The Effect of Methyl Mercury on the Growth of the Green Alga, Coelastrum microporum Naeg. strain 280

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Coelastrum microporum Naeg. is a coenobial green plant in the order Chlorococcales. C. microporum strain 280 is a hetertroph which can be grown in the dark with glucose as the sole carbon source (LYNCH et al. 1967). This paper is concerned with the effects of mercury on this green alga. Mercury is volatile and thus distributed throughout the world atmosphere (COPELAND 1970). GOLDWATER (1971) states that marine algae are able to concentrate mercurials up to more than 100 times the concentration found in sea water. Since methyl mercury is the most toxic of all mercurials to the environment it was used as the toxic principle in these experiments.

Experimental Procedures

Bristol's medium as modified by BOLD (1949) was utilized as the growth medium. However, phosphate solutions were made with 11.876g K_2HPO_4 and 9.078 g of KH₂PO₄ per liter. This adjusted the pH of the medium to about 6. Fifteen hundred ml of this medium was placed in eight 2,000 ml erlenmeyer flasks. To the other flasks 25.1, .025, .0126, .006, .003, and .0008 ppm of methyl mercury were added aseptically after autoclaving, (for laboratory precaustions when using alkyl mercury compounds see Science vol. 172:872, April, 1971). All six flasks were inoculated with 15 ml of inoculum adjusted to a percent transmittance between 9 and 11. They were placed in growth chambers at a temperature which varied from 21 to 22 C with a photoperiod of 16 hr. light and 8 hr. dark at a light intensity of 975 foot candles. The flasks were shaken daily to prevent clumping of the cells and were grown until the controls reached a density of 70% transmittance on a Bausch and Lomb Spectronic 20 at 520 mu. Photosynthetic rates were determined by the method of VERDUIN (1951).

Results

An inhibition of growth in <u>Coelastrum microporum</u> Naeg. strain 280 was noted with .003, .006, .0126, and .025 and 25.1 ppm of methyl mercury chloride. The only concentration of methyl mercury chloride that did not markedly inhibit growth was .0008 ppm. At this concentration, the samples grew much like the controls.

TABLE 1.

The effects of methyl mercury chloride on the growth of Coelastrum microporum Naeg. strain 280.

Concentration of methyl mercury chloride (ppm)	% Transmittance	ul/l of algae
0.000	66.6	182
0.0008	68.6	158
0.003	77.0	125
0.006	93.5	31
0.0126	92.8	34
0.25	96.4	16
25.1	97.6	11

Table one shows the results of these experiments. Controls are the average of 6 samples. The mercury treated samples are the average of two samples. Beyond a level of .006 ppm the growth of this microorganism was drastically reduced falling from 31 ul/1 of algae to 16 ul/1 with the addition of .025 ppm of mercury methyl.

It was noted in all experiments that the samples having high concentrations of methyl mercury chloride stored more starch than the controls. This accumulation of starch was estimated by the relative degree of staining from modified Lugol's iodine solution when examined under a light microscope.

In the first experiment, the controls showed no noticeable accumulation of starch. Samples having concentrations of .025 and 25.1 ppm methyl mercury chloride showed slight to moderate accumulations of starch. The control samples in the second experiment showed no accumulation to moderate accumulations of starch. Samples containing .003 ppm and .006 ppm methyl mercury chloride showed moderate to heavy accumulations of starch. The controls of the third experiments showed slight to moderate accumulations of starch. The samples containing .0008 ppm of methyl mercury showed zero to moderate accumulations of starch. Samples containing .0126 ppm methyl mercury chloride showed moderately heavy to heavy accumulations of starch. It appears

that the addition of methyl mercury to algal cultures disrupts the normal metabolic pathways and leads to a starch accumulation. It may also be true that this starch accumulates as a result of the decrease in total protoplasm (cell growth) the excess carbohydrate being then stored as starch.

Cells grown in higher concentrations of methyl mercury chloride also tended to settle to the bottom of the culture flasks. Only the cells in the samples containing .0008 ppm methyl mercury chloride remained suspended in the flask much as the control samples which contained no methyl mercury chloride. Perhaps the tendency of the algae in the mercury-amended cultures to settle was due to a change in the specific gravity of the cells resulting from an accumulation of starch and/or mercury. Other investigators (PASSOW and ROTHSTEIN 1960) have found that mercury affects the membrane permeability of cells.

A slight increase in gross photosynthetic rates was noted in samples treated with .0008 ppm methyl mercury chloride when compared with the controls. This slight increase in photosynthetic rate could indicate that amounts below the .0005 ppm allowable levels of mercury in lakes, rivers, and ground waters have subtle but significant effects on algal populations.

The green alga, <u>Coelastrum microporum</u>, exhibited some growth at all concentrations of mercury but the cell growth was greatly reduced with increasing amounts. The presence of mercury affected both the accumulation of starch and cell buoyancy. Gross photosynthetic rates were altered at .0008 ppm of this compound.

References

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