To the Graduate Council:

I am submitting herewith a thesis written by Kelly Lynn Felderhoff entitled “A Revision of Tomoceridae (Insecta: Collembola) in Great Smoky Mountains National Park and Southern Appalachians Using Morphological and Molecular Approaches.” I have examined the final electronic copy of this thesis for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Entomology and Plant Pathology.

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(Original signatures are on file with official student records.)

Revision of Tomoceridae (Collembola) in Great Smoky Mountains National Park and Southern Appalachians

A Thesis
Presented for the
Master of Science
Degree
The University of Tennessee, Knoxville

Kelly Lynn Felderhoff
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Dedication

This thesis is dedicated to my family and friends who supported me through this endeavor; to my parents, for their endless love and encouragement, and for instilling the importance of a higher education in me. In particular, this is dedicated to my cousin, Daniel Reiter. His love for the Lord's gift of the natural world will always be remembered.

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I am so grateful to Chris and Sandy Felderhoff, my parents, for their unending love and guidance and for their many sacrifices to support me emotionally and financially throughout my time here at the University of Tennessee. I always knew they were behind me and proud of me. Finally, I thank Brian Oten for his love, his sense of humor, and his amazing support and belief in me. His willingness to help and listen was essential to me completing this project with a smile.

Abstract

Large, heavily scaled tomocerid springtails (Collembola) are abundant in eastern forests, and are important components of the detrital food web. The genus *Pogonognathellus* predominates in the southern Appalachians While a number of well-delimited tomocerid species have been described, others have vague morphological boundaries and appear to be species complexes. Before this study began in 2005, four species were known to occur in Great Smoky Mountains National Park (GSMNP) and the surrounding Appalachians (*P. bidentatus, dubius, elongatus, flavescens*). Also occurring throughout the area is *Tomocerus lamellifera*, a widespread and distinctive species. The goals of this project were to correlate scale patterns and colors, ground color, morphology, and DNA sequences for separation of species; to describe any new species detected; and to redescribe known species. Scale patterns and molecular sequences have not been previously studied in North American Tomoceridae.

We collected 432 specimens from diverse localities and maintained them in culture containers. After a specimen molted, it was photographed to capture the pristine scale pattern and color, then preserved in 100% ethanol (EtOH). The preserved specimen then was re-photographed for ground color (most scales are dislodged in preservative). Selected specimens were dissected and the various appendages (legs, mouthparts, furcula) were slide-mounted for morphological analysis. The torso was used to obtain DNA, from which the 5'-3' exoribonuclease II gene was amplified, sequenced, and analyzed phylogenetically. Freshly collected specimens of *P. flavescens* from the type locality (Sweden), preserved in 100% EtOH, were provided by Arne Fjellberg and included in the analysis. *Tomocerus minor* and *Harlomillsia oculata* (Oncopoduridae) were used as outgroups for the phylogenetic analysis.

A phylogenetic tree based on the molecular sequences was used to indicate relationships that then were tested with morphological characters, chaetotaxy, scale pattern, and color. Putative *P. flavescens* from North America were not grouped with Swedish specimens, raising the possibility that *P. flavescens* does not occur in North America. *Pogonognathellus nigritus*, previously synonymized with *P. elongatus*, was determined to be a valid species, and *P. elongatus* itself was found to be mis-described in most accounts. Two new species were identified: one from a cave in GSMNP which is closely related to a California cave species, and a second from several forest localities (i.e. GSMNP and Mount Mitchell, NC). A group of specimens ascribed to the *flavescens/dubius* complex also appear to represent several undescribed species, but more specimens are needed for further analysis.

Preface

This work and the new scientific names and nomenclatural acts included in it are not being issued for permanent scientific record in the sense of Article 8a(1) of the International Code of Zoological Nomenclature (International Commission on Zoological Nomenclature 1999). Therefore, the names and nomenclatural acts proposed in this thesis are not available in the sense of Article 10a of the Code (ICZN).

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I. Literature Review

i. Overview of Collembola

Springtails are small, wingless hexapods (the largest up to 10 mm) inhabiting diverse, usually humid environments. They occur in moist soil and leaf litter environments, under loose bark, in decaying matter, beneath soil, in moss, in the canopy of trees, and some are strictly cave dwellers. Considered the most widespread and abundant terrestrial arthropods, collembolans remain a much understudied group of hexapods (Hopkin 1997). Only recently has the importance of springtails in ecosystems been recognized. Collembola are consumers primarily of fungal spores, hyphae, and decaying plant tissue, although one or more species have been recorded as consuming feces, filter paper, salt agar, cheese, butter, starch, syrup, and other springtails (Knight and Angel 1967). In turn, springtails are important prey of spiders, beetles, pseudoscorpions, centipedes, and other predacious arthropods. They are important decomposers and recyclers in a healthy ecosystem, and have a heavy influence on soil structure and soil respiration. Depending on their abundance and forest composition, Collembola are responsible for 2% to 30% of forest floor decomposition (Bitzer et al. 2005, Hopkin 1997). Collembolans also are beneficial by grazing on rhizoplane and rhizosphere fungi and reducing root disease (Lussenhop 1996, Nakamura et al. 1992). The effects of pesticides on

Collembola has been studied extensively. Attention to springtail population decline due to residual pesticides in the soil has been the concern of some ecologists (Goto 1972).

Springtails get their common name from the furcula, a tail-like jumping organ on the fourth abdominal segment. The furcula evolved by fusion of a pair of abdominal legs. Many Collembola can instantaneously jump several times their own body length when disturbed. Some species are spectacular leapers; *Pogonognathellus flavescens* Tullberg (Collembola: Tomoceridae) has been documented to jump approximately 20 times the length of its body (Zinkler and Schroff 1989). In many soil-dwelling species, the furcula is reduced or absent. The furcula has three segments: a basal manubrium, paired dentes, and a toothed mucro at the distal end of each dens. Each segment can have important characters for species separation.

On the first abdominal segment, Collembola have a ventral tube, the collophore, which is used for water uptake and as a device for adherence to vertical surfaces. It is from this organ that the name “Collembola” was created, Greek for “glue piston” (Hopkin 1997). This structure is not often used in taxonomy, but in some genera setal numbers and arrangements are species-specific.

Springtails have entognathous mouthparts (enclosed within the buccal cavity), consisting of five components: the labrum (which encloses the mouthparts dorsally), a pair of mandibles, a pair of maxillae, the hypopharynx,

and the labium (which encloses the mouthparts from below) (Goto 1972, Hopkin 1997). The shape and ornamentation of the mandible, maxilla, and labium often are species-specific (Fjellberg 1984, 1998/99).

The eyes are comprised of up to eight small ocelli on each side of the head, with each ocellus consisting of multiple fused ommatidia. Eyes are used primarily for orientation. During migrations on snow *Hypogastrura socialis* Uzel (Hypogastruridae), a snow flea, has been observed sensing the angle of the sun before making a jump (Hågvar 1995). Cave-specific and soil-dwelling species often have reduced numbers of ocelli or are blind (Packard 1877, Hopkin 1997).

Sexual dimorphism is rare in most families of springtails. Gender can be determined by the shape of the genital aperture of the fifth abdominal segment. Fertilization is indirect; males deposit a stalked spermatophore which the females acquire to fertilize themselves. Mating behavior varies widely among families, from males depositing spermatophores at random, to elaborate courtship behavior between a male and a female (Hopkin 1997). Eggs are laid singly or in clusters of up to 100 eggs. On average, a female springtail will produce about 90 eggs in a year (Hale 1965).

Recent debate on the taxonomic placement of Collembola has been vigorous. Collembola form an undoubtedly monophyletic group within the Phylum Arthropoda, placed either in Class Insecta, or Class Parainsecta, or in a separate Class Collembola. More conservative entomologists keep Collembola grouped with Insecta, while some phylogenetic data have suggested they are a

class of their own, related to the Class Crustacea. Their true taxonomic position is still in debate. Fossil Collembola are not uncommon but all springtail fossils, some more than 400 million years old, have modern morphological characteristics. Cladistics has been the most appropriate way to study their relationships (Hopkin 1997) but molecular studies show promise for resolving morphologically similar species.

ii. Tomoceridae

Tomocerid springtails are abundant in eastern forests. Many species attain a length of 5 mm or more, making them among the largest of all collembolans (Folsom 1913). They are easily identified by their arthropleonid body type and four segmented antennae, with the third segment much longer than the fourth. The last two segments are flexible and can coil or twist when disturbed. This characteristic is unique to tomocerids. Most species have six eyes on each side of the head, but occasionally two or fewer occur. All species occurring in the southern Appalachians have 6 + 6 eyes. The length of the fourth abdominal segment is subequal to the third. Currently, 136 species of Tomoceridae are known worldwide, 16 of which are found in the United States (Christiansen and Bellinger 1998).

For tomocerids, the most important food source is fungal spores (Knight and Angel 1967). Early instars are recognizable as tomocerids but generally

cannot be identified to species. Ungual teeth are often very different for the first five instars, and dental spines are constantly changing (Christiansen 1964).

The family Tomoceridae is comprised of 15 genera worldwide, most of which occur in the northern hemisphere (Fanciulli et al. 2000). Five described species were known to occur in GSMNP and the southern Appalachians prior to this study: *Pogonognathellus bidentatus* Folsom, *P. dubius* Christiansen, *P. elongatus* Maynard, *P. flavescens*, and *Tomocerina lamellifera* Mills. The four species in the genus *Pogonognathellus* are difficult to distinguish from one another morphologically. The bodies of tomocerids are covered with colorful scales that may form species-specific patterns. Scale color ranges from gray or brown to deep purple-black; species with distinct patterns often have patches of white scales. Frequently, the scales impart a metallic appearance (Folsom 1913). Ground color, which is seen when the scales are removed, is usually a dull gray or yellow.

Naming and identifying species in Tomoceridae is difficult. Many of the existing species have numerous synonyms. Three of the five species occurring in GSMNP have synonyms. *Tomocerus flavescens*, in which there is much variation, has six synonyms associated with it: *T. alba* Packard, *T. plumbeus* Packard, *T. arcticus* Schött, *T. americanus* Schött, *T. niger* Folsom, and *T. separatus* Folsom. *Pogonognathellus bidentatus* has two synonyms: *P. jeanneli* Bonet and *Tritomurus henroti* Delamare.

iii. Review of Tomoceridae Literature

The first tomocerid described, *Macrotoma flavescens*, was described by Tullberg (1871). Packard (1877) noted a similar cave form that he named *Tomocerus alba*, because of its pale appearance. Later, Packard (1888) named another cave species *T. pallidus*. Both of these descriptions lack even the most simple morphological exposition, and refer only to color and apparent transformation of *T. flavescens* into a cave form. No illustrations were given and type specimens are unknown. These species are unplaceable and will remain so unless new material is collected from the type locality caves. Schött (1893) reported a similar species and named it *T. arcticus*. This taxon later was synonymized with *Pogonognathellus flavescens* (Christiansen and Bellinger 1998).

The first recognizable U.S. species were described by Folsom (1913), who reviewed the known North American Tomoceridae. Eight taxa were recognized: four forms of *Tomocerus flavescens*, *Tomocerus bidentatus*, *Tomocerus vulgaris* Tullberg, *Tomocerus minor* Lubbock, and *Tritomurus californicus* Folsom. Folsom acknowledged the presence of scales but did not depict them in his species descriptions. Mills (1934) described *Tomocerus lamelliferus* (now *Tomocerina lamellifera*). Two species, *T. elongatus* and *T. nigritus*, were described from New York by Maynard (1951). Both of these species are now placed in *Pogonognathellus*. Yosii (1956) transferred *T. elongatus* and *T. nigritus* to a new genus, *Maynardia*, on the basis of long manubrial spines, which other

tomocerids lack. Later workers did not accept this arrangement. Knight (1958) raised *T. flavescens americanus* to species status (*T. americanus*) on the basis of field observations, but did not give a clear morphological basis for separation of this taxon from other *T. flavescens* forms. This change was not accepted by later workers.

In a major revision of the family in North America, Christiansen (1964) did not recognize any of the *T. flavescens* forms and synonymized *T. nigritus* with *T. elongatus*. He also described *T. celsus*, a western cave species, and *T. dubius*, a species very similar to *T. flavescens*. The co-authored monograph of Christiansen and Bellinger (1980, 1998) provided keys and descriptions of all known North American Collembola and summarized North American Tomoceridae, but did not include additional species or insight beyond that of Christiansen (1964).

As early as 1908, students of Collembola have placed distinctive groups of *Tomocerus* species in separate genera (e.g., *Pogonognathus*: Börner 1908). Yosii (1956) revised the family, recognizing several previously established genera and creating new genera. However, most authorities adopted a more conservative approach and either did not recognize these newer genera or kept them as subgenera (Christiansen and Bellinger 1998, Gisin 1960). The genus *Pogonognathellus* was described long ago (Börner 1908) but was not generally accepted until the late 1990s. However, most authors retained it as a subgenus of *Tomocerus*. When first published by Börner in 1908, the subgenus was

named *Pogonognathus* but this name was found to be preoccupied (Mollusca) and was changed to *Pogonognathellus* by Paclt (1944). Currently the genus is separated from other tomocerid genera on the basis of an enlarged inner dental scale, the absence of outer dental basal spine-like setae, and a single dorsal mucronal lamella (Christiansen and Bellinger 1998).

iv. Research Objectives

The goal of this project was to determine the diversity and identities of Tomoceridae in Great Smoky Mountains National Park and surrounding areas by correlations of scale patterns and colors, ground color, morphology, and DNA sequences. The specific objectives of this project were:

(1) Maintain specimens in culture to document color and scale patterns;

(2) Characterize and describe any new species found in Great Smoky Mountains National Park and surrounding areas;

(3) Conduct a molecular phylogenetic analysis that could contribute to species differentiation.

II. The Family Tomoceridae

i. Introduction

Springtails (Order Collembola) are small insects that live in soil and leaf litter of moist environments. They are important decomposers and recyclers in a healthy ecosystem. Up to 100,000 individuals comprising many species in several families can occur in one cubic meter of mesic forest. Great Smoky Mountains National Park (GSMNP), in the southern Appalachians, provides a wide range of habitats where a highly diverse springtail fauna can be found, some of which are undescribed and new to science.

In the southern Appalachians, only two genera of Tomoceridae occur, *Pogonognathellus* and *Tomocerina*. The latter genus is represented by *T. lamellifera*, a distinctive and well-defined species, while *Pogonognathellus* is represented by four species prior to this study.

Members of the family Tomoceridae are unique springtails because their bodies are covered in colorful scales that create patterns or an iridescent appearance. However, when specimens are placed in alcohol, these scales fall off, usually leaving the springtail looking a dull yellow or gray (Fig. 1). Thus, most entomologists never see these patterns and they are rarely described in the literature. Tomocerids are some of the largest springtails (up to 5 or 6 mm) and



Figure 1. Scale loss in preservative. *Pogonognathellus elongatus* (top) and *Pogonognathellus bidentatus* (bottom); habitus (left) and preserved in alcohol with scales dislodged (right).

are found in the detritus of the forest floor, rather than the soil. They consume fungi and decomposing leaf litter. Because of their relatively large size and abundance, tomocerids are among the springtails most likely to be seen by the public. Some of the larger species have been observed climbing ferns or blades of grass and can be collected with a sweep net.

The antennae are comprised of four segments, with the third segment the longest and last segment much shorter than the third. This character distinguishes Tomoceridae from all other Collembola. The third and fourth segments are strongly annulate and the third is very flexible. All tomocerids collected in this study have six eyes per side.

Before this study was initiated in August 2005, five species of tomocerids were known to occur in GSMNP, and three undescribed species were suspected. Of the known species, morphological characteristics set them apart but molecular differences were not known. This project sought not only to describe any new species discovered, but to find additional species that have not yet been documented from these habitats.

The use of genetic variability among insects is a valuable approach in systematics. Many factors influence the genetic structure of a population. First, the surrounding environment can cause certain characteristics to express themselves. Another factor, and perhaps the most important, is genetic drift, or fluctuations in the frequency of alleles of a population. Historical events also play a major role, especially in mountainous terrain such as in GSMNP. Geographical

barriers and habitat change isolate populations resulting in population bottlenecks. This effect is especially applicable to springtails, as their dispersal is largely reliant on passive transport. Genetic variability among tomocerids indicates that, in GSMNP, numerous microspecies may exist that have developed in isolation among the various watersheds and other restricted habitats. Microspecies complexes in the southern Appalachians have been suggested for *Metajapyx* (Diplura), *Eosentomon* (Protura), and for several genera of Carabidae (Coleoptera).

ii. Materials and Methods

Collection

The collecting localities were chosen to sample a variety of areas within GSMNP (collecting permit GRSM-2006-SCI-0011) and the surrounding Appalachian Mountains. Additionally, areas were selected based on accessibility, location in relation to other collection sites, and suitable habitats. Localities within GSMNP are marked in Figure 2 and are given in detail in the species descriptions in Chapter III.

Leaf litter and moss samples of moist areas were collected, as individuals are relatively large and not usually found in the soil. Each sample was

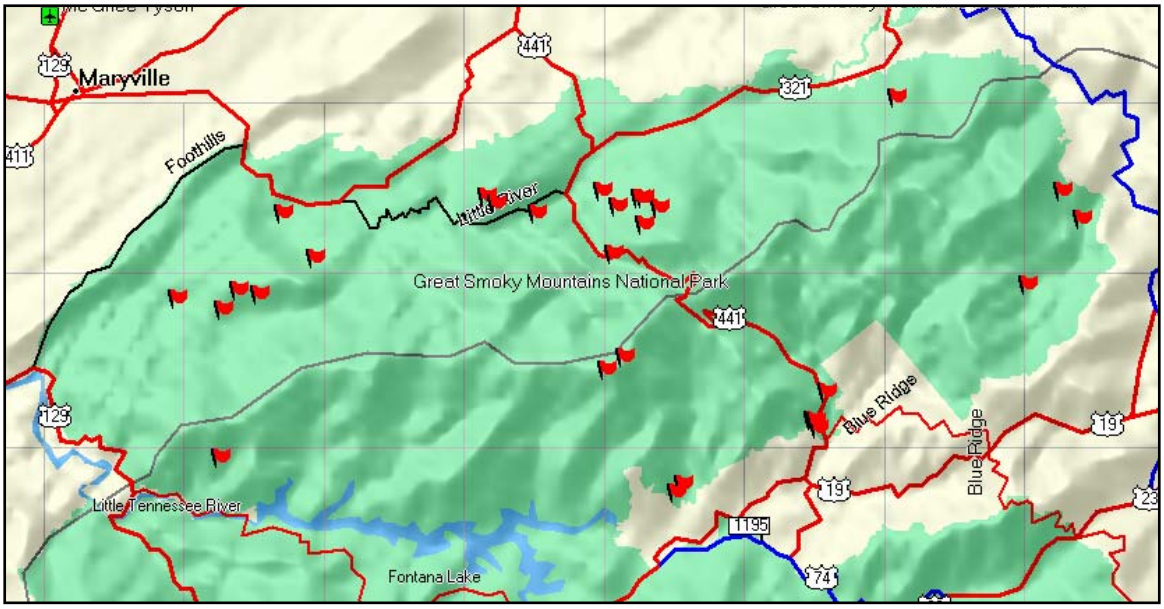


Figure 2. Collecting localities in GSMNP.

deposited in a one or two-gal Hefty® OneZip bag for transport. Samples were placed in Tullgren funnels (Fig. 3) to extract live Collembola. In a Tullgren funnel, material is placed under a light source (15 W light bulb) which heats and dries the organic material. Organisms migrate downward to escape these conditions and fall through the screen supporting the sample. In this project, collection chambers were used to collect live specimens falling through the screen. Typically, this process was complete in 1-2 days. The live collection chambers were made from 90 x 50 mm Pyrex® dishes with a plaster/carbon medium in the bottom, moistened with tap water. The plaster/carbon medium consisted of one part activated carbon powder and one part plaster of Paris (calcium sulfate). This substrate was used in the bottoms of the individual culture chambers used throughout the project.

Culture

Individual specimens were placed in 1-oz. Nalgene® containers with a carbon/plaster medium bottom. The medium functions to maintain moisture and absorb waste. Each specimen was given granules of baker's yeast and small pieces of leaf from the environment in which they were found. Yeast is commonly used as food for springtails in live culture. Through observation, the springtails with a small piece of leaf in the container were found to live longer. Individuals were monitored daily for food, water, and molting.

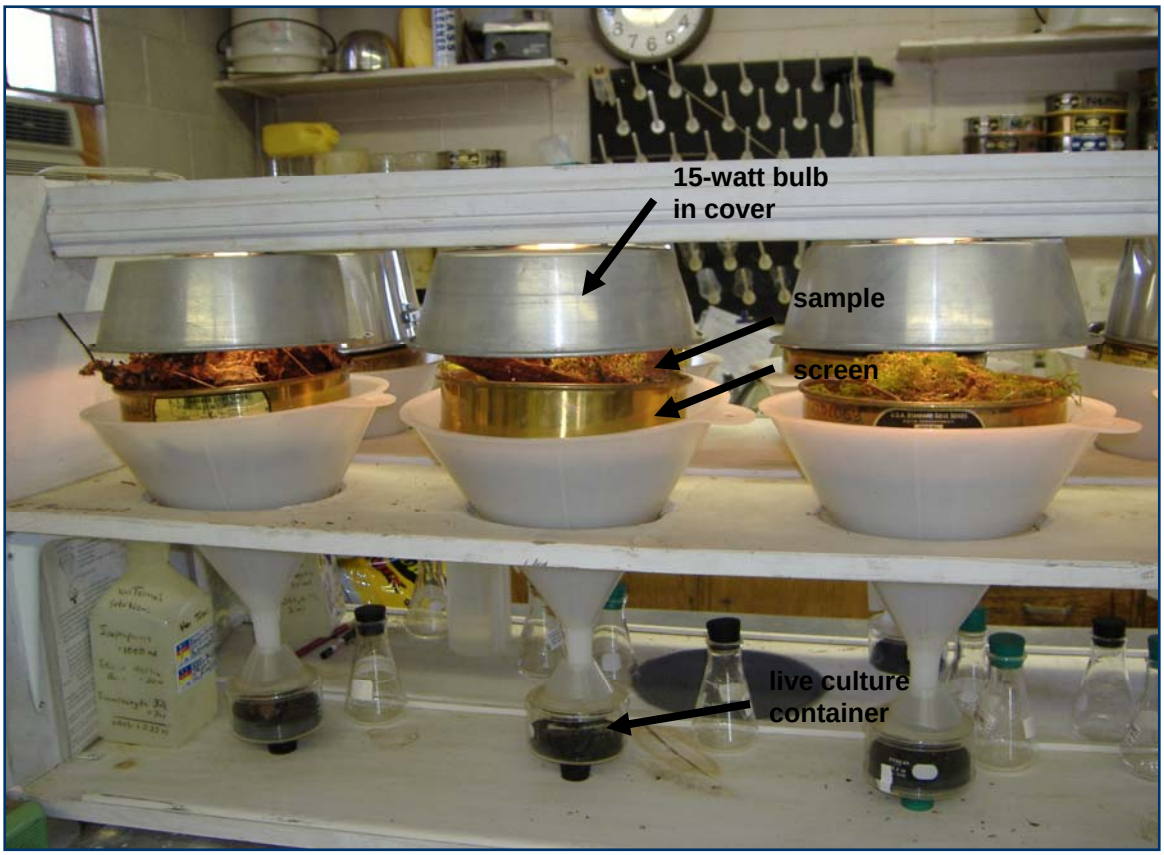


Figure 3. Tullgren funnels used to extract live specimens.

Imaging

If the specimen had molted and acquired a complete scale pattern, digital images were taken from several angles and magnifications. Each specimen was photographed with a 3.34 megapixel Nikon® CoolPix 990 digital camera. Digital files were stored for later reference to scale pattern. After live specimens were photographed, they were preserved in 95% EtOH and photographed again after the body scales had fallen off (Fig. 1).

Morphological analysis

Specimens were prepared for dissection by clearing them in Marc Andre I liquid (30 ml deionized water, 40 g chloral hydrate, 30 ml glacial acetic acid). Specimens were cleared for 5-15 min, then the parts to be mounted were detached with an eye knife and mounted under separate cover slips in Hoyer's medium (125 ml deionized water, 75 g gum Arabic, 500 g chloral hydrate, 50 ml glycerin).

For mouthpart dissections, the head was placed in Andre I and heated in a slide oven at 60°C for 5-10 min to soften the cuticle. The mandibles, maxillae, labial palps, and labrum were removed with the aid of entomology pins and forceps. If the remaining head cuticle was intact, it was also mounted.

To slide-mount the cuticle, alcohol was removed from the body in two sequential baths of tap water of 10-15 min each. The body was placed in 10% KOH (10 g KOH/100 ml H_2O) and allowed to clear before being immersed in two

more baths of water, then transferred back to alcohol. The cuticle was cut open longitudinally along the ventral side, muscle tissue was teased out and removed, and the cuticle was slide-mounted dorsal side up in Hoyer's medium. The morphological characteristics studied include both traditional characteristics and others previously unused in this family.

Body chaetotaxy: The arrangement of setae is often a species-specific characteristic. A typical tomocerid will have between four and 12 macrochaetae on the second thoracic segment (Th. II) and a cluster on the anterior margin. This cluster is not shown in chaetotaxy drawings in this study. Typically, there are three posterior macrochaetae on Th. III, the first abdominal segment (Abd. I), and Abd. II, three posterior macrochaetae and two anterior macrochaetae on Abd. III, two posterior macrochaetae and one anterior macrochaetae on Abd. IV, and three posterior macrochaetae on Abd. V. One bothriothrix is present on Th. II, Th. III, and Abd. III, while two are present on Abd. IV.

Cuticles were slide-mounted and the chaetotaxy was drawn with the aid of a drawing tube mounted on an interference-contrast microscope. Figures of the chaetotaxy were standardized after Christiansen (1964) to facilitate comparisons of species. Figures show the left dorsal side of the body, with Th. II at the top, followed by Th. III, and the first four abdominal segments. The fifth segment was included when the setal bases could be clearly seen.

Dental spines: This character is very useful for identification of species of Tomoceridae. Variations are common in dental spinal composition within a

species; the dental spine formula followed here is that used by Folsom (1913) and Christiansen and Bellinger (1998). In a hypothetical example, 1,2/3-5,1-2, non-underlined numbers indicate small spines, while underlined numbers designate large spines. Numbers preceding the slash indicate spines on the first of the two subsegments of the dens. Similarly, numbers following the slash are the spines on the second subsegment of the dens.

Eyespot: All tomocerids in this area have six eyes per eyespot. The shape of the eyespot can be triangular or trapezoidal and differs among species.

Foot complex: Another very useful characteristic, the foot complex also varies among species. The larger claw, the unguis, and its smaller accessory claw, the unguiculus, may have inner teeth. At the base of the unguis arises a long tenent hair that is clubbed or pointed. Pairs of legs were mounted separately, as variation may occur on different feet of the same specimen.

Labial palp: *Tomocerus* and *Pogonognathellus* have complete labial palps bearing five papillae each with a terminal sensillum and 16 guard setae at the base. Guard setae lack a setal socket and are more transparent than typical setae. The labial palps of several tomocerid specimens representing different species were examined and no qualitative differences were discerned.

Labrum: The labrum was studied on multiple specimens and all had similar morphology (Fig. 4) with a 5,5,4 setal formula, four minute apical setae, and an apical fringe.

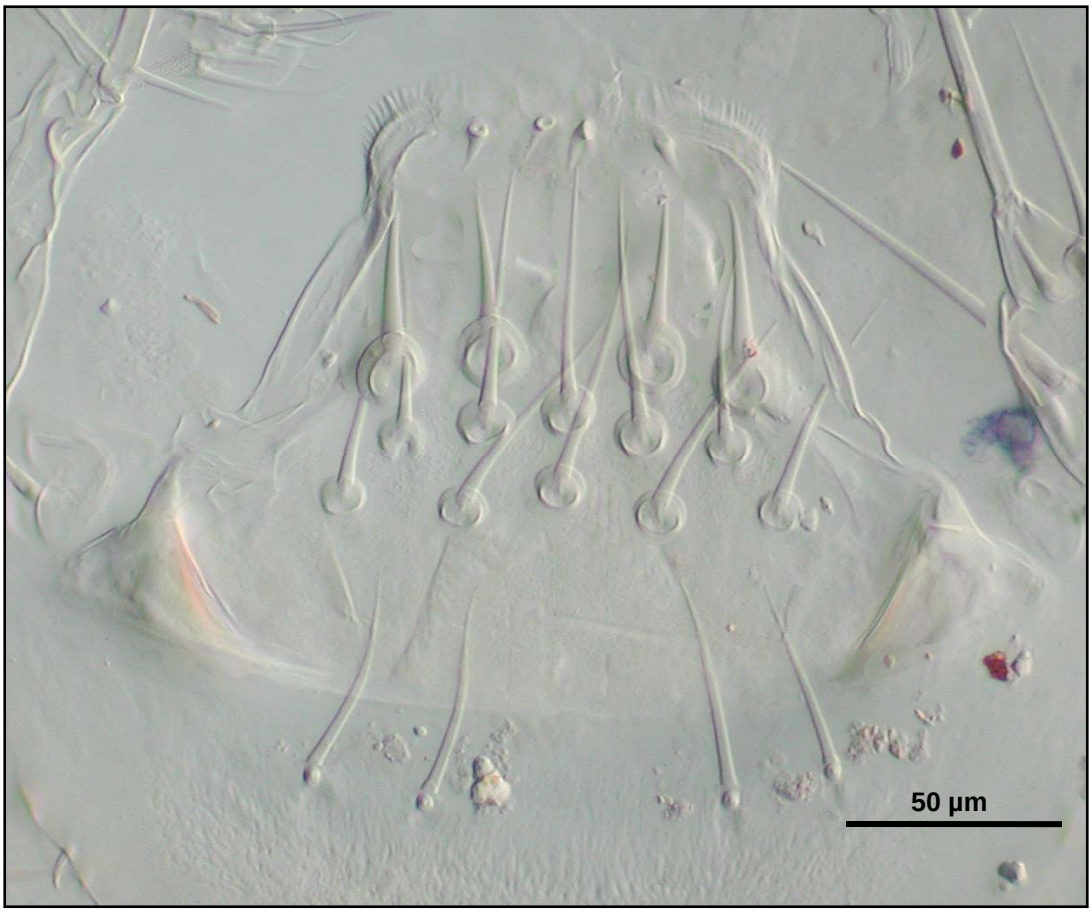


Figure 4. Labrum of *Pogonognathellus bidentatus*, typical of tomocerids.

Legs: The inner surface of the tibiotarsi, especially that of the foretarsus, may have strong, thick spines that are much larger than typical setae. The number of spines varies among species.

Length: Length excluding antennae and furcula was determined by measuring the largest specimens to determine a maximum length.

Mandible: Two mandibles, one with four teeth at the distal end, and the other with five, are typical of the family. The basal part of the head of each mandible is a molar plate (Fig. 5).

Manubrial setae: Two species (*P. elongatus*, *P. nigritus*) possess spine-like setae on the manubrium (Maynard 1951). These setae are not present on other examined *Pogonognathellus* species.

Maxilla: The head of the maxilla consists of six lamellae of varying size and shapes. Several lamellae have well-developed fringes that obscure the smaller lamellae. In *Pogonognathellus*, the long second lamella appears to have species-specific differences.

Mucro: The terminal segment of the furcula has several characters important in species descriptions: the absence or presence of a lamella and its extent, the number of intermediate teeth, the distance of the basal tooth from the base of the mucro, and the relative size of the lateral tooth to the basal tooth.

Holotypes and some paratypes will be deposited in the U.S. National Museum of Natural History (NMNH), Washington, D.C. Other paratypes will be deposited in the University of Tennessee Insect Collection. Voucher specimens

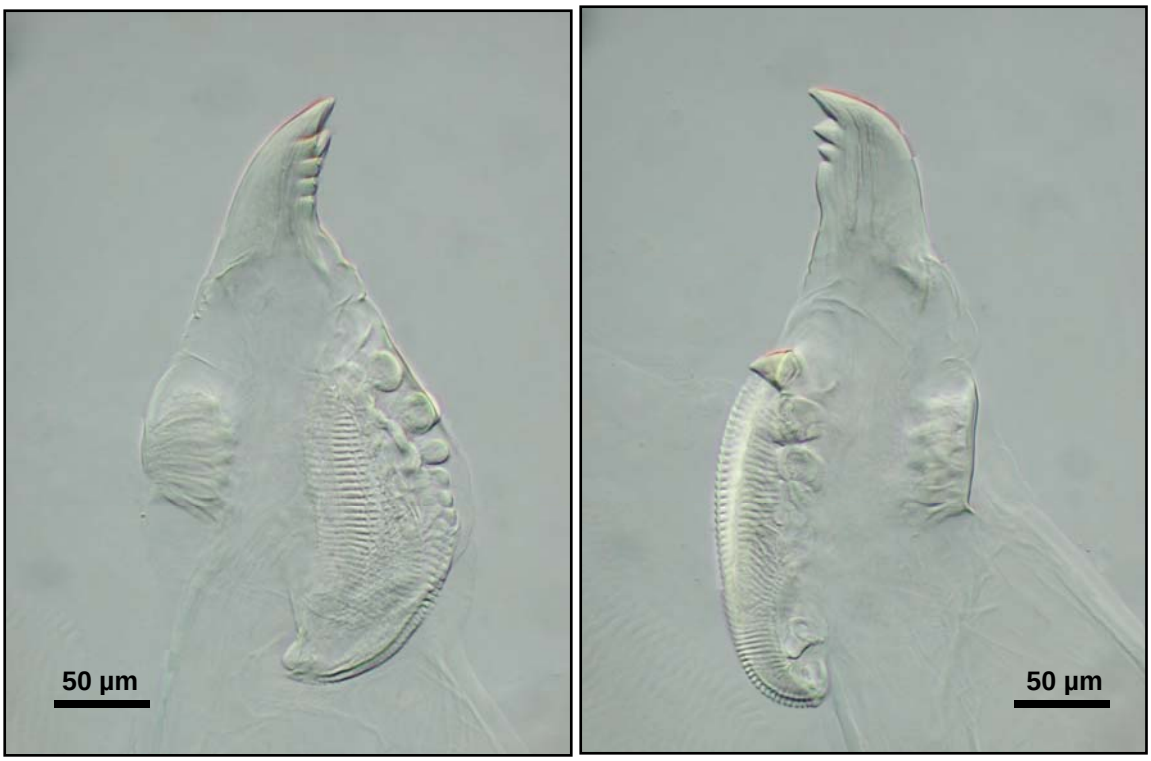


Figure 5. Pair of mandibles of the same specimen of *Pogonognathellus elongatus*, illustrating typical mandibular structure of Tomoceridae.

of all species will be deposited in the University of Tennessee Insect Collection and the GSMNP Natural History Collection.

Molecular analysis

The 5'-3' exoribonuclease II gene was amplified using the polymerase chain reaction (PCR), sequenced, and analyzed phylogenetically. In Collembola, the region of this gene analyzed contains two exons separated by a short intron, a highly variable non-coding region. Therefore, this gene is suitable for use in separating morphologically similar species. The furcula, legs, and head were removed and slide-mounted to serve as vouchers prior to DNA extraction. A majority of the body was used to obtain DNA. The protocol used is described below.

DNA extraction: First, the springtail body was removed from 100% EtOH (preservative) and allowed to dry on a lint-free tissue. In a 1.5 mL plastic vial, 700 µL lysis buffer, 8 µL proteinase K, and the specimen were combined. The specimen was then macerated using a sterile pestle for each individual. This homogenate was incubated in a heat block (Fisher Scientific Isotemp 125D, Pittsburgh, PA) at 55°C overnight.

After being heated overnight, 700 µL of a phenol/chloroform/isoamyl solution (25:24:1) was added to each vial. Each vial was mixed by flicking or inverting several times and then centrifuged (Eppendorf Centrifuge 5417R) at 14,000 x g for 5 min. The aqueous layer (top layer) was pipetted off, placed into

a new 1.5 µL Eppendorf tube, and 650 µL chloroform/isoamyl alcohol (24:1) was added. This second vial was flicked and spun at 14,000 x g for 5 min. The aqueous layer was pipetted into a new 1.5 µL Eppendorf tube to which 65 µL (1/10 volume) of 3M sodium acetate was added to create a DNA salt. The vial was mixed by flicking and briefly centrifuged. An amount of cold (-20°C) isopropyl alcohol equal to the volume already in the vial was added and the tube was inverted several times until the solution turned clear. The solution was spun at 14,000 x g for 5 min to pellet the precipitated DNA salt into a pellet at the bottom of the vial. The eluent was discarded and 1 mL of 70% EtOH was added to the pellet to wash off excess salt. The tube was inverted several times and the eluent poured off, being careful not to pour out the DNA pellet. A subsequent wash with 1 mL 95-100% EtOH was conducted to facilitate drying of the pellet. The vial containing the DNA pellet was laid on its side between Kimwipes® and allowed to air dry, or was dried for 45 min in a Speed Vac (Centrivap Concentrator, Labconco, Kansas City, MO).

DNA resuspension: After the DNA pellet dried, it was suspended by adding 100-150 µL TE, depending on the size of the pellet. It was then heated in the heat block at 55°C for 5 min to fully resuspend the DNA.

Amplification: The DNA template was amplified by PCR using Hotstart Ex Taq (TakaraMirus, Madison, WI). In each 0.5 ml tube, 1.5 µL of the DNA template was mixed with 36 µL ddH_20, 5 µL 10X buffer, 1.5 µL 50 mMol $MgCl_2$, 4 µL of 10 mMol dNTPs, 2.5 µL of forward primer (5'-3' Exo2 335F: 5' –

GAYGAYTGGGTNTTYATGTGYTT – 3'), 2.5 µL of reverse primer (5'-3' Exo2 651R: 5'-CCYTGCCANGCRAAYTTYTT – 3' or 3'R: 5' – GCTSCGTTATTATTATCAGGT – 3'), and 0.2 µL of Taq. The mix was flicked and spun briefly, placed in a thermocycler (Eppendorf Mastercycler), and cycled for approximately 3.5 h using the following parameters: 2 min at 94°C, 5 cycles of 94°C for 30 s, 57°C for 15 s, and 1-2.5 min at 72°C, 10 cycles of 94°C for 30 s, 53°C for 15 s, and 1-2.5 min at 72°C, 33 cycles of 94°C for 30 s, 47°C for 15 s, and 1-2.5 min at 72°C, followed by a 4°C soak.

Following amplification, 8 µL of green loading dye (Fermentas, Glen Burnie, MD) was added to each vial. An agarose gel was prepared by mixing 75 mL of 1XTAE and 0.6 g of agarose (Fisher Biosciences), heating the mixture in a microwave for approximately 40 s, with occasional swirling, until the solution was clear. The mixture was taken out, cooled to 50°-60°C, and poured into an electrophoresis casting tray (Fisher Biosciences Electrophoresis System). Once the gel had set, the lane molds were removed and the gel was placed into proper orientation within the buffer trough; 1X TAE was added to the trough until the gel was fully immersed, and each specimen's reacted PCR solution plus loading dye and 10 µL of a DNA size standard (1 kb ladder, Promega, Fitchburg, WI) were added to the gel lanes. The electrophoresis ran for 45 min at 100 V. The gel was removed and placed on a UV light box for visualization via ethidium bromide contained in the gel and running buffer. The fluorescing DNA bands were

excised from the gel using a fresh razor blade for each specimen. The gel slice containing the DNA was placed into a new 1.5 mL tube and refrigerated.

Purification: Next, the PCR products were purified from the agarose gel. To each tube containing a gel band, 375 µL of QG solubilization buffer (Qiagen Corp., Germantown, MD) and 175 µLof isopropanol were added. This mix was vortexed (Fisher Scientific Mini Vortexer) for two s then heated in the heat block at 55°C until the gel dissolved completely. The liquid was pipetted into silica spin columns (Qiagen) and centrifuged at 11,400 x g for one min to remove melted gel and other contaminants. Contaminants were removed from the bottom portion of the spin column, after which 500 µL buffer QG was added. After 10 min the vial was centrifuged at 11,400 x g for one min and the waste discarded. Wash buffer (730 µL Buffer PE) was added and 15 min later, the tubes were centrifuged again at 11,400 x g for one min and the waste discarded. The vials were dried by centrifugation at 13,000 x g for two min. The silica membrane column was placed into a new vial and 30 µL of elution buffer (10 mmol Tris HCl, pH = 8.5) was added and allowed to stand for 40 min. Tubes were then centrifuged at 13,000 x g for one min. A thin agarose gel was poured in the same manner as the previous gel, using only 50 mL of 1X TAE and 0.5 g agar instead. In order to determine the amount of DNA template to use in the sequencing reaction, 3 µL of loading dye and 2 µL of purified PCR product were placed in a 0.5 ml vial, flicked, and centrifuged briefly. The contents of the vial were electrophoresed in the agarose gel for 45 min, after which the gel was

removed and the DNA bands were photographed using a Gel Logic 200 (Kodak, Rochester, NY) imaging system. The intensity of the PCR band compared to a band of known concentration in the DNA size standard revealed how much DNA was recovered from the gel and therefore how much DNA template to use in the subsequent cycle sequencing reaction.

Sequencing reaction: For each specimen, two vials were labeled, one for the forward primer and one for the reverse primer. Depending on the amount of DNA template required, with the general rule being 10 ng of DNA per 100 bases of fragment length, different amounts of water were added according to the table below:

DNA template (µL)	2	3	4	5	6	7	8	9	10	12	14
H_2O (µL)	11	10	9	8	7	6	5	4	3	1	0

In addition to the template and dd H_2O, 5 µL of 5x dilution buffer (Better Buffer, The Gel Company, San Francisco, CA), 1 µL primer (forward and reverse in separate vials), and 1.1 µL of Big Dye 3.1 (Applied Biosystems, Foster City, CA) solution were added. The 0.5 ml tubes were placed in the thermocycler and cycled according to the following parameters: 1 min at 94°C, 15 cycles of 96°C for 15 s, 50°C for 15 s, and 4 min at 60°C, 25 cycles of 96°C for 15 s, 48.5°C for 15 s, and 4 min at 60°C, 25 cycles of 96°C for 15 s, 47°C for 15 s, and 4 min at 60°C, followed by 60°C for 10 min and a 40°C soak.

Sequencing: Both strands of each product were cycle sequenced in 20 µL reactions using 8-fold diluted Big Dye 3.1 (Applied Biosystems). Sequencing reactions were cleaned using Centri-sep columns (Princeton Separations, Adelphia, NJ), electrophoresed through a 6% polyacrylamide gel using an MJ Research BaseStation Automated DNA Sequencer (Bio-Rad, Hercules, CA), and analyzed using Cartographer 1.2.7 software (Basten et al. 1994). Sequences from opposing strands were reconciled and verified for accuracy using Sequencher 4.2.2 (Gene Codes, Ann Arbor, MI). Sequences are archived in GenBank.

DNA alignment and phylogenetic analysis: Exon-intron boundaries were elucidated using Sequencher. DNA sequences were exported from Sequencher as an unaligned nexus file. MacClade (Maddison and Maddison 2005) was used to create an NBRF-formatted file which was then read into Clustal X (Thompson et al. 2001) for alignment. Alignment was straightforward using the default parameter of the program, except for the intron. The data matrix can be obtained from the author, upon request, as a NEXUS- or NBRF-formatted file.

Nucleotides were subjected to parsimony and Bayesian analysis. Parsimony searches used PAUP*'s (4.0b10) heuristic search option, with TBR branch rearrangement. Characters were treated as unordered and maxtrees was set to increase incrementally. Most-parsimonious trees were found by conducting 1,000 searches, with each search begun from trees acquired by

stepwise addition with a random addition sequence order. The aforementioned intron was excluded in some analyses.

Bayesian analysis was conducted using MrBayes 3 (Ronquist and Huelsenbeck 2003) using the model suggested by Modeltest (Posada and Crandall 1998). Each Markov chain in the Bayesian search was started from a random tree and run for 1×10^6 to 2×10^6 cycles, sampling every $1{,}000^{th}$ cycle from the chain. Four chains were run simultaneously, three hot and one cold, with the initial 1,000 cycles discarded as burn-in. Stationarity was evaluated by monitoring likelihood values graphically. Each simulation was run twice. We used the default settings for the priors on the rate matrix (0-100), branch lengths (0-10), gamma shape parameter (0-10), and the proportion of invariant sites (0-1). Base frequencies were estimated from the data.

Node support was evaluated by nonparametric bootstrap resampling (Felsenstein 1985). Bootstrap values for parsimony analyses were calculated from 500 replicates, with each replicate consisting of three searches starting with a tree built by stepwise addition using the simple addition sequence. A maxtrees of 500 was imposed.

III. Tomoceridae in Great Smoky Mountains National Park and the southern Appalachians

i. Results

Eight species were found in GSMNP and the surrounding areas within the Appalachian Mountains. This list includes the five species previously known to occur (*Pogonognathellus bidentatus, P. dubius, P. elongatus, P. flavescens,* and *Tomocerina lamellifera*), two species new to science (*Pogonognathellus danieli* and *P. mystax*), and one resurrected species (*Pogonognathellus nigritus*).

Key to Tomoceridae collected in this study

1) Dens without spine-like scales at inner base (Fig. 6a), mucro with prominent lamella running from basal tooth to anteapical tooth…….***Tomocerina lamellifera***

1') Dens with pair of spine-like scales at inner base (Fig. 6b)……...**2**

2) Larger spines in the middle of dental spines series (Fig. 6c); tenent hair pointed……....***Pogonognathellus bidentatus***

2') Larger spines at terminal end of dental spine series of 2nd subsegment (Fig. 6d)…….....**3**

3) Ground color a shade of gray, purple, or black……..……**4**

3') Ground color yellow, sometimes with small areas of pigmentation……...**6**

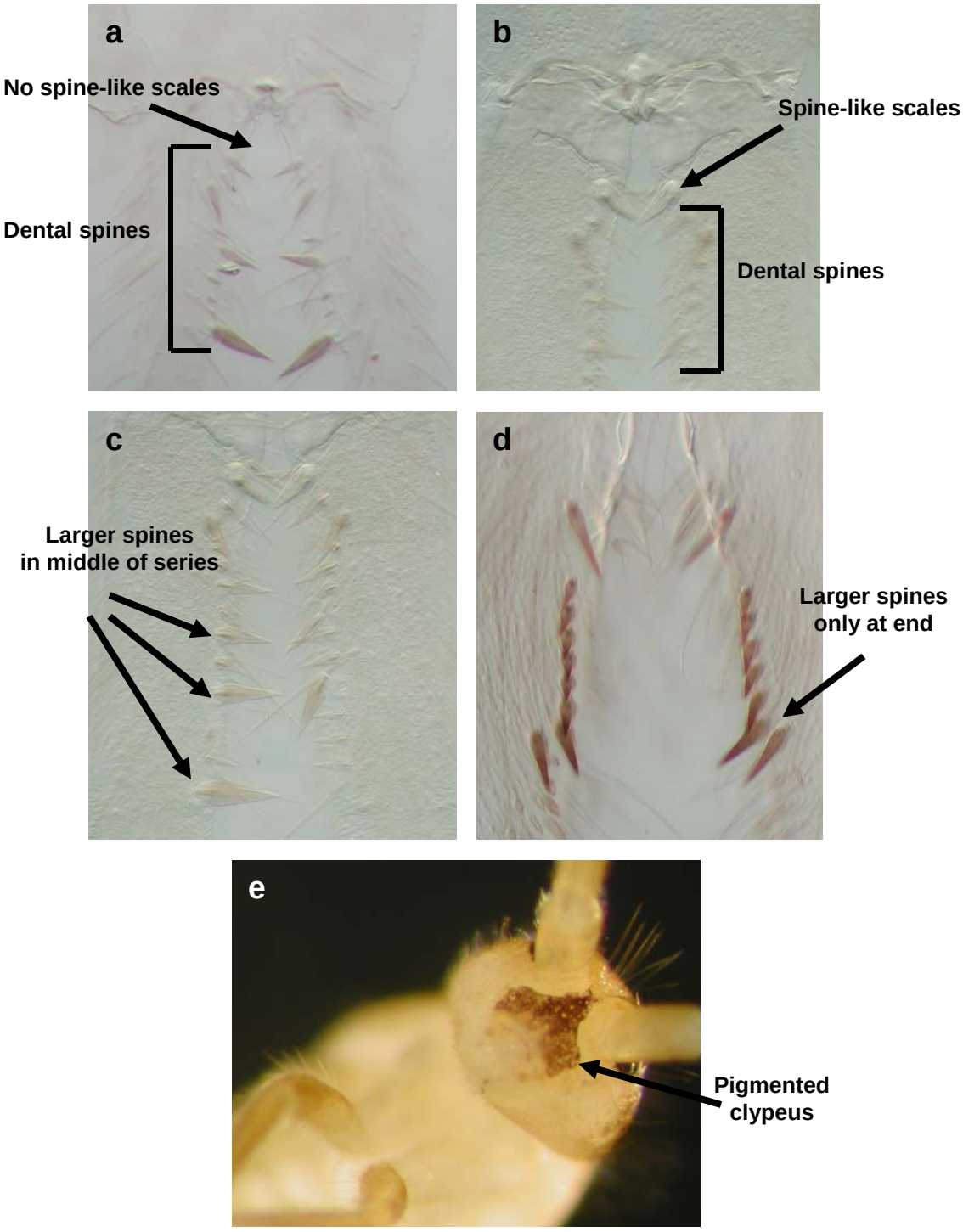


Figure 6 a-e. Figures of the taxonomic key; a) spine-like scales absent at inner base; b) spine-like scales present; c) large spines present in middle of spine series; d) large spines only at terminal end of spine series; e) pigmented clypeus.

4) Ground color pale to dark purple; hind femur with prominent purple band; 3 ungual teeth; 7-9 mucronal teeth……..***P. nigritus***

4') Ground color light to dark patchy gray; 2 ungual teeth, rarely 3……..**5**

5) Abd. III with 2 anterior macrochaetae; 1 macrochaetae on each side of bothriothrix on Abd. III; mesochaetae absent on Abd. V; ground color light to medium gray; 2 ungual teeth; 0-1 unguicular teeth…….***P. dubius***

5') Abd. III with 1 anterior macrochaeta; macrochaeta medial of bothriothrix on Abd. III; mesochaetae present on Abd V; ground color medium to dark gray, almost black; 2 or 3 ungual teeth, 1 unguicular tooth, cave species……***P. danieli***

6) Clypeus with triangular purple pigmentation (Fig. 6e); frequently a purple band on metafemur; 3 or 4 ungual teeth…..……...***P. mystax***

6') Clypeus white to pale yellow; 3 ungual teeth…….**7**

7) Brown pigmented lateral band on Th. II & III; 4 spine-like setae on inner side of tibiotarsus…….***P. elongatus***

7') Brown pigmented bands absent on thorax; inner side of tibiotarsus without spine-like setae…….***P. flavescens* complex**

ii. Molecular data

A parsimony bootstrap/Bayesian inference tree was developed from the sequences from 46 individuals, excluding the two species serving as outgroups (Fig. 7). Several species formed monophyletic groups: *P. elongatus, P. nigritus, P. mystax, and P. danieli*. *Pogonognathellus dubius* and *P. flavescens* did not show close relationships among all of the specimens, indicating these are likely species complexes.

The relationship between *P. flavescens* of Europe and Nearctic *flavescens*-like individuals indicate that true *P. flavescens* may not occur in the United States. *Pogonognathellus bidentatus* showed the most distant relationship to the rest of the ingroup studied. Of the numerous specimens of *P. bidentatus* for which amplifications of the gene were attempted, only one specimen (*P. bidentatus* 371) was successfully sequenced. Attempts to sequence this gene from *Tomocerina lamellifera* also were unsuccessful.

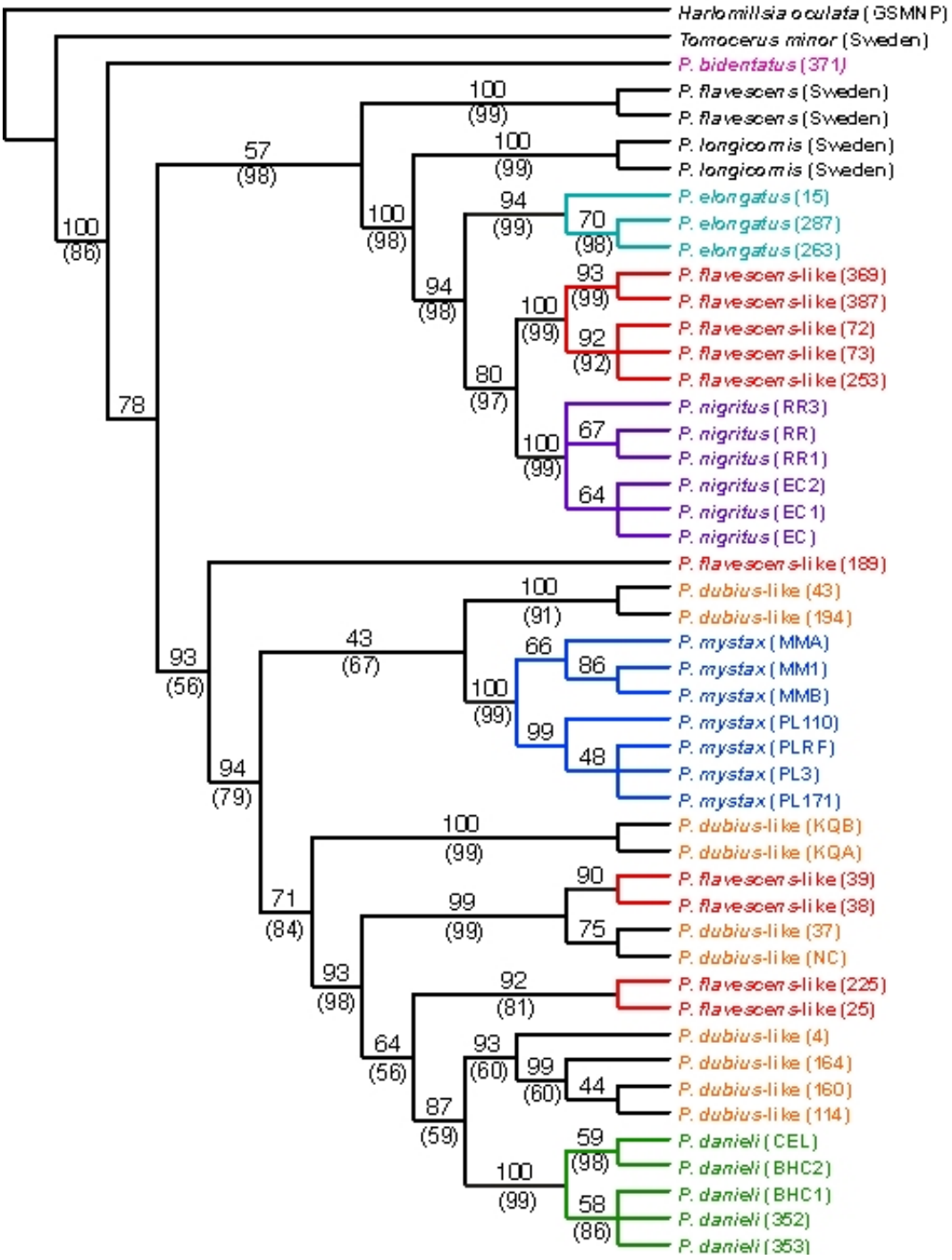


Figure 7. Parsimony bootstrap/Bayesian inference tree; scores above the line indicate confidence of the node using parsimony bootstrap; scores below the line in parentheses are Bayesian scores. The code following the species or species complex indicates the specimen used.

iii. Systematics

***Pogonognathellus bidentatus* (Folsom)**

Figs. 8, 9

Tomocerus bidentatus Folsom, 1913:463, pl. 40 Figs. 7, 8.

= *Tomocerus bidentatus jeanneli* Bonet, 1934:363, Fig. 1.

= *Tritomurus henroti* Delamare, 1949:117, Figs. 1-4.

= *Tomocerus (Pogonognathellus) bidentatus* Christiansen, 1964:668, Figs. 4, 30, 40, 41, 59, 77, 90.

Maximum Length: 5 mm.

Color: Ground color pale yellow to orange (Fig. 8b). Eye patches dark blue to black, trapezoidal. Antennal segments with dark purple to gray pigment or with segments I and II yellow. Tibiotarsi with pale purple to gray pigmentation. Scales medium gray to brown, scale bands at posterior margin of each segment slightly darker (Fig. 8a). Iridescent white scale patches at bothriotrichal origins on Th. II and III and Abd. I, III, and IV.

Head: Antennae shorter than body. Second lamella of maxilla with fringed proximal shelf, lanceolate projection with 3 small denticles, otherwise lacking subapical fringe (Fig. 8d).

Chaetotaxy: Cluster of numerous setae on anterior edge of Th. II. Abd. III, IV & V with 4 posterior macrochaetae. Caudal setae short and straight.

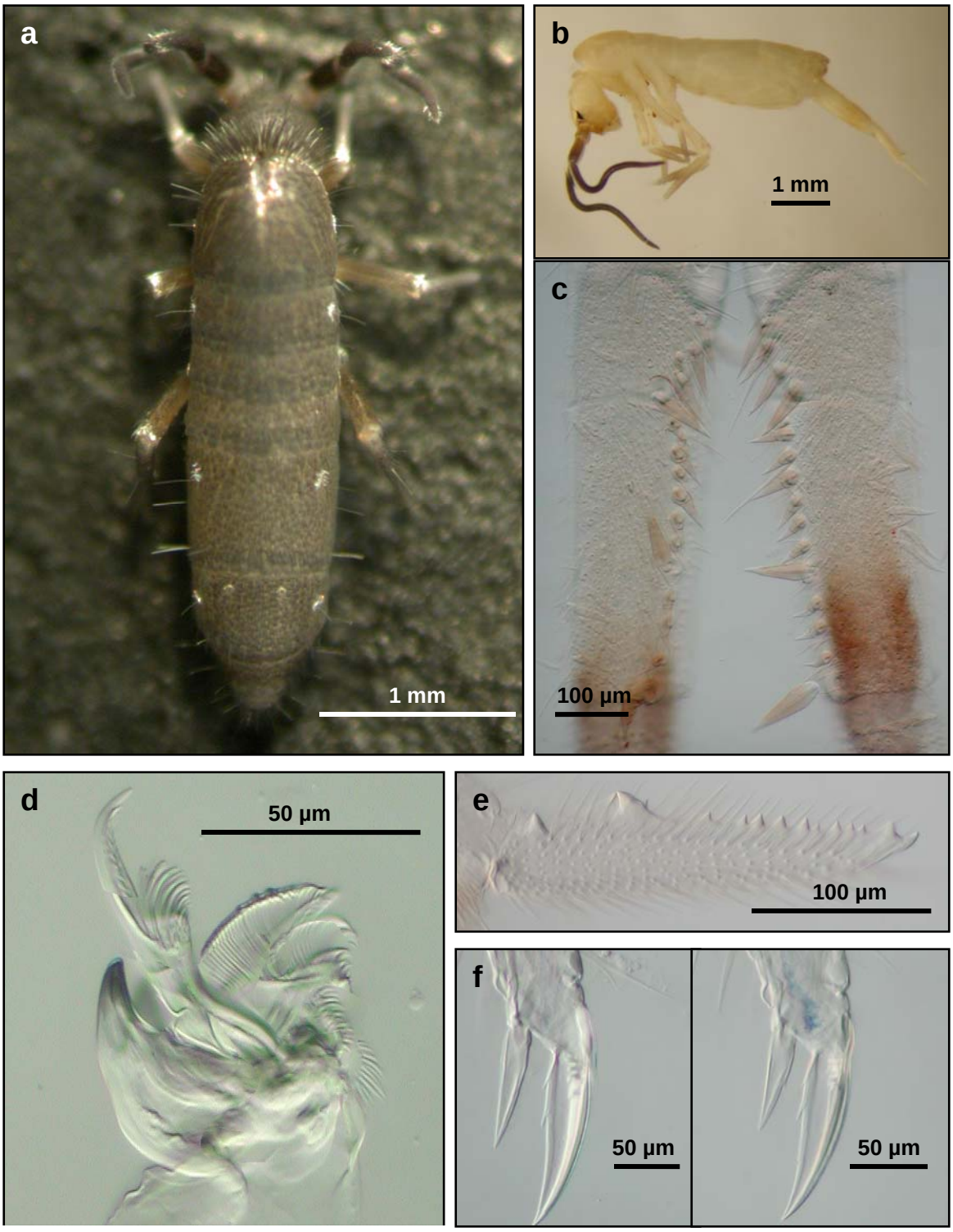


Figure 8 a-f. *Pogonognathellus bidentatus* a) habitus; b) preserved; c) dental spines; d) maxilla; e) mucro; f) foot complex, without and with unguicular tooth.

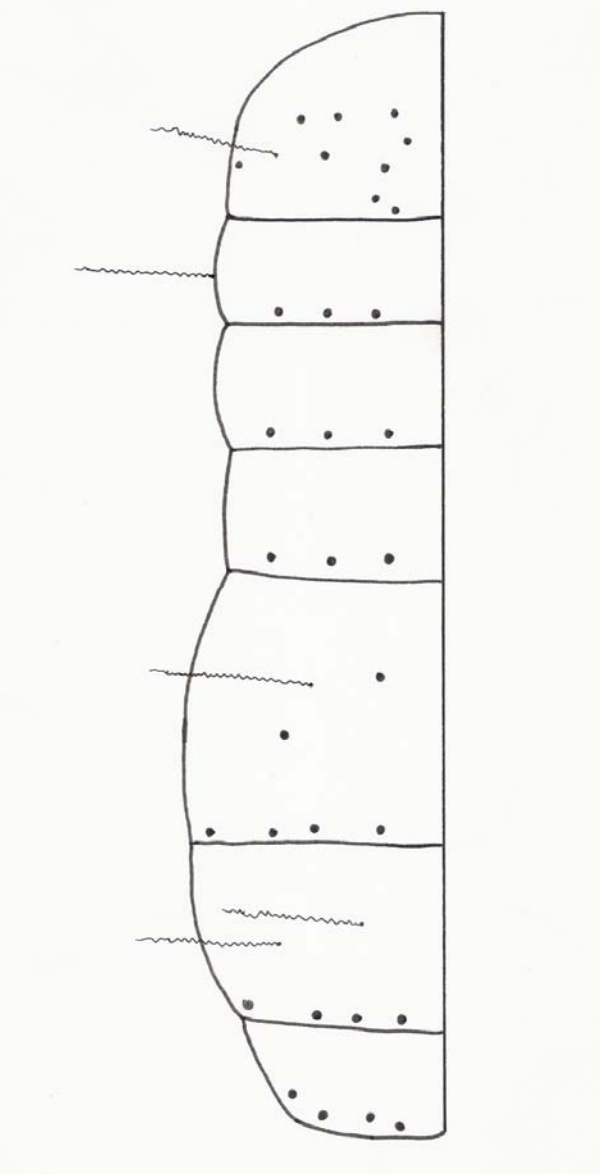


Figure 9. *Pogonognathellus bidentatus*, chaetotaxy. Filled circles indicate macrochaetae; wavy lines indicate bothriotricha.

Legs: Inner ungual teeth usually 2, rarely 3 or 4 (Fig. 8f). Unguiculus lanceolate, usually without inner tooth, infrequently with 1 tooth. Tenent hair pointed or rarely weakly clavate. One or two large, spine-like setae on inner surface of metatibiotarsi.

Furcula: Manubrium lacking spine-like setae. Dens with spine-like scale at inner base. Dental spines brown, smooth to finely striate, terminal spines and some inner spines considerably longer than others, formula 2-5, 1/2-6, 1, 0-2, 0-2, 2-4, 1 (Fig. 8c). Mucro elongate with numerous ciliate setae and basal lamella and with 7-10 intermediate teeth; basal tooth separated from side tooth by more than one-quarter length of mucro (Fig. 8e).

Ecology: Found in moist leaf litter in low and high elevations.

Localities and numbers of specimens studied: Ten specimens, Blount County, Tennessee, GSMNP: 1 specimen, Abrams Falls, leaf litter, 20 August 2006, K. L. Felderhoff and A. B. Hawrysko, collectors; 4 specimens, Gregory Cave, mixed litter from outside cave entrance, 27 July 2006, K. L. Felderhoff, P. J. Long, M. MacCarroll; and 5 specimens, White Oak Sinks, near Blow Hole Cave, leaf litter and sweep net, 28 July 2006, K. L. Felderhoff, P. J. Long, M. MacCarroll. Seven specimens, Sevier County, Tennessee, GSMNP: 2 specimens, Roaring Fork, seep on roadside, 6 September 2006, K. L. Felderhoff and E. C. Bernard; 2 specimens, Clingmans Dome, pitfall trap, 25 October-10 November 2000, C. Parker, I. Stocks, M. Petersen; and 3 specimens, Goshen Prong, pitfall trap, 25 October-10 November 2000, C. Parker, I. Stocks, and M.

Petersen. Two specimens, Knox County, Tennessee, John Sevier Hunter Education Center, Rifle Range Road, mixed pines and hardwoods leaf litter, 25 October 2006, K. L. Felderhoff and E. C. Bernard.

Discussion: Folsom (1913) described the tenent hair as clavate, but this was an error as noted by Christiansen (1964). The possession of a weak inner unguicular tooth is a frequent but not typical variation in the species and the tooth may be present or absent on different feet of the same specimen. Christiansen described the species as primarily a cave form, but specimens seen in this study were collected in leaf litter and pitfall traps, not in caves.

Pogonognathellus bidentatus can be easily recognized by its relatively small eyepatch, compared with other *Pogonognathellus* spp., and the dental spine arrangement with long middle spines. In culture, live *P. bidentatus* can be recognized by the unusual twisting of the antennae (Fig. 8a) when an air current is passed over the specimen. Their antennae draw back closer to the head and curl irregularly when disturbed.

Pogonognathellus danieli **new species**

Fig. 10, 11

Maximum length: 4 mm.

Color: Color (in alcohol) light or dark gray to black on tergites, larger specimens blackish gray (Fig. 10a). Pigment not uniform. Larger specimens

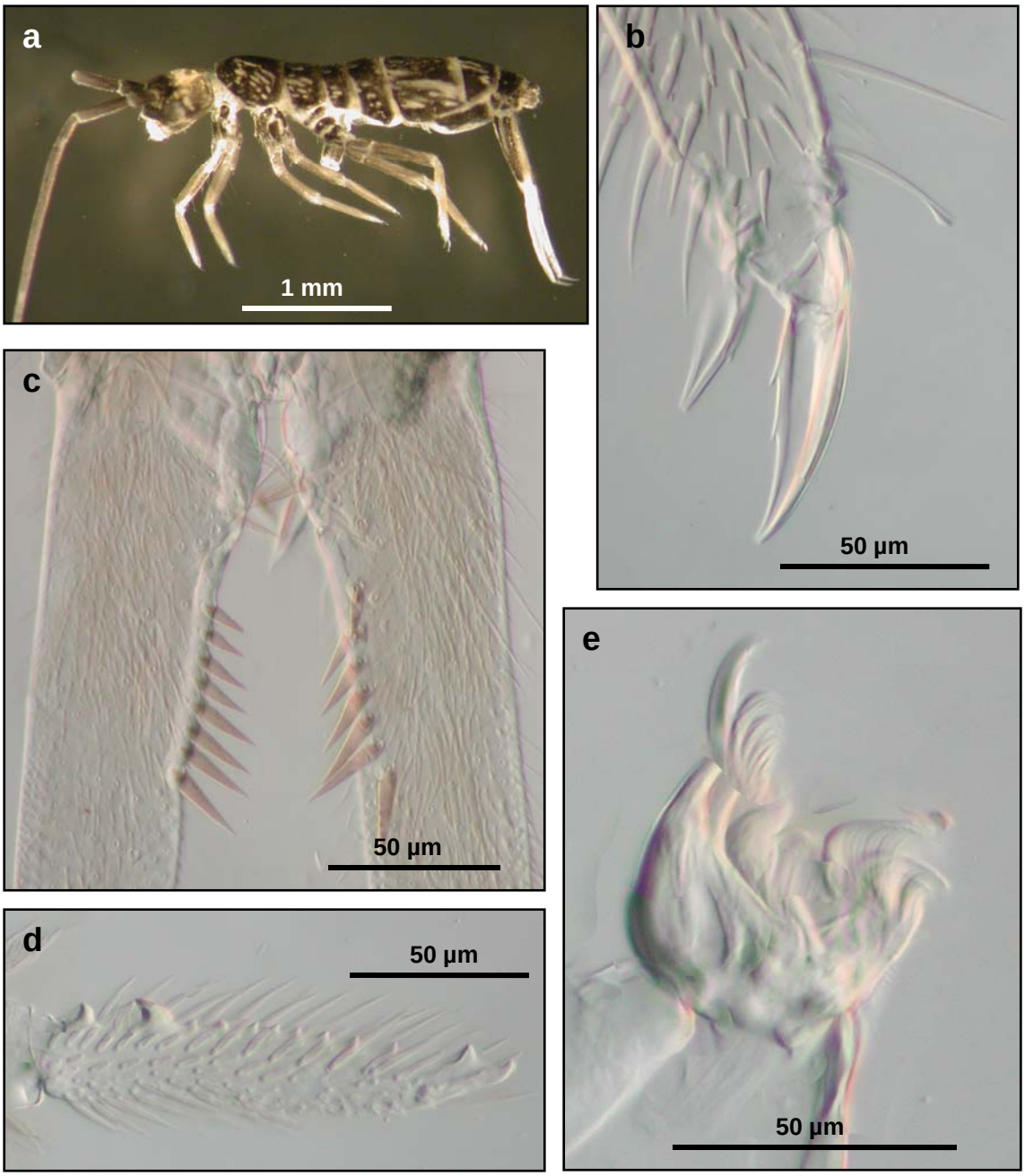


Figure 10 a-e. *Pogonognathellus danieli* a) preserved; b) foot complex; c) dental spines; d) mucro; e) maxilla.

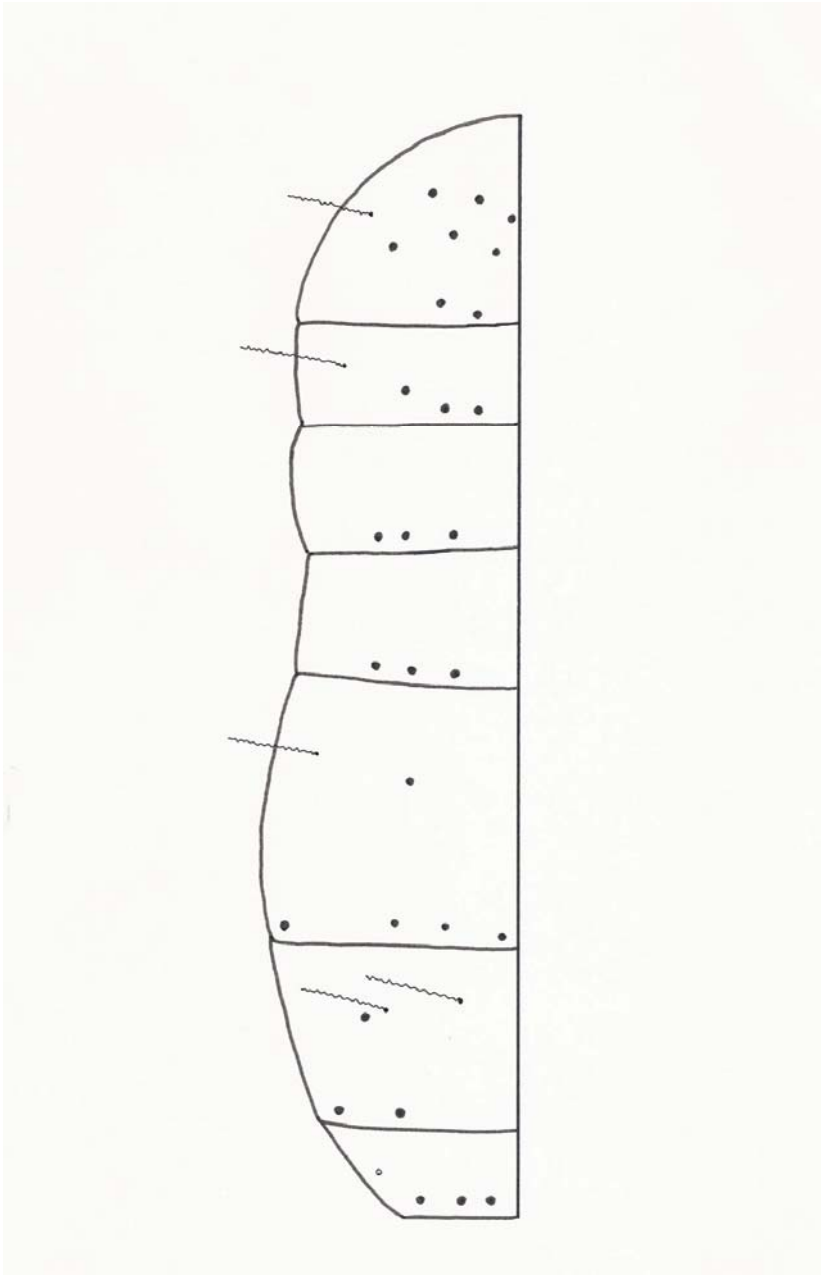


Figure 11. *Pogonognathellus danieli*, chaetotaxy. Filled circles indicate macrochaetae; open circles indicate mesochaetae; wavy lines indicate bothriotricha.

darker. Manubrium dark, dens and mucro pale. Legs dark, intersegmantal membranes pale. Scale pattern unknown.

Head: Antennae shorter than body. Second lamella of maxilla with long fringes to tip, heavier cluster of fringe on basal half (Fig. 10e).

Chaetotaxy: 4 posterior and 1 anterior macrochaetae on Abd. III, 2 bothriotricha medial to anterior macrochaetae on Abd. IV (Fig. 11).

Legs: Inner ungual teeth usually 2, rarely 3. Unguiculus lanceolate, 1 inner tooth (Fig. 10b). Tenent hair weakly clavate, much shorter than unguis. One or two large, spine-like setae on inner surface of metatibiotarsi.

Furcula: Dens with spine-like scale at inner base. Dental spines brown, smooth to finely striate, terminal spines gradually increasing in length, formula 0-3, 0-2/5-7, 1-2 (Fig. 10c). Mucro elongate with numerous ciliate setae, apical and anteapical teeth, basal lamella and 6-8 intermediate teeth; basal tooth one-fourth length of mucro from side tooth, and less than half the size of side tooth (Fig. 10d).

Ecology: Found in leaf litter in limestone cave (karst).

Type specimens: One holotype, 6 paratypes, Blount County, Tennessee, GSMNP, White Oak Blowhole Cave, 28 July 2006, J. Mays, collector. Two specimens, Blount County, Tennessee, GSMNP, White Oak Blowhole Cave, leaf litter from cave entrance drop, 28 July 2006, K. L. Felderhoff, J. Hilten.

Etymology: Named for the author's cousin, Daniel Reiter.

Diagnosis: *Pogonognathellus danieli* is closely related to *P. celsus* Christiansen and is found in a similar habitat. Morphologically, the two differ significantly. The tenent hair of *P. danieli* is always clubbed, whereas *P. celsus* is described as having at least one pointed tenent hair. The maxilla also differs; *P. danieli* has longer fringes that extend half way up the second lamella; *P. celsus* has shorter fringes restricted to the basal fourth. The unguis sometimes has both 2 and 3 inner teeth on the same individual, and frequently on the same pair of legs. Paratype specimens of *P. celsus* were obtained for morphological comparison from K. Christiansen but were not suitable for genetic analysis.

***Pogonognathellus dubius* (Christiansen)**

Figs. 12, 13

Tomocerus (Pogonognathellus) dubius Christiansen, 1964:671, Figs. 3, 25, 61, 79, 94.

Maximum length: 3.5 mm.

Color: Ground color white with light mottled gray pigment on tergites. Gray pigmentation covering all appendages (Fig. 12b). Eye patches black, triangular. Small triangular patch of dark pigment between bases of antennae. Antennal segments with gray pigment. Scales uniform grayish to silver (Fig. 12a).

Head: Antennae shorter than body. Second lamella of maxilla with long fringe to tip, basal half with denser fringes (Fig. 12e).

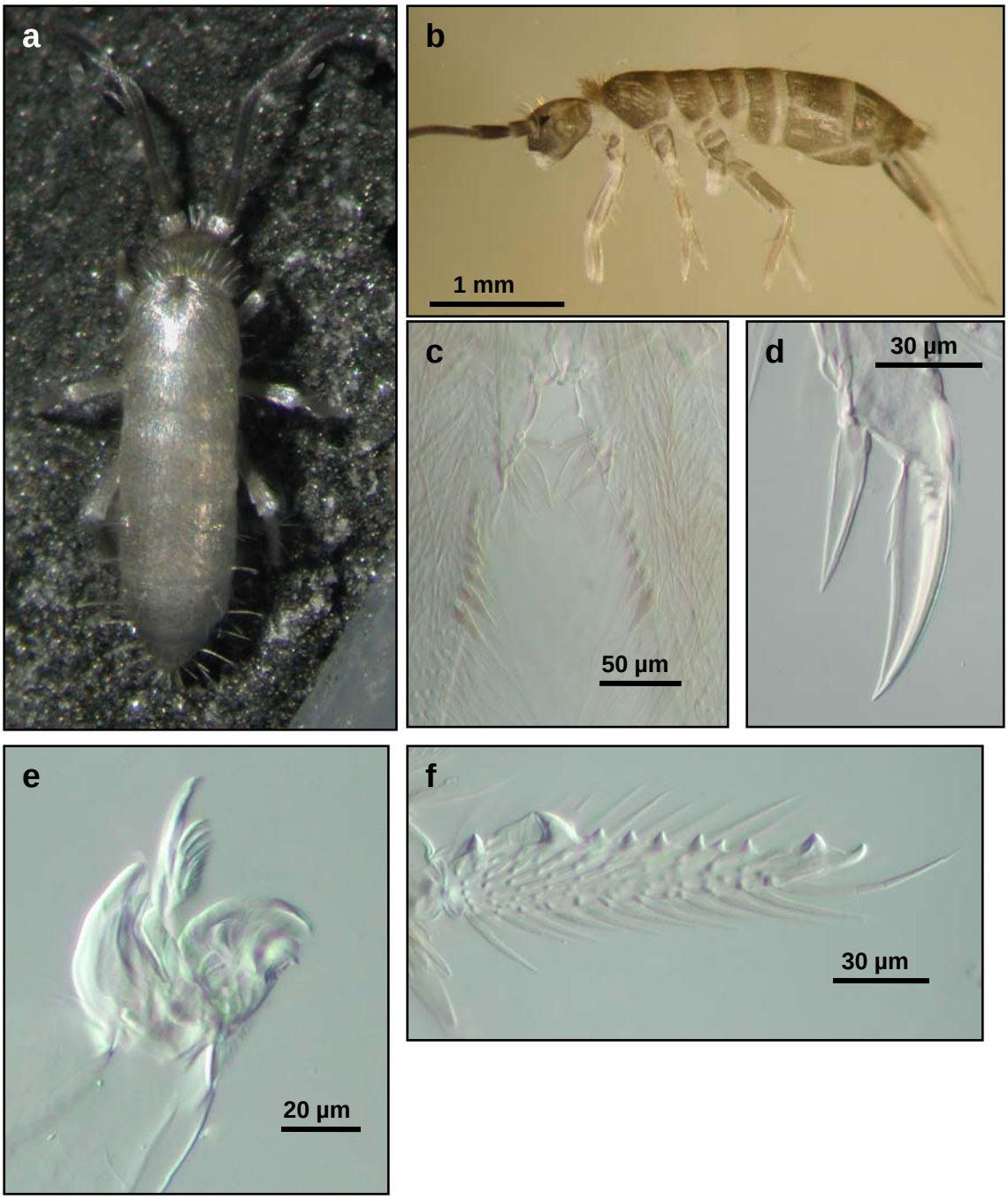


Figure 12. *Pogonognathellus dubius* a) habitus; b) preserved; c) dental spines; d) foot complex; e) maxilla; f) mucro.

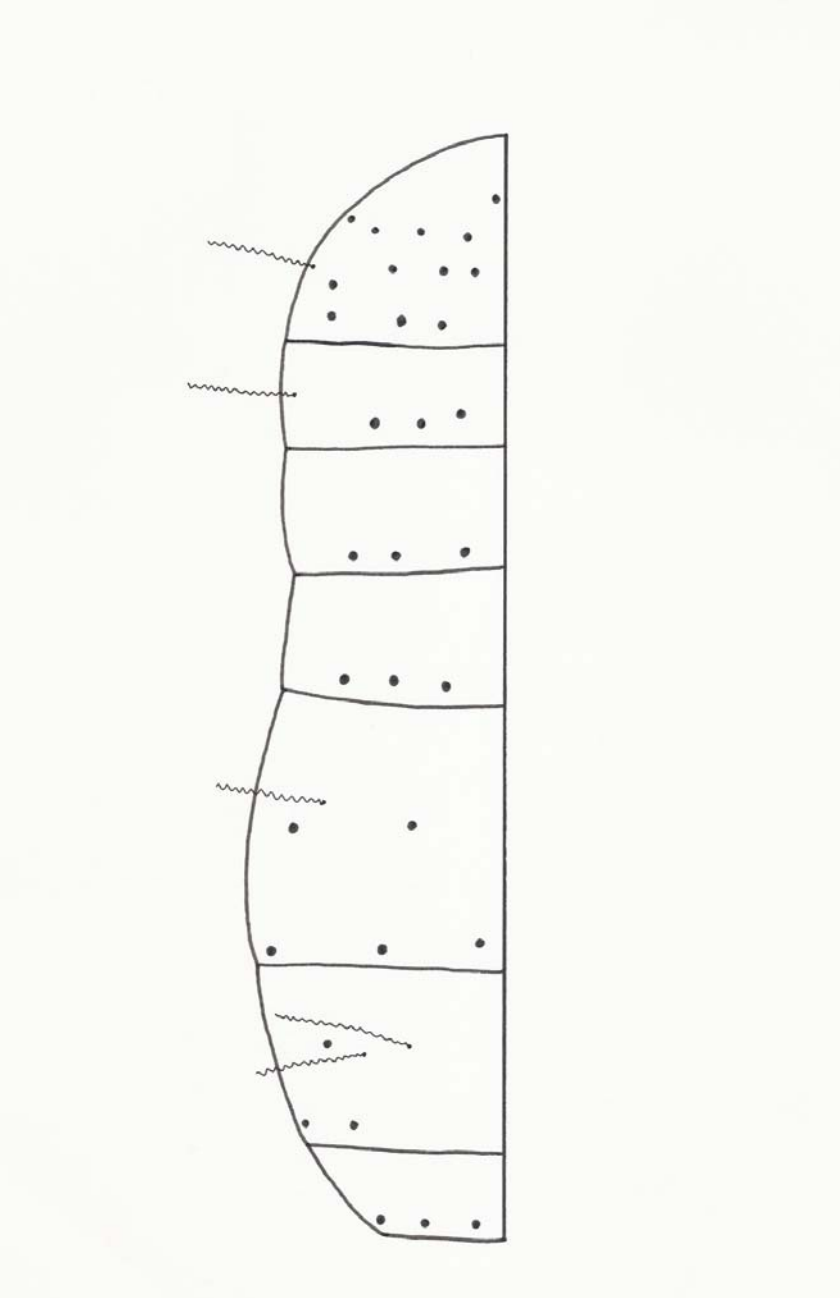


Figure 13. *Pogonognathellus dubius*, chaetotaxy. Filled circles indicate macrochaetae; wavy lines indicate bothriotricha.

Chaetotaxy: 2 bothriotricha medial to anterior macrochaetae on Abd. IV (Fig. 13).

Legs: Unguis with 2 inner teeth. Unguiculus lanceolate, with or without small inner tooth (Fig. 12d). Tenent hair clavate. One large, spine-like seta on inner surface of metatibiotarsus.

Furcula: Dens with spine-like scale at inner base. Dental spines brown, smooth to finely striate, terminal spines considerably longer than others, formula 0-2, 1/4-6, 2-3 (Fig. 12c). Mucro elongate with numerous ciliate setae, basal lamella and 5-7 intermediate teeth; basal tooth at least one-fourth the mucro length from side tooth, and smaller than half its size (Fig. 12f).

Ecology: Found in moist leaf litter in low and high elevations.

Localities and number of specimens studied: Six specimens, Blount County, Tennessee, GSMNP: 4 specimens, Gregory Cave, mixed litter from outside cave entrance, 27 July 2006, K. L. Felderhoff, P. J. Long, M. MacCarroll, collectors; and 2 specimens, White Oak Sinks, near Blow Hole Cave, leaf litter and sweep net, 28 July 2006, K. L. Felderhoff, P. J. Long, M. MacCarroll. One specimen, Sevier County, Tennessee, GSMNP, Roaring Fork, seep on roadside, 6 September 2006, K. L. Felderhoff and E. C. Bernard. One specimen, Swain County, North Carolina, GSMNP, Noland Ridge at Noland Creek headwater monitor site, mixed leaf litter, 6 September 2005, K. L. Felderhoff and E. C. Bernard.

Discussion: *Pogonognathellus dubius* was considered a part of the former *P. flavescens* complex until Christiansen separated it morphologically in his 1964 publication. Based on molecular data, this species is distinct from the *P. flavescens*-like complex found here and the *P. flavescens* from the type locality in Sweden. I identified *P. dubius* based on characters that Christiansen (1964) used to distinguish it from *P. flavescens*. However, specimens identified as *P. dubius* did not cluster in the morphological analysis, suggesting that this species, too, may comprise a complex of species that will require additional analysis.

***Pogonognathellus elongatus* (Maynard)**

Figs. 14 -16

Tomocerus (Pogonognathus) elongatus Maynard, 1951:126, Figs. 223-230.

= *Maynardia elongata* Yosii, 1956: 77.

= *Tomocerus (Pogonognathellus) elongatus* Christiansen, 1964:672-673, Figs. 5, 6, 27, 39, 67.

Maximum Length: 6 mm.

Color: Ground color pale yellow with dark brown lateral band on Th. II and III (Fig. 14c). Eye patches black, trapezoidal. Small pigmented triangle between base of antennae, small pigmented patch between eye and antennal base. Antennal segments with pale purple to gray pigment on distal parts of segments III and IV. Scales medium to dark brown covering dorsal surface, forming a pattern of weakly iridescent, alternating areas of light and dark scale patches on

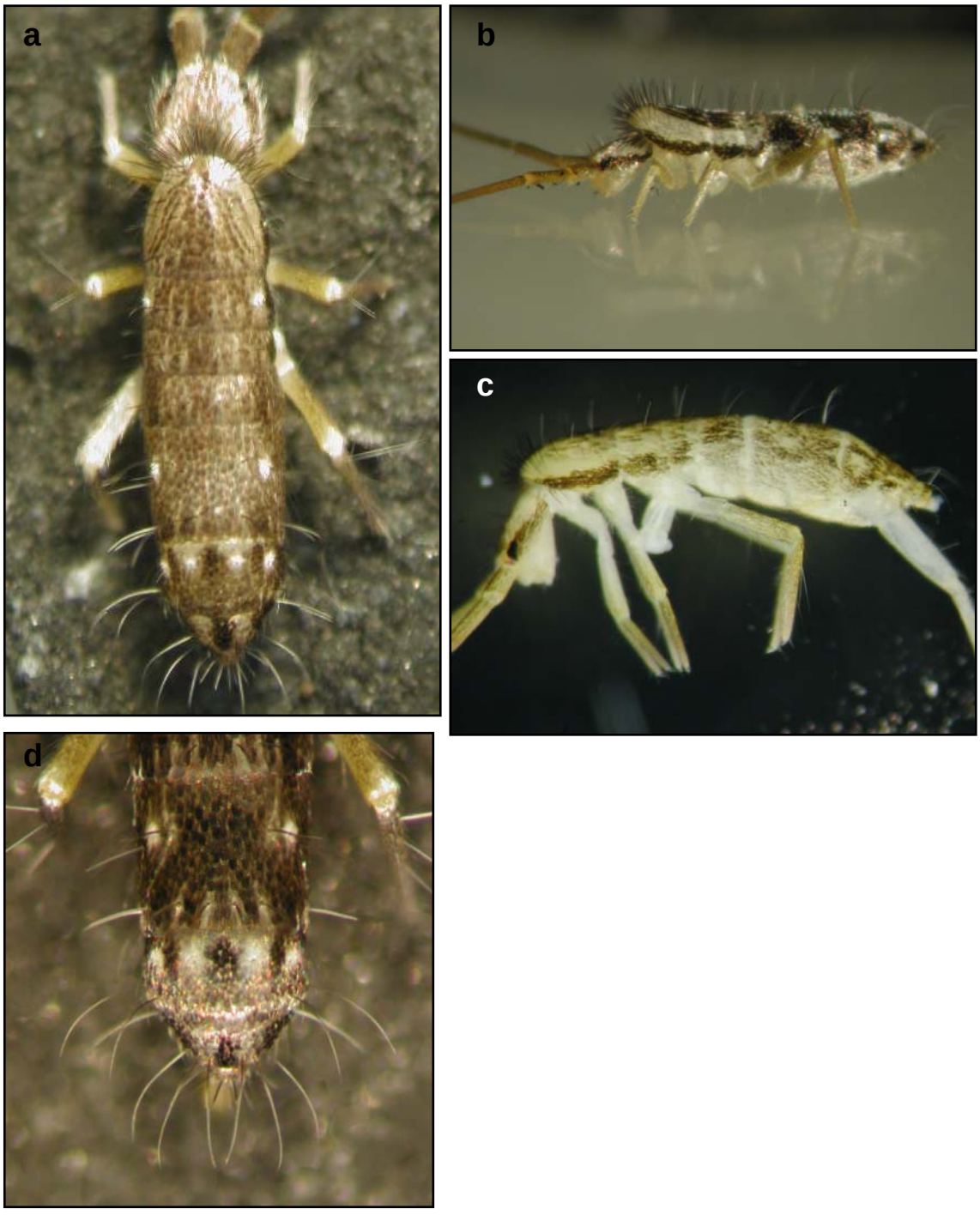


Figure 14 a-d. *Pogonognathellus elongatus* a) habitus; b) habitus, lateral view; c) preserved, lateral view; d) posterior abdomen, close-up of scales and setae.

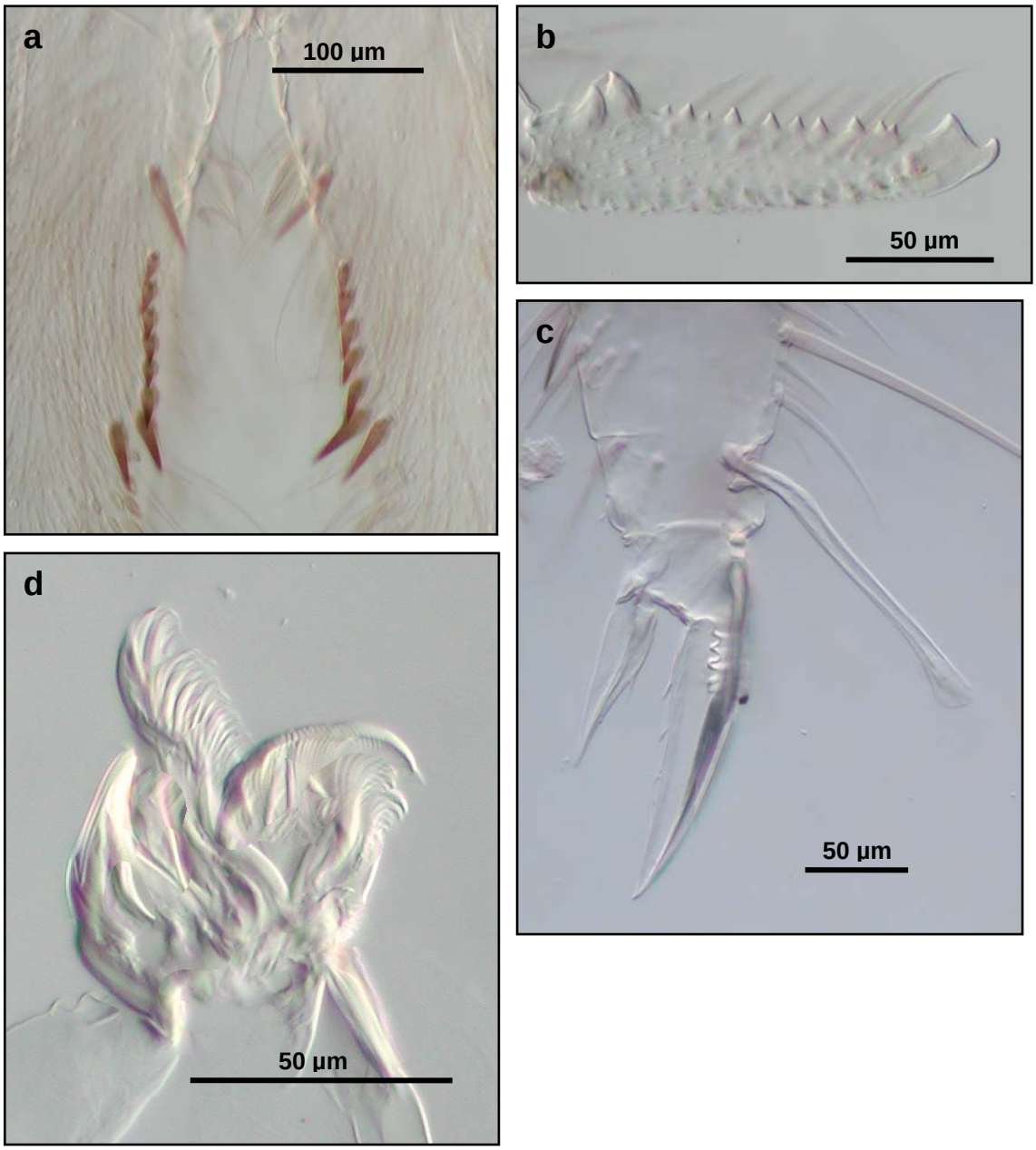


Figure 15a-g. *Pogonognathellus elongatus* morphology a) dental spines; b) mucro; c) foot complex; d) maxillae.

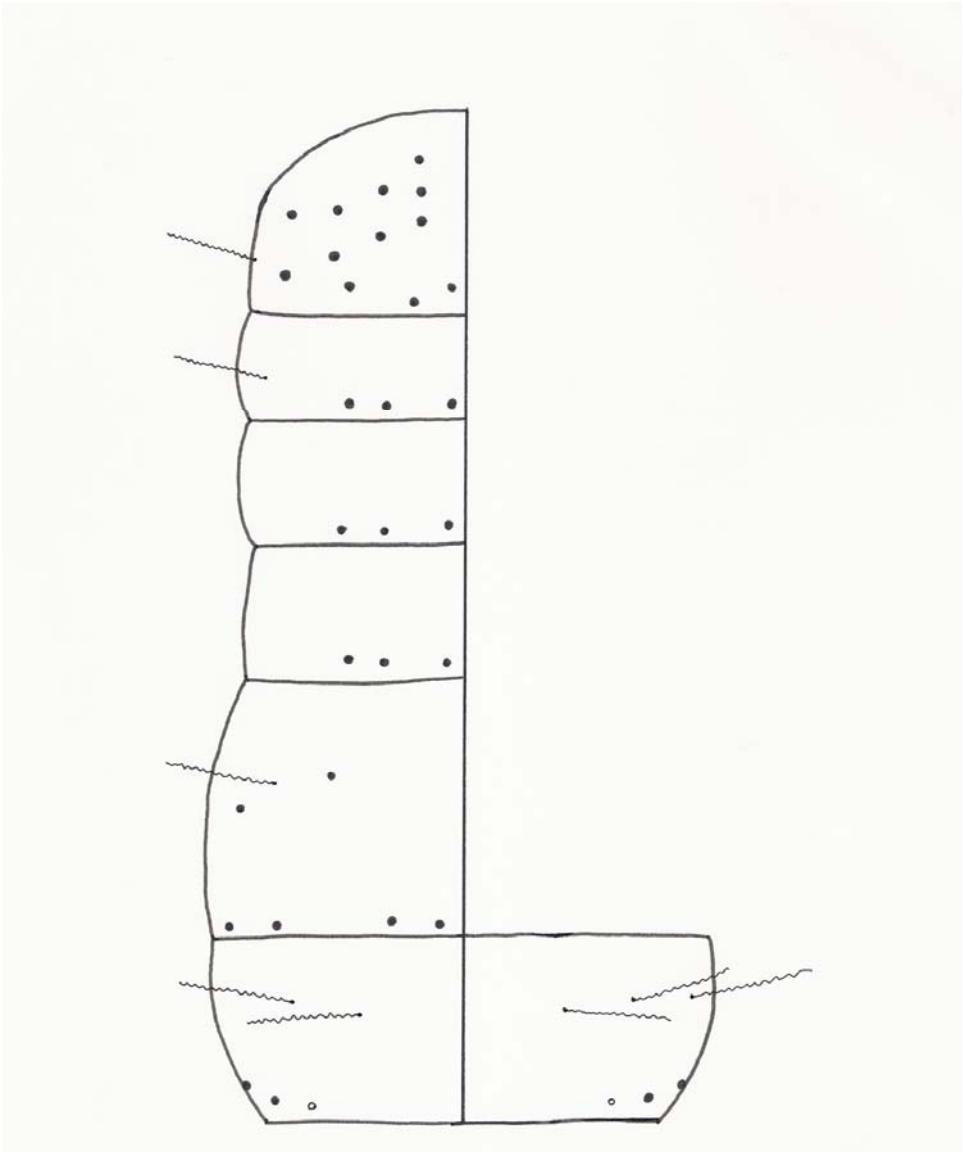


Figure 16. *Pogonognathellus elongatus*, chaetotaxy. Filled circles indicate macrochaetae; open circles indicate mesochaetae; wavy lines indicate bothriotricha. Usual condition of Abd. IV on left, unusual condition with 3 bothriotricha on right.

Abd. IV and V (Fig. 14a). Dark brown lateral band stretching from base of antennae, through eye patch, to Abd. I, with pale yellow or white band directly above (Fig. 14b). Iridescent white scale patches at bothriotrichal origins on Th. III, Abd. III, and Abd. IV (Fig. 14a, 14d).

Head: Antennae longer than body. Second lamella of maxilla with long fringes to the apex (Fig. 15d).

Chaetotaxy: 4 posterior macrochaetae and bothriothrix between 2 anterior macrochaetae on Abd. III. Mesochaetae medial to 2 posterior macrochaetae and frequently 3 bothriotricha on Abd. IV (Fig. 16). Caudal setae of distal segment short and straight, setae of preceding segments long and curved, directed caudally (Fig. 14d).

Legs: Unguis with 3 inner teeth. Unguiculus lanceolate, usually with inner tooth, occasionally 1-2 smaller proximal teeth (Fig. 15c). Tenent hair stout, clavate. Four spine-like setae on inner surface of metatibiotarsus.

Furcula: Manubrium with 2 pairs of spine-like setae, dens with 1 pair. Dens with spine-like scale at inner base. Dental spines brown, smooth to finely striate, terminal spines considerably longer than others, formula 0, 1/6-9, 2 (Fig. 15a). Mucro elongate with numerous ciliate setae, basal lamella, and 7-10 intermediate teeth; basal tooth even with side tooth and over half its size (fig. 15b).

Ecology: Found in moist leaf litter in low and high elevations. This species often climbs up blades of grass or lower branches of trees, and may be collected in Malaise traps.

Localities and numbers of specimens studied: Seventeen specimens, Swain County, North Carolina, GSMNP: 4 specimens, Enloe Cemetery, rotting wood and leaf litter, 3 April 2006, K. L. Felderhoff, collector; 2 specimens, same locality, 19 June 2006, K. L. Felderhoff; 10 specimens Mingus Mill, leaf litter, 19 June 2006, K. L. Felderhoff; Purchase Knob, March 2007, K. L. Felderhoff and E. C. Bernard; and 1 specimen, Noland Ridge at Noland Creek headwater monitor site, mixed leaf litter, 6 September 2005, K. L. Felderhoff and E. C. Bernard. Thirteen specimens, Sevier County, Tennessee, GSMNP: 5 specimens, Roaring Fork Motor Nature Trail, mixed leaf litter, 6 September 2005, K. L. Felderhoff and E. C. Bernard; 4 specimens, litter from seep, 13 March 2006, K. L. Felderhoff and E. Alexander; and 4 specimens, Laurel Falls trail, 11 July 2006, K. L. Felderhoff. Seven specimens, Blount County, Tennessee, GSMNP: 1 specimen, Gregory Cave, leaf litter right outside cave entrance, 27 July 2006, K. L. Felderhoff and M. MacCarroll; 4 specimens, Rainbow Cave, leaf litter collected near waterfall, 28 July 2006, K. L. Felderhoff; and 2 specimens, White Oak Sink, aspirated from leaf litter, 28 July 2006, K. L. Felderhoff. Ten specimens, Mitchell County, North Carolina, Mount Mitchell State Park, small ferns and low branches on side of trail, 12 July 2007, K. L. Felderhoff.

Discussion: This distinctive species is easy to separate from other North American *Pogonognathellus* by its intricate scale pattern, lateral banding on Th. II and III, and unusually long antennae. Christiansen (1964) and Christiansen and Bellinger (1980, 1998) described the many specimens they examined from

across North America as having a blue to gray ground color, and expressed uncertainty over the description by Maynard. In particular, they assumed that Maynard had confused the surfaces of the manubrium, and so did not accept that the manubrium possessed four prominent blunt, spines. This uncertainty was due in part to the unavailability of the type specimens, which were kept in Maynard's private collection until long after he passed away. I was able to borrow types from NMNH and confirm the general accuracy of Maynard's description. Christiansen's blue/gray specimens, ranging from Alaska to Pennsylvania, need to be reexamined to determine their identities.

Among other *Pogonognathellus* species, *P. elongatus* is closely related to *P. nigritus* on the basis of the prominent spines of the manubrium. This character prompted Yosii (1956) to establish a separate genus, *Maynardia*, for these two species. The molecular analysis conducted in this project clearly places these species within the limits of *Pogonognathellus*, confirming the synonymization of *Maynardia* with *Pogonognathellus* by Christiansen (1964).

Some specimens exhibit a prominent, anteriorly projecting mesonotal hump similar to that in several *Lepidocyrtus* species (Entomobryidae). Both females and males can have this hump. The function of this hump is not clear; its presence and size vary among individuals collected in the same location, and seasonality also does not appear to influence its presence. This species has not been successfully reared in culture, so it is not yet possible to follow the

development of individuals from egg to adult to determine if the hump is a developmental or reproductive trait.

This species is distinct from *P. nigritus* (Maynard 1951), which was synonymized with it by Christiansen (1964) due to similar morphological characteristics. Because of the smaller pointed tenent hair proximal to the clubbed tenent hair which often becomes dislodged, this may result in what appears to be a single, pointed hair. This species is one of the largest in the family and can be collected with a variety of collection methods (Malaise trap, sweep net, beating cloth, Tullgren funnel).

***Pogonognathellus flavescens* (Tullberg), species complex**

Fig. 17

Macrotoma flavescens Tullberg, 1871:149.

= *Tomocerus alba* Packard, 1877: 158.

= *Tomocerus plumbeus* Packard, 1888:65.

= *Tomocerus arcticus* Schött, 1893: 43, Figs. 8, 9.

= *Tomocerus americanus* Schött, 1893:169.

= *Tomocerus niger* Folsom, 1902:97, Figs. 46, 47.

= *Tomocerus separatus* Folsom, 1913:460, Figs. 1, 2.

= *Tomocerus (Pogonognathellus) flavescens* Christiansen, 1964:673, Figs. 1, 29, 30, 33-36, 54-58, 76, 78, 91-93.

Maximum length: 6.0 mm.

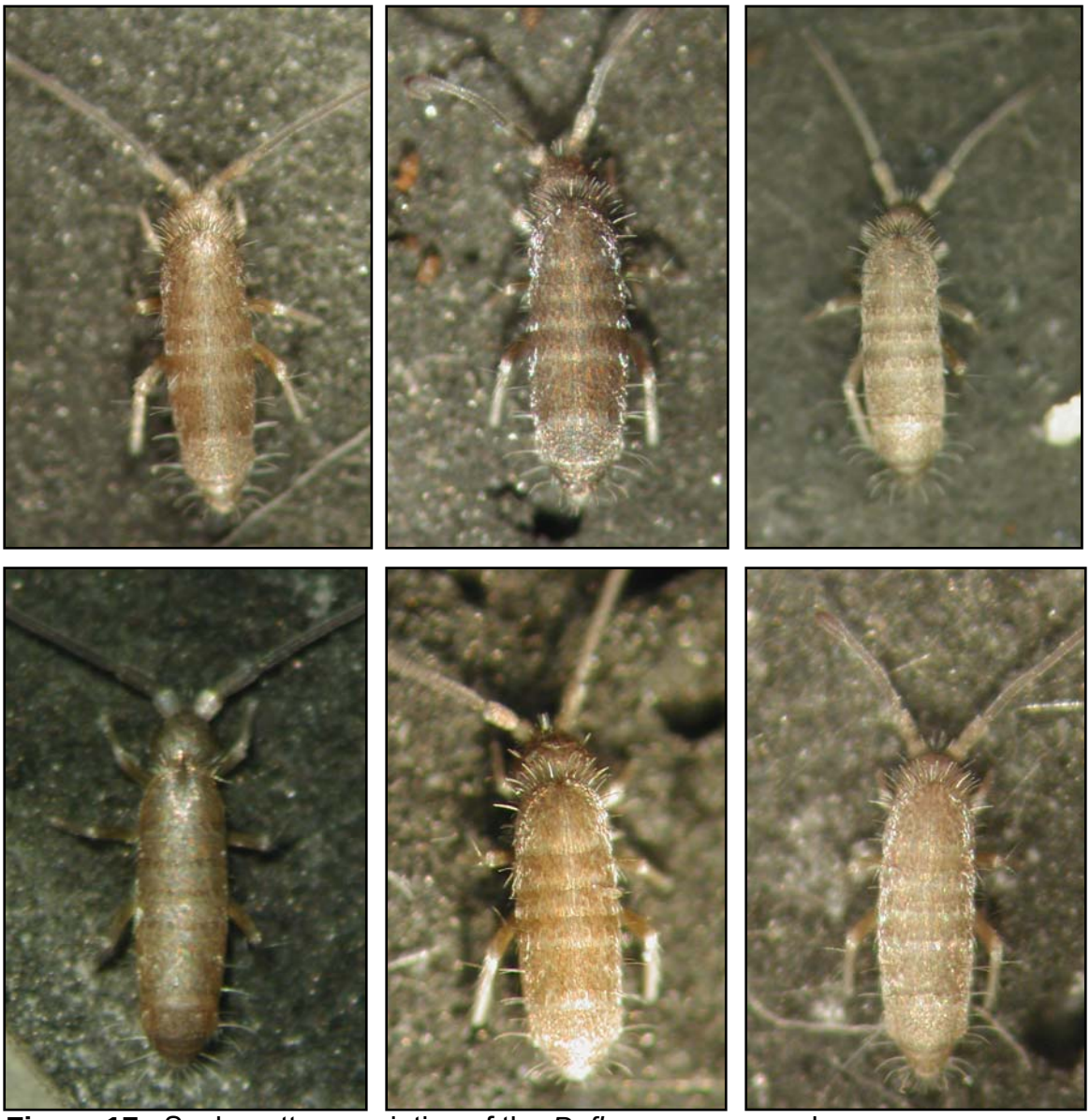


Figure 17. Scale pattern varieties of the *P. flavescens* complex.

Color: Ground color off-white to pale yellow. Eye patches black, trapezoidal. Scales variable: pale to dark brown to silver-gray (Fig. 17).

Head: Antennae shorter than body.

Legs: Unguis with 3 inner teeth. Unguiculus lanceolate, with inner tooth. Tenent hair clavate. One large, spine-like seta on inner surface of metatibiotarsus.

Furcula: Dens with spine-like scale at inner base. Dental spines brown, smooth to finely striate, terminal spines considerably longer than others, formula variable. Mucro elongate with numerous ciliate setae, basal lamella, and variable number of intermediate teeth; basal tooth less than one-fourth length of mucro away from base, and side tooth larger than half the size of basal tooth.

Ecology: Found in a wide variety of habitats, in moist leaf litter and moss, in low and high elevations. This species is a frequent climber on vegetation.

Discussion: Christiansen (1964) studied hundreds of specimens attributable to *P. flavescens* in an attempt to make sense of this complex, concluding that it was a highly variable taxon that could be a group of very similar species not separable by any of the characters he studied. I obtained freshly collected *P. flavescens* from Sweden (type locality) and preserved in 95% EtOH from Arne Fjellberg, for inclusion in this study. On the basis of the molecular analysis, none of the *flavescens*-like specimens collected in this study were conspecific with the European specimens. Thus, the possibility exists that *P. flavescens* may not occur in North America at all, and that all of the many records recorded from

North America are other species. In the molecular analysis, the putative Appalachian *P. flavescens* formed several distinct monophyletic groups sometimes close to putative *P. dubius*. However, not enough specimens were available for an adequate morphological investigation of these apparent taxa. Much of this morphology is based of Christiansen and Bellinger (1998). Many specimens that fit into this complex were found in multiple collecting localities (see Appendix for details). There is much work to be done on the separation of this complex.

***Pogonognathellus mystax* new species**

Figs. 18 - 20

Maximum Length: 4.5 mm.

Color: Ground color white to pale yellow or orange (Fig. 18d). Prominent purple pigmentation patch on clypeus (Fig. 6e). In some populations metafemur occasionally with prominent purple band, and mesofemur with weak purple band (Fig. 18d). Small pigmented spot directly anterior to eyespot. Legs gradually becoming purplish-brown distally. Eye patches black, triangular. Ant. III and IV with purple pigment. Ant. I and II pale yellow, covered in scales. Scales light to medium brown with paler brown to white bands at posterior margins of Th. II and III and all abdominal segments. Iridescent white scale patches at bothriotrichal origins on Th. III, Abd. III and Abd. IV (Fig. 18a,18b,18c).

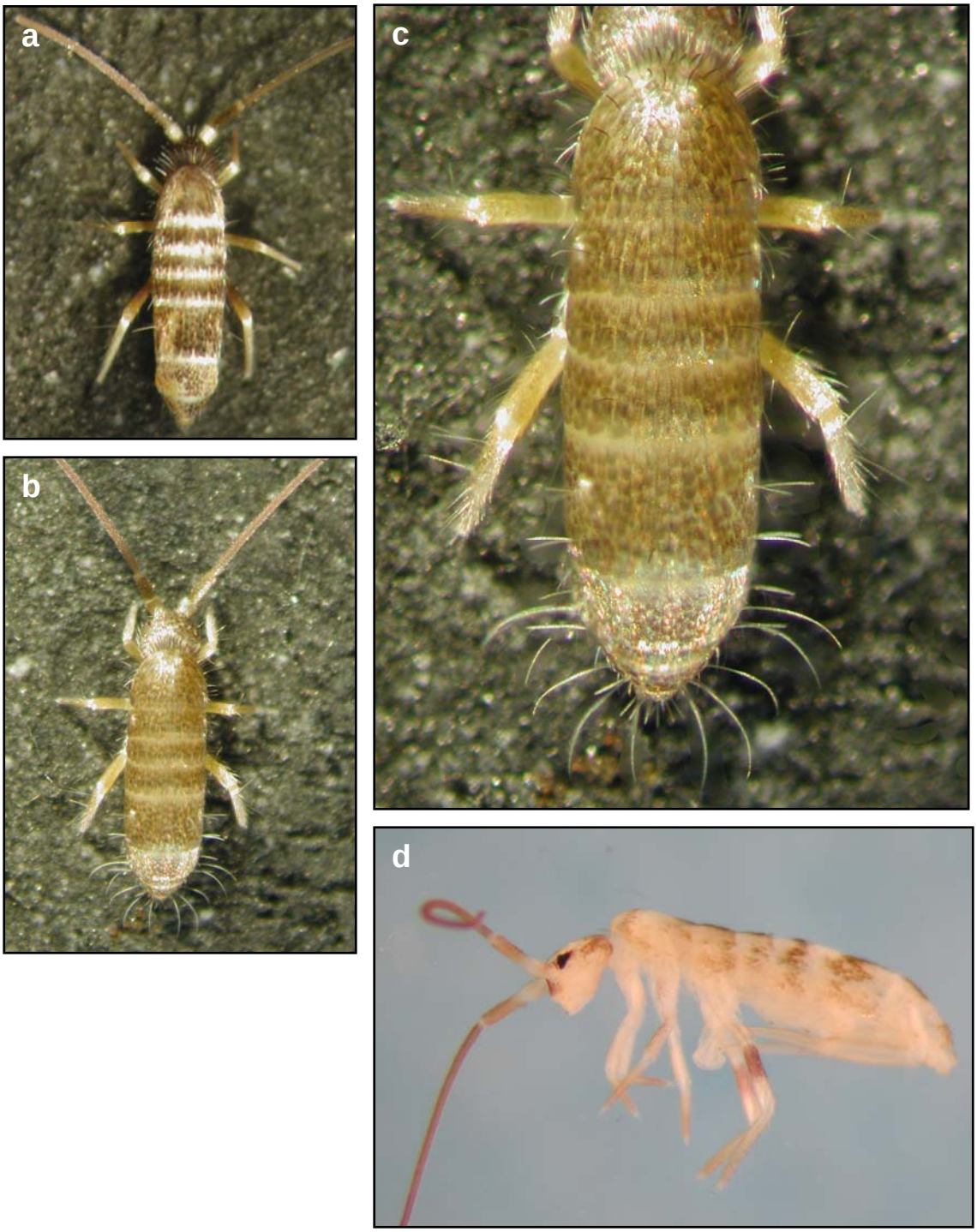


Figure 18 a-d. *Pogonognathellus mystax* a) habitus; b) habitus; c) close up of scale pattern; d) preserved, lateral view.

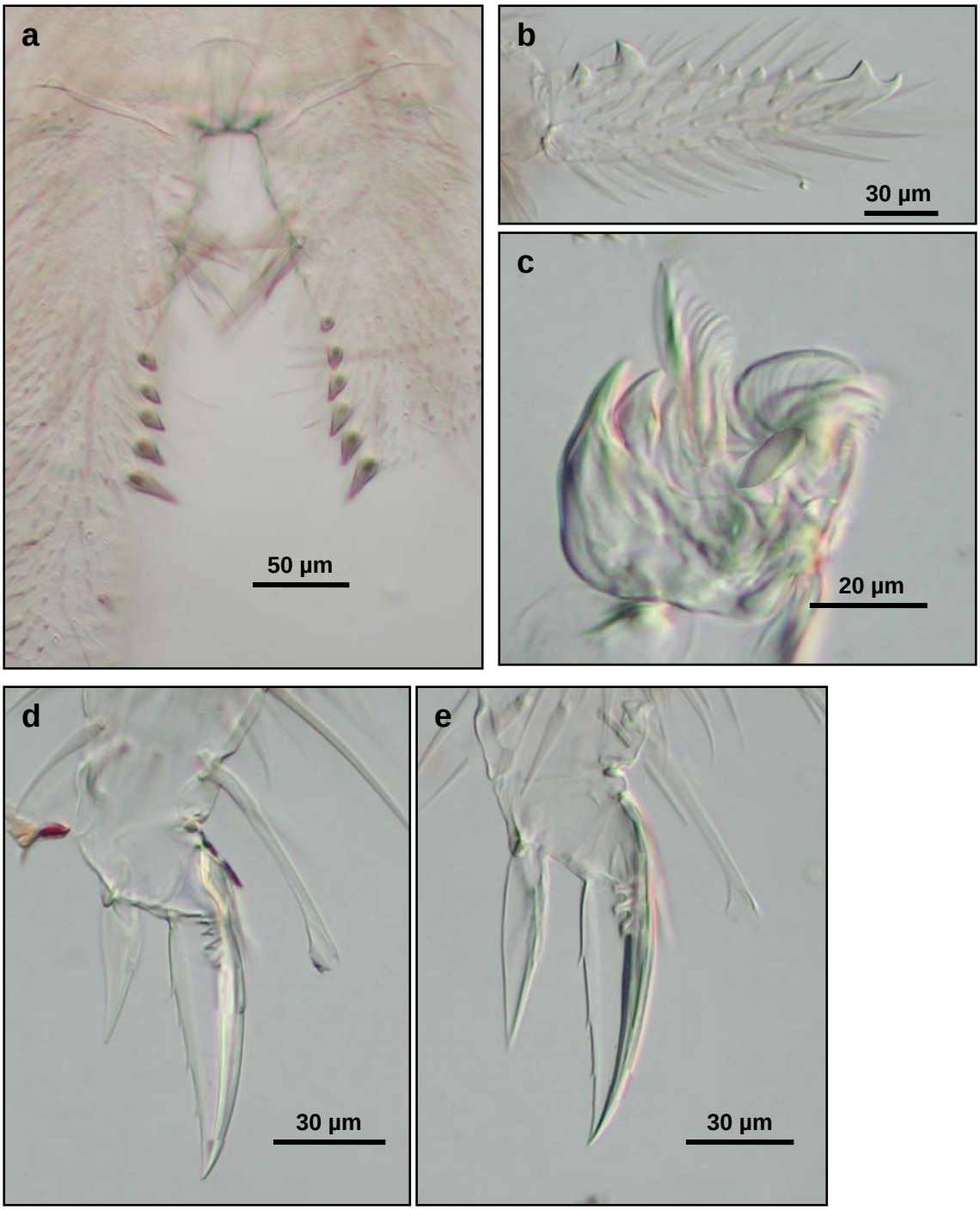


Figure 19a-e. *Pogonognathellus mystax* morphology a) dental spines; b) mucro; c) maxilla; d) foot complex, 4 ungual teeth; e) foot complex, 3 ungual teeth.

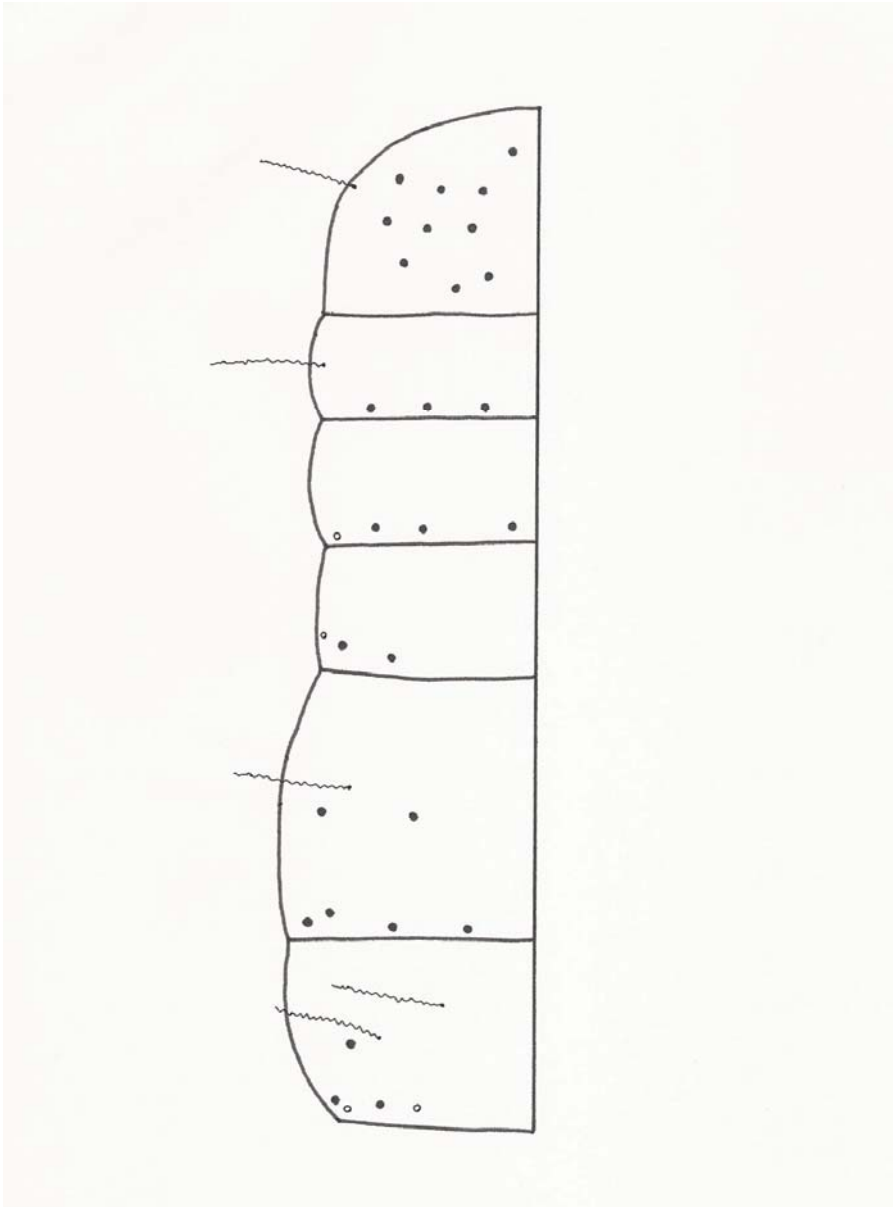


Figure 20. *Pogonognathellus mystax*, chaetotaxy. Filled circles indicate macrochaetae; open circles indicate mesochaetae; wavy lines indicate bothriotricha.

Head: Antennae shorter than body. Second lamella of maxilla with fringes on basal portion (Fig. 19c).

Chaetotaxy: 4 posterior macrochaetae on Abd. III. Lateral mesochaeta on Abd. I and II. 2 posterior mesochaetae associated with the 2 posterior macrochaetae on Abd. IV (Fig. 20). Caudal setae of terminal segment short and straight.

Legs: Unguis with 3 inner teeth, frequently 4 inner teeth (Fig. 19d, 19e) on prolegs or mesolegs; number of teeth sometimes different on ungues of the same pair of legs. Unguiculus lanceolate, with or without small inner tooth. Tenent hair clavate. Five or six large, spine-like setae on inner surface of metatibiotarsus.

Furcula: Dens with spine-like scale at inner base. Dental spines brown, smooth to finely striate, terminal spines considerably longer than others, formula 0-1, 2/3-5, 2 (Fig. 19a). Mucro elongate with numerous ciliate setae, basal lamella, and 6-9 intermediate teeth; basal tooth to base of mucro less than one-fourth the length of mucro. Lateral tooth close to mucro base, level with basal tooth (Fig. 19b).

Ecology: Found in moist leaf litter, in moss, or on low branches at low and high elevations.

Type specimens: Holotype and 26 paratypes, North Carolina, Mitchell County, Mount Mitchell State Park, small ferns and low tree branches along trails, 12 July 2007, K. L. Felderhoff, collector. Ten specimens from following

locations in Sevier County, Tennessee, GSMNP: 3 specimens, Rainbow Falls trailhead, moss on fallen log, 11 May 1999, E. C. Bernard; 1 specimen, Roaring Fork Motor Nature Trail, in seep, 13 March 2006, K. L. Felderhoff; 2 specimens, Little River Road, litter from seep, 20 September 2006, K. L. Felderhoff, 1 specimen, same locality, in moss, 20 September 2006, K. L. Felderhoff, and 3 specimens, same locality, 13 April 1995, R. Hix. Seven specimens, Blount County, Tennessee, Reed Creek, 17 April, 1997, P. J. Obenauer. Two specimens, Swain County, North Carolina, GSMNP: 1 specimen, Noland Ridge at Noland Creek headwater monitor site, mixed leaf litter, 23 June 2006, K. L. Felderhoff, and 1 specimen, Enloe Cemetery, moss on log, 3 April 2006, K. L. Felderhoff. Nine specimens, Avery County, North Carolina, Highway 221 near Crossnore, seep on roadside, 10 March 2006, K. L. Felderhoff.

Etymology: Greek for “moustache”; named for the distinct pigmentation on the clypeus.

Diagnosis: This species is easily distinguished from most other species by the pigmentation on the clypeus. Very similar specimens from other localities with the same pigmented clypeus also have a purple band on the metafemur. These two groups of specimens together form a monophyletic group in the molecular analysis but are segregated within the monophyly into separate subgroups. The significance of this split is yet to be determined.

***Pogonognathellus nigritus* (Maynard)**
Figs. 21, 22

Tomocerus (Pogonognathus) nigritus Maynard, 1951:130, Figs. 231-235.

= *Tomocerus nigrita* Yosii, 1956:77.

= *Tomocerus (Pogonognathellus) elongatus* Christiansen, 1964:672, Figs. 5, 6, 27, 39, 67.

Maximum Length: 5 mm.

Color: Ground color bright or pale purple to brownish-purple, dark purple, or deep purple-black (Fig. 21c). Hind femur with prominent purple band, middle femur with smaller band. Eye patches black, trapezoidal. Antennal segments with purple pigment on segments I and II; segments III and IV pigmented distally. All other appendages white. Scales dark brown to dark gray, strongly metallic. Iridescent white scale patches at bothriotrichal origins on Th. II and III and Abd. I, III, and IV (Fig. 21a, 21b).

Head: Antennae shorter than body. Second lamella of maxilla with fringed shelf, lanceolate projection with 2 small denticles and subapical fringe (fig. 21f).

Chaetotaxy: Lateral mesochaetae on Abd. I & II; Abd. III with 4 posterior macrochaetae; 2 posterior macrochaetae and 1 posterior mesochaetae on Abd. IV; 2 posterior macrochaetae on Abd. V (Fig. 22). Caudal setae of distal segment short and straight, setae of preceding segments long and curved, directed caudally.

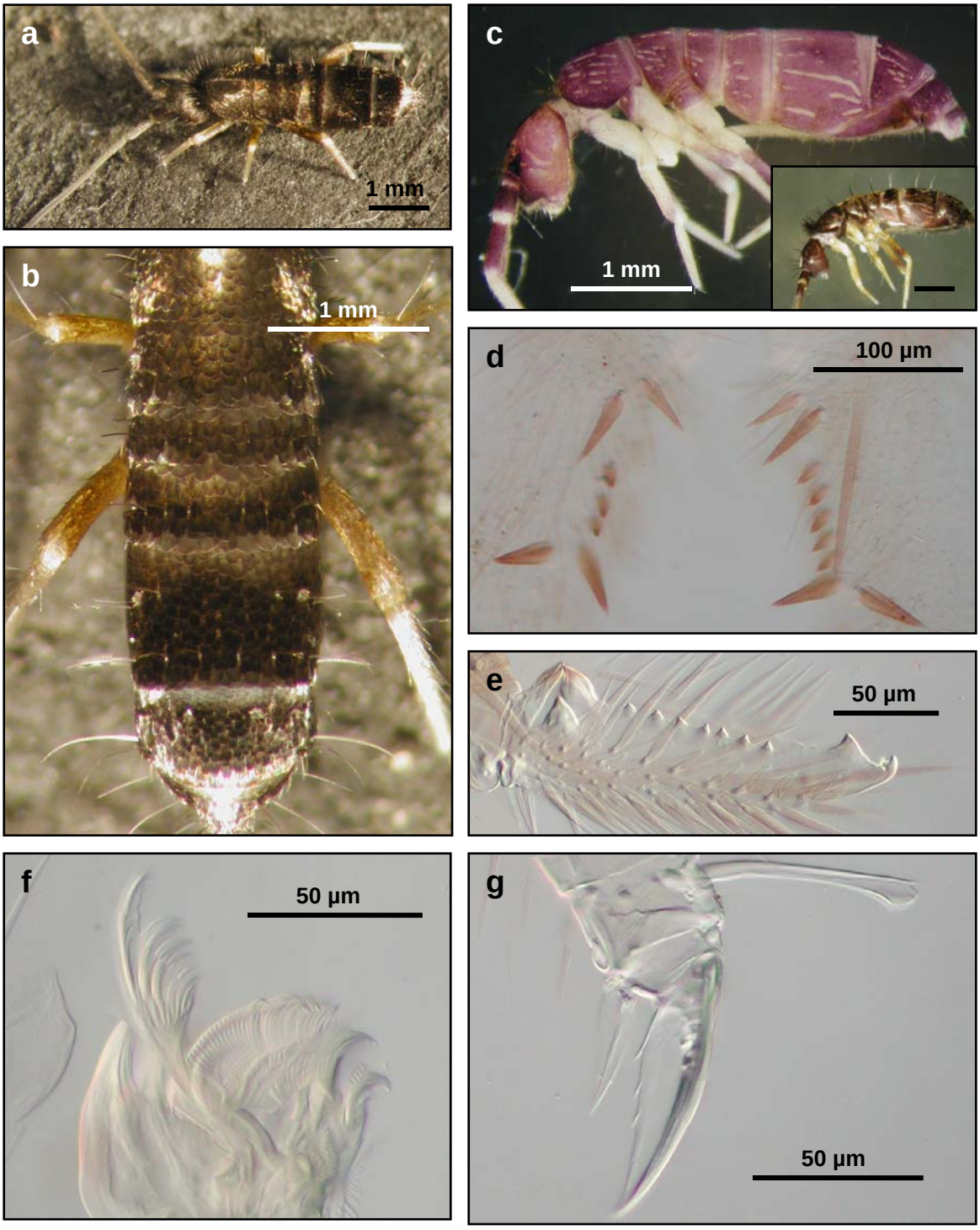


Figure 21 a-g. *Pogonognathellus nigritus* a) habitus; b) scale pattern; c) preserved, two color variations; d) dental spines; e) mucro; f) maxilla; g) foot complex.

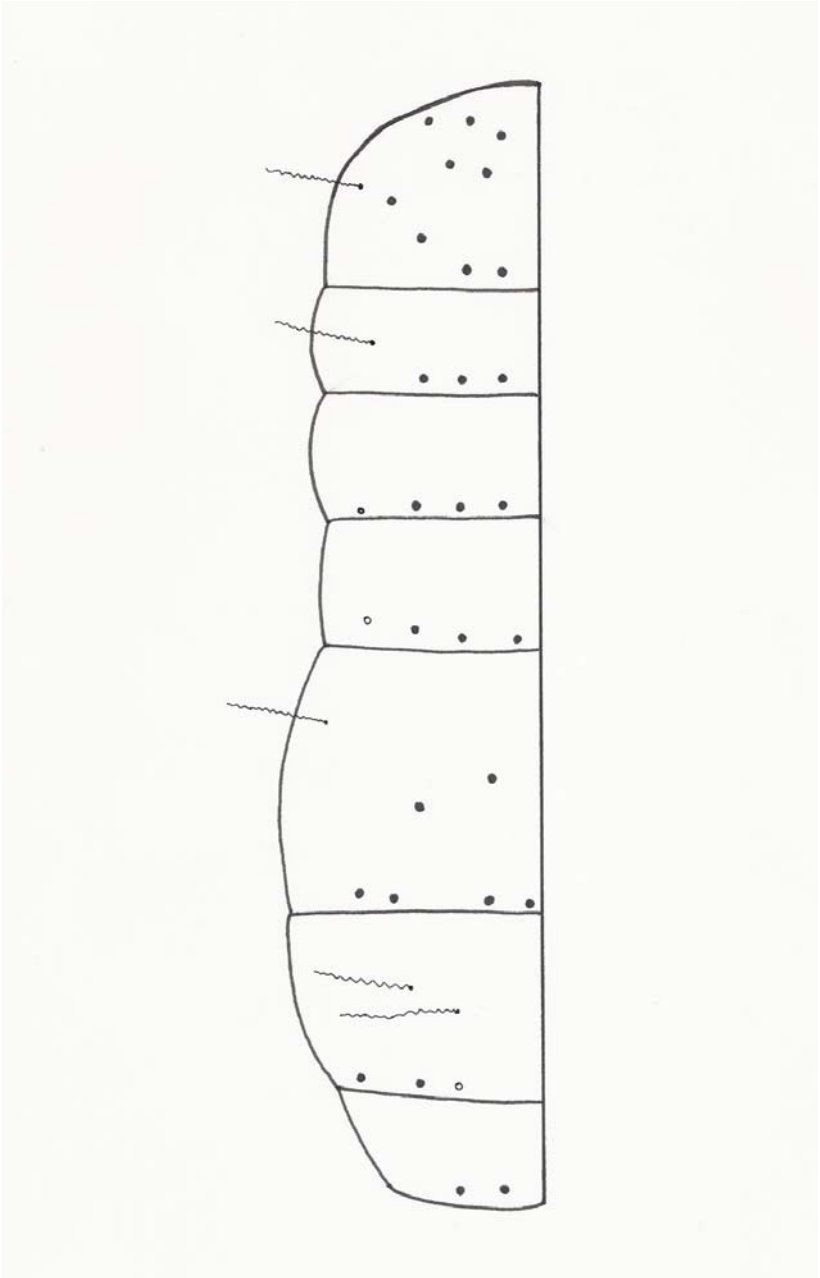


Figure 22. *Pogonognathellus nigritus*, chaetotaxy. Filled circles indicate macrochaetae; open circles indicate mesochaetae; wavy lines indicate bothriotricha.

Legs: Unguis with 3 inner teeth. Unguiculus lanceolate, with or without small inner tooth (Fig. 21g). Tenent hair stout, clavate. Five or six large, spine-like setae on inner surface of metatibiotarsus.

Furcula: Manubrium with 2 pairs of spine-like setae, dens with 1 pair. Dens with spine-like scale at inner base. Dental spines brown, smooth to finely striate, terminal spines considerably longer than others, formula 0, 2/3-5, 1-2 (Fig. 21d). Mucro elongate with numerous ciliate setae, basal lamella and 7-9 intermediate teeth; basal tooth close to side tooth, both teeth near mucronal base (Fig. 21e).

Ecology: Found in moist leaf litter in low and high elevations.

Localities and numbers of specimens studied: Twenty-one specimens, Swain County, North Carolina, GSMNP: 18 specimens, Enloe Cemetery, 15 March 2004, Cherokee High School students, collectors; 1 specimen, same site, January 2007, Cherokee High School students; 1 specimen, Purchase Knob, mixed leaf litter, March 2007, K. L. Felderhoff and E. C. Bernard; and 1 specimen, same locality, 16 October 2003, ATBI Plot, C. Parker, I. Stocks, M. Petersen. Twenty-three specimens, Knox County, Tennessee: 2 specimens, John Sevier Hunter Education Center, Rifle Range Road, mixed pines and hardwoods leaf litter, 25 October 2006, K. L. Felderhoff and E. C. Bernard; 14 specimens, same locality, pitfall trap, site 4, open grasses and vines, 8-11 May 2005, J. H. Lowe and E. E. C. Clebsch; 7 specimens, same locality, pitfall trap, site 3, mixed pine-hardwoods leaf litter, 8-11 May 2005, J. H. Lowe and E. E. C. Clebsch.

Diagnosis: *Pogonognathellus nigritus* differs from its near relative, *P. elongatus*, by many morphological characteristics. The most obvious is ground color; *P. nigritus* is a vivid purple of variable darkness, while *P. elongatus* is typically a pale yellow with a distinct lateral band on Th. II and III. Scale pattern is also very different: *P. nigritus* has deep metallic purple-brown scales; *P. elongatus* has intricate brown and white patterns. Dental spine formula also differs. *Pogonognathellus celsus* has six to nine smaller spines in the dental series; *P. nigritus* never has more than five. Length of the antennae in *P. nigritus* is shorter than the body; whereas, antennal length in *P. elongatus* is longer than the body. *Pogonognathellus nigritus* was described by Maynard (1951) but synonymized with *P. elongatus* by Christiansen (1964) due to apparently similar morphology. Type specimens were not available to Christiansen; although Maynard's collection was deposited in NMNH, *P. nigritus* types apparently no longer exist (D. DeRoche, *in litt*.). While all of these morphological characteristics indicate that Maynard's description is of the species discussed here, there is at least one discrepancy. Maynard described the species as having four or five ungual teeth, but all specimens collected in this study had only three. Ungual teeth can be variable within species of Tomoceridae, and such a difference by itself generally is not considered sufficient for species separation. Therefore, *P. nigritus* is accepted here as a valid species. Nevertheless, collection of specimens from the type locality (Bear Mountain State Park, NY) would be desirable.

***Tomocerina lamellifera* (Mills)**
Figs. 23, 24

Tomocerus lamelliferus Mills, 1934:84, Figs. 134-136.

= *Tomocerina lamellifera* Yosii, 1956.

Maximum Length: 2 mm.

Color: Ground color pale yellow to white, light blue-gray pigmentation on cuticle, darker at anterior end (Fig. 23a). Eye patches black. Antennal segments with blue-gray pigment; segments III and IV darker distally. All other appendages white. Scale pattern a uniform gray.

Head: Antennae shorter than body, second lamella of maxilla with fringe on basal portion, shorter fringe extending to tip.

Chaetotaxy: The chaetotaxy is variable, especially on the fourth abdominal segment (fig. 24).

Legs: Unguis with 3 inner teeth. Unguiculus lanceolate (fig. 23b). Tenent hair clavate. Elongate setae on inner surface of metatibiotarsus.

Furcula: Dens without spine-like scale at inner base. Dental spines brown, smooth; intermediate and terminal spines considerably longer than others, formula 2-3, 1/1, 1, 2-3, 1 (fig. 23c). Mucro elongate and slender with numerous ciliate setae, long lamella extending from basal tooth to anteapical tooth at distal end; 2-4 intermediate teeth (fig. 23d).

Ecology: Found in moist leaf litter at low elevations.

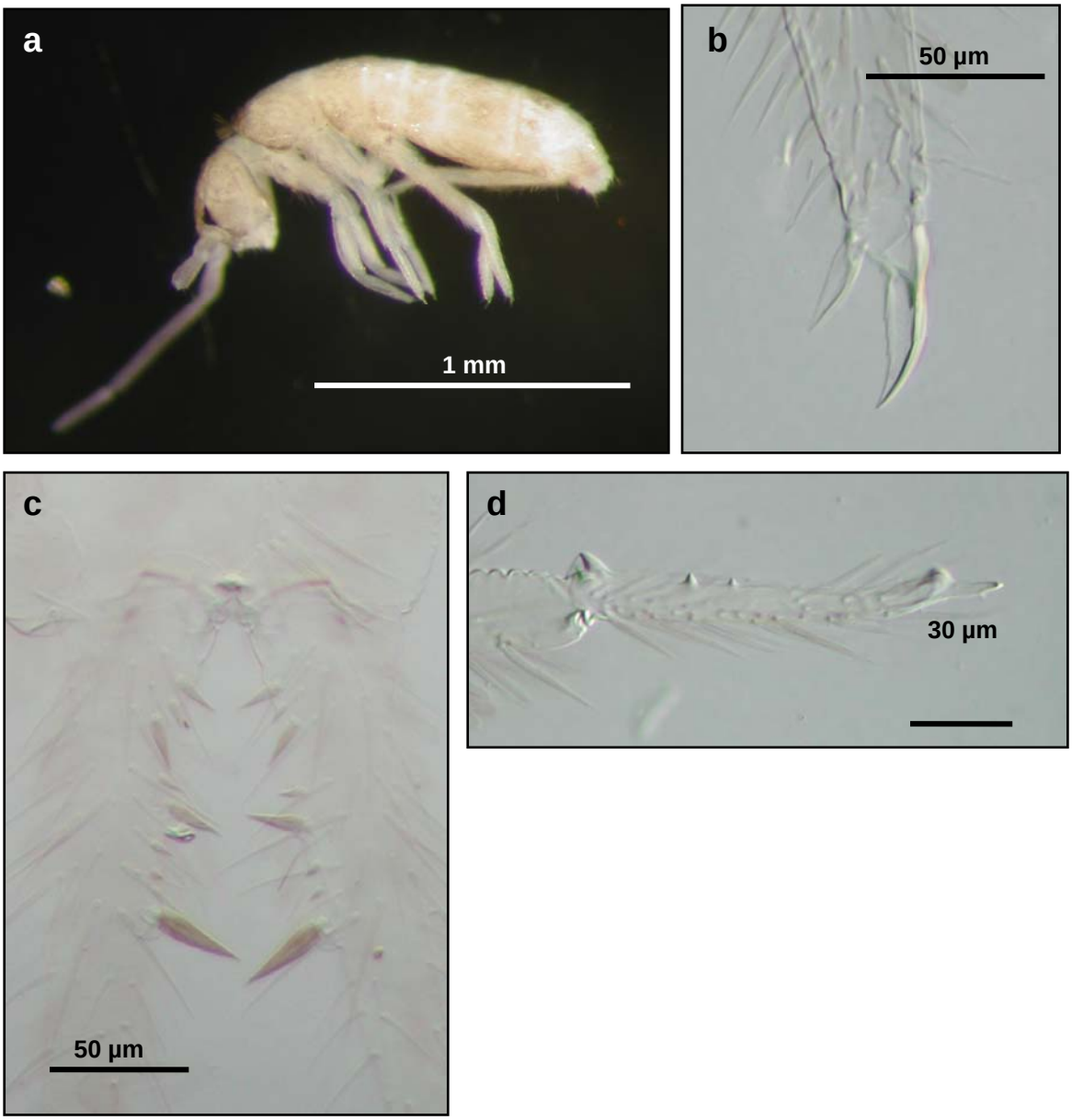


Figure 23. *Tomocerina lamellifera*; a) preserved; b) foot complex; c) dental spines; d) mucro.

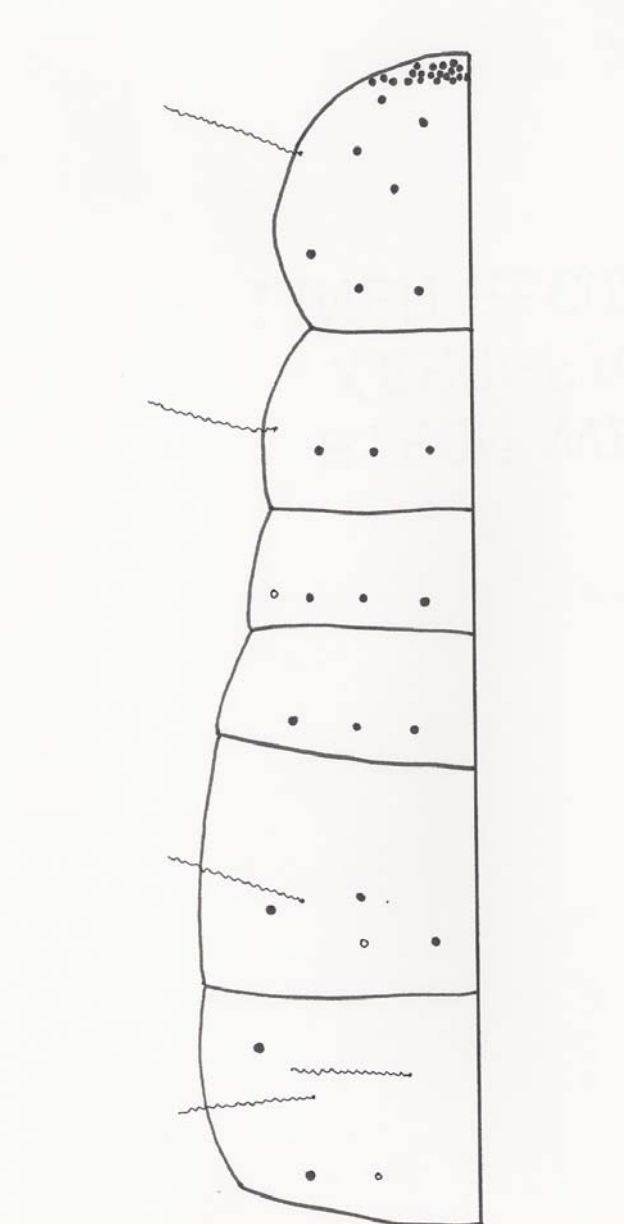


Figure 24. *Tomocerina lamellifera*, chaetotaxy. Filled circles indicate macrochaetae; open circles indicate mesochaetae; wavy lines indicate bothriotricha.

Localities and numbers of specimens studied: Five specimens, Sevier County, Tennessee, GSMNP: 4 specimens, Roaring Fork Motor Nature Trail, seep leaf litter, 13 March 2006, K. L. Felderhoff and E. Alexander, collectors; and 1 specimen, Little River Road, 20 September 2006, K. L. Felderhoff and E. C. Bernard. One specimen, Blount County, Tennessee, Reed Creek, 17 April 1997, P. J. Obenauer.

Diagnosis: *Tomocerina lamellifera* is easily recognized by its small size, relatively short antennae, and distinctive furcula. The dental spines and long, slender mucro are clearly visible with a stereoscope.

IV. Conclusion

This project began as a study to determine the value of scale patterns for the identity of Tomoceridae. Molecular analysis and traditional morphological studies were performed to validate the specificity of the patterns. Taken together, however, the complexity of the family proved to be much greater than expected. Specimens of putative *Pogonognathellus flavescens*, the most commonly identified species in North America, were molecularly much different from the true European *P. flavescens*, suggesting that *P. flavescens* may not occur in the United States and instead is a complex of closely related species. Furthermore, molecular data revealed that *P. dubius* probably is another species complex that will need further attention. Two new species (*P. danieli*, *P. mystax*) and a resurrected species (*P. nigritus*) were suggested by the molecular data. There are likely many more species new to science in the southern Appalachians.

Often, a taxonomist is fortunate enough to notice an obvious apomorphic character that can be used to separate morphologically similar species. Such is the case with *P. mystax* and its separation from the *flavescens* complex by the occurrence of pigmentation on the clypeus, and the separation of *P. nigritus* from *P. elongatus* by distinct differences in body color, among other morphological characters. As with many understudied taxa, tomocerids are a family in which much work is left to be done.

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Appendix

Localities of Foreign and Unknown Specimens

Swedish specimens:

P. flavescens, *P. longicornis*, and *T. minor*

Sweden: SK: Lund Dalby Söderskog, 26 August 2006, A. Fjellberg.

***P. flavescens*-like:**

#25	Sevier County, Tennessee, GSMNP, Rainbow Falls trail, near waterfall, leaf litter, 11 September 2005, K. L. Felderhoff.
#37	Sevier County, Tennessee, GSMNP, Rainbow Falls trail, 30 min up trail, leaf litter, 11 September 2005, K. L. Felderhoff.
#38/#39	Sevier County, Tennessee, GSMNP, Grotto Falls trail, near waterfall, leaf litter from hollow log, 27 September 2005, K. L. Felderhoff and E. Gilch.
#189	Swain County, North Carolina, GSMNP, Enloe Cemetery, 17 290707E 3932327N, leaf litter from riparian zone, 3 April 2006, K. L. Felderhoff, M. MacCarroll, E. C. Bernard.
#225	Swain County, North Carolina, GSMNP, Mingus Mill, leaf litter, 19 June 2006, K. L. Felderhoff.
#253	Swain County, North Carolina, GSMNP, Noland Ridge at Noland Creek headwater monitor site, mixed leaf litter, 23 June 2006, K. L. Felderhoff.
#369	Knox County, Tennessee, John Sevier Hunter Education Center, Rifle Range Road, 17 233976E 3993746N, site 1, mixed pine-hardwoods, 25 October 2006, K. L. Felderhoff and E. C. Bernard.
#387	Knox County, Tennessee, John Sevier Hunter Education Center, Rifle Range Road, 17 234407E 3993216N, site 3, mixed pine-hardwoods leaf litter, 25 October 2006, K. L. Felderhoff and E. C. Bernard.

***P. dubius*-like:**

#4	Sevier County, Tennessee, GSMNP, Roaring Fork Motor Nature Trail, moss, 6 September 2005, K. L. Felderhoff.
#43	Sevier County, Tennessee, GSMNP, Grotto Falls trail, near waterfall, leaf litter from rock face, 27 September 2005, K. L. Felderhoff and E. Gilch.

#114	Swain County, North Carolina, GSMNP, Enloe Cemetery, 17 290707E 3932327N, decomposing wood, 3 April 2006, K. L. Felderhoff, M. MacCarroll, E. C. Bernard.
#160	Swain County, North Carolina, GSMNP, Enloe Cemetery, 17 290707E 3932327N, leaf litter near decomposing log, 3 April 2006, K. L. Felderhoff, M. MacCarroll, E. C. Bernard.
#164	Swain County, North Carolina, GSMNP, Enloe Cemetery, 17 290707E 3932327N, leaf litter, 3 April 2006, K. L. Felderhoff, M. MacCarroll, E. C. Bernard.
#194	Swain County, North Carolina, GSMNP, Enloe Cemetery, 17 290707E 3932327N, moss on decomposing log, 3 April 2006, K. L. Felderhoff, M. MacCarroll, E. C. Bernard.
KQA	Blount County, Tennessee, GSMNP, Gregory Cave, litter outside cave, in vicinity of entrance, 27 July 2006, K. L. Felderhoff and M. MacCarroll.
KQB	Blount County, Tennessee, GSMNP, Gregory Cave, litter outside cave, in vicinity of entrance, 27 July 2006, K. L. Felderhoff and M. MacCarroll.

(Localities for specimens #72 and #73 are unavailable.)

Definitions

bothriothrix	unusually thin, flexible, elongate setae, found in characteristic positions on the cuticle (pl. bothriotricha).
dens	the long proximal segment of the distal arms of the forks of the manubrium bearing the mucro apically.
furcula	the forked spring or leaping appendage borne on the fourth abdominal segment.
manubrium	the large median base of the furcula bearing the dens.
mucro	the third and terminal segment of the furcula, arising from the apex of the dens.
tenent hair	apically expanded setae situated near apices of tibiotarsus, overhanging pretarsus and claws; may be clavate or pointed in tomocerids.
unguis	the dorsal, larger claw-like appendage borne by the pretarsus.
unguiculus	the ventral, smaller claw-like appendage borne by the pretarsus.

Abbreviations

Abd.	Used to denote certain segments of the abdomen.
Ant.	Used to denote certain segments of the antennae.
DNA	Deoxyribonucleic acid.
g	grams.
gal	gallons.
GSMNP	Great Smoky Mountains National Park.
h	hours.
in litt.	In litterace, communication occurring via written letter or email.
kb	kilobases.
min	minutes.
mL	milliliters.
mm	millimeters.
NMNH	U.S. National Museum of Natural History.
Th.	Used to denote certain segments of the thorax.
µL	microliters.
µm	micrometers.
PCR	polymerase chain reaction.
s	seconds.
USDA	United States Department of Agriculture.
V	volts.
x g	Times gravity, referring to the relative centrifugal force.

Vita

Kelly L. Felderhoff was born on April 16, 1983. She was raised in College Station, TX and graduated (cum laude) from A&M Consolidated High School in May of 2001. Kelly graduated in August 2005 from Texas A&M University in College Station, TX with a bachelor's degree in Entomology. While there, she pursued individual research projects studying forensic entomology, completed an internship with the Texas IPM Program, worked in the university's renowned insect museum, published an article in *American Entomologist*, was president of the student-run Undergraduate Entomology Student Organization, and received the Undergraduate Student Award for the Department of Entomology. That same month, she moved to Knoxville, TN to attend the University of Tennessee and pursue her Master's degree in Entomology and Plant Pathology, concentration Entomology. Under the direction of Dr. Ernest Bernard, she completed her degree in December 2007. Kelly is a member of Entomological Society of America and the Gamma Sigma Delta Agricultural Honor Society.