Low Ambient Level Uptake of ¹⁴C-DDT by Three Species of Marine Phytoplankton

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The degree to which marine phytoplankton accumulate DDT residues from the environment must be known in order to assess the possibility of interference with photosynthetic processes (11) and the potential for transfer of these residues to higher levels in marine food chains. Freed (6) recently re-emphasized the need to view accumulation of pesticides in biological materials in terms of a partition coefficient, implying that an equilibrium is reached between ambient and internal concentrations of the pesticide materials. Södergren (9) showed that ¹⁴C-DDT uptake in the fresh water phytoplankter Chlorella sp. was due to rapid, passive absorption, thus indicating a partition mechanism. Uptake studies with a marine diatom reported by Keil and Priester (7) were long term, used concentrations which exceeded normal levels for marine waters, and did not account for losses of material due to codistillation of DDT with water (2). Since high concentrations may affect the apparent partition coefficient of an organism for DDT residues in water, as shown experimentally by Butler (4), it is desirable to determine partition coefficients at concentrations similar to those in the natural environment. We thus undertook to measure 14C-DDT uptake for three species of marine phytoplankton in pure culture.

Methods

Inocula for axenic cultures of <u>Syracosphaera carterae</u> (a coccolithophorid), <u>Amphidinium carteri</u> (a dinoflagellate), and <u>Thalassiosira fluviatilus</u> (a centric diatom) were obtained from the Institute of Marine Resources culture collection in La Jolla, California. These were grown on IMR medium (5) in sterile, 2 liter culture flasks kept at 18[±]l^oC and illuminated daily with 12 hour periods of 550 foot candles of light. Cultures were used in experiments just as they reached maximum total growth.

Replicate 100 ml aliquots of the algal suspension were analysed for particulate oxidizable carbon (10) prior to the uptake experiments. Ring-labelled ¹⁴C-DDT (Nuclear-Chicago CFA-226) was made up to 100 ppm in ethanol solution. Dilutions of this ethanol stock solution were made by adding 100 microliters to fresh, membrane-filtered IMR medium. At the low concentrations used, much of the isotope was effectively lost at the glass-liquid interface, so our determinations of the concentration of ¹⁴C-DDT in the diluted aqueous stock were always less than the nominal concentration based on activity measurements of the added 100 microliters. Repeated subsamples of the diluted aqueous stock varied greatly in activity unless the vessel was kept stoppered and stirred with a magnetic stirrer. Pipettes used for aliquots were prewetted with the solution, since it was found that initial aliquots were always lower in activity. All glassware to receive aqueous solutions were similarly prewetted to prevent adsorptive losses.

For the uptake experiments, 100 ml of the algal suspension was added to a 250 ml flask along with an amount (ca. 20 ml) of the aqueous stock solution of the isotope sufficient to give the desired concentration. Replicates were run for most concentrations. The mixtures were stoppered, agitated, and allowed to equilibrate for a few minutes. Due to the short time of exposure before uptake, there was probably negligible conversion from the parent compound (p,p'-DDT); gas chromatographic checks of the labelled stock showed no evidence of decomposition. The contents of the flask were then filtered onto a glass fiber filter (a few onto membrane filters) and dessicated for 24 hours. Filters were removed from the dessicator, placed in a scintillation vial with 10 ml of toluene scintillation fluid, and counted in a scintillation counter. Replicate 1 ml aliquots from the aqueous stock solution were taken before and after each addition to the algal suspensions and were counted in Bray's solution (3). The total amount of activity added was calculated from the counts found in these aliquots. The concentration of the labelled DDT in the aqueous stock solution declined during the course of each experiment. possibly due to codistillation during the time the vessel was open.

Results and Discussion

The results are summarized in Table I. Note that 16% to 54% of the initially added $^{14}C-DDT$ was removed from the water by the algal cells. The partition coefficients are calculated on the basis of the final equilibrium concentration of the $^{14}C-DDT$ in the medium.

The concentration factor for the marine phytoplankton tested exceeds the estimate of Keil and Freister (7) for <u>Cylindrotheca closterium</u>, even when correction is made for our measurement of algal material in terms of oxidizable carbon. True partition coefficients calculated using known carbon to volume percentages for the algal clones used in these experiments are 2.5×10^4 for <u>S</u>. <u>carterae</u> and <u>T</u>. <u>fluviatilus</u>, and 8.0×10^4 for <u>A</u>. <u>carteri</u>. These values are equivalent to wet weight concentration factors.

Using the estimate of 1.9×10^5 for a relative partition coefficient in combination with a local estimate of the concentration of DDT residues in whole seawater at 15 ppt (8), we obtain an expected value of about 30 ppm (oxidizable carbon) DDT residues. This value is encompassed by the confidence interval (95%) of values obtained by GLC-EC

Table I.		¹⁴ C-DDT Uptake by Three Species of Marine Phytoplankton	Species of	Marine Ph	ytoplankton	
	Total*	Uptake*	Cell C	Equil.	ppb/C tn	Relative Part.
Species	pg ¹⁴ C-DDT	pg 14C-DDT	5m	ppt	algae	
<u>Syracosphaera</u> carterae	274±34(2)	143±31(2)	301	2.0	475	2.3 x 10 ⁵
=	Ξ	112±16(2)	151	2.6	741	2.9 x 10 ⁵
<u>Thalassiosira</u> fluv.	362±20 (2)	238±32 (2)	730	2.4	326	1.4×10^{5}
Ξ	Ŧ	194±15(2)	365	3°3	532	1.6 x 10 ⁵
<u>Amphidinium</u> carteri	32 3±22 (2)	273±6(2)	1226	1.0	223	2.2 x 10 ⁵
=	128(1)	68(1)	961	0.5	70	1.4 x 10 ⁵
Ξ	192 (1)	92 (1)	=	0.8	96	1.2×10^{5}
\star Mean followed with standard deviation of the estimate; number of values shown in parentheses. The symbol pg denotes picograms, or 10 ⁻¹² grams.	andard deviati i picograms , c	on of the estim or 10 ⁻¹² grams.	late; numbei	r of values	shown in p	barentheses.

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analyses of phytoplankton samples from the same approximate time and location as the water concentration determination.

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Literature Cited

- BIGGAR, J.W., G.R. DUTT, and R.L. RIGGS. Bull. Environ. Cont. Toxicol. 2,90 (1967)
- BOWMAN, M.C., F. ACREE, JR., C.S. LOFGREN, and M. BEROZA Science 146,1480 (1964)
- 3. BRAY, G.A. Anal. Biochem. 1,279 (1960)
- 4. BUTLER, P.A. Symposium on the Biological Impact of Pesticides in the Environment, Oregon State University, Corvallis (In Press)
- EPPLEY, R.W., R.W. HOLMES, and J.D.H. STRICKLAND J. Exp. Mar. Biol. Ecol. 1,191 (1968)
- 6. FREED, V.H. Symposium on the Biological Impact of Pesticides in the Environment, Oregon State University, Corvallis (In Press)
- 7. KEIL, J.E. and L.E. PRIESTER. Bull. Environ. Cont. Toxicol. 4,219 (1969)
- ODEMAR, M.W., P.W. WILD, and K.C. WILSON. California Dept. Fish and Game, MRO Ref. No. 68-12 (1968)
- 9. SODERGREN, A. Oikos 19,126 (1968)
- 10. STRICKLAND, J.D.H. and T.R. PARSONS. Fish.Res. Bd. Canada Bull. 167 (1968)
- 11. WURSTER, C.F. Science 159,1474 (1968)