromus inermis* Leyss.
Type locality – Xerothermic sward, Owczary, Poland.
Habitus – Vagrant on upper leaf surfaces.

***Aculodes neglectivagrans* Skoracka, 2005**

Type host – *Calamagrostis neglecta* (Ehrh.) Gaertn., Mey et Schreb., presently referred as a synonym of *Calamagrostis stricta* (Timm) Koeler (The Plant List 2013).
Type locality – Littoral dune, 14 m elev., 10 km W of Kuzrjeka Village, near Turij Cape, S. of Umba, White Sea Coast, Kola Peninsula, Russia. (66° 35.722' N, 34° 42.965' E)
Habitus – Vagrants on upper leaf surfaces.
Reference – Skoracka and Pacyna (2005).

***Aculodes ponticus* Sukhareva, 1986**

Type host – *Eremopyrum distans* (Koch) Nevski.
Type locality – Near Kabardinka Village, Krasnodarski Krai, Russia.
Habitus – Deep grooves between veins on surface of the leaves.

***Aculodes stoloniferae* Skoracka, 2005**

Type host – *Agrostis stolonifera* L. subsp. *straminea* (Hartm.) Tzvel.
Type locality – Taiga, near a stony beach, near Turij Cape, S. of Umba, White Sea. Coast, (66° 37.822' N, 34° 27.345' E) Kola Peninsula, Russia.
Habitus – Vagrants on upper leaf surfaces.
Reference – Skoracka and Pacyna (2005).

***Aculodes sylvatici* Skoracka, Labrzycka and Rector, 2009**

Type host – *Brachypodium sylvaticum* (Huds.) P. Beauv.

Type locality – Velebit mountains, around 24 Km SE of Senj (15° 02.830'E, 44° 56.412'N), Croatia.
Habitus – Vagrants on upper leaf surface.

***Aculodes tsukushiensis* Xue, Song, Hong, 2010**

Type host – *Elymus tsukushiensis* Honda var. *transiens* (Hack.) Osad.
Type locality – Zhouzhi County, Shaanxi Province, P.R. elevation 500 m, (34°03'54"N, 108°19'22"E), China.
Habitus – Vagrant on leaf surfaces. No damage to the host was observed.

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