

# SOME PHYSIOLOGICAL STUDIES ON TWO STRAINS OF THE GREEN ALGA *COELASTRUM MICROPORUM* NAEG.

J. GATES, J. ANDERSON, D. L. LYNCH and M. G. FENWICK  
*Northern Illinois University, DeKalb, Illinois*

**ABSTRACT.**—The effects of agar concentration, temperature, colchicine, and gibberellic acid on the growth and morphology of two strains (280 and 281, Univ. Indiana Algal Coll.) of *C. microporum* Naeg. were studied. Coenobial formation was observed to differ between the two strains as a result of these treatments and conditions.

Motile cells were produced by both strains and hypertrophy of cells grown in colchicine was observed.

Some comparative physiological studies of two strains (Nos. 280 and 281, Univ. Indiana Algal Coll.) of the colonial alga, *Coelastrum microporum* Naeg. show several differences between the two strains; i.e., size and length of time in coenobial form, the ability to utilize various substrates at various temperatures and the ability to grow heterotrophically. The investigations reported below encompass the following areas of investigation: (1) the effect of the alkaloid, colchicine, on growth and morphology, (2) the effect of gibberellic acid on growth and morphology, (3) the effects of solidity of media on coenobial formation, and (4) the apparent production of motile bodies by both strains.

## METHODS AND MATERIALS

All studies were made on organisms grown in a Bristol's medium as modified by Bold with glucose (10 g/l) added, and hereafter referred to as the medium. For the colchicine studies 150 ppm colchicine were added to the medium. Gibberellic acid was added to the medium at the rate of 10 mg/l. All cultures were grown in test tubes. The light source was a bank of fluorescent lights of about 400 fc. For studies on the effects of solidity of medium on the coenobial development, the cultures were grown in petri plates to which Bristol's

and glucose (10 g/l) were added. Agar was added to this medium at the rate of 0.0, 1.5, 2.0, 2.5, and 3%.

An experiment was undertaken to determine the fate of certain motile bodies observed microscopically in various media on which these algae were cultured. Due to their small size pipetting proved impracticable so that a centrifuge technique was used. The procedure was as follows:

1. Examined known stock cultures of 280 and 281 to find a culture rich in these motile bodies.
2. Inoculated these cultures into a centrifuge tube of sterile distilled water.
3. Centrifuged, then with a sterile pipette removed one ml of supernatant.
4. Examined  $\frac{1}{2}$  of this portion on a slide for complete absence of vegetative cells and the presence of motile bodies.
5. Inoculated the remaining fluid on slants.

## RESULTS AND DISCUSSION

**Colchicine.**—Colchicine appeared to have an inhibiting effect on the production of coenobia. Both strains produced large single cells of bizarre shapes sometimes lobed and very granular in appearance. Rate of development in terms of numbers, however, did not appear to be affected. When some of these abnormally large cells were inoculated into fresh medium they reverted back to normal coenobial forms.

**Gibberellic Acid.**—Cultures were inoculated into the medium to which gibberellic acid had been added. (Table 1). The cultures were then grown heterotrophically at either 20°C or 36°C. Gibberellic acid appeared to stimulate growth and increase cell size at both temperatures. Strain 281, however, appeared to be extremely sensitive to this

concentration of gibberellic acid. After 8 weeks the cultures turned white, and upon transfer into the medium no growth occurred in the light. Culture 280 upon transfer grew abundantly under these same conditions. Thus these two strains appear to be differentially sensitive to gibberellic acid.

*Effect of Solidity of Medium.*—The cultures were inoculated into test tubes of medium and petri dishes of medium to which agar had been added at 0.0, 1.5, 2.0, 2.5 and 3.0%. The results obtained were opposite to those of Trainor and Hilton (1963). They reported an increment of coenobial formation corresponding to an increment of solidity of media. The percentage of coenobial formation in our experiments, however, decreased with an increase in solidity of media with the greatest percentage of coenobia occurring in liquid media.

*Production of Motile Bodies.*—When the present investigations with *Coelastrum* were begun a motile stage had not yet been reported in the literature. But our observations indicated that motile cells were being formed. Observations of cells (1500 X) showed very small apparently motile bodies present in the medium. These bodies were of such a size so as to be affected by Brownian movement. A drop of a mixture of HCl and methylene blue was placed under the coverslip and as the front reached the bodies all movement of the type seen earlier stopped. Characteris-

tic Brownian movement continued. These motile bodies have been seen inside the mother cell as well as in the media. They have been seen in various sizes which could represent various stages of development of zoospores and they have been seen leaving the mother cell as well. There appears to be a parallelism between yellowing of the cultures and motility. This yellowing occurs after 1 month in liquid medium and after 2 months on agar slants with the same medium at 400 fc illumination with a bank of fluorescent lights.

Following the procedure outlined under Methods and Materials for the determination of motile bodies, growth was observed after 48 days indicating that these motile bodies were probably a part of the life cycle. Though the results were positive the technique is open to criticism. Since this time a motile stage in *C. microporum* 280 has been reported (Trainor and Burg, 1965). Motility in *C. microporum* 281 has not hitherto been reported.

#### LITERATURE CITED

- TRAINOR, F. R., and C. A. BURG. 1965. Motility in *Scenedesmus dimorphus*, *Scenedesmus obliquus*, and *Coelastrum microporum*. J. Phycol. 1:15-18.  
 ———, and R. L. HILTON. 1963. Culture and *Scenedesmus longus*. Bull. Torrey Bot. Club 90:407-412.

Manuscript received May 26, 1966.

TABLE 1.—Effects of 10 mg/1 of gibberellic acid on the growth and morphology of the two strains of *Coelastrum microporum*.

Strain	Glucose-gibberellic acid -Bristol's		Reinoculated into glucose and Bristol's, light -from tubes grown with gibberellic acid
	20°C	36°C	25°C
280.....	+++	++	++
281.....	++	+++	-
Remarks.....	Increased growth rate, Larger cells. 281 cells lost chlorophyll and became shrunken. 281 remained in coenobial form for a longer period of time.		280 survived and retained chlorophyll. 281— no growth of inoculum