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# NEW FRIDERICIA SPECIES (OLIGOCHAETA: ENCHYTRAEIDAE) FROM VÉRTES MOUNTAINS OF HUNGARY

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The up to now unknown enchytraeid fauna of the Vértes Mountains (part of the Transdanubian Mountains, Hungary) was investigated. Eleven genera, 41 species and one subspecies were identified. One species, *Fridericia mahunkai* sp. n. and one subspecies *Fridericia gamotheca hungarica* ssp. n. are new to science. It is proposed to distinguish three subspecies of *Fridericia gamotheca* Issel, 1905: *F. g. gamotheca, F. g. maroccoiensis* and *F. g. hungarica* ssp. n. The description of *F. cf alata* Nielsen et Christensen, 1959 is provided. The protist *Buetschliellopsis* sp. is reported for the first time from the gut of an enchytraeid from Hungary.

Key words: Fridericia, new species, subspecies, Enchytraeidae, fauna, Vértes Mountains.

## INTRODUCTION

The investigation of the Hungarian enchytraeid fauna has been proceeded intensive since 2001 with the support of the Hungarian Scientific Research Fund (OTKA). First the enchytraeid faunas of the north-eastern mountain ranges (Bükk, Mátra, Zemplén, Börzsöny Mts) were investigated, resulting in the recording of 77 species belonging to 14 genera (Dózsa-Farkas 2005), including five species new to science: *Marionina sexdiverticulata* Dózsa-Farkas, 2002, *Achaeta unibulba* Graefe, Dózsa-Farkas et Christensen, 2005, *Fridericia eiseni* Dózsa-Farkas, 2005, *F. schmelzi* Cech et Dózsa-Farkas, 2005 and *F. crassiductata* Dózsa-Farkas et Cech, 2006. Another 14 species were new for the fauna of Hungary.

In the present paper the faunistic results from the Vértes Mountains are presented and furthermore a new *Fridericia* species and a new subspecies from this area are described.

### MATERIALS AND METHODS

Study area. The Vértes Mountains are part of the Transdanubian Mountains. Their area occupies 314 km<sup>2</sup>. Since 2005 the main part of the range has been designated as the 'Vértes Nature Park'. The average altitude is 350 m a.s.l. Geologically the Vértes Mountains constitute a fairly uniform structure. On the surface of the entire range there are no older rocks than those from the mid-Triassic. The main parent material is dolomite from the upper Triassic. The Vértes Mountains possess a well-developed valley network (1260 km in total length), but in spite of this springs and streams are very rare. Annual sunshine duration is ca 1950–2000 hours. The average annual temperature above the height of 350 m is 8.5 C° (in January -3 C°). The average annual precipitation is between 600 and 700 mm. In winter the land is usually snow-covered (BENI & VISZLÓ 1996).

Hungarian Natural History Museum, Budapest

Different habitats were investigated, ranging from stream banks to different forest types, from meadows to rock grasses and also to the microhabitat of decaying tree trunks. From total, 14 localities 25 macro and microhabitats were sampled (Table 2).

The animals were extracted from the soil by the wet funnel method (O'CONNOR 1962). Worms were first observed and measured alive, than anaesthetized in 30% ethanol and subsequently preserved in 70% ethanol. Later, the specimens were stained with borax-carmine, then passed through an ethanol (70% to absolute) dehydration series, mounted temporarily in clove oil, and later mounted in Euparal in a slide between two coverslips. The important morphological structures *in vivo* were recorded, drawn and photographed using Axio Imager.A2 microscope, using DIC (differential interference contrast) illumination and an AxioCam MRc 5 (Zeiss) digital camera with Axiovision software. The whole-mounted specimens were reinvestigated and photographed, too.

Holotypes and paratypes of new taxa are deposited in the collection of the Department of Systematic Zoology and Ecology, Eötvös Loránd University, Budapest.

## RESULTS

In total 42 species and one subspecies were recorded, belonging to eleven enchytraeid genera (Table 1). Two taxa are considered new to science and described below: *Fridericia mahunkai* sp. n. and *Fridericia gamotheca hungarica* ssp. n. All species represent new records for the Vértes Mountains and two

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Achaeta spp.	F. galba (Hoffmeister, 1843)	
A. eiseni Vejdovský, 1877	F. gamotheca hungarica ssp. n.	
A. pannonica Graefe, 1989	F. hegemon (Vejdovský, 1878)	
Bryodrilus ehlersi Ude, 1892	F. maculatiformis Dózsa-Farkas, 1972	
Buchholzia appendiculata Buchholz, 1862	F. paroniana Issel, 1904	
Enchytraeus christenseni Dózsa-Farkas, 1992	F. ratzeli Eisen, 1879 (F. eiseni, sensu Dózsa-	
E. lacteus Claparede, 1861	Farkas, 2005)	
E. buchholzi sensu lato	F. rendsinata Dózsa-Farkas, 1972	
E. bulbosus Nielsen et Christensen, 1962	F. sylvatica Healy, 1979	
E. variatus Bouguenec et Giani, 1987	F. waldenstroemi Rota et Healy, 1999	
Enchytronia christenseni Dózsa-Farkas, 1970	F. mahunkai sp. n.	
En. parva Nielsen et Christensen, 1959	<i>Fridericia</i> sp. 1	
<i>En.</i> sp. 1	Henlea nasuta (Eisen, 1878)	
Fridericia cf. alata Nielsen et Christensen,	<i>H. perpusilla</i> Friend, 1911	
1959	H. similis Nielsen et Christensen, 1959	
F. argillae Schmelz, 2003	<i>H. ventriculosa</i> (d'Udeken, 1854)	
F. bisetosa (Levinsen, 1884)	Marionina argentea (Michaelsen, 1889)	
F. bulboides Nielsen et Christensen, 1959	M. brendae Rota, 1995	
F. christeri Rota & Healy, 1999	M. communis Nielsen et Christensen, 1959	
F. conculcata Dózsa-Farkas, 1986	Globulidrilus riparius (Bretscher, 1899)	
F. connata Bretscher, 1902	Mesenchytraeus pelicensis Issel, 1905	
F. dura Eisen, 1879 (Fridericia ratzeli sensu	Stercutus niveus Michaelsen, 1888	
Dózsa-Farkas, 2005)		

Table 1. A list of the enchytraeid species of the Vértes Mountains.

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Table 2. Investigated localities, habitats, sampling date with the recorded species.

- Vértestolna, mixed deciduous forest (mostly with oak trees) 01.11.2005 (Buchholzia appendiculata, Enchytraeus buchholzi sensu lato, E. bulbosus, Fridericia paroniana).
- **Near Majk-creek.** 47°29′23″N, 18°20′44″E, 245 m a.s.l., 18.10. 2007.
  - **a:** Alnus trees (Enchytraeus buchholzi sensu lato, Henlea perpusilla, Marionina argentea, Globulidrilus riparius).
  - **b**: under an old oak tree (*Achaeta pannonica, Buchholzia appendiculata, Enchytronia christenseni, Fridericia ratzeli, Marionina argentea, M. brendae*).
- Near Kőhányás village. 47°25′51″N, 18°20′18″E, 293 m a.s.l., 18.11.2007, 27.03.2008.
  - a: mixed deciduous forest (with hornbeam and beech dominance) (Achaeta spp., Achaeta eiseni, A. pannonica, Buchholzia appendiculata, Enchytraeus lacteus, E. buchholzi sensu lato, Enchytronia christenseni, En. parva, En. sp.1, Fridericia galba, F. maculatiformis, F. sylvatica, Marionina brendae, M. communis).
  - **b**: Decaying tree trunks (*Mesenchytraeus pelicensis*).
  - c: Beech wood on dolomit rock hillside (*Achaeta eiseni, Buchholzia appendiculata, Enchytraeus christenseni, E. buchholzi* sensu lato, *Enchytronia christenseni, En.* sp.1., *Fridericia bisetosa, F. paroniana, F. waldenstroemi, Henlea nasuta, Marionina brendae, M. communis*).
- On the top of a small hill, under oaks, 47°27′12″ N, 18°21′29 E, 298 m a.s.l., 18.11. 2007. (*Achaeta eiseni, Fridericia galba, Fridericia ratzeli*).
- **After Labanc-dűlő under Hosszú-Hill.** 47°27′21″N, 18°22′08″E, 268 m a.s.l., 18.11.2007.
  - **a:** Meadow, with thorn- and sloe-bush (*Enchytronia christenseni*, *En. parva*, *Fridericia bulboides*, *Henlea ventriculosa*).
  - **b**: Planted pine forest (*Fridericia bulboides*).
- Csíkvarsai-rét. 47°22′25″N, 18°27′27″E, 138 m a.s.l., 18.11.2007.
  - **a:** Near to the lake-shore (*Globulidrilus riparius*).
    - **b**: Pasture of Hungarian Steppe Cattle (*Achaeta* spp., *Enchytraeus christenseni*, *Fridericia galba*).
- Csákvár, on the hillside. 47°23′30″N, 18°26′05″E, 230 m a.s.l., mixed deciduous forest (mostly with linden, oak and ash). 21.11.2006 (*Achaeta* spp., *Buchholzia appendiculata*, *Fridericia maculatiformis*, *F. paroniana*).
- Csákvár, meadow. 47°23'17"N 18°26'29"E, 209 m a.s.l., 24.11.2009. (Fridericia christeri, F. conculcata, F. ratzeli, F. maculatiformis, Henlea ventriculosa).
- Vértessomló. 47°30′42″N, 18°22′31″E, 243 m a.s.l., young Turkey oak wood on the top of a hill, 27.03.2008. (Buchholzia appendiculata, Enchytraeus buchholzi sensu lato, E. variatus, Enchytronia christenseni, Fridericia connata, F.ratzeli, Stercutus niveus).
- Várgesztes. 47°28'49"N, 18°23'35"E, 265 m a.s.l., oak-hornbeam wood, 27.03.2008. (Achaeta spp., Buchholzia appendiculata, Enchytraeus christenseni, E. buchholzi sensu lato, E. bulbosus, E. variatus, Enchytronia christenseni, Fridericia bisetosa, F. galba, F. hegemon, F. maculatiformis, F. dura, F. rendsinata, F. tubulosa, F. waldenstroemi, F. sp.1, Henlea perpusilla, H. ventriculosa).

Várgesztes. 47°28'49"N, 18°23'47"E, 284 m a.s.l., 23.07.2013.
a: Decaying tree trunks (*Bryodrilus ehlersi, Buchholzia appendiculata, Enchytraeus buchholzi* sensu lato, *Fridericia paroniana*).
b: Soil near to a small creek (*Achaeta eiseni, Enchytronia christenseni, Marionina argentea*).

Table 2 (continued)			
Várgesztes. 47°28′ 58″ N, 18°23′51″ E, 325 m a.s.l., ravine, 27.03.2008.			
a: Oak-hornbeam wood (Buchholzia appendiculata, Enchytraeus christenseni, E. buchholzi sensu lato, Fridericia eiseni, F. galba, F. tubulosa, Henlea nasuta).			
<b>b:</b> Decaying tree trunks (Buchholzia appendiculata, Fridericia galba, F.dura, Henlea nasuta).			
<b>c:</b> On the rocky outcrop (Sedum) (Buchholzia appendiculata, Fridericia rendsinata, Henlea ventriculosa).			
Gánt. 47°27′00″N, 18°23′01″E, 233 m a.s.l., meadow, 27.03.2008 (Buchholzia appendiculata,			
Enchytraeus christenseni, E. buchholzi sensu lato, Fridericia connata, F. maculatiformis, F. paroniana, F. tubulosa, Marionina communis, Henlea ventriculosa)			
Vértesboglár. 47°25′59″N, 31°23′49″E, 160 m a.s.l., meadow, 27.03.2008, 24.11.2009, 23.07. 2013.			
<b>a:</b> Debris at the base of an old willow tree ( <i>Achaeta pannonica, Fridericia</i> cf. alata, F. hege- mon, Henlea nasuta, H. similis).			
<b>b:</b> Meadow under willow trees (Achaeta eiseni, A. pannonica, Enchytraeus christenseni,			
E. buchholzi sensu lato, E. bulbosus, Fridericia maculatiformis, F. paroniana, F. sylvatica,			
F. cf. alata, F. mahunkai sp.n., Henlea ventriculosa, Marionina communis).			
c: Meadow with field bindweed (Enchytraeus buchholzi sensu lato, Fridericia gamotheca			
hungarica ssp. n., F. cf. alata, Henlea ventriculosa).			
d: Soil near to a water canal (Achaeta eiseni, A. pannonica, Enchytraeus bulbosus, Frideri-			

Leg.: on 01.11.2005 Kontschán, J.; on 27.03.2008 and on 24.11.2009 Dózsa-Farkas, K. and Kontschán, J., on 23.07.2013 Dózsa-Farkas, K. and Nagy, G.

cia hegemon, F. cf. alata, Henlea nasuta).

species are also new for the Hungarian fauna (*Fridericia argillae* Schmelz, 2003 and *F. cf. alata* Nielsen et Christensen, 1959). Because the specimens differ in some characters from the description of *F. alata* given by SCHMELZ (2003) a detailed description is also provided below. The status of two *Achaeta* species, the *Enchytronia* sp.1 and *Fridericia* sp.1 have not been ascertained yet. These probably represent new species for science, but further investigations are needed to clarify their status. A list of species collected in the individual samples is given in Table 2.

### DESCRIPTION OF TAXA

# Fridericia mahunkai sp. n.

(Figs 1A–C, 2A–D, 3A–E)

Type material. Type locality: Vértesboglár, 47° 25′ 59″N, 31°23′49″E, 160 m a.s.l., meadow, moderately moist soil under large *Salix* tree, collected by K. Dózsa-Farkas & J. Kontschán, 27.03.2008. Holotype: F.17. slide No. 795 Paratypes: P. 94.1–3, slides No. 794, 796, 797. Deposited in the collection of the Department of Systematic Zoology and Ecology, Eötvös Loránd University, Budapest.

Etymology: Named in honour of my late colleague Dr. Sándor Mahunka.



**Fig. 1.** A–C: *Fridericia mahunkai* sp. n. A = spermatheca, B = penial slits, C = coelomocytes. D–G: *F. gamotheca hungarica* spp. n. D = spermathaecae, E = oesophageal appendage, F = coelomocytes, G = penial slits. H–J: *F. cf. alata* H = spermathaeca, I = coelomocytes, J = penial slits (all figures were made from living specimens; scale bars = 50 μm).

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Diagnosis. The new species can be recognized by the following combination of characters: (1) medium size (13–16 mm *in vivo*), segments 51–58; (2) max. 4 chaetae per bundle; (3) clitellum girdle-shaped: hyalocytes and granulocytes arranged in transverse rows dorsally; (4) four preclitellar nephridia; (5) coelomo-mucocytes a type, lenticytes small; (6) bursal slit longitudinal; (7) large seminal vesicle, (8) no subneural glands; (9) sperm funnel cylindrical, approximately half as long as body diameter, collar as wide as the funnel body, sperm ca 150–160  $\mu$ m long; (10) spermathecae separate entally; have two globular diverticula with stalks oriented entad.

Description. Holotype 10.7 mm long, 360  $\mu$ m wide at VIII and 400  $\mu$ m at the clitellum (fixed), segments 58. Body length of the three paratypes 12.8–15.4 mm, width 290–300  $\mu$ m at VIII and 350–390  $\mu$ m at the clitellum (*in vivo*), length of fixed specimens 8.6 mm (in two specimens the body-end is missing), width 280–380  $\mu$ m at VIII, 310–420  $\mu$ m at the clitellum, segments 51–52. Chaetae: 4 – 4,3,2,(1) : 4 – 4,3,2,(1). As in other *Fridericia* species the chaetae within a bundle are arranged in pairs with the outer being longer and thicker than the inner (35 × 3.5  $\mu$ m against 25 × 2.5  $\mu$ m in the first six segment, later 45 × 4.5  $\mu$ m against 30 × 3  $\mu$ m alike in ventro-lateral and dorso-lateral bundles; only two chaetae per bundle from XX (chaetae in the bundles close to the body end somewhat longer: about 50–52 ×



**Fig. 2.** Micrograph of *Fridericia mahunkai* sp. n. A = brain (marked with arrow), B = epidermal gland cells, C = clitellar region (e = egg, sv = seminal vesicle, sperm funnels marked with arrows), D = segments XIII–XVI (chylus cells marked with arrows, e = egg). (A and D stained, B and C *in vivo*; scale bars = 50 μm).

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4.2–5  $\mu$ m). Head pore at 0/I. Dorsal pores from VII. Epidermal gland cells arranged in 2–3 transverse rows per segment (Fig. 2B). Clitellum in XII–1/2 XIII, girdle-shaped, hyalocytes and granulocytes arranged in rows dorsally (Fig. 2C), weakly developed between the bursal slits. Thickness of body wall 27–37  $\mu$ m (dorsally and preclitellarly thicker, cuticle about 1  $\mu$ m in fixed specimens). Brain egg-shaped, 125–150  $\mu$ m long (fixed), about 2 times longer than wide (Fig. 2A).

Oesophageal appendages (peptonephridia) long, with 2–3 elongated terminal branches. Pharyngeal glands all united dorsally, all with ventral lobes, these well developed in



Fig. 3. Micrograph of *Fridericia mahunkai* sp. n.: A = spermathecae (marked with black arrows, oesophageal appendage marked with white arrow, ph = pharyngeal glands), B = spermathecae, C = nephridium preclitellarly, D = spermathecae in stained slide, E = male copulatory organ (marked with arrow, e = egg). (A–C *in vivo*, D–E stained; scale bars = 50 µm).

V and VI. Chloragocytes from V, brown in vivo. Dorsal vessel from XIX-XX, blood colourless. Midgut pars tumida unusually long from XXVIII-XLIV occupying 5-9 segments. Four pairs of preclitellar nephridia from 6/7 to 9/10, length ratio anteseptale : postseptale 1:1.5-2, anterior origin of efferent duct (Fig. 3C). Coelomo-mucocytes scarce (length 15-34 µm, fixed) with slightly wavy outline and the matrix blured but clearly visible nucleus, a type, lenticytes small: 5–7  $\mu$ m long. Chylus cells (Fig. 2D) between XIV–XV, occupying 2 segments. Seminal vesicle large in XI-XII. Sperm funnels cylindrical (Fig. 2C), about 140-170 µm long and 100–132 µm wide (in vivo) 1.5 times as long as wide. The funnel length in fixed specimens 120–150 µm. Collar slightly narrower or as wide as the funnel. Spermatozoa 150–160 μm long, heads 65–70 μm (in vivo). Diameter of sperm ducts 6 μm (fixed). Male copulatory organs (Fig. 3E) 105–110 µm long, 55–80 µm wide and 70–75 µm high (fixed), the laterally bent bursal slits are longitudinal (Fig. 1B). No subneural glands. Spermathaecae (Figs 3A, B, D): a small sessile ectal gland at the orifice, ectal ducts about 130–190  $\mu$ m long and 13 µm wide in the fixed specimens, widened proximally and projecting into the ampullae (ental bulb about 28-30 µm wide in vivo), canal not widened. Ampullae with two short stalked diverticula inserting on opposite sides of ental bulbs at bases of ampullae and oriented entad. Ampullae open separately into oesophagus. Inner surface of ampullae and stalks of diverticula thick-walled with granular texture. Diverticula have a spherical sperm-containing chamber (37–45 µm in diameter, fixed). Maximum width of spermathaecae (ampulla + diverticula) 110–120 μm (*in vivo*). One or two mature eggs at a time.

Distribution and habitat: Only known from the type locality.

Differential diagnosis. Seven Fridericia species have similar spermathecae with two spermathecal diverticula oriented entad and separately connected with the oesophagus (listed in Dózsa-Farkas 2009, Tab. 9 and in addition the variant of F. galba (Hoffmeister, 1843) with two diverticula). The main differences between other species of this *Fridericia* sub-group and the new species are as follows: F. heliota Zalesskaya, 1990 (Rota et al. 2003), F. alata Nielsen et Christensen, 1959, and F. galba (SCHMELZ 2003) have 5 pairs of preclitellar nephridia and the spermathecal ectal ducts are longer than in the new species. F. nanningensis Xie, Liang et Wang, 2001 (XIE et al. 2001) and F. bubalus Sesma et Dózsa-Farkas, 1993 (Sesma & Dózsa-Farkas 1993) have also 5 pairs of preclitellar nephridia and in addition the oesophageal appendages are different: c type in F. bubalus and b type in F. nanningensis. F. sylvatica Healy, 1979 (HEALY, 1979) has 4 pairs of preclitellar nephridia but the clitellum is only laterally developed and there is an additional pair of ventral pharyngeal gland lobes in VII. F. conculcata Dózsa-Farkas, 1986 (Dózsa-Farkas 1986) has also 4 pairs of preclitellar nephridia but also small subneural glands in XIII-XIV and is lacking lateral chaetae postclitellarly down to XXIV–XXVII.

## **Fridericia gamotheca hungarica** ssp. n. (Figs 1D–G, 4A–F, 5A–G, 6A–F)

Type material. Type locality: Vértesboglár, 47°25′ 59″N, 31°23′49″E, 160 m a.s.l., meadow, with many field bindweed (*Convolvulus arvensis*), collected by Dózsa-Farkas, K. & Nagy, G., 23.07.2013. Holotype: F.18. slide No. 838. Paratypes: P. 95.1–17 slides No.

812–820, 829, 830, 836, 837, 839–842, altogether 17 specimens. Deposited in the collection of the Department of Systematic Zoology and Ecology, Eötvös Loránd University, Budapest.

*Etymology*: The name refers to the country of origin, geographically distant from the sampling sites of the other two subspecies.

Diagnosis. (1) medium-sized species: about 7–11 mm long *in vivo* (6–8 mm fixed), segments 31–37; (2) maximum of chaetae 4–6; (3) coelomocytes b type, lenticytes small; (4) oesophageal appendage with short branches proximally as well as terminally; (5) clitellum girdle-shaped, hyalocytes also present ventrally; (6) dorsal vessel origin in XIII; (7) 5 pairs preclitellar nephridia; (8) sperm funnel pear-shaped, seminal vesicle absent; (9) the two spermathecal ampullae united completely, with two rounded diverticula with short stalks on each side; ectal duct long, with a small sessile ectal gland at the orifice; (10) several mature eggs at a time.

Description. Holotype 5.8 mm long, 340 µm wide at VIII and 350 µm at the clitellum (fixed), segments 37. Body length of paratypes 7–10.4 mm, width 300–450  $\mu$ m at VIII and 350-490 µm at the clitellum (in vivo), length of fixed specimens 5.6-8.6 mm, width 240-400 µm at VIII, 310–460 µm at the clitellum, segments (29), 31–37. Chaetae: 3,4 <sup>2(1)</sup>4,5 3,(2): 4,5,6, - 4,5,3,(2). In some specimens with a maximum of only four chaetae per bundle. Three and two chaetae only in posterior segments, but two chatae in a bundle occure not only terminally in the specimens with a maximum four chaetae. Two (rarely one) small chaetae also in XII, laterally (25–35  $\mu$ m long, 2.5  $\mu$ m wide). The outer chaetae longer and thicker than the inner ones within a bundle (the outer ca 50  $\mu$ m long and 4–5  $\mu$ m wide, the inner ones 30-38 µm long and 3-3.2 µm wide) but in the last 6-10 segments all chaetae are of equal length. Maximum length of the chaetae 57-65 µm in the terminal segments. Head pore at 0/I (Fig. 4C), dorsal pores from VII (Fig. 4D). Epidermal gland cells arranged in 3 transverse rows per segments. Clitellum in XII-1/2XIII, girdle-shaped, hyalocytes and granulocytes arranged in rows (Fig. 5B). In the fully developed adult specimens with eggs the hyalocytes and granulocytes are similarly arranged between the bursal slits too. Thickness of body wall 22–37  $\mu$ m, the longitudinal layer of muscles 10–17  $\mu$ m thick, cuticle about 1  $\mu$ m or < 1 μm (fixed specimens). Brain posteriorly slightly concave, 100–130 μm long (fixed), about 1.7–2 times longer than wide (Figs 4A–B).

Oesophageal appendages (peptonephridia) (Figs 1E & 4E) with some short braches in the proximal part and 3–4 branches terminally. First pair of pharyngeal glands united dorsally, the second and third pairs dorsally separate, all with ventral lobes and even the ventral lobes in IV are not separate from the dorsal lobes. Chloragocytes from V, dark brown (*in vivo*), diameter 25–35  $\mu$ m *in vivo*, 25  $\mu$ m when fixed. Dorsal vessel from XIII, blood colourless. Midgut pars tumida (Fig. 6A) from XXI–XXVI occupying 3 segment lengths. Five pairs of preclitellar nephridia from 6/7 to 10/11, length ratio anteseptale : postseptale 1 : 1.5, anterior origin of efferent duct. Coelomo-mucocytes (length 22–40  $\mu$ m *in vivo*, 20–25  $\mu$ m, fixed) variable, with a few or many refractile granules (Figs 1F & 4F), dark when accumulated, b type, lenticytes small: 5–10  $\mu$ m long. Chylus cells between X–XII, occupying 2 segments (Fig. 5A). Seminal vesicle absent. Sperm funnels pear-shaped (Figs 5C–E), about 100–162  $\mu$ m long *in vivo*, 87–150  $\mu$ m when fixed, and 1.4–1.6 times longer than wide, collar narrower than funnel body and about 10–14  $\mu$ m high. Spermatozoa about 100  $\mu$ m long, heads 40  $\mu$ m (*in vivo*). Diameter of sperm ducts 5  $\mu$ m (fixed). Male copulatory

organs (Fig. 5F) 60–87  $\mu$ m long, 50–60  $\mu$ m wide and 40–50  $\mu$ m high (fixed), the bursal slits with longitudinal and transverse components, the latter branching off the posterior end (Figs 1G & 5G). No subneural glands. Spermathaecae (Figs 1D & 6B–D): one small sessile ectal gland at the orifice of ectal duct (in one case two glands, Fig. 6C), ectal ducts long (about 170–266  $\mu$ m long and 14–18  $\mu$ m wide in the fixed specimens), the two ampullae merging mostly completely. Two rounded diverticula with short stalks on each side of the ampullae. One common dorsal opening into oesophagus. The diverticula and ampullae



**Fig. 4.** Micrograph of *Fridericia gamotheca hungarica* ssp. n. A–B = brain, C = head pore (marked with arrow), D = dorsal pores (the coelomocytes coming out, marked with arrows), E = oesophageal appendages (terminal branches marked with white arrows, proximal branches marked with black arrows), F = coelomocytes (coming out). A, C, E and F *in vivo*, B and D stained; scale bars = 50 μm).

mostly empty but sometimes also containing low quantities of sperm. Two to four mature eggs at a time (Fig. 6E).

In some specimens (including holotype) several protists were observed in the gut in front of the clitellum (Fig. 6F), identified by Dr Júlia Török as *Buetschliellopsis* sp (*Ciliophora: Actinomatida: Hoplitophryidae*). Till now a single species, *Buetschiellopsis enchytrae*, has been described from *Fridericia* sp. (PUYTORAC 1954). In Hungary this genus of protist had not been found in enchytraeids up to now.



**Fig. 5.** Micrograph of *Fridericia gamotheca hungarica* ssp. n.: A = chylus cells (marked with white arrows, penial apparate marked with black arrow), B = clitellar gland cells laterally, C = clitellar field: sperm funnels (marked with arrows, e = egg), D and E = sperm funnels (marked with arrows, F = male copulatory organ (marked with arrow), G = bursal slit (marked with arrow). (A, C, D and G *in vivo*, B and F fixed but not stained; scale bars = 50 µm).

Distribution and habitat: Only known from the type locality.

Differential diagnosis. The new taxon belongs to *F. gammotheca* Issel, 1904. The differences of this species with other closely related species (they belongs to the group of 12 species characterized by proximally fused sper-



**Fig. 6.** Micrograph of *Fridericia gamotheca hungarica* ssp. n.: A = midgut pars tumida (marked with arrows, B–D = spermathecae marked with arrows (an extra small figure shows the ectal duct, which in this case shows exceptionally two glands), D = four mature eggs in the body, F = *Buetschliellopsis* sp. protist in the gut (marked with arrow). A and F stained, B–F *in vivo*; scale bars = 50 µm).

mathecae (Dózsa-Farkas 2009, Tables 1–2, Schmelz & Collado 2013) are as follows: F. connata Bretscher, 1902, F. monochaeta Rota, 1995 and F. brunensis Schlaghamerský, 2007 differ from the new subspecies in the maximum of chaetae in a bundle [only 2 (3)], F. waldenstroemi Rota et Healy, 1999 is much larger (9–15 mm and 40–54 segments). Most similar to F. gamotheca is F. argillae Schmelz, 2003, but the latter smaller and slender (only 3–4 mm long and 150–230 µm wide) and differs from it by its first pair of pharyngeal glands without ventral lobes and the third pair having a posterior projection; in addition, the two spermathecal ampullae are cylindrical, elongate and joining only at their proximal ends. Likewise in F. marginata Schmelz et Collado, 2013 and F. roembkei Schmelz et Collado, 2013 the spermathecal ampullae are united only proximally and chylus cells are located postclitellarly. F. montafonensis Schmelz, 1998 and F. nemoralis Nurminen, 1970 are much larger (60–70 and 40–53 segments, respectively) and their spermathecal diverticula much larger, moreover the chylus cells are present far back postclitellarly in these species. Two species (F. bernini Dózsa-Farkas, 1988 and F. profundicola Dózsa-Farkas, 1991) belong also to the group defined above but they differ from the new subspecies because in F. bernini the spermathecae have more than two diverticula and in *F. profundicola* there are very few mucocytes but numerous large lenticytes, moreover in the latter the spermathaecal diverticula are large, oriented entad and have further small pouch-like protrusions.

The new subspecies can be easily distinguished from the other two subspecies (*F. gamotheca gamotheca* Issel, 1905 and *F. gamotheca maroccoiensis* Dózsa-Farkas, 1989, see discussion) mainly by the following characters: by their size (the length smaller than the length of *F. g. gamotheca* and larger than the length of *F. g. maroccoiensis*) and segment number (it is in *F. g. gamotheca* higher than in the new subspecies, but lower in *F. g. maroccoiensis*), the presence of chaetae in XII laterally, oesophageal appendages with branches not originating not only terminally, coelomocytes of b type , dorsal vessel origin more anteriorly in XIII, a different connection of the dorsal lobes of pharyngeal glands, and by the pear-shaped and smaller sperm funnels (Table 3).

# *Fridericia* cf. *alata* Nielsen et Christensen, 1959 (Figs 1H–J, 7A–F, 8A–F, 9 A–D)

In the locality Vértesboglár (47°25′59″N, 31°23′49″E, 160 m a.s.l., meadow) the dominant species was *F. alata*, but the specimens assigned to this species differed in some respects from the descriptions by NIELSEN AND CHRISTENSEN (1959) and SCHMELZ (2003). Therefore an account of characters in agreement and disagreement with the above descriptions is given.

Size about the same: 10-14 mm long, diameter  $330-400 \mu$ m at VIII and  $370-450 \mu$ m at the clitellum [similarly in SCHMELZ (2003): 12-19 mm and up to 400  $\mu$ m in XII] (*in vivo*),

length of fixed specimens 8–13 mm, segments 42–54 [in SCHMELZ (40)–48–58–(67)]. Similarly the maximum number of chaetae 5 or 6 and in the bundles of the posterior body part mostly two chaetae. In NIELSEN and CHRISTENSEN (1959) the innermost pair of chaetae much smaller than the outer pairs, in SCHMELZ (2003) the inner chaetae in the preclitellar bundles almost as long as the outer ones (this is a mistake, R. M. Schmelz, pers. comm.). In our specimens the outer chaetae longer and thicker than the inner within a bundle (e.g. in a bundle with 6 chaetae in the preclitellar segment the outermost pair of chaetae 63  $\mu$ m long and 5  $\mu$ m wide, the middle one 55  $\mu$ m long and and 4.5  $\mu$ m wide and the inner-



Fig. 7. Micrograph of *Fridericia* cf. *alata*: A = brain, B = head pore (marked with arrow), C = epidermal gland cells, D = nephridium preclitellarly, E = oesophageal appendage of b type, F = colomocytes. (A–D and F *in vivo*, E fixed but not stained; scale bars = 50 μm).

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most only 30  $\mu$ m long and 2.5  $\mu$ m wide). In the terminal segments the maximum length of the chaetae 90–100  $\mu$ m, width 5–7  $\mu$ m. Head pore at 0/I (Fig. 7B), dorsal pores from VII. Contrary to SCHMELZ (2003) the epidermal gland cells not indisting but in the Hungarian specimens large and arranged in 3 transverse rows per segments (Fig. 7C). Body wall 30–40  $\mu$ m, cuticula < 1  $\mu$ m. Clitellum in XII–1/2XIII, girdle-shaped, hyalocytes and granulocytes



Fig. 8. Micrograph of *Fridericia* cf. *alata*: A = pharyngeal glands (the hindmost lobes of the 3rd glands marked with arrows), B = midgut pars tumida (marked with arrows), C = clitel-lar glands in rows dorsally, D = clitellar glands ventrally (before and between the bursal slits weakly developed glands marked with white arrows, the epidermal glandular thickening in XIII marked with black arrow), E = chylus cells in XII–XIV, F = epidermal glandular thickening in XIII (marked with arrow). (A, C, E, F *in vivo*, B and D stained ; scale bars = 50 μm).

arranged in rows (Fig. 8C). Ventrally before and between the male openings the gland cells weakly developed and mostly only granulocytes, after that hyalocytes and granulocytes are present alike (Fig. 8D). Brain (Fig. 7A) anteriorly convex, posteriorly slightly concave or truncate, about 150  $\mu$ m long, 1.8 times longer than wide *in vivo*, 100–135  $\mu$ m long, 1.4–1.7 times longer than wide in the fixed specimens. Oesophageal appendages (Fig. 7E) unbranched, coiled, long (b type) may extend into VII. First and second pair of pharyngeal glands united but the third pair separated dorsally, the 2nd and 3rd pair have long



**Fig. 9.** A–D. Micrograph of *Fridericia* cf. *alata*: A = sperm funnels (marked with black arrows, male copulatory organ marked with white arrow, B = penial slit (marked with arrow), C and D = spermathecae; E–F micrograph of *Fridericia tubulosa* paratype : E = spermatheca, F = male copulatory organs (A–C in vivo, D fixed but not stained, E and F stained; scale bars = 50 μm).

1995, K&H: KOTA & HEALY 1999, 5: SCHMELZ 2003)				
	F. gamotheca gamotheca Issel, 1905	F. gamotheca maroccoiensis Dózsa-Farkas, 1989	F. gamotheca hungarica ssp. n	
Segments	(37-42-50	28–33	(29)-31-37	
Length (mm)	I: 10–12; R: 10–15	5–7	7-10.4	
Diameter in XII (µm)	R: 409–613	400	350-490	
Max. no. of chaetae	6	4	6 (4)	
Lateral chaetae in XII	absent	absent	present	
Oesophageal appendage	unbranched or with short terminal branches	1–2 short termi- nal branches	short proximal and terminal branches	
Coelomo-mucocytes	I: 40–50 μm, R:37–43 μm, with very few granules, not at cell perifery	no size data, a type granules absent	22–40 μm, b type with few granules at cell perifery or many in the whole cell	
Dorsal vessel from	I: XVI; R: (XV)–XVI–XVII	XIV–XVII	XIII	
Preclitellar nephridia	5 pairs	5 pairs	5 pairs	
Pharyngeal glands	S: all united dorsally and all with ventral lobes	S: all separate dorsally	in IV united, in V–VI separate dorsally, all with ventral lobes	
Chylus cells	postclitellarly: R&H: XIII-1/2XV	preclitellarly: S: XI–XII	preclitellarly: X–XII	
Sperm funnel, length : diameter ratio	cylindrical, 3:1, collar as wide as funnel body	cylindrical, 3:1, collar as wide as funnel body	pear-shaped, 1.4– 1.6:1, collar narrower then funnel body	
Bursal slits	longitudinal with transverse component	S: longitudinal, transverse com- ponent not seen	longitudinal with transverse compo- nent	
Shape of diverticula of spermathecae, sperm in it	R: oval (square), S: no sperm in lumina of am- pullae and diverticula	globular, rarely sperm in lumina of diverticula	rounded, sometimes sperm in diverticula and ampullae	
Ectal glands of spermathecae	S: absent, R: absent except 2 specimens	present	present	
Mature eggs at a time	R:2, S: 1	2	2–4	
Occurrence	Italy I: Appennins SW of Modena, R: Tosca- na, Latium, Calabria, S: Galicia: la Coruña	Marocco (Rif, cental Atlas)	Hungary (Vértes Mountains)	

Table 3. Comparison of the three subspecies of *Fridericia gamotheca* (I: Issel 1905, R: Rota1995, R&H: Rota & Healy 1999, S: Schmelz 2003)

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ventral lobes, their distal end lobated and slightly extend into VII (Fig. 8A). Chloragocytes from V, light brown (in vivo), diameter 20–25  $\mu$ m. Dorsal vessel from XVI–XVIII (XIX–XX in SCHMELZ), blood colourless. Midgut pars tumida from XXIX-XXXVII, occupying 5-8 segments (not given in the original description). Five pairs of preclitellar nephridia (Fig. 7D) from 6/7 to 10/11 (but sometimes only four, in one case three pairs were observable), postseptale a little longer than anteseptale, efferent duct rising anteriorly. Coelomocytes (Figs 1I & 7F) numerous, mucocytes with blurred pale vesicles (type a/c) length 28–32  $\mu$ m in vivo, 20–25 µm fixed) lenticytes small, 5–6 µm long. Chylus cells between XII–XV, occupying 2.5–3 segments (Fig. 8E) (contrary to this in SCHMELZ: XIV-1/2XVII). Seminal vesicle large (occupying IX-XII or X-XII). Sperm funnel (Fig. 9A) large, 300-450 µm long in vivo, 250–360 µm when fixed, and 2–3 times longer than broad, collar slightly narrower than funnel body; vas deferens 12–15  $\mu$ m wide, spermatozoa ca 400  $\mu$ m long, heads 100–150  $\mu$ m (in vivo) (no dimensions given by SCHMELZ). Male copulatory organs (Fig. 9A) large, flattened, 220–270  $\mu m$  long, 75–88  $\mu m$  wide and 70–75  $\mu m$  high (fixed), the bursal slits (Figs 1J & 9B) with longitudinal and transverse components (H-shaped after SCHMELZ 2003). No subneural glands, but a glandular thickening of the epidermis occurs in XIII behind the ventral chaetal bundles (Figs 8D, F), having no connection with the ventral nerve cord. Spermathaecae (Figs 1H & 9C-D) nearly agree with the original description: one small (20–25 µm long) sessile ectal gland at the orifice of spermathecal ectal duct, ampullae have two elongate diverticula (about 55–75  $\mu$ m long), all three chambers with distinct lumen. The ectal ducts widening proximally into conical projection into ampullar lumen. Separate openings into oesophagus. The only difference is the length of the ectal ducts: 'the ducts ca. 1.5x body diameter, 400 µm' according to SCHMELZ (2003) but in the present specimens without exception the length of the duct not longer than the body diameter (220–310  $\mu$ m long and 18–20 µm wide, fixed). One to two mature eggs at a time.

67 specimens investigated in vivo, than fixed in 70% ethanol, 26 specimens were mounted in Euparal.

Differential diagnosis. These specimens, found in the Vértes Mountains differed from the description of *F. alata* given by SCHMELZ (2003) in the following characteristics: the epidermal gland cells well developed in three transverse rows (not indistinct); the distal end of ventral lobes at the third pair of pharyngeal glands lobated and slightly extend into VII; dorsal vessel from XVI–XVIII (in contrast to XIX–XX); chylus cells between XII–XV (in contrast to XIV–1/2XVII); a glandular epidermal thickening in XIII midventrally, and finally the length of the spermathecal ectal duct, which is not longer than the diameter of the body (in contrast to 1.5×).

The specimens of *F. alata* described by CHALUPSKÝ (1986) (which identity after opinion of SCHMELZ (2003) is unknown) also differs from our specimens among other things, in occurence of lateral chaetae in XII and reddish blood.

Because *F. tubulosa* Dózsa-Farkas, 1972 is somewhat similar in respect to its spermatheca (especially when alive), and oesophageal appendage, which is also of b type, I made some stained slides of several paratypes of *F. tubulosa* (P. 6). Differences were well visible: the spermathecal diverticula are larger in *F. tubulosa*, the ectal gland very large and duct shorter and stouter, gradually

but remarkably widening proximad, canal also widening, as also pointed out by SCHMELZ (2003) (Fig. 9E). The male copulatory organ in *F. tubulosa* is also rather large, but of a quite different shape (Fig. 9F).

## DISCUSSION

Based on the 42 species found in the Vértes Mountains we can say that the enchytraeid fauna of this area is quite diverse, and in its majority represented by species typical of the Hungarian, or wider Central European fauna. This mountain geologically shows a fairly uniform structure with dolomite rock-bed, but in some parts of the mountains: the southern foothills of the range, we can find submediterranean, and in the north-facing slopes alpinelike climatic conditions. Probably, this is reflected in the occurrence of the newly described subspecies (*F. gamotheca hungarica*). The main distribution area of this species is specifically in the Mediterranean region (Italy, Spain and Morocco). On the other hand, the presence of, e. g., *F. cf. alata* (if it is not a new species) may be connected to the colder climatic conditions, because *F. alata* has been found mainly in northern Europe so far.

*F. gamotheca hungarica* ssp. n. effectively showed the typical traits presented in the original species description (ISSEL 1905), its supplementary description made on the basis of more recent Italian material (Rota 1995, Rota & HEALY 1999), and the same description supplemented by SCHMELZ (2003) on the basis of Spanish material. However, it simultaneously differed in several traits from the latter and from the form I described from Morocco (Dózsa-FARKAS 1989). As SCHMELZ (2003) has already proposed that these differences 'suggest at least a geographic differentiation of the species into a European and a North African subtaxon', I propose to separate this species into three subspecies: *F. gamotheca gamotheca* Issel, 1905, *F. gamotheca maroccoiensis* Dózsa-Farkas, 1989 and *F. gamotheca hungarica* ssp. n. I described the new subspecies accordingly. For a better interpretation I summarized the typical characteristics of the three subspecies in Table 3.

The specimens, found in the Vértes Mountains and described as *F. cf. alata*, differed from the description given by NIELSEN and CHRISTENSEN (1959) and SCHMELZ (2003) but at present I can't decide that these differences are within what can be considered intraspecific variance or a new species come into question, but additional foreign material is necessary to assess the taxonomic status of the Vértes population.

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