



## **Two New Species of Vitronura (Collembola: Neanuridae) from Shanghai, Eastern China, with Dna Barcodes**

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## TWO NEW SPECIES OF *VITRONURA* (COLLEMBOLA: NEANURIDAE) FROM SHANGHAI, EASTERN CHINA, WITH DNA BARCODES

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### ABSTRACT

*Vitronura setaebarbulata* **sp. nov.** and *V. quartadecima* **sp. nov.** are described from Shanghai, China. *V. setaebarbulata* **sp. nov.** is characterized by its heavy barbate macrosetae, very granulated tubercles, dorsointernal tubercles on head coalescent and very close each other, mandible with three big teeth, apical one with denticules and maxilla with one of the 2 lamellae trifurcated. *V. quartadecima* **sp. nov.** is characterized by having 14 tubercles on head, well isolated each other, smooth body setae and mandible with three big teeth, apical one with denticules. The COI barcode sequences were sequenced for four individuals of *V. setaebarbulata* **sp. nov.** and twelve individuals of *V. quartadecima* **sp. nov.** respectively. No genetic divergence between individuals of same species was found, and the divergence of nucleotides between the two new species is 18.54%. An updated key to world species of *Vitronura* is also provided.

Key Words: springtails, taxonomy, key, genetic divergence, macrosetae, tubercle

### RESUMEN

Se describen e ilustran *Vitronura setaebarbulata* **sp. nov.** y *V. quartadecima* **sp. nov.** de Shanghai, China. *V. setaebarbulata* **sp. nov.** se caracteriza por tener gruesas macrosedas barbuladas, tubérculos muy granulados, los tubérculos cefálicos dorsointernos coalescentes, muy cerca uno del otro, mandíbula con tres grandes dientes, el apical con denticulos y maxila con una de ambas lamelas trifurcadas. *V. quartadecima* **sp. nov.** se caracteriza por tener 14 tubérculos cefálicos, todos separados entre ellos, sedas lisas y mandíbulas con tres grandes dientes, el apical con denticulos. Se realizó la secuenciación del código de barras COI de 4 y 12 individuos respectivamente para estas especies. No se encontró divergencia genética entre los individuos de la misma especie, y la divergencia de nucleótidos entre las dos nuevas especies fue de 18.54%. Adicionalmente se proporciona una clave para las especies del mundo de *Vitronura*.

Palabras clave: colémbolos, taxonomía, clave, divergencia genética, macrosedas, tubérculos

The genus *Vitronura* Yosii, 1969 is widely distributed in the world, especially in southeastern and eastern Asia (Yosii 1976; Deharveng & Weiner 1984; Lee & Kim 1990; Smolis & Deharveng 2006; Tanaka & Hasegawa 2010; Jiang & Yin 2011, 2012). All species of the genus have antennal tubercles that are isolated from the frontal tubercle on the head. Among 18 species described to date (Tanaka & Hasegawa 2010; Jiang and Yin 2012), 7 of them are recorded in China (Jiang & Yin 2012). Two new species of *Vitronura* were found in the course of investigations of Collembola from several parks in Shanghai, China, which are described in this paper. DNA barcodes from mitochondrial COI genes were also sequenced

and recorded, in order to give a useful reference for identification and phylogenetic comparisons in the future.

### MATERIALS AND METHODS

Specimens were hand collected by using a feather-pen and aspirator. The habitats are bamboo or broad-leaved forest, where the animals were found under dry leaves or on upper layer of soil. The living animals were picked up directly from leaves and taken to laboratory. Some of them were cleared in Nesbitt's fluid and mounted in Hoyer's solution for identification, other were used for molecular experiment and the cuticles retrieved from

voucher specimens were also mounted in Hoyer's solution for identification. Drawings and measurements were done with the aid of a phase contrast microscope NIKON E600. Terminology for the description of the new taxa follows mainly Deharveng (1983), Deharveng & Weiner (1984), and a modification of Smolis & Deharveng (2006).

#### Abbreviations Used in the Description

General morphology: Abd., abdominal segment; Ant., antennal segment; Cx., coxa; Fe., femur; Scx2, subcoxa 2; Ti, tibiotarsus; Th., thoracic segment; Tr., trochanter; VT, ventral tube.

Groups of setae: Ag, antegenital; An, anal; Fu, furcal; Ve, ventroexternal; Vi, ventrointernal; Vl, ventrolateral.

Tubercles: An, antennal; Fr, frontal; Cl, clypeal; De, dorsoexternal; Di, dorsointernal; DL, dorsolateral; L, lateral; Oc, ocular; So, subocular.

Types of setae: ML, long, blunt macroseta; MI, long, acuminate macroseta/ae; Mc, short blunt macroseta/ae; m, smooth, ordinary setae; me, mesoseta/ae; mi, microsetae; ms, microsensillum/a; S, sensillum/a; sgd, dorsal guard sensillum; sgV, ventral guard sensillum; or, subapical organite of antenna IV; i, acute seta on Ant. IV, thinner and shorter than the other ordinary setae; x, labial papilla x.

SIPPE, Shanghai Institute of Plant Physiology and Ecology. UNAM, Universidad Nacional Autónoma de México.

#### DNA Extraction, Amplification and Sequencing

For DNA barcodes, total DNA was extracted from one single individual by non-destructive DNA extraction methods following Gilbert et al. (2007) with minor modifications. We shortened the incubation time in lysis buffer to 30-45 min, and used a DNA extraction kit (Promega Co.) to purify the total DNA. The mitochondrial COI gene sequence was amplified (658 bp) by primer pair LCO/HCO (Folmer et al. 1994). PCR products were purified and then sequenced directly using both of the amplification primers. DNA sequences were analyzed with the software DNA-STAR (Burland 2000) and deposited in GenBank. The genetic divergences (p-distance) were analyzed using MEGA 4.0 (Tamura et al. 2007).

## RESULTS

*Vitronura* Yosii, 1969

Type species: *Neanura mandarina* Yosii, 1954

#### Diagnosis

*Neanura*-like Collembola, body pale yellow to red in life and white in alcohol. With reduced *Neanura*-type mouthparts. Antennal tubercles

isolated from frontal tubercle on head. Organ of Ant. III flanked by 2 guard setae. Tuberculation of post cephalic tergites (Di, De, DL, L), with some of them fused. Tubercles are well individualized and distinct in living specimens. Tergites of Abd. V with 2 + 2 tubercles (De + Di, DL and L tubercle migrated ventrally) or more rarely 1 + 1 + 1 (Di tubercles merged at midline). Di sometimes vestigial on Abd. IV. Most species have 2 or 3 pairs of eyes.

#### Remarks on Morphology

As mentioned by Deharveng (1983), Deharveng and Weiner (1984) and Smolis and Deharveng (2006), differences between species of this genus are primarily based on mouthparts (such as mandible and maxilla), number of setae on ocular tubercle, unguicular tooth, chaetotaxy, and tubercles Di on head fused or not. Besides that, types of macrosetae maybe important for species separation, but types of short setae on body vary among different individuals of the same species.

*Vitronura setaebarbulata* sp. nov. (Figs. 1-13, Tables 1, 2 and 5)

#### Material Examined

**HOLOTYPE.** Female, eastern China, Shanghai City, Shanghai Botanic Garden, bamboo forest (Bambusoideae spp.), 23-IX-1997, leg. Q.Y. Yue. Paratypes. 4 females and 3 males, same locality as holotype, collected by Q.Y. Yue among 1997-1998 (2 males on 27-VIII-1997, 2 females on 23-IX-1997, 1 male on 27-IX-1997, 2 females on 18-V-1998); 1 female (voucher SH2012009) and 2 males (vouchers SH2012011 and SH2012012), with DNA barcodes sequenced, same locality as holotype, 12-III-2012, leg. Y. Bu and Y. Gao; 2 females, Shanghai, Songjiang County, Zhongjiashan, 6-XI-2002, and 15-VI-2004, leg. Y. M. Yang. Holotype and 10 paratypes are deposited in SIPPE; two paratypes are deposited in UNAM.

**Description.** Adult. Length (n = 13) 1.7 mm (range: 1.1-2.5 mm). Color red when alive for adults and pink for early juveniles and with barbulate macrosetae (Fig. 1; for color image see Supplementary Fig. 1), white in alcohol. Eye patches dark. Postantennal organ absent. Antenna shorter than head diagonal (ratio as 4:5). Tubercles with heavy granulation (Fig. 8). Macrosetae (ML, MI and Mc) with heavy barbulation. MI acuminate, present on lateral tubercles, ML and Mc with blunt tips present almost on all tubercles (Fig. 9). Measurements of setae: long and acuminate barbulate macrosetae (MI) 85-130 µm, long and thick barbulate macrosetae (ML) 80-113 µm, short barbulate macrosetae (Mc) 25-63 µm, mesosetae (me) thin and smooth 20-50 µm, long smooth setae 100 µm, present on last abdomen

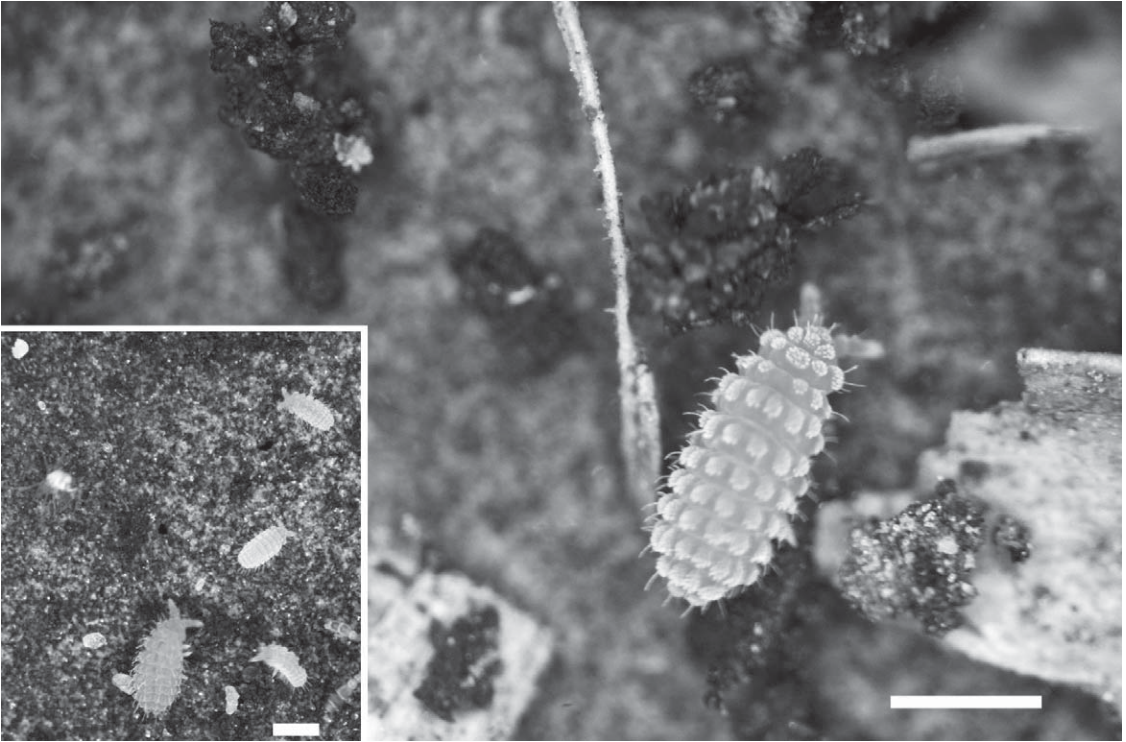


Fig. 1. *Vitronura setaebarbulata* sp. nov., alive; collected from Shanghai Botanic Garden. Inset shows adult with juveniles. Scale bars: 1000  $\mu$ m.

segment, microsetae (mi) tiny, 10  $\mu$ m, and sensorial setae slender, 38-50  $\mu$ m (Fig. 9).

Ant. I with 7 setae: 2 long, thick and ciliate (ML), others smooth, and few subcuticular reticulations. Ant. II with 11 setae: 2 long, thick and ciliate setae (ML), and others smooth. Ant. III and IV dorsally fused, sensorial organ of Ant. III with Sgv and Sgd similar, 2 microsensilla on separate dorso-lateral groove and ventral microsensillum very close to Sgv (Fig. 6). Ant. IV with 8 sensilla and 12 large smooth setae, i seta, trilobed apical vesicle present, subapical organite (or) with global tip, without ventral modified setae (Fig. 6).

Mouth cone short (Fig. 10). Labrum with 0/2, 2 smooth setae of different sizes. Labium with 7 pairs of setae, labial organ, seta A and L absent (Fig. 10), papilla  $\times$  absent. Mandible with 3 big teeth, apical one with 4-5 denticules (Fig. 4). Maxilla styliform, with 2 lamellae, one of them trifurcated (Fig. 3).

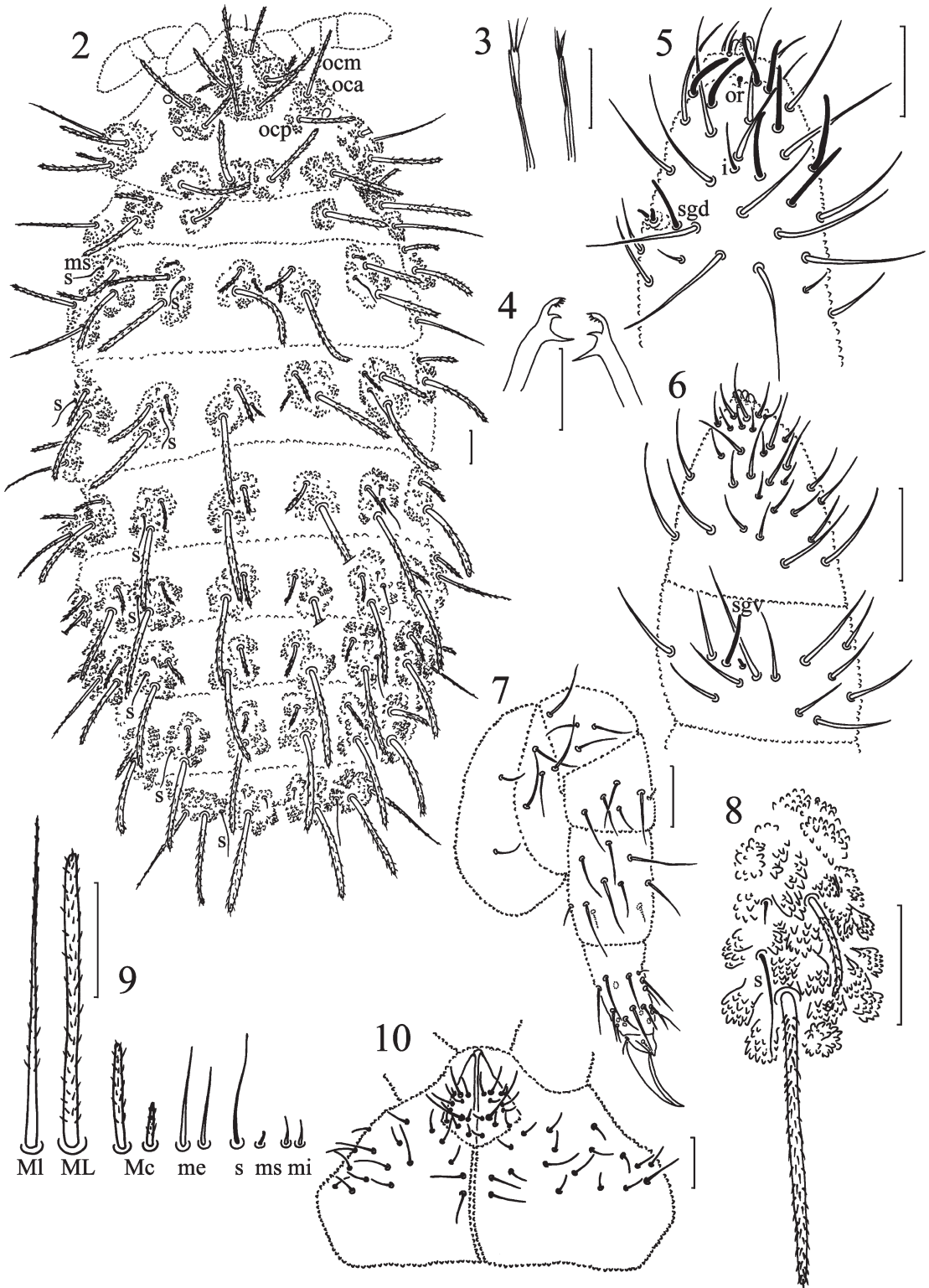
Head with 2 + 2 weakly pigmented eyes. Ocular tubercle with 3 ocular setae, Oca is microseta (mi), Ocm (ML) and Ocp (Mc) are barbulate macrosetae (Fig. 2).

Body setae types as in Fig. 9. Cephalic tubercles and chaetotaxy in Fig. 2 and Table 1. Clypeal tubercle (Cl) with 2 barbulate macrosetae (ML), 2 smooth setae (mi). Antennal tubercles (An) sepa-

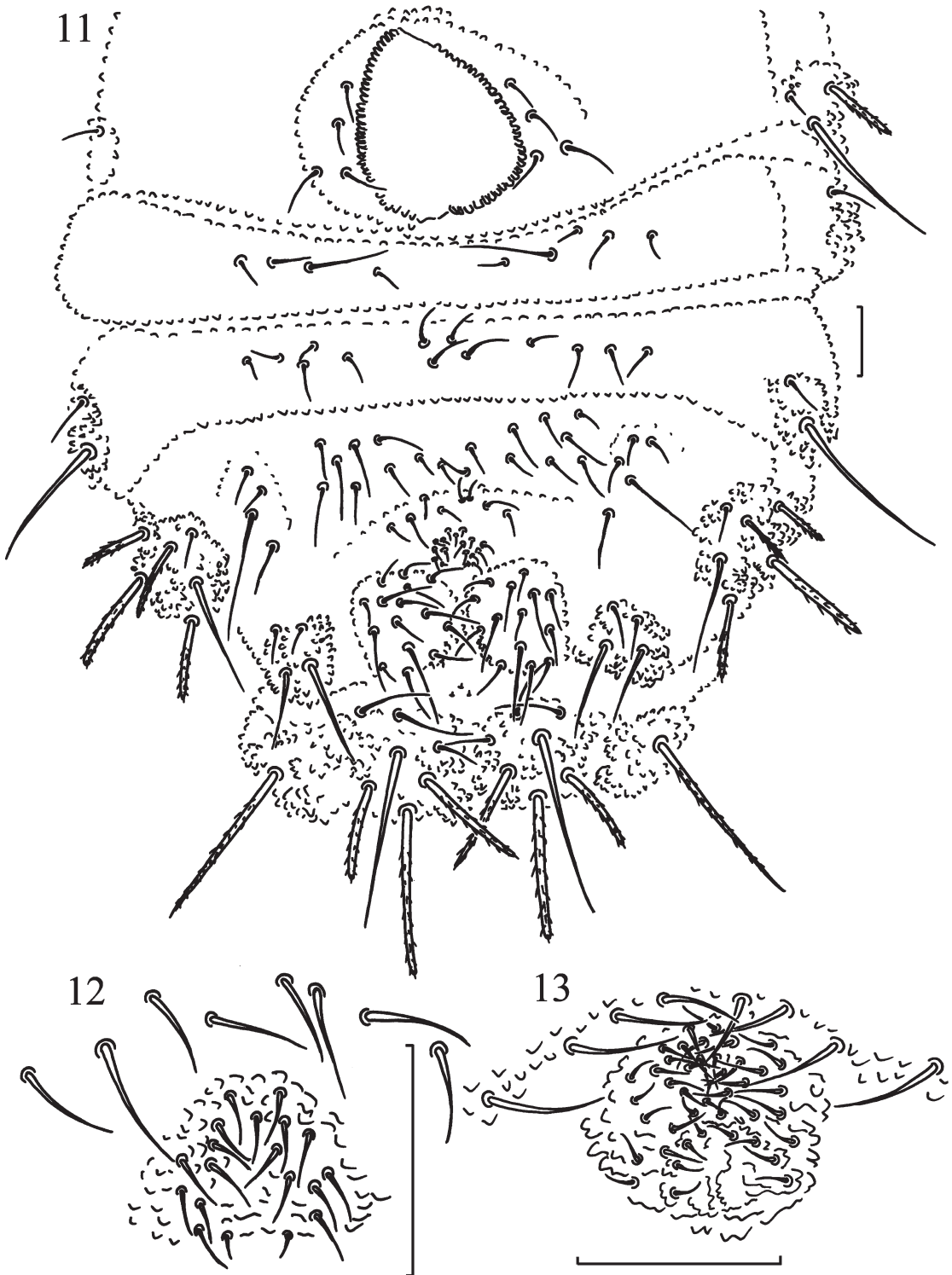
ately, with 4 setae, 2 barbulate macrosetae (ML, Mc) and 2 smooth setae (mi). Frontal tubercle (Fr) with 2 long barbulate macrosetae (ML) and one smooth microseta (mi). Cephalic dorsal dorso-internal (Di) very close each other, coalescent, with 2 long barbulate macrosetae (ML), dorsoexternal (De) with one barbulate macroseta (ML) and 2 smooth microsetae (mi), dorsolateral (DL), lateral (L) and subocular tubercles (So) fused with about 12 setae: 2 long barbulate macrosetae (ML), 1 long and acuminate barbulate macroseta (ML), 2 short barbulate macrosetae (Mc) and about 7 smooth setae (me, mi).

Number of tubercles from Th. I to Abd. VI on half notum is: 3, 4, 4/4, 4, 4, 2, 1 (Figs. 2 and 11). Thoracic and abdominal chaetotaxy as in Fig. 2 and Table 2. Tubercles setae formula on Th. I-Abd. V as Di: 033/22223; De: 1,3+s,4+s/3+s, 3+s, 3+s, 2+s; DL: 2,3+s+ms, 3+s/2222; L: 133/33364. Sensory setae formula on Th. I-Abd. V as 0, 2+ms, 2/1111. Tubercles De+DL of Abd. V with 1 sensillum and 3 setae (2ML, m). Each tubercle on Abd. VI with 7 setae (2ML, Mc, 4m) (Fig. 11).

Subcoxae with 0, 2, 2 setae, coxae with 3, 7, 8 setae, trochanters with 6, 6, 6 setae; femora with 12-13, 11-12, 10 setae, and tibiotarsi with 19, 19 and 18 setae on legs I, II and III respectively (Table 2). Tibiotarsi without tenent hairs (Fig. 7). Unguis without tooth (Fig. 7).



Figs. 2-10. *Vitronura setaebarbulata* sp. nov. 2. Dorsal chaetotaxy of the body; 3. Maxilla; 4. Mandible; 5. Ant. III-IV, dorsal view; 6. Ant. III-IV, ventral view; 7. Leg III; 8. De tubercle of Abd. I; 9. Setae types; 10. Ventral view of head and labium. Scale bars: 50 µm in Figs. 2, 5-10, 10 µm in Figs. 3 and 4.



Figs. 11-13. *Vitronura setaebarbulata* sp. nov. 11. Ventral view of Abd. I-VI and tergite VI; 12. Female genital plate; 13. male genital plate. Scale bars: 50  $\mu$ m.

TABLE 1. CEPHALIC TUBERCLES AND CHAETOTAXY OF *VITRONURA SETAEBARBULATA* SP. NOV.

Tubercles	Number and type of setae	Number and names of setae
Cl	2ML, 2Mc	2F, 2G
An	1ML, 0-1Mc, 2-3 mi	B, C, D, E
Fr	2ML, Mc	2A, O
Oc	ML, Mc, mi	Ocm, Ocp, Oca
Di	ML	Di1
De	ML, 2mi	De1, De2, Di2
DL+L+So	2ML, 1ML, 2Mc, 7me+mi	Uncertain

Chaetotaxy of legs and sterna as shown in Table 2. Ventral tube with 4+4 setae (Fig. 11). Anal tubercle with 14 setae and 3 setulae (Fig. 11). Furcular vestige with 4-5 setae (Fig. 11). Female with 3-4 pairs pregenital setae, 15-27 circumgenital setae and 1+1 eugenital setae (Fig. 12); male with 4 pairs of pregenital setae, about 40 circumgenital setae and 4 + 4 eugenital setae (Fig. 13).

Variation. There is a variation of the type of shortest setae on each body tubercles. In some individuals shortest setae are short barbate macrosetae (Mc), in others those shortest setae are smooth microsetae (mi). This variation does not depend on age, sex or body size of the specimens. Chaetotaxy of Abd. V of 2 females from Zhongjishan are 2, 5\*, 4 (Others are 3, 4\*, 4, Table 2), other variations for the chaetotaxy of ventral side of the abdomen segments are in Table 2.

#### Etymology

The species is named after the long heavy barbate macrosetae on the body.

#### Ecology

In moist litter and leaves under bamboo (Bambusoideae spp.) or lily magnolia, *Magnolia li-*

*liiflora* Desrous (Magnoliales: Magnoliaceae) in Shanghai Botanic Garden.

#### DNA Barcodes

The standard DNA barcodes (658 base pairs) from Mitochondrial COI gene was amplified and sequenced for 4 individuals from the Shanghai Botanical Garden. There is no divergence between individuals of this population. The sequences were deposited at GenBank with accession number JX027483-JX027486.

#### Remarks

The new species is similar to *V. tubercula* Lee and Kim, 1990 from Taibei with big barbate macrosetae, well developed tubercles and weakly fused Di tubercles on head. But they can be separated by the mouthparts (mandible with 3 smooth teeth in *V. tubercula* vs. 3 big teeth and the apical one with 4-5 small denticules in *V. setaebarbulata* sp. nov.; maxilla with 2 simple lamellae in *V. tubercula* vs. 2 lamellae with one of them trifurcate in *V. setaebarbulata* sp. nov.), and body chaetotaxy and size (Table 5).

*Vitronura quartadecima* sp. nov. (Figs. 14-25, Tables 3-5)

TABLE 2. BODY TUBERCLES AND CHAETOTAXY OF *VITRONURA SETAEBARBULATA* SP. NOV.

	Terga					Legs			
	Di	De	DL	L	Scx2	Cx	Tr	Fe	Ti
Th. I	—	1	2	1	0	3	6	12(13)	19
Th. II	3	4 <sup>1</sup>	4 <sup>1</sup> +ms	3	2	7	6	11(12)	19
Th. III	3	5 <sup>2</sup>	4 <sup>1</sup>	3	2	8	6	10	18
	Terga					Sterna			
Abd. I	2	4 <sup>1</sup>	2	3	VT	4			
Abd. II	2	4 <sup>1</sup>	2	3	Ve	4-5		Vl	—
Abd. III	2	4 <sup>1</sup>	2	3	Ve	4(5)		Fu	4-5
Abd. IV	2	3 <sup>3</sup>	2	6	Ve	9 (8)		Vl	4-5(6)
Abd. V	3	4 <sup>1</sup>		4	Ag	3-4		Vl	—
Abd. VI	7				Ve	14		An	3mi

<sup>1</sup>3 + S; <sup>2</sup>4 + S; <sup>3</sup>2 + S.

TABLE 3. CEPHALIC TUBERCLES AND CHAETOTAXY OF *VITRONURA QUARTEDECIMA* SP. NOV.

Tubercles	Number and type of setae	Number and Names of setae
Cl	2ML, 2me	2F, 2G
An	1ML, 3me	B, C D E
Fr	2ML, Mc/me	2A, O
Oc	ML, Mc, me	Ocm, Ocp, Oca
Di	ML	Di1
De	ML, 2me	De1, De2, Di2
DL	ML, 4me	DL1, uncertain
L	ML, 2me	uncertain
So <sup>1</sup>	1ML, 9me	uncertain

<sup>1</sup>So tubercle absent, but in this area the setae still present.

Material Examined

HOLOTYPE. female: eastern China, Shanghai City, Shanghai Botanic Garden, sweet olive plantation (*Osmanthus* spp.), 28-X-1997, leg. Q. Y. Yue. Paratypes. 2 females and 1 male, same locality as holotype, collected on 28-X-1997, 5-XI-1997, and 10-XI-1997, respectively; 2 females on 23-IX-1997, 1 male on 27-IX-1997, 2 females on 18-V-1998, leg. Q. Y. Yue. 8 females (vouchers SH2012001-SH2012005, SH2012022-SH2012024), garden of Shanghai Institute of Plant Physiology and Ecology, bamboo plantation (*Bambusoideae* spp.), with DNA barcodes sequenced, 9-III-2012, leg. Y. Bu and Y. Gao; 2 females (voucher SH2012018, SH2012020) and 2 males (voucher SH2012019, SH2012021), with DNA barcodes sequenced, Shanghai Zoo, 24-III-2012, leg. Y. Bu and Y. Gao. Holotype and 18 paratypes are deposited in SIPPE; two paratypes are deposited in UNAM.

Description. Length of adult ( $n = 21$ ) 2.1 mm (range: 0.9-3.3 mm). Color red in living specimens (Fig. 14; for color image see Supplementary Fig.

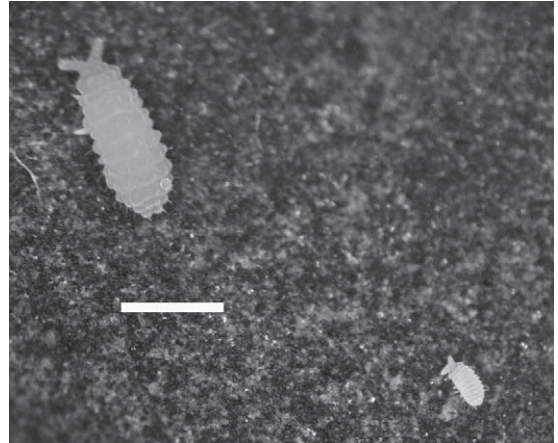


Fig. 14. *Vitronura quartadecima* sp. nov. alive; collected from Shanghai Zoo. Scale bars: 1000 µm.

14) and with well-distributed pink tubercles, pink for early juveniles, white in alcohol. Eye patches dark (Fig. 14). Postantennal organ absent. Antenna shorter than diagonal of head (ratio as 3:5). Tubercles with granulation distributed around long macrosetae and subcuticular reticulation. All body setae smooth. Blunt macrosetae (ML and Mc) with middle axle, present on most body tubercles, acuminate macrosetae (ML) present on lateral tubercles. Measurements of setae: long and acuminate macrosetae (ML) 80-115 µm, long and thick macrosetae (ML) 75-100 µm, short macrosetae (Mc) 30-50 µm, mesosetae (me) 20-50 µm, and sensorial setae slender, 50-75 µm (Fig. 18).

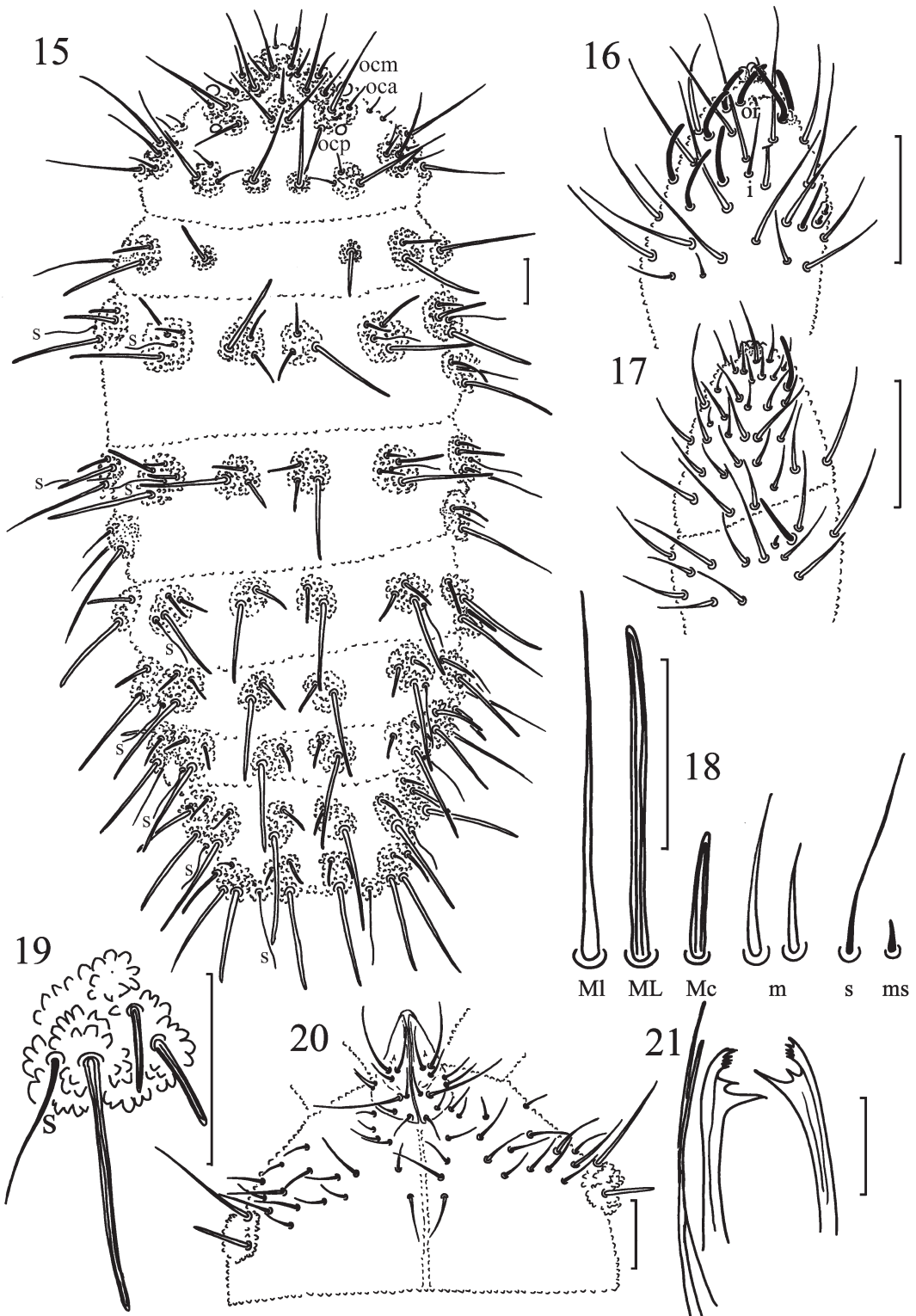
Ant. I with 7 setae, 2 longer than others; Ant. II with 11 setae, 2 longer than others. Ant. III and IV dorsally fused, sensorial organ of Ant. III with Sgv and Sgd similar, 2 microsensilla on separate dorso-lateral groove and ventral microsensillum very close to Sgv (Figs. 16 and 17). Ant. IV with 8 sensilla and 12 large smooth setae, i seta, trilobed

TABLE 4. BODY TUBERCLES AND CHAETOTAXY OF *VITRONURA QUARTEDECIMA* SP. NOV.

	Terga				Legs				
	Di	De	DL	L	Scx2	Cx	Tr	Fe	Ti
Th. I	—	1	2	1	0	3	6	12	19
Th. II	3	4 <sup>1</sup>	4 <sup>1</sup> +ms	3	2	7	6	11	19
Th. III	3	5 <sup>2</sup>	4 <sup>1</sup>	3	2	8	6	10	18
	Terga				Sterna				
Abd. I	2	4 <sup>1</sup>	2	3	VT	4			
Abd. II	2	4 <sup>1</sup>	2	3	Ve	4(5)		V1	—
Abd. III	2	4 <sup>1</sup>	2	3	Ve	4(5)		Fu	4-6
Abd. IV	2	3 <sup>3</sup>	3	4	Ve	8(9)		V1	5(6)
Abd. V	3	5 <sup>2</sup>		4	Ag	3-4		V1	1
Abd. VI	7				Ve	14		An	3mi

<sup>1</sup>3+S; <sup>2</sup>4+S; <sup>3</sup>2+S.





Figs. 15-21. *Vitronura quartadecima* sp. nov. 15. Dorsal chaetotaxy; 16. Ant. III-IV, dorsal view; 17. Ant. III-IV, ventral view; 18. Setae types; 19. De tubercle of Abd. I; 20. ventral view of head and labium; 21. Maxilla and mandible. Scale bars: 50  $\mu$ m in Figs. 15-20, 10  $\mu$ m in Fig. 21.

TABLE 5. COMPARISON OF *VITRONURA SETAEBARBULATA* SP. NOV., *VITRONURA QUARTADECIMA* SP. NOV. AND TWO SIMILAR SPECIES.

Species	<i>V. tubercula</i>	<i>V. setaebarbulata</i> sp. nov.	<i>V. quartadecima</i> sp. nov.	<i>V. giselae</i>
Size (mm)	3	1.7	2.1	0.9-1.4
Tubercle formula	11/688/888662	11/688/888862	14/688/888862	12/688/888862
Mandible teeth	3 smooth	3 with denticules	3 with denticules	3 smooth
Maxilla lamellae	2	2 (trifurcate)	2	?
Th. III De setae	4	5	5	5
Oc setae	2	3	3	3
Fu setae	3	4-5	4-6	4
Abd. V setae	3,5,3	3,4,4	3,5,4	3,5,4
ML Type	serrated	barbulate	smooth	smooth

apical vesicle and subapical organ (or) present, without ventral modified setae (Fig. 17).

Labrum with 0/2, 2 smooth setae of different sizes. Mouth cone long (Fig. 20), labium with 7 pairs of setae, labial organ, seta A absent, papilla x present (Fig. 20). Mandible with 3 big teeth as *V. setaebarbulata* sp. nov. (Fig. 21). Maxilla styli-form, with 2 simple lamellae (Fig. 21).

2 + 2 weakly pigmented eyes. Ocular tubercle with 3 ocular setae; Oca is a microseta (me), Ocm (ML) and Ocp (Mc) are macrosetae.

Types of setae on body as in Fig. 18. Cephalic tubercles and chaetotaxy in Fig. 15 and Table 3. Head with 14 tubercles. Clypeal tubercle (Cl) with 2 macrosetae (ML), 2 smooth setae (mi). Antennal tubercles (An) isolated, each with 4 setae: one macroseta (ML) and 3 mesosetae (me). Frontal tubercle (Fr) with 2 long macrosetae (ML) and one short macroseta (Mc). Cephalic dorsal dorsointernal tubercle (Di), with one long macroseta (ML), dorsoexternal (De) with one long macroseta (ML) and 2 mesosetae (me), dorsolateral (DL) with 5 setae (ML, 4me), lateral (L) with 3 setae (ML, 2me). Subocular area not tubercle shaped (So), with about 10 setae: one long macroseta (ML) and about 9 mesosetae (me).

Number of tubercles from Th. I to Abd. VI on half notum is: 3, 4, 4/4, 4, 4, 3, 1 (Figs. 15, 22). Thoracic and abdominal chaetotaxy as in Fig. 15 and Table 4. Tubercles setae formula on Th. I-Abd. V as Di: 033/22223; De: 1, 3+s, 4+s/3+s, 3+s, 3+s, 2+s; DL: 2, 3+s+ms, 3+s/2223; L: 133/33344. Sensory setae formula on Th. I-Abd. IV as 0, 2+ms, 2/11111. Tubercles De+DL of Abd. V composed by two adjacent tubercles, with one sensillum on small tubercle and 4 setae (2ML, Mc, mi) on large tubercle. Each tubercle on Abd. VI with 7 setae (2ML, 2MI, 3m) (Fig. 22).

Subcoxae with 0, 2, 2 setae, coxae with 3, 7, 8 setae, trochanters with 6, 6, 6 setae, femora with 12, 11, 10 setae, tibiotarsi with 19, 19 and 18 setae on legs I, II and III, respectively (Table 4), tenent hairs absent (Fig. 23). Unguis without tooth (Fig. 23).

Chaetotaxy of legs and sterna as in Table 4. Ventral tube with 4+4 setae (Fig. 22). Anal tuber-

cle with 14 setae and 3 setulae (Fig. 22). Furcular vestige with 4-6 setae (Fig. 22). Male with 4 pairs pregenital setae, about 36 circumgenital setae and 4 + 4 eugenital setae (Fig. 24). Female with 3-4 pairs pregenital setae, 17-20 circumgenital setae and 1+1 eugenital setae (Fig. 25).

Variation. Lateral tubercles on Abd. V of one female from Shanghai Zoo with 5 setae. Other variations for the chaetotaxy of ventral side of the abdomen segments as shown in Table 4.

#### Etymology

The species is named *V. quartadecima* for the 14 tubercles on the head.

#### Ecology

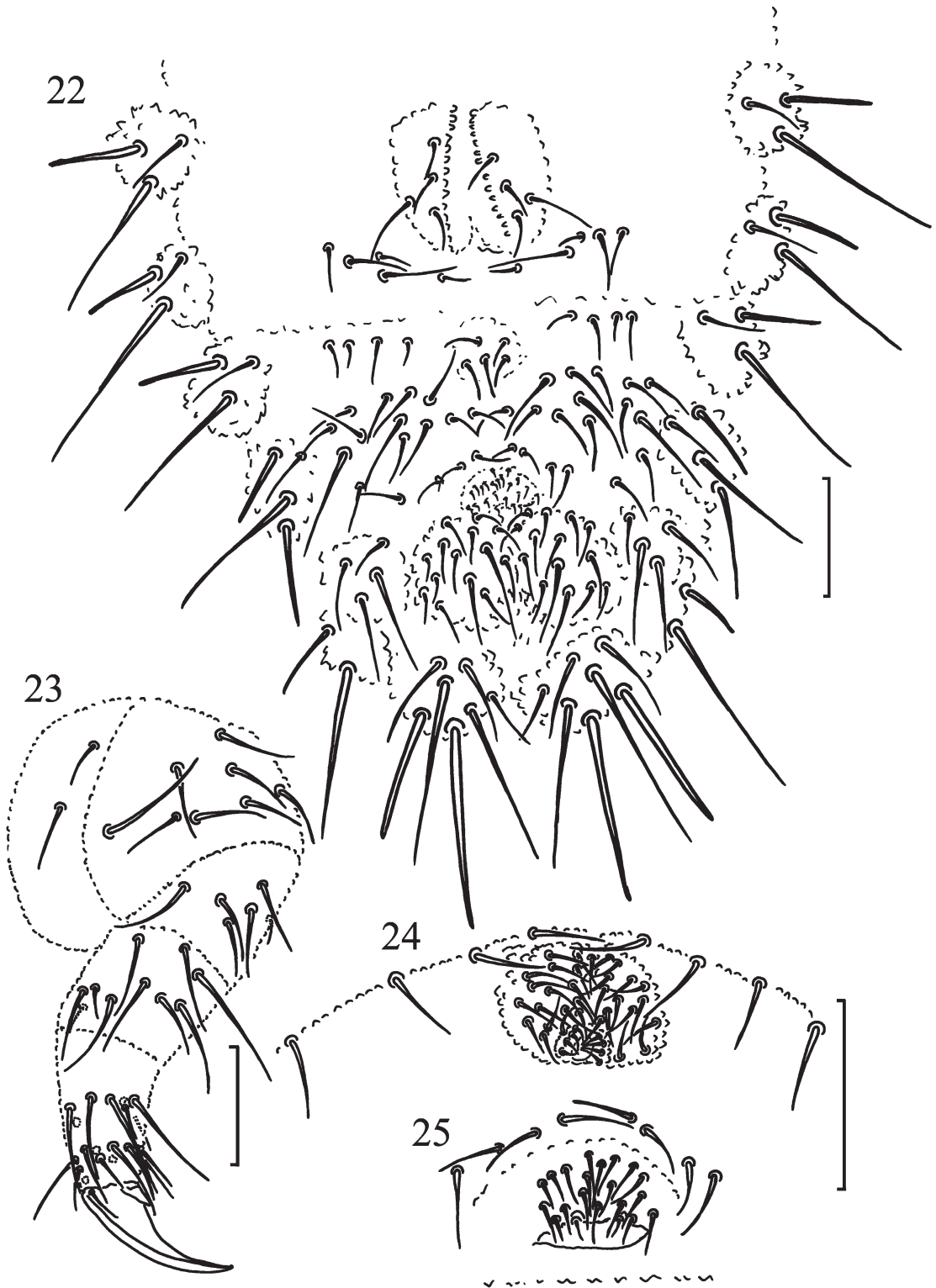
In moist litter and leaves under bamboo (Bambusoideae spp.) or sweet olive (*Osmanthus* spp.; Lamiales: Oleaceae) of several gardens in Shanghai.

#### DNA Barcode

The standard DNA barcodes sequences (658 base pairs) from Mitochondrial COI gene was amplified and sequenced for 12 paratypes (No. N1-5, Nx1-3, N 18-21) of *Vitronura quartadecima* sp. nov. The gene sequences between SIPPE population and Shanghai Zoo populations are completely identical. All sequences were deposited at GenBank with the accession number JX027487-JX027498.

#### Remarks

The new species is close to *V. giselae* (Gisin, 1950) (cosmopolitan species), with smooth axle macrosetae, ventral tube with 4+4 setae and some dorsal chaetotaxy. But they can be separated by the tubercles numbers on the head (12 in *V. giselae* vs. 14 in *V. quartadecima* sp. nov.) and mandible (3 smooth teeth in *V. giselae* vs. 3



Figs. 22-25. *Vitronura quartadecima* sp. nov. 22. Ventral view of Abd. I-VI and tergite VI; 23. Leg III; 24. Male genital plate; 25. Female genital plate. Scale bars: 50  $\mu$ m.

big teeth and the apical one with denticules in *V. quartadecima* **sp. nov.** (Table 5).

The different numbers of nucleotides for barcode sequences between *Vitronura setaebarbulata* **sp. nov.** and *Vitronura quartadecima* **sp.**

**nov.** are 122 sites, and the genetic divergence is 18.54%. The different numbers of coded amino acids for this sequence between these 2 new species is 3, and divergence of coded amino acids is 1.37%.

UPDATED KEY TO SPECIES OF *VITRONURA* YOSII, 1969 (MODIFIED FROM SMOLIS AND DEHARVENG, 2006 AND JIANG AND YIN, 2012)

- 1. Cephalic tubercle Oc with 1 or 2 setae . . . . . 2
- Cephalic tubercle Oc with 3 setae. . . . . 12
- 2. Cephalic tubercle Oc with 1 seta. . . . . 3
- Cephalic tubercle Oc with 2 setae. . . . . 4
- 3. Cephalic tubercle An with 1 seta . . . . . *V. mascula* Smolis & Deharveng, 2006; Vietnam
- Cephalic tubercle An with 2 setae. . . . . *V. shaanxiensis* Jiang & Yin, 2011; China: Shaanxi
- 4. Tubercles Di on head fused along midline. . . . . *V. tubercula* Lee & Kim, 1990; China: Taiwan
- Tubercles Di on head separate. . . . . 5
- 5. Seta O on head present. . . . . 9
- Seta O on head absent . . . . . 6
- 6. Tubercles Di on Abd. V fused along midline . . . . . *V. sinica* Yosii, 1976; China: Hongkong
- Tubercles Di on Abd. V separate . . . . . 7
- 7. Tubercle De of Th. II with 5 setae. . . . . *V. luzonica* Yosii, 1976; Philippines
- Tubercle De of Th. II with 4 setae. . . . . 8
- 8. Tubercle An of head with C and E setae. . . . .
- . . . . . *V. gressitti* Cassagnau & Deharveng, 1981; Papua New Guinea, Brunei
- Tubercle An of head without C and E setae . . . . .
- . . . . . *V. kunigamiensis* Tanaka & Hasegawa, 2010; Japan
- 9. Tubercles Di on Th. I with 2 setae . . . . . *V. latior* (Rusek, 1967); China:Guangdong
- Tubercles Di on Th. I with 1 seta . . . . . 10
- 10. Claw with inner tooth . . . . . 11
- Claw without inner tooth . . . . . *V. mandarina* (Yosii, 1954); Japan, Solomon Islands
- 11. Tubercle Di on Abd. V with 2 setae . . . . . *V. namhaeiensis* Lee, 1974; Korea
- Tubercle Di on Abd. V with 3 setae . . . . . *V. pygmaea* (Yosii, 1954); China:Taiwan, Japan, Indonesia
- 12. Cephalic tubercle De with 2 setae . . . . . *V. joanna* (Coates, 1968); South Africa
- Cephalic tubercle De with 3 setae. . . . . 13
- 13. Cephalic tubercles Di fused or weakly fused along midline. . . . . 14
- Cephalic tubercles Di distinctly separate . . . . . 16
- 14. Cephalic seta O on head present . . . . . 15

- Cephalic seta O on head absent . . . . . *V. singaporiensis* (Yosii, 1959); China:taiwan, Southeast Asia
15. Tubercle Di of Abd. I present . . . . . *V. macgillivrayi* (Denis, 1933); Costa Rica
- Tubercle Di of Abd. I absent . . . . . *V. setaebarbulata* **sp. nov.**; China: Shanghai
16. Claw with inner tooth . . . . . 17
- Claw without inner tooth . . . . . 18
17. Mandible with 5 denticles, tubercle De+DL of Abd. V with 5 setae. . . . . *V. qingchengensis* Jiang & Yin, 2012; China: Sichuan
- Mandible with 3 denticles, tubercle De+DL of Abd. V with 4 setae . . . . . *dentata* Deharveng & Weiner, 1984; North Korea
18. Head with 14 tubercles. . . . . *V. quartadecima* **sp. nov.**; China: Shanghai
- Head with 12 tubercles. . . . . 19
19. Tubercle Ve of Abd. IV with 7-9 setae; labrum distally ogival . . . . . *V. acuta* Deharveng & Weiner, 1984; North Korea
- Tubercle Ve of Abd. IV with 10 setae; labrum distally non-ogival . . . . . *V. giselae* (Gisin, 1950); Switzerland, Kenya, Mexico

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