The Effects of Various Aroclor Fractions on the Population Growth of *Chlorella pyrenoidosa*

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Introduction

Polychlorinated biphenyls (PCBs), because of their characteristic chemical properties, are among the most stable and persistent chemicals found in the environment (GUSTAFSON 1970). Monitoring studies of the abiotic environment have detected PCBs in rainwater, sewage outfalls, marine and fresh water bodies, and the atmosphere (BIDLEMAN, et al., 1974; HAMMOND 1972).

Extensive residue analyses of PCBs have been done for fish (HAMMOND 1972; RISEBROUGH, et al., 1972; CARR, et al., 1972), and birds (JOHNSTON 1973; JENSEN, et al., 1969; HAMMOND 1972) but much less work has been done on organisms near or at the base of the food pyramid. MOSSER et al., (1972) have studied the toxicity of PCBs to mixed cultures of selected marine unicellular algae and MORGAN (1972) studied the effects of the PCB Aroclor 1242 on Chlamydomonas reinhardtii. KIEL et al., (1971) studied effects on the diatom Cylindrothecz closterium. Other than the above studies, work on PCBs as they affect primary producers is lacking.

This paper reports the results of studies performed on Chlorella pyrenoidosa in the laboratory using various PCBs at concentrations of 1 ppm and 100 ppb. Chlorella pyrenoidosa was selected for study because of its wide occurrence in nature, because its physiology is better understood than that of most other species of unicellular algae, because it is a fresh water organism representative of those at the base of the ecological food pyramid, and because it is a common pollution alga.

The objective of our research was to examine what effects, if any, would be exerted on the population growth of Chlorella pyrenoidosa by various fractions and concentrations of the PCB Aroclor.

Methods

Cultures of Chlorella pyrenoidosa were obtained from the American Type Culture Collection (Strain #7516). Sterile cultures were maintained and experiments performed in liquid

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Bristol's medium (STEIN 1973). All stock and experimental cultures were kept on a rotary shaker under continuous illumination by fluorescent light of 165 foot-candles. PCBs, when added, were introduced into the medium in the organic solvent acetone. Chlorella censuses were made directly with a Spencer Bright-Line Beubauer hemacytometer.

The polychlorinated biphenyls studied all share the trade name Aroclor[1] (Monsanto Company). Aroclor fractions 1232, 1242, 1254, and 1268 were used in these experiments. The first two digits indicate that the compounds are biphenyls and the last two digits refer to the weight-percentage chlorine content of each Aroclor fraction.

Results and Discussion

An experiment was performed to test the effect of Aroclors of differing chlorine content on Chlorella population growth. Aroclors 1242, 1254, and 1268 were tested each at a concentration of 1 ppm. All treatments and controls were established in triplicate. Controls contained 0.1% concentration of acetone. The innoculum culture was 5 days old with a density of 27.05×10^6 cells/ml.

Aroclor 1242 treated populations were 64% lower in population density than the controls at 8 hours. Aroclor 1254 produced a 45% reduction and Aroclor 1268 resulted in a 36% reduction at 8 hours (Table I). Throughout the remainder of the experiment, Aroclor 1242 populations remained at much lower densities than the control but the difference between controls and experimentals markedly decreased as populations grew. The Aroclor 1254 treated populations were equal to the controls by 129 hours. Aroclor 1268 was equal to control populations by 56 hours (Table I).

These results indicate that the initial perturbation caused by each treatment was inversely related to the chlorine content of the PCB. Similar results have been obtained by LICHTENSTEIN et al. (1973) and SIVILINGAM et al. (1973). However, considerable disagreement with the theory that toxicity is inversely related to chlorine content exists (BICKERS et al., 1972).

One theory of PCB toxicity and chlorine content is that sublethal effects are directly correlated with chlorine content whereas lethal effects are inversely correlated (PEAKALL, et al., 1970). The results of this experiment are in agreement with this theory only if Chlorella cells were killed by PCB concentrations. However, the actual cause of the observed population reductions is unknown at this time.

The experiment just discussed involved the use of PCBs at concentrations of 1 ppm. An experiment was performed to compare

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[1]Obtained from Analabs.

TABLE I

Population growth of *Chlorella* treated with various Aroclors at 1 ppm.

Hours	Control		Aroclor 1242			Aroclor 1254			Aroclor 1268		
	$\bar{X}^1$	CV^2	$\bar{X}$	CV	R^3	$\bar{X}$	CV	R	$\bar{X}$	CV	R
08	0.22	43	0.08	20	.36	0.12	28	.54	0.14	4	.64
21	0.26	6	0.10	10	.38	0.17	7	.65	0.13	50	.50
44	1.17	23	0.65	57	.56	0.78	26	.67	1.05	22	.90
56	3.45	6	2.71	23	.78	2.34	26	.68	3.72	5	1.08
73	8.60	3	7.67	28	.89	7.08	11	.82	8.94	1	1.04
103	18.69	3	16.10	6	.86	16.05	14	.86	17.65	2	.94
129	28.32	7	20.92	25	.74	28.73	14	1.01	26.25	5	.93
191	46.05	3	28.04	18	.61	46.71	8	1.01	43.21	1	.94

[1] Cells/ml. x 10^6
[2] Coefficient of Variability
[3] Ratios of Treatment Means to Control Means

effects at this concentration with a lower concentration of 100 ppb. Three replicates each were established of a control + 0.1% acetone, Aroclor 1232 at 1 ppm. and Aroclor 1232 at 100 ppb.

The ratio of the Aroclor 1 ppm treatment to the control mean showed a 23% drop below the control in these treatment populations at 17 hours (Table II). This drop had increased to 46% at 46 hours. The populations then rapidly began approaching control levels. The 100 ppb treatment never fell below an 18% drop at 46 hours. This drop may not have been statistically significant considering the small cell counts and variability (Table II). This experiment demonstrated that the lower concentration of PCB had decidedly less effect on the population growth of the Chlorella.

TABLE II

Population growth of Chlorella treated with Aroclor 1232 at differing concentrations

Hours	Control		Aroclor 1 ppm			Aroclor 100 ppb		
	$\bar{X}$[1]	CV(%)[2]	$\bar{X}$	CV(%)	R[3]	$\bar{X}$	CV(%)	R
17	0.22	--	0.17	--	.77	0.21	--	.95
32	0.23	--	0.17	--	.74	0.23	--	1.00
46	1.82	8	0.99	12	.54	1.50	9	.82
73	6.66	4	3.99	15	.60	6.33	9	.95
94	11.03	3	7.79	7	.71	9.88	.5	.90
119	16.31	5	15.34	6	.94	16.18	3	.99
190	33.45	10	34.81	18	1.04	35.25	14	1.05
263	77.60	13	73.60	14	.95	80.00	7	1.03

[1]Cells/Ml. x 10^6
[2]Coefficients of Variability
[3]Ratios of Treatment Means to Control Means

The results of this study indicate that the PCBs tested exerted temporary depressing effects on the population growth of Chlorella pyrenoidosa and only at a concentration substantially higher than that found currently in most ecosystems.

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