Diversity of sandflagellates in the Oslofjord

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This study was carried out in the Marine Botany department of the Biology

Institute, University of Oslo, under the supervision of Professor Jahn Throndsen and partly

in collaboration with the "Sand Flagellates" research branch of the MARE project.

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Eider Zubizarreta Garai

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1.0 Introduction

1.1 Aim of this master thesis

This thesis seeks to document the diversity of auto- and heterotrophic flagellates in two sandy beaches from the Oslofjord and give a brief insight on the physical and chemical factors that influence the sediment and the organisms that it hosts.

1.2 What is a benthic flagellate

Benthic flagellates in soft bottoms, also known as psammobic* flagellates, are free-living unicellular eukaryotic organisms with one or more flagella that inhabit the interstices of the sediment particles or have temporary connection to the substratum (Fig. 1, Fig.2).

Within this group, we find both non-photosynthetic (protozoans) and photosynthetic representatives (considered as algal cells) which most commonly find place in the following classification groups: dinoflagellates, (auto and heterotrophs), cryptomonads, haptophytes, euglenids and taxa of uncertain taxonomic affiliation. Most of the organisms here fall into the concept of protista, but some others (dinoflagellates and euglenids) fit well in the traditional group of the Algae.

The range of size of these organisms varies in the order of 2 μ m to 50 μ m (Patterson *et al.* 1989), cells exceeding these values were also recorded. We deal, therefore, with nanoplankton (2-20 μ m) and microplankton (20-200 μ m) sized cells.

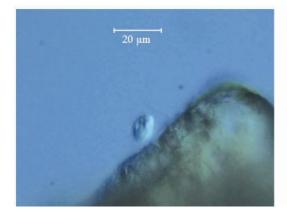


Fig 1. Flagellate on particle surface

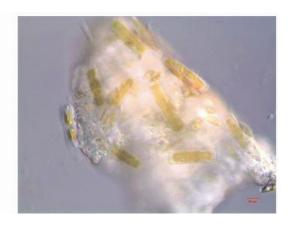


Fig 2. Diatoms on sand grain

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^{*} Psammon: sand; psammobic: sand dwelling organisms.

Living in the sandy sediments of a beach implies having developed adaptive morphologies and behavioural patterns. Most organisms adopt one of two adaptive strategies as for motion: tactic species exploit surface-water interfaces by gliding, and swimming species move freely within the pores (Patterson, Larsen & Corliss, 1989). Morphologically, flat ones dominate.

Regarding the biology of these organisms, we find that only few are photosynthetic, the great majority consisting of heterotrophs. According to Patterson *et al*, 1993, the conditions in the depths of the water column, are comparable to those found in the sand which combines both low light and low O_2 levels, giving rise to a relative increase in heterotrophic forms of life.

1.3 Benthic flagellates in the marine food web

In contrast to the extensive investigations carried out on the protists belonging to the marine pelagic communities, those from sediments of the litoral areas have been subject of few studies. A reason for this may lie in the heterogeneity of the sand environment and the difficulty in extracting the organisms from it (Fenchel, 1987). The group of benthic ciliates too have been paid more attention. But the contribution of the flagellates to other ecosystems has suggested that they may prove to be ecologically more important than ciliates (Andersen and Fenchel, 1985; Fenchel 1970).

Heterotrophic flagellates, the most abundant and studied group among the benthic sand-dwelling flagellates, play an important role in the so called "microbial loop" as they function as predators on bacteria and diatoms, and as prey for larger zoobenthos. In addition, their excretions serve in the remineralization of essential elements and this way promote phytoplankton growth (Won Je Lee, 2001). Their smaller size brings about a higher degree of dissolved organic matter excretion opposed to larger protists such as ciliates, because excretion rate is inversely related to size (Dolan, 1997). Therefore, their major contribution to the marine food web comes with their nutrient recycling ability, along with their innate prey-predator property.

As for autotrophic species, such as some *Amphidinium* and *Gymnodinium* dinoflagellates and euglenophytes, together with diatoms and cyanobacteria, they are likely to be the most important contributors to primary production in the interstitial zone (Flø Jørgensen *et al.*, 2004). Their ecological significance needs, however, to be further investigated, as ecological studies of benthic flagellates have mainly focused on the role and abundance of heterotrophic species (Fenchel, 1967, 1969, Larsen and Patterson 1990, Lee and Patterson 2002).

1.4 Characterization of the sand habitat, some ecology

The beach appears at first glance as an inhospitable space, but it conceals an enormous variety and a highly adapted community. The interstitial marine habitat is a mechanically unstable, porous habitat, which accumulates metabolic energy from two main sources: carbon-fixing activity from autoctonous photosynthetic organisms in the euphotic layers and from the degradation and reassimilation of organic material from the overlying water (Patterson *et al* 1989). It is an open system which hosts not only autochtonous but also alochtonous organisms from the overlaying plankton, and a dynamic system because the sediment suffers a continuous mechanical disturbance by the action of the waves and tides which flagellates need to cope with.

Regarding the physical aspect of this habitat, the size of particles plays a critical part because it influences the porosity, permeability and capillarity of the sediment (Fenchel, 1969). Porosity is defined as the interstitial space, which is of little importance to the biota (Patterson *et al.* 1989). Oppositely, permeability, understood as the ability of the water to drain through the sediment, and capillarity, as the property to retain water, are of great significance in the diversity, distribution and abundance of sand biota in general. Larsen (1985) suggests, for example, that sand particles below 125 µm in diameter host low numbers of dinoflagellates and that even the proportion of armoured and unarmoured dinoflagellates may be a consequence of the particle size.

On the other hand, small particles are readily maintained in suspension by turbulence and drifted away. Thus, fine sediments form only in sheltered areas. Particulate organic matter follows the same fate: it is suspended and drifted to low turbulent zones, where it remains adsorbed to the fine sediment particles thanks to their high internal surface area. Consequently, silt and muddy shores will be nutrient-rich and coarser sandy shores, poorer (Patterson *et al.* 1989).

At the same time, finer sediments, due to a scarce water renewal, an active microbial break-down, a low photosynthesis caused by a low penetration of light, and little air that makes it through the compressed sediment, result in a poorly oxygenated habitat. Whereas, coarser sediments, which originate in higher turbulent shores, have higher O₂ levels due to a lower organic content and a stronger aerial drainage.

The supply of oxygen into the different layers of the sand is, indeed, the most influential of all chemical factors on the biology of sediments. (Little, 2000). The resultant oxygen profile is again a consequence of other factors: the above mentioned, nature of the sediment, the decomposing and production (C-fixing) activity, and the bioturbation by burrowers and the light intensity of the radiation penetrating the sediment.

The latter, is affected by the depth of the overlying water, climatic conditions and again by the sediment type. The light attenuates fast through the sand, and already at a few mm into the sediment, the bottom of the euphotic zone (1% of the total incident illumination) is reached (Patterson *et al* 1989). This leaves only a fine layer for the obligate photosynthetic ones to live in. Even so, some autotrophs (diatoms) may be found deeper in the sand, which could be explained by attributing them facultative heterotrophic abilities or vertical migration.

Factors such as temperature and salinity vary enormously daily and seasonally and therefore are not attributed significant power in driving diversity. These organisms tackle salinities up to 50‰ which result from tide and wave withdrawal and thus desiccation (Jacob Larsen, pers. com.).

In regards to the zonation of sandy shores, different criteria have been used for this purpose. McLachlan and Jaramillo (1995, Fig.3 below) pay attention to the strength of the wave action against the shore and raise the concepts of reactive and dissipative beaches. In the latter, the force of the tides and waves is so strong that it attenuates itself while creating a long and smooth surf zone, where the sand will eventually be fine. In reactive beaches, the force of tides and waves is weak and so it will be diminished readily and forming a short and sloped stretch. The sand of reactive beaches is much coarser. McLachlan also defines the sand as dry, damp and saturated regarding the degree of moist.

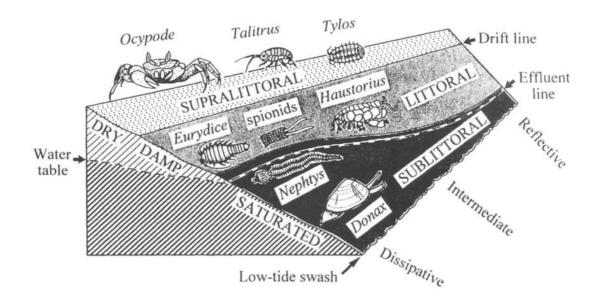


Fig. 3 Scheme of zonation of sandy shores (After McLachlan and Jaramillo, 1995)

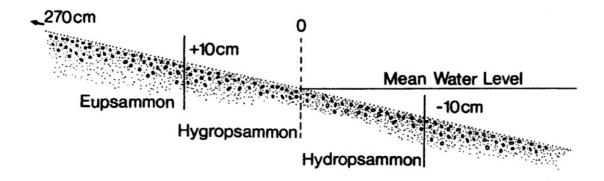


Fig. 4 Diagrammatic representation of marine psammon zonation (Wiszniewski, 1937)

Wiszniewski (1937, Fig. 4 above) distinguishes different zones of the beach on the basis of the water content raising the terms: Hydropsammon to the completely saturated sand, Hygropsammon to that partially saturated sand due to capillarity and splashing of the waves, and Eupsammon, to the dry sand.

The study presented in this thesis, focuses on the Hygropsammon zone.

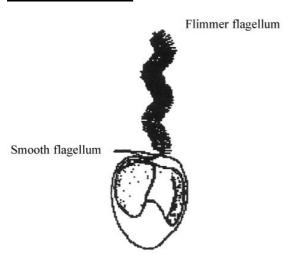
1.5 Short review of previous work on sand flagellates in Norway

The interstitial flagellates are receiving increasing attention, but the Norwegian sandy shores are yet little studied in comparison to those from tidal flats and muddy beaches in other countries in Scandinavia, like Denmark (Kaas et al 1985, Larsen 1985, Larsen 1987) and Finland (Vørs 1992). If observations concerning sand flagellates have been carried out in Norway, they have not been published yet. Previous to the present study, only Grimsrud's (2001) work in the Oslofjord is known to me.

1.6. Overview of the systematics and expected taxa

The systematics of the different flagellate groups encountered will be addressed in the following section.

Heterokontophyta



The main characteristic of this division is the two different flagella they possess (see schematical drawing to the left). One is smooth and shorter and it is projected backwards or to the side, while the other one is longer, directed into the direction of swimming and covered with thin, tubular, stiff hairs (mastigonems).

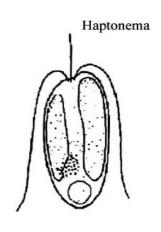
Among the sand flagellates from this group *Olisthodicus luteus* is usually the most common and it is classified within the class Raphidophyceae (see schematical drawing to the right, ventral and profile sides).





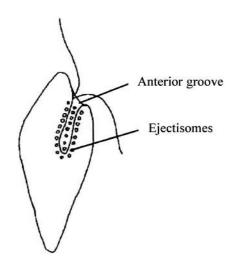
Haptophyta (cl. Prymnesiophyceae)

The algae from this division are autotrophic with two chloroplasts, and have two smooth flagella of equal or unequal length. In addition they have a filament-like structure coming out near the flagellar insertion area (see diagram on the right). Sometimes it is too short to be observed in the light microscope. It can be long or short, and the long ones can coil. It is thought to function as a prey-catching and attaching structure.



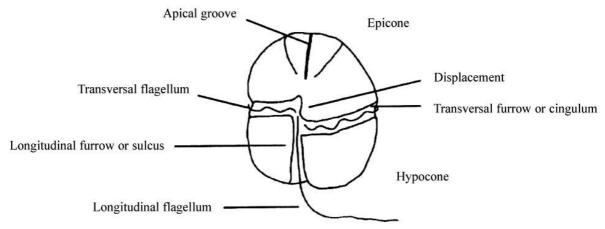
Cryptophyta

Cryptomonads are naked cells with two slightly unequal flagella emerging from an anterior pocket, vestibulum (see drawing on the right). Cryptomonads are distinguished by the presence of characteristic extrusomes called ejectisomes, which consist of two connected spiral ribbons held under tension. Large ejectisomes, visible under the light microscope, are associated with the furrow and gullet. Most of them, have one or two chloroplasts, but there are a few colourless: ex. the genus *Goniomonas* Stein 1878. Cryptomonads may have very delicate surface scales which are only revealed by applying special techniques.



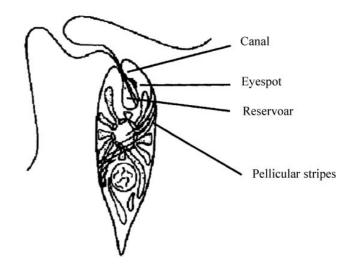
Dinophyta

Dinoflagellates are unicellular forms with two dissimilar flagella. One of these extends towards the posterior, called the longitudinal flagellum, while the other forms a lateral circle, called the transversal flagellum. In many forms these are set into grooves, called the sulcus and cingulum (see figure below). The transverse flagellum provides most of the force propelling the cell, and often imparts to it a distinctive whirling motion. Dinoflagellates have a complex cell cover called amphiesma, composed of flattened vesicles called alveoli. In some forms, these contain cellulose plates that make up a sort of armour called the theca. These come in various shapes and arrangements, depending on the species and sometimes stage of the dinoflagellate. Species that belong to the sand habitat are mostly unarmoured, and many of them have an apical groove. Autotrophic as well as heterotrophic representatives are commonly observed from the following genera: *Amphidinium*, *Gymnodinium*, *Gyrodinium* and *Katodinium*.

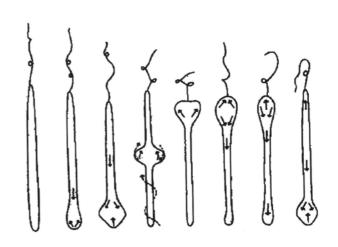


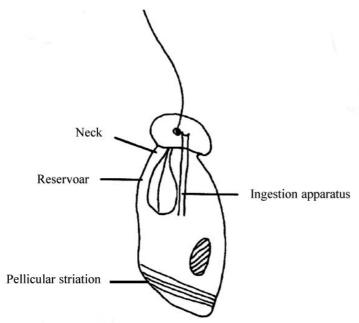
Euglenophyta (Euglenozoa)

These organisms can be both auto- or heterotrophs and are characterized by having one, two or multiple of two number of flagella coming out from an intracellular pocket known as the reservoir. It narrows in a canal as it approaches the outside of the cell. The cell surface is usually striped and this shape is known as pellicula (see figure on the right). Autrotrophic species have an eyespot which help them light-orientate.



A proteineous structure in the cell membrane, called pellicula, which consists of striae that travel along the cell surface, is responsible for the characteristic euglenoid movement (see schematical drawing to the right).

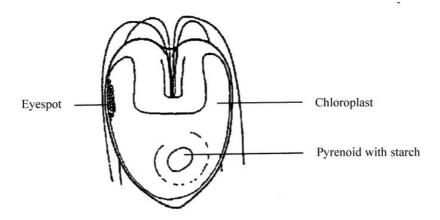




In the sediments most euglenophytes speciess are colourless from diversity. They extensive usually possess an ingestion organelle formed by two longitudinal rods and share the other same features with the rest of the Euglenophytes. To the left, a sketch of one of the heterotrophic genera (Urceolus) is presented. Other genera like Anisonema, Dinema, Notosolenus, Petalomonas, Ploeotia, etc, are also represented in the sediment.

Chlorophyta

These are photosynthetic green-coloured cells with two or multiple of two number of flagella, and an eyespot and a pyrenoid within a single chloroplast. Storage products, in form of starch appear associated with the pyrenoid. Two main class are common in the interstitices of marine sediments: Prasinophyceae with main genus *Pyramimonas* (see figure below) and Chlorophyceae, with *Chlamydomonas* as the principal representative.



2.0 MATERIAL AND METHODS

Two localities of the Oslofjord were sampled and screened for sand flagellates: Huk and Hulvika. They represent two typical sandy beaches in the Oslofjord.

2.1 Description of the localities

The two localities investigated are situated within the Oslofjord: Huk, in the Bygdøy peninsula in Oslo (Fig. 5, 6) and Hulvika, in the eastern part of the Oslofjord, south from Oslo city (Fig. 7, 8).

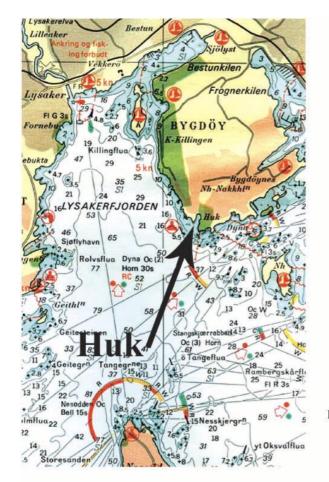
Huk is a sandy beach divided by a natural overhanging rock covered in macroalgae and blue mussels at its littoral belt (Fig. 10). It is located in the inner part of the Oslofjord and faces the south-southwest. It is a quite sheltered beach but with a pronounced exposure to pollution due to the densely inhabited area in which it is included, and with a slight wave action produced by a constant passing of heavy ships to the harbour of Oslo. It is, besides, a part of a recreational area exploited during the summer and weekends.

Sand-grain size is somewhat coarse with strong variation within the surf zone, where patches of different particle size are often observed. Broken mussels shells accumulate in the surf zone.

Hulvika is a narrow, south-southwest oriented bay (Fig. 12). In the inner part, a little freshwater stream runs through the beach from agriculture fields. It flows abundantly during the snow melting period and appears almost dry in the wintertime. On a local basis, it influences the salinity of the surface water body, especially when the wind and the weather are such that the surface layer presses against the beach: e.g. at strong south or west winds and currents. In spring, however, the north and east components of the winds are the most important.

The bay is shallow, up to 3 m at the deepest close to the beach; about 20 m far towards the open fjord it becomes deeper, 14 m maximum. The grain size is heterogeneous, the beach appears as clusters of different particle size.

When the south wind blows right towards the bay, the waves become stronger, as well as at passing boats. This effect gets amplified in the inner part of the bay due to the reflection of the waves on the rocky walls that flank this narrow branch of the fjord.



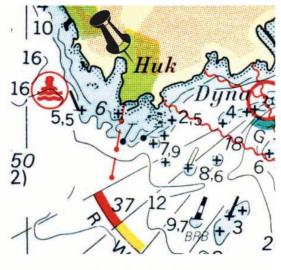


Fig. 6 Huk in detail

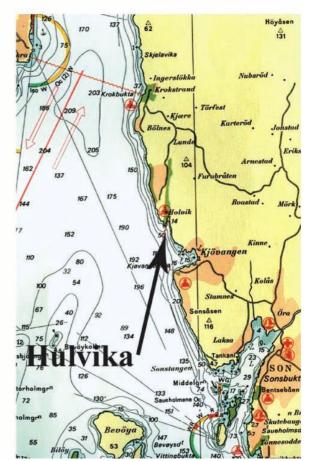
Fig. 5 Map of the inner Oslofjord showing Huk

Fig. 7 Map of the Oslofjord showing Hulvika

190 Holoik
190 115 2+15 11

Kjøvangbukta

Fig. 8 Hulvika in detail



11



Fig. 10 Sampling at Huk



Fig 11. Hulvika beach flanked by rocky walls on both sides

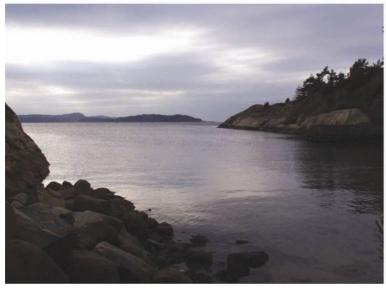


Fig 12. Hulvika, a narrow bay.

2.2 Collecting procedure and parameters measured

Sampling of the sand was performed throughout the first semester of the year 2005. Hulvika was sampled approximately every four weeks and Huk every three. The samples were collected in the daytime, between 11:00-15:00.

The sand was collected in the surf zone, that is, in the narrow range where the tide meets the beach. This sand is constantly moisten by the waves, but not completely submerged. However, sometimes it was required to get the sand from around the stripe submerged 10 cm. This was done at rising tide because the sand at this depth had been covered with water long enough to expect it be populated with psammobic organisms. These organisms may have come from deeper layers in the sand or from nearby wet sand. Generally, however, it was preferable to sample at falling tide.

The sand was collected by means of a plastic bailer with a ca. 20 cm wide and flat mouth right after the withdrawal of the waves (Fig.13). Only the uppermost 0.5 cm to 1 cm sand was collected in order to avoid hypoxic or anoxic sand from depths where light could not reach. This anoxicity was easily detected by the dark colour of the sand and the smell of sulfhydric acid (H₂S) that it expelled. To keep the sand from desiccating, it was stored with water to its saturation.

The sand was transferred into plastic 20 cm-diameter Petri dishes and into 100 ml borosilicate glass reagent bottles with the help of a kitchen spoon. They were stored in climate rooms at the same temperature as originally collected.

An YSI Model 63 (YSI Inc., Yellow Springs, Ohio, USA) was used to measure salinity, temperature, pH and conductivity. These parameters were registered placing the instrument's probe in the water lying immediately above the sand.

2.3 Extraction methodology and microscopy

For a better observation of psammobic organisms, they need to be taken out of the sand preferably by their own means. Shaking of the sand to extract the organisms out, proved to be a bad method in that few of them survived and that the resultant water was full of particles and dead organic matter. Therefore, organisms were desired to be obtained alive, intact and in a clear water or substrate.

Two extraction methods were applied for this purpose. One would be used for further viewing in the light microscope and the other one for the Scanning Electron Microscopy (SEM).



Fig. 13 Sampling sequence



Fig 14 Petri dish showing cover slips on the sand

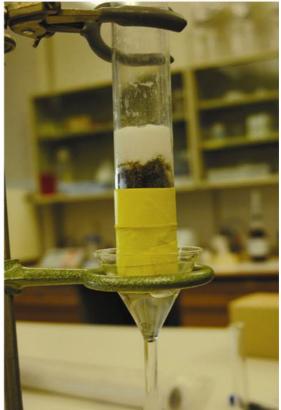


Fig 15 Ice method set up

2.3.1 Coverslip method and Light Microscopy

2.3.1.a) Coverslip method

This method is based on the adherence of the cells to the coverslips.

Previously submerged in alcohol and left to dry, these coverslips were placed on the sand surface kept in Petri dishes (Fig. 14) for at least two hours before being observed in the light microscope. The purpose of this was to let the flagellates adhere to the glass. They swam from the interstices of the grains to the fine layer of the seawater between the sand and the coverslip. They either adhere themselves to the glass, or stay swimming in the water that gets adhered to the coverslip when this one is grabbed out of the sand with a pair of forceps. Coverslips were then put on slides and brought for observation in the light microscope.

The coverslips were observed thoughout several days. Therefore, the time for the organisms to adhere onto the glass varied from 2 hours up to even two weeks.

2.3.1.b) Observation in LM and digital photography

Observation in the light microscope was carried out in a Nikon Microphot-FX microscope; for digital photography, a Spot Insight Color model 3.2.0 (see Diagnostic Intruments from Nikon), in the beginning, and a Nikon D70, later in the study, were employed. As for optics, phase contrast at 10x resulted best in screening for cells. For deeper details, DIC (differential interpherence contrast) was used for both at 20 and 40x objectives.

Observation at 20x resulted usually in an optimum as to viewing the cell shape good enough but failed as to giving further details (surface texture, flagellar insertion, etc) to identify it down to genus or species levels.

Therefore, the combination of the three objectives was necessary for drawing and taking notes and concluding in a taxon. In many cases, however, light microscopy did not suffice for species or genus clarification. In these cases, it was desired to view the cells at higher magnification in the SEM. This demanded a specific cell extraction method in which cells would be collected in water, as clear as possible, and a further treatment for preparing the material for SEM.

2.3.2 Melting ice method and Scanning Electron Microscopy

2.3.2.a) Melting ice method

The melting ice method (Uhlig, 1964) consists of a steep temperature gradient built up in the substratum through the addition of ice from above, which forces flagellates to abandon the interstices of the sand.

The set up was built as follows:

A 5 cm diameter plastic cylinder void inside worked as the main body. A 90 μ m pore size mesh was tightly strapped to one end with a powerful tape. This cylinder was vertically sustained by means of a laboratory clamp where the mesh would face downwards.

About 25 ml of sand was transferred into the tube with the help of a teaspoon. The first dripping of the sand water itself through the mesh was not collected, due to high amount of dirt. On top of the sand, some cotton would go to help the drainage from the ice be administrated homogeneously to the sand. Finally, a filtered sea water ice block was put. This ice was obtained by freezing around 50 ml of filtered seawater in another 5 cm diameter plastic cylinder in upright position. This way the shape of the ice suited perfectly in the set-up cylinder (Fig. 15, 16).

A ca 7 cm diameter Petri dish filled with 5 to 10 ml of filtered seawater waited below the set-up for the flagellates to flow downwards and traverse the mesh to end up in it. The water of the Petri dish required being in contact with the mesh of the cylinder, so that the cells flew themselves through it into the water contained in the Petri dish. As the ice was melting and the water level rising, new Petri dishes were replaced.

This water was immediately observed in a Nikon model TMS inverted microscope in order to transfer the flagellates into a less dirty and smaller amount of seawater. The water was screened for cells using 20x phase contrast. All the flagellates were indiscriminately sucked with a pipette and transferred to a clear smaller water volume contained in a pointed-end short glass sampling tube. This procedure was performed with all the Petri dishes resulting from the melting of the ice block, usually not more than three. It was intended to collect as many cells as possible, but around 300 cells were considered to be sufficient to compensate the loss that originated from the further preparation process for SEM.

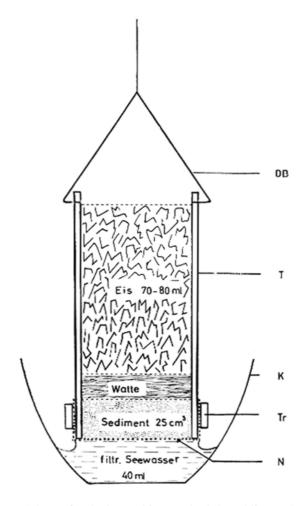


Fig 16. Scheme for the ice melting method (by Uhlig 1964).

2.3.2.b) Preparation and observation in the SEM

The organisms collected through the melting ice method were fixed with a combination of two fixatives: 1% iodine (Lugol) and 0,25% Glutaraldehyde. This was chosen following the advice of Wenche Eikrem at the Marine Biology Department at UiO. For the next steps different variants were performed. In the beginning, the fixed material followed centrifugation and then was rinsed with caccodilat acid 0.1M several times (at least 3 times). The realization that centrifugation might lead to a shedding of the flagella, it was decided to avoid centrifuge in the next preparations. In order to replace the concentration of the cells by a centrifuge, the fixed material was left to sediment between each rinsing and post-fixing steps.

Post-fixing was fulfilled with OsO_4 during 1h and underwent rinsing with cacodylate buffer right after.

As for finding a physical support to the cells, two alternatives were tested to find the most suitable one for these particular flagellates. One part followed filtration through a "nucle pore" filter, and a second one was left to sediment in 8 mm diameter poly-L-lysin cover slips in a wet chamber. These two variants met again in the serial ethanol dehydration step. The filters and the coverslips underwent 50%, 70%, 90% and 96% serie for 10 minutes each and 4 times at 100% for 15 minutes each. Then, the critical point drying took place. This was suspected to be a critical step when it came to loosing material, but was proved to be the best alternative for a better dehydration in comparison to dehydrating the cells with Hexamethyldisilazane.

Then, both the filters and the coverslips were mounted on a stub by means of carbon filters and brought to the gold-palladium coating apparatus. The material was then ready to be observed in the SEM.

2.4 Identification of the species

Identification was mostly based on Larsen & Patterson (1991), Herdman (1920-24), Hulburt (1957) and Campbell (1973), but in many cases assisted by other works listed in the reference list. Whenever appropriate a reference to the literature consulted is included with the species comments.

As the proficiency of methods and ability to identify the species were increasing throughout the sampling period, the seasonal occurrence cannot be presented for many of the species.

3.0 RESULTS

3.1 Description of the species found in the Light Microscope

The species encountered are presented below in accordance with their systematical position following Larsen & Patterson (1991) with some recently published changes. Species which was readily identified and in full accordance with the original descriptions or those of Larsen & Patterson (1991) are only briefly commented upon, whereas taxa which showed deviating features/ characters are treated more extensively.

Species found in the present study are listed below (based on botanical nomenclature) followed by the descriptions of the species.

Div. Heterokontophyta

Class Raphidophyceae Norris 1982

Order Chattonellales Throndsen 1993

Olisthodiscus luteus N. Carter 1937

Class Dictyochophyceae (P.C. Silva 1980) Silva 1982

Order Pedinellales Zimmermann et al. 1984

Actinomonas mirabilis Kent 1880 / Pteridomonas danica Larsen and

Patterson 1990

Div. Haptophyta

Class Prymnesiophyceae Hibberd 1976

Order Isochrysidales Pascher 1910

Dicrateria gilva M. Parke 1949

Order Prymnesiales Papenfuss 1955

Prymnesium nemamethecum Pienaar & Birkhead 1994

Div. Cryptophyta

Class Cryptophyceae Fritsch 1927

Order Cryptophyceae Engler 1903

Chroomonas diploccoca Butcher 1959

Cryptomonas sp.

Order Goniomonadales Novarino & Lucas 1993

Goniomonas amphinema Larsen and Patterson 1990

G. pacifica Larsen and Patterson 1990

Class Dinophyceae Fritsch, 1929= Dinoflagellida Bütschli 1885

Order Gymnodiniales Lemmermann 1910

Genus Amphidinium Claparède & Lachmann 1859

Amphidinium bipes Herdman 1924

A. britannicum (Herdman) Lebour 1925 = Togula britannica, T. compacta,

T. jolla (Herdman) M. Flø Jørgensen, S. Murray, & N. Daugbjerg 2004

A. cf. carterae Hulburt 1957

A. herdmanii Kofoid and Swezy 1921

A. herdmanii Kofoid and Swezy 1921

A.poecilochroum J. Larsen 1985

A. psittacus J. Larsen 1985

A. testudo Herdman 1924

A. cf. trulla Jørgensen & Murray 2004 (= A. operculatum Claparède &

Lachmann, 1858)

Genus Glenodinium (Ehrenberg) Stein 1883

Glenodinium cf. foliaceum sensu Conrad et Kufferath

Genus Chilodinium Massart 1920

Chilodinium cruciatum (Massart 1900/1901) Massart 1920

Genus Gymnodinium Stein 1883

Gymnodinium cf. danicans Campbell 1973

G. variabile Herdman 1924

Gymnodinium sp.

Genus Gyrodinium Kofoid & Swezy

G. cf. estuariale Hulburt 1957

G. cf. lebourae Herdman 1924

Gyrodinium resplendens Hulburt 1957

G. uncatenum Hulburt 1957

Genus Katodinium Fott 1957

Katodinium cf. fungiforme (Assimova 1926) Loeblic 1965

Katodinium glandula (Herdman 1924) Loeblich 1965

Order Prorocentrales

Genus Prorocentrum Dodge 1975

Prorocentrum cf. lima (Ehrenberg) Dodge 1975

Div. Euglenophyta

Class Euglenophyceae Schoenichen 1925= Euglenida Bütschli 1884

Order Sphenomonadales Leedale 1967

Genus Anisonema Dujardin 1841

Anisonema cf. acinus Dujardin 1841

A. glaciale J. Larsen and Patterson 1990

A. prosgeobium Skuja 1939

Genus Metanema Senn 1900

cf. Metanema strenuum (Skuja 1948) Larsen 1987

Genus Notosolenus Stokes 1884

Notosolenus apocamptus Stokes 1884

N. hemicircularis Lee and Patterson 2000

N. urceolatus J. Larsen and Patterson 1990

cf. Notosolenus sp.

Genus Petalomonas Stein 1878

Petalomonas cf. cantuscygni Cann and Pennick 1986

P. minuta Hollande 1942

P. poosilla J. Larsen and Patterson 1990

Order Eutreptiales Leedale 1967

Genus Cyclidiopsis Korschikow 1917

Cyclidiopsis acus Korschikow 1917

Genus Eutreptiella da Cunha 1913

Eutreptiella sp.

Euglenophyta sp.

Order Heteronematales Leedale 1967

Genus Dinema Perty 1852

Dinema litoralis Skuja 1939

D. valida J. Larsen and Patterson 1990

Dinema sp.

Genus Heteronema Dujardin 1841

Heteronema exaratum J. Larsen and Patterson 1990

H. ovale Kahl 1928

Genus Jenningsia Lee, Blackmore and Patterson 1999

Jenningsia cf. macrostoma (Ekebom et al. 1996) Lee et al. 1999

Genus Peranema Dujardin 1841

Peranema dolichonema J. Larsen and Patterson 1990

Genus Ploeotia Leedale 1969

Ploeotia adhaerens J. Larsen and Patterson 1990

P. corrugata J. Larsen and Patterson 1990

P. pseudanisonema J. Larsen and Patterson 1990

P. tenuis J. Larsen and Patterson 1990

P. vitrea Dujardin, 1841 emend. Farmer and Triemer 1987

Genus Urceolus Mereschkowsky 1879

Urceolus cornutus J. Larsen and Patterson 1990

U. cf. cristatus J. Larsen and Patterson 1990

U. cyclostomus (Stein, 1878) Mereschkowsky 1878

U. sabulosus (Stokes 1886) J. Larsen and Patterson 1990

Div. Chlorophyta

Class Prasinophyceae T. Christensen 1962 ex Moestrup & Throndsen 1988

Order Chlorodendrales Fritsch 1917

Genus Nephroselmis Stein 1878

Nephroselmis cf. rotunda (N. Carter) Fott

Genus Pyramimonas Schmarda 1850

Pyramimonas cf. orientalis Butcher 1959

Class Chlorophyceae sensu Mattox et Stewart 1984

Order Volvocales Oltmanns 1904

Genus Chlamydomonas Ehrenberg 1834

Chlamydomonas cf. nonpulsata Butcher 1959

Group related to multicellular taxa (Lee et al. 2000)

Class Choanoflagellatea sensu Cavalier-Smith (2002)

Order Choanoflagellida W.S. Kent 1880

Acanthocorbis sp. S. Hara & E. Takahashi 1984

Species of uncertain taxonomy: Insertae sedis

Amastigomonas mutabilis (Griessmann) Molina and Nerad 1991

Amastigomonas debruynei de Saedeleer 1931

Bodo designis Skuja 1948

B. cf. saliens J. Larsen and Patterson 1990

Colpodella unguis Patterson and Simpson 1996

Cryptaulax cf. marina Throndsen 1969

Discocelis saleuta Vørs 1988

Metopion fluens J. Larsen and Patterson 1990

Metromonas simplex (Griessmann) Larsen and Patterson 1990

Peltomonas volitans Vlk 1942

Phyllomitus granulatus J. Larsen and Patterson 1990

Protaspis gemmifera J. Larsen and Patterson 1990

Protaspis obliqua J. Larsen and Patterson 1990

Protaspis tegere J. Larsen and Patterson 1990

Rhynchomonas nasuta (Stokes 1888) Klebs 1892

DIV. HETEROKONTOPHYTA

Class Raphidophyceae Norris 1982

Order Chattonellales Throndsen 1993

Genus Olisthodiscus N. Carter 1937

Olisthodiscus luteus N. Carter 1937 (non O. luteus sensu Leadbeater 1969)

Plate 1, Fig. (m); Plate 2, Fig. (2)

<u>Description:</u> The cell shape was elliptical. Flagella were inserted subapically, about at one fifth part of the cell length from the apex. The length of the flagella were similar to the body length. It contained 6-12 yellow-brownish chloroplasts on the lateral edge, leaving space to the nucleus in the mid-front of the cell. Chloroplasts were never placed on the ventral or the dorsal edges.

<u>Size:</u> They varied in size, ranging from 16 to 24 μ m in length and 8 μ m to 12 μ m in width. <u>Observation</u>: The cells were always observed in contact with the substratum, never swimming freely. They were observed in both localities and they could at times be extremely abundant, especially from April and on.

Comments: The species found and identified in this study matched with those described by Carter (1937) who named it *Olisthodiscus luteus*. It was for many years confused with the planktonic *Heterosigma akashiwo* (Hada) Hada ex Hara & Chihara (1987) and was wrongly reported from coastal waters in many countries (see e.g. Hara *et al.*1985). The taxon *Olisthodiscus carterae* Hulburt (1965) is also assumed to be a synonym for *Heterosigma akashiwo* (Hara & Chihara 1987). The first fine structure study ascribed to *O. luteus* (Leadbeater 1969) was also unfortunately based on *H. akashiwo*. The known distribution of *O. luteus* N. Carter is restricted to tidal mud or sandy beaches in Europe and Japan. Grimsrud (2001) recorded it in Hulvika in May.

Class Dictyochophyceae (P.C. Silva 1980) Silva 1982 Order Pedinellales Zimmermann, Moestrup & Hällfors 1984 Genus Actinomonas Kent 1880 / Pteridomonas Penard 1890

Actinomonas mirabilis Kent 1880/Pteridomonas danica Larsen& Patterson 1990 Plate 1, Fig. (n)

Description: cells were colourless, round and had a single thick flagellum emerging from a small depression in the anterior part of the cell. It contained a ring of numerous tentacles inserted around the flagellum, a few emerging from the posterior part, and a ca. 15 μ m long stalk as a posterior projection. The anterior part of the cell was slightly broader than the posterior part. The cell could drag particles attached to the trailing pseudopodium.

Size: 4- 6 µm in diameter.

<u>Observation</u>: They were observed a few times in Huk and always in adjunction to the substratum (only seen on cover slips), occasionally trailing on its long posterior stalk.

<u>Comments</u>: The flagellum had an undulating beat and this made it easy to notice.

Since *Actinomonas mirabilis* is indistinguishable from *Pteridomonas danica* in the light microscope, both species were named together as only one observation.

DIV. HAPTOPHYTA

Class Prymnesiophyceae Hibberd 1976 Order Isochrysidales Pascher 1910

Genus Dicrateria M. Parke 1949

Dicrateria gilva M. Parke 1949

Plate 1, Fig. (g); Plate 2, Fig. (5)

<u>Description</u>: Round cells with a non external or emergent haptonema. Two homodynamic flagella $25 \mu m$ long. It contained two chloroplasts on the sides of the cell and a big central particle that may have been chrysolaminaran.

Size: 11 µm in diameter.

Observation: It was observed a few times and only in February and March in Huk.

<u>Comments</u>: This species has been reported from plankton (Throndsen 1997), and since it was observed a few times only, it may well be that it came with the free water together with the sand and that it was not strictly a psammobic species.

Order Prymnesiales Papenfuss 1955

Genus Prymnesium Massart & Conrad 1926

Prymnesium nemamethecum Pienaar & Birkhead 1994

Plate 1, Fig. (h); Plate 2, Fig. (6)

<u>Description</u>: Cell shape was oblong to round with distinctive brown-yellow chloroplast. It had two sub-equal flagella which swung around the cell when swimming. One was longer than the other and both moved showing undulating movements. The haptonema appeared thickened due to the organic scales on its surface, measured about 3-4 μ m long and was used to attach to the substrate. Most of the times they appeared anchored to the coverslip and it was only possible to see their posterior part pointing away from the substratum.

Size: $10-16 \mu m \log and 6-10 \mu m \text{ wide}$.

<u>Observation</u>: This species was very abundant in Huk in the beginning of the study (February and March). It was also observed later in the spring and beginning of summer but never in such high numbers. It was also present in Hulvika.

<u>Comments</u>: *Prymnesium* species are hard to tell apart unless one brings their scales into observation in the electron microscope. Similar specimens of *Prymnesium* have previously been observed in sand from Hulvika (Grimsrud 2001), the identity was then verified by EM observations on the scale structure.

DIV. CRYPTOPHYTA

Class Cryptophyceae Fritsch 1927

Order Cryptomonadales Engler 1903

Genus Chroomonas Hansgirg 1885

Chroomonas diploccoca Butcher 1959

Plate 1, Fig. (a), (b); Plate 2, Fig. (1); Plate 18, Fig. (1), (2)

Description: Blue-green elliptical cells, dorsally convex and slightly flat ventrally. In the anterior part, it showed a long visible tubular gullet from which both flagella emerged with a length of ca 7-8 μ m. Two very conspicuous refractive bodies were located in the lower part of the cell body. They appeared as two coupled rings. The two rows of ejectisomes were easy to notice but they appeared to the eye as a single one.

Size: 9-11 μm long and 6-7 μm wide.

Observation: They were observed in big amounts in May and June and only in Huk.

<u>Comments</u>: Butcher found this species in salt-marsh lagoons in England, and the present study shows that it also inhabit the sand interstices.

Genus Cryptomonas Ehrenberg 1832

Cryptomonas sp.

Plate 1, Fig. (c); Plate 2, Fig. (2) Plate 18, Fig. (1), (2)

<u>Description</u>: Elongated, ovoid and dorsiventrally compressed cells with yellow brown chloroplasts. Two or three rows of ejectisomes were visible in the furrow area, not clear. Two flagella of ca. 10-12 μ m emerged from the gullet, a little bit to the right side of the vestibulum. They were very fast swimmers.

<u>Size</u>: 22-25 μm long and 10-12 μm wide.

Observation: It was observed only in Huk but several times.

<u>Comments</u>: *Cryptomonas* has usually been reported as freshwater genus. The samples from the present study were collected in a marine environment, probably very influenced by the nearby freshwater stream. This fact may be the reason why the present taxon was encountered there.

Class Goniomonadeae Cavalier-Smith 1993 (= Cyathomonadea Cavalier-Smith)

Order Goniomonadales Novarino & Lucas 1993

Genus Goniomonas Stein 1878

Goniomonas amphinema J. Larsen and Patterson 1990

Plate 1, Fig. (f); Plate 2, Fig. (3)

<u>Description</u>: Cells were round to oblong ventrally but a little flattened laterally. Two flagella inserted in an anterior lateral pocket, one directed anteriorly, one posteriorly. The two flagella were unequal in length, the longer one trailed over the body and was slightly longer than the cell. The shorter flagellum pointed forward. Sometimes several longitudinal ridges could be noticed but they were not as conspicuous as in *G. pacifica*. A transverse row of ejectisomes was relatively easy to see because it refracted the light.

<u>Size</u>: 5-7 μm.

<u>Observation</u>: It was found in both localities and in almost all of the samples throughout the present study.

<u>Comments</u>: *G. amphinema* resembles *G. pacifica* very much, therefore it is hard to tell them apart in the light microscope. The smaller size and shorter flagella distinguish *G. amphinema* from the other.

Goniomonas pacifica J. Larsen and Patterson 1990

Plate 1, Fig. (d) (f); Plate 2, Fig. (4)

<u>Description</u>: Colourless and rounded cells with lateral compression, truncate anteriorly and rounded posteriorly. Cells with several distinctive longitudinal ridges on the surface and a clear transverse row of ejectisomes. Two flagella of equal length emerged from a small anterior depression and were directed anteriorly. When swimming the flagella diverged.

<u>Size</u>: 8-10 μm.

<u>Observation</u>: This species was observed in both localities and in almost all of the samples all through the study period.

<u>Comments</u>: *G. pacifica* could be distinguished from *G. amphinema*, by size and flagellar length, where as in *G. pacifica* the flagella have equal length and are directed forward.

Plate 1. Cryptophyta, Haptophyta, Chlorophyta, Heterokontophyta, Choanoflagellata

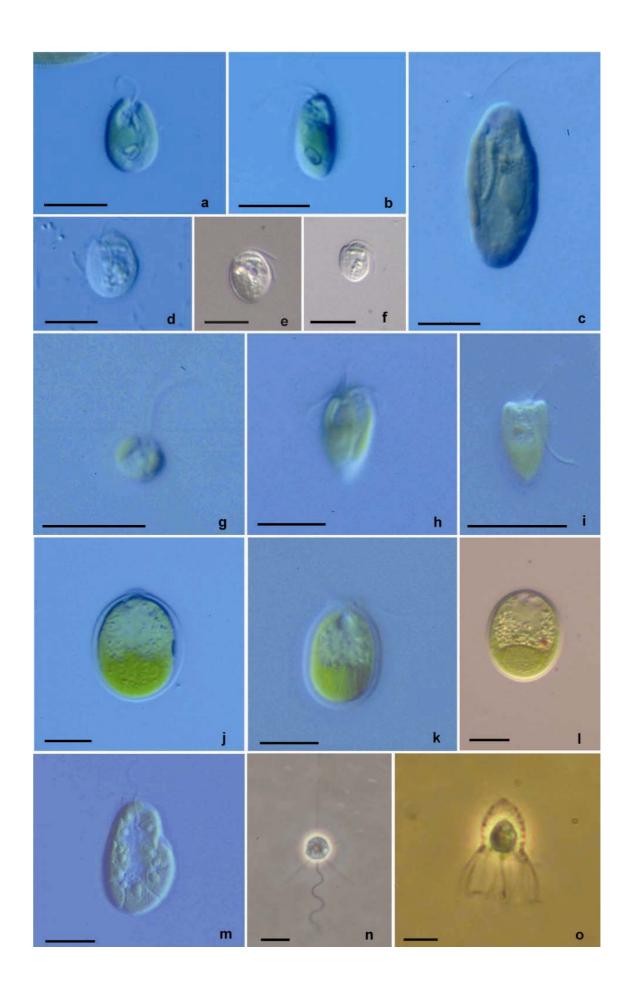
Scale bar= 10 µm

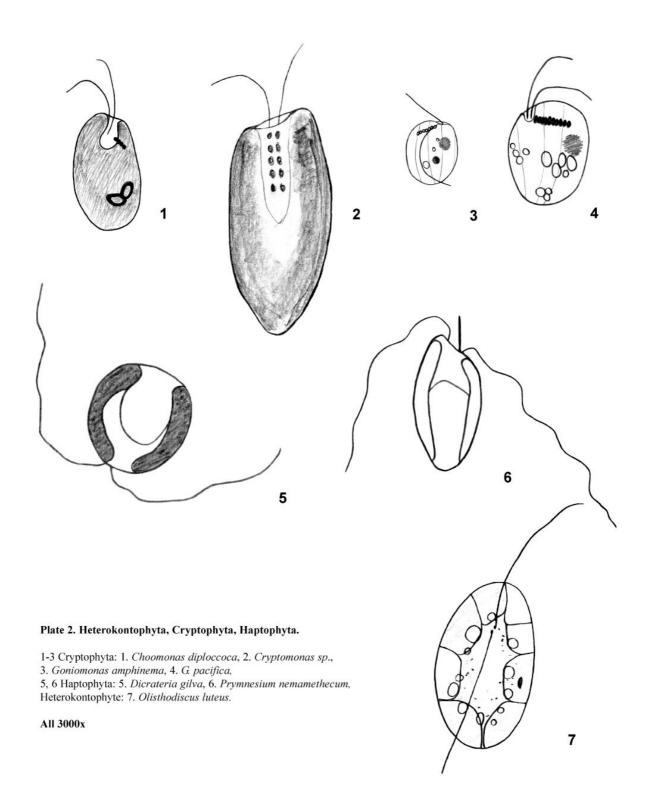
⁽a) (b) Chroomonas diploccoca, (c) Cryptomonas sp., (d) (e) Goniomonas pacifica, (f) G. amphinema,

⁽g) Dicrateria gilva, (h) Prymnesium nemamethecum,

⁽i) Pyramimonas sp.1, (j) Chlamydoimonas cf. nonpulsata, (k) Ch. sp., (l) Ch. sp.

⁽m) Olisthodiscus luteus, (n) Actinomonas mirabilis / Pteridomonas danica, Choanoflagellata: (o) Acanthocorbis sp.





DIV. DINOPHYTA

Class Dinophyceae Fritsch 1929= Dinoflagellida Bütschli 1885 Order Gymnodiniales Lemmermann 1910

Genus Amphidinium Claparède & Lachmann 1859

Amphidinium bipes Herdman 1924

Plate 3, Fig. (a); Plate 4, Fig. (1); Plate 17, Fig. (1), (2), (3)

<u>Description</u>: Colourless cell, with a typical *Amphidinium* epicone but more reduced. Dorsiventrally flattened and with a deep invagination in the posterior part where the sulcus began, creating an effect of two legs. Always swam in close connection to the substrate trailing on its longitudinal flagellum.

Size: 30 µm long µm 24 wide.

<u>Observation</u>: It was only observed in Huk and few times spread all along the study period and in few numbers.

<u>Comments</u>: Could readily be identified due to its size and the shape formed by the two conspicuous "legs". It has been found with and without plastids, and the ones with chloroplasts appear to be more abundant in sandy beaches. However, in this study only colourless individuals have been observed.

Amphidinium britannicum (Herdman) Lebour 1925 = Togula britannica, T. compacta, T. jolla (Herdman) Flø Jørgensen, S. Murray & Daugbjerg 2004 Plate 3, Fig. (b), (c); Plate 4, Fig. (2)

<u>Description</u>: Dorsiventrally flattened cells with many yellow-brown chloroplasts and a strongly descending cingulum which made the epicone asymmetrical. Cells varied a lot in size and shape, ranging from ovate to elongated, having both apex and antapex rounded. The furrow system was well visible, where the transversal one had a great displacement. The longitudinal furrow traversed all the way down to the antapex. Sometimes red inclusions were visible in the centre of the cell. Most cells were still, but few a swam for a while in a spiral movement and then the longitudinal flagellum could be easily discerned. The chloroplasts were elongated and numerous, and were situated radially. The nucleus was placed in the middle.

<u>Size</u>: 28-38 μm long, 22-32 wide.

<u>Observation</u>: They were present in both localities starting from around March until June. They were observed in tens of them and sometimes motile cells appeared surrounded by a hyaline layer, as temporary cysts.

<u>Comments</u>: Amphidinium britannicum has recently been studied by morphological and molecular methods by Flø Jørgensen et al. (2004) who revealed a species complex consisting of three species included in a new genus called *Togula* Flø Jørgensen, S. Murray & N. Daugbjerg. The genus *Amphidinium* was emended and now only consists of species

with minute left-deflected epicone, which has excluded from this genus what it was considered *A. britannicum* before. Flø Jørgensen *et. al* erected the new genus *Togula* and described three different species that were before included as one: *T. britannica*, *T. compacta* and *T. jolla*. The paper came to our knowledge too late to be applied in the identification work, and hence the records are all referred to *A. britannicum* sensu Lebour (1925). The variation in size observed in the present material indicate, that more than one species of *Togula* may have been present, but according to Flø Jørgensen (pers. com.) the illustrated specimens belong either to *T. compacta* or *T. jolla*.

Amphidinium cf. carterae Hulburt 1957

Plate 3, Fig. (d); Plate 4, Fig. (5)

<u>Description</u>: Cells oval, flattened, slightly tapered posterioly. Small epicone with a little deflection to the left. Several green-yellow chloroplasts or probably a single reticulate one with multiple lobes placed around a central pyrenoid. Nucleus in the hypocone.

Size: 22 µm long.

Observation: It was observed once in the beginning of May in Hulvika.

<u>Comments</u>: *A. carterae* and *A. operculatum* are similar species with main differences in size and chloroplast arrangement. In the light microscope, it is difficult to see the plastids, therefore the main characteristic that can tell them apart is size, where *A. carterae* is slightly smaller than *A. operculatum*. This cell was identified first as *A. klebsii* sensu Carter (1937) a species which later was named *A. carterae* Hulburt (1957) because its size suited better these two descriptions than that of *A. operculatum*.

Amphidinium herdmanii Kofoid and Swezy 1921

Plate 3, Fig. (f); Plate 4, Fig. (6)

<u>Description</u>: Cells were ovoid to somewhat rectangular. The epicone was reduced and it was broad and the biggest part was included in the hypocone. The furrow system had an Y shape on the ventral side. Brown-yellow chloroplasts radiating from the centre. Nucleus was very big and it was placed in the posterior part, occupying most part of the hypocone. The sulcus opened all the way down to the antapex in a wide base giving a sense of protuberances on both sides of the furrow. This protrusions are much smaller than in *A. bipes*, in which they resulted in a two leg-type structure.

Size: 25-27 µm long and 20-22 µm wide

<u>Observation</u>: Cells of *A. herdmanii* were observed in low numbers in both localities, but the species was more common in Huk. It appeared from February until June.

<u>Comments</u>: It is an easily identifiable species due to its characteristic shape and sulcus opening posteriorly.

Amphidinium pellucidum Kofoid and Swezy 1921 (= Gymnodinium venator Flø Jørgensen & Murray 2004)

Plate 3, Fig. (j), (k); Plate 4, Fig. (8)

<u>Description</u>: Cells oval from the ventral side, more rounded apex and a little more square antapex. Dorso-ventrally flattened. Reduced epicone; ca ¼ of the cell length. Heterotrophic cells with yellow-orange-brown coloured inclusions in the posterior part of the cytoplasm. Nucleus located anteriorly. These cells were very active, they prowled among other flagellates and diatoms.

Size: 26-32 µm long and 18-22 µm wide.

Observation: observed only in Huk in June.

<u>Comments:</u> Most probably belonged to the plankton based on its rare occurrence in the samples and its free swimming style. Recently changed name to *Gymnodinium venator*. Like many other species, this one was previously included within *Amphidinium* and since it did not any longer fit this genus' new definition, it needed be revised and replaced. It has been still included within the rest of the *Amphidinium* species in the present study, because by the time we got to know about this new name, all the plates and descriptions were already organized and placed.

Amphidinium poecilochroum J. Larsen 1985

Plate 3, Fig. (1); Plate 4, Fig. (9)

<u>Description</u>: coloured and oval cell, rounded posteriorly; with a strongly reduced epicone, and narrower than the hypocone which gave the cell an impression of having a round hypocone. It contained a few yellow-green inclusions and a single big chloroplast in the central part of the cell. The longitudinal furrow was evident.

Size: 18 µm long and 13 µm wide.

Observation: Observed once in Huk in May.

Amphidinium psittacus J. Larsen 1985

Plate 3, Fig. (m); Plate 4, Fig. (4)

<u>Description</u>: This cell was elongated, with a length to width ratio of 2.0, and asymmetrical with the cingulum descending. Epicone strongly reduced; took 1/3 to ½ of the total cell length. The cell was heterotrophic and there were no visible inclusions.

Size: 11 µm long and 5-6 µm wide.

Observation: Only observed once in Huk in February.

<u>Comments</u>: Only a single cell was observed but it was an easy one to identify due to its small size, shape and because it differs well from the rest of the *Amphidinium*.

Amphidinium testudo Herdman 1924

Plate 3, Fig. (i); Plate 4, Fig. (10)

<u>Description</u>: Oval cells which lost their shape quickly when they fastened tightly to the substrate. They were ventrally flattened. The epicone was strongly reduced. There was a circumferential groove running around the anterior part of the cell, and a second one along the length of the body. They contained many green-brownish chloroplasts that seemed to be located around the centre of the cell. This species was most commonly observed attached to the substratum, only a few times was it seen swimming freely.

<u>Size</u>: 34-42 μm long and 24-30 μm wide.

Observation: It was observed regularly in both localities in spring time.

<u>Comments</u>: A. testudo was easy to identify but could also be easily mistaken for A. corrugatum Larsen & Patterson 1990 because they both have the same appearance except that A. testudo is slightly smaller and does not have longitudinal surface ridges on the dorsal side and the little crack on the epicone as A. corrugatum does.

Amphidinium cf. trulla Jørgensen & Murray 2004 (= A. operculatum Claparède & Lachmann 1858)

Plate 3, Fig. (e); Plate 4, Fig. (7)

<u>Description</u>: Cell were oval to round. A small epicone protruded from the hypocone. Flattened dorsiventrally and with greenish-yellow chloroplasts arranged radially around a central pyrenoid. Nucleus located posteriorly.

<u>Size</u>: 24-28 μm long and 18-20 μm wide

Observation: In both localities, few cells in all samples, April to June, not in the winter.

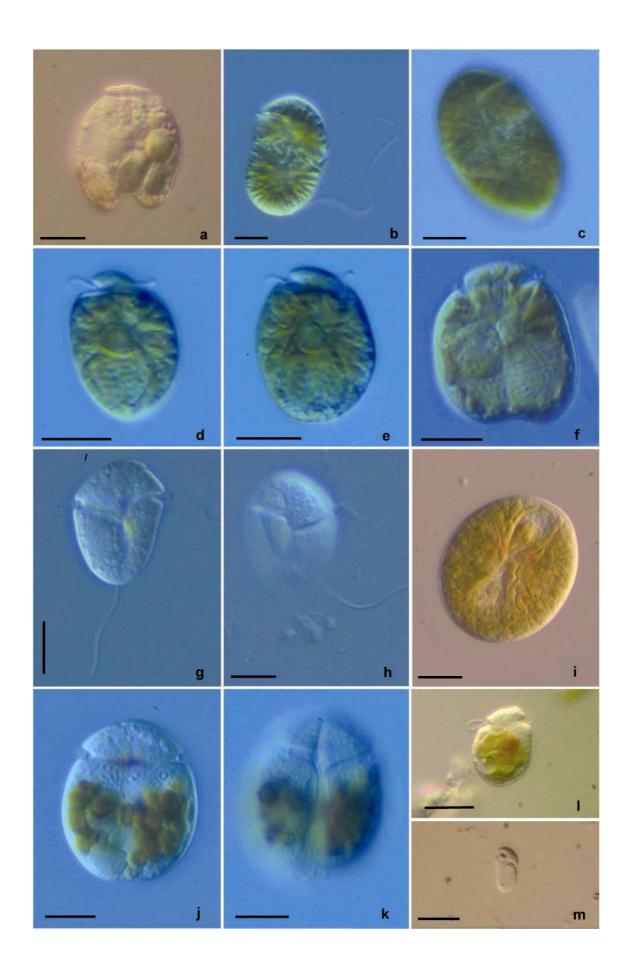
<u>Comments</u>: As mentioned before, this species is very similar to *A. carterae* Hulburt, except that *A.* cf. *trulla* had a bigger size and the plastids placed radially around the central pyrenoid. This was observed many times and therefore we concluded in *A.* cf. *trulla*.

Amphidinium trulla, as well as A. massartii Jørgensen & Murray 2004, have recently been separated and renamed as two different taxa from the previously described A. operculatum Claparède & Lachmann, 1858. The two new species are difficult to distinguish but A. trulla is a more common species in Kattegat and Skagerrak (Mårten Flø Jørgensen and Jahn Throndsen, pers. com.) than is A. massartii, thus we doubtfully concluded in A. trulla.

Plate 3. Amphidinium (including the newly renamed Togula, Chilodinium cruciatum and Gymnodinium venator).

(a) Amphidinium. bipes, (b), (c) Togula compacta or T. jolla (=A. britannicum) from the ventral and lateral sides, respectively, (d) A cf. carterae, (e) A. trulla (=A. operculatum), (f) A. herdmanii, (g), (h) Chilodinium cruciatum, ventral outline and scheme of the furrow system, in specular view, (i) A. testudo, (j), (k) A. pellucidum (=Gymnodinium venator) and furrow system, respectively; sulcus becomes less distinct as it approaches the posterior end, (l) A. poecilochroum, (m) A. psittacus.

Scale bar= 10 µm



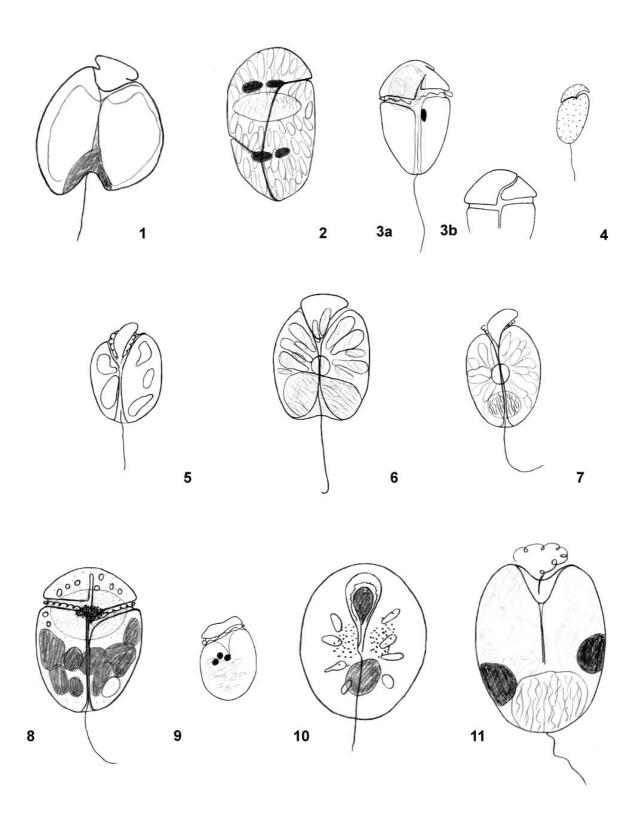


Plate 4. Dinoflagellates: Amphidinium (other renamed taxa included before in the Amphidinium genus) and Prorocentrum

Amphidinium: 1. A. bipes, 2. A. britannicum (=Togula compacta or T. jolla), 4. A. psittacus, 5. A. cf. carterae, 6. A. herdmanii, 7. A. cf. trulla (=A. operculatum), 8. Gymnodinium venator (=A. pellucidum), 9. A. poecilochroum, 10. A. testudo. Renamed taxa included before in Amphidinium: 3a, 3b. Chilodinium cruciatum (=G. cruciatum) general cell outline and furrow system, Prorocentrum: 11. P. cf. lima (= Exuviaella marina).

all 1500X

Genus Glenodinium (Ehrenberg) Stein 1883

Glenodinium cf. foliaceum sensu Conrad et Kufferath

Plate 5, Fig. (a); Plate 6, Fig. (1)

<u>Description</u>: Dorsiventrally flattened and ventrally rounded cell with the apex somewhat pointed. Epicone with triangular shape and took 50% of the whole cell length. Cingulum in the middle of the cell and had no displacement. Sulcus was visible but it seemed to be more indistinctive when it approached the antapex. It had ca 20 green-yellow chloroplasts organized around a central nucleus. A red triangular shaped eyespot was very visible at the mid-sulcal area. The longitudinal fagellum was ca 1.5 times the body length. A thin cell wall could hardly be distinguished around the cytoplasm. It was a very active swimmer.

Size: 24 µm long and 18 µm wide.

Observation: only a single cell was observed in Hulvika at the end of June.

<u>Comments</u>: This species was difficult to find in the literature on sandy or muddy shores. A very similar species was identified in Conrad and Kufferath 1954, a study on shallow waters. The rareness of its occurrence in the sand and its fast swimming behaviour may indicate that it came with the free waters collected together with the sand and that it did not indeed belong to the sand environment.

Genus Chilodinium Massart 1920

Chilodinium cruciatum (Massart) Massart 1920 (= Gymnodinium cruciatum Massart 1900-1901, Chilodinium cruciatum Massart in Conrad 1926, non Gymnodinium cruciatum Thompson 1950

Plate 3, Fig. (g), (h); Plate 4, Fig. (3)

<u>Description</u>: Colourless, ellipsoidal cell, more rounded anteriorly, tapering posteriorly and with a conspicuous orange eyespot in the hypocone. The transversal furrow, $2 \mu m$ broad and displaced about the same width as the furrow; the longitudinal one extended to the apex as a sigmoidal apical groove creating a little notch on the left side of the cell. Sulcus ran from posterior end to 2/5 from anterior end. Slightly rough surface. Nucleus in the anterior part. Swimming actively.

Size: 25 µm long and 15 µm wide.

Observation: Only observed once in the end of June in Hulvika.

Comments: Its special features could make it easy to tell it apart from other species, but it was difficult to find any description that would fit such a cell. In Conrad's study from 1926, the species was larger and wider, $40\text{-}50~\mu\text{m} \times 30\text{-}40~\mu\text{m}$, than the cell found in my sample. However, according to the cell outline from his illustrations (see Fig. 17), it shows with no doubt that it is the same organism.

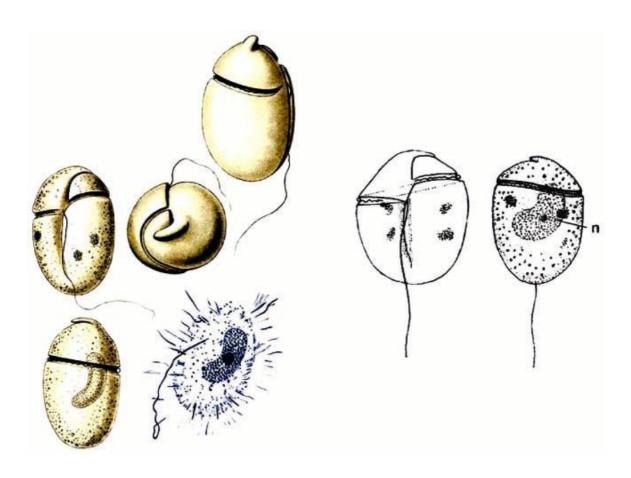


Fig. 17 Illustrations of Chilodinium cruciatum (Massart) Massart from Conrad 1926 and Massart 1920.

Genus Gymnodinium Stein 1883

Gymnodinium cf. danicans Campbell 1973

Plate 5, Fig. (m); Plate 6, Fig. (6)

Description: Non-coloured, rounded cell with the epicone slightly pointed. The cingulum was in the middle of the cell and had a little displacement which left the right side of it at a higher level. Sulcus broad, and ran markedly all the way down to the antapex. No plastids and no nucleus were visible. They were many of them swimming all around at high speed. Size: 15 μm long and 13μm wide.

Observation: It was observed in Hulvika in great numbers through all the study period. Comments: It was very difficult to find a non-swimming cell, therefore the few details observed were drawn from the photographs taken of them. According to work reported by M. Hoppenrath, from the Alfred Wegener Institute of Polar and Marine Research, http://www.awi-bremerhaven.de/Benthic/CoastalEco/list of species/Benthic microflora/dinoflagellates.html, a G. danicans was recorded from intertidal zones of sandy shores in the Wadden Sea area.

Gymnodinium variabile Herdman 1924

Plate 5, Fig. (f); Plate 6, Fig. (3)

<u>Description</u>: Cell shape could vary from oval to more rounded and it was dorsiventrally flattened. The epicone was slightly larger than the hypocone and ended more pointed than the hypocone which was completely rounded. Epicone and hypocone of same width. The cingulum was situated a little below the centre of the cell and with a slight displacement. The sulcus was less pronounced but extended all the way down to the antapex. Many elongated yellow-brown chloroplasts were distributed through all the cytoplasm. It contained red coloured refractive spots in the hypocone. The nucleus was $16~\mu m$ in diameter and occupied the anterior part, the epicone. It was commonly seen surrounded by a mucilage layer. Only immobile individuals were observed, never swimming cells.

Size: 38 µm long and 30 µm wide.

Observation: It was observed twice in Hulvika in February and March and in small numbers.

<u>Comments</u>: Even though it was variable in shape, the organization of its chloroplasts, the red refractive bodies and its passivity made it easy to identify.

Gymnodinium sp.

Plate 5, Fig. (b), (c); Plate 6, Fig. (2)

<u>Description</u>: Cell oval to rhomboidal with rounded apex and antapex. Slightly flattened dorsiventrally. Epicone was dome-shaped. The sulcus produced a slight invagination in the posterior end of the hypocone. Epicone equal to the hypocone in length, both with same width. The furrow system was without or with a slight displacement only. Both longitudinal and transversal furrows were narrow, about 2 μ m wide. The epicone surface was corrugated, with around 5 longitudinal ridges, parallel to each other (see photo, plate 5, Fig. (c)). Spherical nucleus 10 μ m wide, situated anteriorly, occupied a great part of the epicone, Yellow-brown chloroplasts distributed randomly in the cytoplasm, specially in the hypocone. Two oval red refractive eyespots below the cingulum. Active swimmer.

Size: 28 µm long and 18 µm wide.

Observation: Observed only once in Hulvika, end of June.

<u>Comments</u>: The present cell did not look like any other *Gymnodinium* observed during the sampling period of the current study. It was identified down to *Gymnodinium* genus based on its straight furrow system.

Genus Gyrodinium Kofoid & Swezy 1921

Gyrodinium cf. estuariale Hulburt 1957

Plate 5, Fig. (g), (h), (i)

Description: Cells were oval to round ventrally. The grooves were very broad, sulcus approximately 2-3 μm and cingulum 2 μm . The transversal furrow had a displacement of 2 μm , same width as the cingulum itself. They contain no chloroplasts. Some cells were filled with food inclusions. They were fast swimmers.

Size: 12-15 µm long and 10-12 µm wide.

Observation: It was observed several times in Hulvika in spring and beginning of summer.

<u>Comments</u>: *Gyrodinium estuariale* usually contains chloroplasts but the cells observed were completely colourless except for some orange food particles in some individuals which gave them some colour. This species has been reported from planktonic (Wadden Sea, M. Hoppenrath, web information from Alfred Wegener Institute, Helgoland) and benthic (Botany Bay, NW Australia, M. Hoppenrath and S. Murray; web information from Marine Biological Laboratory, Woods Hole) habitats.

Gyrodinium cf. lebourae Herdman 1924

Plate 5, Fig. (n); Plate 6, Fig. (7); Plate 17, Fig. (7)

<u>Description</u>: Small dinoflagellate, oval from the ventral side and slightly dorsiventrally flattened. Cingulum was strongly displaced, beginning approximately 0.3 of the cell length from the apex, rising initially, then falling across the dorsal side of the cell, ending approximately 0.6 of the cell length from the apex. Seen from the ventral side it adopted an S shape. Both cingulum and sulcus were broad and measured ca. 3.5 μm. Epicone protrudes slightly from the hypocone. It had no chloroplasts and the nucleus was not visible. It possessed an elongated red-orange eyespot situated at the beginning of the sulcus, around 5 μm long. They were very fast swimmers.

Size: 10-14 µm long and 7-10 µm wide.

Observation: It was observed only in Hulvika in great numbers from around April and on.

<u>Comments</u>: They were very hard to study because they were very active swimmers. When they stayed still it took short time before they lost their shape and collapsed.

Gyrodinium resplendens Hulburt 1957

Plate 5, Fig. (d), (e); Plate 6, Fig. (4a), (4b)

<u>Description</u>: Naked dinoflagellate of oval to rhomboidal shape. Girdle displaced three times its width or 1/3 body length, was straight and had a distinctive extension of the groove towards the apex (apical groove) which curled somewhat. The furrows were narrow, 2-3 μ m wide, and they barely broadened in their distal part. Many elliptical shaped yellow-brown chloroplasts organized around the central part. Nucleus situated posteriorly.

Size: 40-42 µm long and 32-34 µm wide.

Observation: Only observed in one sample from Hulvika around the end of May.

<u>Comments</u>: It was easy to identify due to its pronounced apical groove. It only occurred once and other studies in benthic flagellates do not show this species, therefore it most probably came with the free waters that mixed with the sand at sampling.

Gyrodinium uncatenum Hulburt 1957

Plate 5, Fig. (j), (k); Plate 6, Fig. (5a), (5b)

<u>Description</u>: Oval shape in all its profiles. Strongly displaced cingulum which curled ventrally. Sulcus was curled as well. Both furrows were narrow, not broader than 2.5 μ m. The furrow system drew a characteristic outline (see drawings, Plate 6, Fig. 5a and 5b). Several elliptical dark yellow-brown chloroplasts arranged around the centre of the cell. Nucleus was not noticed.

Size: 34-42 µm long and 24-30 µm wide.

Observation: It was only observed once in Hulvika in May.

<u>Comments</u>: *G. uncatenum* was slightly larger than *G. resplendens* and lacked a pronounced apical furrow. It was readily identified due to its characteristic displaced girdle that could be seen in all views.

Genus Katodinium Fott 1957

Katodinium cf. fungiforme (Assimova 1926) Loeblich 1965

Plate Z, Fig. (y), (x); Plate 6, Fig. (8)

<u>Description</u>: Ovoid cell from the ventral side and slightly flattened dorsiventrally. No thecal plates could be observed. The hypocone was smaller than the epicone, in the ratio of 0.3-0.4. The epicone very rounded and without any visible notch. Cingulum was 2 μ m wide and sulcus a little narrower than that. Nucleus was located in the hypocone and was ca. 5 μ m diameter. Chloroplasts were absent; it contained small colourless vesicles as well as orange reflective food inclusions. Cells were fast swimming.

Size: 15 µm long and 12 µm wide.

Observation: Observed once in Hulvika in May. Only found in that sample.

<u>Comments</u>: *Katodinium fungiforme* and *K. asymmetricum* (Massart 1920) Loeblich 1965 are distinguished by size and the apical notch in the latter one. In the cell found in this study, no notch could be observed and therefore it was concluded that it could probably be *K. fungiforme*. The size, however, was more close to that of *K. asymmetricum*.

K. glandula (Herdman 1934) Loeblich 1965 was excluded because this species is much smaller and has a narrower hypocone.

Katodinium glandula (Herdman 1924) Loeblich 1965

Plate 5, Fig. (1); Plate 6, Fig. (9)

Description: Ventrally ovoid and dorsiventrally slightly flattened. A general globular appearance. The hypocone was almost as wide as the epicone but the latter one was longer, a little less than 2/3 of the total cell length. The epicone was dome-shaped and possessed a distinctive notch to the left of the apex. The cingulum was straight and very narrow, ca 1.5 μm wide. On the ventral side, in the middle, the cingulum drew a concavity in the epicone margin of it, that is, the cingulum broadened itself breaking its straight path across the ventral side (see drawing, Plate 6, Fig. 9). There was not a marked sulcus, only a trace of a sulcus was visible in the anterior part of the hypocone at the cingular level, just where the above mentioned wide area was drawn. Chloroplasts were absent; food particles occasionally present. The nucleus was in the anterior part, and occupied great part of the epicone, ca. 8 μm diameter. It swam quite fast and often rolled around itself.

Size: 22-24 µm long and 16-18 µm wide.

Observation: Observed in Huk, only as solitary cells, in January and in June.

<u>Comments</u>: *Katodinium glandula* is characterised by an apical notch and its motion style.

Order Prorocentrales

Genus Prorocentrum Dodge 1975

Prorocentrum cf. lima (Ehrenberg) Dodge 1975

Fig. 18; Plate 4, Fig. (11)

<u>Description</u>: Armoured photosynthetic species, composed with two concave valves of smooth surface. Cells ovate and more or less compressed (14-18 µm) in lateral view. Flagella located in the anterior end, in a shallow V-shaped depression. One longitudinal flagellum and another transversal which could be seen undulating in a circle off the apex.

Yellow-brown plastids, posterior nucleus and a big vacuole anteriorly. It was a very slow swimmer.

Size: 36-44 µm long and 26-34 µm wide.

Observation: In both localities, April and on. Many cells. Comments: This is most probably the taxon described first by Cienkowski 1881 who identified it as *Exuviaella marina* and later it has changed name several times: *Exuviaella lima* (Ehrenberg) Bütschli 1885 *Prorocentrum marinum* Dodge et Bibby vide Dodge 1982, *Exuviaella marina* var *lima* (Ehrenberg) Schiller 1933. The most recent name is *Prorocentrum lima*. There are slight differences among the benthic *Prorocentrum* species, and therefore I could not be certain about the exact taxon.

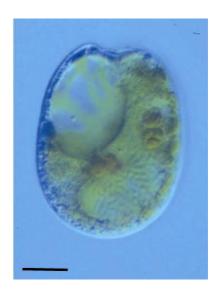
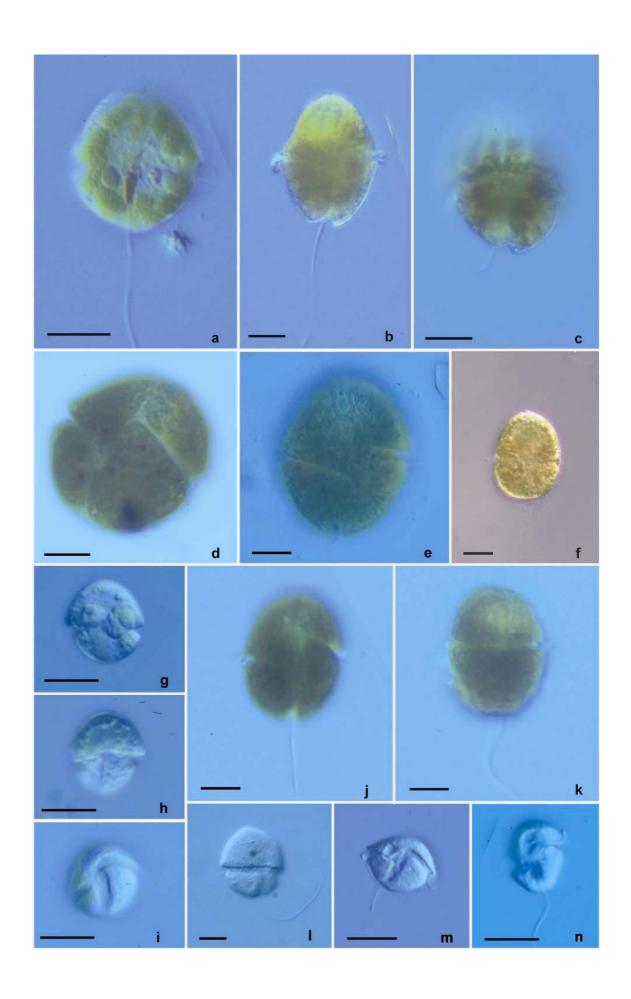
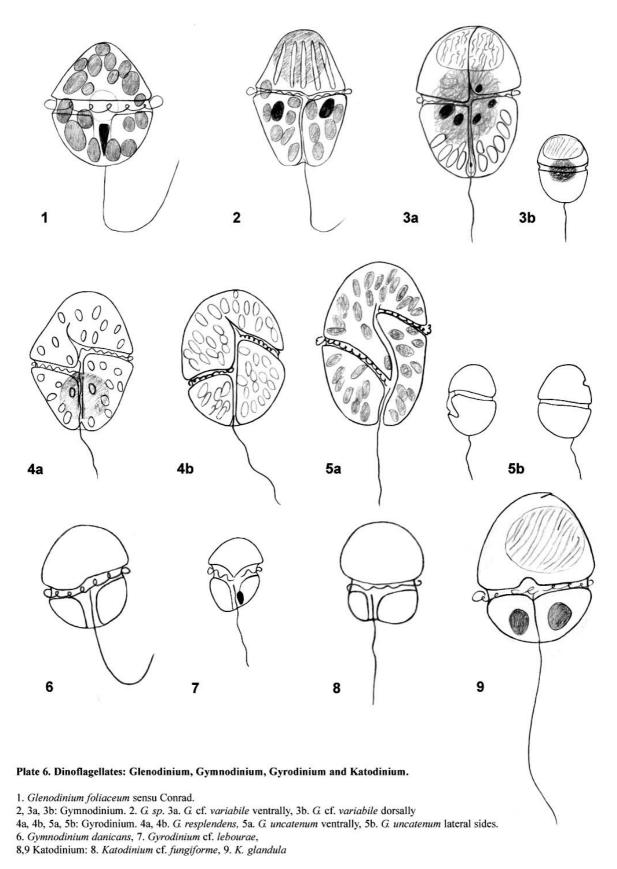


Fig. 18 Prorocentrum cf. lima

Plate 5. Dinoflagellates (a) Glenodinium cf. foliaceum, (b), (c) Gymnodinium sp., (d) (e) Gyrodinium resplendens, (f) Gymnodinium variabile, (g), (h), (i) Gyrodinium cf. estuarile, (j) Gyrodinium uncatenum, ventrally (k) G. uncatenum laterally, (l) Katodinium glandula, (m) Gymnodinium cf danicans, (n) Gyrodinium cf. lebourae.
Scale bar= 10 μm





Glenodinium, Gymnodinium 1500x; Gyrodinium 1000x; smalls and Katodinium 2000x

DIV. EUGLENOPHYTA

Class Euglenophyceae Schoenichen, 1925= Euglenida Bütschli 1884

Order Sphenomonadales Leedale 1967

Genus Anisonema Dujardin 1841

Anisonema cf. acinus Dujardin 1841

Plate 7, Fig. (a); Plate 8, Fig. (1)

<u>Description</u>: Cell outline is like a barley grain, very elongated. Dorsiventrally flattened. Seven to nine distinct longitudinal pellicular stripes on each of the ventral and dorsal sides. No ingestion organelle present. The anterior flagellum was about 1.5 times cell length and beat freely from side to side. The posterior trailing flagellum was 3 times the cell length and its base was thickened. The cells glided smoothly.

Size: 17 µm long and 7 µm wide.

Observation: It was at least observed once in Huk in June.

Comments: This cell's shape and size did not fit well in the descriptions for *Anisonema* acinus which ranged between 22 and 30 μ m, but the number of the pellicular stripes, flagellar lengths and the lack of the ingestion organelle indicated it was most probably *A. acinus*.

Anisonema glaciale J. Larsen and Patterson 1990

Plate 7, Fig. (b); Plate 8, Fig. (3)

<u>Description</u>: Colourless. Cell body oval ventrally, dorsiventrally flattened and with about 7-8 longitudinal surface striae on the ventral side. Anterior flagellum 1.5, and posterior flagellum 3.0 times the cell length. The anterior flagellum with a thickened base. Red refractive bodies antapically. The cells moved forward with a rapid sleek gliding, the anterior flagellum sweeping from side to side.

Size: 25 µm long and 15 µm wide.

Observation: It was observed in Huk in the beginning of June, few individuals.

Anisonema prosgeobium Skuja 1939

Plate 7, Fig. (c); Plate 8, Fig. (2)

<u>Description</u>: The surface was smooth. Anterior flagellum 1.5 and posterior one 3 to 3.5 times the body length. Heterotrophic. Nucleus on the right hand side of the cell and several coloured inclusions present anteriorly and few dispersed in the cytoplasm.

Size: 34µm long and 20 µm wide.

Observation: Observed in Huk in June. Few cells.

<u>Comments</u>: Very similar to *A. acinus* but with no pellicular striation, this is regarded as a distinguishing feature.

Genus Metanema Senn 1900

cf. Metanema strenuum (Skuja 1948) Larsen 1987 (= Anisonema strenuum Skuja 1948)

Plate 7, Fig. (q); Plate 8, Fig. (10)

<u>Description</u>: Cells flattened dorsiventrally and ovate ventrally with pointed anterior and posterior protuberance. Flexible cell. Two equal flagella, slightly longer than the cell; the anterior flagellum directed towards the right and the posterior one to the left, as two arms in opposite direction. With surface striation. No digestion organelle visible. With skidding motion.

Size: 21 µm long and 15 µm wide.

Observation: This cell was most probably seen in both localities throughout the entire work.

<u>Comments</u>: All through the study period, all the cells with the above described appearance were included in the taxon *Heteronema exaratum*. It was not until the end of the work that it came to my knowledge the difference between the two. *Metanema strenuum* should be bigger in size, more rounded and should lack of an ingestion apparatus. Even though I remember having seen two similar cells, one much bigger than the other one, and without an ingestion organelle, now, I can only leave it as cf. *Metanema strenuum*, which is the most common *Metanema* found in benthic habitats.

Plate 7. Anisonema, Dinema, Heteronema, Metanema, Peranema, Ploeotia

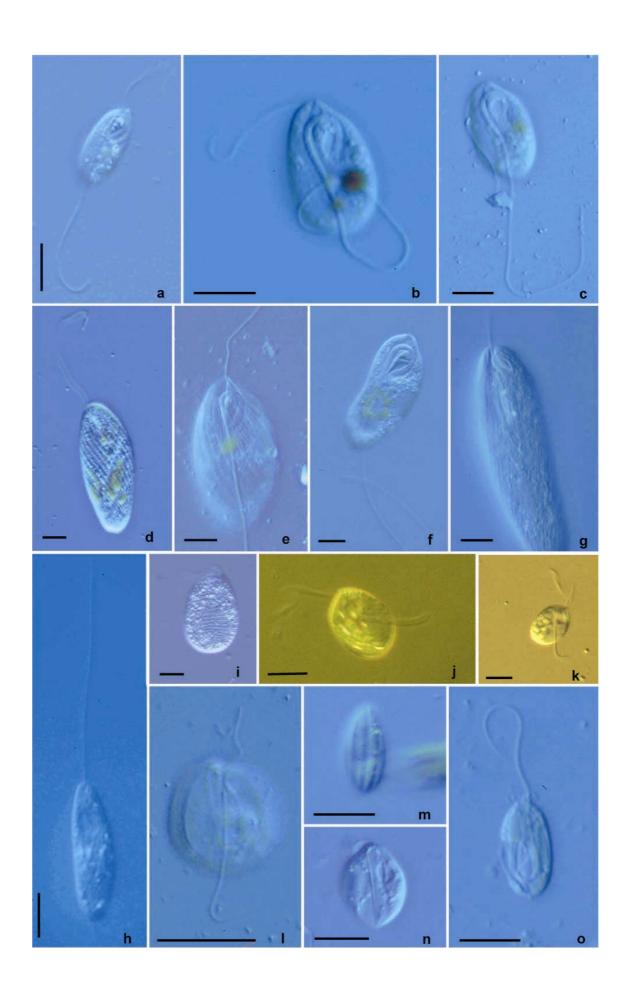
Scale bar=10µm

⁽a) Anisonema cf. acinus, (b) A. glaciale, (c) A. prosgeobium, (d) Dinema litoralis, (e) D. valida pellicular striation, (f) D. valida long posterior flagellum, (g) D. sp.

⁽h) Peranema dolichonema,

⁽i) Heteronema ovale, (j) H.. exaratum (k) cf. Metanema strenuum,

⁽¹⁾ Ploeotia adhaerens, (m) P. corrugata, (n) P. tenuis, (o) P. pseudanisonema.



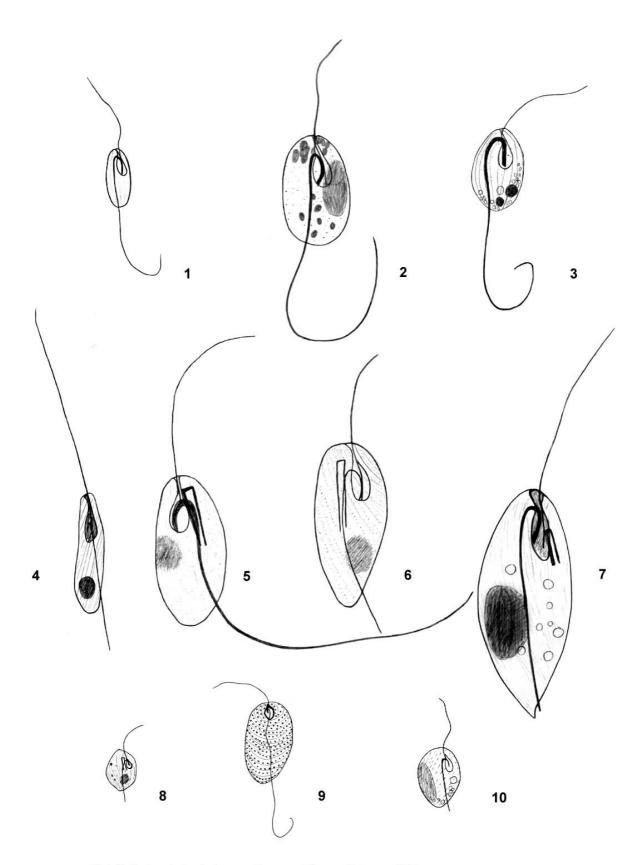


Plate 8. Euglenophytes: Anisonema, Peranema, Dinema, Metanema, Heteronema.

- 1-3, Anisonema: 1. *Anisonema acinus*, 2. *A. prosgeobium*, 3. *A. glaciale* 4. *Peranema dolichonema*, 5,6 Dinema: 5. *D. valida*, 6. *D. litoralis*. 7. *D. sp.*, 8, 9 Heteronema: 8. *H. exaratum*, 9. *H. ovale*, 10. cf. *Metanema strenuum*

all 1000x

Genus Notosolenus Stokes 1884

Notosolenus apocamptus Stokes 1884

Plate 10, Fig. (e); Plate 9, Fig. (1); Plate 13, Fig (1)

<u>Description</u>: Cells oval and flattened. The anterior part of the cell narrowed and posterior end rounded. A deep longitudinal dorsal groove ran along the entire cell. Two flagella emerged from the reservoir, the anterior one was 1-1.5, the posterior one ca. 0.4 times the cell length. Some small vesicles located antapically. As all *Notosolenus* species, it glided with the anterior flagellum extended.

Size: 10 μm long and 6 μm wide.

Observation: It was observed at least once, when identified, in a sample from Hulvika.

<u>Comments</u>: It is very probable that *Notosolenus apocamptus* was present in more samples too, but due to the time consuming efforts required for the species identification in *Notosolenus* as well as *Petalomonas*, not all the encountered specimens of the genera could be given the attention needed for a species identification.

Notosolenus hemicircularis Lee and Patterson 2000

Plate 10, Fig. (f); Plate 9, Fig. (2); Plate 13, Fig. (2)

<u>Description</u>: Elongated cell, pointed posteriorly, anteriorly with a short neck associated with a semicircular collar. Five dorsal keels ran along the cell. Ventrally 3-4 ridges. The two flagella were unequal in length, the anterior one was 1.3 times the cell length and the recurrent posterior one 0.5-0.6 the cell body. Gliding movement.

Size: 12 μm long and 7 μm wide.

Observation: Observed twice in Hulvika in June.

<u>Comments</u>: This species had a peculiar collar which made it easy to identify, and hence not easily overlooked.

Notosolenus urceolatus J. Larsen and Patterson 1990

Plate 10, Fig. (g); Plate 9, Fig. (3); Plate 13, Fig (3)

<u>Description</u>: Cell outline was broad posteriorly and narrower anteriorly with a small neck around the flagellar canal. Three dorsal keels, three ventral ridges and 2 lateral. Two flagella of unequal length, anterior one was slightly longer than the cell body, the posterior flagellum 0.7 times the cell length. The nucleus laid in the central left side. Gliding motion with anterior flagellum directed forward.

Size: 9-15 μm long and 7-11 μm wide.

Observation: It was observed several times in Hulvika in May and June

<u>Comments</u>: It was very common in the last sample from Hulvika, in June. It was not until then that this species was identified and paid attention for, therefore, it could have been present in other samples as well.

Notosolenus urceolatus is distinguished from *N. hemicircularis*, by its larger size, absence of a collar and in having three not five dorsal keels.

cf. Notosolenus sp.

Plate 10, Fig. (h); Plate 9, Fig. (4); Plate 13, Fig (4)

<u>Description</u>: Oval shape, but more pointed anteriorly. Posterior part broader and not rounded. Very well developed reservoir that occupied 0.3 part of the cell length. Anterior flagellum ca. 1.5-2.0 times longer than the cell. Posterior flagellum could not be studied because the cell suddenly disappeared.

Size: 10 μm long and 6-7 μm wide.

Observation: At least once in Huk in June.

<u>Comments</u>: Since the posterior flagellum could not be seen, there may be a doubt whether this cell was a *Notosolenus*, which had lost its recurrent flagellum, or *Petalomonas*, which lack a second emergent flagellum.

Genus Petalomonas Stein 1878

Petalomonas cf. cantuscygni Cann and Pennick 1986

Plate 10, Fig. (g); Plate 9, Fig. (5); Plate 13, Fig (5)

<u>Description</u>: Flattened dorsiventrally. Longitudinal keels 5-6. Single emergent flagellum of same size as cell length or slightly shorter. Ingestion apparatus well defined and reached almost the posterior end of the cell, it was ca. 0.7 of the cell length. Vesicles antapically.

Size: 13 µm long and 10 µm wide.

Observation: At least once in Huk, beginning of June.

<u>Comments</u>: As it was commented for *Notosolenus* species, *Petalomonas* too is a genus that requires a lot of training in its species' identification due to its small size and few distinctive features. Thus, it may have been present in more samples too.

Petalomonas minuta Hollande 1942

Plate 10, Fig. (g); Plate 9, Fig. (6); Plate 13, Fig (6)

<u>Description</u>: Cell outline was elliptical and flattened. A deep longitudinal groove ran along the dorsal face of the cell. A single flagellum inserted in a reservoir in the right side of the cell and was about the same length as the cell. Nucleus in the left side of the cell. Gliding.

Size: 10 µm long and 7 µm wide.

Observation: Observed in both localities in June.

<u>Comments</u>: The presence of *P. minuta* as well as other *Petalomonas* and *Notosolenus* species were noticed at the end of the study period because the difficulty in their identification. This means they could well have been present in earlier samples too.

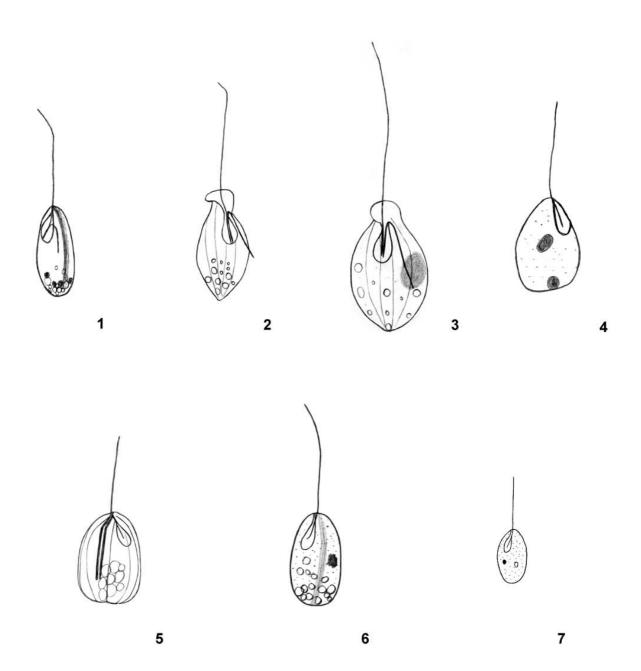


Plate 9. Euglenophytes: Notosolenus and Petalomonas

1-3 Notosolenus: 1. N. apocamptus, 2. N. hemicircularis, 3. N. urceolatus, 4. cf. Notosolenus sp., 5-7 Petalomonas: 5. P. cantuscygni, 6. P. minuta, 6. P. poosilla.

Notosolenus and Petalomonas: 3000x

Petalomonas poosilla J. Larsen and Patterson 1990

Plate 10, Fig. (g); Plate 9, Fig. (7); Plate 13, Fig (7)

<u>Description</u>: Cell were ovate to ellipsoidal and flattened. A single flagellum emerged from the reservoir with about the same length as the cell. Reservoir in the right side and nucleus in the left side of the cell. The cells move by gliding.

Size: 5-7 μm long and 4-5 μm wide.

Observation: In both localities in June.

Order Eutreptiales sensu Leedale 1967

Genus Cyclidiopsis Korschikow 1917

Cyclidiopsis acus Korschikow 1917

Plate 10, Fig. (a), Plate 11, Fig. (1) and Plate 11, Fig. (1)

<u>Description</u>: Colourless euglenid. Extremely long and thin, fusiform or elongated. Only one flagellum emergent from a wide apical canal. Reservoir and canal measured 15 to 20 μ m and were very visible. Pellicula striation very fine, almost invisible, and with oblique pattern. Flagellum long, 1-1.5 times the cell length and had whip-like beats. Needle-like structured paramylon grains about 25-30 μ m long were distributed all through the cytoplasm. Nucleus long and in a central position. Eye-spot absent. Very metabolic, with pronounced euglenoid movement. Moved by gliding.

Size: up to 130 µm long and 7 µm wide when completely distended.

Observation: It was only observed once in a sample from Hulvika from May.

<u>Comments</u>: *Cyclidiopsis acus* is an unexpected species to be found in Hulvika as it is usually confined to freshwater environments. This may indicate influence of freshwater at least in part of the beach in Hulvika.

Genus Eutreptiella da Cunha 1913

Eutreptiella sp.

Plate 10, Fig. (g); Plate 11, Fig. (3); Plate 18, Fig. (3); Fig. 19

Description: Phototrophic cylindrical cells with a pointed posterior end. Two unequal flagella inserted apically, a short posterior one was 0.6 and the anterior one 1- 1.5 times the cell. Both would beat actively to propel the cell through the water. The reservoir was shallow, ca. 7 μm. Many bright green coloured chloroplasts were visible in the cytoplasm. A distinct red eyespot or stigma located outside the plastids and by the reservoir. A pyrenoid of 4-6 μm in diameter and with 2 paramylon shields around was present in the centre of the cell. A large nucleus was located below the pyrenoid. Pellicular striation shallow, but clearly visible, ca. 6-7 striae per μm. Active swimmers and metabolic cells. They could shed the flagella and loose shape quickly forming structures called palmelloids (Fig. 19).

Size: 38-40 μm long and 10-12 μm wide.

Observation: Observed in Huk several times in May and June. Many times they appeared in great numbers as palmelloids.

<u>Comments</u>: It was the only autotrophic euglenid observed in the current study. It was quick and when it stopped it formed a palmelloid or blew up so fast that it made it hard to study in detail.

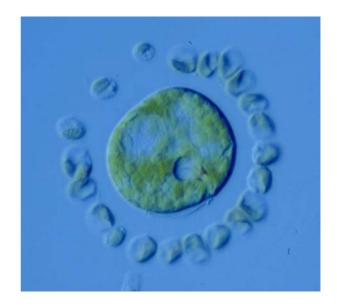


Fig. 19 Eutreptiella sp. in palmelloid form

Euglenozoa sp.

Plate 11, Fig. (4)

<u>Description</u>: Colourless cell. Elliptical to round. Flattened dorsiventrally, ventral and dorsal side concave, as two valves. Cell surrounded by a kind of membranous rim in which transversal lines were drawn. This rim was interrupted by the canal from which two unequal flagella emerged. The reservoir was short and no distinction could be observed between the canal and the reservoir itself. Anterior flagellum, which steered the cell and was thicker, of about the same as the cell length and the recurrent flagellum a little less than 2.0 times the cell length. Smooth gliding.

Size: 18-22 µm long and 16-20 µm wide.

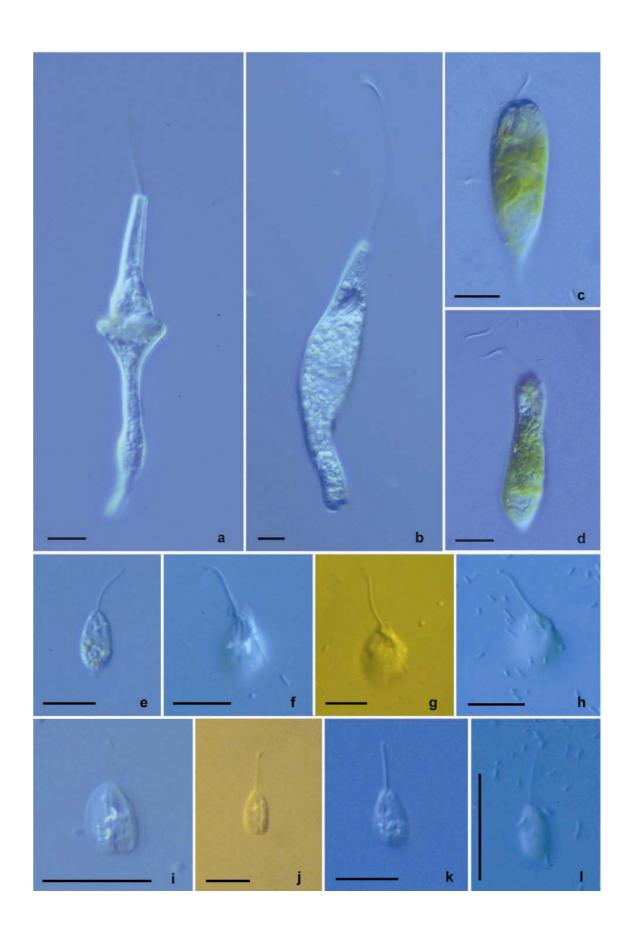
Observation: Only in Hulvika, once in February.

<u>Comments</u>: The reservoir structure, the insertion of the flagella and the motion style indicated it was an Euglenozoa, but it could not be further classified because nothing like it could be found in the literature.

Plate 10. Cyclidiopsis, Jenningsia, Eutreptiella, Notosolenus, Petalomonas

- (a) Cyclidiopsis acus, (b) Jenningsia sp., (c) (d) Eutreptiella sp.
- (e) Notosolenus apocamptus, (f) N. hemicircularis, (g) N. urceolatus, (h) N. sp.
- (i) Petalomonas cf. cantuscygni, (j), (k) P. minuta, (l) P. poosilla.

Scale bar=10µm



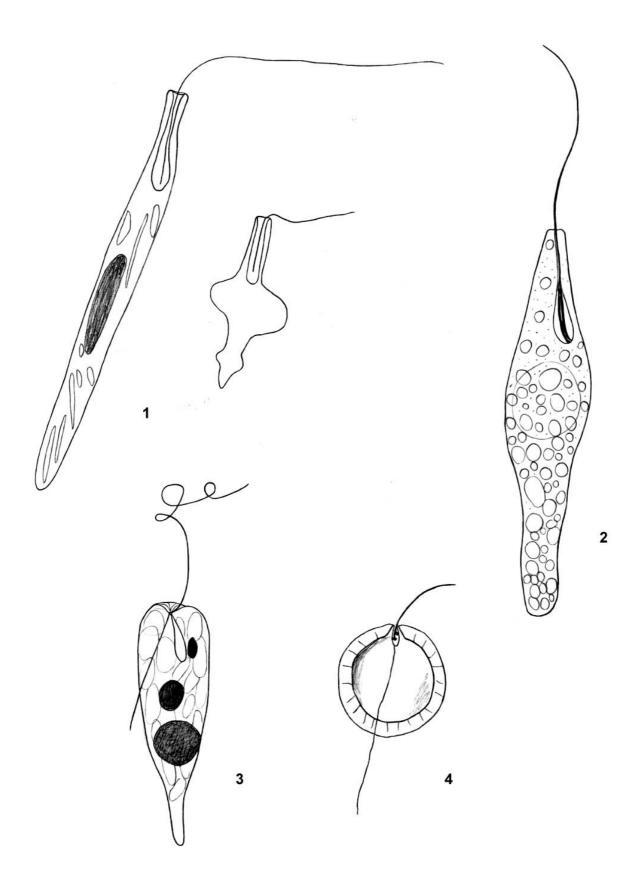


Plate 11. Euglenophytes: Cyclidiopsis, Jenningsia, Eutreptiella, Euglenophyta sp.

- 1. Cyclidiopsis acus (1000x) straight and elastical positions, 2. Jenningsia sp. (1000x), 3. Eutreptiella sp. (2000x),
- 4. Euglenophyta sp. (2000x).

Order Heteronematales Leedale 1967

Genus Dinema Perty 1852

Dinema litoralis Skuja 1939

Plate 7, Fig. (d); Plate 8, Fig. (6)

<u>Description</u>: Ovate cell, narrower on the posterior end. Helix-shaped pellicular striation, almost longitudinal and very conspicuous. Anterior flagellum less than 1.5 times the cell length and recurrent one slightly longer than the cell. Ingestion organelle with two narrow rods, extending less than halfway down the cell, and situated to the right of the cell. Nucleus spherical and situated in the posterior part. Smooth gliding interrupted by sudden stops. Phagotrophic, may consume diatoms as big as itself, ca. $50 \, \mu m$.

<u>Size</u>: 45-60 μm long and 18-25 μm wide.

Observation: Observed several times in both localities, in April, May and June.

<u>Comments</u>: This is an easy species to identify. It usually appears with ingested diatoms inside.

Dinema valida J. Larsen and Patterson 1990

Plate 7, Fig. (e); Plate 8, Fig. (5)

<u>Description</u>: Ovate cell. Quite wide almost longitudinal striations on both faces and these follow an S-helix. The anterior flagellum was as long as the cell or slightly longer and beat with a sweeping motion. The posterior flagellum was between 3.0 and 3.5 times the cell length, very thickened in its basis and emerged as a hook from the flagellar pocket. Ingestion apparatus present, had two rods and extended halfway down the cell. Phagotrophic. Nucleus in the right side of the cell. Gliding motion.

Size: 46 µm long and 22 µm wide.

Observation: Once in Huk in June.

<u>Comments</u>: *Dinema valida* is distinguished from *D. litorale* by its widely spaced pellicular striation and its strong recurrent flagellum.

Dinema sp.

Plate 7, Fig. (g); Plate 8, Fig. (7)

<u>Description</u>: Elongated, rounded anteriorly, tapered posteriorly, very pointed. Pellicular striation fine, almost longitudinal and narrowly spaced. Posterior and anterior flagella about the same length as the cell, thickened recurrent flagellum. Ingestion apparatus with two short rods, to the left of the flagellar pocket. Flagellar insertion pocket well developed and oblique. Large nucleus on the right side of the cell. Gliding cell with sudden squirming movements.

<u>Size</u>: 70 μm long and 22 μm wide. Observation: Once in Huk in June. <u>Comments</u>: The shape, cell surface and size pointed to *Dinema*. However, this cell was larger than any other *Dinema* species found earlier in the sampling period. The recurrent flagellum could be a distinctive feature in identifying it because it was as long as the cell, which is not common in this genus, but I found no species description within *Dinema* that would fit this characteristic.

Genus *Heteronema* Dujardin 1841

Heteronema exaratum J. Larsen and Patterson 1990

Plate 7, Fig. (j); Plate 8 Fig. (8)

Description: Cells ovate to round, apex and antapex slightly pointed. Dorsiventrally flattened. Rigid, with little or none plasticity. Canal opening subapically. Small ingestion organelle of about 4 μ m long situated to the right of the flagellar pocket. Conspicuous pellicular striation following an S-helix. Flagella similar in length, slightly longer than the cell and pointing in different direction when moving. The anterior flagellum pointed to the right and forward and the posterior one trailed and pointed to the left. Reservoir in the left side of the cell. Nucleus in the centre. Swimming motion, not gliding. Extremely fast swimmers in comparison to other euglenoids. The cell and flagella advanced as one, moving as a whole with the anterior flagellum held in an arc. In immotile cells, the flagella coiled up.

Size: 11-14 µm long and 10-12 µm wide.

Observation: In both localities several times through the study period.

<u>Comments</u>: *Heteronema exaratum* was easy to recognize due to its peculiar way of moving, its small size and the length of its flagella. However, almost at the end of the work, I realized that I might have mixed it up sometimes with *Metanema strenuum*, which has a similar appearance but lacks of an ingestion apparatus. It can be assured, however, that *H. exaratum* was observed at least once in Huk.

Heteronema ovale Kahl 1928

Plate 7, Fig. (i); Plate 8, Fig. (9)

<u>Description</u>: Oval cells with a pointed posterior end visible when the cell stretched and elongated. Canal opening subapically. Flattened with pellicular striation oblique, following an S-helix, often associated with refractive bodies parallel to each striae. Flexible cells. Two flagella, anterior one about the same length as the cell and the recurrent one up to twice the cell length. Ingestion organelle with two rods, extending about half way to the posterior end. Skidding type of motion with the anterior flagellum held in an arc and beating freely while the recurrent flagellum retained a loose contact with the substratum, moving vigorously in a plane parallel to the substratum. It squirmed very often.

<u>Size</u>: 22-32 μm long and 18-26 μm wide.

Observation: In Huk and Hulvika, frequently from February to June.

<u>Comments</u>: *Heteronema ovale* was easy to recognize by its squirming and skidding behaviour, its pellicular striation accompanied by refractile granules and its pointed posterior end. *Heteronema ovale* could be confused with the recently described species, *H. larseni* Lee and Patterson, 2000, but the former is distinguished by its smaller size.

Genus Jenningsia Lee, Blackmore and Patterson 1999

Jenningsia cf. macrostoma (Ekebom et al. 1996) Lee et al. 1999

Plate 10, Fig. (b); Plate 11 Fig. (2)

<u>Description</u>: Long cell, fusiforme shaped. Rounded posteriorly, pointed anteriorly. Fine pellicular striations, almost longitudinal. Ingestion organelle with two poorly developed short rods in the front of the cell. Flagellar pocket located on the left side of the cell, it only extended down to a 0.2 or 0.3 part of the cell length. A very thickened single flagellum projected from the canal and was as long as the cell length, or slightly shorter. It beat freely. Refractile granules were randomly distributed inside the cell. Nucleus located in the centre of the cell. The cell glided with a squirming movement.

Size: Up to 130 long when completely stretched and 15-20 µm wide.

Observation: Once in a sample from Huk, in the beginning of June.

Comments: It appears to be slightly bigger than its original description which gives a size range between 80 and 114 μ m. However, the rest of the morphological and behavioural characteristics fit well with the description. *Jenningsia* cf. *macrostoma* appeared only once which indicates that it is a rare species which together with its peculiarity, made it easy to distinguish from the rest of the euglenoids, but still hard to find in the literature due to its rareness.

Genus *Peranema* Dujardin 1841

Peranema dolichonema J. Larsen and Patterson 1990

Plate 7, Fig. (h); Plate 8, Fig. (4)

<u>Description</u>: Cells long sack-shaped with longitudinal pellicle striations. Ingestion organelle not observed. Posterior end of the cell rounded. Anterior flagellum almost twice the length of the cell. Posterior flagellum slightly longer than the cell. The reservoir went deep down in the cell. Nucleus in the posterior part. The recurrent flagellum stayed attached to the body as if it fits in a groove and only the posterior short part was free.

<u>Size</u>: 30-40 μm long and 16-20 μm wide.

Observation: Huk and Hulvika, once in each locality, in June and May respectively.

<u>Comments</u>: *Peranema dolichonema* is distinguished from other large colourless euglenoids, by its sack shape, and the recurrent flagellum of which the posterior part lies free from the cell body.

Genus Ploeotia Leedale 1969

Ploeotia adhaerens J. Larsen and Patterson 1990

Plate 7, Fig. (1); Plate 13, Fig. (8)

<u>Description</u>: Cell round and very flattened dorsiventrally with four prominent or raised longitudinal ridges in irregular trace, on both ventral and dorsal sides as crests. Posterior end with a blunt protrusion. Two unequal flagella emerged from a flagellar groove, anterior flagellum as long as the cell and the posterior one 1.5 times the cell length. Posterior flagellum base not thickened. Ingestion apparatus with two long rods on the right side, they deepened down to the posterior end and the distance between them decreased posteriorly. Nucleus on the right side of the cell. Gliding motion with anterior flagellum sweeping.

Size: 23 µm diameter.

Observation: Once in Huk in June.

<u>Comments</u>: *Ploeotia adhaerens* can be identified by its regular rounded outline, the long and posteriorly tapering ingestion organelle and the surface crests.

Ploeotia corrugata J. Larsen and Patterson 1990

Plate 7, Fig. (m); Plate 13, Fig. (10)

<u>Description</u>: Cell outline oval to elongated. Dorsally convex and ventrally flattened. Two lateral and 4 ventral thick corrugated longitudinal ridges. Posterior end with indentation. Two flagella of unequal length, the anterior flagellum, which beat rapidly from side to side, was ca 0.6 times the cell length and the posterior one about 1.7 times. Ingestion organelle extended from the right anterior to the left posterior side and was difficult to observe. Nucleus on the right side of the cell. Movement by smooth gliding.

Size: 12-14 μm long and 7-9 μm wide.

Observation: Twice in Huk in April and June.

<u>Comments</u>: The corrugated appearance of the dorsal side and the indented posterior end are distinguishing features for *Ploeotia corrugata*.

Ploeotia pseudanisonema J. Larsen and Patterson 1990

Plate 7, Fig. (o); Plate 13, Fig. (11)

<u>Description</u>: Oblong cell, flattened dorsiventrally. With 3-4 fine longitudinal grooves on the ventral face. Anterior flagellum about 1.5 times the cell length and with sweeping motion, recurrent flagellum about 4 times the cell length and thickened in its base, tapering distally. Flagellar canal and reservoir in the left side and reached shallow down in the cell. Ingestion organelle with two conspicuous rods extending almost to the posterior end of the cell. Nucleus on the right hand side. Gliding type of movement.

Size: 16 μm long and 7 μm wide.

Observation: Once in Huk in June.

<u>Comments</u>: *Ploeotia pseudanisonema* can be easily distinguished by the length of its recurrent flagellum.

Ploeotia tenuis J. Larsen and Patterson 1990

Plate 7, Fig. (n); Plate, 13 Fig. (9)

<u>Description</u>: Cell outline oblong to roundish. Flattened. Posterior end pointed. Fine longitudinal grooves, 2-3, ventrally. Anterior flagellum about the cell length and recurrent one 2.5 times the cell. Ingestion organelle very well developed with two long rods that extended the length of the cell and formed a hook in its base.

<u>Size</u>: 16 μm long and 13 μm wide.

Observation: Once in Hulvika in May.

Ploeotia vitrea Dujardin 1841 emend. Farmer and Triemer 1987

Fig. 20; Plate 13, Fig. (12)

<u>Description</u>: Cell outline oval, not very flattened. Posterior end pointed and anterior part slightly obtuse. Four ventral, two lateral and four more dorsal refractive, prominent and longitudinal ridges consisted of double fine ridges. The anterior flagellum was slightly shorter than the cell and beat freely from side to side, posterior flagellum was thick and almost twice as long as the cell. Ingestion apparatus very well developed, tapered posteriorly, with an anterior protrusion in the right rod and as long as the cell and slightly oblique, inserted from the anterior mid part to the posterior left side. Nucleus in the mid-left side. Reservoir difficult to observe and located at the left hand side of the cell. Food inclusions distributed randomly in the cytoplasm.

Size: About 50 μm long and 30 μm wide.

Observation: Observed once in Huk, beginning of June.

<u>Comments</u>: *Ploeotia vitrea* has usually been described as being half size the present cell. However, features found in the current cell, such as the 10 doubled refractive ridges, flagellar lengths and the ingestion organelle were in accordance with the description for this taxon.



Fig. 20 Ploeotia vitrea.

Genus Urceolus Mereschkowsky 1879

Urceolus cornutus J. Larsen and Patterson 1990

Plate 12 Fig. (a), (b), (c); Plate 13, Fig (14)

<u>Description</u>: Cell sack-shaped. Anterior collar with a wide opening, about $16 \mu m$ in diameter and with a regular outline. Pellicular striations fine, closely spaced and in S-helix, closely spaced and following a pattern such: one distinct intercalated with one less-marked striae. Single flagellum emerging from the inside of the collar and about the length of the cell. Ingestion organelle well developed with two rods connected in the base of the collar and diverging from one another posteriorly. Nucleus located in the central-posterior part. No adhered detritus particles. One cell was extremely metabolic.

Size: 27 μm long and 14 μm wide.

Observation: Observed twice in Huk in June

<u>Comments</u>: *Urceolus cornutus* has the following distinguishing features from other *Urceolus* species: the pellicular striations are very compact and fine, the collar has a more regular outline.

Urceolus cf. cristatus J. Larsen and Patterson 1990

Plate 12 Fig. (g), (h); Plate 13, Fig (16)

<u>Description</u>: Sack-shaped cell. Pellicle stripes not visible except on the expanded part of the collar. Flagellum length almost twice as long as the cell. Ingestion apparatus not observed. Refractive detrital material adhered to the cell surface obscured details of the body surface and gave the cell a glassy, bright pearl colour. Nucleus located laterally.

Size: 27 µm long and 22 µm wide.

Observation: Once in Hulvika in May.

<u>Comments</u>: The prominent longitudinal ridge that extends the length of the flagella best characterizes this one from the rest of the *Urceolus* species. However, this feature was not observed, probably due to the enormous amount of detrital particles adhered to the cell surface. Other characteristics, however, such as, the smaller size in comparison to *U. sabulosus* and *U. cornutus*, and detritus level on cell surface.

Urceolus cyclostomus (Stein 1878) Mereschkowsky 1878

Plate 12 Fig. (i); Plate 13, Fig (13)

<u>Description</u>: Colourless, plastic cell. Flask-shaped with a stout posterior region. It had a funnel-like neck with a collar of 16 μm in diameter and an irregular outline. Pellicular striation in S-helix. A single 55 μm long flagellum with its base thickened emerged from the hollow neck. It reached inward the posterior third of the cell body. Ingestion organelle with two parallel rods, 12-14 μm long. Nucleus big, 15 μm in diameter, located posteriorly.

Size: About 45 μm long and 25 μm wide.

Observation: Once in Hulvika and probably another time in Huk (*U. cf. cyclostomus*), both in June.

Urceolus sabulosus (Stokes 1886) J. Larsen and Patterson 1990

Plate 12 Fig. (d), (e), (f); Plate 13, Fig (15)

<u>Description</u>: Cell sack-shaped. Pointed posteriorly. Flagellum about the length of the cell or slightly longer. Ingestion organelle could not be well observed. Reservoir very distinct and reached down half of the cell length. Fine pellicular striation visible only on the collar surface. Collar with irregular outline and its circular border thickened. Surface somewhat covered with adherent particles. Nucleus not observed. A red inclusion situated to the left of the cell. Ingestion vacuoles present. Elastic cell with squirming movements.

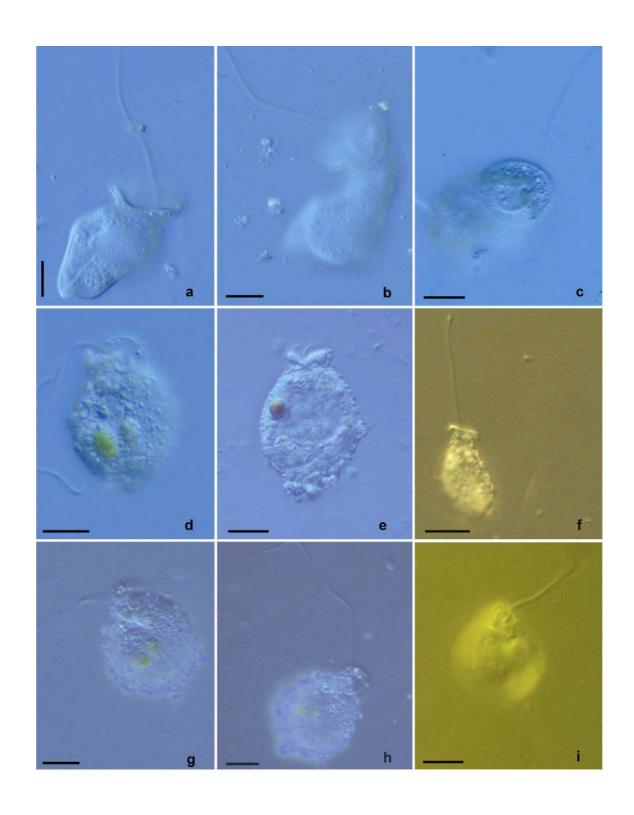
Size: $36-40 \mu m$ long and $20-22 \mu m$ wide.

Observation: Twice in Hulvika in May and June.

Plate 12. Urceolus

Scale bar=10 µm

⁽a) Urceolus cornutus, (b) U. cornutus, pellicular striation, (c) U. cornutus, collar detail (d) Urceolus sabulosus, (e) U. sabulosus, collar ventrally, (f) U. sabulosus, flagellum length (g), (h) U. cf. cristatus, (i) U. cyclostomus.



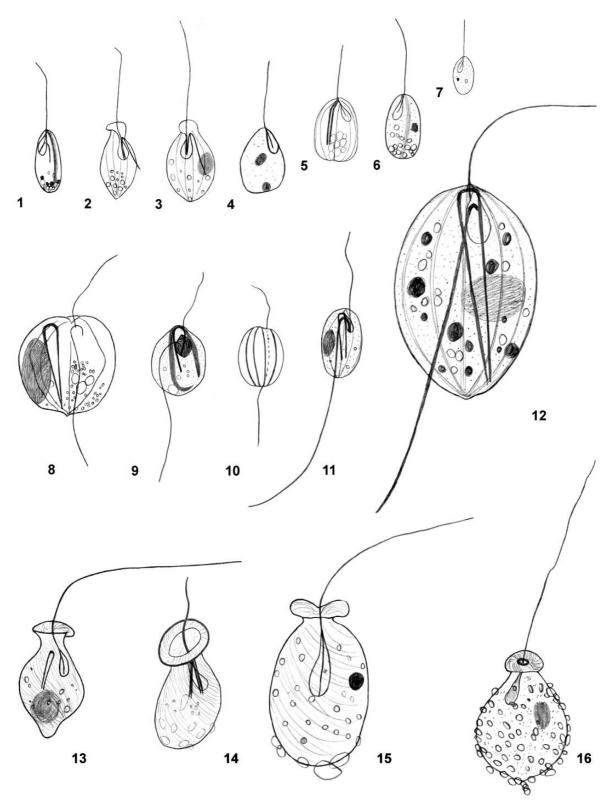


Plate 13. Euglenophytes: Notosolenus, Petalomonas, Ploeotia, Urceolus.

1-4 Notosolenus: 1. N. apocamptus, 2. N. hemicircularis, 3. N. urceolatus, 4. N. sp.,

5-7 Petalomonas: 5. *P. cantuscygni*, 6. *P. minuta*, 7. *P. poosilla*, 8-12 Ploeotia: 8. *Pl. adhaerens*, 9. *Pl. tenuis*, 10. *Pl. corrugata* in dorsal view, 11. *Pl. pseudoanisonema*, 12. *Pl. vitrea*, 13-16 Urceolus: 13. *U. cyclostomus*, 14. *U. cornutus*, 15. *U. sabulosus*, 16. *U. cristatus*

Notosolenus and Petalomonas: 2000x; Ploeotia and Urceolus 1500x

DIV. CHLOROPHYTA

Class Prasinophyceae T. Christensen, 1962 ex Moestrup & Throndsen 1988

Order Chlorodendrales Fritsch 1917

Genus Nephroselmis Stein 1878

Nephroselmis cf. *rotunda* (N.Carter) Fott

Plate 14, Fig. (2)

<u>Description</u>: Autotrophic, bean-shaped cell. Two unequal flagella inserted laterally, in an invagination, shorter one directed anteriorly during swimming and longer one trailing. Single green-yellow chloroplast, with a red eyespot in the centre of the cell, in association with the chloroplast. Pyrenoid present.

Size: 7 µm long and 6 µm wide..

Observation: Once in Hulvika in February.

<u>Comments</u>: The present identification into *Nephroselmis* cf. *rotunda* was suggested by Jahn Throndsen according to his experience with the Prasinophyceae. It can be easily mixed with *N. pyriformis* (N. Carter) Ettl 1982, and only the morphology of its scales observed in TEM could tell them apart. Since this was not performed in this case, I can only assure the genus Nephroselmis, characteristic by its kidney shape, its small size and the layout of the flagella. On the other hand, since it was only observed once, the present *Nephroselmis* cell was most probably originally from the free water bodies neighbouring the sampled sand and was brought mixed with it into the lab.

Genus Pyramimonas Schmarda 1850

Pyramimonas cf. orientalis Butcher 1959

Plate 14, Fig. (3)

<u>Description</u>: Four-lobed cells with an inversely pyramidal shape, tapering posteriorly but posterior end rounded. Four flagella inserted in an anterior pit in the centre of the pyramid base and pointing posteriorly, about 1.0 times the cell length. A large green chloroplast took up most of the cell volume, and a large pyrenoid surrounded by starch shields took part of the chloroplast volume in the posterior part of the cell. Big double eyespots in the centre of the cell. Granules in the surroundings of the invagination.

Size: 16 µm long and 10 µm wide.

Observation: At least once in Huk in June.

<u>Comments</u>: Though the morphology of this genus is typical and common to almost all the species, *Pyramimonas* species identification depends very much on the ultra structure of flagella and body scales revealed only in the electron microscope.

Pyramimonas sp. 1

Plate 1, Fig. (i); Plate 14, Fig. (4)

<u>Description</u>: Small, four-lobed inversely pyramidal-shaped cell, very pointed posteriorly. Four flagella emerging from an anterior invagination and pointing to the sides, as long as the cell length. Single green chloroplast. Pyrenoid posteriorly, not-shielded. The double eyespot is situated by the central invagination.

<u>Size</u>: 8 μm long and 5 μm wide.

Observation: in Huk in June.

Pyramimonas sp. 2

Plate 14, Fig. (5)

<u>Description</u>: four-lobed oval shaped cell. Posterior part rounded, as well as anterior part. Four flagella emerging from an anterior depression and directed posteriorly, slightly longer than the cell length. Large green chloroplast occupying a big part of the total cell volume. Basal pyrenoid strongly shielded and surrounded by ring-like structures. Two small eyespots in the chloroplast-free area, by the anterior invagination area. Small granules anteriorly.

Size: 17 μm long and 14 μm wide.

Observation: Once in Hulvika in June.

Class Chlorophyceae sensu Mattox et Stewart 1984

Order Volvocales Oltmanns 1904

Genus Chlamydomonas Ehrenberg 1834

Chlamydomonas cf. nonpulsata Butcher 1959

Plate 1, Fig. (j); Plate 14, Fig. (1)

<u>Description</u>: Rounded cell, flattened dorsiventrally, surrounded by a sheath and with an anterior papilla. Two equal flagella slightly shorter than the cell length emerging from the papilla area. Single large green chloroplast. Nucleus central. Red-orange stigma laterally located. Basal pyrenoid. Granules all over the cell cytoplasm.

Size: 20-24 μm long and 14-16 μm wide.

Observation: At least once, when carefully observed, in Huk, beginning of June.

<u>Comments</u>: Like *Pyramimonas*, *Chlamydomonas* is a very difficult genus for species identification. The time available did not allow for such time consuming identification work and these taxa were therefore given less priority than the other flagellates encountered.

Other *Chlamydomonas* cells were also present in the samples of which I only took photographs (see Plate 1, Fig. (k), (l)).

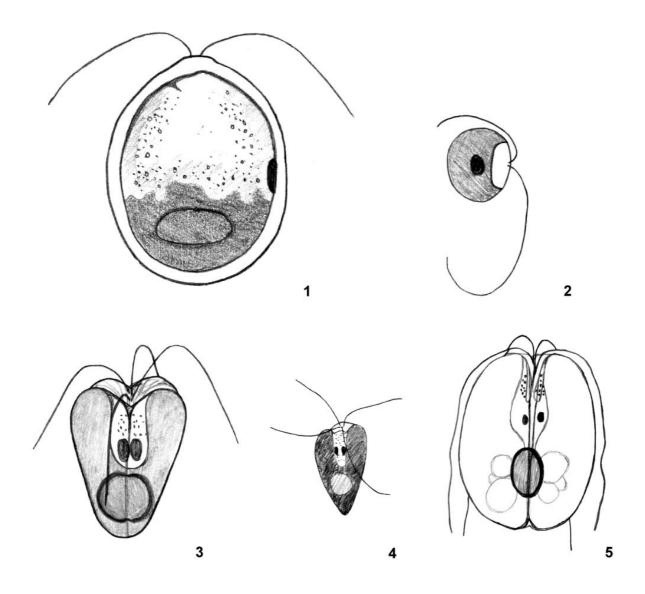


Plate 14. Chlorophyta.

- 1. Chlamydomonas cf. non-pulsata, 2. Nephroselmis cf. rotunda, 3. Pyramimonas cf. orientalis, 4. Pyramimonas sp.1, 5. Pyramimonas sp.2.

All 3000x

Group related to multicellular taxa (Lee et al. 2000)

Class Choanoflagellatea sensu Cavalier-Smith (2002) Order Choanoflagellida W.S. Kent 1880

Acanthocorbis sp. S. Hara & E. Takahashi 1984

Plate 1, Fig. (o)

<u>Description:</u> Lorica around the cell of 8 μ m long and 5-6 μ m wide. Eleven to 14 transversal costae and ca. 15 longitudinal costae. Single flagellum comes out of the lorica.

Size: 15-16 µm long.

Observation: Only observed twice in Huk, in February and March.

<u>Comments</u>: Though choanoflagellates have mostly been reported from free water masses,

Norris (1965) showed a rich occurrence of species living neustonic in tidal pools.

Species of uncertain taxonomic position: Insertae sedis

Amastigomonas mutabilis (Griessmann 1913) Molina and Nerad 1991

Plate 15, Fig. (h); Plate 16, Fig. (14), (15)

<u>Description</u>: Elliptical cells, flattened dorsiventrally. Two flagella subapically inserted. Laterally directed anterior snout from which the anterior flagellum extended about 0.5 times the cell length. The recurrent flagellum extended 1.5 times the cell length, trailing under the cell body to which it attached loosely in a longitudinal slight groove. Granules alongside the recurrent flagellum. Nucleus situated subapically near the right margin of the cell. Gliding motion and great flexibility.

Size: 10-11 µm long and 5 µm wide.

Observation: Observed in Hulvika in March and June. Few individuals.

<u>Comments</u>: It is also known as *Rhynchomonas mutabilis* Griessmann 1913 and *Thecamonas mutabilis* Larsen and Patterson 1990. *Amastigomonas mutabilis* is similar to *A. debruynei* de Saedeleer 1931, but can be distinguished by its bigger size, its longer flagellum and because of its ventral granules alongside the recurrent flagellum.

Amastigomonas debruynei de Saedeleer 1931

Plate 16, Fig. (16)

<u>Description</u>: Ovate cell, dorsiventrally flattened and flexible. With a snout emerging from the anterior right margin of the cell. Anterior flagellum emerging from this snout, 1 μ m, tip region narrower than the rest. Recurrent flagellum also inserted subapically and trailing under the cell, about 1.5 times the cell length. Cell metabolic. Gliding motion.

Size: 7 μm long and 5-6 μm wide.

Observation: Observed at least once in Hulvika, in May.

Comments: It has also been known as *Thecamonas trahens* Larsen and Patterson 1990.

Bodo designis Skuja 1948

Plate 15, Fig. (a); Plate 16, Fig. (1)

<u>Description</u>: Elliptical cell body with two flagella inserted subapically. The apical part of the body being set off as a rostrum. Anterior flagellum, half the length of the cell, posterior one slightly longer than 1.5 times the cell length and trailing and the tip adhering to the substrate. Moving with a rotating, skidding or gliding motion.

Size: 6-8 µm long and 3-4 µm wide.

Observation: Often observed in both localities.

Bodo cf. saliens J. Larsen and Patterson 1990

Plate 15, Fig. (b); Plate 16, Fig. (2)

<u>Description</u>: Elliptical-shaped cells with two unequal flagella emerging subapically from a lateral pocket. Anterior flagellum shorter than body length. Posterior flagellum twice as long as the cell length. Cell swam in rapid darts and straight lines.

Size: 7 μm long and 4 μm wide.

Observation: Observed in Hulvika at least once.

<u>Comments</u>: Bodonids' identification is difficult due to their reduced size and their similar appearance.

Colpodella unguis Patterson and Simpson 1996

Plate 15, Fig. (e); Plate 16, Fig. (7)

<u>Description</u>: Bean-shaped in profile. The anterior flagellum inserted in a deep, triangular curving depression; slightly shorter than the cell length, directed laterally and posteriorly. The posterior flagellum about 1.5 times the cell length, inserted at the top of a shallow longitudinal groove on one face of the cell and directed posteriorly. Swimming rapidly.

Size: 10 µm long and 5 µm wide.

Observation: Once in Huk in June.

Comments: Rare species. Its peculiar and different appearance eases the identification.

Cryptaulax cf. marina Throndsen 1969

Plate 15, Fig. (f), (g); Plate 16, Fig. (6)

<u>Description</u>: Cells elliptical, pointed posteriorly. Flagella inserted near the anterior end, in a groove or pocket-like structure. The anterior flagellum slightly shorter than the cell length and pointed anteriorly in a curve. Recurrent flagellum twice as long as the cell body and twisted round the cell following a spiral groove. Vacuoles observed. Rotating motion with close adherence to the substratum.

Size: 12-16 µm long and 4-5 µm wide.

Observation: Observed few times in Hulvika, in March and June.

Discocelis saleuta Vørs 1988

Plate 16, Fig. (8)

Description: Disc-shaped and colourless cell, difficult to notice. Flattened dorsiventrally. Two flagella emerging from a concave anterior margin of the cell. The anterior flagellum, shorter than 1 μ m. The recurrent flagellum, slightly longer then the cell, trailed behind the cell. Vacuoles present. Smooth gliding motion, in close contact with the substrate.

Size: 5 µm in diameter.

Observation: Observed few times in in Hulvika.

Metopion fluens J. Larsen and Patterson 1990

Plate 15, Fig. (i); Plate 16, Fig. (9)

<u>Description</u>: Cell outline ovate. Laterally compressed with a small rostrum anterior to the flagellar insertion. One flagellum 1.5 times the cell length emerging from behind the rostrum was observed. Moved by gliding.

Size: 3-6 µm in diameter.

Observation: Few times in both localities spread all through the sampling period.

<u>Comments</u>: Easy to identify due to its shape and small size. It is so tiny that the light microscope allows to observe only the general cell outline and the long flagellum. The very short second flagellum is difficult to notice. Not a common species.

Metromonas simplex (Griessmann 1913) Larsen and Patterson 1990

Plate 16, Fig. (11)

<u>Description</u>: Pear-shaped, very compressed dorsiventrally. A flagellum twice as long as the cell length emerged from the narrower end of the cell, curving at the distal end and attaching to the substratum. The second flagellum is very short and difficult to observe. The cell swung slowly from side to side like a pendulum. Vesicles in the cytoplasm.

Size: 6 μm long and 5 μm wide.

Observation: Few times in both localities and in different samples.

<u>Comments</u>: Impossible to mistake for other species due to its characteristic pendular movement. Rare.

Peltomonas volitans Vlk 1942

Plate 16, Fig. (12)

<u>Description</u>: Dorso-ventrally flattened, with a concavity in the front part of the cell from which one anteriorly projecting flagellum emerged. Flagellum 20 µm long.

Size: 5-6 µm in diameter.

Observation: At least once in Huk in February.

<u>Comments:</u> Morphologically, it is a very simple organism, easy to identify. Rare species.

Phyllomitus granulatus J. Larsen and Patterson 1990

Plate 16, Fig. (5)

<u>Description</u>: Sack-shaped cell. Rounded posteriorly, more pointed anteriorly. Two flagella emerging subapically from a pocket. Anterior flagellum beat with a sine-wave, was about 1.0 times the length of the cell and directed to the front and slightly to the right. Posterior flagellum equal to the cell length and trailing behind the cell. Slightly flexible cell. No refractile granules observed on the surface.

Size: 8 µm long and 5 µm wide.

Observation: Once in Hulvika in May.

Comments: Rare species.

Protaspis gemmifera J. Larsen and Patterson 1990

<u>Description</u>: Cell outline roundish and dorsiventrally flattened. Cell surface rather warty. Two flagella inserted subapically in an indistinct ventral furrow. Anterior flagellum as long as the cell length and posterior one twice as long. Posterior flagellum trailing behind the cell and anterior one beating rapidly. Nucleus situated anteriorly, below the flagellar insertion. Cell moved by gliding.

<u>Size</u>: 13-16 μm long and 11-14 μm wide.

Observation: Once or twice in each of the localities.

<u>Comments</u>: Differed from the other Protaspis by its smaller size, its rounded ouline, its warty surface and the lack of a ventral furrow and an anterior protuberance. Not as common as *Protaspis obliqua* J. Larsen and Patterson 1990 and *Protaspis tegere* J. Larsen and Patterson 1990

Protaspis obliqua J. Larsen and Patterson 1990

Plate 15, Fig. (o), (p); Plate 16, Fig. (17)

<u>Description</u>: Cells oval to roundish, flattened dorsiventrally. Cell indented anterioly showing a small protrusion that protruded to the right of the cell. Two flagella inserted under the protrusion. Anterior flagellum half the cell length and the posterior one 1.5 times the length of the cell. Rounded nucleus located subapically in a median position. Some cells with food vacuoles.

Size: 20-25 µm long and 12-16 µm wide.

Observation: Randomly in both localities throughout the whole study period.

<u>Comments</u>: *Protaspis obliqua* was distinguished by its larger size compared to *P. gemmifera* (Larsen and Patterson 1990), its anterior indentation and the position and rounded shape of the nucleus.

Protaspis tegere J. Larsen and Patterson 1990

Plate 15, Fig. (q), (r); Plate 16, Fig. (18)

<u>Description</u>: Oblong, ovate cells. Slightly flattened. A longitudinal median ventral groove extended from the flagellar insertion depression to the posterior end of the cell. Two flagella, unequal in size, inserted subapically. The anterior flagellum inserted slightly anterior to the posterior flagellum and was slightly shorter than the length of the cell. The posterior flagellum was 1.5 times the cell length. Nucleus was disc-shaped and located in the right side of the cell. Gliding motion.

<u>Size</u>: 16-24 μm long and 10-16 μm wide.

Observation: Frequently observed in both localities through all the study period.

<u>Comments</u>: *Protaspis tegere* differed from other *Protaspis* species by its longitudinal ventral furrow and its more elongated shape.

Rhynchomonas nasuta (Stokes 1888) Klebs 1892

Plate 15, Fig. (j); Plate 16 Fig. (13)

<u>Description</u>: Ovoid cell body, metabolic. With a bulbous motile snout or proboscis at the lateral anterior margin of cell, appearing as a developed rostrum. The proboscis moved from side to side. One trailing flagellum inserted at the base of the snout, extended 3 times as long as the length of the cell and trailed behind during locomotion. Anterior flagellum lies alongside the snout and is difficult to see. Cells glided.

Size: 6 µm long and 5 µm wide.

Observation: Observed few times in Hulvika and Huk.

Comments: Rhynchomonas nasuta is characterized by its well developed snout.

Protist 1

Plate 15, Fig. (k)

<u>Description</u>: Rounded cell, flattened, with an invagination in the anterior margin of the cell which created an indentation. Slightly posterior to this area, a single 20 μm long flagellum emerged from a shallow depression and trailed behind the cell. All along the circular margin of the cell, a row of granules occured. It appeared as if this row protruded from the cell's flat surface. Food vacuoles present. Cell moved by gliding.

Size: 14 µm in diameter.

Observation: Once in Huk in May.

Protist 2

Plate 15, Fig. (1); Plate 16, Fig. (10)

<u>Description</u>: Oval cells with an anterior depression from which two flagella emerged. Anterior one, which moved rapidly from side to side, was directed forward and to the left,

and posterior one was trailing behind and oriented to the right of the cell. Both flagella equal in length to the cell. A ventral longitudinal groove ran from the depression down to almost the antapex. A line of granules were drawn longitudinally, almost parallel to the groove and to the left side of it. It moved smoothly in big circles.

Size: 11 µm long and 6-7 µm wide.

Observation: Numerous in a sample from May in Hulvika only.

Protist 3

Plate 15, Fig. (m), (n); Fig. 21.

<u>Description</u>: Roundish, totally colourless cell, flattened. Rough surface. In lateral view, it appeared tapering distally, as if there were three different levels (see Fig. 21, lateral view). Anterior margin of the cell with an invagination from which four flagella emerged. Two flagella, 2.0 times the cell length, coiled parallely to one another around the ventral side (see sketch in Plate 15, Fig. (m)), and the other two, as long as the length of the cell, directed forward. Some cells with orange food inclusions. Smooth gliding.

Size: 20-22 µm in diameter.

Observation: in Huk, once in February and June.

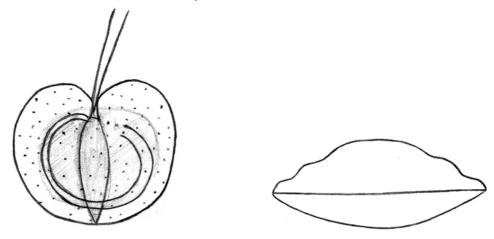


Fig. 21 Protist 3. Ventral and lateral views.

Protist 4

Plate 16, Fig. (14)

<u>Description</u>: Elliptical cells, flattened dorsiventrally. 2 flagella subapically inserted. Anterior flagellum projected forward, slightly shorter than the length of the cell. The recurrent flagellum 1.5 times the cell length, trailing under the cell body to which it attached loosely in a longitudinal shallow groove, following part of the plasmalema. A line of granules occurred alongside the recurrent flagellum. Gliding motion.

<u>Size</u>: 10 μm long and 8 μm wide.

Observation: Twice in Hulvika in March.

<u>Comments</u>: It should be easy to identify it due to the peculiar way the posterior flagellum trails parallel to the plasmalemma, but I found no species description in to which this organism would fit. Jahn Throndsen suggested it could be *Amastigomonas mutabilis* (Griessmann) Molina and Nerad, but even though it shared similarities with the mentioned taxon, I have chosen to leave it outside as a different species or as an unknown one.

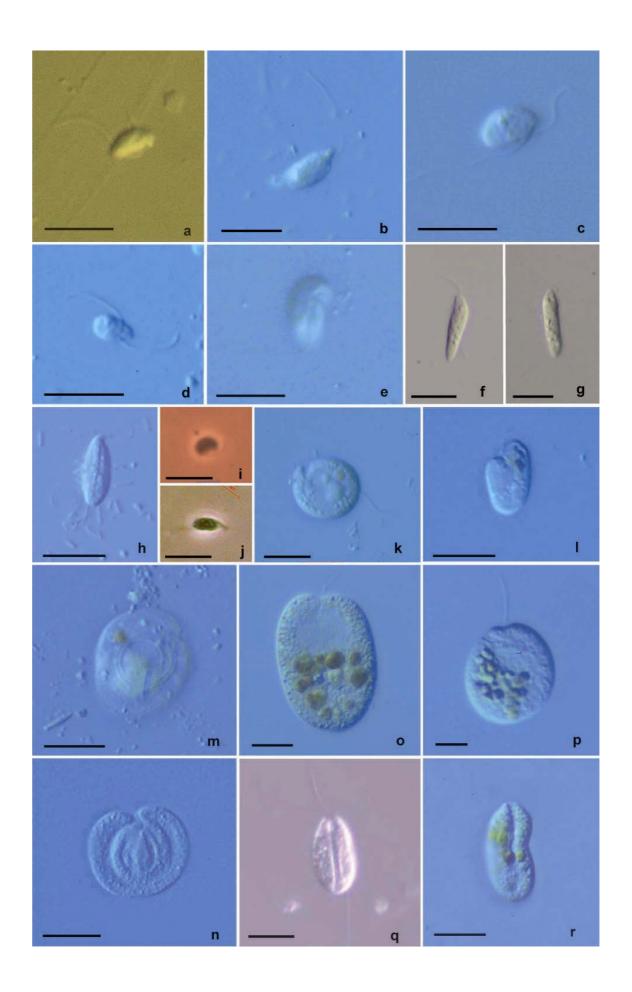
Plate 15. Insertae sedis group.

(a) Bodo designis, (b) B.cf. saliens, (c) B.sp., (d) Bordnamonas tropicana, (e) Colpodella unguis,

(f) (g) Cryptaulax cf. marina, (h) Amastigomonas mutabilis, (i) Metopion fluens, (j) Rhynchomonas nasuta,

(k) Protist 1, (l) Protist 2, (m) (n) Protist 3, (o) (p) Protaspis obliqua, (q) (r) Protaspis tegere.

Scale bar= 10 µm



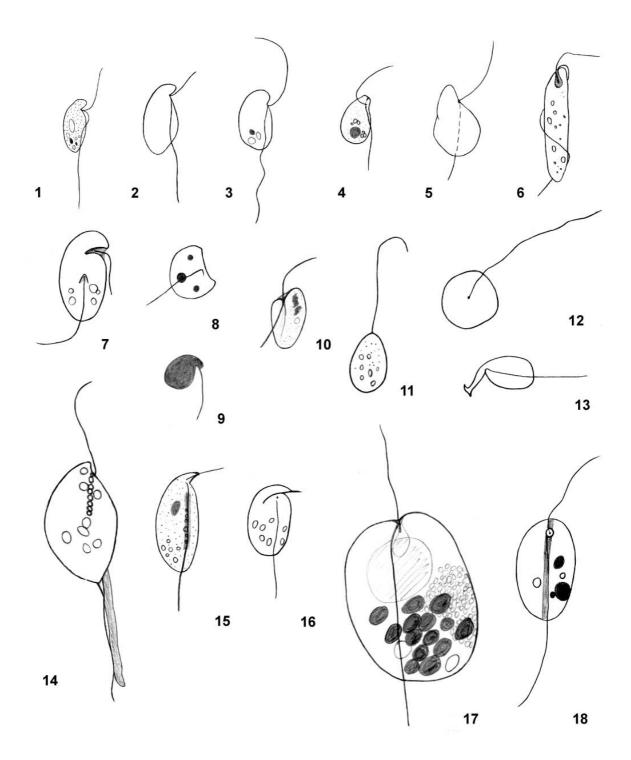


Plate 16. Insertae sedis group.

1-3 Bodo: 1. B. designis, 2. B. saliens, 3. B. sp., 4. Bordnamonas tropicana, 5. Phyllomitus granulatus, 6. Cryptaulax cf. marina, 7. Colpodella unguis, 8. Discocelis saleuta, 9. Metopions fluens, 10. Protist 2, 11. Metromonas simplex, 12. Peltomonas volitans, 13. Rhynchomonas nasuta, 14-16 Amastigomonas: 14. Protist 4, 15. A. mutabilis, 16. A. debruynei, 17,18 Protaspis: 17. P. obliqua, 18. P. tegere.

All 3000x, Protaspis 1500x

3.2 Results from the Scanning Electron Microscope

The preparations for the SEM resulted in showing few of the total cells isolated for such purpose. The majority of the cells got washed away from the physical support they were contained in, be it poly-L-lisin coverslips or be it "nucle pore" filters. Only few of them remained adhered. Usually, the biggest flagellates succeeded: gymnodinioid dinoflagellates and some euglenophytes. I found it very difficult to identify species using the SEM. The organisms showed a totally different image compared with how they looked like in the light microscope, and one needs to get aquainted with this new appearance. Among the dinoflagellates, however, four taxa could be identified down to genus level, or lower. As for euglenophytes, only *Eutreptiella* sp. was recognized thanks to its singular shape and surface striation.

Followingly, I present two plates that gather some photographs of a few flagellates in the scanning electron microscope.

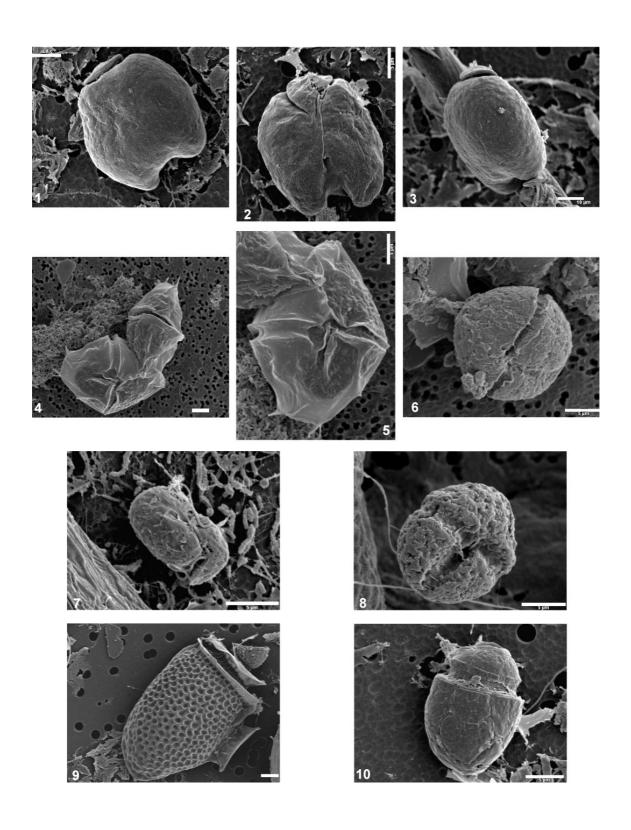


Plate 17. Dinoflagellates in scanning electron microscopy.

1-3 Amphidinium bipes, dorsal, ventral, and lateral view respectively, 4, 5 "semi-thecate" dinoflagellate, 6. unarmoured dinoflagellate: cf. *Gymnodinium/Glenodinium*, 7. *Gyrodinium* cf. *lebourae*, 8. another unarmored dinoflagellate, 9. *Dinophysis sp.* (from plankton), 10. Thecate dinoflagellate: cf. *Katodinium fungiforme*.

Scale bar= 5 µm

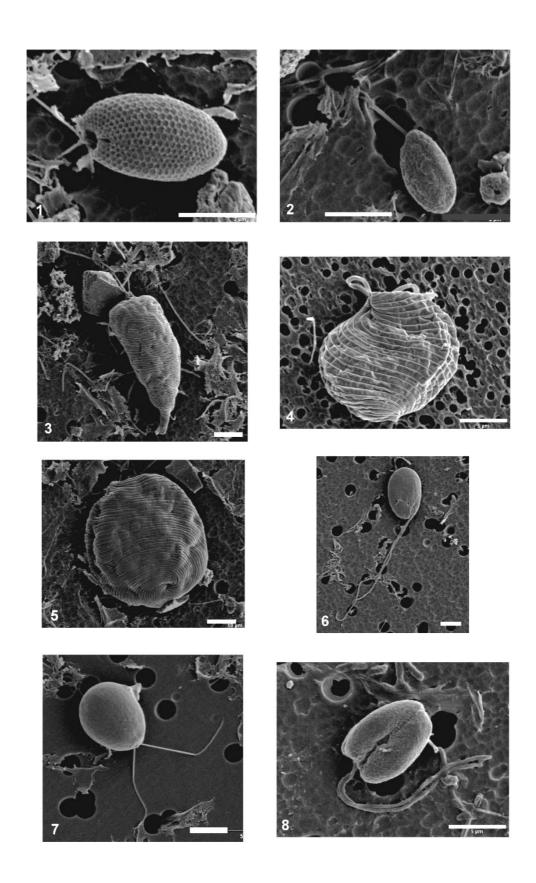


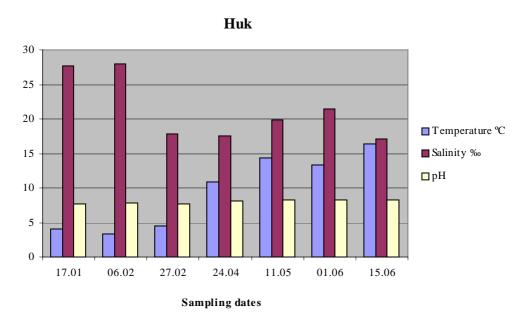
Plate 18. Cryptophytes, Euglenophytes and Chlorophytes.

1, 2. Unidentified cryptomonads, 3. Eutreptiella sp., 4. Unknown euglenoid I, 5. Unknown euglenoid II, 6. Unidentified species, 7. Chlamydomonas sp. 8. Pyramimonas sp.

Scale bar= $5 \mu m$

3.3 Hydrography

Temperature, salinity and pH data measured in the surf zones, where samples were collected, are presented here in two graphs (Fig. 22), corresponding to the two localities studied: Huk and Hulvika. While the pH shows minor variations only, temperature and salinity vary throughout the sampling period. As a general trend, temperature increases from wintertime to summer in both localities, with a remarked drop in this increase on 27th February in Hulvika. Salinity does not seem to follow any trend in neither of the two localities, especially in Hulvika.



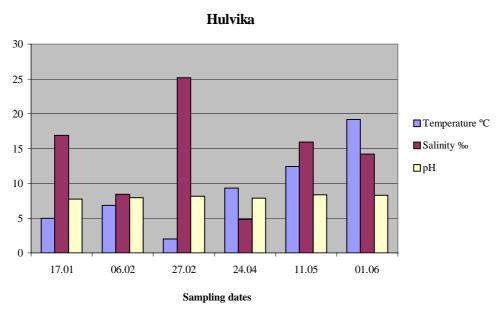


Fig. 22 Graphs showing values of measured variables in Huk and Hulvika.

3.4 Comments on the observations

As it is shown in the following species list, euglenoids in general were more varied in Huk, and the types of *Ploeotia*, *Anisonema* and *Dinema*, in particular, were more frequent than in Hulvika. The genus *Notosolenus* was, however, better represented in Hulvika. As regards dinoflagellates, Hulvika was richer, especially concerning *Gymnodinium* and *Gyrodinium* taxa, while the sand from Huk had a higher diversity of *Amphidinium* species. On the other hand, cryptophytes were more common in Huk.

HUK

HETEROKONTOPHYTA

Actinomonas mirabilis/ Pteridomonas danica

Olisthodiscus luteus

НАРТОРНҮТА

Dicrateria gilva

Prymnesium nemamethecum

CRYPTOPHYTA

Chroomonas diplococca

Chryptomonas sp

Goniomonas amphinema

G. pacifica

DINOPHYTA

Amphidinium bipes

A. britannicum (=Togula britannica, T. compacta, T. Jolla)

A. herdmanii

A. poecilochroum

A. psittacus

A. testudo

A. cf. trulla (=A. operculatum)

Gymnodinium venator(=Amphidinium pellucidum) Katodinium glandula (=Massartia asymmetrica)

Prorocentrum cf. lima (Exuviaella marina)

EUGLENOPHYTA

Anisonema cf. acinus

A. glaciale

A. prosgeobium Dinema litoralis

D. valida

D. sp.

Eutreptiella sp.

Heteronema exaratum

H. ovale

Jenningsia cf. macrostoma

cf. Metanema strenuum

Notosolenus sp

Peranema dolichonema

Petalomonas cf. cantuscygni

P. minuta

P. posilla

Ploeotia adhaerens

P. corrugata

P. pseudanisonema

P. vitrea

Urceolus cyclostomus

U. cornutus

CHLOROPHYTA

Chlamydomonas cf. nonpulsata

Pyramimonas cf. orientalis

P. sp. 1

CHOANOFLAGELLATA

Acanthocorbis sp.

INSERTA SEDIS

Bodo designis

Bordnamonas tropicana

Colpodella unguis

Cryptaulax cf. elegans Discocelis saleuta

Metopion fluens

Metromonas simplex Peltomonas volitans

Protaspis gemmifera

P. obliqua

P. tegere

Rhynchomonas nasuta

Protist 1, 3

HULVIKA

HETEROKONTOPHYTA

Olisthodiscus luteus

HAPTOPHYTA

Prymnesium nemamethecum

CRYPTOPHYTA

Goniomonas amphinema

G. pacifica

DINOPHYTA

Amphidinium britannicum (=Togula britannica, T. compacta,

T. Ĵolla)

Amphidinium cf. carterae

A. herdmanii

A. testudo

A. cf. trulla (=A. operculatum)

Glenodinium cf. foliaceum

Chilodinium cruciatum (= Gymnodinium cruciatum)

G. cf. danicans

G. variabile

G. sp.

Gyrodinium cf. estuarile

G. cf. lebourae

G. resplendens

G. uncatenatum

Katodinium cf. fungiforme

Prorocentrum cf. lima (Exuaviaella marina)

EUGLENOPHYTA

Cyclidiopsis acus

Dinema litoralis

Euglenophyta sp.

Heteronema exaratum

H. ovale

cf. Metanema strenuum

Notosolenus apocamtpus

N. urceolatus

N. hemicircularis

Peranema dolichonema

Petalomonas minuta

P. poosilla Ploeotia tenuis

Urceolus cf, cristatus

U. cyclostomus

U. sabulosus

CHLOROPHYTA

Nephoselmis cf. rotunda

Pyramimonas sp. 2

INSERTA SEDIS

Amastigomonas debruynei

A. mutabilis Bodo designis

B. cf. saliens

Bordnamonas tropicana Cryptaulax cf. marina

Cryptaulax elegans

Discocelis saleuta Metopion fluens

Metromonas simplex Phyllomitus granulatus

Protaspis gemmifera

Protaspis obliqua

Protaspis tegere Rhynchomonas nasuta

Protist 2, 4

4.0 DISCUSSION

4.1 Evaluation of the material and methods

4.1.1 Collection of the material

This was a diversity study and therefore only a few abiotic variables (temperature, salinity and pH) were measured, though with quite high precision. These variables meant to serve only for background information, that is, to reveal some general characteristics of the biotope which these organisms inhabited during the sampling period. Neither the collection of the sand nor the measurements of the variables took place at the same tidal conditions or at exact time. This might have influenced the relative composition of the sand communities as some species are more mobile than others. It is has been shown, however, that 98% of the population of flagellates is found in the uppermost 1 cm layer of the sediment at falling tide (Kingston 1999a). Since this layer has actually been wholly sampled in the present study, little or no substantial difference is to be expected from sample to sample in this respect. Anyhow, the current work never intended to present a comparative study, but rather give a general information of the flagellate diversity in the sand of the two Oslofjord beaches, regardless of seasonality, time interval or spacial dimension.

4.1.2 Extraction methodology

Due to the small size of the organisms studied, critical observations had to be done on sand free preparations. Thus the extraction of flagellates from the sand became a most important issue.

The coverslips that had remained for long in the petri dishes began to desiccate and even though water was added, the intervals between the desiccation and the watering might have caused the death of some cells. However, since the observation took place from the very first moment up to even 2 weeks after the collection, it is assumed to be enough time for having had the opportunity to observe most of the species, even those that could die from dessication later on.

The extraction from the coverslip method compared to the ice-melting method, proved to be better for the use in the light microscopy, because it was able to concentrate more cells. The ice-method appeared to extract many cells out of the sand but they were

contained in a relatively large amount of water that could not be directly used for observation in a coverslip. However, the ice-melting method succeeded in extracting the organisms into sufficiently sand and debris free water, ready for subsequent fixing procedures.

4.1.3 Identification

There has inevitably been a subjective selection as to which cells have been chosen for observation and identification. This means that species with extra difficulty in their identification have been granted less attention and therefore less effort has been spent on them. Consequently, several different taxa may have been included in a single *Genus* spp. and other species could not be identified at all, leading to an insufficient understanding of the diversity that occurs in the samples.

Genera such as *Amphidinium*, some *Gymnodinium* and *Gyrodinium*, *Anisonema*, *Dinema*, *Peranema*, *Heteronema*, *Notosolenus*, *Ploeotia*, *Urceolus*, *Protaspis*, a.o., were more readily identified than *Chlamydomonas*, *Pyramimonas* and minute specimens, which resulted in a more reduced attention to the latter ones.

On the other hand, no fixed material was used because only living material could demonstrate indispensable features like movement, behaviour and real cell shape. Experience showed that most recurrent species could be identified by a single glance and the identification verified by known details observed during shorter periods of immobility. Identification could in general not be made on fixed material as many of the salient features are lost during the procedure.

Though most of the species encountered have been recorded in similar localities elsewhere, only a few have previously been reported from the Norwegian coast. Some species as, e.g., *Cryptaulax marina* Throndsen is originally described from shallow areas in Norway (Throndsen 1969), whereas others, like *Chilodinium cruciatum* (Massart) Massart and *Cyclidiopsis acus* Korschikow are only occasionally reported globally.

4.1.4 Observations

In the following, I will present and discuss some of the general features revealed through the 5 months-period observations. These features refer to all sort of qualities of the flagellates and their distribution: characteristics, locomotion, position among flagellates, composition in different sand grain sizes, Huk versus Hulvika flagellate composition, etc.

In the first observations soon after the collection of the samples, larger euglenoids and dinoflagellates, especially those that appeared only once in the whole study, such as *Glenodinium foliaceum*, *Chilodinium cruciatum*, *Gyrodinium resplendens*, *Gyrodinium uncatenum* and *Katodinium* dominated. These taxa began, however, to diminish in number and species diversity as the sample grew older. After a few days, and until the samples were discarded, small heterotrophs began to be more common while the above mentioned dinoflagellates disappeared.

Species of *Amphidinium*, *Gymnodinium* and to a lesser extent, some heterotrophic euglenoid genera such as *Heteronema*, *Ploeotia* and *Anisonema*, showed to be extremely resistant to be sucked out with a pipette. This fact was observed when trying to isolate cells by a pipette for later fixation during the preparations for SEM. They glided or swam closely above the substratum and as soon as they "felt" a sucking current, they strongly attached to it with their posterior flagellum. Sometimes, they remained attached even after blowing them through the pipette. This may show how well they cope with the action of the waves in their natural habitat.

In general, the swimming velocities were extremely high for observation in the microscope. Dinoflagellates, such as *Gymnodinium*, *Gyrodinium* and *Katodinium*, and the cryptomonad *Cryptomonas* sp. swam rapidly, giving no chance to study them while swimming.

Different modes of movement were observed within the different flagellates in the inverted light microscope. Gymnodinium danicans, Gyrodinium cf. lebourae, Katodinium cf. fungiforme and K. glandula, all of which were relatively small cells, were among the dinoflagellates which swam and slided by rolling around at a high speed. On the contrary, species belonging to genus Amphidinium, had no spiral rotating motion pattern, they glided over the surface and when they settled or rested on the substrate, they held on to it tightly with the longitudinal flagellum. A. bipes did no rolling at all, while A. herdmanii was, among the Amphidinium species, the one rolling the most. Concerning heterotrophic euglenoids' movement, they mostly crawled by adhering and dragging their posterior flagellum or cell body on the substratum whereas some advanced with sudden pulses. This was the case of Anisonema, Dinema, some Heteronema, Notosolenus, Petalomonas, Peranema, Ploeotia, and Urceolus. The haptophyte Prymnesium nemamethecum swam by beating both flagella homogeneously by the two sides of the cell. Intermittently, they adhered to the substrate, either the coverslip or the slide, by their haptonema and many times stood "up-side-down" with the flagella extended opposite to one another, showing, therefore, the posterior end of the cell to the observer. In general, gliding over a substrate and crawling modes of movement dominated over free-swimming modes. This emphasize that these organisms are adapted to life on a solid but mobile substrate, to which they need to remain attached or alternatively to be small enough to swim freely in the interstices.

Flagellates demonstrated a disliking for mixing with others. As soon as two flagellates encountered, they turned round spasmodically and continued their way. Contrarily, pennate diatoms kept their direction and pushed until whatever the obstacle was, got run over and disappeared from their way.

Regarding predation, it was observed that ciliates housed ingested diatoms inside, which occupied a great proportion of their total cell volume. Some large euglenoids, like *Dinema*, also showed this feeding habit. Other flagellates had food vacuoles in their cytoplasms but I did never observe them ingesting other flagellates or diatoms.

When it comes to species composition in the interstitial environment, some notes can be added. It varied from sample to sample, seasonally and according to sand particle size. One of the outstanding characteristics was that heterotrophs dominated well over the autotrophs in number, biomass and species variety, especially in the winter months. In March, photosynthetic species began to appear in higher numbers, but yet not exceeding the colourless ones, except in those samples where *A. britannicum* (=Togula britannica, T. compacta, T. jolla) was present in great numbers. Among dinoflagellates, generally, those from the genus Amphidinium dominated among the Gymnodiniales, although when Gyrodinium cf. lebourae was present, it did it in so large numbers that it may have exceeded the Amphidinium. As they were much smaller, however, Amphidinium may have been dominant in biomass. Among euglenoids, Notosolenus, Petalomonas and Ploeotia were the most common genera, but they were also with the lower biomass.

According to Fernandez-Lebourans *et al.* 2003, the low biomass and diversity of the autotrophic community may be due to the limitation of light, the high consumption by the ciliates or the bacterial and viral control.

As for the trends in species composition according to particle sizes, the following feature was noticed in two samples of different sand grain size, collected same day and same place in Hulvika, close to each other. Finer sand appeared to house a higher diversity than the coarser one; more species and higher number of dinoflagellates were observed, especially from the genera *Amphidinium* and *Gymnodinium*. This fact corroborates indeed that granulometric characteristics have a marked effect on the composition of the psammobic biota and may even indicate that coarser sand is more poor both with regard to diversity and abundance, supposedly due to a lower content of particulate and dissolved organic matter.

4.2 Ecological approach to the results

The methods applied and the data obtained on the abiotic conditions do not suffice for a thourough ecological discussion of the results. Some related conclusions may, however be drawn.

4.2.1 Influence of the hydrography and other variables on the diversity

The data regarding hydrography only tells us that temperature increases from January to June which must be caused by a higher light intensity and longer photoperiod corresponding to spring and summer. As common for the coastal areas of Northern Europe there is a very marked increase in temperature from winter when air temperature is down to 10 centrigrades below zero (occasionally even lower) rising with season to about 20 degrees above zero in June. Sea water temperature will commonly be from minus 2 in winter to plus 15-20 centigrades in summer. There is usually a strong termocline in the spring-summer-early autumn which implies that the nutrients salts (from the free water masses) for autotrophic species are scarce and the sand community probably depend mostly on nutrients supplied by remineralisation in the sand. Salinity followed no trend because this variable is ruled by the input of freshwater from rain and streams and by the desiccation due to tide and waves cycle stages. For the heterotrophic benthic flagellates neither salinity nor temperature appear to influence the community, because they can tackle great variations in both variables without being affected significantly and a proof of this is that many species are reported as having world-wide distribution. However, more extensive and careful taxonomic work is required for achieving reliable biogeographical generalizations (Patterson et al. 1989).

A noted increase in relative numbers of autotrophic species was evident through the sampling period. This fact, can be explained by an increase of light intensity and daylength and a subsequent increase of photosynthetical capability. Alongside, this would lead to a higher production of oxygen in the interstices of the sand and therefore a more oxygenic environment. From the information in Patterson *et al.* (1989) it seems evident that oxygen and light are the main driving forces in the succession of species from low abundance and domination by heterotrophic species to higher abundance and increasing importance of autotrophic forms.

4.3 Concluding remarks

The present investigation showed that the biotopes sampled were rich in species diversity, and continued sampling would most probably have revealed more species in addition to those which were just observed, but not identified this time. The observed diversity was dertermined not only by the quality of the sample, but also by the conditions related to the methods applied for observation. Thus it is important to consider the conditions met by the flagellates in the Petri dishes and during the ice method extraction. The success of the coverslip method probably depends on an early establishment of a bacterial community on the coverslip surface, seeded from the sand, and serving as food for the grazing heterotrophic flagellates, This also explaines why it takes some time before the flagellates collect on the glass (Jacob Larsen pers.com.). The coverslip method worked well to give preparations suitable for light microscopy and revealed the great species diversity in the samples. In order to provide material for detailed studies of the flagellate constituents of the psammobic communities by the ice extraction method, the material either has to be gently concentrated by mechanical means (e.g. centrifugation or filtering), or even better brought into unicellular culture.

For future studies with electron microscopy, molecular or chemical analysis included, however, it is highly needed to develop more efficient extraction methods.

5.0 SUMMARY

The present thesis seeks to document the diversity of the flagellates in some sediments of the Oslofjord. The analysis of these sediments lasted from January 2005 until June of the same year. The sand samples were collected from the surf zone, that is, the wave zone, of two beaches: Huk, in the Bygdøy peninsula, and Hulvika, south to the city of Oslo. Huk was sampled seven times, whereas, Hulvika six times. Along with the sediment samples, three variables were measured: temperature, salinity and pH.

Samples were immediately transferred into plastic petri dishes and coverslip put on top. These coverslips were examined in the light microscope for identification of species. Besides, some samples were processed for extracting the organisms by means of the so-called ice-method. The water extracted by this method would be employed for the preparations for scanning electron microscopy.

All in all, 85 different flagellate taxa were found in the two localities, 71 of which were identified down to species level in the light microscope. Of the total taxa 22 were autotrophs, while 63 were heterotrophic species. In general, the autotrophs began to increase in species diversity and abundance with the arrival of spring and summertime. The Huk location showed a higher diversity in euglenoids than Hulvika. Finer sand proved to contain more species of some dinoflagellate genera as compared to coarser sand.

As for the variables measured, temperature followed an increase with season, while salinity showed an irregular pattern. No correlation could be demonstrated between the occurrences of species, both abundance and diversity, with the environmental variables measured. Most probably, increase of light due to seasonality and consequently a higher oxygen concentration, were the causes for the succession of species from low abundance and heterotrophic specimens to higher abundance and autotrophic specimens.

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