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Changes of photosynthetic production of organic carbon in Antarctic marine diatoms influenced by Aroclor 1254

ABSTRACT: Experiments have been carried out on the influence exerted by Aroclor 1254 upon the photosynthetic production of organic ^{14}C by an assemblage of marine Antarctic diatoms (*Thalassiosira* sp. 48%, *Nitzschia* sp. 21%, *Chaetoceros* sp. 15% and *Corethron criophilum* 10%). Samples of various numbers of cells per cm^3 of water have been used. Incorporation of $^{14}CO_2$ by the diatoms proved to be proportional to the increased number of cells in the sample only at the lowest levels of concentration in per cm^3. Further increase of the level of ^{14}C in diatoms has not been found as number of cells in the sample kept growing. Calculation of brutto photosynthesis has indicated that low concentration of Aroclor 1254 (0,01 to 1 ppm) may stimulate the photosynthetic incorporation of carbon, yet the photosynthetic release of carbon from cells within the photorespiratory process is stimulated to a higher degree. High concentration of Aroclor (1 to 50 ppm) inhibit the brutto assimilation, yet the release of carbon during the photorespiratory process is inhibited to a higher degree. A hypothesis is being considered implying that the relation between the intensity of photosynthesis and intensity of photorespiration may vary according to the rate of concentration of Aroclor.

Key words: Aroclor 1254, Antarctic marine diatoms, photosynthetic production of organic carbon.

Introduction

Experimental use of assemblages of cells of varying composition as well as the use of samples with varied concentration of cells has caused great difficulties in comparing the results obtained in various laboratories studying the influence exerted by chlorinated hydrocarbons (CHs) on the photoassimilation of $^{14}CO_2$ by diatoms. Actually, such a factor as the rate of concentration of cells in the sample may essentially influence the assimilation of $^{14}CO_2$. This is the reason why methodical investigation has been carried out to determine the dependence of assimilation of $^{14}CO_2$ of the number of cells in a sample. For these experiments we have chosen a compound whose impact upon the photosynthesis was the lowest, i.e. the Aroclor 1254 (Łukowski, Bystrzejewska, Ligowski 1989). At the same time a closer look was taken at the mechanism of the said compound by the diatoms. An attempt has also been made to determine the influence exerted by Aroclor 1254 upon the following two components of photosynthesis:

1) intensity of photosynthetic incorporation of $^{14}CO_2$,
2) intensity of photosynthetic release of ^{14}C in the photorespiratory process and extracellular release of photosynthetic ^{14}C-products.

Material and methods

In the experiment the phytoplankton was used that had been hauled by using Copenhagen type net with a mesh size of 55 μm from a water column 0—100 m in the South Shetland Islands region. The phytoplankton contained nothing but diatoms. The cell suspension has been transferred in portions of 50 cm^3 into waterproof, transparent plastic containers placed in the artificially lit chamber. Light intensity inside those plastic containers amounted up to 2000 lx. The containers were constantly cooled to the temperature of 2°C by continuous flow of outboard water.

The chlorinated hydrocarbon Aroclor 1254 dissolved in acetone was then added to the containers with suspension of diatoms. In 24 hours after its introduction (12 hrs of light and 12 hrs darkness) 5 μCi of ^{14}C (37 000 Bq) was added to each portion and then the samples were exposed to light for 4 hrs. The assimilation process of $^{14}CO_2$ has been interrupted by addition of formaline. The samples were then filtrated through the Millipore membrane filters of 1,2 μm pore diameter, whereas the cells that remained on the filters were used to estimate the radioactivity of the incorporated ^{14}C by means of a Geiger-Müller counter. The radioactivity of the samples was expressed in Bq. For control samples were used that have been treated with acetone only (100 μl) as well as samples without acetone.

The influence of Aroclor 1254 on photoassimilation of $^{14}CO_2$ was examined in relation to the number of diatom ells in the sample. The following diatoms were dominant inside the sample: *Thalassiosira* sp. (abt. 48%), *Nitzschia* sp. (abt. 21%), *Chaetoceros* sp. (abt. 15%), *Corethron criophilum* (abt. 10%). Concentration of cells within the consecutive samples was following: 2450, 4800 and 7500 cells in 1 cm^3.

Intensity of incorporation of $^{14}CO_2$ by diatoms was determined, and the loss in photosynthetic production of organic carbon (photosynthetic loss = = PL) in the light in the photorespiratory process and by extracellular release of photosynthetic products was calculated:

$$PL = P_A - P_B,$$

where:

P_A — intensity of photosynthetic production of ^{14}C-organic by a cell from a sample of the density of 2400 cells/cm^3,

P_B — intensity of photosynthetic production of ^{14}C-organic by a cell from a sample of the density of 7500 cells/cm^3.

Results and discussion

The results obtained made it possible to determine the relation between the assimilation of $^{14}CO_2$ and the density of cells. It appeared that only in low concentrations the radioactivity increased along with cell number; in higher density of cells a decrease of radioactivity was observed (Fig. 1). Since the amount of the initially applied radioisotope has been 20-fold greater than the level of radioactivity incorporated by the diatoms, we can rule out the possibility that in the samples of the highest cell density the substrate for the photosynthetic assimilation was missing.

In the case of the Aroclor 1254-treated samples it might appear that the accumulation of the lower radioactivity by the greater number of diatoms than the amount accumulated by the smaller number of cells was caused by the fact that with high density of cells only a part of them was subject to toxication. This is, however, contradicted by the character of the control curve.

A possibility has also been considered that the cells overshadowed each other in the high-density samples; it is namely well known that the influence exerted by CH_s upon the photosynthetic assimilation of carbon depends to a large extent on intensity of light (Farlane, Gloschenko and Hawis 1972). Probably the continuous mixing up of samples resulting from movements of the ship all throughout the experiment was insufficient therefore the higher cell density could result in the changes of photosynthesis and an

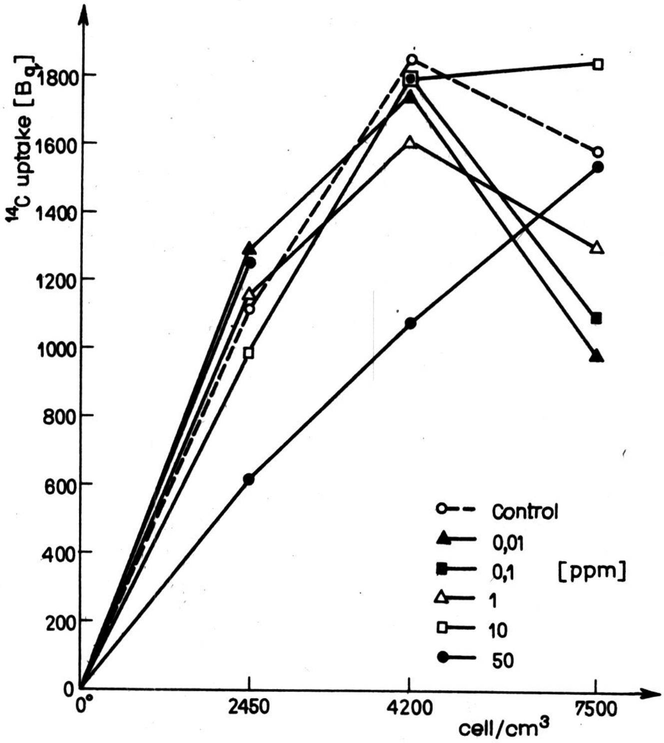

Fig. 1. The incorporation of ^{14}C in the process of photosynthesis of marine Antarctic diatoms in relation to the number of cells in 1 cm^3 under influence of different concentrations of Aroclor 1254

increase of extracellular release of photosynthetic products (Nalewajko and Lean 1977). One has to consider the excretion of the assimilated $^{14}CO_2$ to the environment, since the photosynthetising cells do not retain the total of the assimilated carbon but release part of it to the environment at the same time. The release of the previously assimilated carbon takes place in process of extracellular release of phytosynthetic products (Chróst 1986) and during the photorespiratory process, simultaneous to the photosynthesis, where some diatoms release CO_2 while others release the ions HCO_3^-, and still others release the glicolate that belongs to the intermediate metabolites of the photorespiratory path (Dowton and Treguna 1968, Kowalik and Schmid 1971, Dohler and Koch 1972, Stabenau 1972).

The inter-relations between the photosynthetic and the photorespiratory processes influence significantly the ratio ^{12}C to ^{14}C in the environment surrounding the cells. Samples with varied concentration of cells but of constant volume may thus substantially differ in the ^{12}C to ^{14}C ratio. Measurements of assimilation of $^{14}CO_2$ by plants reveal a netto phytosynthesis since it is diminished by an amount of ^{14}C released during the photorespiratory process. Hence the differences in amounts of the ^{14}C incorporated by a single cell from a sample of low density and the level of ^{14}C found in a cell from a sample of high density may present an approximately estimated measure of the release of ^{14}C within the photorespiratory process and in process of extracellular release of photosynthetic products (Fig. 1).

Admitting such assumptions we have analyzed the influence of Aroclor 1254 upon the netto photosynthesis calculated per cell derived from samples of low density, where incorporation of ^{14}C was dependant on number of cells as well as on the influence upon the photorespiration measured by the difference in radioactivity of such a cell and the radioactivity of a cell derived from a sample of the highest density (Fig. 2).

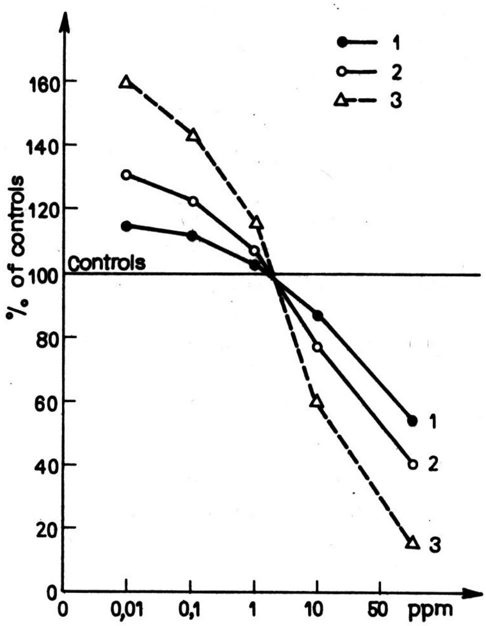

Fig. 2. The influence of Aroclor 1254 on photosynthetic ^{14}C-incorporation in the cell and on the photosynthetic release; 1 — netto photosynthesis, 2 — brutto photosynthesis, 3 — PL--photosynthetic release

The Aroclor 1254 has not inhibited the netto photosynthesis in diatoms until their concentration reached more than 1 ppm. The obtained calculation suggests at the same time that concentrations of Aroclor lower than 1 ppm will stimulate intensity of the release of ^{14}C in the photorespiratory process while this stimulation diminishes along with the increasing concentration of Aroclor. Concentrations of Aroclor higher than 1 ppm will inhibit that process along with increased concentration of the said compound.

Calculations of the brutto photosynthesis which require taking into account the corrections for losses of $^{14}CO_2$ due to photorespiration show that low concentration of Aroclor 1254 may stimulate the photosynthetic incorporation of carbon, while that stimulation seems to be weaker than that in the photorespiration. High concentrations of Aroclor inhibit the brutto photosynthetic assimilation $^{14}CO_2$, yet they inhibit the release of incorporated ^{14}C in the photorespiratory processs to a still greater extent. At the same time the mechanism points to a possibility of altered relations between intensity of photosynthesis and intensity of photorespiration caused by changes in the concentration of Aroclor. The relation between the photosynthetic incorporation of carbon and its release due to photorespiration points to the state of dynamic balance between those two processes. The influence of Arclor 1254 concentration upon the changes in that balance may present the measure of the toxic effect of the applied compound and points to the regulatory mechanism acting within the cells subjected to the action by the Aroclor.

The inhibition of photosynthesis by Aroclor 1254 in concentrations of over 1 ppm may be explained by the damages to the chloroplasts. According to Moore and Harriss (1972) the total disintegration of chloroplasts occurs within 24 hrs when the concentration of this compound reaches 1 ppm. Under such conditions it is not only the photosynthetic process that is upset but the same thing happens to the photorespiratory process occurring within the chloroplasts. The results of our calculations have confirmed the fact that the Aroclor induced inhibition of netto photosynthesis is accompanied by inhibition of extracellular release of organic ^{14}C.

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Streszczenie

Prowadzono doświadczenia nad wpływem Arocloru 1254 na fotosyntetyczną produkcję ^{14}C-organicznego u okrzemek zawartych w fitoplanktonie sieciowym pobieranym z morskich wód antarktycznych. Do badań wykorzystano próby o różnej gęstości komórek w zbiorowisku złożonym głównie z przedstawicieli: *Thalassiosira* sp. — 48%, *Nitzschia* sp. — 21%, *Chaetoceros* sp. — 15%, *Corethron criophilum* — 10%. Włączanie ^{14}C przez okrzemki było proporcjonalne do wzrostu ilości komórek w próbie tylko przy niskim zagęszczeniu komórek w cm^3. Przy wzroście liczby komórek w testowanych próbach nie stwierdzono zwiększania poziomu ^{14}C w okrzemkach. Wyniki takie uzyskano zarówno dla prób kontrolnych, jak i zawierających Aroclor 1254, z wyjątkiem prób, których stężenie tego związku w wodzie było najwyższe (50 ppm). Dyskutowane są przyczyny zróżnicowanego efektu Arocloru 1254 w zależności od ilości komórek w próbie. Obliczenia fotosyntezy brutto wskazują, że niskie stężenie Arocloru 1254 (0.01—0.1 pp) mogą stymulować fotosyntetyczne włączanie węgla, jednak bardziej stymulują uwalnianie węgla z komórek w procesie fotooddychania oraz w procesie pozakomórkowego wydzielania produktów fotosyntezy. Wysokie stężenia Arocloru 1254 (1—50 ppm) hamują asymilację węgla brutto, jednak w większym jeszcze stopniu hamują uwalnianie węgla w procesie fotooddychania oraz przyżyciowego wydzielania pozakomórkowego. Sformułowano hipotezę, że w zależności od stężenia Arocloru 1254 zmienia się relacja pomiędzy natężeniem fotosyntezy a intensywnością fotooddychania i pozakomórkowego wydzielania węgla organicznego.