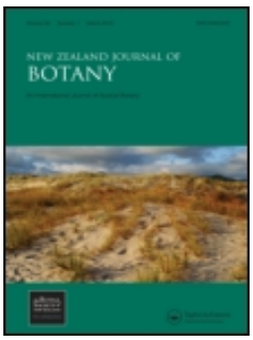

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SOME NEW AND UNCOMMON *CHLAMYDOMONAS* SPECIES FROM NEW ZEALAND

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SUMMARY

Seven taxa of *Chlamydomonas*, including two new species, are described. The taxa were recovered from soils in New Zealand, from Macquarie Island, and from sediment in an unnamed lake on the Antarctic Continent.

INTRODUCTION AND METHODS

Until recently only five species of *Chlamydomonas* had been recorded in New Zealand, all from aquatic habitats (Chapman *et al.*, 1957), but during a study of the algae in soils from New Zealand, the Subantarctic Islands, and the Antarctic Continent, *Chlamydomonas* species were isolated from a few of the samples and grown in unialgal cultures. They include the following taxa—two are new species, three are forms of species known from English soils, and two are widely distributed in aquatic and terrestrial habitats—

Subgenus *Chlamydomonas*

Chlamydomonas chlorostellata n. sp., on Tekoa soil (N.Z.). Forms of this species were found in a rainwater puddle in Christchurch and in soil from Green Valley in Spit Bay, Heard Island, in the South Indian Ocean;

C. subangulosa Fritsch & John forma, on Ahuriri soil (N.Z.);

C. reinhardti Dang., on Tekoa soil, at depth of 3 cm;

C. snowiae Printz, from sediment in an unnamed Antarctic lake.

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*Working for Soil Bureau at the University of Canterbury, Christchurch, under a grant from D.S.I.R.

Subgenus *Chlamydella*

C. rima n. sp., on Waiareka soil (N.Z.);

C. macrostellata Lund forma, on Macquarie Island;

C. moewusii Gerloff forma *microstigmata* Lund, on Temuka soil (N.Z.).

Apart from the sediment from the Antarctic lake, all the soils were covered by indigenous or induced grassland and the samples were collected as cubes (about $15 \times 15 \times 8$ cm); they were cut out of the turf or peat with a sterile knife, packed in plastic bags and kept as cool as possible during the journey to Christchurch. Tekoa soil was examined in greater detail than the others; turf was collected in spring, summer, and autumn 1956 and 1957 in an undisturbed state with a cylindrical soil sampler; the cores were 10 cm in diameter and 8 cm deep. Algae on the surface were studied and also those at 3 cm, the soil at this depth being removed from each core with a sterile cork-borer. The New Zealand specimens were examined and cultured a few days after they were collected, but material from Macquarie Island and the Antarctic Continent was received three or four months after it had been collected.

Soils were examined by direct observation. Sub-samples were inoculated on to nutrient agar, or placed in covered petri dishes and moistened with nutrient medium (Flint, 1958). *Chlamydomonas* spp. were isolated from these plates and resulting unialgal cultures were grown in Detmer's medium (Bold, 1942), on the medium that Pringsheim designed for *Micrasterias*, in soil/water medium (Pringsheim, 1946), or in Juller's solution (as modified by Pocock, 1960). Soil extract (De, 1939), micro-elements (Miller and Fogg, 1957) and iron in EDTA (as sodium iron complex) to give a concentration of 1 p.p.m. Fe, were added to each medium. At first the cultures were grown at room temperature in indirect daylight, but since April 1961 they have been kept in an insulated room where the temperature varies between 20° and 21°C and illumination is provided for 16 hours daily by two fluorescent tubes (5 ft, 80 W, white or warm white tubes) and one 15 W incandescent bulb, which together give a maximum intensity of 212 foot-candles. Slopes of the unialgal cultures were sent by airmail from New Zealand to Czechoslovakia where they were grown in Czurda's standard medium with soil-water (Ettl, 1959), one series of cultures being illuminated by daylight and the other with two 40 W fluorescent tubes. No attempt was made to prepare bacteria-free cultures.

Chemical analyses of New Zealand soils were carried out by the Soil Bureau according to the methods described by Metson (1956) and results are expressed on the basis of oven-dry weight of soil.

The geographical position of the six habitats from which *Chlamydomonas* was isolated is shown in Fig. 1. The most northerly habitat is situated at about 39° S (No. 1, Ahuriri soil) and the most southerly at about 78° S (No. 6, Antarctic Continent), and although all but the latter are now covered by grassland, there are marked differences in the physical

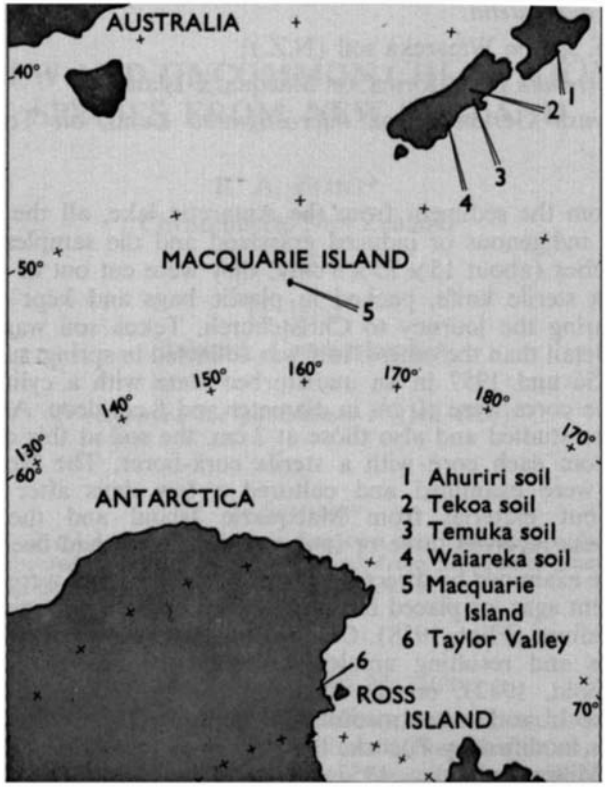

FIG. 1—Places at which soils were collected.

and chemical characteristics of the soils, in the climates under which they were formed, and in the parent materials from which they were derived. The available information is summarised in Table 1. Each habitat is discussed in relation to the species found on it.

Two features are common to all the species of *Chlamydomonas* in this study; all contain two apical contractile vacuoles lying in a plane at right-angles to that of the flagella, and each species reproduces asexually by forming four daughter cells; the plane of division is initially longitudinal but the protoplast rotates through 90° within the mother-cell wall.

DESCRIPTION OF THE SPECIES

Chlamydomonas chlorostellata Flint and Ettl n. sp. (Fig. 2)

Cellulae regulariter ellipsoideae, interdum ellipsoideo-cylindricae, polis utrisque rotundatis; membrana valde crassa, in basi cellulae a protoplasto saepe distanti; papilla clara hemisphaerica; flagellis usque ad 1.5 plo cellulae longioribus;

TABLE 1—Chemical analyses of New Zealand soils made by Soil Bureau, D.S.I.R., according to methods given by Metson (1956)

Soil	Altitude (m)	Average annual rainfall (mm)	Depth of sample (cm)	pH	Organic C % (1)	Total N % (1)	C/N	P citric, mg P % (2)	Cation exchange (N—ammonium acetate at pH 7.0)							$CaCO_3$ % (6)	Total soluble salts % (7)
									CEC me% (3)	BEC me% (3)	BS % (4)	Ca me % (5)	Mg me % (5)	K me % (5)	Na me % (5)		
Ahuriri	2	890	0–7.6	7.5–7.9	3.0	0.26	12	22	22.3	—	—	—	5.6	1.19	1.0	0.83	1.49
Tekoa	730	1630	5–10	5.3	5.7	0.34	17	7	23.0	10	46	7.8	2.3	0.4	0.3	—	—
Temuka	14	640	0–7.6	7.0	8.3	0.70	12	16	34.0	33	97	23.0	4.7	2.4	0.2	—	—
Waiareka	35	590	0–7.6	7.0–7.8	7.3	0.69	11	20	77.0	68	88	42.0	21.0	1.8	0.3	—	—

(1) Organic C % and total N % = organic carbon and total nitrogen/100 g soil.

(2) P citric mg P % = phosphorus extracted by 1% citric acid, mg P/100 g soil.

(3) CEC me % and TEB me % = cation exchange capacity and total exchangeable bases respectively, in milliequivalents/100 g soil.

(4) BS % = percentage base saturation $= \frac{\text{TEB}}{\text{CEC}} \times 100$.

(5) Ca me %, Mg me %, K me %, Na me % = exchangeable calcium, magnesium, potassium, and sodium respectively in milliequivalents/100 g soil.

(6) $CaCO_3$ % = calcium and magnesium carbonates expressed as percent $CaCO_3$.

(7) Total soluble salts % = soluble salts dissolved in a 1:5 ratio of soil : water extract and determined gravimetrically.

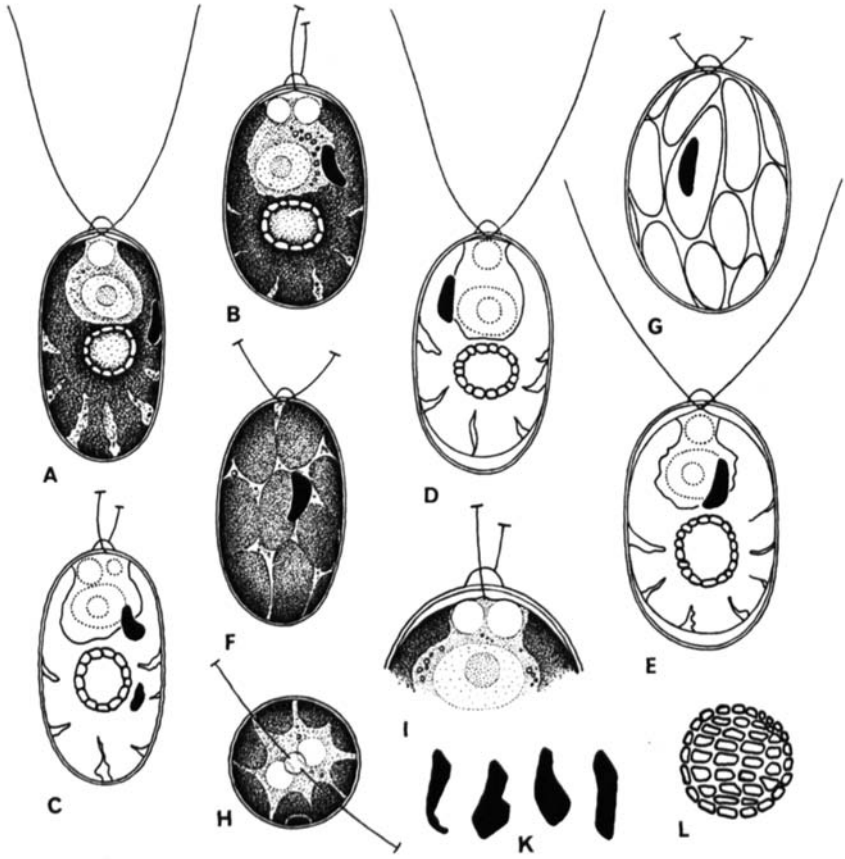

FIG. 2—*Chlamydomonas chlorostellata* n.sp. A–E. Different vegetative cells in optical section with the clear stellate chromatophore; F. G. The surface of the chromatophore; H. Apical view of the cell; I. Detail of the anterior end; K. Different shapes of the stigma; L. Starch grains of the pyrenoid.

chromatophori stelliformiter dissecto, laciniis a centro radialiter excurrentis; pyrenoide magno cum parvis granulis amylaceis in centro chromatophori; stigmate ellipsoideo magno in parte anteriori cellulae; nucleo nucleolato supra pyrenoidem; binis vacuolis pulsantibus. Propagatio asexualis divisione transversali protoplasti in 4 cellulas filiales. Cellulae 18–24 μ longae, 11–17 μ latae. Typus figura nostra 2.

The culture is kept in our collection (Ettl, A19).

In side-view (optical longitudinal section) the cells are regularly elliptical (sometimes they are almost cylindrical with rounded ends) and in optical transverse section they are circular. The cell wall is smooth, robust, colourless, and posteriorly it often stands away from the protoplast. Flagella (one and a half times as long as the body of the cell) pass through the relatively large, clear, hemispherical papilla. The chromato-

phore is bright green; in young cells it is cup-shaped with a very thick base, but as shallow or deep radial incisions develop it becomes lobed and appears to be stellate. The lobes touch the wall and in surface-view the cells seem to contain a number of separate discoid chromatophores (Fig. 2 F, G). A conspicuous basal pyrenoid is embedded in the central part of the chromatophore and surrounded by a number of small starch grains arranged in horizontal rows. The red stigma is large (4.5–6.5 μ long), elliptical or slightly rounded, and lies at the middle or towards the apical end of the cell. The nucleus is spherical and anterior to the pyrenoid. The cells are 18–24 μ long, 11–17 μ broad—they are often 22×15 μ.

During asexual reproduction the chromatophore loses its characteristic structure and the pyrenoid and stigma disappear, arising later in the fully developed daughter cells. Sexual reproduction and non-motile stages were not seen.

Our species differs from all those previously described. Although it resembles some which contain a stellate chromatophore, there are significant differences. *C. rotula* Playfair has neither papilla nor stigma and the lobes of its chromatophore are delicate and more widely separated. *C. sectilis* Korsch. lacks a papilla and the anterior lobes of the chromatophore are larger than the posterior. Although the mature cells of *C. meslinii* Bourr. have a dissected chromatophore, it is initially agloëform, the lobes are delicate and arranged in three rings, the anterior part of the cell is colourless, the stigma is filiform, and there are differences in the pyrenoid. *C. chlorostellata* seems therefore to be an independent species.

C. chlorostellata was isolated in spring 1956 and again in spring 1957 from the surface of samples of Tekoa soil collected at Bealey, at about 800 m above sea level in the eastern foothills of the Southern Alps (*see* Fig. 1, No. 2) (Thornton, 1958). The soil is a yellow-brown earth, described as a strongly leached silt loam. It is acid with a low concentration of phosphate and high C/N ratio. There is a tendency towards the formation of mor at the surface. The effect of the relatively high rainfall (about 1,630 mm p.a.) is offset by steep slope, rapid drainage, and a northerly aspect, so in summer the soil can become dry. Algae were rarely seen in the samples by direct observation, but species of the following genera appeared in cultures—*Ankistrodesmus, Botrydiopsis, Cylindrocystis, Euglena, Interfilum, Mesotaenium, Pleurochloris, Stigonema*. The grassland replaces beech forest (*Nothofagus solandri* var. *cliffortioides*) which was destroyed by fire about 100 years ago, and it includes indigenous species of *Agropyron, Danthonia, Festuca*, and *Poa* and the introduced grasses *Agrostis tenuis* and *Anthoxanthum odoratum* (Thornton, 1958).

Chlamydomonas reinhardti Dang. was also isolated from Tekoa soil in spring 1956, from 3 cm below the surface.

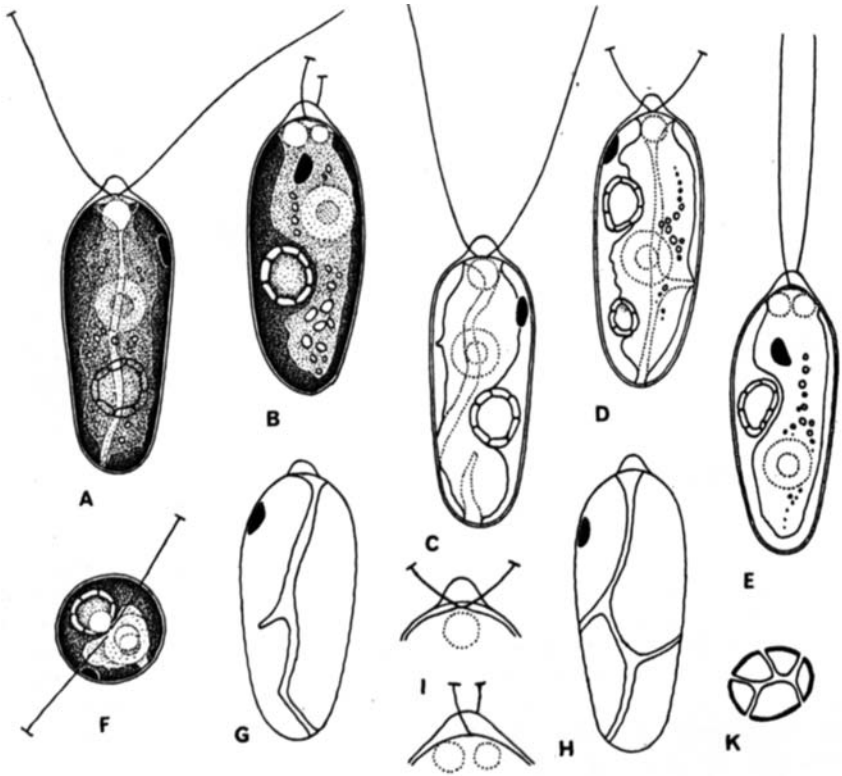

FIG. 3—*Chlamydomonas rima* n.sp. A–E. Vegetative cells (D. One cell with double pyrenoid); F. Apical view of the cell; G–H. The surface of the chromatophore with the fissure; I. Details of the papilla from two sides. K. Starch grains of the pyrenoid.

Chlamydomonas rima Flint and Ettl n. sp. (Fig. 3)

Cellulae longe ellipsoideae vel longe inverse ovoidcae usque ad rapaeformis. Membrana modice crassa, papilla magna, late rotundata: flagellis cellulae longitudinem aequantibus vel paulum longioribus; chromatophoro principio ollaeformi sulco profundi longitudinali dissecto, interdum sulcis brevibus etiam librate laciniato; pyrenoide uno laterali; stigmate claro in fronte cellulae; nucleo nucleolato in sito variabili; binis vacuolis pulsantibus. Propagatio asexualis divisione protoplasti transversa in 4 cellulas filiales. Cellulae 13–20 μ longae, 5–11 μ latae. Typus figura nostra 3.

The culture is kept in our collection (Ettl, A31).

In side-view the cells are elongated and either elliptical or obovate elliptical, usually resembling the form of a sugar-beet, in which the broad anterior end tapers slightly to the narrower base. The colourless cell wall is smooth, moderately thick, with a conspicuous helmet-like papilla which in one view (in the plane perpendicular to that of the flagella) is rounded and in the other view is narrower and clearly detached from the cell

(Fig. 3, I). The flagella are as long as or a little longer than the body of the cell. The bright green chromatophore is parietal but similar to the cup-shaped type; on one side there is a narrow longitudinal fissure which divides it into two parts. The fissure is asymmetrical; sometimes it is curved and sometimes branched so that the chromatophore is horizontally lobed. The part of the chromatophore opposite the longitudinal fissure is more robust and contains a local thickening in which the pyrenoid is lodged. The position of the thickening and pyrenoid vary; usually they are median but they often occur either towards the anterior or the posterior end of the cell. Two pyrenoids were very rarely seen in this species. The orange-red stigma is very conspicuous, slightly elongated, and situated about a third of the cell's length behind the apex. The nucleus is spherical and lies in the anterior or posterior part of the cell, depending on the position of the pyrenoid and the excavation in the chromatophore. The cells are 13–20 μ long, 5–11 μ broad, and most frequently 16.5×7.5 μ.

During asexual reproduction the chromatophore loses its characteristic features and the pyrenoid and stigma disappear. When division is completed they reappear and the daughter and mature cells closely resemble each other. Sexual reproduction and non-motile stages were not observed.

Although *C. rima* is superficially similar to two known species (*C. concinna* Gerloff and *C. moewusii* Gerloff) it differs from both in shape, chromatophore, and papilla. *C. concinna* is cylindrical with broadly rounded ends and has a hemispherical papilla, while *C. moewusii* is either cylindrical with rounded apices or symmetrically elliptical, and it has a papilla that is flattened in one view and rounded in the other. In both these species the chromatophore has a simple outline without lobes or fissures.

C. rima was isolated in autumn 1960 from a sample of Waiareka soil from Oamaru (Fig. 1, No. 4). The soil is a yellow-grey earth, formed in association with basic igneous rocks. It is described as a very weakly leached brown granular clay which is alkaline; exchange capacity, base-saturation, and exchangeable Ca and Mg are high. The annual precipitation is about 590 mm. The algal flora included diatoms, spherical unicellular green algae, species of *Microcoleus* and *Nostoc*; the dominant grasses in the pasture were *Dactylis glomerata* and *Festuca arundinaceae*.

Chlamydomonas subangulosa Fritsch and John forma (Fig. 4 A–D)

This species was erected by Fritsch and John (1942) who isolated it from three acid soils in England. As their account and illustrations are incomplete, our material is described in detail. The cells are laterally flattened—a feature which is obvious when they are seen in apical view (the flattening was not mentioned in the original diagnosis, but it is shown in the figure of daughter cells). In side-view the cells are broadly elliptical with rounded ends. The cell wall is thick, colourless, and there is a broad, shallow papilla through which the flagella emerge. In the drawing of Fritsch and John the flagella originate from two widely separated points, whereas in cells of our cultures they arise from a small process at the

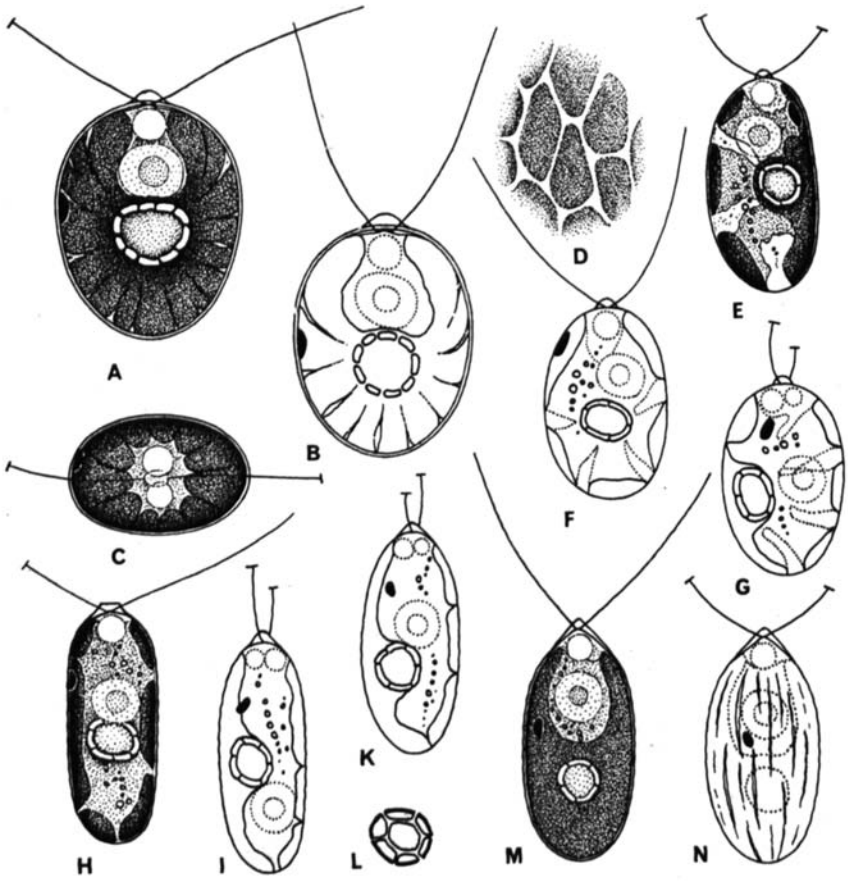

FIG. 4—A–D. *Chlamydomonas subangulosa* Fritsch & John forma.
A, B. The wider side of the cells; C. Apical view of a cell; D. Surface of the chromatophore; E–G. *Chlamydomonas macrostellata* Lund forma; H–L. *Chlamydomonas moewusii* f. *microstigmata* Lund; L. Starch grains; M, N. *Chlamydomonas snowiae* Printz.

apex of the protoplast, diverge at a wide angle, and pass out through the flat papilla. The chromatophore is massive, cup-shaped, and the thick posterior part extends up to the middle of the cell. The whole structure is lobed or striated by radial incisions which give it a stellate appearance. The incisions are narrow, and the closely set lobes touch the wall. In surface-view the cells seem to contain a number of separate discoid chromatophores. The large round or elliptical pyrenoid is lodged in the basal part of the chromatophore and there is a conspicuous red stigma at the equator of the cell. The nucleus is central and anterior to the pyrenoid. The cells are the same size as the type (usually 22 μ long and 16 μ broad)

and asexual reproduction is also similar. Sexual reproduction and non-motile stages were not seen.

This alga was isolated from Ahuriri soil, near Napier, in winter 1960 (Fig. 1, No. 1). The gleyed soil is recent and estuarine and was raised about 2 m above sea level during an earthquake in 1931; the land is still being reclaimed. It is a very weakly leached alkaline clay loam containing free $CaCO_3$ and has high exchangeable Mg. Mean annual rainfall is 890 mm. The algal flora included diatoms, species of *Kentrosphaeria* and *Pleurochloris*, and the pasture contained *Lolium* spp. and *Trifolium repens*.

C. subangulosa occurred in Britain in very acid sandy heath soils at Oxshott and Breckland (John, 1942; Fritsch and John, 1942); the latter soils had a high C/N ratio and total exchangeable bases were very low (Watt, 1940). Moreover the samples had been air-dried and stored in bottles for nearly three years before they were cultured, which suggests that some kind of drought-resisting cell is formed in this species.

Chlamydomonas macrostellata Lund forma (Fig. 4 E–G)

The cells are elliptical or elliptical-ovoid, sometimes slightly elongated or a little asymmetrical, one side being more convex than the other. The cells are circular in optical transverse section. The cell wall is very thin, the papilla is nearly hemispherical and the flagella emerge through it. The much-dissected chromatophore lies in a lateral position, the lobes extending radially from its centre. The number and size of the lobes are indefinite. A round or elliptical pyrenoid is lodged in the central part of the chromatophore but laterally in the cell. The stigma is red, elliptical, and situated about a quarter of the cell's length behind the apex. Asexual reproduction is similar to that described for the type. The cells are 10–12.5 μ long and 6–7.5 μ broad, and in our material they differ from the type by their small but conspicuous papilla, their distinct and clearly seen stigma, and their smaller size.

This alga was isolated from peat on Macquarie Island, which lies in latitude 54° 30′ S and has a cold temperate climate (without a warm season), the air temperature varying between −8°C and +11°C (Fig. 1, No. 5). The average precipitation is about 1,000 mm p.a., but the sky is usually overcast and the humidity is always above 90%; even in mid-summer the mean daily sunshine is only 2.8 hours (Taylor, 1955). The peat probably contains wind-blown salt as Bunt (1965) recorded for other sites. The sample was collected at Green Gorge, about 270 m from the shore, at about 15 m above sea level in short tussock which was probably grazed by rabbits and frequented by birds. The pH value of the sample on arrival in New Zealand was 5.3–6.3. Other algae isolated from this habitat include species of *Ankistrodesmus*, *Chlorococcum*, and *Cosmarium*. It was interesting to find the desmid, because very few have been seen in soils from the Antarctic or Subantarctic Islands (personal observations). There were many empty diatom frustules in the peat, which may have formed part of the original flora.

Chlamydomonas moewusii Gerloff forma **microstigmata** Lund (Fig. 4 H–L)

This form of *C. moewusii* was found on bare soil in England and is distinguished from the type of the species by a small linear stigma and an irregularly perforated chromatophore (Lund, 1947). The cells in our culture were, in general, similar to Lund's form, but in some respects differed from it. They are more elliptical and, in contrast to the English form, only scarcely cylindrical. The cell wall is very thin and the papilla is typical of the species. The flagella are a little shorter than the cells. The bright green chromatophore is cup-shaped but with an elongated anterior portion, and its inside edge is undulated, grooved, and often irregularly perforated. The pyrenoid is lateral, lying in a thickening of the chromatophore and there is a very small indistinct stigma on the anterior part of the chromatophore. The nucleus is either anterior or posterior to the centre, depending on the position of the pyrenoid. The cells are usually 14 μ long and 6.5 μ broad.

C. moewusii forma *microstigmata* was isolated from the surface of Temuka soil collected in February 1961 from a swampy depression in a field at Lincoln College, Christchurch (Fig. 1, No. 3). The soil is described as a weakly leached southern gley silt loam; the pH is 7.0, and base saturation and exchangeable Ca are high. Mean annual rainfall is about 640 mm. The algal flora included species of *Dactylococcus, Nitzschia,* and *Vaucheria;* the pasture contained *Holcus lanatus, Lolium* spp., *Poa* sp., and *Trifolium repens.*

Chlamydomonas snowiae Printz (Fig. 4 M, N)

The species is distinguished by its conical anterior end. The cells in our culture agree more with Pascher's illustration (1927, fig. 174) than with Printz's diagnosis. The compact cup-shaped chromatophore has a delicate striation on its surface caused by very fine grooves, some long, some short, arranged longitudinally. Our cells are 15–17 μ long and 8–10 μ broad, and apart from the chromatophore, they are similar to the type in their structure and in the method of asexual reproduction. The culture grew best in Juller's modified solution (Pocock, 1960).

C. snowiae was isolated from sediment collected in November 1959 from the bottom of a small unnamed Antarctic lake 5 or 6 km from the mouth of the Taylor Dry Valley (Fig. 1, No. 6). At the sampling point the water was 15–30 cm deep. The lake occupied a hollow on a moraine composed of granite, dolerite, and metamorphic rocks (Claridge, pers. comm.). The pH of the sediment was 7.7, but nothing else is known of the physical and chemical nature of the lake water. The surrounding material resembles profile No. 34 described by Claridge (1965), where free $CaCO_3$ is present and the pH value is 9.8. Analyses of lakes, ice, and meltwater in Taylor Dry Valley (Angino *et al.*, 1964, 1965) (Table 2) and of the soils (Claridge, 1965) suggest that the water in the unnamed lake might be rich in Cl, Na, and Mg. For most of the year the lake would be frozen and only free of ice for about two months. There is continuous

TABLE 2—Chemical analyses of ice from Taylor Valley, Antarctica, quoted from Angino *et al.* (1965)

		Parts per million					
	pH	PO_4	Ca	Mg	K	Na	Cl
Taylor Glacier ice (melted)	7.1	16.0	20	104	0	0	561
Taylor Glacier meltwater from snout of glacier	7.0	0	18	1	1.1	7.0	5–10

darkness for at least four months, and even in summer, cloud much reduces the amount of light (Hatherton, 1961). Diatoms were present in the sample and species of *Chlorococcum* and *Chlorosarcinopsis* were isolated from the sediment.

This is the first record of *C. snowiae* from the Antarctic Continent. Although algae have been recovered from 70% (101) of the samples of soil from the region of McMurdo Sound, *Chlamydomonas* was not among them (personal observations). Other species of *Chlamydomonas* have been recorded from McMurdo Sound by Wille (1928) who summarises earlier reports, and by Holm-Hansen (1964), but always in aquatic habitats.

C. snowiae occurs in both hemispheres. It has been recorded in Australia in temporary pools (Playfair, 1923; Moewus, 1953) and in soil (Phillipson, 1935). In studies of algae in English soils (Bristol, 1920; Bristol Roach, 1927) this species was isolated from samples that were air-dried before they were cultured. It occurs in Californian sewage oxidation ponds (Silva and Papenfuss, 1953); it was also isolated from a soil in California and shown to be homothallic in culture (Smith, 1946).

DISCUSSION

With the exception of *C. snowiae* these species grew on soil under grass, and apart from *C. chlorostellata*, which was isolated in the spring of two successive years, each was found once. With so few observations it is uncertain whether the species are temporary or permanent elements in the communities, and so little is known about the needs of most of them that beyond describing the habitats nothing constructive can be said about their ecology.

John (1942) and Fritsch and John (1942) studied the algae of 24 soils (under grasslands, forests, heaths, fen, and moor) and found *Chlamydomonas* on 20 of them. Lund (1947) examined the algae on 58 bare soils and recorded *Chlamydomonas* on 23 of them. Altogether 26 taxa of *Chlamydomonas* were identified in these investigations, but none of them

was recorded by all the authors and 75% of the taxa were restricted to a single soil. Lund reports that none of his taxa of *Chlamydomonas* appeared regularly in any particular type of soil.

Pure cultures of several species of *Chlamydomonas*, including *C. reinhardti* (which occurred on Tekoa soil) have often been used in research in genetics and physiology and although it remains to be seen how far the facts thus obtained are true of a species in nature, some of the characteristics disclosed by this kind of work seem to be ecologically important. The following information on the needs and behaviour of *C. reinhardti* was extracted from a few papers:

Nutrition

a. Under some conditions *C. reinhardti* multiplies rapidly, its generation time being 5 hours (Levine and Ebersold, 1960). Under other circumstances it divides every 6.6–9.5 hours (Sager and Granick, 1953).

b. It is facultatively heterotrophic, growing in the dark (Smith, 1948), on acetate, when it divides every 14–18 hours (depending on the medium) (Sager and Granick, 1953).

c. It does not need vitamin B_{12}, thiamine or biotin, in the culture medium.

d. Its growth is unaffected by pH varying between 5.0–8.5 (Gerloff, 1940) and pH 5.5–8.0 (Sager and Granick, 1953).

e. It cannot use nitrate as a source of nitrogen (Syrett, 1962), and Proctor (1957) shows it prefers ammonium salts as a source of nitrogen.

Permeability

a. *C. reinhardti* is usually killed by streptomycin (Sager, 1960), but it is less sensitive than some blue-green algae to chloramphenicol (Echelin and Morris, 1965).

b. It may be inhibited by substances which *Chlorella vulgaris* and *Anacystis nidulans* excrete into culture medium (Proctor, 1957).

c. The herbicide-O-methylthreonine does not inhibit the formation of photosynthetic pigments in *C. reinhardti* (Hutner and Provasoli, 1964).

Excretions from living and dead cells of C. reinhardti

a. Oxidisable material excreted by living cells stimulates the growth of *Chlorella pyrenoidosa* (Allen, 1955).

b. The products of dead cells (fatty acids?) inhibit the growth of *Haematococcus pluvialis* (and perhaps of *Scenedesmus quadricauda*) (Proctor, 1957).

Biology and reproduction

a. *Chlamydomonas reinhardti* is positively phototactic to light of low intensity and negatively phototactic when the intensity exceeds

170 foot-candles. An "eyeless" mutant was slightly sensitive to light (Hartshorne, 1953).

b. Sexual reproduction is isogamous and heterothallic. Gametes are formed when the supply of nitrogen is depleted (Sager and Granick, 1953; Trainor, 1959; Tsubo, 1961).

c. Light may (Smith, 1951; Trainor, 1959) or may not (Sager and Granick, 1953) be necessary for the formation of gametes.

d. Gametes are not attracted by gaseous substances in coal gas (e.g., ethylene, acetylene) and they show no chemotaxis when mixed in intra- or interspecific combinations (Tsubo, 1961).

e. Gametes of opposite mating types pair and fuse very quickly (Levine and Ebersold, 1960).

f. Light of high intensity does not induce polyploidy in this species (Levine and Ebersold, 1960).

g. Ripening of the zygote needs light which, according to its intensity and duration, partly determines the thickness of the zygote wall (Levine and Ebersold, 1960).

Although these observations refer to *C. reinhardti* in culture, they help to explain the widespread distribution of the species. From an ecological point of view the characteristics which seem to be particularly important are its capacity to multiply rapidly, to tolerate a wide variation in pH, and to be independent of an external source of vitamins. The ability of this species to use ammonium salts as a source of nitrogen would be an advantage in a waterlogged soil where nitrifying organisms and nitrates are absent, and its facultatively heterotrophic nutrition (acetates have been found in soils (Vallentyne, 1957)) would allow the alga to grow in the dark. Nitrogen is essential for vegetative multiplication of *C. reinhardti*, but its absence leads to the development of gametes and thick-walled zygotes. In this form the alga can survive in an environment that is temporarily unfavourable for its vegetative growth.

In more general terms these experiments on *C. reinhardti* emphasise that "the effect of the abiotic environment on organisms, the effect of organisms on the environment and the interaction of organisms all contribute to the biogeochemical cycles" (Provasoli, 1958), and when as much is known about the rare species as is known about *C. reinhardti*, it may be possible to reach a better understanding of their distribution.

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