

***Urotricha spetai* nov. spec., a new Plankton Ciliate (Ciliophora, Prostomatea) from a Fishpond in the Seidlwinkel Valley, Rauris, Austrian Central Alps*)**

Wilhelm FOISSNER

The genus *Urotricha* contains about 30 well-defined species, most having been studied in vivo and as silver preparations. This applies also to *Urotricha spetai* nov. spec., a medium-sized ($\sim 50 \times 45 \mu\text{m}$) congener with the following main characteristics: body conspicuous because very broadly obovate; extrusomes bluntly fusiform and $5 \mu\text{m}$ long, forming a distinct fringe; on average 42 ciliary rows, 4 caudal cilia in line, 21 oral dikinetids, and 3 adoral membranelles. *Urotricha spetai* is possibly most closely related to *U. castalia* and *U. matthesi*.

FOISSNER W., 2012: *Urotricha spetai* nov. spec., ein neues planktisches Ciliat (Ciliophora, Prostomatea) aus einem Fischteich im Seidlwinkel, Rauris, österreichische Zentralalpen.

Die Gattung *Urotricha* enthält etwa 30 gut definierte Arten, von denen die meisten in vivo und in Silberpräparaten untersucht worden sind. Das gilt auch für *U. spetai*, eine mittelgroße ($\sim 50 \times 45 \mu\text{m}$) Art mit folgenden Hauptmerkmalen: Gestalt auffallend, weil sehr breit obovate; Extrusome plump spindelförmig und $5 \mu\text{m}$ lang, bilden einen deutlichen Saum; im Mittel 42 Wimpernreihen, 4 Caudalcilien, 21 orale Dikinetiden und 3 adorale Membranellen. *Urotricha spetai* ist vermutlich nahe verwandt mit *U. castalia* und *U. matthesi*.

Key words: Alpha-taxonomy, biodiversity, silver impregnation, *Urotricha castalia*, *Urotricha matthesi*.

Introduction

The genus *Urotricha* contains very small ($\leq 20 \mu\text{m}$) to middle-sized ($\sim 100 \mu\text{m}$) prostomatids with the following characteristics (FOISSNER and PFISTER 1997): “Plagiocampidae (?) with direct and indirect silverline pattern and somatic ciliary rows distinctly shortened posteriorly, leaving blank a more or less wide area occupied by one or many caudal cilia. Circumoral kinety (undulating membrane) circular and closed. Adoral membranelles (brosse) underneath circumoral kinety, interrupt somatic ciliary rows, usually enklitolph-dextiotrop and composed of two to many short, oblique rows arranged one after the other. Most species swim rapidly and can perform conspicuous jumps”.

Presently, the genus contains about 30 well-defined species, most of which were investigated in silver preparations and were reviewed by FOISSNER (1997), FOISSNER & PFISTER (1997), FOISSNER et al. (1999), and SONNTAG & FOISSNER (2004). The new species, *U. spetai*, attracted my attention by a unique combination of features, i.e., a very broadly obovate body and four elongated caudal cilia.

The ciliates of the Austrian Central Alps are comparatively well known because I studied them for years. About 200 species have been found in pools and streams and about 90 in various soils (FOISSNER 1980, FOISSNER & PEER 1985). One third of this species was undescribed. These are impressive numbers. In spite of this, a glance into the plankton

*) Dedicated to my mentor and friend, Hofrat Univ.-Doz. Dr. Franz SPETA, former head of the Biologiezentrum of the Oberösterreichische Landesmuseum in Linz, on occasion of his 70th birthday.

of a fishpond in the Rauris area immediately revealed a further new species, *U. spetai*, described in this contribution.

Materials and methods

Urotricha spetai was discovered in a fertilized fishpond in the Seidlwinkeltal, Rauris, Austrian Central Alps on 29th May 2010 (further details, see type locality). There were two ponds close together, but *U. spetai* occurred only in that which was nearer to the street. The ciliate was obtained by filtering about 15 l pond water through a plankton net with 12 µm mesh size. *Urotricha spetai* was moderately abundant in the filtered sample (500 ml), which was transported to the Salzburg laboratory, where the in vivo observations and silver preparations were performed, according to the protocols given by FOISSNER (1991). Culture attempts failed.

Counts and measurements of silvered specimens were performed at a magnification of 1,000 ×. In vivo measurements were conducted at magnifications of 100 – 1,000 ×. Illustrations of live specimens were based on free-hand sketches and micrographs; those of impregnated cells were made with a drawing device.

Results

Urotricha spetai nov. spec.

Diagnosis: Size about 50 × 45 µm in vivo; does not jump. Shape conspicuous because broadly to very broadly obovate and non-ciliated posterior portion hardly set off from body proper. Macronucleus broadly ellipsoidal. Excretory pore of contractile vacuole subterminal, outside of row formed by caudal cilia. Extrusomes bluntly fusiform, 4–5 µm long, form a distinct fringe. On average 42 ciliary rows extending 78% of body length; usually 4 elongated caudal cilia, forming a short row on posterior pole. Oral opening about 10 µm across in vivo, surrounded by an average of 21 dikinetids; 3 oblique adoral membranelles composed of 6, 5, and 4 bristles, respectively.

Type locality: Plankton of a fishpond in the Austrian Central Alps, Rauris area, Seidlwinkel valley, 47°9' 9.60"N/12°53'27.86"E, 1293 m above sea level.

Type material: 1 holotype and 3 paratype slides with silver nitrate-impregnated (Chatton-Lwoff method), environmental specimens have been deposited in the Biologiezentrum of the Oberösterreichische Landesmuseum in Linz (LI). Furthermore, 4 paratype slides with protargol-impregnated (FOISSNER's method) specimens have been deposited in the same locality.

Dedication: I dedicate this species to Hofrat Univ.-Doz. Dr. Franz SPETA on the occasion of his 70th birthday, appreciating the employment of a protistologist as a curator in the Biology Centre of the Upper Austrian Museum in Linz (LI).

Description: Size 40–60 × 35–55 µm in vivo, usually about 55 × 50 µm, length: width ratio thus 1:1 – 1.2:1, on average 1.1:1 (Table 1). Shape conspicuous because broadly to very broadly obovate or obampulliform when the posterior, non-ciliated portion is slightly constricted; unflattened. Anterior end very broadly rounded, posterior narrowly rounded, never acute (Fig. 1, 6–8, 12, 14–23). Nuclear apparatus in or near mid-body, slightly nearer to ventral side. Macronucleus globular to ellipsoidal, on average broadly ellipsoidal, i.e., 13 × 10 µm in protargol preparations; nucleoli not recognizable in vivo.

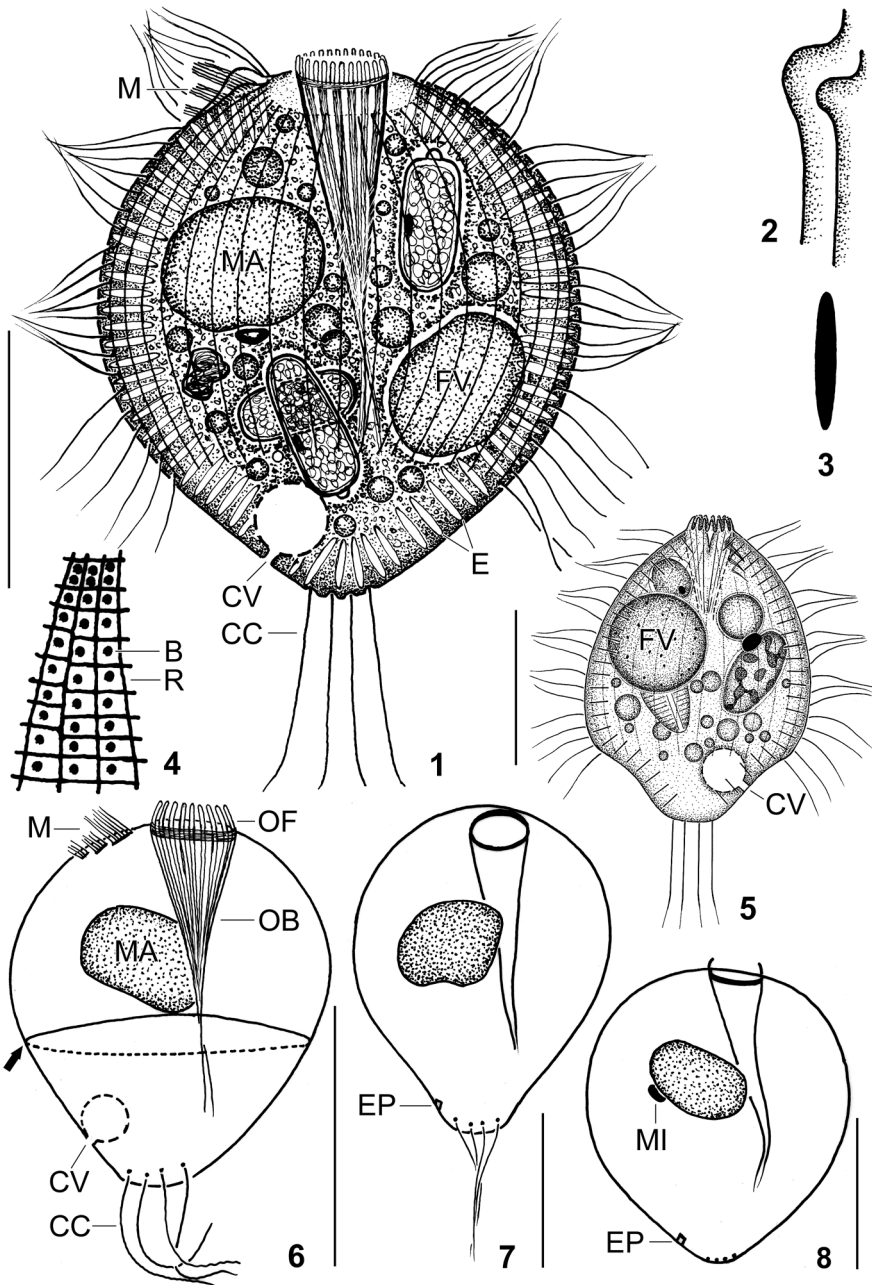


Fig. 1–8: *Urotricha spetai* (1–4, 6–8) and *U. matthesi tristicha* (5, from FOISSNER & PFISTER 1997) from life (1–5) and after protargol impregnation (6–8). 1: Left side overview of a representative specimen, length 50 μm . 2: Ridges between adoral membranelles. 3: Extrusome, length 5 μm . 4: Cortex pattern. 5: Overview. 6–8: Shape and size variability. The arrow in Fig. 6 marks the end of the ciliary rows. B – basal body, CC – caudal cilia, CV – contractile vacuole, E – extrusomes, EP – excretory pore, FV – food vacuole, M – adoral membranelles, MA – macronucleus, MI – micronucleus, OB – oral basket, OF – oral flaps, R – ridges. Scale bars 20 μm (Fig. 5–8) and 25 μm (Fig. 1).

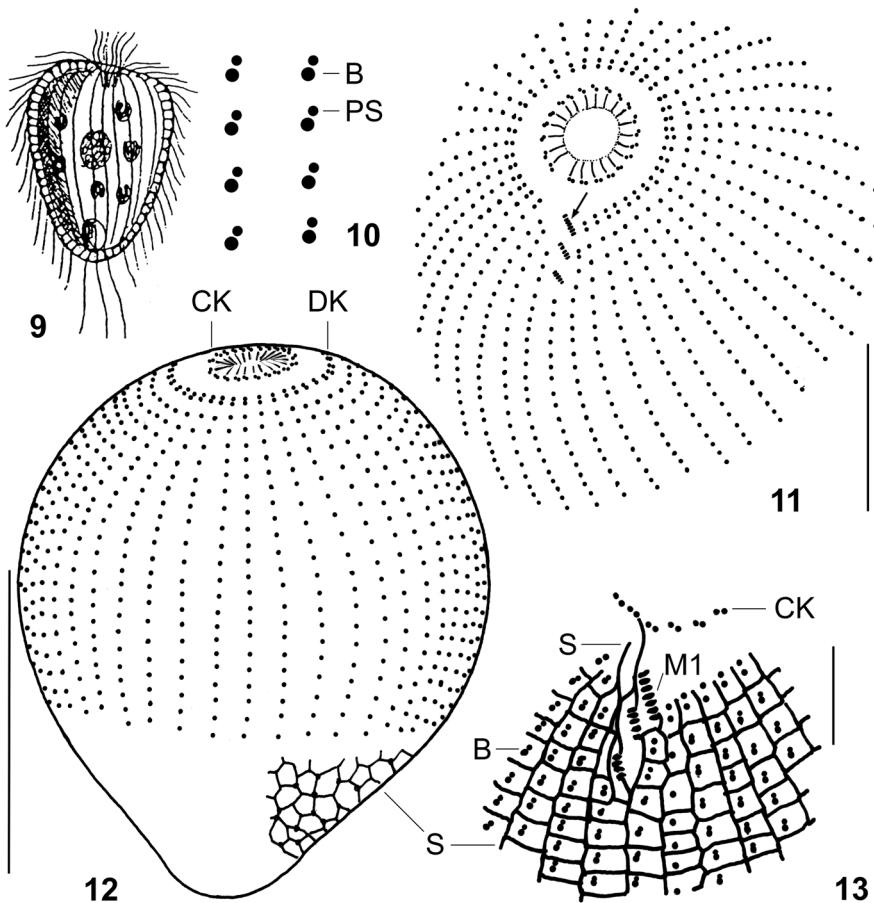


Fig. 9–13: *Urotricha alveolata* (9, from FOISSNER & PFISTER 1997) and *U. spetai* (10–13) from life (9) and after silver nitrate impregnation. 9: This species has large cortical alveoli, length 30–40 μm . 10: Detail of somatic ciliary rows. 11: Oblique frontal view of holotype specimen, showing the arrangement of the oral and somatic ciliary pattern. The arrow marks the adoral membranelles. 12: Lateral overview, showing the very broadly obovate body shape and the somatic ciliary pattern. 13: Silverline and basal body pattern in the area of the adoral membranelles. B – basal body, CK – circumoral kinety, DK – dikinetics, M1 – adoral membranelle 1, PS – parasomal sac, S – silverlines. Scale bars 5 μm (Fig. 13), 15 μm (Fig. 11), and 25 μm (Fig. 12).

Micronucleus likely lenticular, about 3 μm in size (Fig. 1, 6–8, 21, 22, 24, 26, 27, 32, 39, Table 1). Contractile vacuole subterminal anterior of caudal cilia, with single excretory pore on ventral side, i.e., the brosse area (Fig. 1, 6–8, 23, 27). Extrusomes as numerous as cilia, producing conspicuous fringe; one each in transverse cortical ridges between cilia; appear as bright dots in surface view; individual extrusomes bluntly fusiform, 4–5 μm long, compact (Fig. 1, 3, 17, 25, 27–29, 31, 43). Cortex rigid, with ridge and silverline pattern as typical for urotrichs (FOISSNER & PFISTER 1997, FOISSNER et al. 1999), i.e., quadrangular meshes with cilia right of centre; non-ciliated posterior portion with polygonal

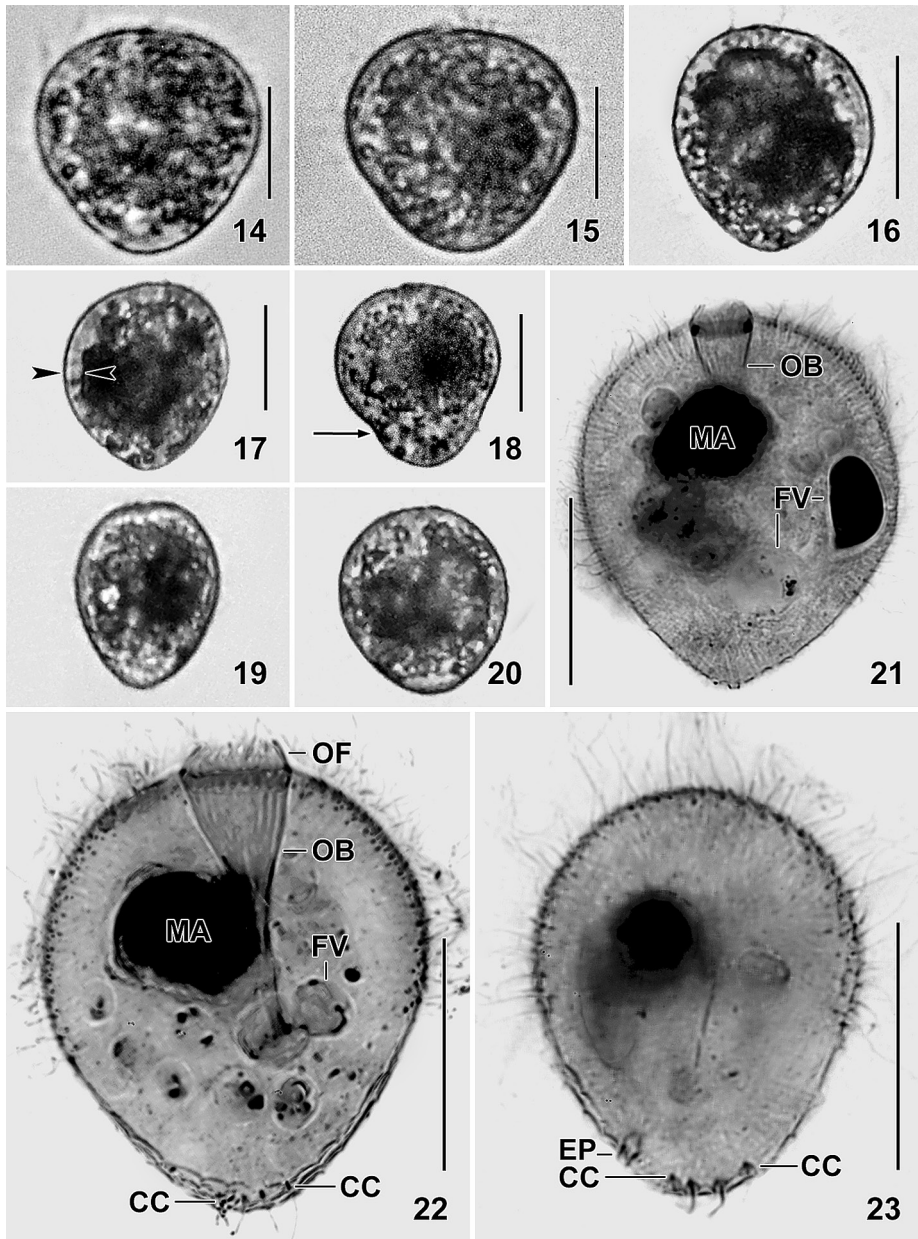


Fig. 14–23: *Urotricha spetai* from life (14–20) and after protargol impregnation (21–23). 14–20: Freely motile specimens, showing the characteristic obovate to broadly obovate body shape. The specimens appear spotted due to various food inclusions. The arrowheads mark the fringe produced by the extrusomes. Only one of the seven specimens shown has the non-ciliated posterior body portion plug-like set off (18, arrow). 21–23: The broadly obovate body shape is perfectly preserved in protargol preparations, in which, inter alia, the oral basket and the basal bodies of the four caudal cilia are recognizable. CC – caudal cilia, EP – excretory pore, FV – food vacuoles, MA – macronucleus, OB – oral basket, OF – oral flaps. Scale bars 25 μ m.

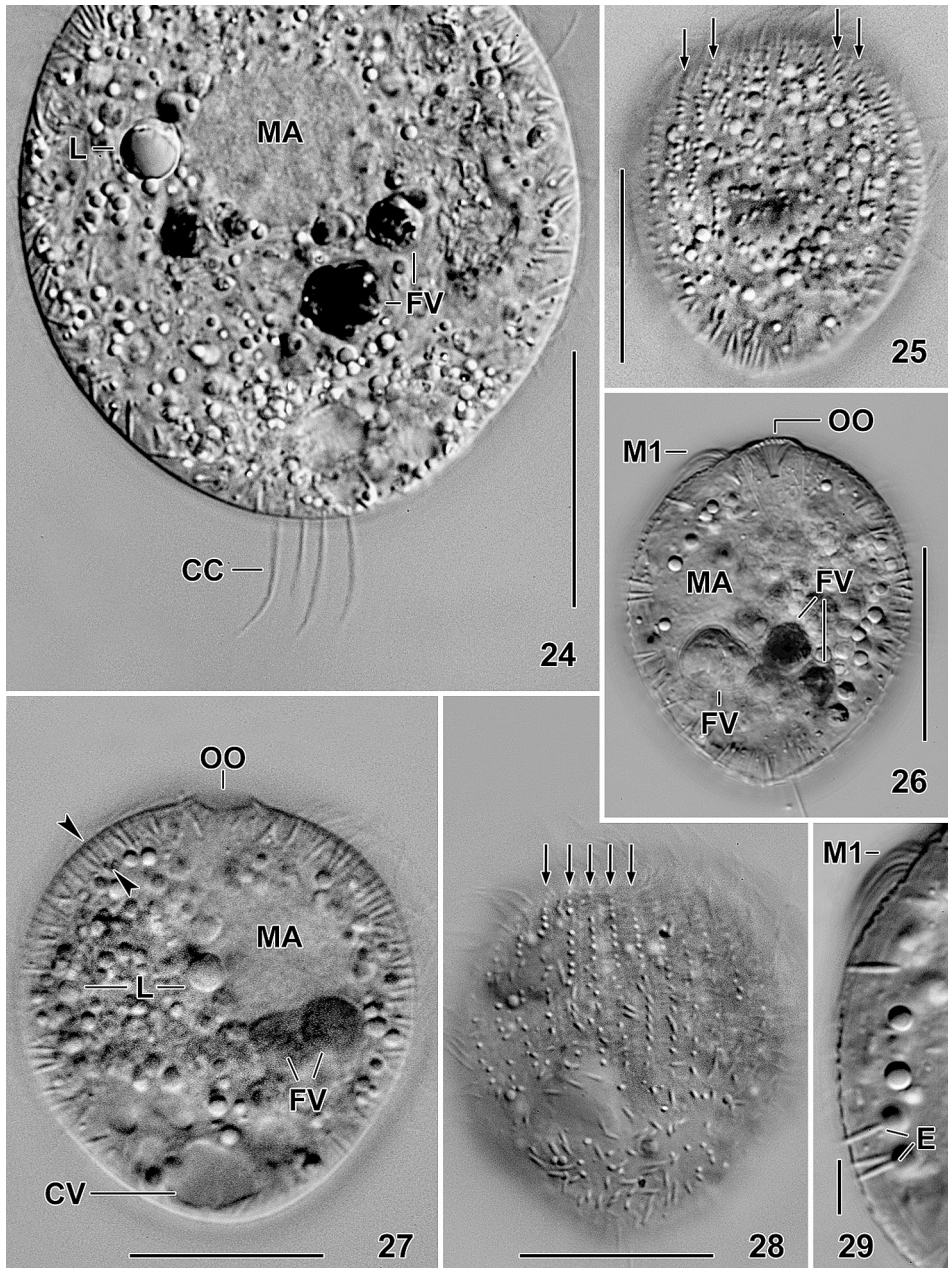


Fig. 24–29: *Urotricha spetai*, live micrographs of slightly squashed specimens. 24: *U. spetai* has four caudal cilia and deeply orange food vacuoles. 25–28: Cells focused to surface and centre to show main cell organelles and the distinct granule rows (arrows) and conspicuous fringe (opposed arrowheads) produced by the extrusomes, respectively. 29: Adoral membranelle 1 and some bluntly fusiform, 4–5 µm long extrusomes. CC – caudal cilia, CV – contractile vacuole, E – extrusomes, FV – food vacuoles, L – lipid droplets, M1 – adoral membranelle 1, MA – macronucleus, OO – oral opening, Scale bars 5 µm (29) and 25 µm (24–28).

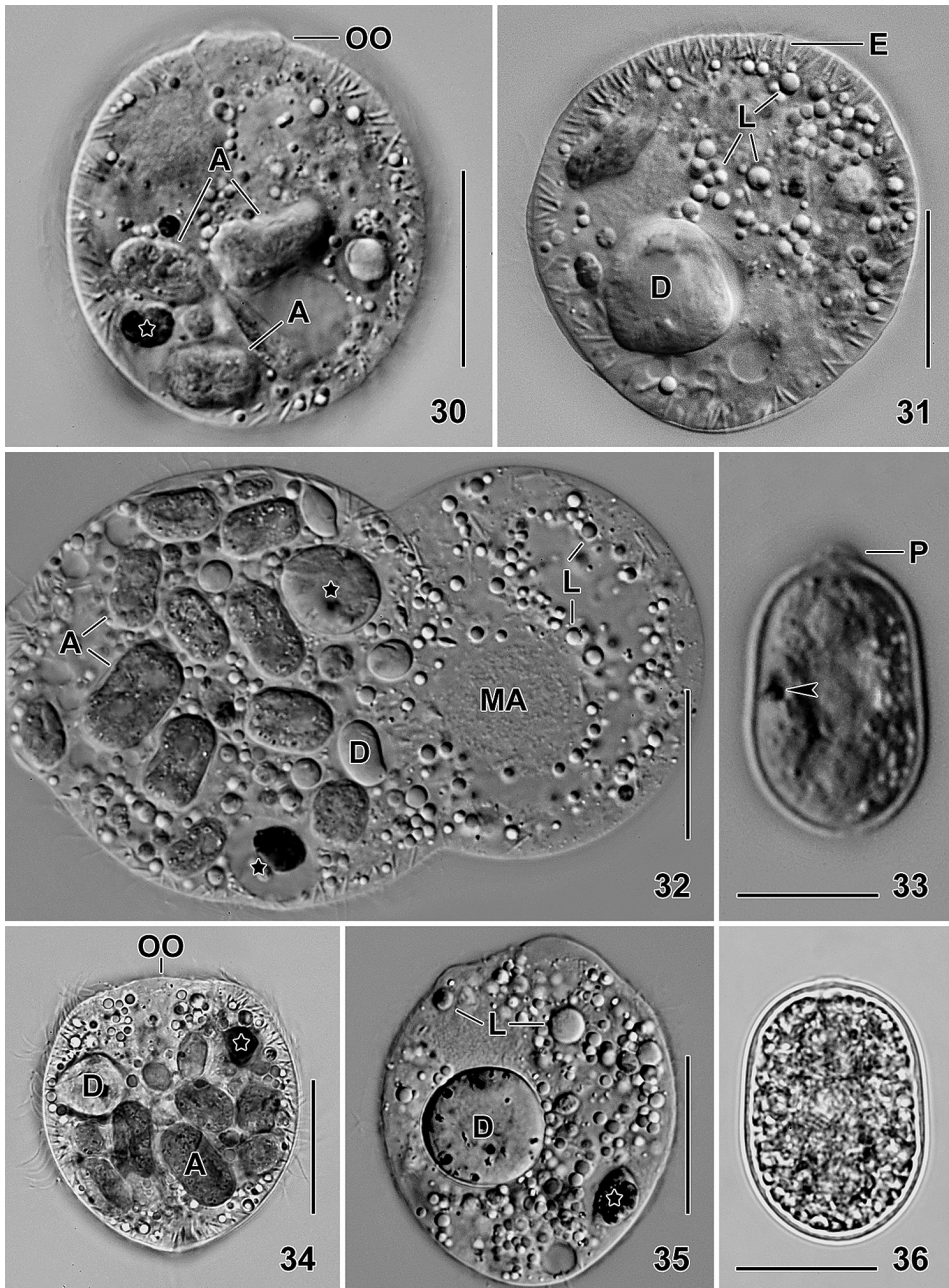


Fig. 30–36: *Urotricha spetai*, food and food digestion from life. When collected, *U. spetai* fed mainly on a 15–25 µm long *Chlamydomonas (steinii?)* with the eyespot in mid-body (33, arrow-head; 36). Many algae (A) are simultaneously digested, first becoming bright, orange globules (D) that are decomposed to lipid droplets (L), leaving deeply orange remnants (asterisks). A – algae, still recognizable, D – decomposing algae, E – extrusomes, L – lipid droplets, MA – macronucleus, OO – oral opening, P – papilla. Scale bars 10 µm (33, 36) and 25 µm (30–32, 34, 35).

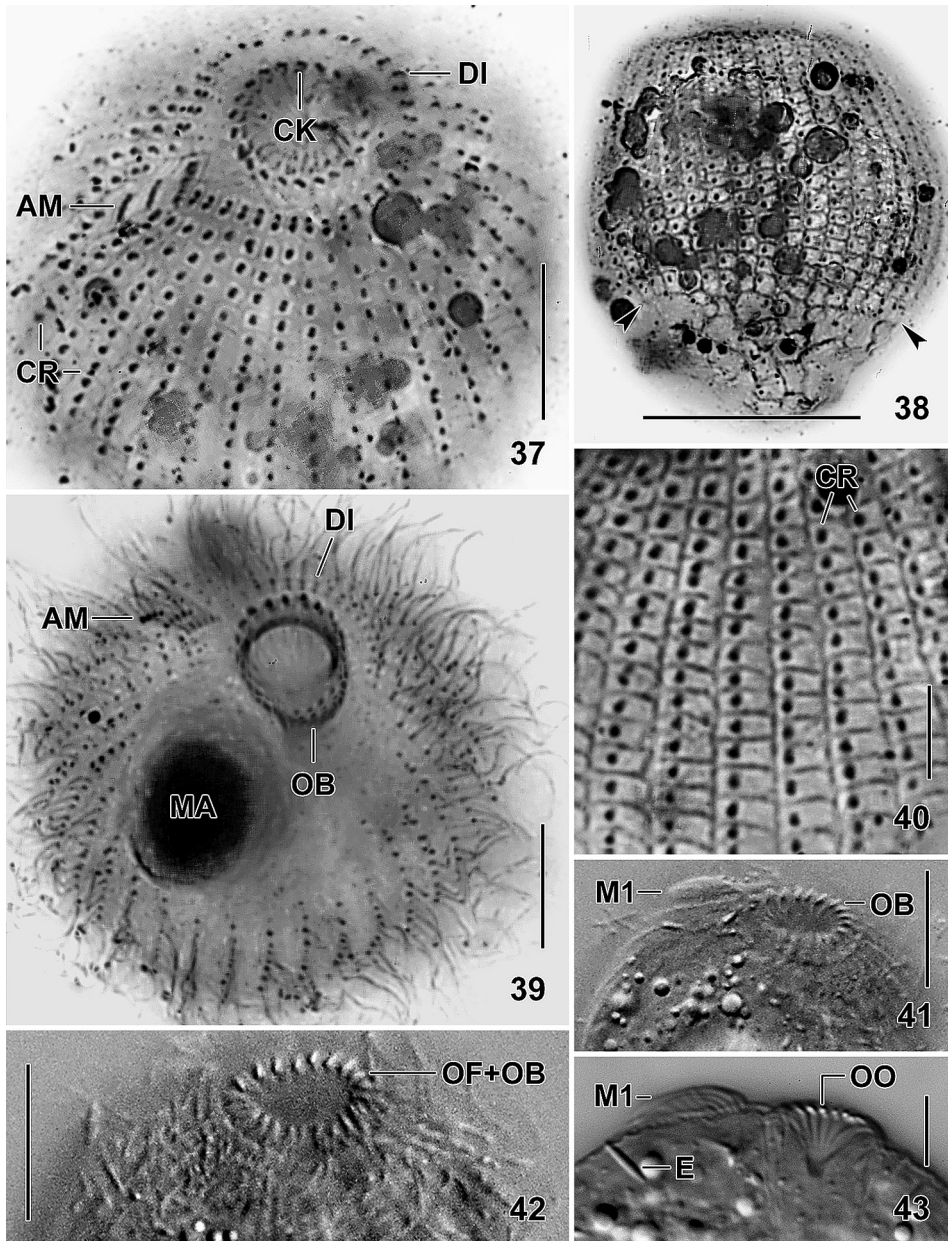


Fig. 37–43: *Urotricha spetai* from life (41–43) and after Chatton-Lwoff silver nitrate (37, 38, 40) and protargol (39) impregnation. 37, 39: Anterior polar views, showing the oral structures, especially the oral basket and the three minute adoral membranelles. 38, 40: Silverline and cortex pattern. Arrowheads mark the non-ciliated posterior portion, containing polygonal silverline meshes. 41–43: Oral structures, especially the oral basket and the rather long cilia of membranelle 1. AM – adoral membranelles, CK – circumoral kinety, CR – ciliary (basal body) rows, DI – dikinetid, E – extrusome, M1 – membranelle 1, MA – macronucleus, OB – oral basket, OF – oral flaps. Scale bars 5 μ m (40, 43), 10 μ m (37, 39, 41, 42), and 25 μ m (38).

Abb. 1–8, Seite 175: *Urotricha spetai* (1–4, 6–8) und *U. matthesi tristicha* (5, aus FOISSNER & PFISTER 1997) in vivo (1–5) und nach Protargol Imprägnation (6–8). 1: Links laterale Übersicht eines repräsentativen Exemplars, Länge 50 µm. 2: Rippen zwischen den adoralen Membranellen. 3. Extrusom, Länge 5 µm. 4: Cortex Muster. 5: Übersicht. 6–8: Variabilität von Körperform und Körpergröße. Der Pfeil in Abb. 6 markiert das Ende der Wimpernreihen. B – Basalkörper, CC – Caudalcilien, CV – kontraktile Vakuole, E – Extrusome, EP – Exkretionsporus, FV – Nahrungsvakuole, M – adorale Membranellen, MA – Makronucleus, MI – Mikronucleus, OB – Mundtrichter, OF – Mundklappen, R – Rippen. Maßstriche 20 µm (Fig. 5–8) und 25 µm (Fig. 1).

Abb. 9–13, Seite 176: *Urotricha alveolata* (9, aus FOISSNER & PFISTER 1997) und *U. spetai* (10–13) in vivo (9) und nach Silbernitrat Imprägnation. 9: Diese Art besitzt große corticale Alveolen, Länge 30–40 µm. 10: Details der somatischen Wimpernreihen. 11: Schräger Blick auf das orale und somatische Wimpernmuster des Holotyps. Der Pfeil weist auf die adoralen Membranellen. 12: Lateraler Überblick. Beachte die breit-obovate Körperform und das somatische Wimpernmuster. 13: Silberlinien und Basalkörper Muster im Bereich der adoralen Membranellen. B – Basalkörper, CK – circumorale Wimpernreihe, DK – Dikinetiden, M1 – adorale Membranelle 1, PS – parasomaler Sack, S – Silberlinien. Maßstriche 5 µm (Abb. 13), 15 µm (Abb. 11) und 25 µm (Abb. 12).

Abb. 14–23, Seite 177: *Urotricha spetai* in vivo (14–20) und nach Protargol Imprägnation (21–23). 14–20: Freischwimmende Individuen, die die charakteristische obovate bis breit-obovate Körperform zeigen. Die Zellen erscheinen gescheckt wegen der verschiedenen Nahrungseinschlüsse. Die Pfeilspitzen markieren den von den Extrusomen gebildeten Saum. Nur bei einem der sieben abgebildeten Individuen ist der unbewimperte hintere Körperteil zapfenartig abgesetzt (18, Pfeil). 21–23: Die breit-obovate Körperform ist in den Protargolpräparaten perfekt erhalten. Man erkennt, unter anderem, den Mundtrichter und die Basalkörper der vier Caudalcilien. CC – Caudalcilien, EP – Exkretionsporus, FV – Nahrungsvakuolen, MA – Makronucleus, OB – Mundtrichter, OF – Mundklappen. Maßstriche 25 µm.

Abb. 24–29, Seite 178: *Urotricha spetai*, lebende, leicht gequetschte Zellen. 24. *U. spetai* hat vier Caudalcilien und grüne oder tief orange gefärbte Nahrungsvakuolen. 25–28: Diese Photos, bei denen der Fokus auf der Zelloberfläche oder dem Zellzentrum liegt, zeigen wichtige Organellen. Die Extrusome bilden deutliche Körnchenreihen (Pfeile) und einen auffällenden Saum (gegenüber liegende Pfeilspitzen). 29: Adorale Membranelle 1 und einige stumpf-spindelförmige, 4–5 µm lange Extrusome. CC – Caudalcilien, CV – kontraktile Vakuole, E – Extrusome, FV – Nahrungsvakuolen, L – Fetttropfen, M1 – adorale Membranelle 1, MA – Makronucleus, OO – Mundöffnung. Maßstriche 5 µm (29) und 25 µm (24–28).

Abb. 30–36, Seite 179: *Urotricha spetai*, Nahrung und Verdauung in vivo. Die frisch gefangenen Zellen ernährten sich hauptsächlich von einer 15–25 µm langen *Chlamydomonas (steinii?)* mit einem Augenfleck in Körpermitte (33, Pfeilspitze; 36). Viele Algen (A) werden gleichzeitig verdaut, dabei werden sie zuerst glänzende, orange Kugeln (D), die dann zu Fetttropfen (L) abgebaut werden, von denen tief orange gefärbte, unverdauliche Reste übrig bleiben (Sternchen). A – noch erkennbare Algen, D – in Verdauung befindliche Algen, E – Extrusome, L – Fetttropfen, MA – Makronucleus, OO – Mundöffnung, P – Papille. Maßstriche 10 µm (33, 36) und 25 µm (30–32, 34, 35).

Abb. 37–43, Seite 180: *Urotricha spetai* in vivo (41–43) und nach CHATTON-LWOFF Silbernitrat (37, 38, 40) und Protargol Imprägnation (39). 37, 39: Vordere Polansichten, die die Mundstrukturen zeigen, besonders den Mundtrichter und die drei kleinen adoralen Membranellen. 38, 40: Silberlinien- und Cortextmuster. Die Pfeilspitzen weisen auf den unbewimperten hinteren Teil, in dem sich ein polygonales Silberliniennetz ausbreitet. 41–43: Mundstrukturen, nämlich der Mundtrichter und die ziemlich langen Wimpern der adoralen Membranelle 1. AM – adorale Membranellen, CK – circumorale Wimpernreihe, CR – Wimpern(Basalkörper)reihen, DI – Dikinetide, E – Extrusome, M1 – Membranelle 1, MA – Makronucleus, OB – Mundtrichter, OF – Mundklappen. Maßstriche 5 µm (40, 43), 10 µm (37, 39, 41, 42) und 25 µm (38).

silverline pattern (Fig. 4, 13, 38, 40). Cytoplasm colourless but most specimens dark at low magnification ($< 100\times$) due to many refractive food inclusions, making cells spotted under transmitted light at higher magnification. Possibly feeds mainly or exclusively on an about 20 μm long *Chlamydomonas* (*C. steinii*?) becoming inflated and more or less orange during digestion; sometimes full of colourless and orange lipid droplets (Fig. 1, 14–22, 24, 26, 27, 30–36). Swims rapidly but does not jump.

Somatic ciliary pattern as typical for genus. Cilia 8–10 μm long in vivo, arranged in equidistant, meridional rows, leaving blank posterior, obconical 22% of cell; number of cilia/row highly variable, on average 27; each row commences with a dikinetid, i.e., two narrowly spaced granules, of which only the posterior one is ciliated. On posterior pole a line of three to five, usually four caudal cilia 15–20 μm long and with fine, laterally curved distal end (Fig. 1, 6, 10–12, 22, 23, 37–40, Table 1).

Tab. 1: Morphometric data on *Urotricha spetai*. – Tab. 1: Morphometrische Daten von *Urotricha spetai*.

Characteristics ^a	Method	$\bar{x}$	M	SD	CV	Min	Max	n
Body, length	CHL	46.0	46.0	4.7	10.3	36.0	52.0	21
Body, width	CHL	42.0	42.0	4.4	10.6	31.0	50.0	21
Body length: width, ratio	CHL	1.1	1.1	0.1	5.1	1.0	1.2	21
Anterior body end to end of ciliary rows	CHL	35.8	37.0	4.8	13.5	22.0	41.0	21
Macronucleus, length	P	13.8	13.0	1.9	13.9	11.0	18.0	21
Macronucleus, width	P	9.6	10.0	1.3	13.5	8.0	12.0	21
Micronucleus, length	P	3.2	3.0	0.5	16.5	2.5	4.0	15
Micronucleus, width	P	2.4	2.5	0.4	15.3	2.0	3.0	15
Oral opening, diameter ^b	CHL	8.2	8.0	0.7	8.3	7.0	10.0	21
Blank area between begin of ciliary rows, $\emptyset$	CHL	13.1	13.0	1.3	10.1	10.0	16.0	21
Oral basket, length	P	29.2	30.0	4.8	16.3	18.0	37.0	21
Oral basket, width	P	6.4	6.0	–	–	6.0	7.0	21
Adoral organelle 1, length	CHL	2.9	3.0	–	–	2.5	3.5	16
Adoral organelle 1, number of kinetids	P	6.3	6.0	0.9	14.4	4.0	8.0	21
Adoral organelle 2, length	CHL	2.2	2.0	–	–	2.0	2.5	16
Adoral organelle 2, number of kinetids	P	4.9	5.0	0.7	13.5	3.0	6.0	21
Adoral organelle 3, length	CHL	1.8	2.0	–	–	1.5	2.5	16
Adoral organelle 3, number of kinetids	P	4.0	4.0	0.6	13.7	3.0	5.0	21
Somatic ciliary rows, number	CHL	41.8	41.0	1.9	4.6	40.0	46.0	19
Kinetids in a dorsal kinety, number ^c	CHL	27.1	26.0	6.1	22.6	15.0	42.0	21
Caudal cilia, number	P	4.0	4.0	–	–	3.0	5.0	21
Circumoral dikinetids, number	CHL	21.2	22.0	1.9	9.0	18.0	25.0	17

^a Data based on mounted, silver-impregnated, and randomly selected environmental specimens. Measurements in μm . CHL – Chatton-Lwoff silver nitrate impregnation, as described in FOISSNER (1991), CV – coefficient of variation in %, $\emptyset$ – diameter, M – median, Max – maximum, Min – minimum, n – number of specimens investigated, P – protargol impregnation, protocol A, as described in FOISSNER (1991), SD – standard deviation, $\bar{x}$ – arithmetic mean.

^b Measured between circumoral dikinetids and somatic dikinetids.

^c Dikinetid at begin of kineties counted as 1 kinetid.

Oral apparatus as typical for genus. Oral opening in centre of anterior pole, surrounded by 18–25 circumoral dikinetids associated with 2–3 μm long flaps. Oral funnel about 10 μm across in vivo, extends to near body end and slightly obliquely to dorsal side (Fig. 1, 6–8, 11, 12, 21, 22, 26, 27, 37, 39, 41–43, Table 1). Three minute, obliquely arranged adoral membranelles (“brosse”) inserted in anterior end of somatic ciliature, separated from each other by two about 3 μm high, hump-shaped ridges (Fig. 1, 2, 11, 13, 37, 39, Table 1). Membranelle 1 (M1) composed of an average of six stiff, about 5 μm long cilia; M2 composed of an average of five stiff, 2–3 μm long cilia; M3 as M2 but composed of only four cilia on average (Fig. 6, 11, 13, 26, 29, 41, 43 Table 1).

Occurrence and ecology: As yet found only at type locality. Not psychrophilic because surviving room temperature for two days. Although there were various planktonic algae in the pond, the food vacuoles contained only *Chlamydomonas*, indicating at least a preference for this kind of food.

Discussion

Urotricha spetai is a distinct species due to the very broadly obovate body shape and the four caudal cilia. Thus, only four congeners have some similarities: *U. castalia*, as redescribed by FOISSNER & PFISTER (1997); *U. matthesi* KRAINER, 1995; *U. matthesi tristicha* FOISSNER & PFISTER, 1997; and *U. alveolata* KAHL, 1926.

Urotricha castalia differs from *U. spetai* by body shape (ellipsoidal to very bluntly fusiform vs. very broadly obovate), the location of the excretory pore of the contractile vacuole (usually in circle formed by the caudal cilia vs. outside caudal cilia), the extrusomes (two size types, rod-shaped vs. one type, bluntly fusiform), and the number and arrangement of the caudal cirri (on average 7–9, forming a circle vs. 4 arranged in line).

Urotricha matthesi matthesi differs from *U. spetai* by body shape (broadly ellipsoidal vs. very broadly obovate), the shape of the extrusomes (rod-like vs. bluntly fusiform), the number and length of the ciliary rows (on average 32 and posteriorly shortened by $\leq 10\%$ vs. 42 and shortened by 22%), the number of circumoral dikinetids (on average ~ 15 vs. 21), and the number of adoral membranelles (5–6 vs. 3).

Urotricha matthesi tristicha (Fig. 5) differs from *U. spetai* (Fig. 1) by body shape (conical to broadly fusiform vs. very broadly obovate), the non-ciliated posterior body portion (plug-like set off from body proper vs. not set off), the size and shape of the extrusomes (2.5 μm long rods vs. 4–5 μm long and bluntly fusiform), and the number of ciliary rows (31 vs. 42 on average).

Urotricha alveolata (Fig. 9) possibly belongs to the genus *Pelagothrix* FOISSNER et al., 2002 and is insufficiently known. It is only $30 \times 20 \mu\text{m}$ in size (vs. $50 \times 45 \mu\text{m}$), has a distinct layer of cortical alveoli (vs. indistinct = as usual), and lacks distinct extrusomes (vs. distinct, forming a conspicuous fringe).

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Address:

Univ.-Prof. Dr. Wilhelm FOISSNER, Universität Salzburg, FB Organismische Biologie, Hellbrunnerstrasse 34, A–5020 Salzburg, Austria. E-Mail: wilhelm.foissner@sbg.ac.at.