References

1. N. Nakanishi, M. Koreeda, S. Sasak, M. L. Chang, H. Y. Hsu, *Chem. Commun.* No. 24, 915 (1966); T. Ohtaki, R. D. Milkman, C. M. Williams, *Proc. Nat. Acad. Sci. U.S.* **58**, 981 (1967); J. Jizba, V. Herout, F. Sorm, *Tetrahedron Lett.* **1967**, 1689 (1967); H. Rimpler and G. Shulz, *ibid.*, p. 2033; T. Takemoto, Y. Hikino, K. Nomoto, H. Nikino, *ibid.*, p. 3191; H. Hoffmeister, G. Heinrich, G. B. Staal, W. J. van der Burg, *Naturwissenschaften* **54**, 471 (1967); S. Imai, S. Fujuka, M. Nakanishi, M. Koreeda, T. Kurokawa, *Steroids* **10**, 557 (1967); H. Hoffmeister, K. Nakanishi, M. Koreeda, H. Y. Hsu, *J. Insect Physiol.* **14**, 53 (1968).
2. J. N. Kaplanis, M. J. Thompson, W. E. Robbins, B. M. Bryce, *Science* **157**, 1436 (1967).
3. M. N. Galbraith and D. H. S. Horn, *Chem. Commun.* No. 24, 905 (1966).
4. P. E. Ellis, *Anim. Behav.* **11**, 142 (1963).
5. G. B. Staal, thesis, Laboratorium voor Entomologie, Wageningen, Netherlands (1961).
6. M. J. Thompson, J. N. Kaplanis, W. E. Robbins, R. J. Yamamoto, *Chem. Commun.* No. 13, 650 (1967).
7. P. Hocks, G. Schulz, E. Watzke, P. Karlson, *Naturwissenschaften* **54**, 44 (1967).
8. H. Hoffmeister, C. Rufer, H. Ammon, *Z. Naturforsch.* **20b**, 130 (1965).
9. D. B. Carlisle, D. J. Osborne, P. E. Ellis, J. E. Moorhouse, *Nature* **200**, 1230 (1963).
10. V. B. Wigglesworth, *J. Exp. Biol.* **29**, 561 (1952).
11. M. J. Wells, *Quart. J. Microscop. Sci.* **95**, 231 (1954).
12. C. M. Williams, *Biol. Bull.* **103**, 120 (1952).
13. P. Karlson, *Vitamins and Hormones* **14**, 227 (1956).
14. W. Etkin, *Science* **139**, 810 (1963); *Neuroendrocrinology* **1**, 45 (1966).

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DDT Reduces Photosynthesis by Marine Phytoplankton

Abstract. *Concentrations of DDT [1,1,1-trichloro-2,2-bis(p-chlorophenyl) ethane] as low as a few parts per billion in water reduced photosynthesis in laboratory cultures of four species of coastal and oceanic phytoplankton representing four major classes of algae, and in a natural phytoplankton community from Woods Hole, Massachusetts. Toxicity to diatoms increased as cell concentration decreased. This inhibition may be of ecological importance.*

Recently it has become apparent that DDT (*1*) and its derivatives are among the most widely distributed synthetic chemicals on Earth. They are found not only in soils (*2*), runoff water (*3*), air, and rainwater (*4*), but also in most animals analyzed from widely diverse parts of the world, including Antarctica (*5*, *6*). Residues of DDT were recently reported in marine organisms of both the Atlantic and Pacific oceans, including pelagic birds at the top of wholly oceanic food chains (*7*); this and other evidence suggest widespread contamination of marine plankton by these chemicals. While components of the zooplankton have been shown to be highly susceptible to DDT (*8*, *9*), very little is known of its effects on phytoplankton. Since a substantial part of the world's photosynthesis is performed by phytoplankton (*10*), interference with this process could be important to the biosphere.

In order to determine the effects of DDT on photosynthesis by marine phytoplankton, species important as food organisms, representing four classes of phytoplankton, were chosen from laboratory cultures maintained at Woods Hole Oceanographic Institution (WHOI). These included the diatom *Skeletonema costatum*, isolated from Long Island Sound (WHOI clone "Skel"); the coccolithophore *Coccolithus huxleyi* from the Sargasso Sea (32°10′N,64°30′W; "BT-6"); the green alga *Pyramimonas* sp. from the Sargasso Sea (33°11′N,65°15′W; "13-10 Pyr"); and the neritic dinoflagellate *Peridinium trochoideum* ("Peri"). In addition, water from Vineyard Sound (Woods Hole) was tested, containing a typical neritic phytoplankton community dominated by various diatoms.

Fluorescent lights (5000 lux) on a cycle of 14 hours light and 10 hours dark were used at 17°C. The four laboratory cultures were grown axenically in half-strength medium "f" (*11*), an enriched sea water. In 125-ml erlenmeyer flasks, 50-ml portions of medium were inoculated to yield the appropriate cell concentrations, and these were cultured for 24 hours. This initial culture period was omitted with *Pyramimonas* and the Vineyard Sound water; the latter was filtered through 50-μ mesh to remove zooplankton and was enriched to "f/100." To each flask 1 or 2 μl of pure *p,p′*-DDT (*1*) in an ethanolic solution was then added to yield the desired concentrations of DDT. Control flasks received equal amounts of pure ethanol, with no detectable effect on results. After culturing for 20 to 24 more hours, ^{14}C-bicarbonate (*12*) was added, and the algae were illuminated for 4 to 5 more hours. A few controls were run in darkness. Cultures were then collected on 0.8-μ Millipore filters, by use of slightly reduced pressure, and washed three times with filtered sea water; the filtered cultures were dried and radiation was counted. The radioactivity retained by the filtered cells is related to the amount of carbon fixed by photosynthesis (*13*).

Cell concentrations were chosen to approximate densities found in nature. To facilitate qualitative comparisons between the four phytoplankton species, cell concentrations were calculated to yield approximately equal total cell areas at the end of the experiment. Cell area roughly correlates with metabolic activity in phytoplankton (*14*).

The data from these experiments are shown in Fig. 1; dark uptake of ^{14}C

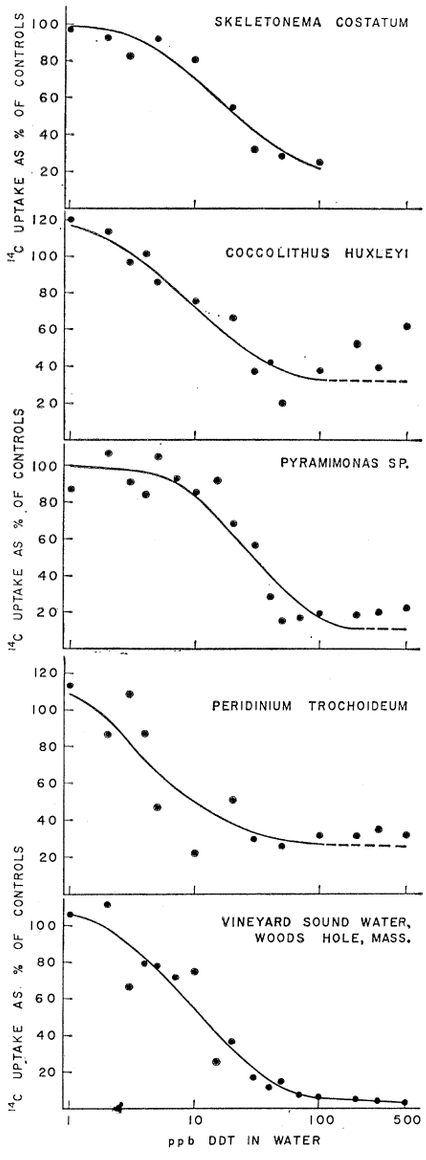

Fig. 1. Photosynthesis by phytoplankton at various concentrations of DDT, measured by uptake of ^{14}C (percentages) relative to uptake by controls. The following are, respectively, calculated cell concentrations (per milliliter) at the end of each experiment, mean counts per minute (cpm) for controls, uptake in the dark (percentages) relative to uptake by controls, and probabilities that a negative linear regression is random: *Skeletonema*, 1600, 1390 cpm, 1.1 percent, $P = .052$; *Coccolithus*, 4920, 1435 cpm, 39.7 percent, $P = .00074$; *Pyramimonas*, 2100, 564 cpm, 22.8 percent, $P = .032$; *Peridinium*, 240, 533 cpm, 53.4 percent, $P = .059$; Vineyard Sound water (as obtained), 6200 cpm, 0.6 percent, $P = .0011$.

was subtracted from all results. Probabilities (P) that a negative linear regression was random ranged from .0007 to .06, with $P < .0001$ for these five experiments combined. Data from DDT concentrations greater than 100 parts per billion (ppb) have been omitted from the regression calculations since they are not on the linear part of the curve and may be subject to anomalies caused by the limited solubility (1.2 ppb) of DDT in water (*15*). The fact that DDT could be immobilized as a solid when the solubility limit was so greatly exceeded may explain the slightly increased uptake of ^{14}C in some instances at 200 to 500 ppb of DDT.

Presumably the effect on phytoplankton occurs after absorption of DDT by the cells, a process to be expected from the very low solubility of DDT in water and its higher solubility in lipid-containing biological material. One might therefore expect increased effect with decreasing cell concentration, caused by an increased amount of DDT per cell. Accordingly in one experiment the concentration of DDT was held constant at various cell concentrations. The validity of the hypothesis is shown by the results of 10 ppb of DDT (Fig. 2): photosynthesis by *Skeletonema* decreased to half that by the controls as the cell concentration was reduced by two orders of magnitude.

The fact that these data apparently follow sigmoid curves is typical of dose-response relations (*16*) and suggests the absence of a threshold concentration of DDT below which no effects occur. Experimental scatter produced some uptake of ^{14}C above 100 percent at low concentrations of DDT, however. This should not be interpreted as a low-level stimulatory effect, a possibility that cannot be evaluated from these data.

It is known that DDT reduces Hill reaction activity in barley chloroplasts (*17*). Photosynthesis by two species of algae and by a natural phytoplankton community was diminished by 1000 ppb of DDT (*8*), but it now appears that at least some phytoplankton species are sensitive to much lower concentrations.

Evaluation of the significance of these findings to phytoplankton communities in nature is not simple, although the near-ubiquity of DDT residues implies a potential for widespread effects. Representative concentrations of DDT residues within natural phytoplankton communities are not generally known.

Water remote from a site of application of DDT usually averages less than 1 ppb (*3*), but concentrations in water may be misleading. Natural waters carry DDT in suspension, as well as particulates to which DDT is adsorbed at concentrations 10^4 to 10^5 times those in the water (*6*). In my experiments half the DDT may be lost by codistillation within 24 hours (*18*), so that DDT concentrations were probably much lower than is indicated (Figs. 1 and 2) when uptake of ^{14}C was measured.

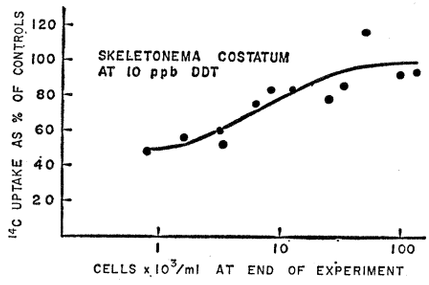

Fig. 2. Photosynthesis in *Skeletonema* at 10 ppb of DDT at various cell concentrations; $P = .032$.

Absorption of DDT by the cells further reduces the concentration in the water. In nature, concentrations in water are likely to be more constant, the DDT being replaced from persistent residues in mud, detritus, runoff water, and other sources as it is absorbed by cells. Thus a given concentration in natural waters would be of greater biological importance than the same initial concentration in vitro under the conditions of these experiments.

Water near a direct application of DDT to the environment, however, commonly contains concentrations comparable to those employed by me. Treatment with DDT of a Florida salt marsh yielded water containing up to 133 ppb for 1 week (*19*), some California waters contained as much as 22 ppb (*6*), and 14 to 20 ppb of DDT was added directly to Clear Lake, California (*20*). Under such circumstances, and especially where cell concentrations are low, photosynthesis and growth by phytoplankton (*21*) are probably reduced, so that the base of the food chain is diminished and higher organisms are affected.

Selective toxic stress by DDT on certain algae may alter the species composition of a natural phytoplankton community (*21*). This floral imbalance could favor species normally suppressed by others, producing population explosions and dominance of the community by one or a few species. This process would aggravate the problems of eutrophication caused by over-fertilization from sewage, agricultural runoff, and other artificial sources; it may partially explain the appearance of algal blooms during recent years within certain closed bodies of water. Such effects are insidious and their cause may be obscure, yet they may be ecologically more important than the obvious, direct mortality of larger organisms that is so often reported.

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References and Notes

1. 1,1,1-Trichloro-2,2-bis(*p*-chlorophenyl)ethane.
2. C. A. Edwards, *Residue Rev.* **13**, 83 (1966); H. Cole, D. Barry, D. E. H. Frear, A. Bradford, *Bull. Environ. Contam. Toxicol.* **2**, 127 (1967); G. M. Woodwell and F. T. Martin, *Science* **145**, 481 (1964).
3. L. Weaver, C. G. Gunnerson, A. W. Breidenbach, J. J. Lichtenberg, *Public Health Rep.* **80**, 481 (1965).
4. D. C. Abbott, R. B. Harrison, J. O'G. Tatton, J. Thomson, *Nature* **208**, 1317 (1965); *ibid.* **211**, 259 (1966); G. A. Wheatley and J. A. Hardman, *ibid.* **207**, 486 (1965); P. Antommaria, M. Corn, L. DeMaio, *Science* **150**, 1476 (1965).
5. G. M. Woodwell, C. F. Wurster, P. A. Isaacson, *Science* **156**, 821 (1967); N. W. Moore and J. O'G. Tatton, *Nature* **207**, 42 (1965); W. J. L. Sladen, C. M. Menzie, W. L. Reichel, *ibid.* **210**, 670 (1966 → J. Robinson, A. Richardson, A. N. Crabtree, J. C. Coulson, G. R. Potts, *ibid.* **214**, 1307 (1967); P. A. Butler, in *Trans. North American Wildlife Natural Resources Conf. 31st 14–16 March 1966*, pp. 184–9.
6. J. O. Keith and E. G. Hunt, in *Trans. North American Wildlife Natural Resources Conf. 31st 14–16 March 1966*, pp. 150–77.
7. R. W. Risebrough, D. B. Menzel, D. J. Martin, H. S. Olcott, *Nature* **216**, 589 (1967); C. F. Wurster and D. B. Wingate, *Science* **159**, 979 (1968).
8. P. A. Butler, *U.S. Fish Wildlife Serv. Circ. 167* (1963), pp. 12–20.
9. V. L. Loosanoff, J. E. Hanks, A. E. Ganaros, *Science* **125**, 1092 (1957); D. S. Grosch, *ibid.* **155**, 592 (1967); H. O. Sanders and O. B. Cope, *Trans. Amer. Fisheries Soc.* **95**, 165 (1966).
10. C. S. Yentsch, *Oceanog. Marine Biol. Ann. Rev.* **1**, 157 (1963).
11. R. R. L. Guillard and J. H. Ryther, *Can. J. Microbiol.* **8**, 229 (1962).
12. Comprising 0.1 to 0.5 ml of a sodium ^{14}C-bicarbonate solution, 10 μc/ml, pH 9.5; New England Nuclear Corp., Boston, Mass.
13. E. S. Nielsen, *J. Conseil Conseil Perm. Intern. Exploration Mer* **18**, 117 (1952); M. S. Doty and M. Oguri, *Pubbl. Staz. Zool. Napoli Suppl.* **31**, 70 (1959).
14. T. J. Smayda, *Inter-Amer. Trop. Tuna Comm. Bull.* **9**, 467 (1965).
15. M. C. Bowman, F. Acree, M. K. Corbett, *J. Agr. Food Chem.* **8**, 406 (1960).
16. W. W. Westerfeld, *Science* **123**, 1017 (1956).
17. P. D. Lawler and L. J. Rogers, *Nature* **215**, 1515 (1967).
18. F. Acree, M. Beroza, M. C. Bowman, *J. Agr. Food Chem.* **11**, 278 (1963).
19. R. A. Croker and A. J. Wilson, *Trans. Amer. Fisheries Soc.* **94**, 152 (1965).
20. E. G. Hunt and A. I. Bischoff, *Calif. Fish Game* **46**, 91 (1960).
21. N. Lazaroff and R. B. Moore, *J. Phycol. Suppl.* **2**, 7 (1966); E. L. Bishop, *Public Health Rep.* **62**, 1263 (1947).
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