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Effect of Hexachlorobenzene (HCB) on Photosynthetic Oxygen Evolution and Respiration of *Chlorella pyrenoidosa*

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Hexachlorobenzene (HCB) has gained significance through its appearance in the global ecosystem as an environmental contaminant. In a recent review LEONI and D'ARCA (1976) presented data on the HCB contamination in Italy where residues of this pollutant have been found in surface waters, soil, and livers of birds found dead, as well as in cows milk and human adipose tissue.

Only little is known about the persistence of HCB in the environment but from studies of FREITAG et al. (1974) and ISENSEE et al. (1976) it may be concluded that HCB is even more stable than dieldrin and DDT. However, incubation of HCB at a dose level of 3.74 ppm in an Erlenmeyer flask with Chlorella pyrenoidosa for one month with agitation only once a day leads to an intensive metabolism to pentachlorophenol and other polar metabolites (F. GEIKE, unpublished results).

Although increasing information about the harmful effects of HCB on animals and on the induction of microsomal enzymes by HCB is available only little is known about the action of this environmental pollutant on plants. Recently, GEIKE and PARASHER (1976) were able to show that HCB depressed growth of the freshwater alga Chlorella pyrenoidosa in short-term experiments while an exposure to HCB over a period of three months enhanced growth. In the present paper it is shown that photosynthetic oxygen evolution is depressed by HCB while respiration is only slightly decreased.

MATERIALS AND METHODS

Chlorella pyrenoidosa strain 211-8b from the Pringsheim algae collection, University of Göttingen, was grown as described previously by GEIKE and PARASHER (1976). Algal cells equivalent to 1.0 mg chlorophyll were given into culture tubes containing the culture medium after adding the appropriate amount of HCB in acetone solution (1.0 ml) and were incubated under continuous light and aeration with an air stream of 7.5 l / h for 72 h at 30°C.

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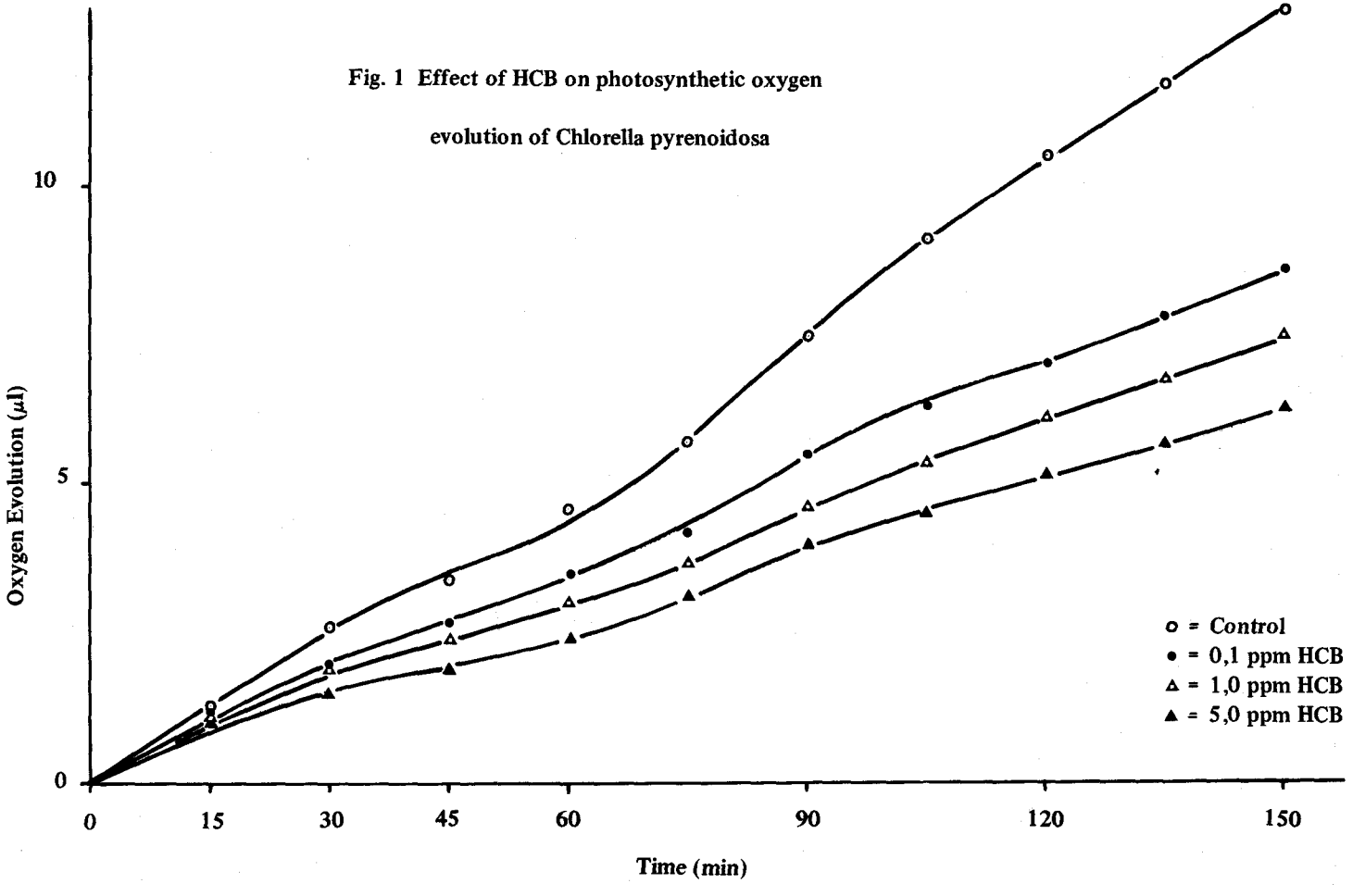
Fig. 1 Effect of HCB on photosynthetic oxygen evolution of Chlorella pyrenoidosa



The controls received the same amount of acetone. The cells were harvested by centrifugation and resuspended in the supernatant culture medium to give a suspension equivalent to 0.1 mg chlorophyll / ml.

The studies on the effect of HCB on photosynthetic oxygen evolution and respiration of algal cells were performed in conical Warburg vessels of approximately 13 ml and a central inner well. In both series of experiments each vessel received cells equivalent to 0.2 mg chlorophyll in a total volume of 3.0 ml and the estimation of oxygen evolution and oxygen consumption was performed by manometric techniques according to the method described by UMBREIT et al. (1972). In photosynthesis experiments the algal cells were illuminated with 20000 Lux at 30°C while the cells during estimation of oxygen consumption were incubated at 30°C in the dark.

RESULTS AND DISCUSSION

The effect of HCB application on photosynthesis and respiration was studied in experiments with 0, 0.1, 1.0, and 5.0 ppm HCB in the nutrient solution which contained 0.33 % acetone. The results presented here are representative values from experiments repeatedly performed with similar results. In Fig. 1 the effect of HCB on photosynthetic oxygen evolution is shown. Incubation of the freshwater alga Chlorella pyrenoidosa over a period of 72 h and determination of the rate of photosynthesis thereafter revealed an inhibition of this process by the environmental pollutant. The rate of inhibition after measuring photosynthesis for 2 h was approximately 33.3, 42, and 51 % for 0.1, 1.0, and 5.0 ppm, respectively. These results are in good agreement with changes in ultrastructure after exposure of Chlorella cells to HCB where severe damage to cell membranes and especially a disintegration of the thylakoid system after exposure of the cells to 10 ppm HCB was observed (PARASHER et al. 1978). About 60 % of the cells of the culture were showing damage. These results, again, are in accordance with the present results which reveal that a dose level of 5.0 ppm HCB causes an inhibition in photosynthesis of about 50 %.

Studies on the oxygen consumption of Chlorella cells after exposure to HCB revealed that HCB has only a slightly negative effect on respiration of the cells at higher concentrations. A dose level of 5.0 ppm HCB showed a maximum inhibition of about 10 % (Tab. 1). These results again correspond to the ultrastructural observations of PARASHER et al. (1978) showing that mitochondria were in most cases damaged only after the thylakoid system had disintegrated. From experiments in this laboratory, however, it is known that respiration of

Tetrahymena pyriformis and of plant tissue other than

TABLE 1

Effect of HCB on oxygen consumption of Chlorella pyrenoidosa cells

HCB concentration (ppm)	Rate of oxygen consumption (μl O_2)		
	Time of readings (h)		
	1	2	3
Control	1.61 ± 0.02	3.41 ± 0.07	7.74 ± 0.08
0.1	1.54 ± 0.02	3.32 ± 0.03	7.75 ± 0.08
1.0	1.50 ± 0.01	3.28 ± 0.02	7.30 ± 0.06
5.0	1.44 ± 0.02	3.20 ± 0.03	7.13 ± 0.07

photosynthetic is inhibited by an exposure to HCB. Further investigations on this aspect, therefore, are necessary.

SUMMARY

In experiments with Chlorella pyrenoidosa it was shown that an exposure of these cells to HCB at a dose level of 0.1, 1.0, and 5.0 ppm caused an inhibition of photosynthetic oxygen evolution of 33.3, 42, and 51 %, respectively. Respiration, however, was inhibited only slightly, if at all.

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