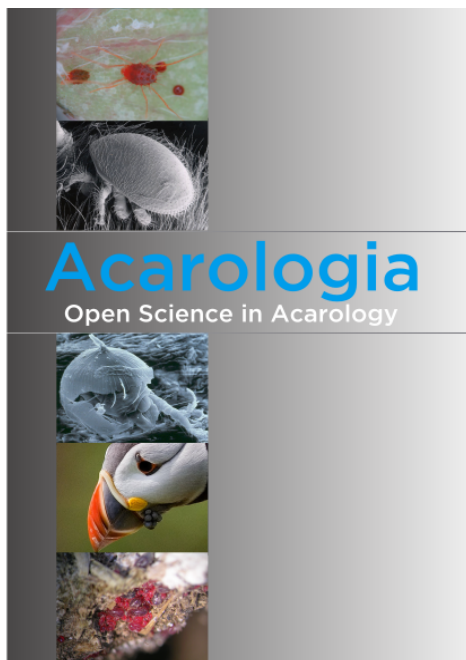




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# *Agistemus collyerae* (Acari: Trombidiformes: Stigmaeidae) in South Africa: first record, introduction pathways and a re-description including additional life stages

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## ABSTRACT

*Agistemus collyerae* Gonzáles-Rodrigues (Stigmaeidae) is reported for the first time in South Africa on grapevines in the Western Cape Province. We investigate a possible introduction pathway of this mite as a contaminant on imported agricultural goods. Based on South African interception data, we report new country records of *A. collyerae* in the USA, Chile, Yemen, Spain and France. We re-describe it based on the South African specimens, including first-time descriptions of the male and deutonymph. Female specimens have shorter dorsal setae than those documented in the original descriptions, but we do not consider this enough evidence to justify a new species.

**Keywords** grapevine, predatory mite, *Agistemus collyerae*, South Africa, new record

**Zoobank** <http://zoobank.org/96B9114B-6665-400A-9D00-465FF77F3974>

## Introduction

Many mite species have been transported and introduced beyond their natural ranges, due in part to the ease with which they can accidentally be transported on internationally traded commodities (Navajas & Ochoa 2013). Many agricultural pest species have been introduced this way, but less is known about the introduction of non-native predators in agricultural systems (Palevsky *et al.* 2013; Saccaggi *et al.* 2016).

In 2015 South African authorities detected a newly introduced pest of quarantine status on grapevine, *Brevipalpus lewisi* (Trombidiformes: Tenuipalpidae) (Saccaggi *et al.* 2017). During the delimiting survey that followed this detection, a non-native predator, *Agistemus collyerae* (Trombidiformes: Stigmaeidae) was found on some samples around the same location where *B. lewisi* was first detected. The new discovery of two non-native mite species raised questions as to the introduction pathways of both mites. We investigated the presence and identity of mites on agricultural imports to South Africa. During this investigation, we noted differences in the specimens examined and found that the male and deutonymph of *A. collyerae* had not been described.

*Agistemus collyerae* falls within the family Stigmaeidae, which consists of 577 species in 34 genera, with *Agistemus* the third largest genus with 87 species, after *Stigmaeus* (133 species) and *Eustigmaeus* (124 species) (Fan *et al.* 2016). Stigmaeidae are present in all geographical regions, with most species described from the Palaearctic (289 species). Africa has only 45 Stigmaeidae species described, with most of those stemming from South Africa (Fan *et al.*

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2016). Members of this family are considered economically important predators of mites and small insects (Santos & Laing 1985; Gerson *et al.* 2003; Fan and Zhang 2005; Fan & Flechtmann 2015).

*Agistemus collyerae* González-Rodríguez 1963 was described from New Zealand, where it is assumed to be native (González-Rodríguez 1963). It is also recorded from Australia, Iran, Portugal, Turkey and Italy (Fan *et al.* 2016). It has been recorded from various cultivated and wild plants, and is known to feed on *Tetranychus lambi* (Tetranychidae) and *Aculus fockeui* [syn. *Aculus cornutus*] (Eriophyidae) (Fan and Zhang 2005).

Here we report *Agistemus collyerae* (Acari: Trombidiformes: Stigmaeidae) for the first time from South Africa. Possible introduction pathways of *A. collyerae* into South Africa are investigated and discussed. We re-describe the species based on South African specimens, including for the first time a description of the male and deutonymph.

## Materials and methods

### Plant and mite collection

Plant samples were collected from grapevine (*Vitis vinifera*, various cultivars) throughout South Africa as part of a nation-wide survey from March 2015 – November 2016 (for more details see Saccaggi *et al.* 2017). Shoots and leaves from each plant were collected, sealed in plastic bags, and transported to the laboratory for microscopic inspection. Plant parts were examined under a stereomicroscope for the presence of mites. Any mites found were preserved in 70% ethanol until clearing in lactic acid and mounting on glass slides in PVA mounting medium, following the general protocols outlined in Krantz & Walter (2009).

### Specimen examination

Slide-mounted specimens were identified by EU using a compound microscope with phase contrast and measured with a Zeiss Image Analysing System. All measurements are in micrometers (µm). Specimens were identified as *Agistemus collyerae* using the keys to Raphignathoidea and Stigmaeidae provided in Fan and Zhang (2005) and Fan *et al.* (2016). Specimens were compared to the original description of *Agistemus collyerae* by González-Rodríguez (1963) and re-descriptions by Fan and Zhang (2005) and Yeşilayer and Çobanoğlu (2013). Specimens from South Africa, New Zealand, Australia, Italy, USA, Chile, Yemen, Spain and France were examined and compared to each other.

### Examination of introduction pathways

The introduction pathways of *Agistemus collyerae* to South Africa were investigated by searching for records of mites detected on imported plant products. We examined the interception databases maintained by the Plant Health Diagnostic Division of South Africa's Department of Agriculture, Forestry and Fisheries (DAFF). These databases contain records of all organisms (mites, insects, nematodes, fungi, bacteria, viruses and phytoplasmas) detected on imported plant material by DAFF since 2004 (Saccaggi and Pieterse 2013). Additional *A. collyerae* specimens intercepted from other countries before 2004 were found in the South African National Collection of Arachnida (NCA), housed at the Agricultural Research Council (ARC) at Roodeplaat, Pretoria, Gauteng Province.

Specimens of *A. collyerae* detected on imported material were compared to published descriptions as described above, to specimens from New Zealand, and to the specimens collected in this study.

## Results and discussion

New species record: *Agistemus collyerae*

This is the first report of *Agistemus collyerae* (Trombidiformes: Stigmaeidae) from grapevine in South Africa. It was detected on three samples collected in March and April 2015 in the Western Cape Province. It was found in association with *Brevipalpus lewisi*, *B. obovatus* (Trombidiformes: Tenuipalpidae) (Saccaggi *et al.* 2017) and *Colomerus cf. vitis* (Trombidiformes: Eriophyidae), on which it is assumed to be feeding.

### Re-description

#### Super Family Raphignathoidea Kramer

#### Family Stigmaeidae Oudemans

#### Genus *Agistemus* Summer

#### *Agistemus collyerae* González-Rodrigues (Figs. 1-8)

*Agistemus collyerae* González-Rodrigues, 1963: 349; 1965: 34; Fan & Zhang, 2005: 41; Ferreira *et al.*, 2007: 56; Rahmani *et al.*, 2012: 105; Beyzavi *et al.*, 2013: 397; Yeşilayer and Çobanoğlu, 2013: 96; Fan *et al.*, 2016: 31. *Zetzellia collyerae* (González-Rodrigues) Wood, 1967: 131.

Material examined — Eight females, two males and one deutonymph from grapevine, Wellington, 16 April 2015; one female from Malmesbury, 16 March 2015; and one female from Riebeek West, 17 April 2015. All these locations are within the Western Cape Province, South Africa. Specimens are deposited in the National Collection of Arachnida (NCA) in Pretoria, South Africa. Measurements are given in Table 1. Other features are described below.

Diagnosis (Female) — Dorsal shields reticulated, setae *c1* and *d1* shorter than distances to setae *d1* and *e1*, respectively; setal formulae of genua 3-0-0-0, femora 4-4-2-2 and tibiae 6-6-6-4, coxa IV with one seta.

**Female** (Figures 1-5, Table 1) (n=10 for qualitative characters) — Idiosoma oval.

Dorsum (Figures 1-2) — Dorsal shields (fig 1-2A) covered with thick-walled reticulations, each cell with about 11-14 vacuoles; setae short, barbed (fig 2B) and gradually becoming longer caudally; prodorsum with one pair of eyes and one pair of postocular bodies. Setae *c2* are situated on small spindle-shaped platelets.

Gnathosoma (Figures 3B-C) — Subcapitulum (fig 3C) with 2 pairs of setae (*m* and *n*) and two pairs of oral setae (*or1* and *or2*). Chelicerae free, with movable digits. Palp six-segmented (fig 3B); palp tarsus with four simple setae, one solenidion, one tridentate and one simple eupathidium; palp tibia with two smooth setae, one stout claw and one seta-like accessory claw; palp genu with one long simple seta; palp femur with two serrate setae; palp trochanter without setae and palp coxa with a small spine.

Venter (Figure 3A) — Covered with striae with three ventral setae (*1a*, *3a* and *4a*). Anogenital area (fig 3A) with two pairs of aggenital setae (*ag1-2*), on a smooth horse-shoe like area, one pair of very long genital setae (*g*) stretching past setae *ps3* and three pairs of anal setae (*ps1-3*), latter all smooth except for *ps3*.

Legs (Figures 4-5) — Setal formulae of leg segments: coxae 2+*elcp*-1-2-1, trochanters 1-1-1-1, femora 4-4-2-2, genua 3 (including *k*)-0-0-0, tibiae 5+1 $\varphi$  - 5+1 $\varphi$  - 5+1 $\varphi$  - 4, tarsi 11+1 $\omega$  - 9+1 $\omega$  - 7+1 $\omega$  - 7.

**Male** (Figures 6-8, Table 1) (n=2) — Idiosoma oval, similar to female.

Dorsum (Figure 6A) — All dorsal shields reticulated with setae short and barbed with setae *f* longest. Median shield with six pairs of setae, *f* included.

Gnathosoma (Figure 6C) — Like that of female but more robust. Palp femur with a spur on inner margin.



**Figure 1** Female *Agistemus collyerae*: Dorsal view (photo).

Venter (Figure 6B) — Like that of the female except that setae *ag2* and *g* are absent.

Legs (Figures 7-8) — Leg chaetotaxy similar to that of the female, except for tarsi: 11+1 $\omega$ +1 $\omega$  – 9+1 $\omega$ +1 $\omega$  – 7+1 $\omega$  – 7+1 $\omega$ .

**Deutonymph** (Table 1) (n=1) — Idiosoma oval, similar to female.

Dorsum — Dorsal shields weakly reticulate, prodorsum with eyes and postocular bodies. Dorsal setae also barbed and gradually become longer caudally.

Gnathosoma — Like that of female.

Legs — Leg chaetotaxy like that of the female, except for trochanters 1 – 1 – 1 – 0.

Remarks — The South African specimens resemble the original descriptions and re-descriptions in most respects, except for the lengths of the dorsal setae and the leg solenidia (see Table 1). The lengths of dorsal setae in the published descriptions are 32 – 46  $\mu$ m in González-Rodríguez

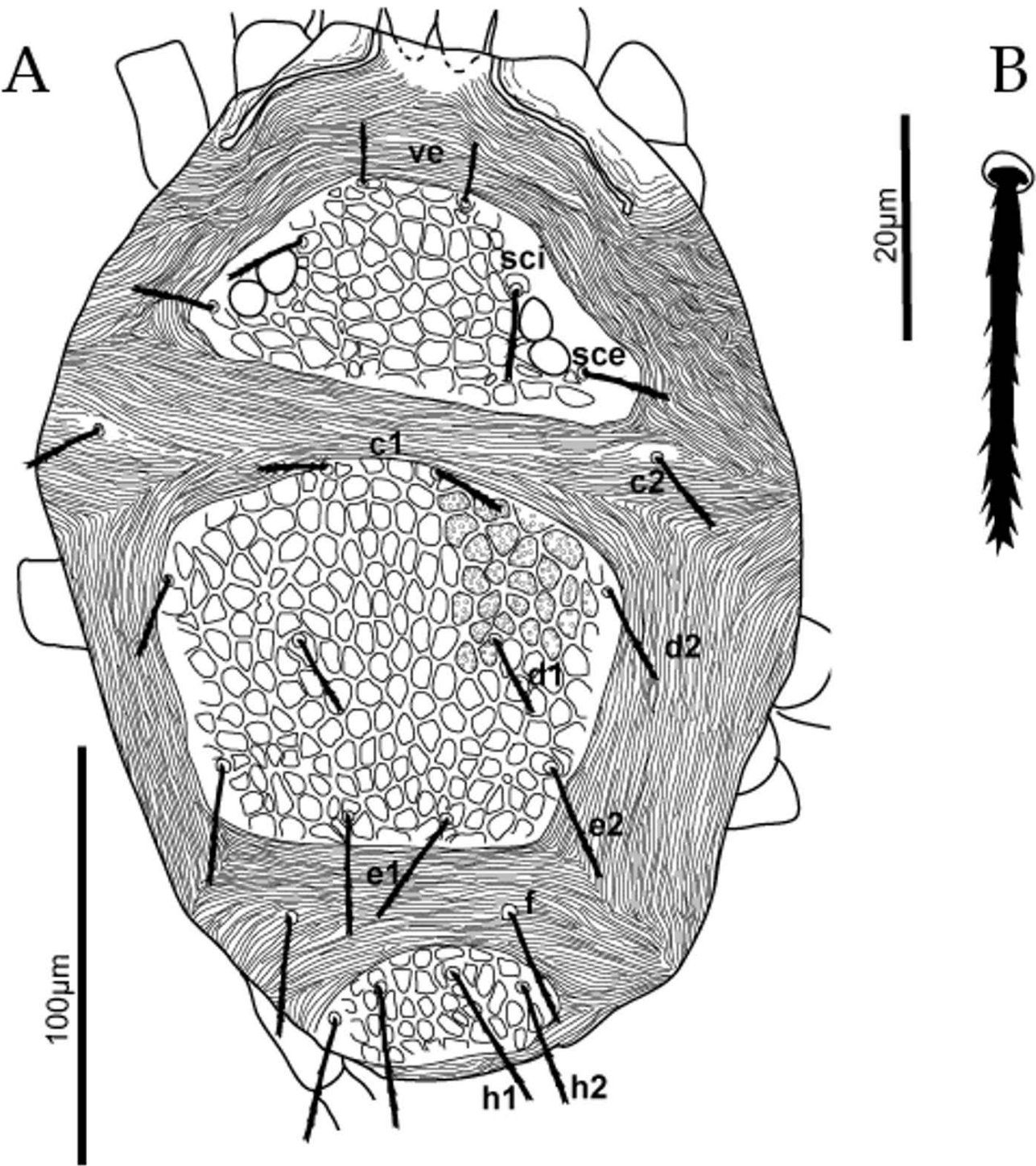
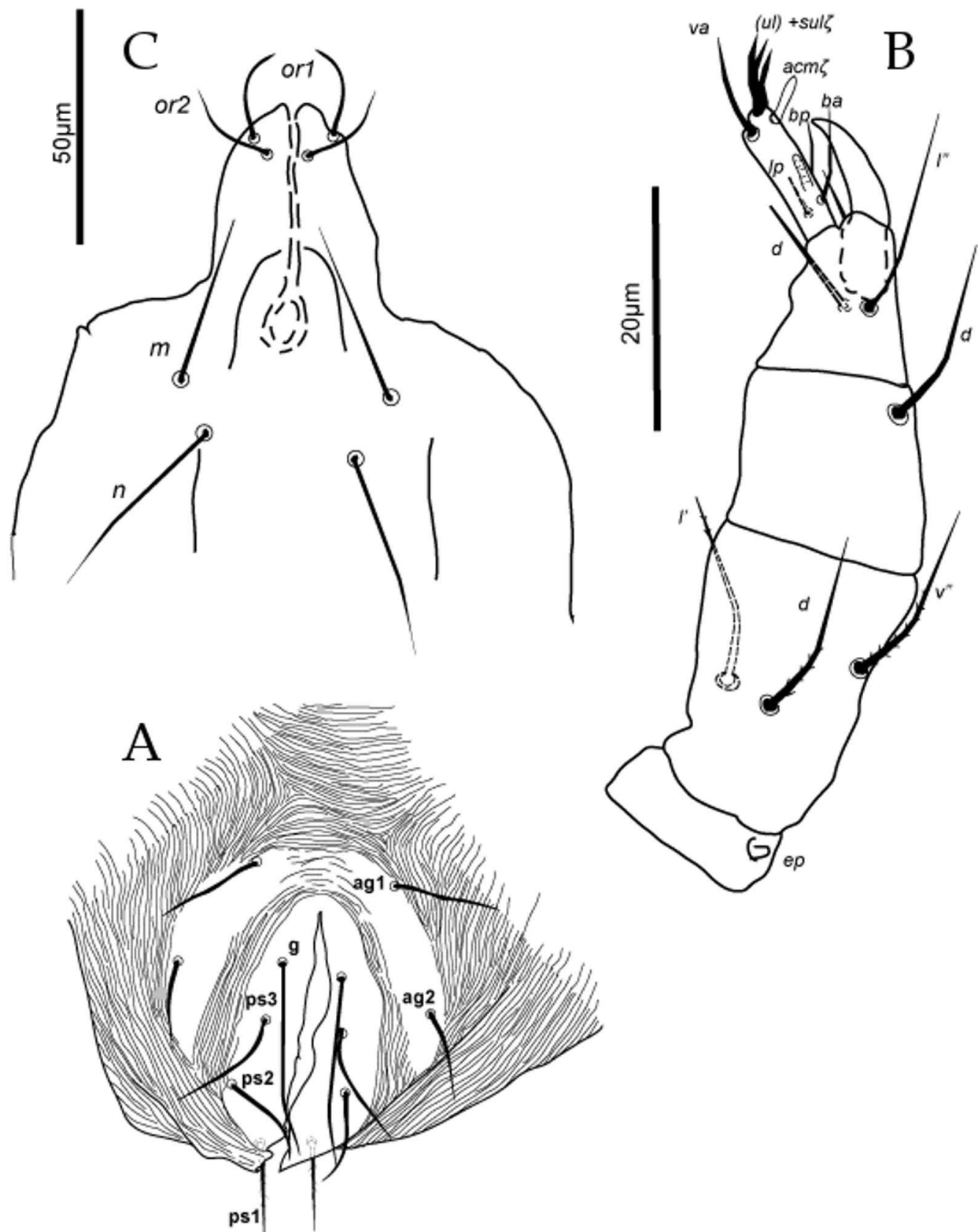
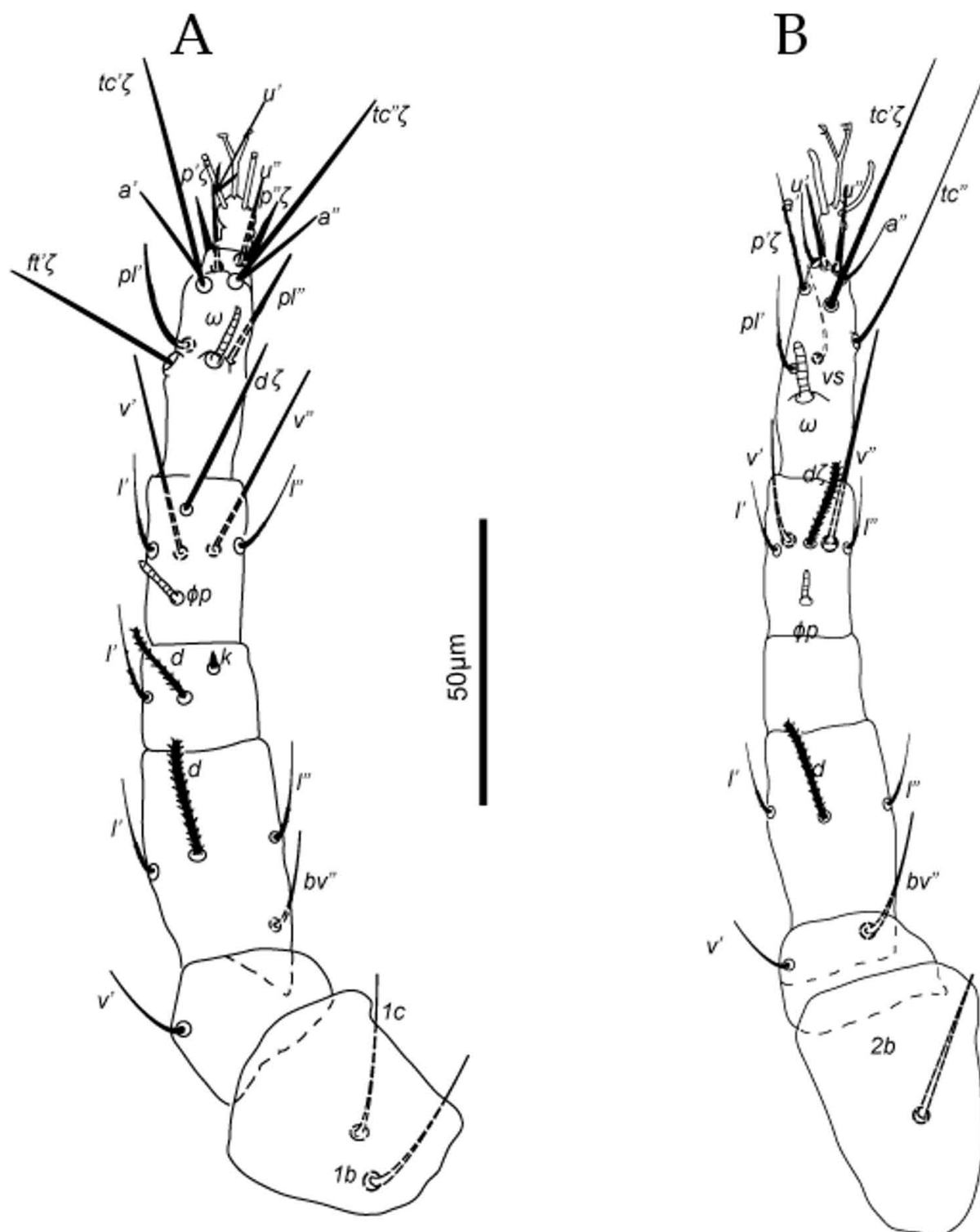


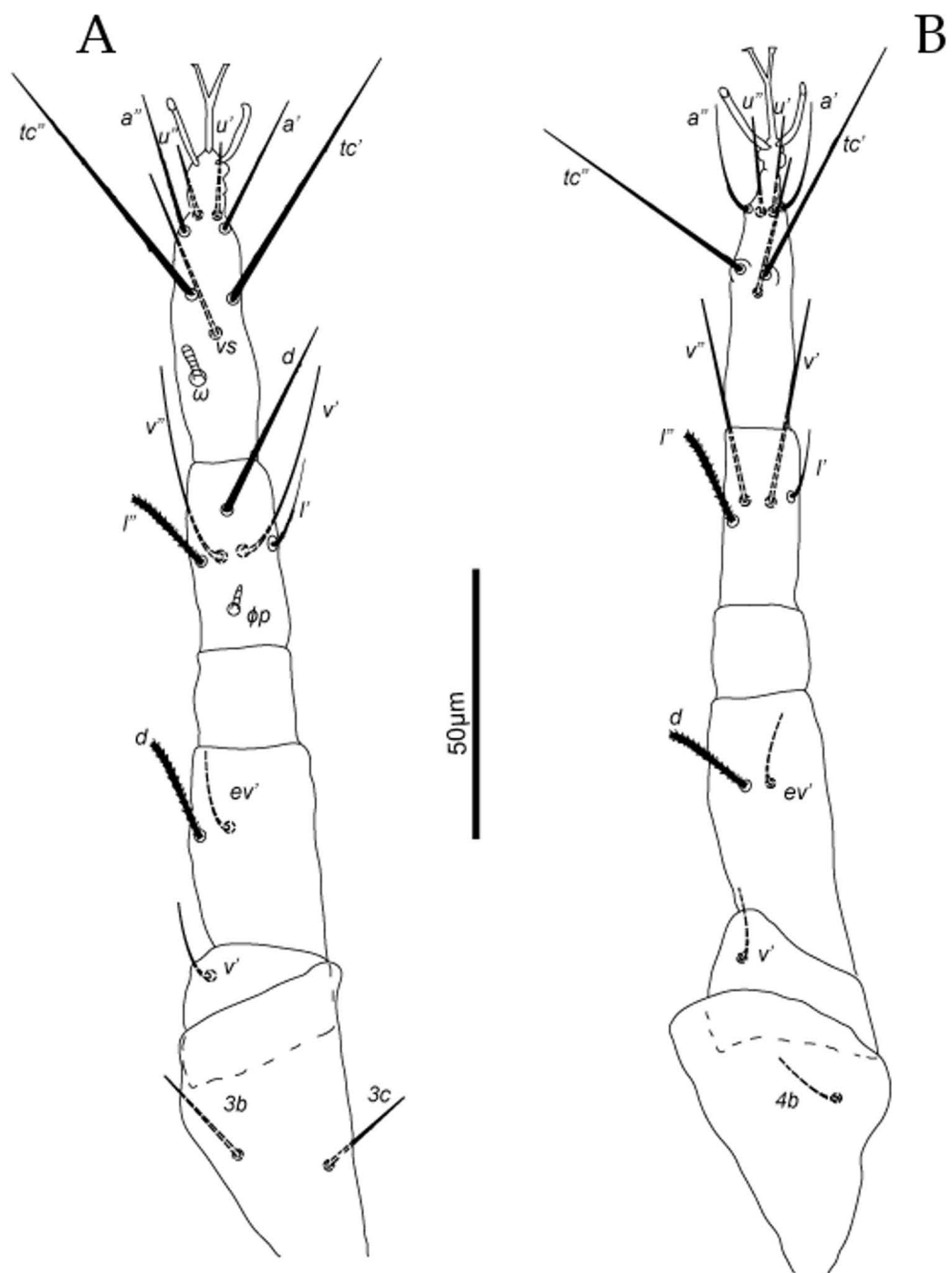
Figure 2 Female *Agistemus collyerae*: A – dorsum; B – dorsal setae.



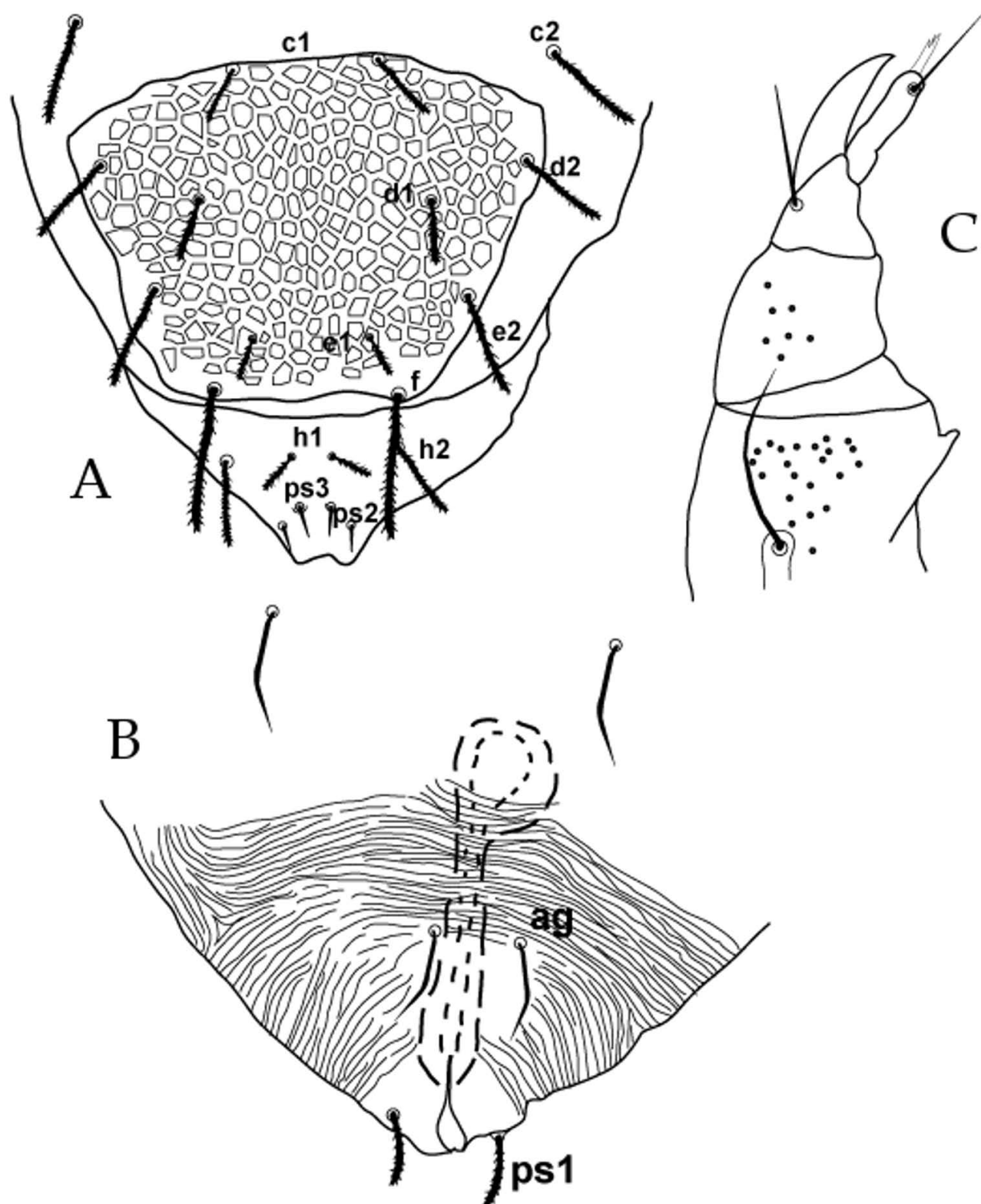
**Figure 3** Female *Agistemus collyerae*: A – venter; B – palp; C – subcapitulum.



**Figure 4** Female *Agistemus collyerae*: A – leg I; B – leg II.



**Figure 5** Female *Agistemus collyerae*: A – leg III; B – leg IV.



**Figure 6** Male *Agistemus collyerae*: A – dorsum; B – venter; C – gnathosoma.

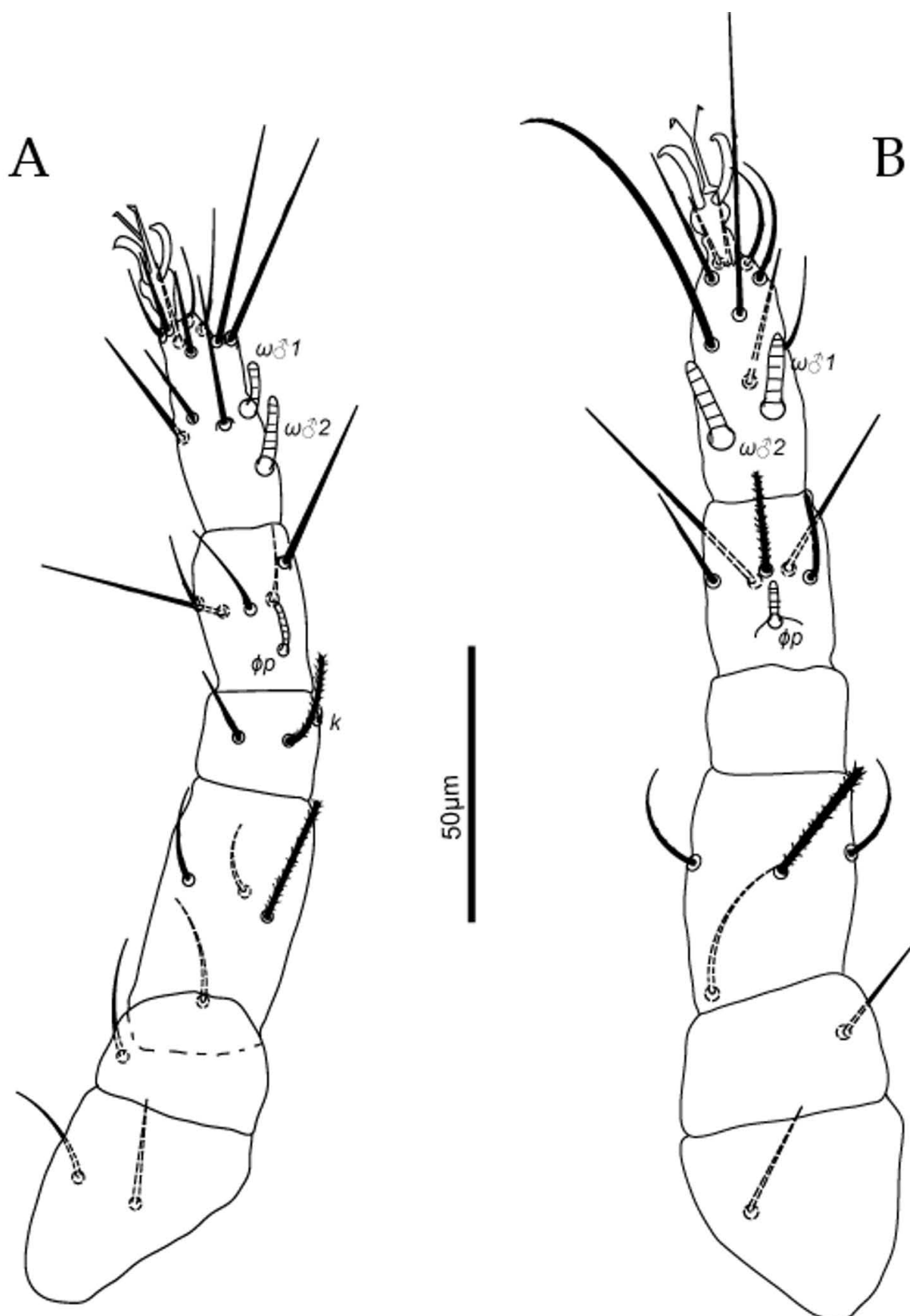
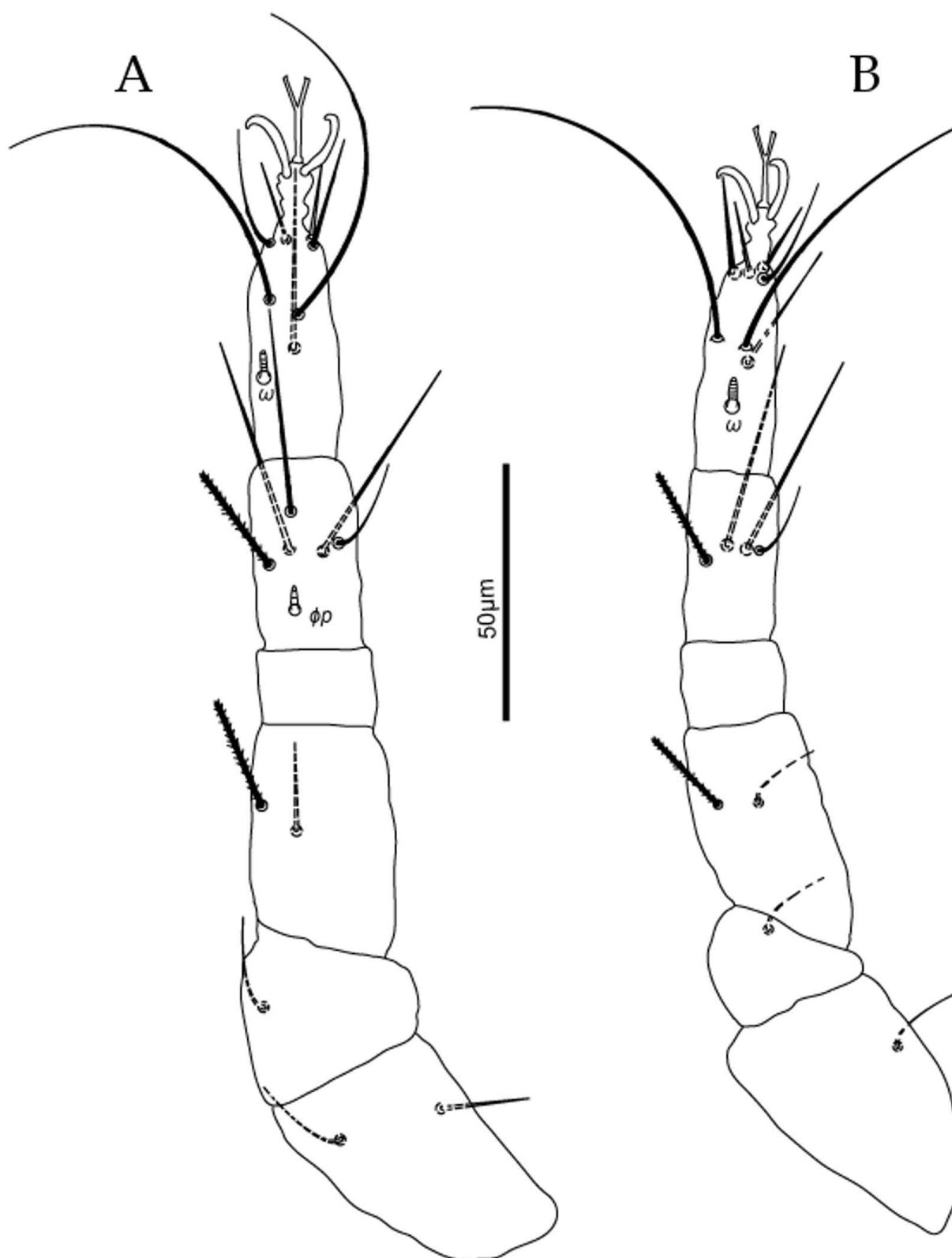


Figure 7 Male *Agistemus collyerae*: A – leg I; B – leg II.



**Figure 8** Male *Agistemus collyerae*: A – leg III; B – leg IV.

(1963), 28 – 50  $\mu\text{m}$  in Fan and Zhang (2005) and 30 – 46  $\mu\text{m}$  in Yeşilayer & Çobanoglu (2013). Specimens from the native range in New Zealand, measured in this study (Table 1), have dorsal setal lengths from 24 to 46  $\mu\text{m}$ . Except for *h2*, the lengths of the dorsal setae in the South African specimens are markedly shorter than the same setae in the published descriptions or the New Zealand specimens, with ranges usually not overlapping. The dorsal setae of the South African specimens range from 15 to 38  $\mu\text{m}$ . However, the leg chaetotaxy, which is considered unique for this species (González-Rodrigues 1963, 1965), is identical to the descriptions. Additionally, the setal ratios, dorsal shields ornamentation and anogenital area are similar, and we do not consider the difference in setal and solenidia lengths enough evidence to justify a new species. This is also the first time that a male and deutonymph are described. The male is unique in having a spur on the inner margin of the palp femur and apparently having only one pair of ag setae instead of two pairs as in the female.

## Introduction pathways

From analysis of the records kept by DAFF and the NCA, eighteen detections of *A. collyerae* on imported agricultural goods have been recorded, with the first recorded detection in 1991. These have not been reported previously, as interception of non-quarantine species (such as predatory mites) is usually not considered to be of economic concern (Saccaggi *et al.* 2016). Table 2 shows a list of these detections. In addition to detections on material from Australia and Italy, *A. collyerae* was also detected from the USA, Chile, Yemen, Spain and France, countries where it has previously not been known to occur. Thus this paper also serves to document the presence of *A. collyerae* in these countries. The unreported presence of *A. collyerae* in these countries is not unusual, as new introductions of small or cryptic organisms (such as mites) are difficult to detect and therefore often go unnoticed. Usually such introductions are noticed either because of damage to a crop (Navia *et al.* 2010) or during a routine survey, as was the case here. The introduction data and pathway of entry of *A. collyerae* to South Africa is unknown, but it is likely that it entered as a contaminant on agricultural goods, as is common for terrestrial invertebrates (Faulkner *et al.* 2015).

When these intercepted *A. collyerae* specimens were compared to published descriptions and specimens from South Africa and New Zealand, the female dorsal setae lengths were more similar to those of the South African specimens, 15 – 40  $\mu\text{m}$  in length, than to the New Zealand specimens or published descriptions. The males and deutonymphs intercepted were also comparable to those newly described from the South African samples.

We initially suspected that this discrepancy in female dorsal setal lengths may have been due to differences in methods of measurements used in the published descriptions and the current study. However, after obtaining specimens from the native range in New Zealand and measuring them ourselves, it seems that this may reflect a true difference between populations. If this is the case, it suggests that at least two lineages of *A. collyerae* exist, one restricted to New Zealand and the other spread globally to other countries. It is plausible that a strain or race of *A. collyerae* with these shorter setal lengths was transported out of New Zealand to a secondary country, which then acted as a “distribution hub” for the species to spread to other countries. It is increasingly recognised that this type of global spread of a species, sometimes termed the “bridgehead effect”, is extremely common, and that many exotic species populations are a mixture of introductions and back-introductions from different origins (Garnas *et al.* 2016). Further investigation into possible morphological similarities, possible transport routes and population genetic linkages would be necessary to investigate this possibility.

The frequency with which non-quarantine organisms (such as predatory mites) are present on imported goods is unknown. It is highly likely that live specimens of non-quarantine mites are able to enter a country relatively easily, increasing the chances of establishment. The ecological and economic effects of introducing predatory arthropods into a novel region are poorly studied. In many cases they are seen as benign or even beneficial introductions,

**Table 1** Measurements of *Agistemus collyerae* collected from South Africa, and comparison with New Zealand female specimens. All measurements are lengths unless otherwise noted. Leg length was measured from coxa to tip of empodium. Measurements are given in  $\mu\text{m}$ . SA = South Africa; NZ = New Zealand.

|                               | NZ female<br>(n=6) | SA female<br>(n=8) | SA male<br>(n = 2) | SA deutonymphs<br>(n=1) |
|-------------------------------|--------------------|--------------------|--------------------|-------------------------|
| <b>BODY SIZE</b>              |                    |                    |                    |                         |
| Length (excl. gnathosoma)     | 224 – 298          | 234 – 262          | 193 – 201          | 188                     |
| Length (incl. gnathosoma)     | 323 – 416          | 335 – 379          | 308                | 277                     |
| Width (at level of c2)        | 171 – 242          | 172 – 207          | 156 – 160          | 137                     |
| <b>GNATHOSOMA</b>             |                    |                    |                    |                         |
| <i>m</i>                      | 21 – 25            | 18 – 23            | 20 – 23            | 18                      |
| <i>n</i>                      | 23 – 27            | 18 – 27            | 18 – 19            | 19                      |
| <i>or1</i>                    | 11 – 12            | 11 – 14            | 12 – 13            | 10                      |
| <i>or2</i>                    | 11 – 12            | 10 – 14            | 10 – 14            | 9                       |
| Moveable cheliceral digit     | 31 – 36            | 36 – 39            | 35 – 40            |                         |
| Palp                          | 72 – 80            | 71 – 85            | 86 – 89            | 63                      |
| Palp tibial claw              | 13 – 18            | 15 – 17            | 19 – 20            | 7                       |
| <b>DORSUM</b>                 |                    |                    |                    |                         |
| Eyes (diameter)               |                    | 9 – 10             |                    | 8                       |
| Post-ocular bodies (diameter) |                    | 6 – 8              |                    | 7                       |
| <i>ve</i>                     | 24 – 27            | 15 – 17            | 15 – 16            | 13                      |
| <i>sci</i>                    | 30 – 39            | 21 – 28            | 23                 | 19                      |
| <i>sce</i>                    | 31 – 38            | 23 – 28            | 22 – 24            | 18                      |
| <i>c1</i>                     | 24 – 31            | 20 – 26            | 15 – 17            | 15                      |
| <i>c2</i>                     | 35 – 39            | 21 – 31            | 23 – 27            | 18                      |
| <i>d1</i>                     | 30 – 37            | 20 – 25            | 17                 | 17                      |
| <i>d2</i>                     | 32 – 39            | 23 – 28            | 23 – 26            | 16                      |
| <i>e1</i>                     | 42 – 46            | 29 – 34            | 12 – 13            | 22                      |
| <i>e2</i>                     | 42 – 45            | 27 – 34            | 25 – 27            | 24                      |
| <i>f</i>                      | 34 – 42            | 31 – 35            | 32 – 34            | 25                      |
| <i>h1</i>                     | 38 – 43            | 34 – 39            | 13 – 14            | 26                      |
| <i>h2</i>                     | 32 – 38            | 32 – 36            | 21 – 26            | 22                      |
| <i>ve – ve</i>                | 27 – 45            | 28 – 34            | 30 – 31            | 33                      |
| <i>c1 – c1</i>                | 36 – 43            | 26 – 36            | 34 – 40            | 26                      |
| <i>d1 – d1</i>                | 61 – 73            | 52 – 64            | 54 – 56            | 48                      |
| <i>e1 – e1</i>                | 33 – 34            | 24 – 34            | 27                 | 22                      |
| <i>f – f</i>                  | 65 – 80            | 58 – 65            | 38 – 42            | 44                      |
| <i>h1 – h1</i>                | 15 – 18            | 13 – 21            | 9                  | 13                      |
| <b>VENTER</b>                 |                    |                    |                    |                         |
| <i>g</i>                      | 35 – 39            | 27 – 37            | -                  |                         |
| Aedeagus                      | -                  | -                  | 48 – 55            | -                       |
| <b>LEGS</b>                   |                    |                    |                    |                         |
| Leg I                         | 185 – 200          | 184 – 195          | 187 – 191          | 148                     |
| $\omega\text{I}$              | 13 – 16            | 9 – 12             | 9 – 10             | 7                       |
| $\omega\text{I}\sigma$        | -                  | -                  | 10 – 13            | -                       |
| $\phi\text{I}$                | 10 – 14            | 8 – 11             | 9                  | 5                       |
| leg II                        | 166 – 189          | 168 – 188          | 169 – 174          | 138                     |
| $\omega\text{II}$             | 10 – 13            | 6 – 10             | 9 – 11             | 5                       |
| $\omega\text{II}\sigma$       | -                  | -                  | 11 – 13            | -                       |
| $\phi\text{II}$               | 9 – 10             | 4 – 6              | 5 – 6              | 3                       |
| leg III                       | 169 – 187          | 173 – 194          | 166 – 175          | 141                     |
| $\omega\text{III}$            | 8 – 10             | 5 – 6              | 5                  | 3                       |
| $\phi\text{III}$              | 7 – 10             | 3 – 5              | 4                  | 2                       |
| leg IV                        | 185 – 210          | 188 – 198          | 184 – 186          | 144                     |
| $\omega\text{IV}\sigma$       | -                  | -                  | 5 – 6              | -                       |

**Table 2** *Agistemus collyerae* specimens detected on agricultural imports from other countries by South Africa.

| country   | crop                                                        | commodity       | import date(s)               | specimens                 |
|-----------|-------------------------------------------------------------|-----------------|------------------------------|---------------------------|
| Yemen     | citrus (unspecified <i>Citrus</i> sp.)                      | budwood         | Aug 1991                     | 1 female                  |
| Australia | mandarin orange ( <i>Citrus reticulata</i> )                | budwood         | Jan 2013                     | 2 deutonymphs             |
|           | nectarine ( <i>Prunus persica</i> var. <i>nucipersica</i> ) | budwood         | Feb 2007, Jan 2010, Feb 2010 | 5 females, 6 males        |
|           | peach ( <i>Prunus persica</i> )                             | budwood         | Jan 2010, Mar 2013           | 2 females, 1 male         |
|           | citrus (unspecified <i>Citrus</i> sp.)                      | budwood         | Jan 2010                     | 3 females                 |
| Italy     | olive ( <i>Olea europaea</i> )                              | rooted cuttings | Oct 2007                     | Unable to locate specimen |
| Spain     | apricot ( <i>Prunus armeniaca</i> )                         | fresh fruit     | Jun 2009                     | 1 female                  |
|           | grapes ( <i>Vitis vinifera</i> )                            | fresh fruit     | Oct 2015                     | 1 female                  |
| Chile     | grapevine ( <i>Vitis vinifera</i> )                         | budwood         | Mar 2014                     | 1 female                  |
| USA       | nectarine ( <i>Prunus persica</i> var. <i>nucipersica</i> ) | budwood         | Nov 2010                     | 2 females                 |
|           | peach ( <i>Prunus persica</i> )                             | budwood         | Nov 2011, Jan 2012, Nov 2012 | 2 females, 1 male         |
|           | citrus (unspecified <i>Citrus</i> sp.)                      | budwood         | May 2016                     | 1 female                  |
| France    | nectarine ( <i>Prunus persica</i> var. <i>nucipersica</i> ) | budwood         | Jan 2017                     | 1 female                  |

enhancing biological control of pest species (Palevsky *et al.* 2013; Hajek *et al.* 2016). However, the ecological effects are largely unknown. There are some notable cases in which introduction of an arthropod predator to enhance biocontrol has generated significant negative impacts – the case of the Harlequin ladybug (*Harmonia axyridis* (Coccinellidae)) being a prime example (Stals & Prinsloo, 2007; Kenis *et al.* 2016). Mite biology and ecology have not been studied extensively enough to understand nor predict the impact an introduced predatory species may have.

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