Classical and Modern Criteria for Determining Species of Cryptophyceae

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Abstracts

The criteria for discrimination between species of Cryptophyceae are briefly reviewed. New observations on morphological variation and the effect of fixatives are included. The present knowledge allows a better use of morphological features like the chloroplast, pyrenoids and periplast structure, and a criterion like the phycobilin-pigments, in species determination. All criteria in use are included in a check-list.

Flagellates belonging to the Cryptophyceae are among the most common phytoplankton algae in lakes, in brackish waters, in the marine coastal waters and in the open sea. Due to their fragility and sensitivity towards chemical fixatives, and their lack of conspicuous discriminating morphological features, they are frequently missed or left out from routine phytoplankton surveys. Since their systematics at species level is inadequate, there are artificial, subjective or situation-dependent distinctive criteria between "species." This lack of conspicuous morphological features frequently has led the practising phytoplankton workers to classify them among "flagellates" or "sp. indet."

The Cryptophyceae are certainly of significance for the productivity and the pelagic food chains of the lacustrine and marine environments (e.g. HOLLIGAN & HARBOUR 1977, GILBERT & BOGDAN 1981, THRONDSEN 1983 and references in KLAVENESS 1984). Accordingly, the questions of their speciation and variability are not only of systematic, taxonomic, evolutionary and biogeographical interest, but also a prerequisite for furthering progress in pelagic food web research, where the microflagellates presently are in focus (e.g. PORTER et al. 1985).

Recent progress in research on cryptomonads has most notably included: their fine structure, with its systematic and evolutionary significance (relevant reviews are those of GANTT 1980, MELKONIAN 1984, SANTORE 1984, — new contributions include those of MEYER & PIENAAR 1984a, b and SANTORE 1985); their pigments (review by GANTT 1979 — new applications by GIESKES & KRAAY 1983, STEWART & FARMER 1984); their role as endosymbionts (OAKLEY & TAYLOR 1978, GRAIN et al. 1982, WILCOX & WEDEMAYER 1984); and their physiology (ANTIA 1980).

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There are, however, less information available that ties up the accumulated knowledge with recent discoveries and the implications for practical systematic work at species level. Neither geographically nor ecologically has the distribution of the Cryptophyceae been reviewed. This knowledge is necessary to estimate their relative importance for the food-chains in the different ecosystems of the world. This paper briefly reviews information available for determining to species the pelagic marine and freshwater representatives of Cryptophyceae. The purpose is to stimulate interest and prevent further negligence of these interesting and probably important links in all pelagic food-webs.

Materials and Methods
Where the original work is presented, the methods are based upon standard techniques for light- and electron microscopies (not always considered sufficient for cryptomonads, according to SANTORE (1984)). Culture methods include the medium of GUILLARD & LORENTZEN (1972), or enriched seawater (stored seawater with the same nitrogen, phosphorus, trace metal and vitamin enrichment). The pigment spectra are made as in KLAVENESS (1982): based upon the recommendation of GANTT (1980) and done by the method of SIEGELMAN & KYCIA (1978). Light photomicrographs were taken with a dry 40× Zeiss Neofluar objective, employing a flash at 1/1500 sec.

Cultures are held in the author’s laboratory in Oslo. Investigators are welcome to have any culture sent for corroborating results or for their own research, provided they are able to keep them in culture. This is usually not very difficult, but as my strains mostly are new and recent isolates, some require special care and frequent attention. Just for this reason, they ought to be far more interesting than the old culture collection strains that have been held in culture for (sometimes) half a century.

Criteria for Discrimination at Species Level
Cell shape distinguishes the class from all other flagellate algae, and in cryptomonads “the general cell form is the most important feature for the determination of species” (JAVORNICKY 1971). A source of some confusion may be inconsequent use of terminology between authors. The terminology of the cryptomonad cells, as clarified by BUTCHER (1967, his fig. II), is recommended and may be repeated from Fig. 1. The distance between the lateral sides is the cell width, the distance between dorsum and venter is the cell thickness. Cell length is the linear distance from apex to antapex. The reniform type of cells, like Protochrysis Pascher and Sennia Pasch. (=Heniselmis Parke) may be included in this series as apically-antapically compressed cells (Fig. 1D).

PRINGSHIEIM (1963, 1968) repeatedly called attention to the twisted or screw-like structure that may be present in living cells (e.g. CONRAD 1942, Cryptomonas obtorta; C. borealis in CAMPBELL 1973, or C. curvata in FOTT & ETTL 1959). This twisting may disappear upon fixation (PRINGSHIEIM 1963). The degree of curving of cell body is not a constant feature (see PRINGSHIEIM 1968).
Plate I shows cells from a culture of a *Cryptomonas* sp., isolated as a single cell in 1980. The upper two rows are living cells viewed from the lateral (upper) and dorso-ventral (second row) aspect, respectively. When growing, the cells are rather elongated, but of variable shape (left). When growth ceases, resources are redistributed between cell compartments; cells attain a different light transmission and refraction, cell shape may change drastically and some cells may proceed toward what appears to be a resting stage (right). Similar stages are found in natural habitats; deviating cell shapes may have been described as separate species (e.g. *Cryptomonas globosa*, Christen 1958).

![Diagram of cryptomonad cells](image)

Fig. 1. Views of different types of cryptomonad cells.
A: An uncompressed cell as it turns on its vertical axis: 1, ventral; 2, lateroventral; 3, lateral; 4, dorsal; 5, dorso-lateral; 6, ventro-lateral.
B: A dorsally compressed form: 1, ventral; 2, latero-ventral; 3, lateral.
C: A laterally compressed form: 1, ventral; 2, latero-ventral; 3, lateral.
D: Reniform type of cryptomonad cell, an apically-antapically compressed form: 1, dorsal; 2, lateral.
A-C, from Butcher (1967, fig. II, reprinted with permission of the Controller of Her Majesty's Stationery Office; British Crown Copyright). D, original.

Third row of Plate I shows cells fixed by adding a drop of osmium tetroxide solution (2%) to 10 ml of culture. The first three pictures show the lateral aspect, and the three next the dorso-ventral aspect. Although cells may approximately retain their shape, the general impression is that the cells swell slightly and thereby change their general appearance. The same seems to be true for cells fixed by the conventional Lugol's solution as used for quantitative phytoplankton work (lower row). Apart from being stained strongly by the fixative, the cells appear swollen compared to the live cells, both from the lateral (Plate I, a, t, u) and dorso-ventral (Plate I, v, w, x) sides. Fixation also changes or obscures the appearance of important cytological details (see below).

Cell size is variable, even within clonal cultures, due to the unequal division. Willén et al. (1980) found a pronounced size and form variation during the year for *Rhodomonas* species in lakes Vättern and Mälaren, and the cells differed in size also between the two lakes.
Also the cell width is "to some extent influenced by the state of nutrition" (Pringsheim 1968). Anton & Duthie (1981), in their analysis of a large, Lugol-fixed Canadian material, found the length/breadth ratio (L/B ratio) a "significant taxonomic character" (and critical for certain species), in spite of avoiding to differentiate between width and thickness of cells. Presumably, the ratio may tend to be fairly constant for a given species, since fixed cells of a kind mostly tend to come to rest in the same position. Cell volumes, for biomass estimation, are commonly calculated from closest geometrical shapes, or approximated therefrom (Willén 1976).

Resting stages or resting spores in the Cryptophyceae have been observed occasionally. The earlier observations are reviewed by Ettl et al. (1967). Further records are those of Wawrik (1969, 1971), Javornicky & Hindak (1970) and Heyning (1976). Lichtlé (1979, 1980) studied the effect of external conditions upon encystment and excystment in Cryptomonas rufescens; the only modern experimental study of the subject. Nitrogen deficiency and a high level of illumination may induce formation of the resting stage. The cysts may germinate under favorable conditions, each giving rise to two or four fully differentiated Cryptomonas cells (Lichtlé 1980). Whether the ability to form morphologically differentiated cysts is a species-specific property remains to be shown. So far, the presence or absence of cysts has been of little value in the efforts of identification to species level.

Formation of palmellae or non-motile stages enclosed within a common polysaccharide jelly has, on the other hand, been treated as a property specific to some species. Cryptomonads are frequently said to become more or less immotile and embedded in a jelly at the onset of cell division, e.g. C. ovata, obovata, platyuris, tetrapyrenoidosa, parapyrenoidifera, etc. (Huber-Pestalozzi 1950). There are, however, cryptomonads that are permanently embedded in a jelly in the vegetative stage, and rarely present motile cells. This is the case for several soil-living strains isolated by Hindak (pers. comm.). In oligotrophic lakes, colonies of two or four cells embedded in a jelly are not uncommonly encountered (Gran & Ruud 1927, Klaveness unpubl.). Santore (1978) briefly reviews the literature on the formation of palmellae in Cryptophyceae.

Shape and size of gullet are also frequently invoked as a significant taxonomic feature. Pringsheim (1968) was critical to the use of gullet length, a viewpoint supported by Anton & Duthie (1981). According to Butcher (1967), "the nature and structure of the depression-furrow-gullet system form the only sound basis for primary classification." Hence, in his fundamental paper, genera are classified according to furrow/gullet and number of trichocyst rows. The location of ejectosomes in the gullet may be a specific property (cf. C. ovata in Butcher 1967).

In spite of the electron microscopical approach, there are still some ambiguity with regard to furrow/gullet relations in some smaller species due to problems of fixation. Santore (1984) believes that a furrow is an artefact; a rupture along the gullet. Klaveness (1981) and Gillott & Gibbs (1983) found that some species do have a ventral furrow leading into the gullet region, in agreement with the observations of earlier, critical light microscopists. Plate
II, e shows the ventral aspect of *Cryptomonas curvata* Ehr. (= *C. rostratiformis* Skuja), where a furrow is apparent.

Also flagellar length is known to vary among species (Butcher 1967, Pringsheim 1968), although "some species are characterized by a definite flagella to cell length ratio" (Anton & Duthie 1981). Butcher's (1967) positioning of the flagellar insertion at the base of the furrow-gullet ("at least in some cases"), is not the case for most of the species investigated by electron microscopy so far. The vestibular region is the site of flagellar insertion — "emerging from the sub-apical furrow opening on the ventral side" (Santore 1985).

Number and position of chloroplasts have been subject to discussion due to the frequent observations of a bridge between the two main chloroplast bodies in *Cryptomonas* spp. (Plate I, k). Pringsheim (1968) seems to agree with Hollande (1942) and Joyon (1963) that there may be only one chloroplast (with a narrow bridge), and that the absence of a bridge between the two chloroplast halves may indicate a cell division forthcoming (cf. also Fott & Etzl 1959, Jawornicky 1971, 1978). Santore (1985), however, is of the opinion that there are two chloroplasts in *Cryptomonas*, documented by his observations of two nucleomorphs in each cell. Etzl (1980b) was able to list a number of common species with a single bilobate chromatophore, and two chromatophores, respectively.

A previously unnoted lateral positioning of chloroplasts led Frisch (1914) to describe a new species, *Cryptomonas anomala*. This position of chloroplasts is not at all anomalous, as it occurs in numerous other species as well (cf. Huber-Pestalozzi 1950, Jawornicky 1976, Etzl 1980a, b, etc.). The chloroplast position should, however, be carefully noted when studying cryptomonads.

Absence or presence of pyrenoids and their number are the main specific character of several species of *Cryptomonas* (e.g. "tetrapyrenoidosa"), while a pyrenoid is a fairly regular feature in those smaller genera with one dorsal chromatophore only (*Rhodomonas, Chromonas*). The pyrenoids need careful identification by means of good optics; there are several very obvious misidentifications in the literature. Pyrenoids are more or less bulging from the inner surface of the chloroplasts. Starch grains or shells around pyrenoids may be identified, but they do not always give the expected blue tint when stained with iodine (a reddish-brown color is common as well), and in cells with abundant starch the product is located all along the chloroplasts and not only in the proximity of pyrenoids. Etzl (1980a) recommended staining with Azocarmine G to enhance details of the pyrenoid. Figure 2 gives an overview of pyrenoid shapes found among Cryptophyceae.

"Maupas' ovals," two strongly light-breaking bodies frequently found inside the *Cryptomonas* cell, are probably of lysosomal nature (Lucas 1970). Their recognition is dependent upon the cell's growth conditions (Pringsheim 1968). Their presence or absence is thus not a reliable feature for systematic purposes. Santore (1984) discusses the "Corps de Maupas," and suggests the term to be abandoned.

"Hansgirg (1885, p. 230) was the first to create confusion by the use of color as a generic character in Cryptophyceae" (Pringsheim 1944). Cell color has been rejected as a
Fig. 2. Schematic overview of pyrenoid types found among Cryptophyceae.
Chloroplast membrane arrangements are indicated inside cells by thick line (chloroplast envelope pair of membranes) and thinner line (chloroplast endoplasmic reticulum pair of membranes). Dorsal (d), ventral (v) and lateral (l) sides are indicated. Starch grains are shaded, nucleomorph may be indicated as dotted circle.

A-C: Cells with one dorsal chloroplast, seen in optical section from the latera.
D-G: Cells with one or two chloroplasts, seen in optical section from the ventral side.
D: Cell similar to C (turned). E: The pyrenoid, with nucleomorph in center, forms a bridge between the two lobes of a single plastid ("Pyrenomonas," SANTORE 1985).
F: Cell with two chloroplasts (or two chloroplast lobes), each with "bulging" pyrenoid.
G: Cell with several smaller, less well defined pyrenoids, not easily recognized by light microscopy (KLAVENESS unpubl., see also SANTORE 1985).

useful taxonomic character by recent authorities (BUTCHER 1967, PRINGSHEIM 1944, 1968) due to great variability between strains/samples and with growth conditions and age of culture. But progress in pigment analysis may revive biliprotein pigment composition as a useful character among the Cryptophyceae. GANTT's (1979) review is a good starting point. At least five different biliprotein pigment groups may be discerned. Since a crude buffer extraction (SIEGELMAN & KYCIA 1978) yields a pigment extract adequate for identification to one of the above groups, the technique should be applied whenever a culture is established (GANTT 1980). It remains to be seen whether a genus like Rhodomonas (rejected by BUTCHER 1967) may be retained on a biliprotein pigment composition different from other cryptomonads. PRINGSHEIM (1968) remarks that variations in the phycobiliprotein pigmentation is not considered as a valid taxonomic feature at generic level in blue-green and red algae. But, contrary to these groups where different shades or colors may develop as a result of variations in relative amounts of at least two different biliprotein-pigments (e.g. KLAVENESS & SKULBERG 1962), the cryptomonads always possess only one phycobiliprotein pigment (GANTT 1979). Figure 3 shows diffuse light spectral scans of living cells, and conventional absorbance spectrophotometer scans.
Fig. 3. Pigments in Cryptophyceae as indicated by simple spectrophotometric methods.

A: Diffuse light spectral scans of cells from pure cultures of *Rhodomonas* sp., *Chroomonas* sp. and *Cryptomonas* sp. The spectra reflect the absorbances of chlorophyll *a* (ca. 435 nm and 676–679 nm), carotenoids (ca. 460 and 480–490 nm), phycoerythrins (ca. 550 nm (*Rhodomonas*) or 566 nm (*Cryptomonas*)) and phycoeyanin (*Chroomonas*, 585 and 646 nm).

B: Conventional absorbance spectrophotometry scans of phosphate buffer extracts of biliprotein pigments from same cultures. The *Rhodomonas* extracts has a wide peak at about 550 nm, *Cryptomonas* at 566 nm and *Chroomonas* two peaks at 585 and 646 nm.

of phosphate buffer extracts, from three different cultures in my possession. Unfortunately, the different kinds of pigments are not necessarily specific to different genera.

Chlorophylls and carotenoids need more elaborate processing (chromatography) to be identified, and there have so far been few indications that they are of practical taxonomic value within the class. The carotenoid alloxanthin is peculiar to the class Cryptophyceae (see LIAAEN-JENSEN 1978), and has been used in practical marine work to identify their relative contribution to the plankton community (GIESKES & KRAAY 1983).

The structure of the cell’s periplast may be distinctive (GANTT 1971) and different types may be recognized by scanning electron microscopy (Plate II): rectangular or hexagonal plate-patterns (SANTORE 1977), papillate (MUNAWAR & BISTRICKI 1979, figs. 5–7, KLAVENESS 1982, Plate II, e in this paper) or smooth as in *Katablepharis* (MUNAWAR & BISTRICKI 1979, fig. 12, corroborated by unpublished Norwegian material) or the tiny *Hillea marina* Butcher as pictured by CHANG (1983).
### TABLE 1. CRITERIA EMPLOYED FOR IDENTIFYING CRYPTOPHYCEAE TO

**Light microscopy**

- **Cell shape (seen from lateral)**
  1) a. spherical  
  b. ellipsoidal  
  c. ovoid (=ovate)  
  d. obovoid (=obovate)  
  e. cylindrical  
  f. lanceolate  
  g. fusiform  
  h. reniform  
  i. other

- **Cell length, μm (median of 30 cells)**
  3) a. < 5  
  b. 5-9  
  c. 10-14  
  d. 15-19  
  e. 20-24  
  f. 25-34  
  g. 35-44  
  h. 45-60

- **Cell width**
  4) a. more than cell thickness  
  b. about equal to cell thickness  
  c. less than cell thickness

- **Length-breadth ratio (Lugol-fixed cells)**
  5) a. <1.2  
  b. 1.3-1.5  
  c. 1.6-1.9  
  d. 2.0-2.3  
  e. 2.4-2.6  
  f. 2.7-2.9  
  g. 3.0-

**Palmellae**

- 6) a. permanently present  
  b. occasionally present  
  c. not observed

**Resting stages**

- 7) a. observed  
  b. not observed

**Flagella**

- 8) a. nearly of equal length  
  b. of unequal length

**Flagella/cell length ratio**

- 9) a. >1  
  b. ca. 1  
  c. 1/2-1  
  d. <1/2

- **Gullet/cell length ratio (when applicable)**
  10) a. >1/2  
  b. ca. 1/2  
  c. <1/2

- **Ejectosomes in furrow-gullet region**
  11) a. one transversal row or annuler  
  b. two longitudinal rows  
  c. three longitudinal rows  
  d. several rows (oblique)

**Position of ejectosomes in gullet (when present)**

- 12) a. in upper gullet only  
  b. in whole gullet region  
  c. in lowermost part of gullet only  
  d. otherwise

- **Maupas' ovals**
  13) a. clearly visible  
  b. not visible

- **Chloroplast number**
  14) a. 0  
  b. 1  
  c. 2  
  d. several

- **Chloroplast shape**
  15) a. entire  
  b. two lobes  
  c. irregular

- **Chloroplast location**
  16) a. dorsal  
  b. dorso-ventral  
  c. bilateral  
  d. otherwise

**Pyrenoids**

- 17) a. 0  
  b. 1  
  c. 2  
  d. 3-5  
  e. several

**Starch grains associated with pyrenoid**

- 18) a. several and irregular  
  b. few and regular  
  c. two cup-shaped  
  d. one cup-shaped  
  e. starch not present

**Visual colour or tendency of cell (bright field)**

- 19) a. red  
  b. brown  
  c. yellow  
  d. green  
  e. blue
SPECIES LEVEL. CATEGORIES ARE SUGGESTED FOR EACH CRITERION.

f. colourless
Lipoid vesicles
20) a. absent
    b. apical
    c. antapical
    d. both
    e. otherwise

Scanning electron microscopy
Periplast structure
21) a. hexagonal plates
    b. rectangular plates
    c. papillae
    d. smooth
    e. otherwise

Gullet opening
22) a. absent
    b. apical
    c. lateral
23) a. via furrow
    b. without furrow
    c. furrow only

Transmission electron microscopy
Periplast structure (in section)
24) a. normal
    b. deviant

Mastigonemes
25) a. not present
    b. on one flagellum only
    c. on both flagella
26) a. similar on both flagella
    b. unequal

Flagellar scales
27) a. absent
    b. present

Flagellar root system
28) a. with rhizostyle, passing nucleus in groove
    b. without rhizostyle

Thylakoids per lamella (typical)
29) a. one
    b. two
    c. three
    d. variable

Nucleomorph
30) a. present
    b. not found

Pyrenoid shape
31) a. irregular
    b. bulging
    c. pyriform

Chemical methods
Phycobiliprotein pigment
32) a. none
    b. PE I
    c. PE II
    d. PE III
    e. PC I
    f. PC II
    g. other

Ecology and growth characteristics
Habitat
33) a. soil
    b. marsh
    c. pond
    d. lake
    e. brackish water
    f. inshore/neritic seawater
    g. oceanic
    h. other
34) a. oligotrophic locality
    b. mesotrophic locality
    c. eutrophic locality
    d. dystrophic locality
    e. saprobic locality
    f. other

Temperature (°C)
35) a. < 5
    b. 6-10
    c. 11-15
    d. 16-20
    e. 21-25
    f. > 25

Salinity (%)
36) a. < 1
    b. 1-15
    c. 16-35

pH
37) a. < 5.0
    b. 5.1-7.0
    c. 7.1-9.0
    d. > 9.1

Water colour (mg Pt/l)
38) a. < 15
    b. 15-40
    c. > 40
Among the periplasts with plate-patterns, there appear to be two different types. Type I has a thin dense plate just beneath and closely adjacent to the plasmalemma and an electron-opaque layer of similar width and density located outside the plasmalemma. Type 2 has plates 5-15 nm in width located a short distance below the plasmalemma (see GANTT 1979, GILLOT & GIBBS 1983). The papillate periplast may be a modification of type 2: a thin plate appears a short distance below the plasmalemma (KLAVENESS 1982, unpubl.). Cyathomonas may have a further modified periplast (HAUSMANN 1979). This species is phagotrophic and may take up and digest bacteria (MIGNOT 1965, SCHUSTER 1968, CYNAR & SIEBURTH in prep., KLAVENESS unpubl.) When studied by the scanning electron microscope, the cellular surface reveals plate structure (MARGULIS & SCHWARTZ 1982).

Artefacts due to problems of fixation can not yet be excluded (e.g. a slight shrinkage might exaggerate subperiplastidal structures normally not visible on perfectly fixed cells), but results so far indicate the periplast structure to be of high potential interest to the taxonomist. Also the "papillate" periplast structure may be a transient feature, due to the method of fixation. But in some large species the proteinaceous plates appear in section to be very thin and inconspicuous, thus rendering the periplastidal euctosomes protruding as "papillae."

Among the marine species of Cryptophyceae, few have been depicted by scanning electron microscopy, even among those that have been in culture. Some marine investigators have successfully included scanning electron microscopy of their samples; these papers may be consulted for techniques of sampling and fixation (e.g. BOOTH et al. 1982, CHANG 1983). As with freshwater species, some specialists may identify marine representatives to species level by light microscopy alone (e.g. THRONDSEN 1969, 1983), but electron microscopy is needed to identify additional distinctive criteria.

HIBBERD (1979) and MOESTRUP (1982) have reviewed different features of the cryptophycean flagella, and recently PENNICK (1981) and SANTORE (1983) called attention to what appears to be organic scales on the flagella. The garniture of mastigonemes and scales may be different between species, and of potential value to taxonomy when a larger number of species have been studied.

Other features, specific to the Cryptophyceae in general, may be the mode of nuclear division (OAKLEY 1978), the nucleomorph (GREENWOOD 1974, GILLOT & GIBBS 1980, MCKERRACHER & GIBBS 1982), the mitochondrial reticulum (SANTORE & GREENWOOD 1977), etc. At present it is difficult to connect taxonomy at species level to these features, unless rather extravagant techniques are called upon. Contractile vacuoles are easily seen in freshwater species. Some strains of Chroomonas (see SANTORE 1984) and Cryptomonas (HINDAK pers. comm.) may have a distinct eyespot.

Table 1 is a check-list of classical and modern diacritical features in use, with options suggested for each feature. In practical work, all available information are recorded although a number may be of very limited or no use. It is the skilful and experienced taxonomist's task to distinguish between the useful and the useless.
Discussion

On dealing with phytoplankton for biomass estimation or in a community context, it is embarrassing to find oneself left with a large group of phytoplankton as “undetermined flagellates,” “sp. indet.,” etc. The inclusion of such an indetermined group in plankton lists is considered “acceptable,” but certainly do not further the progress of knowledge. As a considerable number of the “indeterminable” species may belong to the Cryptophyceae, a presentation of available criteria may aid in the selection, and a future crystallization, of taxonomic concepts useful in practical work. When necessary precautions are taken during sampling and fixation, and the natural variation is adequately mapped, the Cryptophyceae may be no more difficult to deal with than dinoflagellates.

Among Cryptophyceae, authors have described dozens of “species,” only mere ecotypes, local variants, temperature modifications, infected cells or culture strain variants. Too little attention has been paid to the natural size- and shape-variability. A modern taxonomist must apply all means available to elucidate the total variation encountered in nature, and in a modern sense also the potential in the strain or clone’s genetic set-up. But, beware of describing types from entirely artificial conditions, under extreme influences or suffering from decades of isolation under “optimal” conditions (in a culture collection, for example). The type should, ideally, represent the momentum of all the specific potential variability, from which all other entities belonging to that species may be deduced; the type should be described from its “most natural habitat.”

The total number of valid species of Cryptophyceae will drop when the existing species are screened critically. New species should not be launched unless very thorough investigations have been undertaken.

The fundamental problem of speciation in apparently asexual organisms has not so far been ready for discussion in the case of the Cryptophyceae—the biological species concept, defining a species as the members of a common gene pool, is difficult to apply. This “species problem” is discussed at length in the literature, the situation for cryptomonads are symtomatic for numerous protozoans in general. At least some well defined morphological species are widespread in nature; “in between” are numerous “species” with less distinct morphology, apparently forming a continuum, at least between some morphotypes. It is astounding and puzzling that some of the most distinct species among freshwater forms, confined to isolated bodies of water on different continents, may be recognized morphologically as the same taxonomic entity. Have they evolved an ability to retain their genetic make-up in spite of being asexual? Or have common and widespread sexual processes barely been observed (cf. WAWRIK 1969)?

HANSON (1977) discusses the problems of speciation in asexual Protozoa, and cites MAYR (1963), whose provocative concept of “ecospecies” may well apply for the Cryptophyceae. At the present state of knowledge, the recognized species may as well be survivors from a continuum of forms, clustered around adaptive peaks—“the ecological factors have given the former continuum a taxonomic structure.” This situation may change as soon as culture tecniques and experimental investigations are applied.
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Explanation of Plates

Plate I. Cells derived from a single cell isolate of Cryptomonas sp.
a-f: Living cells from one of the laterals.  g-l: Dorso-ventral aspects of living cells.
Note the two chloroplast lobes, a "bridge" between them is visible in k.  m-r: Cells
fixed with osmium tetroxide, lateral aspects (m-o) and dorso-ventral aspects (p-r).
s-x: Cells fixed with Lugol’s solution, oriented in the same manner: s-u lateral, v-x
dorso-ventral.  Bar (lower left) = 10 μm.

Plate II. Scanning electron micrographs showing the periplast structure of cryptomonad cells.
a: Chroomonas sp with rectangular “plate” structures.  As is shown in higher magnifi-
cation in d, the “plates” have a rugulose surface.  b: A large cell of Cryptomonas
curvata (= rostratiformis) with no plate structures of the periplast, but showing a papil-
late or “warty” structure.  Details at higher magnification is shown in e, where also
the apical vestibulum elongated into a furrow, is shown.  c. Another example of Chromo-
monas sp. with hexagonal plate structures on the cell’s surface.  a, c and d: bar = 1 μm,
b and e: bar = 5 μm.