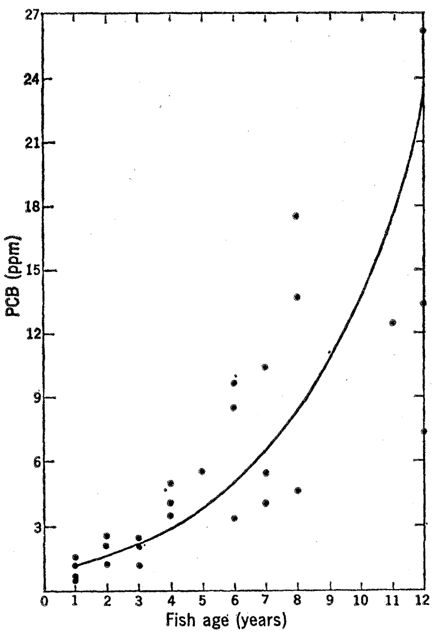

Fig. 1. The concentration of PCB's in Cayuga Lake trout as a function of age.

ppm). The mass spectra obtained from both a Perkin-Elmer 270 and a Finnigan 1015 quadrapole gas chromatograph–mass spectrometer were essentially identical with the mass spectra of the corresponding isomers of Aroclor 1254 standard.

There are many sources of PCB's in the environment, and it is not known what combination of these leads to contamination of Cayuga lake trout. The increase in PCB concentration with trout age is the same trend noted for DDT residues in this series of fish. The larger variation in PCB concentrations among individual 11- or 12-year-old fish may be due to greater differences among foraging, metabolic, and excretory capabilities of these older fish.

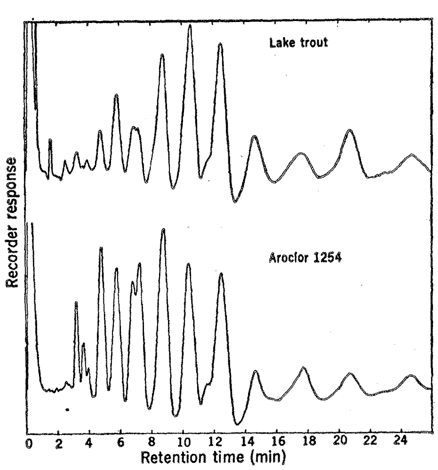

Fig. 2. Gas chromatograms of a 12-year-old Cayuga Lake trout (injected sample represents 0.12 mg of fish) and 1 ng of Aroclor 1254 standard.

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30 June 1972

Atmospheric Carbon Dioxide: Its Role in Maintaining Phytoplankton Standing Crops

Abstract. *The rate of invasion of carbon dioxide into an artificially eutrophic Canadian Shield lake with insufficient internal sources of carbon was determined by two methods: measuring the carbon : nitrogen : phosphorus ratios of seston after weekly additions of nitrogen and phosphorus, and measuring the loss of radon-222 tracer from the epilimnion. Both methods gave an invasion rate of about 0.2 gram of carbon per square meter per day. The results demonstrate that invasion of atmospheric carbon dioxide may be sufficient to permit eutrophication of any body of water receiving an adequate supply of phosphorus and nitrogen.*

The atmosphere is frequently considered to be an insignificant source of CO_2 for phytoplankton production (*1*). Yet, by adding phosphate and nitrate we have been able to increase by almost two orders of magnitude the standing crop of phytoplankton in a small Canadian Shield lake which has extremely low concentrations of natural dissolved inorganic carbon (less than 0.6 mg/liter in the epilimnion in midsummer). The lake appears to have obtained the carbon necessary to produce the algal bloom from the atmosphere (*2, 3*). We report here measurements of the invasion rate of atmospheric CO_2 into the waters of this experimentally enriched lake. The results were obtained between 4 and 18 August 1970.

Lake 227, a small oligotrophic lake in the experimental lakes area of the Fisheries Research Board near Kenora, Ontario, was enriched with 0.34 g of phosphorus (as Na_2HPO_4) and 5.0 g of nitrogen (as $NaNO_3$) per square meter per year in 1969, and 0.48 g of phosphorus (as H_3PO_4) and 7.2 g of nitrogen (as $NaNO_3$) per square meter per year in 1970. The chemical additions were made in 17 weekly increments in 1969 and 21 weekly increments in 1970. The midsummer phytoplankton standing crop, as measured by chlorophyll *a* concentrations, increased from 1 to 3 μg/liter in 1968 (before fertilization) to 50 to 100 μg/liter after the additions. The uptake of CO_2 by this phytoplankton caused an increase in the *p*H of the euphotic zone from normal values of 6 to 7 to values ranging from 9.5 to 10.2 during summer, so that very little of the dissolved inorganic carbon was present as gaseous CO_2 (*2, 3*). The concentration of dissolved inorganic carbon ($\Sigma CO_2 = CO_2 + HCO_3^- + CO_3^{2-}$), however, did not change greatly after fertilization. The midsummer concentrations of total CO_2 in the epilimnion ranged from 20 to 50 μmole/liter (0.24 to 0.60 mg/liter). As a result of the high *p*H and low concentration of CO_2, the partial pressure of CO_2 in epilimnetic waters (calculated from the *p*H, total CO_2 concentration, and temperature) was far below that of the atmosphere (Fig. 1); this created a pronounced concentration gradient from the atmosphere into the lake.

It is impossible to calculate the magnitude of CO_2 exchange between the atmosphere and the water from the difference in partial pressures alone.

Chemical, biological, and physical conditions in the water must also be taken into account. These may in turn be affected by uptake of CO_2 by organisms in the lake. For these reasons the following procedures were employed to calculate the flux of CO_2 from the atmosphere into the lake.

In the first method we took advantage of the simplicity of the Lake 227 system. The drainage basin is devoid of any significant calcareous material (*4*). The input of phosphorus and nitrogen during the period was known, since virtually all of it was supplied by the weekly fertilizations. Inflow from natural sources and outflow were measured as well (*3*). A complete determination of carbon, phosphorus, and nitrogen was done on the lake at weekly intervals.

The period selected for this calculation was 4 to 18 August 1970, when the radon experiments mentioned below were done. The phytoplankton standing crop was at its maximum for the summer (at a chlorophyll a concentration of 90 μg/liter), and inflows and outflows to the lake were completely dry due to lack of precipitation during the period and the preceding weeks.

The weights of suspended and dissolved carbon and phosphorus in the epilimnion of the lake at the beginning and end of the period are given in Table 1. There was an increase of 6.0 kg of carbon and 1.29 kg of phosphorus during the period. We also know that 2.28 kg of phosphorus, as dissolved phosphate, was added to the epilimnion during the period. This was taken up by phytoplankton within a few hours after addition (*3*). By subtracting the increase in total phosphorus, we find that the lake lost 0.99 kg of phosphorus during the period. Since the outflow was not running, and removal in the littoral zone of the lake is insignificant (*5*), the phosphorus must have been removed by phytoplankton and sedimented.

Checking the ratio of carbon to phosphorus in suspended matter (Table 1), we find it to average 125 : 1 by weight. Since no carbon was added as fertilizer, inflows were absent, and the return of carbon from the hypolimnion and sediments was negligible (*6*), the atmosphere was the only possible source.

If we assume an average carbon : phosphorus ratio of 125 : 1 in sedimenting matter, 124 kg of carbon must

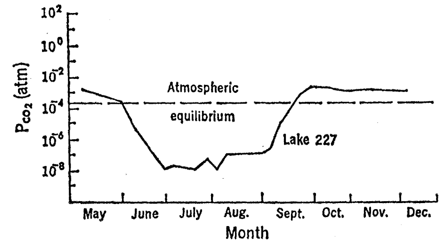

Fig. 1. (Solid line) Partial pressure of CO_2 at a depth of 1 m in Lake 227, calculated from total CO_2, *p*H, and temperature; (dashed line) partial pressure of atmospheric CO_2.

have entered the lake from the atmosphere to balance the known loss of phosphorus. An additional 6 kg of carbon were required to account for the observed carbon increase in the seston of the epilimnion, so that an average invasion of 0.19 ± 0.05 g of carbon per square meter of lake surface per day was required. This is enough to account for the carbon utilized in photosynthesis during the period (*5*). Hence, atmospheric carbon is a significant factor in the eutrophication of Lake 227.

For large lakes and oceans, where all inflows, outflows, and exchanges cannot be accurately measured, a direct elemental balance of the above type is impossible. To measure gas exchange in such situations, Broecker and his colleagues (*7*) have used ^{222}Rn, a biologically inert nuclide which is generated within natural bodies of water by the radioactive decay of ^{226}Ra. A calculation of the CO_2 flux from radon measurements is difficult, however, since the rate of CO_2 exchange is enhanced by the reaction of CO_2 with water and OH^- ions to form HCO_3^- and CO_3^{2-} (*8*). We therefore performed a series of measurements both in Lake 227 and in a large polyethylene tank, to evaluate the method for use in large-scale gas exchange work.

A large and variable quantity of radon reaches Lake 227 from groundwater, which makes it impossible to use the distribution of natural radon and radium to measure the exchange rate. For this reason we diluted approximately 9.7 mc of ^{226}Ra (as dissolved $RaBr_2$) to 50 liters and distributed it throughout the epilimnion of the lake in late July. This brought the activity of the epilimnetic water from approximately 0.5 to approximately 200 disintegrations per minute per liter. The epilimnion of the lake was allowed to mix for 6 days after the addition to ensure uniform lateral distribution of the radium and regeneration of any radon lost during the handling and spiking procedure. After this, sets of samples were taken at 3-day intervals and analyzed for ^{222}Rn and ^{226}Ra according to the methods of Broecker *et al.* (*7*). The samples were taken in the epilimnion and upper hypolimnion at depths of 0, 1, 2, 2.5, 3, and 5 m. Since the partial pressure of radon in the water greatly exceeded that in the atmosphere, radon evasion to the atmosphere could be estimated by measuring the radon depletion in surface waters.

Although the experiment was complicated by preferential sorption of radium to the lake bottom in the littoral zone and uptake of radium by seston, we were able to place limits on the rate of radon loss. The limits, expressed in terms of the velocity of an imaginary piston rising through the water column and sweeping before it the contained gas, were 0.15 to 0.60 m/day, with an average of about 0.4 m/day (*9*). Expressed in terms of the thickness of a stagnant boundary layer, this represents a value of 295 μm (*10, 11*). In the absence of chemical enhancement, this boundary layer would yield an invasion of carbon of 0.04 g/m^2-day. Therefore, the chemical enhancement factor for CO_2 required to explain the carbon found by elemental balance is approximately 3.

This result was checked by an addi-

Table 1. Weights of suspended and dissolved carbon and phosphorus and the carbon : phosphorus ratio in suspended matter in the epilimnion of Lake 227 during the experimental period in 1970.

Date	Suspended (kg)		Dissolved (kg)		Total (kg)		C : P (by weight)
	C	P	C	P	C	P	
4 August	382.3	2.49	23.2	0.62	405.5	3.11	154
18 August	360.8	3.56	50.7	0.83	411.5	4.39	101
Difference	− 21.5	1.07	27.5	0.21	6.0	1.29	

tional experiment. A polyethylene tank 10 cm deep and 200 cm in diameter was shielded from direct sunlight, filled with distilled water, and exposed to the air. Radium was added, and the turbulent conditions were varied by changing the rate at which water was recirculated within the tank until the rate of radon loss yielded a piston velocity equal to that observed for Lake 227. Potassium hydroxide was then added, bringing the *p*H to approximately 10 (that of the Lake 227 epilimnion). The change in CO_2 content with time was monitored. The rate of CO_2 invasion was found to be between three and four times that expected in the absence of any chemical enhancement for these physical conditions (*11*). Hence, the enhancement factor calculated from the tank experiment agrees satisfactorily with that calculated from the elemental balance data for Lake 227.

We conclude that invasion of CO_2 from the atmosphere supplies sufficient carbon to Lake 227 to produce an algal bloom in proportion to the amount of phosphorus and nitrogen added. Since the lake had an unusually low natural carbon content and is extremely well protected from wind, we further conclude that invasion of CO_2 from the atmosphere is likely to be sufficient for the eutrophication of any lake receiving sufficient supplies of nitrogen and phosphorus.

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6. D. R. S. Lean and D. W. Schindler (unpublished results) have studied movement of ^{14}C in labeled sediments and water columns for a period of 1 year. Less than 1 percent of the carbon that falls to the sediments and hypolimnion from the epilimnion ever reaches the euphotic zone of the lake again.
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$$F = (C^{\text{lake}} - P^{\text{atm}}\alpha)V$$

where F is the flux of the gas (moles per square meter), C is the concentration of the gas in the water (moles per cubic meter), P is the partial pressure of the gas in the atmosphere (atmospheres), α is the solubility of the gas in water (moles per cubic meter), and V is the piston velocity (meters per day). Since V has units of distance over time it may be written as the diffusion constant D for radon (that is, 1.36×10^{-5} cm²/sec), divided by a distance term Z, which has been described as representing "a stagnant boundary layer" on the water surface (*11*) and is used in models for gas exchange rates. Once Z is determined by radon measurements, F may be estimated for CO_2 gas because P, α, and D are known for that gas (here we have used 1.92×10^{-5} cm²/sec as the molecular diffusion constant for CO_2).
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Lead Poisoning: Rapid Formation of Intranuclear Inclusions

Abstract. *A single dose of lead (0.05 milligram per gram of body weight) induced characteristic intranuclear inclusions in the epithelium of proximal tubules in rat kidney within 1 to 6 days. The development of the intranuclear inclusions is thus an acute manifestation of lead poisoning, not a delayed one, as has been thought hitherto. Cytoplasmic structures resembling the intranuclear inclusions and situated in the vicinity of endoplasmic reticulum were regularly found in cells bearing the pathognomonic intranuclear inclusions. The latter and the cytoplasmic structures may be derived from a common precursor, perhaps a soluble protein-lead complex.*

In man and in various animals the pathognomonic morphological feature of chronic intoxication with lead is the presence of characteristic intranuclear inclusions in proximal tubular epithelial cells of the kidneys (*1*). The inclusion bodies are eosinophilic and do not appear to contain DNA or RNA but do contain protein, presumably of nonhistone type (*2*). Chemical analysis of inclusions separated from cell homogenates by centrifugation has indicated the presence of lead as well as of protein in the inclusions (*3*). The development of these intranuclear inclusions in proximal tubular epithelial cells has generally been connected with chronic lead poisoning and has been thought to require 4 to 8 weeks of exposure to lead (*1–3*). We have found, however, that in rat kidneys typical intranuclear inclusions occur as early as 24 hours after a single intraperitoneal injection of lead. This indicates that the development of the pathognomonic intranuclear inclusions in renal tubular epithelium is an acute manifestation of lead poisoning.

Young adult female Sprague-Dawley rats, weighing 220 to 270 g, were injected intraperitoneally with lead acetate in sterile water, in doses of 0.05 to 0.20 mg of lead per gram of body weight. From 1 to 6 days after the single dose of lead, tissue was taken from the cortex of each kidney, fixed for 3 hours in 2 percent glutaraldehyde in Millonig's (*4*) buffer (*p*H 7.35) and then for 1 hour in 1 percent osmium tetroxide in Millonig's buffer, and subsequently embedded in Epon 812 epoxy resin (*5*). Thin sections were prepared for electron microscopy and were examined with a Siemens Elmiskop 101 at 80 kv after staining with Reynolds' stain (*6*) or after staining with saturated aqueous uranyl acetate followed by Reynolds' stain.

Results of a survey of kidneys from 38 rats are shown in Table 1. Intranuclear inclusions were detected in 10 to 25 percent of epithelial cells in proximal tubules as early as 1 day after the injection of lead. These inclusions were found only in the epithelium of proximal tubules and not elsewhere in the nephron. The percentage of proximal tubular cells with intranuclear inclusions during the 6-day period was not significantly changed by increasing the dose of lead. As seen by electron microscopy of thin sections, most of