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# Morphological variation of *Fuscheria terricola* BERGER et al., 1983 (Ciliophora, Haptoria)

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A b s t r a c t : The morphology and infraciliature of *Fuscheria terricola* BERGER et al., 1983 were studied using a high-power oil objective and differential interference contrast optics. The infraciliature and the nuclear apparatus were revealed by protargol impregnation. This species was studied in two populations from flooded soils in Suchohrad and Jakubovské rybníky, Western Slovakia. Both populations match the authoritative description by BERGER et al. (1983), especially, in body size and number of ciliary rows. However, data from the present investigation, as well as those from the literature on *F. terricola* show that this species has many more or less distinct morphotypes and most of the variability concerns the extrusomal size (length 3-10  $\mu$ m), the body shape (bottle-shaped to cylindroidal), the shape of the macronucleus (ellipsoidal to horseshoe-shaped), the number of the micronuclei (1-3), as well as the fine structure and the pattern of the dorsal brush.

K e y w o r d s : Flooded soil, morphometry, morphotype, Slovakia, soil ciliates.

# Introduction

KAHL (1930, 1931) brought together most haptorids known to that time, discovered and described many haptorid taxa, and emphasized that their diversity is much greater than previously recognized. In general, the use of KAHL's morphospecies concept in modern ciliate taxonomy has revealed many distinct species (e.g. PETZ et al. 1995, FOISSNER et al. 2002, FOISSNER & XU 2006, SONG & WILBERT 1989 etc.). However, several ciliate species have displayed more or less distinct morphotypes which are often interpreted as part of the natural variability of a species (e.g. FOISSNER et al. 2001). Also, the growing data on *Fuscheria terricola* BERGER et al., 1983, a commonly found haptorid in terrestrial habitats worldwide, have begun to indicate that this species covers a "mass" of morphotypes or even species hardly recognizable at morphological level.

During a survey on soil ciliate fauna in Slovakia, two *F. terricola* populations from flooded soils in Western Slovakia were studied in detail. The present paper gives notes on morphological variation of the species studied.

# Material and methods

Sampling and culture methods: Fuscheria terricola was studied in two

populations from flooded regions in Western Slovakia. Namely, one population was found in a leaf-litter sample consisted of poplar litter, with a pH of 6.0, which I collected on April 16, 2004 at a pond dam with tree cover composed of poplars and willows in Jakubovské rybníky, at 150 m above sea level in Borská nížina lowland (48°25'N, 16°58'E). The other isolate was obtained from a sample composed of poplar and ash leafs with a pH of 5.0, collected on February 2, 2004 in hard-meadow forest in Suchohrad, Borská nížina lowland (48°24'N, 16°51'E).

The samples were collected and processed as described in FOISSNER et al. (2002), that is, with the non-flooded Petri dish method.

Morphological methods: The ciliates were studied in vivo and after protargol preparation. Body shapes of live specimens were drawn from preparations without coverslip. Details were studied on slightly to heavily squeezed individuals, using an oil immersion objective and interference contrast. Live measurements were made at magnifications of  $100 \times to 1,000 \times$ . The infraciliature was revealed with the protargol method according to protocol A in FOISSNER (1991). Counts and measurements on prepared specimens were performed at a magnification of  $1,000 \times$ . Illustrations of live specimens are based on freehand sketches and represent summaries of the observations of live and prepared cells, while those of prepared cells were made with a drawing device. Morphometry is based on well-impregnated specimens, and derived parameters were calculated according to statistics textbooks.

Terminology is basically according to FOISSNER & FOISSNER (1988) and FOISSNER & XU (2006).

#### Results

#### Fuscheria terricola BERGER, FOISSNER & ADAM, 1983

1983 Fuscheria terricola BERGER, FOISSNER & ADAM, J. Protozool. **30**: 529 (original description; description of ontogenesis).

1988 *Fuscheria terricola* BERGER et al., 1983 – FOISSNER & FOISSNER, Arch. Protistenk. **135**: 213-235 (description of fine structures; revision of haptorids).

2002 *Fuscheria terricola* BERGER et al., 1983 – FOISSNER et al., Denisia **5**: 191 (description of a Namibian population with short extrusomes).

M a t e r i a l: This species was studied in two populations, namely from Jakubovské rybníky (pond dam) and Suchohrad (hard-meadow forest). The later population matches the original description very well in the main taxonomical features and is thus described very briefly. Since the isolate from Jakubovské rybníky differs in several taxonomical features such as the fine structure of the dorsal brush and the number of the micronuclei, it is described in detail.

Morphological description of Jakubovské rybníky population: Size about  $60-90 \times 25-45 \mu m$ , usually about  $80 \times 35 \mu m$  in vivo. Outline elliptical or slightly asymmetrical, length:width ratio about 2.4:1 on average in protargol preparations, posterior body end more broadly rounded than the anterior one (Fig. 1). Macronucleus in mid-body or slightly below it, U-shaped (Figs 4, 5, 13-17), rarely Jshaped (Figs 11, 18, 20), with globular nucleoli. Micronucleus globular, about 3.4  $\mu m$ across, near macronucleus; out of 28 protargol-impregnated specimens only two





**Figs 1-10**: *Fuscheria terricola* (Jakubovské rybníky population) from life (1, 4-10) and after protargol impregnation (2, 3). 1: Left side view of a representative specimen. **2**, **3**: Ciliary pattern of dorsal and ventral side and nuclear apparatus. **4**, **5**: Lateral views showing variants of body shape and nuclear pattern. **6**: Structure of dorsal brush. **7**: Oral bulge extrusomes are nail-shaped, 4-6.5 µm long. **8**: Anterior body portion. **9**: Frontal view of oral bulge filled with extrusomes. **10**: Surface view showing cortical granulation. CG – cortical granules, CV – contractile vacuole, B2 – dorsal brush row 2, Ex – extrusomes, MA – macronucleus, MT – monokinetidal bristle tail of brush row 2, N – nematodesmata, OB – oral bulge, SK – somatic kinety. Scale bars 30 µm.







**Figs 22-30**: *Fuscheria terricola* from several populations. **22**: Left side view of a representative specimen from Suchohrad population. **23**, **24**: Ciliary pattern of dorsal and ventral side of Suchohrad population specimen. **25-27**: Specimen from type, Salzburg (from BERGER et al. 1983) and Namibian population (from Foissner et al. 2002). **28**: Specimen after protargol preparation from Suchohrad population. **29**, **30**: Specimen after protargol preparation from Jakubovské rybníky population. B1 – dorsal brush row 1, Ex – extrusomes, N – nematodesmata, SK – somatic kinety. Scale bars 30 μm.

Characteristics	Mean	М	SD	CV	SE	Min	Max	n
Body, length	71.7	71.1	7.8	10.9	1.5	56.2	89.1	28
Body, width	30.4	31.2	4.9	16.0	0.9	20.3	42.2	28
Body length:width, ratio	2.4	2.4	0.5	19.9	0.1	1.6	3.8	28
Oral bulge, width	5.2	4.8	0.7	13.8	0.1	3.9	6.3	28
Oral bulge, height	1.6	1.6	0.4	23.0	0.1	1.0	2.3	28
Oral basket, length of longest nematodesmal bundle	17.2	16.4	3.8	21.9	0.8	11.0	28.1	23
Anterior body end to macronucleus, distance	31.0	31.2	7.9	25.4	1.5	12.5	46.9	28
Macronucleus, length	27.5	26.6	6.2	22.3	1.2	18.7	48.4	28
Macronucleus, width	8.3	8.6	1.0	12.0	0.2	6.2	9.7	28
Macronucleus, number	1.0	1.0	0.0	0.0	0.0	1.0	1.0	28
Micronuclei, largest diameter	3.4	3.3	0.6	16.8	0.1	2.5	5.0	24
Micronuclei, number	1.2	1.0	0.5	41.3	0.1	1.0	3.0	24
Circumoral kinety to end of brush row 1, distance	10.1	10.9	2.4	24.1	0.5	4.7	14.1	23
Circumoral kinety to end of brush row 2, distance	8.4	7.8	1.9	22.1	0.4	4.7	12.5	23
Ciliary rows, number	17.1	17.0	2.0	11.9	0.4	13.0	22.0	28
Ciliated kinetids in a lateral kinety, number	43.3	44.5	6.1	14.2	1.2	32.0	56.0	28
Oralized somatic monokinetids in a kinety, number	6.4	6.0	1.5	23.8	0.3	4.0	11.0	28
Dikinetids in brush row 1, number	13.2	13.0	2.4	18.1	0.6	10.0	16.0	18
Dikinetids in brush row 2, number	9.4	9.5	2.4	25.3	0.6	6.0	15.0	18
Brush rows, number	2.0	2.0	0.0	0.0	0.0	2.0	2.0	28

Table 1: Morphometric data on Fuscheria terricola population from Jakubovské rybníky.

Measurements in  $\mu$ m. Data based on randomly selected protargol-impregnated specimens from non-flooded Petri dish culture. CV – coefficient of variation in %, M – median, Max – maximum, Mean – arithmetic mean, Min – minimum, n – number of individuals investigated, SD – standard deviation, SE – standard error of arithmetic mean.

possessed 2 micronuclei (Fig. 19) and one had 3 micronuclei. Extrusomes arranged centrally in oral bulge, nail-shaped and 4-6.5  $\mu$ m long (Figs 7, 9). Many developing extrusomes scattered in cytoplasm, fusiform, clavate or rod-like after protargol impregnation (Figs 11, 13).

Cilia about 7  $\mu$ m long in vivo, rather widely spaced, that is, on average 43 ciliated kinetids in an ordinary kinety; arranged in 17 longitudinal, equidistant rows commencing

underneath circumoral kinety and extending to posterior polar area (Figs 2, 3, 11-14). Dorsal brush two-rowed, occupies about 14% of body length; dikinetidal portion of row 1 slightly longer than portion of row 2 (Table 1). Row 1 on average 10.1  $\mu$ m long, composed of on average of 13.2 dikinetids associated with clavate anterior bristles gradually increasing in length from 3  $\mu$ m anteriorly to 4.5  $\mu$ m in half of brush length and rod-shaped posterior bristles (0.5  $\mu$ m long). Brush row 2 usually commences with 1-2 monokinetids, 8.4  $\mu$ m long on average, individual dikinetids associated with ellipsoidal anterior bristles of uniform length (3  $\mu$ m) and conspicuously shorter (0.5  $\mu$ m long) rod-shaped posterior bristles (Fig. 6); continues posteriorly with monokinetidal bristle tail extending to mid-body having ellipsoidal bristles, about 3  $\mu$ m long (Figs 1, 8).

Oral apparatus apical, oral bulge ring-shaped and inconspicuous because only 5.5 µm wide and 2 µm high in vivo, mouth centre slightly depressed. Circumoral kinety at base of oral bulge, composed of rather widely spaced dikinetids at the top of each ciliary row (Figs 2, 3, 11-14). Nematodesmata originate from circumoral basal bodies and 4-11 ciliated basal bodies in anterior region of all somatic kineties (oralized somatic kinetids); oral basket inconspicuous in vivo as composed of small bundles (Fig. 8).

Notes on morphology of Suchohrad population: Size about  $90 \times 40 \mu m$ , length:width ratio 2.2:1 in protargol preparations. Macronucleus ellipsoidal, typically accompanied by single micronucleus about 2.7  $\mu m$  across. Extrusomes nail-shaped, only distal portion impregnated with protargol, about 5  $\mu m$  long. On average 15 somatic ciliary rows. Dense group of on average 7 kinetosomes in anterior part of second somatic kinety left of brush row 2 (Figs 23, 24, 28). Two ciliary rows anteriorly differentiated to dorsal brush: brush row 1 with about 8 dikinetids; brush row 2 with 4 dikinetids. Both rows with same bristle composition, that is, dorsal bristles of uniform length and shape. Row 2 continues posteriorly with monokinetidal tail extending to mid-body (Fig. 22).

# Discussion

Morphological variation: *Fuscheria terricola* is highly variable, especially, in body (bottle-shaped to cylindroidal) and macronuclear shape (horseshoe-shaped, stretched or helical), as mentioned in the original description (BERGER et al. 1983). Moreover, growing morphologic data have shown that the variability also concerns the length of the extrusomes (length 3-10  $\mu$ m), the number of the micronuclei (1-3), the fine structure and pattern of the dorsal brush, and the presence/absence of a subapical ciliary condensation in the second or third kinety left of brush row 2 (FOISSNER et al. 2002, present paper). However, *F. terricola* populations investigated in detail share well all main taxonomical features, viz., body size ( $85 \times 30 \mu$ m), number of ciliary rows (on average 16), single macronucleus localized in mid-body, shape and arrangement of ripe extrusomes, and contractile vacuole pattern. Wherefore, a population outstanding in several characteristics (indistinct subapical ciliary condensation and different structure of dorsal brush), isolated from Jakubovské rybníky, was found to be a variation of *F. terricola* and was identified as conspecific with this species, on the basis of the usually high variability revealed in *F. terricola* earlier.

Comparison of Fuscheria terricola populations: My data on the population from Suchohrad match almost perfectly the original descriptions by BERGER

et al. (1983) so that the identification is beyond doubt. Thus, only insignificant differences between the Suchohrad and the type population in the body size  $(90 \times 40 \ \mu m \ vs.)$  $80-100 \times 27 \ \mu\text{m}$ ), the length of the extrusomes (5  $\mu$ m vs. 5-7  $\mu$ m), the structure of the dorsal brush (brush row 1 with 8 vs. 10 dikinetids; brush row 2 with 4 vs. 5 dikinetids), were found. However, F. terricola isolate found at locality Jakubovské rybníky differs from other F. terricola populations by having much more micronuclei (1-3 vs. invariably 1), different structure of the dorsal brush and an indistinct subapical ciliary condensation. Namely, this isolate has brush row 2 only slightly shorter than row 1, viz., row 2 consisted of an average of 9.4 dikinetids and is an average of 8.4 µm in length, whilst row 1 is composed of an average of 13.2 dikinetids and measures 10.1 µm on average. Whereas, type (Austrian), Namibian and Suchohrad populations possess brush row 2 occupying only half of the length of row 1, that is, row 1 is composed of an average of 9.6 dikinetids and measures an average of 9.1 µm and row 2 consisted of 4 dikinetids on average and is about 4.4 µm in length (based on average values from three populations described in BERGER et al. 1983 and FOISSNER et al. 2002). Moreover, this isolate differs also in the shape of the dorsal bristles, namely the row 1 dikinetids are associated with clavate anterior bristles gradually increasing in length and rod-shaped posterior bristles, whilst each dikinetid of row 2 is associated with an ellipsoidal anterior bristles and a conspicuously shorter rod-shaped posterior one, but anterior and posterior bristles of both rows are of uniform length and shape in all other F. terricola populations.

C o m p a r i s o n w i th r e l a t e d s p e c i e s : *Fuscheria* species are very similar at first glance, chiefly in body shape, nuclear and contractile vacuole pattern. Nevertheless, they differ mainly in the number of the ciliary rows, the structure of the dorsal brush, and the length of the extrusomes (Table 2). *Fuscheria lacustris* cannot be confused with *F*. *terricola* because is much smaller ( $50 \times 30 \mu m vs. 85 \times 30 \mu m$ ) and possesses a higher number of ciliary rows (25 vs. 16). *Fuscheria nodosa* differs from *F. terricola* by a smaller cell size ( $65 \times 30 \mu m vs. 85 \times 30 \mu m$ ), longer extrusomes ( $10-13 \mu m vs. 3-10 \mu m$ ), a higher number of ciliary rows (27 vs. 16), and many more micronuclei (3-6 vs. 1-3). *Fuscheria marina* is distinguished by the marine habitats, the body size ( $120 \times 40 \mu m vs. 85 \times 30 \mu m$ ), and the number of somatic kineties (36 vs. 16).

Occurrence and ecology: *Fuscheria terricola* is a common inhabitant of terrestrial habitats. BERGER et al. (1983) discovered it in the soil of a bottomland near Grafenwörth, Austria and in the soil from the Schlossalm, Salzburg, Austria. Later, it was found in terrestrial habitats from Holoarctic, Paleotripic and Neotropic regions and Antarctica, but there are no records so far from Australian region (FOISSNER 1998). FOISSNER et al. (2002) isolated their populations in mud and soil from Benin and Namibia, Africa. I found *F. terricola* in flooded soils from two localities in Western Slovakia (further details on the sample sites, see materials and methods). It feeds on small- to medium-sized ciliates (FOISSNER 1998).

Species <sup>1</sup>	Average size in vivo	Extruso- me size	Ciliary rows, number	Dorsal brush	Ecology
Fuscheria lacustris SONG & WILBERT 1989	$50\times 30~\mu m$	9-12 µm	25 (22-30)	Row 2 is one third of row 1 length	Freshwater
<i>Fuscheria marina</i> PETZ et al. 1995	$120\times 40~\mu m$	5-6 µm	36 (34-39)	Row 2 is one fifth of row 1 length	Marine
<i>Fuscheria nodosa</i> FOISSNER 1980	$65\times 30~\mu m$	10-13 μm	27 (24-30)	Row 2 slightly shorter than row 1	Freshwater
<i>Fuscheria terricola</i> BERGER et al. 1983	$85\times 30~\mu m$	3-10 μm	16 (12-24)	Row 2 is half of row 1 length or both rows of the same length	Terrestrial habitats

Table 2: Comparison of main taxonomic features in Fuscheria.

<sup>1</sup> Data on *F. lacustris* are from SONG & WILBERT (1989); those on *F. marina* are from PETZ et al. (1995); those on *F. nodosa* from FOISSNER (1980) and FOISSNER & O'DONOGHUE (1990); those on *F. terricola* are from BERGER et al. (1983), FOISSNER et al. (2002) and present investigation.

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# Zusammenfassung

Zwei Populationen von *Fuscheria terricola* BERGER et al., 1983 aus Böden der Westslowakei wurden morphologisch untersucht, wobei Lebendbeobachtung und Protargolimprägnation angewendet wurden. Die beiden Populationen entsprechen in Zellgröße und Anzahl der Cilienreihen der Originalbeschreibung. Andere taxonomische Merkmale zeigen hingegen eine größere Variabilität und deuten auf die Präsenz verschiedener Morphotypen hin. So zeigen die neuen Ergebnisse zusammen mit Literaturdaten eine hohe Variabilität in der Größe der Extrusomen (3-10 µm lang), in der Körperform (flaschenförmig bis zylindrisch), der Form des Makronukleus (elliptisch bis hufeisenförmig), der Anzahl der Mikronuklei (1-3) als auch in der Feinstruktur und dem Muster der Dorsalbürste. Ob *F. terricola* eine sehr variable Art ist oder aus vielen schwer differenzierbaren (kryptischen) Arten besteht, muss mit molekularbiologischen Methoden überprüft werden.

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