### 'Sewage fungus'

A field and microscopic guide

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#### Introduction

- Mainly heterotrophic micro-organisms
- Sensitive indicators of organic pollution
- Matrix of filamentous bacteria, fungi and/or stalked protozoa
- Can be present either as an almost pure monoculture of one species or as a mixed growth of several species
- Growth form varies due to species type and severity of organic pollution
- Species present can indicate type of organic pollution
- Colour varies widely from white to brown or pink
- Can be used to identify organic pollution sources several kilometres away
- Assessment below substrate particularly useful as lack of competition with autotrophic organisms
- Microscopic examination required to confirm identification

#### References

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Curtis EJC (1969) Sewage fungus: its nature and effects, Water Research, Vol. 3, 289-311.

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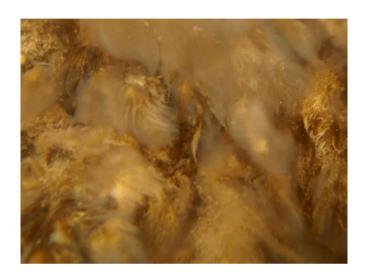
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Tomlinson TG and Williams IL (1975) Fungi. In Ecological Aspects of Used Water Treatment. I. The Organisms and their Ecology, pp 93-152.

Van Veen WL, Mulder EG and Deinema MH (1978) The *Sphaerotilus-Leptothrix* Group of Bacteria, Microbiological Reviews, 329-356.

#### Main sewage fungus taxa and growth forms

The taxa below are the main sewage fungus organisms found in freshwaters and typical growth forms. One or more taxa can be present in sewage fungus outbreaks and microscopic examination is required to confirm identification. Further information on growth forms, environmental conditions and key identification features can be found on pages 12 to 25.



**Sphaerotilus natans**Filamentous bacterium
Slimy fronds



**Zoogloeal bacterium**Ill defined taxonomically
Jelly-like gelatinous mass



**Beggiatoa alba**Filamentous bacterium
Thin white film

#### Main sewage fungus taxa and growth forms



Fusarium aquaeductuum
Filamentous fungus
Imparts pink or red colouration



**Geotrichum candidum**Filamentous fungus
Soft texture loosely following contours of stones



**Leptomitus lacteus**Filamentous fungus
Overlapping cotton wool-like
streamers



Carchesium polypinum Stalked protozoan Short 2-3mm tufts

#### Assessment methodology

Record cover and density above and below substrate as follows:

#### Cover

None

Local - <30%

Widespread - 30 – 60%

Extensive - >60%

#### Density

Trace - Present but only just detectable

Thin - Obvious presence but substrate not obscured

Thick - Thick enough to fully obscure substrate

Massive - Occupies a significant proportion of the water column

#### Recording

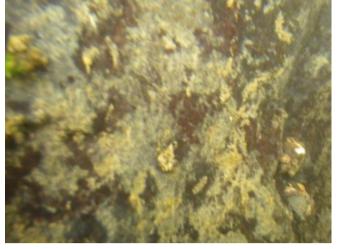
It is also useful to record associated parameters on the recording sheet shown on page 10. All parameters follow BIOSYS methods. Take photographs to show cover, density and growth form. The standard issue Pentax Optio WG series camera is particularly useful, as it can take close up underwater photographs. Photographs can also be used as exhibits in witness statements.

#### Samples for analysis

Take representative samples of sewage fungus in a small amount of water. A 30ml plastic polypropylene vial (shown right) with a conical base is ideal for samples. Write sample site details on side of vial with a permanent pen. If immediate analysis is not possible keep samples refrigerated.



#### Examples of sewage fungus densities



Trace
Present but only just detectable



Thin
Obvious presence but substrate
not obscured



Thick
Thick enough to fully obscure substrate



Massive
Occupies a significant
proportion of the water column

#### Sewage fungus recording sheet

				Site ID:		Date:		Time:	
Water Body: Site Name:						NGR (GPS):			
						Sampler	:		
Photos: Yes / No  TURBIDITY: Tick box ODOUR: Tick box SEWAG									
CLEAR: Water not visibly turbid NONE: No discernible on			dour NONE: No lit		litter present				
SLIGHT: Visible turbidity but no SLIGHT: Odour detectable within the									
MODERN TE O: III + III +						ł			
HIGH: Visibility limited to 10cm depth					GROSS: >60% of possible area				
box for cover and de	ensity			OCHRE: Tick box	for cove	r and den	sity		
	•						-	-1- 1	
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	and obscur	es fine			0% of			and obscures fine	
				area				tices between gravel	
sized particles				EXTENSIVE: >60%			sized particles	-	
SEWAGE FUNGUS ABOVE STONES: Tick box for presence and density						W STON	IES: Tick box for p	resence and densit	у
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obscured				<30% of area			obscured		
THICK: Thick enough t substrate	to fully obs	cure the		WIDESPREAD: 30 - 60 area	0% of		THICK: Thick enough substrate	to fully obscure the	
MASSIVE: Occupies a significant				EXTENSIVE: >60%					
	Laborato	rv:				Date:		Time:	
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FUNGUS BELOW STONES: Field comments  Laboratory:  Date: Time:

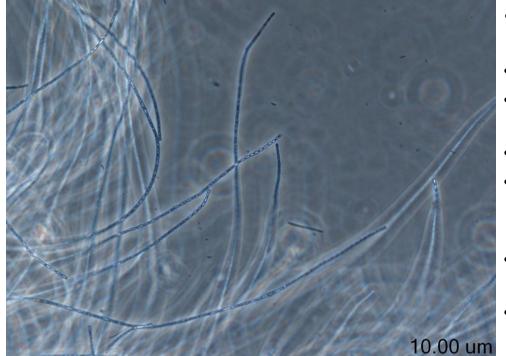
### Main sewage fungus taxa



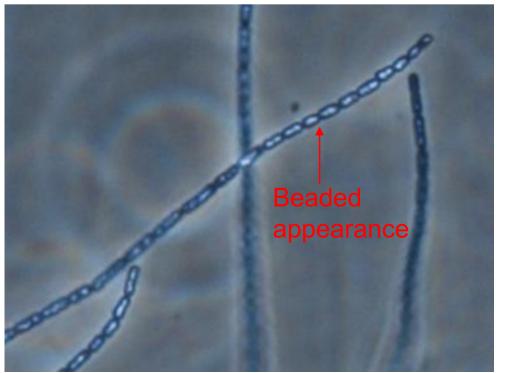


- Filamentous bacterium
- Requires aerobic conditions
- Requires flowing water
- Prefers pH between 6.8-9.0
- Growth form variable from short tufts to slimy fronds
- Tolerates a wide range of organic conditions









#### Sphaerotilus natans

- Filamentous bacterium
- Unbranched filaments with rod shaped cells
- 1-2 µm wide filaments
- Characteristic beaded appearance especially at ends of filaments
- Cells within a closely fitting sheath
- Sheath varies in thickness and can obscure beaded appearance
- False branching can be present
- In lower organic conditions false branching common and thin sheath typical
- In higher organic conditions false branching weakly developed or absent and thick sheath typical
- Zigzag growth may indicate an intermittent discharge

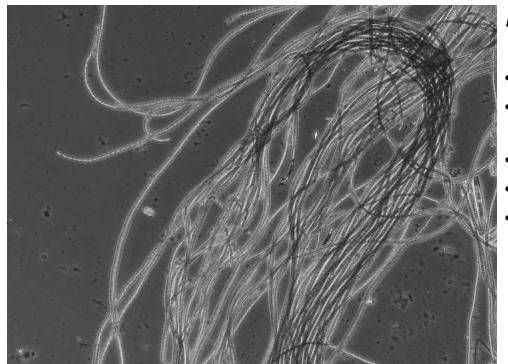


# White film Anoxic sediment



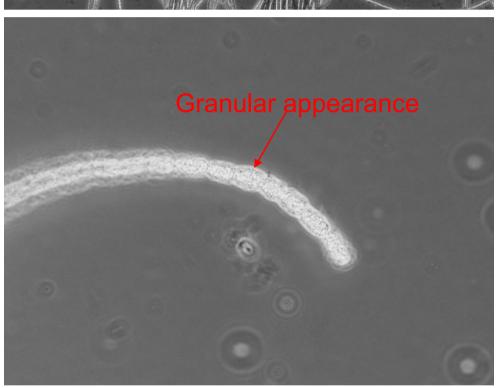
#### Beggiatoa alba

- Filamentous bacterium
- Forms a thin white film on surface of substrate
- Prefers low dissolved oxygen
- Prevalent in high organic conditions
- Oxidises hydrogen sulphide
- Characteristic of slow flowing waters
- Can be found as a monoculture in faster flowing waters where it forms very long fine filaments
- Tolerant of saline conditions



#### Beggiatoa alba

- Filamentous bacterium
- Unbranched filaments with rod shaped cells
- 3-4 µm wide filaments
- Motile filaments
- Sulphur granules stored within cells increasing in number with age

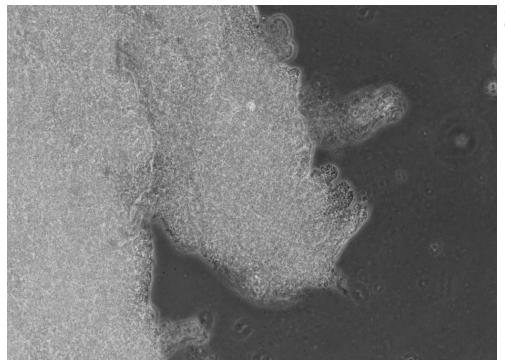






#### Zoogloeal bacteria

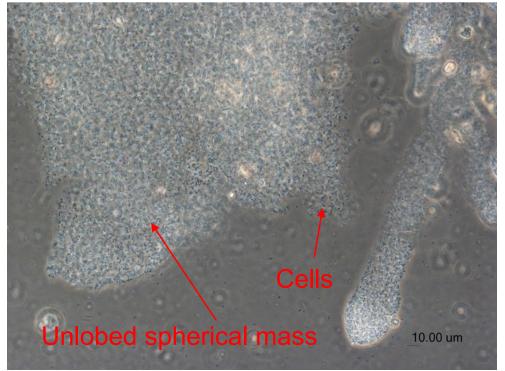
- Not well defined taxonomically
- Jelly-like forming thick gelatinous mass
- Restricted to slow flowing waters
- Prevalent in high organic conditions



#### Zoogloeal bacteria

- Not well defined taxonomically
- Cells embedded in a gelatinous matrix
- Forms lobed and unlobed spherical masses

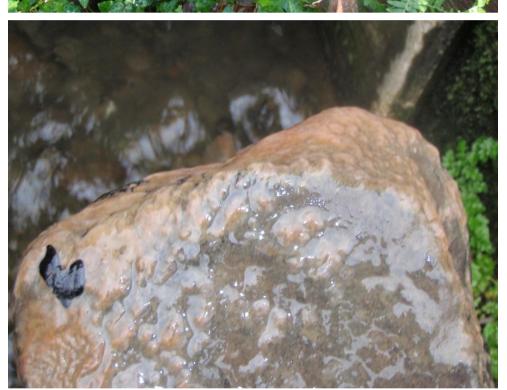


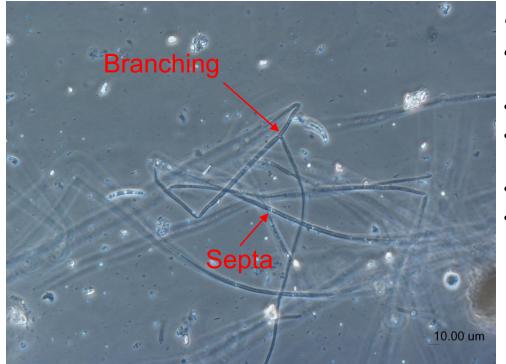




#### Fusarium aquaeductuum

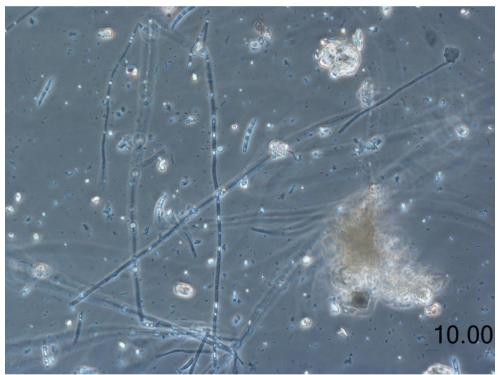
- Filamentous fungus
- Requires high dissolved oxygen
- Prefers acid pH
- Imparts pink or red colouration to growth
- Rarely dominant





#### Fusarium aquaeductuum

- Filamentous fungus
- Septate branched filaments
- 5 µm wide filaments
- Boat-shaped spores freely produced



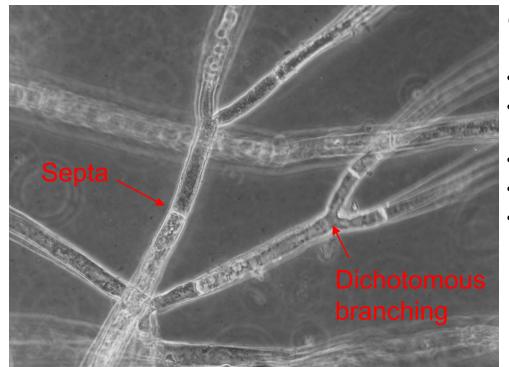






#### Geotrichum candidum

- Filamentous fungus
- Grey to brown in colour
  - Soft texture
- Growth loosely follows contours of stones
- Able to oxidise lactic acid
- Growth supported by ammonia, asparagine and urea
- Often associated with dairy products, silage and pickling wastes
- Prevalent in high organic conditions



#### Geotrichum candidum

- Filamentous fungus
- Septate branched filaments
- 5-10 µm wide filaments
- Dichotomous branching
- Brick-shaped arthrospores may be present

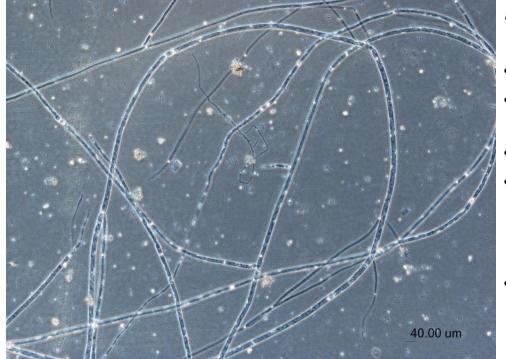


#### Leptomitus lacteus

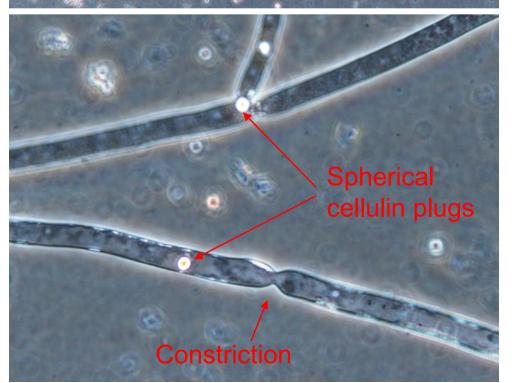
- Filamentous fungus
- Forms long characteristic streamers with overlapping cotton-wool like growths
- Requires high dissolved oxygen
- Prefers moderate to fast flow
- Prefers acid pH







# Branching 20.00 um



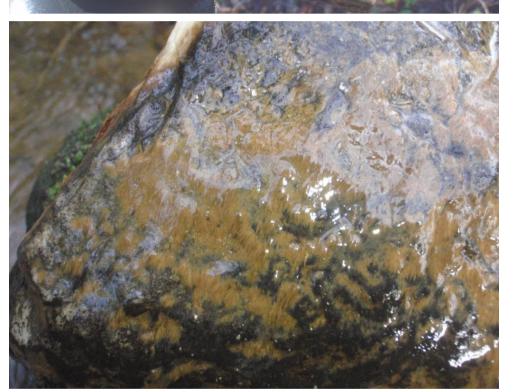
#### Leptomitus lacteus

- Filamentous fungus
- Non-septate coarse branching filaments
- 8-15 µm wide filaments
- Characteristic constrictions at intervals with spherical cellulin plugs
- Cellulin plugs block the constrictions and prevent cytoplasm flowing away from the growing apices of the filaments



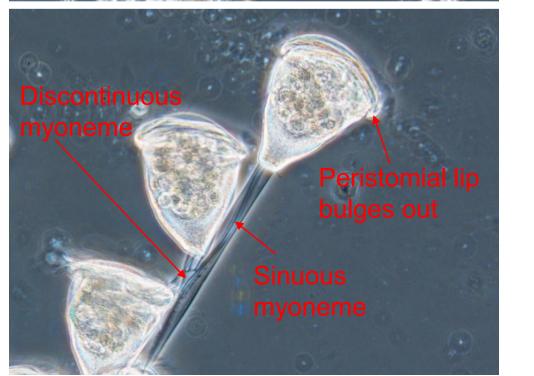
#### Carchesium polypinum

- Stalked protozoan
- Bacteria feeder
- Growth form of short tufts of 2-3 mm
- Prevalent in low organic conditions and recovery zones





# Bell-shaped zoold Branched stalk 20.00 um



#### Carchesium polypinum

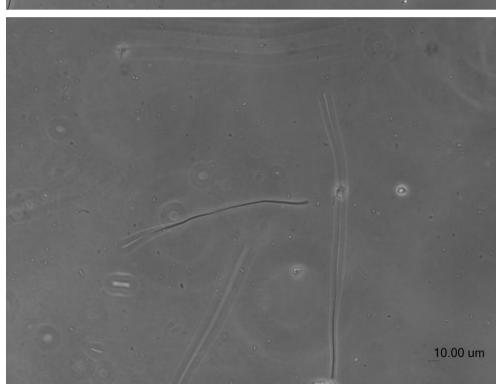
- Stalked protozoan
- Inverted bell-shaped zooids at ends of stalks
- Branched stalks
- Zooids can become separated from stalks, especially in degraded samples
- Stalks can contract independent of each other
- Stalks contract spirally
- Stalk with discontinuous myoneme
- Sinuous myoneme
- Zooids 100-125 μm long
- Zooid peristomial lip bulges out
- Zooid with C-shaped macronucleus
- Zooid with smooth surface

### Other sewage fungus taxa



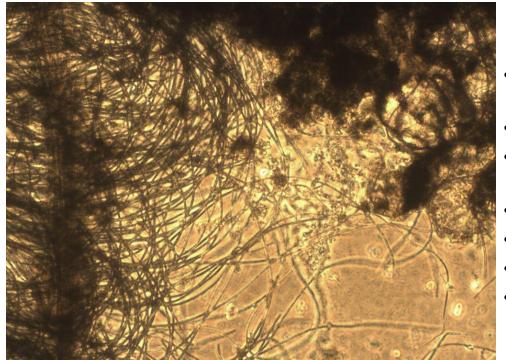
#### Flexibacter spp.

- Filamentous bacterium
- Unbranched filaments
- Motile with whole filament bending and flexing
- No sulphur granules in cells
- Rarely dominant



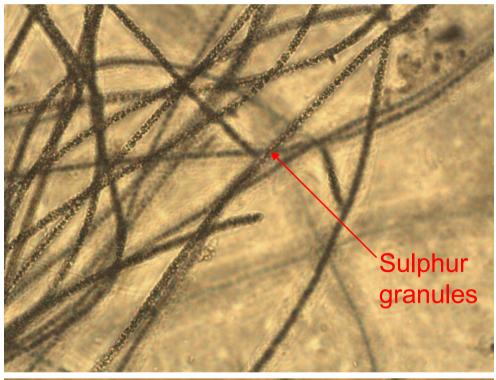
#### Thiothrix II

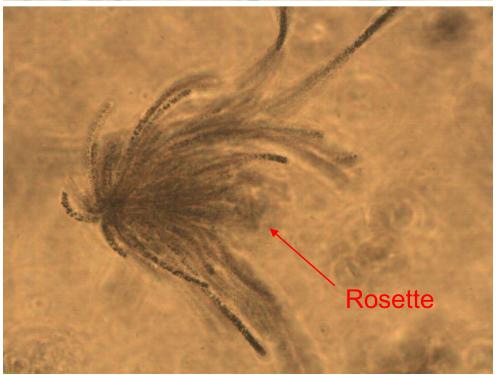
- Gammaproteobacteria
- Uses low molecular carbon sources (shortchain fatty acids and alcohols) as well as reduced sulphur compounds
- Does not grow in anoxic conditions
- Found associated with cows bedded on waste gypsum in West Wales



#### Thiothrix II

- Straight or bowed filaments
- 1 um wide filaments
- False branching and rosette formation
- Sulphur granules evident
- No obvious septa
- Not motile
- Filaments do not taper





#### Achyla spp.

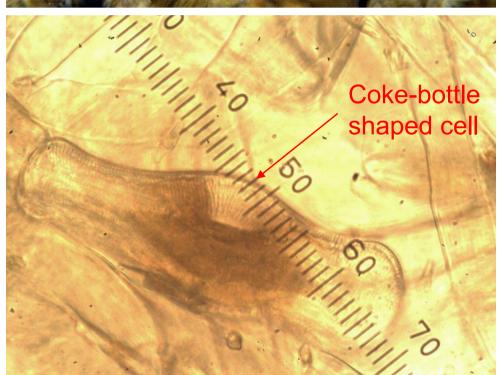
- Filamentous phycomycete fungus
- Similar to Leptomitus but wider filaments and no spherical cellulin plugs
- Tends to grow near source of silage effluent with which it is closely associated

#### Flavobacterium spp.

- Filamentous bacterium
- Unbranched filaments
- 0.5-1 µm wide filaments lying parallel to each other
- Filaments lying close together separated by 10-12µm
- Individual cells 0.5-1 µm wide by 10-50µm long
- Occasionally forming pink or yellow growths
- Requires organic nitrogen source for growth
- Rarely dominant

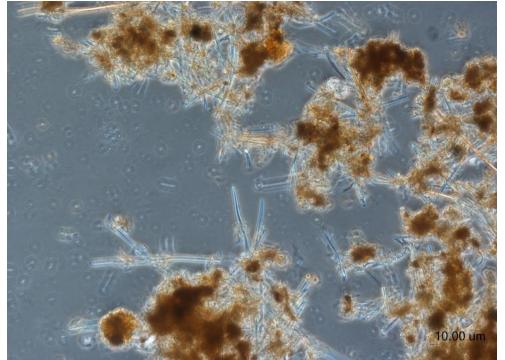
## Taxa commonly mistaken for sewage fungus

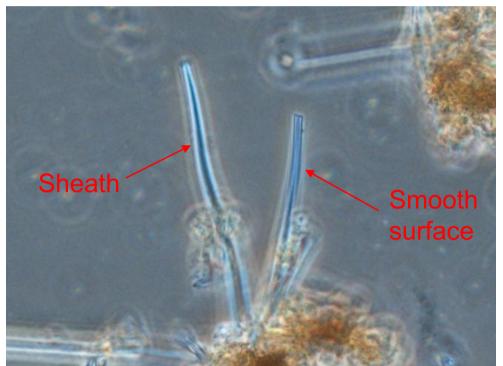




#### Didymosphenia spp.

- Stalked diatom
- Generally occurs as offwhite cohesive lumps or mats
- Prefers cool oligotrophic waters
- Not slimy
- Feels spongy and scratchy, like cotton wool
- Does not indicate pollution
- Obvious 'coke-bottle' shaped diatom cells embedded in a mass of extracellular stalks





#### Leptothrix ochracea

- Filamentous iron bacterium
- 2-3 µm wide filaments
- Grows in slow flowing waters high in ferrous iron and low in organic matter
- Requires ferrous (iron) salts, oxygen and carbon dioxide for growth
- Growth of bacterium results in accumulation and sedimentation of orange-brown ferric hydroxide in sheath surrounding filament
- Sheath has smooth surface
- Growth results in many empty sheaths