THE SYSTEMATICS ASSOCIATION SPECIAL
VOLUME 79

Biogeography of Microscopic Organisms

Is Everything Small Everywhere?

EDITED BY

DIEGO FONTANETO

Imperial College London

CAMBRIDGE
UNIVERSITY PRESS

CAMBRIDGE UNIVERSITY PRESS
Cambridge, New York, Melbourne, Madrid, Cape Town,
Singapore, São Paulo, Delhi, Tokyo, Mexico City

Cambridge University Press
The Edinburgh Building, Cambridge CB2 8RU, UK

Published in the United States of America by Cambridge University Press, New York

www.cambridge.org
Information on this title: www.cambridge.org/9780521766708

First published 2011

Printed in the United Kingdom at the University Press, Cambridge

A catalogue record for this publication is available from the British Library

ISBN 978-0-521-76670-8 Hardback

5

Dispersal of protists: the role of cysts and human introductions

WILHELM FOISSNER

FB Organismische Biologie, Universität Salzburg, Salzburg, Austria

5.1 Introduction

While the distribution of flowering plants and larger animals is easy to determine, this is almost impossible in microorganisms, which are smaller than human beings by a factor of 1.8×10^6, assuming an average size of 100 μm and 180 cm, respectively. Thus, the subject has been searched with varied success and in heated debates (Foissner, 2004; Fenchel and Finlay, 2005), resulting in two hypotheses: the 'cosmopolitan model' (Finlay, 2002; Finlay et al., 2004) and the 'moderate endemicity model', which suggests that one-third of protists has restricted distribution (Foissner, 1999, 2006, 2008). The cosmopolitan model is based on ecological theory, while the moderate endemicity model emphasises flagship species which are so showy, or so novel, that it is unlikely that they would be overlooked if indeed they were widely distributed (Tyler, 1996). The debate has stimulated many investigations whose conclusions frequently read as follows (Bass et al., 2007): 'Our results strongly suggest that geographic dispersal in macroorganisms and microbes is not fundamentally different: some taxa show restricted and/or patchy distributions while others are clearly cosmopolitan.

Biogeography of Microscopic Organisms: Is Everything Small Everywhere?, ed. Diego Fontaneto. Published by Cambridge University Press.

These results are concordant with the 'moderate endemicity model' of microbial biogeography. Rare or continentally endemic microbes may be ecologically significant and potentially of conservational concern. We also demonstrate that strains with identical 18S but different ITS1 rDNA sequences can differ significantly in terms of morphological and important physiological characteristics, providing strong additional support for global protist biodiversity being significantly higher than previously thought.'

Thus, there is hardly any need to enlarge this subject again (for recent reviews, see Dolan, 2005; Martiny et al., 2006; Foissner, 2006, 2008; Caron, 2009). In contrast, little attention has been paid to the reasons why certain species are cosmopolitan and others are not, as evident from the reviews just cited. Wilkinson (2001) and Smith et al. (2008) suggested size and/or air currents as important dispersal factors. However, this has been abandoned by Foissner (2008). He emphasised that microfungi, mushrooms, mosses and ferns are not cosmopolitan although their dispersal means, the spores, are very abundant and usually less than 100 μm in size, corresponding to the trophic and cystic size of most protists (but see Chapters 8–12).

Thus, the reasons for cosmopolitan or restricted distribution must be different. In my opinion, the most important factors are the resting cysts, the geological history and human introductions. The overwhelming structural and chemical diversity of resting cysts becomes meaningful if one considers cysts not only as a simple dormant stage but as dispersal means. This has been widely neglected in the 'everything is everywhere' debate, and thus I shall devote half of this review to the demonstration of cyst diversity, hoping to revive cystology (Gutiérrez and Walker, 1983). The second important factor is the break of Pangaea into Laurasia and Gondwana about 120 million years ago. This has been discussed in several reviews (Foissner, 2006; Smith et al., 2008) and is thus excluded from the present one. The third main factor is human introductions, frequently underweighted by protist biogeographers (Foissner, 2006).

5.2 Dispersal of protists

I recognise four main routes: dispersal in active (non-encysted) state; dispersal by protective resting cysts; dispersal by humans; and dispersal by geological processes, especially the break of Pangaea and continental drift. As mentioned in section 5.1, the palaeobiogeographic route is not treated here.

5.2.1 Dispersal in active state

Usually, live protists are very fragile. Thus, it is reasonable that dispersal occurs mainly in the cystic state (see section 5.2.2). Nonetheless, dispersal in the active state is possibly also rather common, especially in marine environments, where

large water currents might disperse species over large areas or even globally. However, benthic and planktonic foraminifera have distinct areals (Darling and Wade, 2008; Pawlowski and Holzman, 2008), just as other marine protists, for instance, the coccolithophores (Winter et al., 1994).

On land, step-by-step dispersal might be of considerable significance, especially in euryoecious species and at local, regional and continental scales (Green and Bohannan, 2006). Many experiments show that new habitats are often colonised within a few weeks (for reviews, see Maguire, 1963, 1971). Unfortunately, species have been rarely identified, for instance, by Wanner and Dunger (2001) and Meisterfeld (1997), who studied testacean communities from reforested opencast mining sites. Colonisation was fast, but only euryoecious species developed, and most humus-specific species disappeared within a year at a site that was amended with humus from a primary forest to stimulate succession. This matches my (unpublished) observations on ciliates. Only nine euryoecious species colonised three small, artificial ponds within a year, in spite of excess food (for details, see legend to Fig 5.1), and few freshwater ciliate species survived when added to soil (Foissner, 1987, table 15).

Some of the most widespread ciliates, e.g. *Glaucoma scintillans*, *Colpidium colpoda* and species of the *Paramecium aurelia* and the *Tetrahymena pyriformis*-complex very likely lack the ability to produce resting cysts, although they have

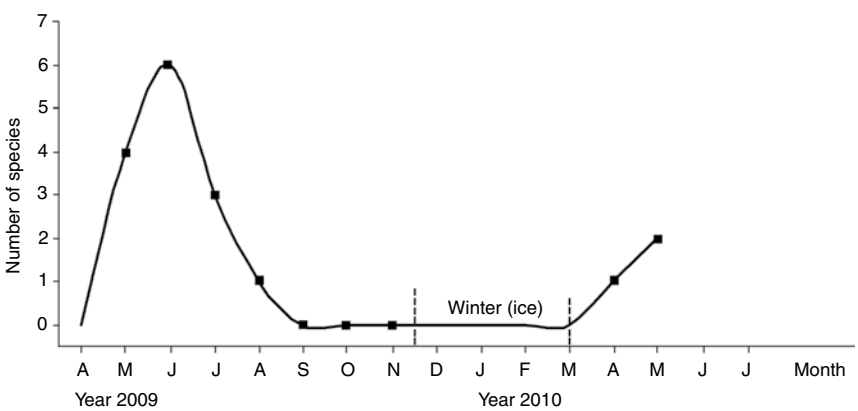


Fig 5.1 Number of ciliate species developing in three artificial ponds containing 1.5 l, 6 l and 12 l tap water and 0.1 g, 0.4 g and 0.8 g porridge oats. The experiment started on 1 April 2009 and is still running. A detailed description will be published later. Altogether, nine species were recognised: *Apocyclidium terricola*, *Chilodonella uncinata*, *Colpoda inflata*, *Epistylis opercularia*, *Odontochlamys alpestris*, *Pseudochilodonopsis algivora*, *Stylonychia pustulata*, *Tetrahymena rostrata* and *Vorticella infusionum*. With the exception of *E. opercularia*, which is possibly an 'air ciliate', all species are common, euryoecious inhabitants occurring in both limnetic and terrestrial habitats. Note the disappearance of the ciliates during a bloom of various algae and cyanobacteria.

been reported in both *Paramecium* (for a review, see Wichterman, 1986) and *Tetrahymena pyriformis* (Nilsson, 2005). However, the evidence is not convincing and not supported by my data. I never found any *Paramecium* in over 1000 air-dried and then rewetted soil samples from a great variety of habitats globally, including soil from flood plains and the surface of dry, ephemeral puddles (Foissner, 1998; Chao et al., 2006). Likewise, I did not find any *Paramecium* in about 200 samples from tank bromeliads of Central and South America, although it occurred in rivers and streams nearby (Foissner, unpublished). Further, my own experiments with *G. scintillans* and *Colpidium kleinii* failed. Thus, I join that group of scientists who believes that certain *Paramecium* and *Tetrahymena* species cannot make protective resting cysts.

Certainly, cystless species are a challenge to all dispersal models, including my cyst theory. While wide dispersal in the active state could be possible in encased species or in species with a thick cortex, as in *Paramecium*, this appears unlikely for fragile species like *Tetrahymena* and *Glaucoma*. I speculate that some of these species, especially those with a wide ecological range, may have distributed step by step or are older than the break of Pangaea. Further, we cannot exclude that such species were originally able to perform anabiosis (anhydrobiosis), i.e. to dry up without forming a special cyst, and becoming viable again when water becomes available. Although anhydrobiosis is extremely rare in present-day ciliates (I know it from only one *Podophrya*-like suctorian ciliate), it might have been more common in certain developmental stages of the species millions of years ago.

As protists are very small and thus of low weight, it is widely believed that air currents and animal vectors are the main distribution agents (Maguire, 1963; Cowling, 1994; Hamilton and Lenton, 1998; Wilkinson, 2001; Smith et al., 2008). Wilkinson (2001) showed by a detailed analysis of Arctic and Antarctic testacean communities that only large species (> 150 μm) are possibly not cosmopolitan.

All the data and hypotheses reviewed above, and many more not mentioned, are in conflict with a simple fact (Foissner, 2006, 2008; Fig 5.2; see also Chapters 9–12): mushrooms, mosses, ferns, lichens and horsetails have restricted distributions although their distribution means (spores) are produced in masses and in the size of most protists (≤ 100 μm). Further, hundreds of bacterial and fungal pests had regional or continental distribution before they were dispersed by humans. This is why I believe that, for example, air currents and the size of the organisms have little influence on their distribution. This has been supported by a study on microscopic fungi (Taylor et al., 2006). Actually, we do not know the amount of stable populations established by dispersal in the active state. Based on the data discussed above, step-by-step distribution of both, in active and cystic states, may play a significant role in at least the euryoecious species and if many similar habitats occur in a certain region.

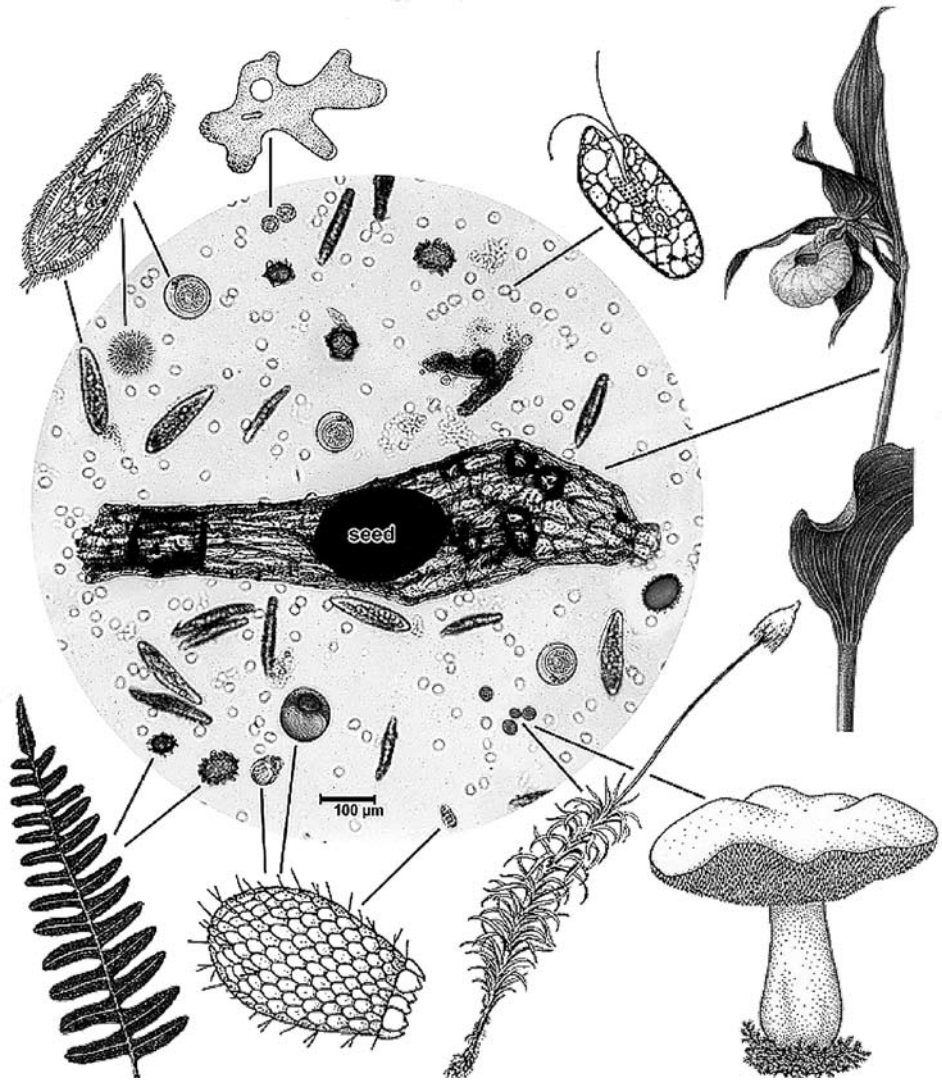


Fig 5.2 This figure compares, at about the same magnification, trophic and cystic protists (ciliates, flagellates, naked and testate amoebae) with spores of macrofungi (mushrooms), mosses, ferns and the minute seed of an orchid (*Vanda caerulescens*). Obviously, all are of minute size and very abundant, for instance, a single *Agaricus campestris* (mushroom) releases 1.6×10^{10} spores within 6 days (Webster, 1983), which exceeds the abundance of ciliates in 1 m^2 of forest soil by several orders of magnitude (Meyer et al., 1989). While nobody denies that mushrooms, mosses and ferns have biogeographies, protists are widely assumed to be cosmopolitan because their small size and high abundance favour air dispersal, an opinion flawed by this figure. Further, protist cysts lack adaptations for air dispersal, while seeds of many flowering plants have such adaptations, for instance, the orchid seed shown which has wings of large-sized, air-filled cells. Reproduced with permission from Foissner (2008).

5.2.2 Dispersal by resting cysts

Many protists can produce a dormant stage, named protective resting cyst, resting cyst (my preferred term), cyst, spore or stomatocyst, depending on the group under investigation. Resting cysts are widely assumed to be the major dispersal agents of unicellular organisms because they are much more stable than live cells (for reviews, see Corliss and Esser, 1974; Foissner, 1987; Gutiérrez and Martin-González, 2002). However, the biogeographic research and discussion ignored almost completely the very different morphological and physiological properties of resting cysts, depending on intrinsic (phylogenetic) and extrinsic (habitats s.l.) factors. Thus, I shall review here some recent studies, showing the overwhelming resting cyst diversity. For instance, the resting cyst of *Maryna umbrellata* is covered with glass granules, representing the first record of biomineralised silicon in ciliates (Foissner et al., 2009). It was just this diversity and some 'simple' observations reported below, which convinced me that cysts are possibly the most important factor for the dispersal of species (cosmopolitan or of restricted distribution) and for their presence/absence in a certain habitat, at a certain time, and under certain environmental conditions.

Unfortunately, our knowledge on the physiology, morphology and macromolecular composition of resting cysts is very limited. Thus, it is not yet possible to ascribe a certain function to the individual cyst layers. However, some general knowledge is available and has been reviewed by Corliss and Esser (1974), Foissner (1987, 2005, 2009), Gutiérrez and Martin-González (2002), Gutiérrez et al. (2003), and Foissner et al. (2005). Very briefly, a 'typical' cyst of a ciliate consists of a pericyst, an ectocyst, a mesocyst, an endocyst and, in certain taxa, a metacyst (Figs 5.11, 5.27). The chemical composition of these layers is known in only a few species (for an example, see Fig 5.11). Generally, acid mucopolysaccharides are frequent in the pericyst, while proteins, glycoproteins, glycogen and chitin are frequent in the mesocyst and endocyst. Unfortunately, the chemical composition of the ectocyst, which is often very thin, is unknown.

5.3 Resting cysts of ciliates from rain forests and hot deserts

Table 5.1 shows cyst survival of soil ciliates from rain forests in Borneo and Malaysia and from various habitats of Namibia, including the Namib Desert (Foissner et al., 2002). In the Namibian samples, there was no loss of species when the air-dried samples were stored for up to seven years, while most species of the rain forest soil could be not activated when the air-dried samples were older than a year, suggesting that the resting cysts died. Obviously, rain-forest ciliates have

Table 5.1 Ciliate species numbers in air-dried and rewetted[a] soil habitats of Namibia[b] and in rain forests of Borneo and Malay.

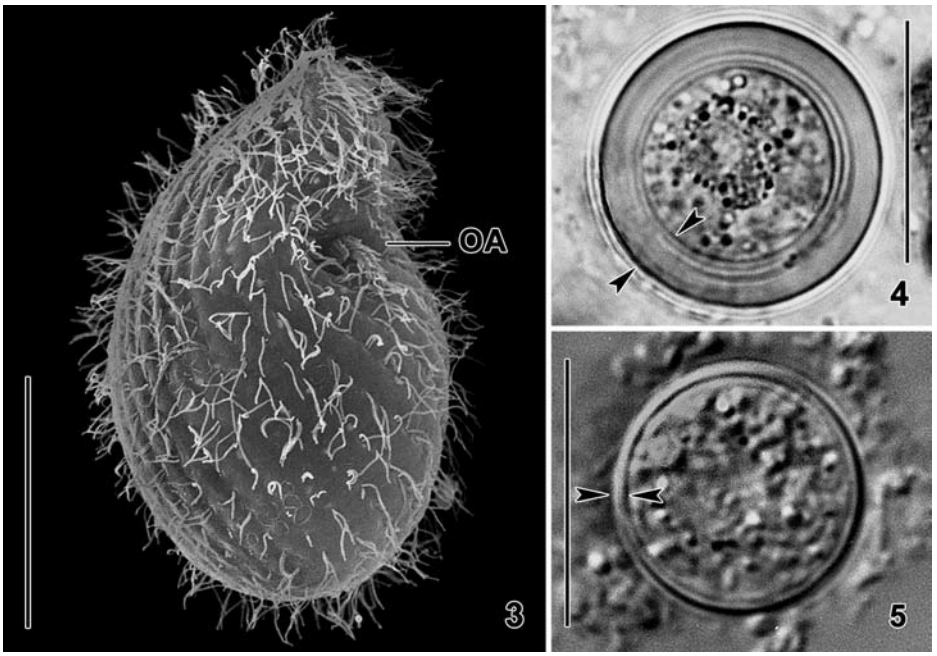
	Namibia		Rain forests	
Time elapsed since collection	**Number of species ($\bar{x}$)**	**Number of samples**	**Number of species ($\bar{x}$)**	**Number of samples**
≤ 10 h	None	Many	25.0	8
Up to 9 months	30.8	23	30.0	7
Up to 65 months	25.9	10	6.4	5
Up to 82 months	41.5	17	1.8	5

[a] Non-flooded Petri dish method as described in Foissner et al. (2002).

[b] Only 'typical' dry soil habitats were selected from the 73 samples investigated, viz., the samples: 1, 2, 4, 5, 7, 8, 9, 11, 12, 13, 16, 17, 18, 20, 23, 24, 26, 27, 29, 31, 32, 33, 35, 36, 37, 38, 39, 41, 42, 43, 44, 48, 49, 50, 53, 54, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 67, 69, 70, 73.

'weak' cysts not adapted to long periods of dryness. Accordingly, they have little chance to disperse via cysts over large areas. This contrasts with the 'strong' cysts from Namibia. The meaning of 'weak' and 'strong' is demonstrated by *Exocolpoda augustini* (Figs 5.3–5.5) collected in Austria and the dry west coast of Namibia (Foissner et al., 2002: site 37). While ordinary cysts with a rather thin (0.5–1 µm) wall are produced under the moderate Austrian climate (Fig 5.5), the wall of the Namibian specimens is about 7 µm thick (Fig 5.4), surpassing the volume of the encysted cell proper three times (3500 μm^3 vs. 14 000 μm^3).

These data match observations from laboratory cultures, where frequently most of the cysts made do not excyst when fresh medium and food are added, and cells sometimes lose the ability to make cysts at all, especially on prolonged cultivation. This makes sense under the constant laboratory conditions, where the populations select for non-encysters or switch off the encystment genes. However, the matter is complex, i.e. both encystment and excystment are influenced by many factors, as shown for example by Meier-Tackmann (1982) and Meier-Tackmann and Wenzel (1988) in a common soil ciliate, *Colpoda cucullus*, and by Müller et al. (2006) in *Meseres corlissi*, a plankton ciliate from ephemeral fresh waters (Fig 5.22). The often highly varying, 'mysterious' encystment and excystment rates give support to the 'scout theory' of Epstein (2009). This theory suggests that microbial populations consist of a mix of active and dormant cells. Faced with an adverse environmental change, more cells are included into dormancy, and survive the challenge. Individual cells would then periodically exit dormancy as a result of infrequent and essentially random events, such as a change in the expression of a master regulatory gene. I call such awakened cells 'scouts'. If the adverse conditions persist, the scout dies. If a scout forms under



Figs 5.3–5.5 *Exocolpoda augustini*, a small, terrestrial ciliate (**3**), makes extremely thick-walled resting cysts (**4**, opposed arrowheads) in the Namib desert, but makes ordinary-walled cysts in Austria (**5**, opposed arrowheads). OA – entrance to oral apparatus. Scale bars 25 µm. From Foissner et al. (2002) and original (**5**).

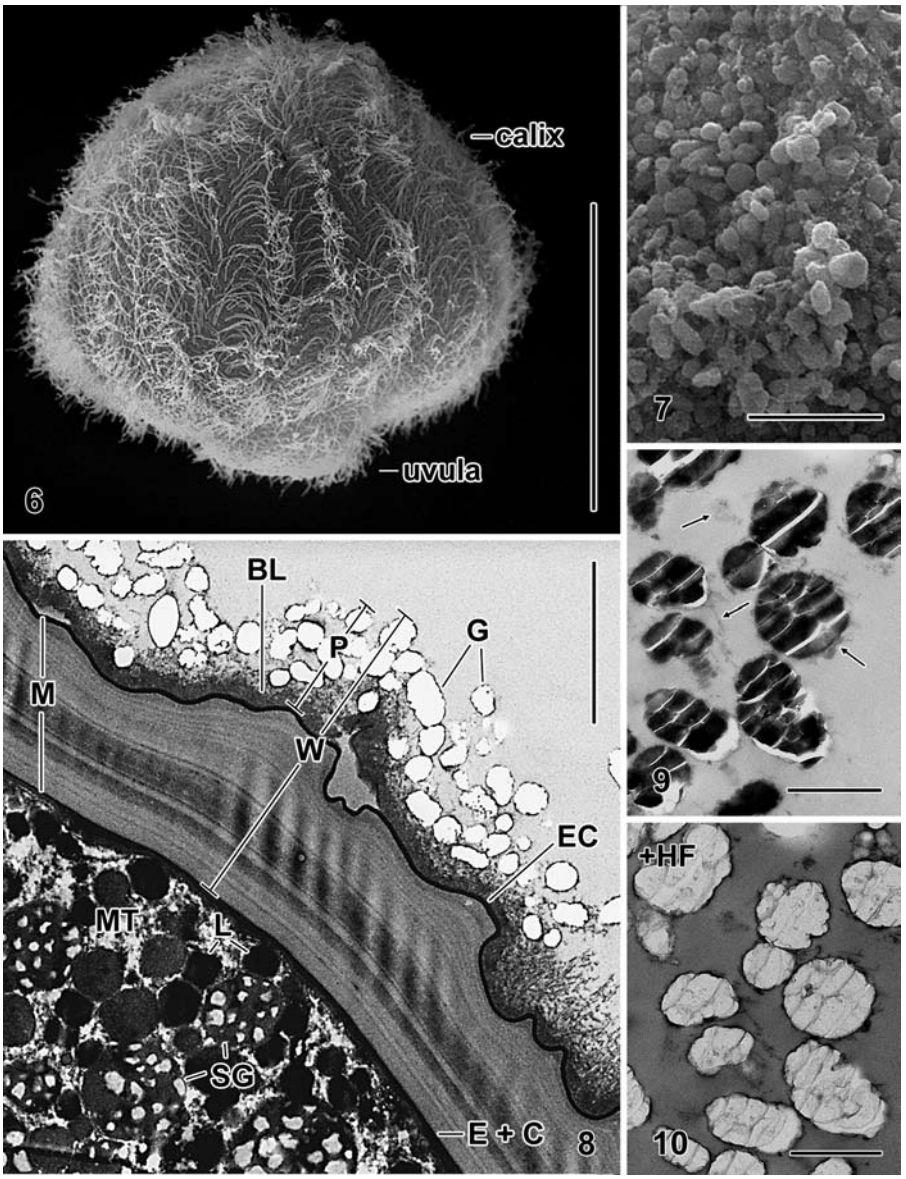
growth-permissive conditions, it starts a new population. In some species, scouts might even use growth-inducing signalling compounds to wake up the rest of the dormant population.

5.4 Some remarkable ciliate resting cysts

5.4.1 *Maryna umbrellata* (Colpodea)

This mushroom-shaped ciliate is about 100 µm in size, and is common in ephemeral limnetic habitats, such as rock pools and meadow puddles (Figs 5.6–5.10). The globular resting cyst is conspicuous because it is as large as the trophic cell. *Maryna umbrellata* is restricted to the northern hemisphere. In Africa, Australia, Central America and South America occurs a similar but smaller species having larger (up to 5 µm) silicon granules.

The fine structure and chemical composition of the resting cyst of *M. umbrellata* were studied by Foissner (2009) and Foissner et al. (2009). This showed the following peculiarities: (i) the cyst wall is about 13 µm thick and thus amounts for half of the total cyst volume (Fig 5.8); (ii) the external, about 4 µm-thick layer is made



Figs 5.6–5.10 *Maryna umbrellata*, a *c.* 100 µm-sized colpodid ciliate, typically living in ephemeral puddles, makes globular resting cysts covered with a layer of glass (silicon) granules (**7**–**10**). **6**: Overview of a trophic specimen in the SEM. **7**: The cyst surface is covered by about 1 µm-sized glass granules well recognisable in the SEM. **8**: The resting cyst has a *c.* 13 µm-thick wall composed of many layers, each having a specific macromolecular composition (see Fig 5.11). The glass granules (G) were solved by hydrofluoric acid. **9**, **10**: Glass layer before and after treatment with hydrofluoric acid. Arrows mark mucous material that holds together the silicon granules. BL – basal layer, EC – ectocyst, E + C – endocyst and ciliate cortex, G – glass granules, L – lipid droplets, M – mesocyst, MT – mitochondria, P – pericyst, SG – spongy globules, W – cyst wall. Scale bars 1 µm (**9**, **10**), 4 µm (**8**), 5 µm (**7**), and 50 µm (**6**). With permission from Foissner (2009) and Foissner et al. (2009).

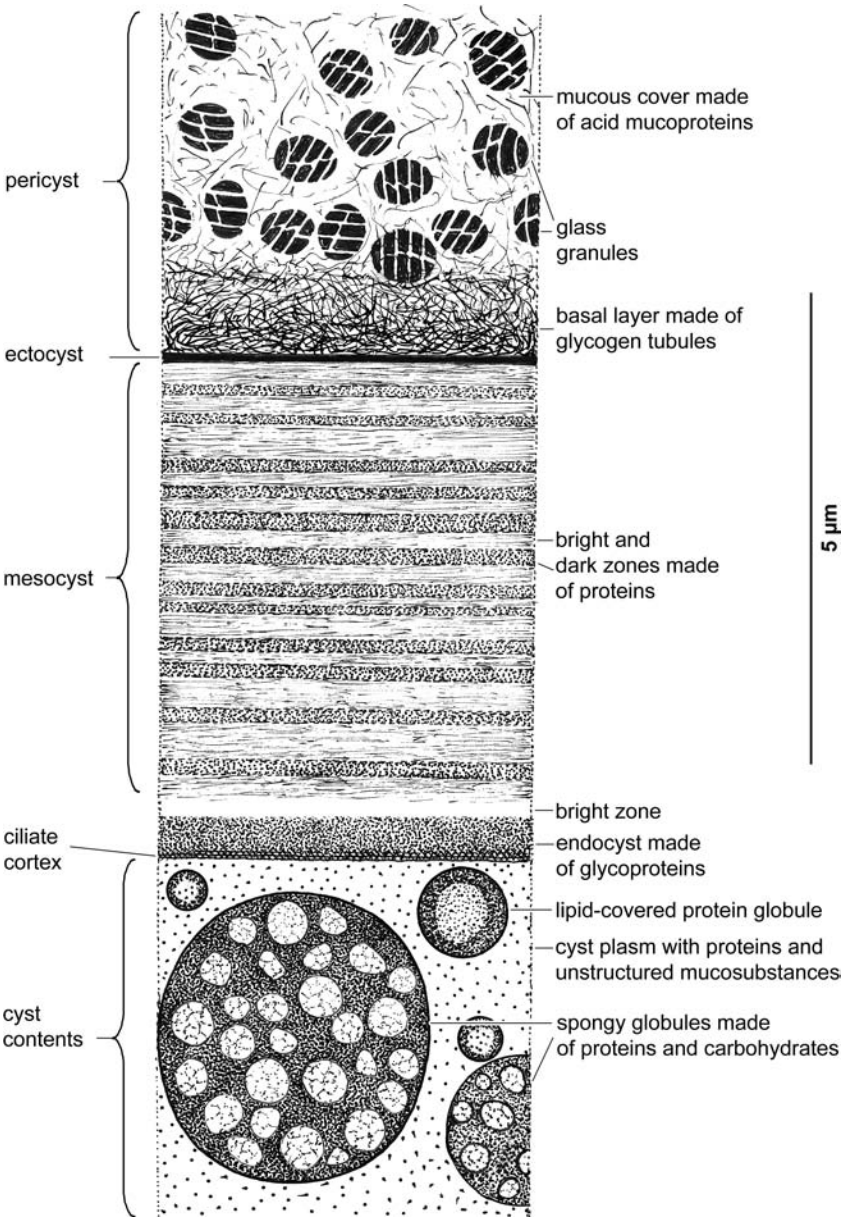


Fig 5.11 *Maryna umbrellata*, scheme of resting cyst based on light-and electron-microscopical observations and cytochemical tests (compare Figs 5.6–5.10). The complex cyst wall very likely determines the dispersal success, i.e. results in a cosmopolitan or restricted distribution. With permission from Foissner (2009).

of minute glass (silicon) granules with a size of about 1 μm (Figs 5.8–5.10); (iii) the mesocyst and endocyst show a high elasticity; (iv) the cytoplasm is studded with about 4 μm-sized globules consisting of a proteinaceous matrix burrowed by electron-lucent strands of glycogen (Figs 5.8, 5.11); (v) the fluid portion of the cyst plasm contains large amounts of acid mucopolysaccharides possibly originating from decomposed mucocysts; and (vi) the ectocyst precursors are released via the parasomal sacs of the kinetids. The most remarkable feature, the glass granules, is produced by the trophic cell and released during the early encystment processes.

Most, possibly all of these peculiarities are related to the ephemeral nature of the habitat, for instance, the thick wall protects the cell from desiccation, while the high elasticity and the glass cover might prevent the cell from mechanical stress (Yang et al., 2009), for instance, when cysts and sand are mixed by a storm. Certainly, all these properties will influence excystment and cyst viability, and thus the dispersal success.

5.4.2 *Pseudomaryna australiensis* and *Sandmanniella terricola* (Colpodea)

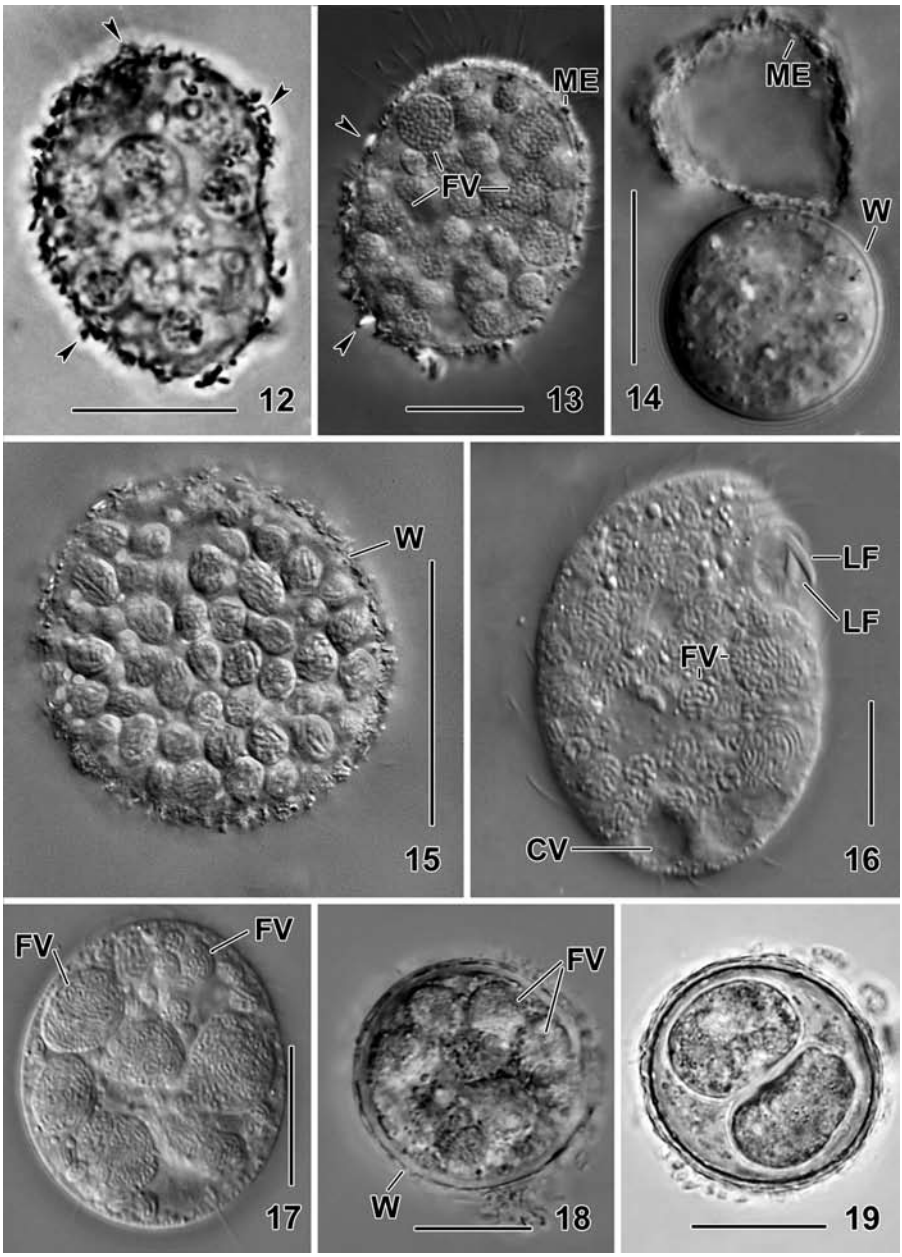
These small ciliates (~50 μm), which live in floodplain soils (Figs 5.12–5.19), have been described by Foissner (2003) and Foissner and Stoeck (2009). One of these, *P. australiensis* lives in a mineralic envelope, making cells and cysts appearing like inorganic soil particles, possibly protecting them from predators (Figs 5.12–5.14). Possibly, *P. australiensis* and *S. terricola* are restricted to the Australian and African region, respectively.

Before encysting, most ciliates digest food and expell the remnants, thus becoming rather hyaline when entering the cystic stage. *Pseudomaryna australiensis* and *S. terricola* do the opposite: they feed, but do not digest, in the trophic stage, becoming packed with large, compact food vacuoles (Figs 5.13, 5.15–5.17), which they digest in the resting cyst, using the energy provided for division (Figs 5.18, 5.19). Possibly, this is an extreme adaptation to the ephemeral nature of the habitat, making it possible to use even very short periods of optimal environmental conditions. The *P. australiensis* and *S. terricola* way must not be mixed with the division cysts of, for example *Colpoda*, which are covered by a temporary, very thin wall entirely different from that of the resting cysts (Foissner, 1993).

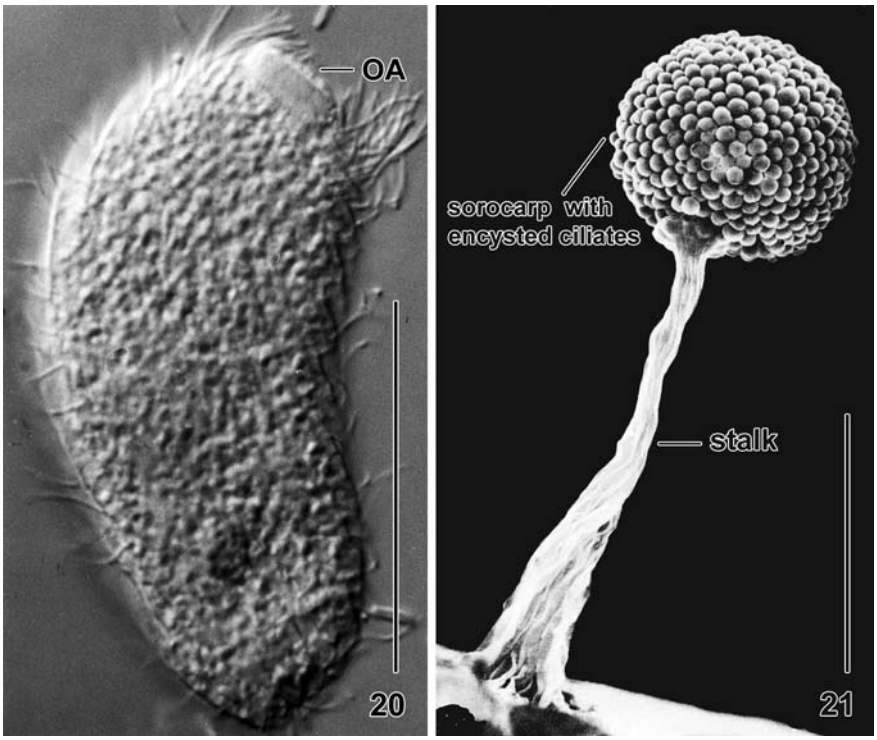
5.4.3 *Sorogena stoianovitchae* (Colpodea)

This curious ciliate lives on rotting foliage of plants (Figs 5.20, 5.21). It has a size of 30–70 × 20–45 μm and belongs to the class Colpodea, possibly representing a distinct order (Foissner, 1993; Foissner and Stoeck, 2009).

Sorogena stoianovitchae is the only ciliate that undergoes fruiting body development, and thus was initially thought to be related to the slime molds (Bradbury and Olive, 1980). The development process can be classified into five stages (Olive



Figs 5.12–5.19 *Pseudomaryna australiensis* (**12**–**15**) and *Sandmanniella terricola* (**16**–**19**) from life. Both species collect bacteria, forming large, compact food vacuoles which are not digested in the trophic (**12**, **13**, **16**, **17**) but in the cystic (**14**, **15**, **18**) stage, where they also divide (**19**). *P. australiensis* has a mineralic envelope (some particles marked by arrowheads), making it looking like a soil particle (**12**–**15**). CV – contractile vacuole, FV – food vacuoles, LF – left oral ciliary field, ME – mineralic envelope, W – cyst wall. Scale bars 20 µm. With permission from Foissner (2003) and Foissner and Stoeck (2009).



Figs 5.20, 5.21 *Sorogena stoianovitchae* from life (**20**) and in the scanning electron microscope (**21**). **20**: Right side overview of a trophont, showing the dome-shaped oral entrance (OA). **21**: Uniquely, *S. stoianovitchae* develops aerial sorocarps, quite similar to those of slime moulds. Scale bars 30 µm (**20**) and 200 µm (**21**). With permission from Bardele et al. (1991) and Olive and Blanton (1980).

and Blanton, 1980; Sugimoto and Endoh, 2008): aggregation, compact aggregation, secretion of a mucous matrix, stalk elongation and completion of the fruiting body. When *S. stoianovitchae* is mildly starved, several hundreds of cells aggregate beneath the water surface, and the aggregate develops into an aerial fruiting body, in which the individual cells encyst, forming a very thin wall (Blanton and Olive, 1983). Essential requirements for fruiting body development are high cell density, a light–dark cycle, and a dark period of more than 8 consecutive hours. In addition, the initial aggregation begins during the night and sunrise (light) triggers the subsequent development. The stalk of the sorocarp is composed of a matrix of complex protein-polysaccharide molecules (Blanton et al., 1983). Recently, Sugimoto and Endoh (2008) analysed the genes involved in fruiting body development. A BLASTX search revealed that sequences with high identity for extracellular proteins (mucin, proteophosphoglycan) or membrane proteins are likely candidates for aggregating material, mucous matrix and stalk material.

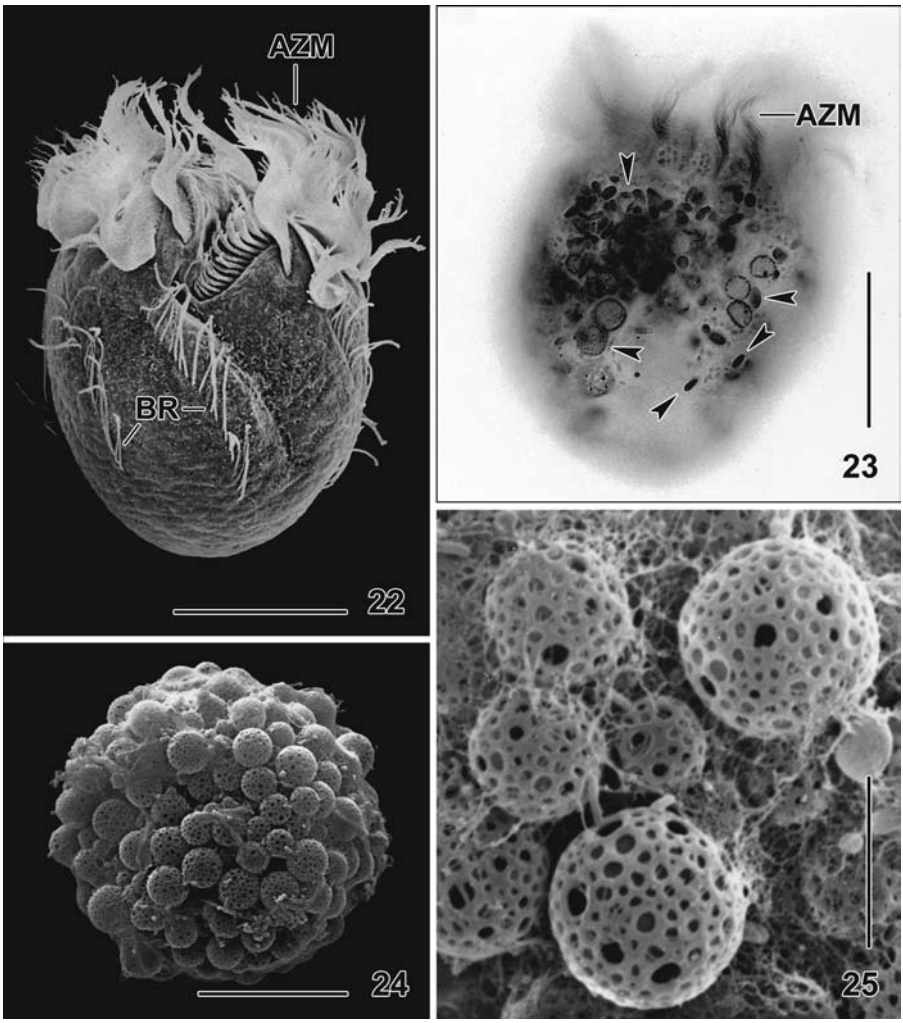
Table 5.2 State of 100 protargol-impregnated *Meseres corlissi* specimens from an exponentially growing culture.

State of specimens	Proportion (%)
Ordinary specimens	40
Ordinary dividing specimens	6
Dividing specimens with cyst wall precursors	2
Specimens with cyst wall precursors	23
Specimens with few food vacuoles	17
Specimens with few food vacuoles and with cyst wall precursors	12

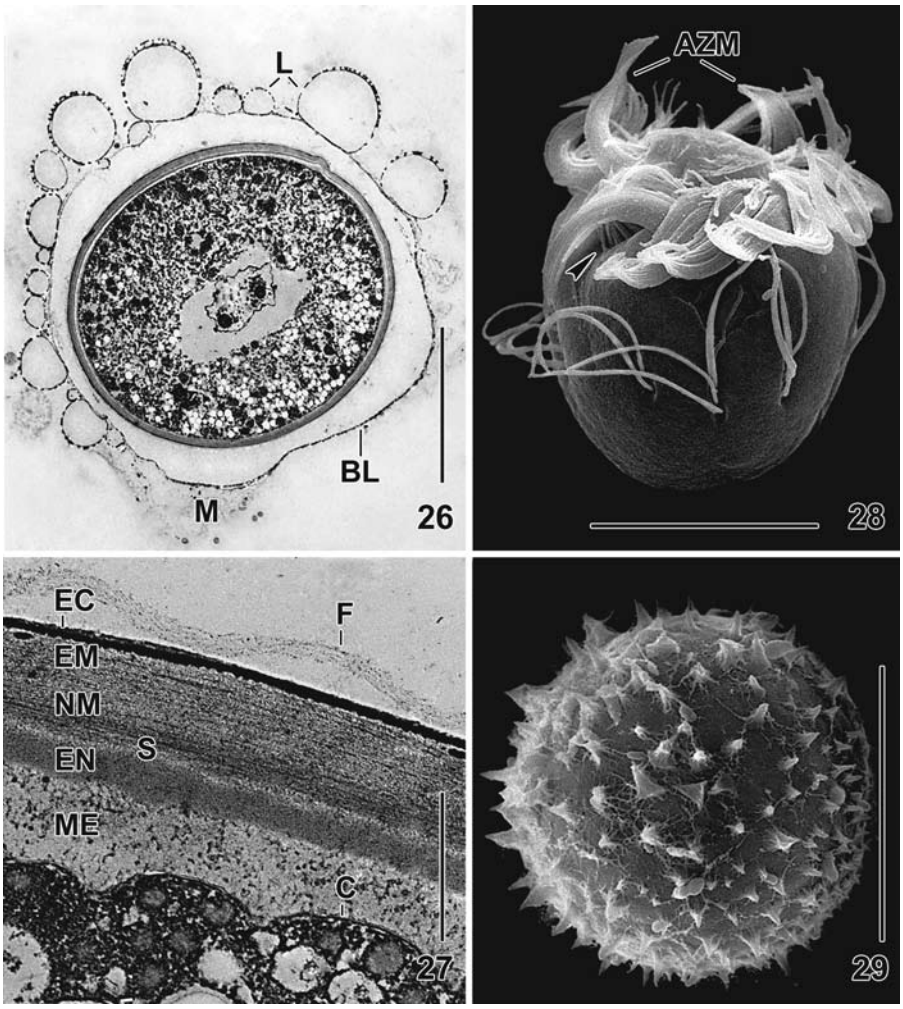
5.4.4 *Meseres corlissi* and *Halteria grandinella* (oligotrichs)

Meseres (Figs 5.22–27) and *Halteria* (Figs 5.28–5.29) are closely related morphologically (Petz and Foissner, 1992) and genetically (Katz et al., 2005). This is sustained by their resting cysts, especially the occurrence and fine structure of the lepidosomes (extracellular, organic structures produced intracellularly by trophic and/or cystic protist species; Foissner et al., 2005). Further, the cysts share a considerable overall similarity, that is, the wall is composed of five layers with similar fine structure (Foissner et al., 2007).

However, there are also conspicuous differences: (i) the lepidosomes are spherical in *Meseres* (Figs 5.24–5.26), while conical in *Halteria* (Fig 5.29); (ii) the lepidosomes of *Meseres* are located in a slimy 'basal layer' (Fig 5.26), while those of *Halteria*, which lacks a basal layer, are attached to the ectocyst; (iii) *Meseres* has a bright (non-osmiophilic) zone between mesocyst and endocyst (Fig 5.27), while both are close together in *Halteria* (Foissner et al., 2007); (iv) *Halteria* lacks the chitin present in *Meseres*, which is unexpected considering the close morphologic and genetic relationship; (v) *Meseres* has five complex types of cyst wall precursors (Foissner and Pichler, 2006), while *Halteria* has possibly only three or four because it lacks the basal layer and the bright zone between mesocyst and endocyst (see items ii and iii); (vi) The 'curious structures', very likely reserve bodies produced by the autophagous vacuoles, have a different shape (Foissner, 2005; Foissner et al., 2007); and (vii) in contrast to *Halteria*, *Meseres* produces part of the cyst wall precursors in the morphostatic condition and even in dividing specimens (Table 5.2, Fig 5.23). This ability, which I term 'precursor stocking', may explain why *Meseres* is able to encyst within one hour, in spite of the complexity of the process (Foissner and Pichler, 2006). Precursor stocking is possibly more common than recognised, i.e. I observed it also in some haptorid ciliates (Foissner, unpublished).



Figs 5.22–5.25 *Meseres corlissi* in the scanning electron microscope (**22**, **24**, **25**) and after silver (protargol) impregnation (**23**). **22**: Ventral overview, showing the conspicuous adoral zone of membranelles (AZM) and widely spaced somatic ciliary rows consisting of stiff bristles (BR). **23**: A morphostatic cell, as recognisable by the adoral zone of membranelles (AZM), which has numerous cyst wall precursors in the cytoplasm (arrowheads), including fully developed lepidosomes (see next figures). This phenomen is called 'precursor stocking'. **24**, **25**: The globular resting cyst is covered by about 200 spherical lepidosomes, i.e. organic scales produced by the Golgi apparatus and present also in 23% of non-encysting specimens (Table 5.2; Fig 5.23, precursor stocking). The lepidosomes have an average diameter of 6 µm and have a reticular wall (**25**). Scale bars 5 µm (**25**), 20 µm (**24**), and 30 µm (**22**, **23**). With permission from Petz and Foissner (1992), Foissner et al. (2005) and Foissner and Pichler (2006).



Figs 5.26–5.29 *Meseres corlissi* (**26**, **27**) and *Halteria grandinella* (**28**, **29**) in the transmission (**26**, **27**) and scanning (**28**, **29**) electron microscope. See also Figs 5.22–5.25. **26**, **27**: *Meseres corlissi* has a complex cyst wall, consisting of (from outside to inside) lepidosomes (L) embedded in a slimy matrix (M), a basal layer (BL), a microfibrillar layer (F), an ectocyst (EC), an ectomesocyst (EM), an endomesocyst (NM), an endocyst (EN) and a metacyst (ME). The cortex (C) of the ciliate is maintained. **28**: Left side view with end of adoral zone of membranelles (AZM) marked by an arrowhead. Note the long jumping bristles. **29**: Like *Meseres, Halteria* has lepidosomes on the surface of the resting cyst. However, the lepidosomes are globular in *Meseres*, while conical in *Halteria*. Scale bars 1 µm (**27**), 15 µm (**28**, **29**), and 20 µm (**26**). With permission from Foissner (2005) and Foissner et al. (2007).

The differences in the cyst structure of *Meseres* and *Halteria*, especially the complex lepidosomes and the presence of a chitin layer in the former, might at least partially explain their different ecology. Although both are cosmopolitan (Katz et al., 2005; Weisse et al., 2008), *Meseres* is very rare and possibly restricted to ephemeral freshwater habitats, while *Halteria* is one of the most common ciliates occurring in a wide variety of ephemeral and permanent limnetic environments (Foissner et al., 1991; Weisse et al., 2008).

As an inhabitant of ephemeral habitats, *Meseres* needs a 'stronger' cyst wall than *Halteria*. Indeed, the wall is twice as thick (1241 nm vs. 660 nm) and has a higher complexity (see above). While the chemical composition and the function of the lepidosomes, whose genesis and release takes a lot of energy, is still obscure, the chitin layer might be helpful in protecting the cell from mechanical and water stress as well as from bacterial decomposition because chitin is a very resistant matter. Finally, precursor stocking is an excellent way to use even short periods of good environmental conditions.

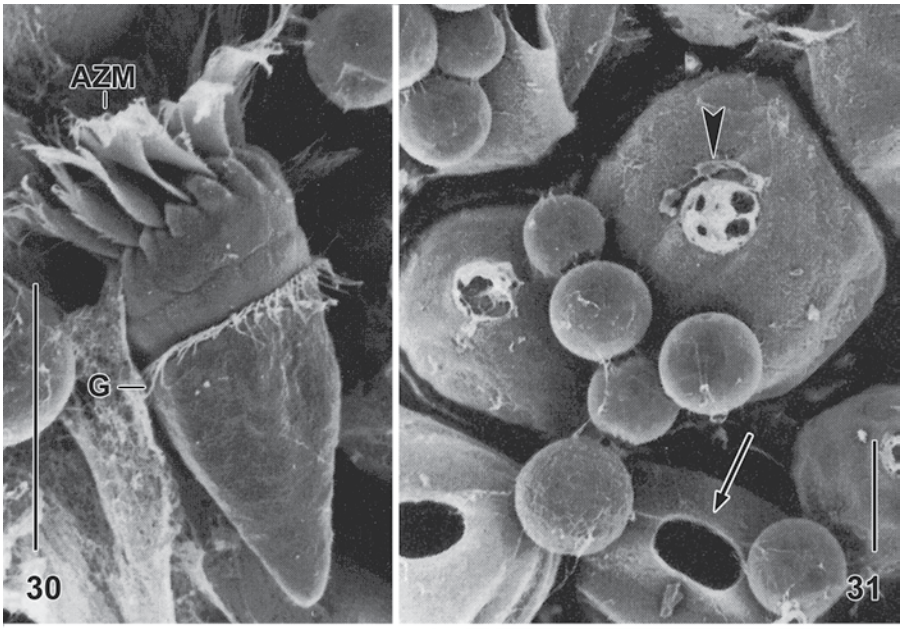
5.4.5 *Strombidium oculatum* (oligotrichs)

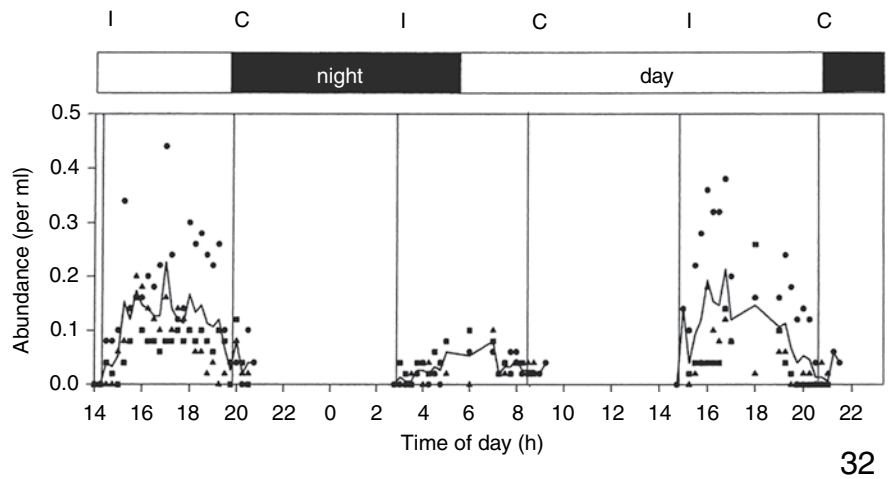
Strombidium oculatum is a tide-pool ciliate and an impressive example of circatidal encystment, first described by Fauré-Fremiet (1948) and later studied in detail by Jonsson (1994) and Montagnes et al. (2002). The ciliate, which has an obconical shape and is about 80 × 40 µm in size (Fig 5.30), is possibly restricted to the northern hemisphere (Agatha, S., pers. comm.). Usually, it is green due to sequestered chloroplasts and has a distinct, red eyespot composed of stigma obtained from chlorophyte prey. The cysts are flattened spheres, about 50 µm in diameter, and in the middle of the top surface there is a 10 µm-wide escape opening closed with a spumiform plug (Fig 5.31; Jonsson, 1994; Montagnes et al., 2002). Unfortunately, the fine structure and chemical composition of the cyst wall and the plug have not yet been investigated.

The circatidal behaviour runs as follows (Fig 5.32, Jonsson, 1994; Montagnes et al., 2002): for about 6 h, at low tide, *S. oculatum* is free-swimming in pools, and about 20–60 min before flushing of the pools it encysts on a substrate. Encystment lasts for about 19 h: two high tides and one intervening low tide. Excystment then occurs the next day about 30–40 min after the pools are isolated. Cells divide almost immediately after excysting, allowing the ciliate population to rapidly exploit potential food resources. Experiments and field observations revealed that *S. oculatum* responds phototaxically and exhibits seasonal trends in population dynamics with very low abundances in winter.

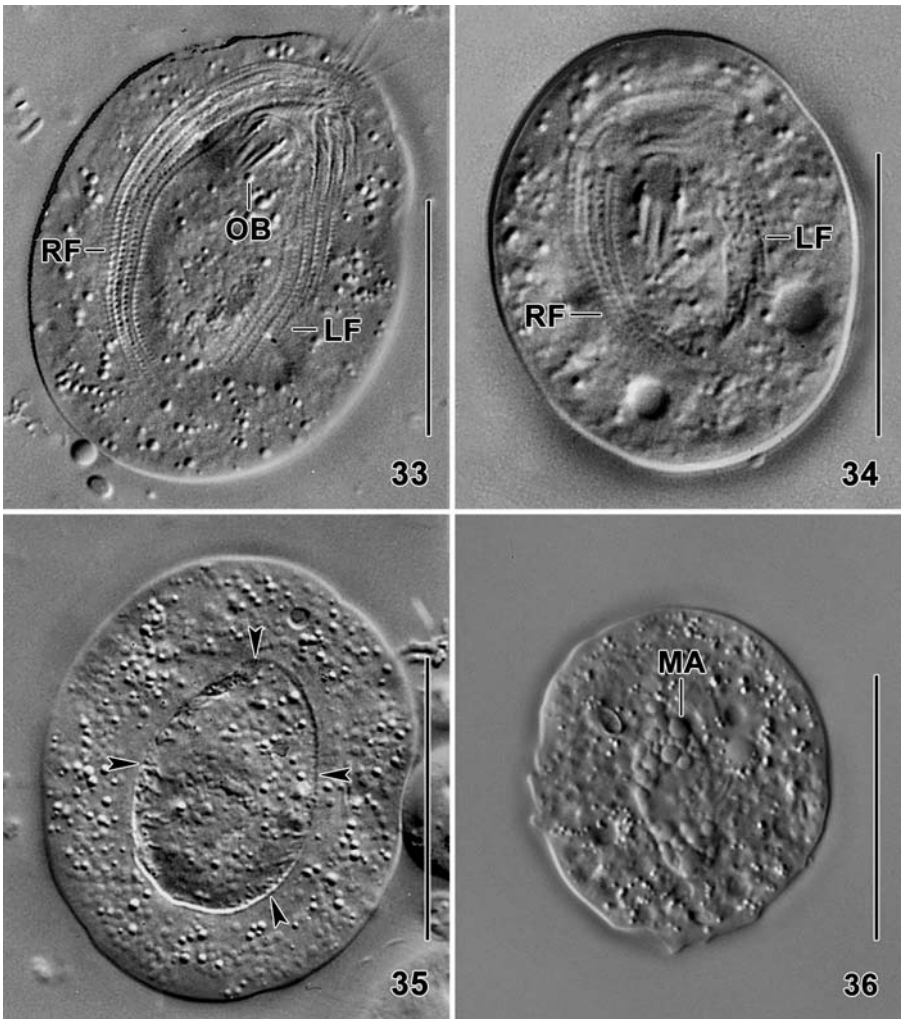
5.4.6 *Odontochlamys* spp. (Chilodonellidae)

These are small (~50 µm), bacterivorous ciliates living in terrestrial and limnetic habitats (Fig 5.33). They are remarkable in having the ability to change within a few





Figs 5.30–5.32 *Strombidium oculatum* in the scanning electron microscope. **30**: Lateral overview, showing the conspicuous adoral zone of membranelles (AZM) and the girdle ciliary row (G). **31**: Resting cysts are closed by a fibrous lid (arrowhead). When excysting, the lid disappears (arrow). **32**: Field observations of the change in abundance of *Strombidium oculatum* over the day–night cycle in three replicate tide pools (■, ●, ▲) over ~3 low and ~3 high tides. The solid line is the mean abundance of ciliates in the three pools. Vertical lines represent when pools were isolated by the outgoing tide (I) and covered by the incoming tide (C). Days and nights were delineated by sunrise and sunset. Scale bars 15 µm (**31**) and 25 µm (**30**). With permission from Jonsson (1994) and Montagnes et al. (2002).



Figs 5.33–5.36 *Odontochlamys* spp. can encyst within 10 min. **33**: Ventral view of a trophic specimen, showing the oral basket (OB) and the right and left ciliary field (LF, RF). **34**: When encystment commences, the cell rounds up and the dorsal side begins to vault over the ventral one. **35**: Middle stage, showing that the dorsal side (margin marked by arrowheads) vaulted over most of the ventral side. **36**: Young resting cyst without distinct wall, showing the macronucleus (MA). Scale bars 25 μm. With permission from Blatterer and Foissner (1992) (**33**–**35**) and original (**36**).

minutes from the active into the cystic state. Thus, encystment can be observed under the microscope (Figs 5.34–5.36). For details, see figure captions.

Obviously, fast encystment is a strategy very helpful in ephemeral habitats, such as moss, leaf litter and small ponds, where these ciliates usually occur. Looking at

the examples provided in this brief review, it becomes obvious that ciliates evolved several quite different strategies to survive in ephemeral habitats. It is likely that many more wait to be discovered.

5.5 Dispersal by humans

Biogeographic changes due to human activities have been largely ignored in the discussion of protist distribution, although a number of examples have been well known for a long time. For example, several tropical and Indopacific species of foraminifera entered the Mediterranean Sea via the Suez Canal (Lesseps' immigrants) and tropical aquaria. Moreover, it is likely that certain toxic dinoflagellates spread by human activities (Hallegraeff and Bolch, 1992). In rotifers, many of which have a similar size as ciliates, *Brachionus havanaensis* and *Keratella americana* have been introduced to southeast Asia by human activities (Segers, 2001). On the other hand, alpine zooplankton richness and genetic diversity have been only slightly influenced by anthropogenic stress and fish introduction, possibly due to the ability to produce long-lived resting stages withstanding unfavourable conditions (Winder et al., 2001).

Shipping (ballast water), the transport of goods and the construction of canals are three major reasons for the artificial dispersal of protists. Millions of tonnes of water and many thousands of tonnes of soil are transported across the world each year. Hallegraeff and Bolch (1992) and Hülsmann and Galil (2002) suppose that since the introduction of water as ballast in the middle of the nineteenth century, many protists may have spread globally, unheeded by protozoologists. The diatoms *Odontella sinensis* and *Coscinodiscus wailesii* entered the North Sea and the Baltic Sea rather recently, together with their parasites (Kühn, 1997; Hülsmann and Galil, 2002). Likewise, *Lagenophrys cochinensis*, an ectosymbiotic ciliate of wood-boring, marine isopods, has probably been transported from New Zealand to California in wooden ship hulls rather recently (Clamp, 2003), while the coccolithophore *Emiliania huxleyi* invaded the Black Sea about 1500 years ago (Winter et al., 1994). Elliott (1973) proposed that a species of the *Tetrahymena pyriformis* complex entered the Pacific Islands when man migrated westward from South and perhaps Central America. The same might have happened more recently with *Paramecium quadecaurelia*, a member of the *P. aurelia* sibling species complex. This species, which was known only from Australia, was recently reported from a pond of the city of Windhoek, the capital of Namibia (Przybós et al., 2003). Dispersal by ship's ballast water might also be responsible for the occurrence of four euryhaline psammobiontic (obligate sand-dwelling) testate amoeba species in the Great Lakes, Canada (Nicholls and MacIsaac, 2004), while marine dinoflagellates possibly cannot establish viable populations in these lakes (Fahnenstiel et al., 2009).

Another impressive example is the appearance of *Hydrodictyon* in New Zealand where this very distinctive alga had never been seen before. It was found in a pond belonging to a hatchery supplying fish and aquatic plants to aquarists. Obviously, *Hydrodictyon* had been imported together with fish or aquatic plants from East Asia (Kristiansen, 1996).

Freshwater diatoms show several impressive examples of human-mediated introductions. They have been reviewed by Vanormelingen et al. (2008), whose text I quote here: '*Asterionella formosa* is a widespread planktonic morphospecies and is frequently considered to be cosmopolitan; it is often seasonally dominant in eutrophic lakes. Detailed analysis of fossil material from 21 sediment cores (14 lakes) from New Zealand showed no trace of *A. formosa* in pre-European sediments, although it is now widespread, occurring in 45% of lakes for which phytoplankton records are available. The most likely vector for the introduction of *A. formosa* is the introduction of salmon ova into New Zealand lakes in the second half of the nineteenth century (Harper, 1994). It is highly unlikely that the species was extremely (i.e. not detectably) rare before European settlement as it is a species that can occur across a wide range of environmental conditions, from oligo- to eutrophic (Harper, 1994; Van Dam et al., 1994). Interestingly, *A. formosa* is also absent from other lake cores in Australasia, which might rule out environmental change due to the introduction of mammalian grazers as a cause for its sudden appearance. The recent spread in New Zealand of another exotic diatom, *Didymosphenia geminata* (Kilroy et al., 2007), is occurring long after the main human-induced environmental changes. Other convincing evidence for human mediated introduction of species (and hence previous dispersal limitation) among diatoms include the appearance of *Thalassiosira baltica* in the Laurentian Great Lakes (Edlund et al., 2000), and the North-American species *Gomphoneis minuta* and *Encyonema triangulum* in France (Coste and Ector, 2000).'

Wilkinson (2010) admonishes me for the lack of examples of human introductions in soil. There are none, unfortunately! And Wilkinson (2010) also did not provide any. However, he provided a solid discussion of soil introductions, some anecdotal evidence, and several suggestions to overcome the methodological problems which, indeed, are much more serious than in limnetic and marine environments.

5.6 Conclusions

Protist distribution is best described with the moderate endemicity model. But little information is available on the reasons why certain species are cosmopolitan and others are not. I argue that the break of Pangaea into Laurasia and Gondwana, the structure and physiology of the resting cysts, and human introductions are

Table 5.3 Percentages of dispersal routes of protists. Based on the calculation of Foissner (2008) that one-third of ciliates possibly have restricted distribution.

Dispersal routes	Amount (%)
Cosmopolitan distribution due to step-by-step dispersal and human introductions	35
Cosmopolitan distribution due to geological processes, euryoecious lifestyle and others	30
Restricted distribution due to morphological and physiological peculiarities of the resting cysts, break-up of Pangaea and insufficient time to disperse in young species	35

the most important factors for the dispersal of species (cosmopolitan or restricted distribution) and for their presence/absence in a certain habitat, at a certain time, and under certain environmental conditions. The same morphospecies may have different resting cysts, depending on the habitat in which the trophic cells live. Thus, long-range dispersal by air currents or animal vectors will not produce viable populations, for instance, ciliate species which evolved in rain forests will very likely not survive in Central Europe because their 'weak' cysts die during transport or do not withstand the different climate. This is substantiated by a comparative analysis of desiccation resistance of ciliate cysts from Namibia and rain forests in Borneo and the Malay Peninsula. Then, I present seven examples from ciliate resting cysts, showing their overwhelming morphological and physiological diversity, for instance, precursor stocking in *Meseres corlissi*, a glass cover in *Maryna umbrellata* and the circatidal cyst production of *Strombidium oculatum*. Step-by-step distribution and human introductions possibly also play a considerable role in the dispersal of species, especially at local, regional and continental scales. Several examples are provided. Table 5.3 shows rather speculative percentages on the contribution of several dispersal routes.

Acknowledgements

Financial support was provided by the Austrian Science Foundation (FWF, projects P20360-B17 and P19699-B17). The technical assistance of Mag. Barbara Harl is greatly acknowledged.

References

Bass, D., Richards, T.A., Matthai, L., Marsh, V., Cavalier-Smith, T. (2007). DNA evidence for global dispersal and probable endemicity of protozoa. *BMC Evolutionary Biology* **7**, 162.

Bardele, C.F., Foissner, W., Blanton, R.L. (1991). Morphology, morphogenesis and systematic position of the sorocarp forming ciliate *Sorogena stoianovitchae* Bradbury and Olive, 1980. *Journal of Protozoology* **38**, 7–17.

Blanton, R.L., Olive, L.S. (1983). Ultrastructure of aerial stalk formation by the ciliated protozoan *Sorogena stoianovitchae*. *Protoplasma* **116**, 125–135.

Blanton, R.L., Warner, S.A., Olive, L.S. (1983). The structure and composition of the stalk of the ciliated protozoan *Sorogena stoianovitchae*. *Journal of Protozoology* **30**, 617–624.

Blatterer, H., Foissner, W. (1992). Morphology and infraciliature of some cyrtophorid ciliates (Protozoa, Ciliophora) from freshwater and soil. *Archiv für Protistenkunde* **142**, 101–118.

Bradbury, P.C., Olive, L.S. (1980). Fine structure of the feeding stage of a sorogenic ciliate, *Sorogena stoianovitchae* gen. n., sp. n. *Journal of Protozoology* **27**, 267–277.

Caron, D.A. (2009). Past president's address: protistan biogeography: why all the fuss? *Journal of Eukaryotic Microbiology* **56**, 105–112.

Chao, A., Li, P.C., Agatha, S., Foissner, W. (2006). A statistical approach to estimate soil ciliate diversity and distribution based on data from five continents. *Oikos* **114**, 479–493.

Clamp, J.C. (2003). Ecology and geographic variation in *Lagenophrys cochinensis* (Ciliophora, Peritricha, Lagenophryidae), a widely distributed ectosymbiont of wood-boring, marine isopods. *Journal of Eukaryotic Microbiology* **50**, Abstract 82.

Corliss, J.O., Esser, S.C. (1974). Comments on the role of the cyst in the life cycle and survival of free-living protozoa. *Transactions of the American Microscopical Society* **93**, 578–593.

Coste, M., Ector, L. (2000). Diatomées invasives exotiques ou rares en France: principales observations effectuées au cours des dernières décennies. *Systematics and Geography of Plants* **70**, 373–400.

Cowling, A.J. (1994). Protozoan distribution and adaptation. In Darbyshire, J.F. (ed.), *Soil Protozoa, pp. 5–42*. Wallingford: CAB International.

Darling, K.F., Wade, C.M. (2008). The genetic diversity of planktic foraminifera and the global distribution of ribosomal RNA genotypes. *Marine Micropaleontology* **67**, 216–238.

Dolan, J.R. (2005). An introduction to the biogeography of aquatic microbes. *Aquatic Microbial Ecology* **41**, 39–48.

Edlund, M.B., Taylor, C.M., Schelske, C.L. et al. (2000). *Thalassiosira baltica* (Grunow) Ostenfeld (Bacillariophyta), a new exotic species in the Great Lakes. *Canadian Journal of Fisheries and Aquatic Sciences* **57**, 610–615.

Elliott, A.M. (1973). Life cycle and distribution of *Tetrahymena*. In Elliott, A.M. (ed.), *Biology of Tetrahymena, pp. 259–268*. Stroudsburg, PA: Hutchinson & Ross.

Epstein, S.S. (2009). Microbial awakenings. *Nature* **457**, 1083.

Fahnenstiel, G., Hong, Y., Millie, D., Doblin, M., Johengen, T., Reid, D. (2009). Marine dinoflagellate cysts in the ballast tank sediments of ships entering the Laurentian Great Lakes. *Verhandlungen der Internationalen Vereinigung für Limnologie* **30**, 1035–1038.

Fauré-Fremiet, E. (1948). Le rythme de marée du *Strombidium oculatum* Gruber. *Bulletin Biologique de la France et de la Belgique* **82**, 3–23.

Fenchel, T., Finlay, B.J. (2005). Cosmopolitanism and microbes. *SILnews* **44**, 5.

Finlay, B.J. (2002). Global dispersal of free-living microbial eukaryote species. *Science* **296**, 1061–1063.

Finlay, B.J., Esteban, G.F., Fenchel, T. (2004). Protist diversity is different? *Protist* **155**, 15–22.

Foissner, W. (1987). Soil protozoa: fundamental problems, ecological significance, adaptations in ciliates and testaceans, bioindicators, and guide to the literature. *Progress in Protistology* **2**, 69–212.

Foissner, W. (1993). Colpodea (Ciliophora). *Fischer, Stuttgart, Protozoenfauna* **4**, I–X + 798 pp.

Foissner, W. (1998). An updated compilation of world soil ciliates (Protozoa, Ciliophora), with ecological notes, new records, and descriptions of new species. *European Journal of Protistology* **34**, 195–235.

Foissner, W. (1999). Protist diversity: estimates of the near-imponderable. *Protist* **150**, 363–368.

Foissner, W. (2003). *Pseudomaryna australiensis* nov. gen., nov. spec. and *Colpoda brasiliensis* nov. spec., two new colpodids (Ciliophora, Colpodea) with a mineral envelope. *European Journal of Protistology* **39**, 199–212.

Foissner, W. (2004). Ubiquity and cosmopolitanism of protists questioned. *SILnews* **43**, 6–7.

Foissner, W. (2005). The unusual, lepidosome-coated resting cyst of *Meseres corlissi* (Ciliophora: Oligotrichea): transmission electron microscopy and phylogeny. *Acta Protozoologica* **44**, 217–230.

Foissner, W. (2006). Biogeography and dispersal of micro-organisms: a review emphasizing protists. *Acta Protozoologica* **45**, 111–136.

Foissner, W. (2008). Protist diversity and distribution: some basic considerations. *Biodiversity and Conservation* **17**, 235–242.

Foissner, W. (2009). The stunning, glass-covered resting cyst of *Maryna umbrellata* (Ciliophora, Colpodea). *Acta Protozoologica* **48**, 223–243.

Foissner, W., Pichler, M. (2006). The unusual, lepidosome-coated resting cyst of *Meseres corlissi* (Ciliophora: Oligotrichea): genesis of four complex types of wall precursors and assemblage of the cyst wall. *Acta Protozoologica* **45**, 339–366.

Foissner, W., Stoeck, T. (2009). Morphological and molecular characterization of a new protist family, Sandmanniellidae n. fam. (Ciliophora, Colpodea), with description of *Sandmanniella terricola* n. g., n. sp. from the Chobe floodplain in Botswana. *Journal of Eukaryotic Microbiology* **56**, 472–483.

Foissner, W., Blatterer, H., Berger, H., Kohmann, F. (1991). Taxonomische und ökologische Revision der Ciliaten des Saprobiensystems – Band I: Cyrtophorida, Oligotrichida, Hypotrichia, Colpodea. *Informationsberichte des Bayerischen Landesamtes für Wasserwirtschaft, München* **1/91**, 478 pp.

Foissner, W., Agatha, S., Berger, H. (2002). Soil ciliates (Protozoa, Ciliophora) from Namibia (Southwest Africa), with emphasis on two contrasting environments, the Etosha region and the Namib Desert. *Denisia* **5**, 1–1459.

Foissner, W., Müller H., Weisse, T. (2005). The unusual, lepidosome-coated resting cyst of *Meseres corlissi* (Ciliophora, Oligotrichea): light

and scanning electron microscopy, cytochemistry. *Acta Protozoologica* **44**, 201–215.

Foissner, W., Müller, H., Agatha, S. (2007). A comparative fine structural and phylogenetic analysis of resting cysts in oligotrich and hypotrich Spirotrichea (Ciliophora). *European Journal of Protistology* **43**, 295–314.

Foissner, W., Weissenbacher, B., Krautgartner, W.-D., Lütz-Meindl, U. (2009). A cover of glass: first report of biomineralized silicon in a ciliate, *Maryna umbrellata* (Ciliophora: Colpodea). *Journal of Eukaryotic Microbiology* **56**, 519–530.

Green, J., Bohannan, B.J.M. (2006). Spatial scaling of microbial biodiversity. *Trends in Ecology and Evolution* **21**, 501–507.

Gutiérrez, J.C., Walker, G.K. (1983). Cystology: a new area in protozoology. *Proceedings of the 5th European Conference on Ciliate Biology*, Geneva (unpaged abstract).

Gutiérrez, J.C., Martin-González, A. (2002). Ciliate encystment-excystment cycle: A response to environmental stress. In Gutiérrez, J.C. (ed.), *Microbial Development Under Environmental Stress, pp. 29–49*. Research Signpost 37/611 (2), Fort P.O., Trivandrum-695 023, Kerala, India.

Gutiérrez, J.C., Díaz, S., Ortega, R., Martín-González, A. (2003). Ciliate resting cyst walls: a comparative review. *Recent Research Developments in Microbiology* **7**, 361–379.

Hallegraeff, G., Bolch, C.J. (1992). Transport of diatom and dinoflagellate resting spores in ships' ballast water: implications for plankton biogeography and aquaculture. *Journal of Plankton Research* **14**, 1067–1084.

Hamilton, W.D., Lenton, T.M. (1998). Spora and gaia: how microbes fly with their clouds. *Ethology Ecology and Evolution* **10**, 1–16.

Harper, M.A. (1994). Did Europeans introduce *Asterionella formosa* Hassall to New Zealand? In Kociolek, J.P. (ed.), *Proceedings of the 11th International Diatom Symposium 1990, pp. 479–484*. San Francisco, CA: California Academy of Sciences.

Hülsmann, N., Galil, B.S. (2002). Protists – a dominant component of the ballast-transported biota. In Leppäkoski, E. et al. (eds.), *Invasive Aquatic Species of Europe, pp. 20–26*. Dordrecht: Kluwer Academic.

Jonsson, P.R. (1994). Tidal rhythm of cyst formation in the rock pool ciliate *Strombidium oculatum* Gruber (Ciliophora, Oligotrichida): a description of the functional biology and an analysis of the tidal synchronization of encystment. *Journal of Experimental Marine Biology and Ecology* **175**, 77–103.

Katz, L.A., McManus, G.B., Snoeyenbos-West, L.O. *et al*. (2005). Reframing the "everything is everywhere" debate: evidence for high gene flow and diversity in ciliate morphospecies. *Aquatic Microbial Ecology* **41**, 55–65.

Kilroy, C., Biggs, B.J., Vieglais, C.C. (2007). *Didymosphenia geminata* in New Zealand: a science response to help manage an unwanted, invasive freshwater diatom. *Abstract book, ASLO Aquatic Sciences meeting*. Santa Fe (New Mexico), 4–9/02/2007.

Kristiansen, J. (1996). Dispersal of freshwater algae – a review. *Hydrobiologia* **336**, 151–157.

Kühn, S.F. (1997). *Victoriniella multiformis*, gen. et spec. nov. (incerta sedis), a polymorphic parasitoid protist infecting the marine diatom

Coscinodiscus wailesii Gran and Angst (North Sea, German Bight). *Archiv für Protistenkunde* **148**, 115–123.

Maguire, B., Jr. (1963). The passive dispersal of small aquatic organisms and their colonization of isolated bodies of water. *Ecological Monographs* **33**, 161–185.

Maguire, B., Jr. (1971). Community structure of protozoans and algae with particular emphasis on recently colonized bodies of water. In Cairns, J., Jr. (ed.), *The Structure and Function of Fresh-water Microbial Communities,* pp. 121–149. Research Division Monograph 3, Virginia Polytechnic Institute and State University, Blacksburg, Virginia 24061.

Martiny, J.B.H., Bohannan, B.J.M., Brown, J.H. et al. (2006). Microbial biogeography: putting microorganisms on the map. *Nature* **4**, 102–112.

Meier-Tackmann, D. (1982). Untersuchungen über die physiologische Funktion der Cystenhülle und die Resistenz der dünnwandigen Dauercysten von *Colpoda cucullus* O.F. Müller (Holotricha, Ciliata). *Zoologischer Anzeiger* **208**, 1–29.

Meier-Tackmann, D., Wenzel, F. (1988). Daten über Wettergeschehen und Reaktivierbarkeit der Dauercysten von *Colpoda cucullus* O.F. Müller (Holotricha, Ciliata). *Zoologischer Anzeiger* **220**, 277–290.

Meisterfeld, R. (1997). Thekamöben – ihr Potential für Ökosystemforschung und Bioindikation. *Abhandlungen und Berichte der Gesellschaft für Naturkunde, Görlitz* **69**, 87–95.

Meyer, E., Foissner, W., Aescht, E. (1989). Vielfalt und Leistung der Tiere im Waldboden. *Österreichische Forstzeitung* **3**, 15–18.

Montagnes, D.J.S., Wilson, D., Brooks, S.J., Lowe, C., Campey, M. (2002). Cyclical behaviour of the tide-pool ciliate *Stombidium oculatum*. *Aquatic Microbial Ecology* **28**, 55–68.

Müller, H., Foissner, W., Weisse, T. (2006). Role of soil in the life cycle of *Meseres corlissi* (Ciliophora: Oligotrichea): experiments with two clonal strains from the type locality, an astatic meadow pond. *Aquatic Microbial Ecology* **42**, 199–208.

Nicholls, K.H., MacIsaac, H.J. (2004). Euryhaline, sand-dwelling testate rhizopods in the Great Lakes. *Journal of Great Lakes Research* **30**, 123–132.

Nilsson, J.R. (2005). Ethanol affects endocytosis and proliferation of *Tetrahymena pyriformis* GL and promotes encystment. *Acta Protozoologica* **44**, 293–299.

Olive, L.S., Blanton, R.L. (1980). Aerial sorocarp development by the aggregative ciliate, *Sorogena stoianovitchae*. *Journal of Protozoology* **27**, 293–299.

Pawlowski, J., Holzman, M. (2008). Diversity and geographic distribution of benthic foraminifera: a molecular perspective. *Biodiversity and Conservation* **17**, 317–328.

Petz, W., Foissner, W. (1992). Morphology and morphogenesis of *Strobilidium caudatum* (Fromentel) *Meseres corlissi* n. sp., *Halteria grandinella* (Müller), and *Strombidium rehwaldi* n. sp., and a proposed phylogenetic system for oligotrich ciliates (Protozoa, Ciliophora). *Journal of Protozoology* **39**, 159–176.

Przybós, E., Hori, M., Fokin, S.I. (2003). Strains of *Paramecium quadecaurelia* from Namibia, Africa; genetic and molecular studies. *Acta Protozoologica* **42**, 357–360.

Segers, H. (2001). Zoogeography of the Southeast Asian Rotifera. *Hydrobiologia* **446/447**, 233–246.

Smith, H.G., Bobrov, A., Lara, E. (2008). Diversity and biogeography of testate amoebae. *Biodiversity and Conservation* **17**, 329–343.

Sugimoto, H., Endoh, H. (2008). Differentially expressed genes during fruiting body development in the aggregative ciliate *Sorogena stoianovitchae* (Ciliophora: Colpodea). *Journal of Eukaryotic Microbiology* **55**, 110–116.

Taylor, J.W., Turner, E., Townsend, J.P., Dettman, J.R., Jacobson, D. (2006). Eukaryotic microbes, species recognition and the geographic limits of species: examples from the kingdom Fungi. *Philosophical Transactions of the Royal Society B* **361**, 1947–1963.

Tyler, P.A. (1996). Endemism in freshwater algae with special reference to the Australian region. *Hydrobiologia* **336**, 1–9.

Van Dam, H., Mertens, A., Sinkeldam, J. (1994). A coded checklist and ecological indicator values of freshwater diatoms from The Netherlands. *Netherlands Journal of Aquatic Ecology* **28**, 117–133.

Vanormelingen, P., Verleyen, E., Vyverman, W. (2008). The diversity and distribution of diatoms: from cosmopolitanism to narrow endemism. *Biodiversity and Conservation* **17**, 393–405.

Wanner, M., Dunger, W. (2001). Biological activity of soils from reclaimed open-cast coal mining areas in Upper Lusatia using testate amoebae (protists) as indicators. *Ecological Engineering* **17**, 323–330.

Weisse, T., Strüder-Kypke, C., Berger, H., Foissner, W. (2008). Genetic, morphological, and ecological diversity of spatially separated clones of *Meseres corlissi* Petz & Foissner, 1992 (Ciliophora, Spirotrichea). *Journal of Eukaryotic Microbiology* **55**, 257–270.

Webster, J. (1983). *Pilze. Eine Einführung*. Berlin: Springer.

Wichterman, R. (1986). *The Biology of Paramecium*, 2nd Edition. New York: Plenum Press.

Wilkinson, D.M. (2001). What is the upper size limit for cosmopolitan distribution in free-living microorganisms? *Journal of Biogeography* **28**, 285–291.

Wilkinson, D.M. (2010). Have we underestimated the importance of humans in the biogeography of free-living terrestrial microorganisms? *Journal of Biogeography* **37**, 393–397.

Winder, M., Monaghan, M.T., Spaak, P. (2001). Have human impacts changed alpine zooplankton diversity over the past 100 years? *Arctic, Antarctic, and Alpine Research* **33**, 467–475.

Winter, A., Jordan, R.W., Roth, P.H. (1994). Biogeography of living coccolithophores in ocean waters. In Winter, A., Siesser, W.G. (eds.), *Coccolithophores*, pp. 161–177. Cambridge: Cambridge University Press.

Yang, S.H., Lee, K.-B., Kong, B., Kim, J.-H., Kim, H.-S., Choi, I.S. (2009). Biomimetic encapsulation of individual cells with silica. *Angewandte Chemie* **121**, 9324–9327.