DDT RESIDUES IN SEAWATER AND PARTICULATE MATTER IN THE CALIFORNIA CURRENT SYSTEM

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ABSTRACT

Continuous samples of seawater and organic particulate material collected along linear transects in the California current system were analyzed for DDT residues. DDT residue concentrations in whole seawater, as determined by continuous-flow, liquid-liquid extraction, ranged from 2.3×10^{-12} g/ml off Oregon and Washington, to 5.6×10^{-12} g/ml off southern California. Geographical patterns in these concentration values are discussed in relation to mechanisms of land-sea DDT residue transfer. DDT residue concentrations in particulate material collected by continuous-flow centrifugation and filtration of the centrifugal pellet onto GFC-glass-fiber filters, ranged from 1.2 to 5.7×10^{-6} g/g carbon (with one exception). These values were related to the density of the standing crops. DDT residues in this particulate fraction accounted for less than 10% of the DDT residues in the whole seawater samples. Residues which are fixed to particles of less than 1-2 μ in diameter may account for the balance of the DDT residues in the whole water samples. Experimental results are described which implicate adsorption as the uptake mechanism for algal cells; these experiments also support the idea that <1-2 μ diameter particles carry most of the DDT residues in whole seawater.

DDT and its metabolites have dispersed into the ocean and are found in high concentrations in the predators of oceanic food chains. Theoretical considerations predict a net transfer of extant DDT residues to the oceans, via atmospheric and river currents (Smith, 1970). In view of the well-known chemical stability of the principal constituents of the DDT complex, *p,p'*-DDT, DDD, and especially DDE, it is not surprising that levels of DDT residues in marine plankton samples have risen during the past decade (Cox, 1970a). No published data are available, however, on concentrations of DDT residues in seawater and in oceanic particulate matter. Chlorinated pesticides have been found in concentrations up to 13×10^{-9} g/ml in surface slicks in Biscayne Bay, Fla., and at concentrations of about $^{-12}$ g/ml in the surrounding waters (Seba and Corcoran, 1969). Measurements of DDT concentration in the open ocean are needed to construct a systematic account of DDT residue transport to the pelagic environment of the ocean, and to estimate the ultimate transport of DDT residues to the sediments.

METHODS AND MATERIALS

Samples of water and particulate material were collected during cruises of the RV *Proteus* in May 1970 from Monterey Bay, Calif., to San Diego, Calif., passing outside the islands off the southern California coast and returning closer to shore through the Santa Barbara Channel. A second cruise was made in September 1970 from Vancouver, British Columbia, to just off the mouth of San Francisco Bay, Calif. Figures 1 and 2 show the cruise tracks and the station enumeration for these cruises.

Sampling was continuous and was done while the ship was underway. Water was obtained from the shipboard seawater system (PVC and Teflon) which pumped water from about 1-2 m below the surface. The stream was first filtered through a 0.176-mm mesh net to remove larger zooplankton from the sampled water. The stream was then split; part of the water was directed into a peristaltic pump which metered the flow of particle-bearing water into a continuous-flow, internal recycle and recovery, liquid-liquid extracter of the type described by Kahn and Wayman (1964). Flow rates through the liquid-liquid extracter averaged 480 ml/hr.

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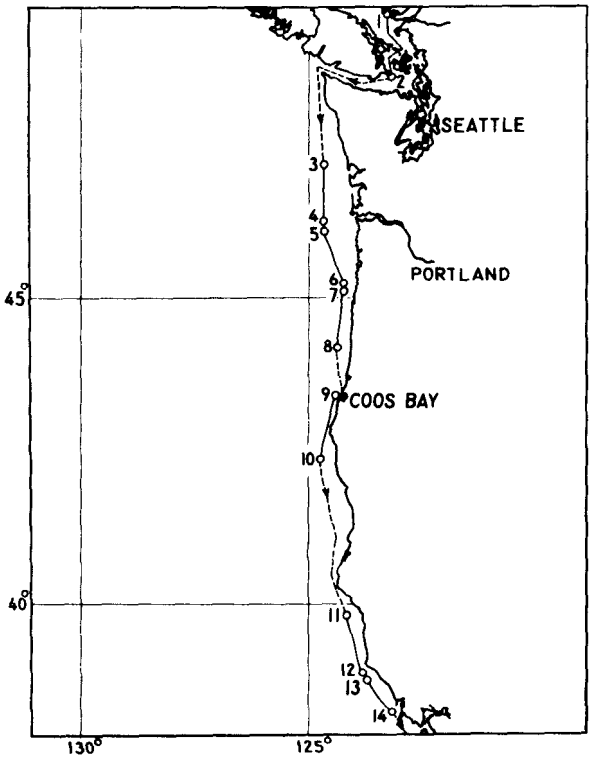

FIGURE 1.—Chart of the transects from Vancouver, British Columbia, to San Francisco Bay, Calif. See Tables 1 and 3 for station data.

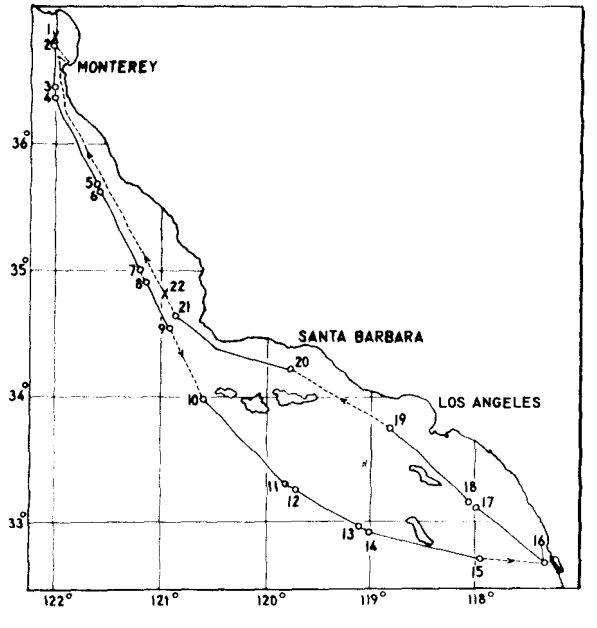

FIGURE 2.—Chart of the transects from Monterey Bay, Calif., to the southern California area. See Tables 2 and 4 for station data.

Since only one extracter was used for water extraction, the possibility existed for incomplete recovery of the DDT residues in the water passing through the device. Repeated tests of the extraction efficiency using a large carboy of oceanic seawater labelled with low levels of ^{14}C-DDT (ca. 5×10^{-12} g/ml) gave an extraction efficiency of 83% (±5%) at the flow rate settings which were used with apparatus at sea. A Variac setting of 70 was used, which produced an internal recycle rate of 900 ml/hr. The magnetic stirrer rate, which affects the degree of fracture of the solvent droplets, was kept constant.

At the end of a particular run, the contents of the centrifuge tubes were transferred to combusted 4.25 cm Whatman GFC filter papers[2] and stored in glass petri dishes at —15° C until analysis. The samples from one tube were analyzed for particulate carbon by the wet combustion method of Strickland and Parsons (1968). The samples from the other tube were analyzed for DDT residues according to previously described methods (Cox, 1970a).

At the end of a water extraction run, the flask containing about 100 ml of hexane, the water extract, was removed and stored until processing. The extract was condensed to 100 μliters after dehydration by passage through an Na_2SO_4 column. The Na_2SO_4 was specially rinsed with solvent and combusted at 350° C to remove interfering impurities normally present in the reagent salt (Lamar, Goerlitz, and Law, 1966). The condensed extract was spotted on an alumina chromatoplate, which was developed in 5% benzene in hexane, so that the solvent front moved 10 cm from the origin. Centimeter wide zones were stripped from the chromatoplates which corresponded to zones expected to contain *p,p'*-DDT, DDD, and DDE according to spots on parallel chromatograms with pure standards. ^{14}C-DDT and ^{14}C-DDE were also used to determine R_f zones. These zones

[2] The use of trade names is merely to facilitate descriptions; no endorsement is implied.

were eluted with a small amount of 20% benzene in hexane into test tubes. The eluates were analyzed individually by the same gas chromatographic techniques used for the particulate samples.

All glassware was combusted at 350° C overnight to remove interfering contaminants. All solvents were nannograde or pesticide quality. A hexane blank was run through the same procedure to detect systematic errors from any of the steps after the initial extraction. No correction was found to be necessary.

RESULTS AND DISCUSSION

WHOLE SEAWATER EXTRACTS

Comparisons of DDT residue concentrations in the particulate samples obtained by the centrifugation/filtration method described above (hereafter referred to as the particulate material) are meaningless when they purport to describe geographical differences since these concentrations change according to the density of the standing crop of the particulate material (Cox, 1970a). Comparisons of the concentrations of DDT residues in whole seawater (Tables 1 and 2) reveal some significant geographical differences. Water in the southern California region appears to have a higher DDT residue concentration. Water off Oregon and Washington has lower concentrations, and there is no evidence of high DDT residue levels adjacent to the mouth of the Columbia River. The relative uniformity of the DDT residue concentrations for this northern cruise (Table 1) suggests a diffuse source of the residues, possibly from atmospheric fallout. Direct measurements of the DDT content of dust in the atmosphere over the Atlantic Ocean (Risebrough, Hugget, Griffin, and Goldberg, 1968) and measurements of DDT residues in rainwater (Tarrant and Tatton, 1968; Yates, Holswade, and Higer, 1970) implicate aerial transport as an important mechanism of land-sea DDT residue transfer. Published calculations based on annual rainfall statistics and probable DDT residue concentrations in rainwater predict the concentration of DDT residues in the surface mixed layer of the oceans to be 5×10^{-12} g/ml (Smith, 1970). This estimate is within a factor range of 0.5 to 1.1 of the results presented in Tables 1 and 2.

Atmospheric fallout may be important in areas remote from river systems draining agricultural areas or in areas remote from waste dumping of highly populated areas. Sewage outfalls near large centers of population, such as the southern California area, contribute a large share of the DDT residue input to the ocean. When the outfall is below the pycnocline, the DDT residues in the effluent may settle with the particles comprising the solid component of the sewage and thus enter the benthic environment. This may account for the high DDT levels found in the livers of bottom dwelling fish in the southern California region, as compared to pelagic species (figures released by the California Department of Fish and Game in 1970). Sedimentation of organic particulate material from the surface layers represent an additional input to the benthos.

Input of DDT residues to the mixed layer is represented by the following sources: (1) sewage input by vertical transport of material from below the pycnocline or by direct input from shallower outfalls, (2) input from terrestrial runoff water which bears fallout particles, and (3) direct input from fallout of particles over the water. The relative importance of these

TABLE 1.—DDT residue concentrations in seawater obtained by liquid-liquid whole water extracts from transects shown in Figure 1.

Stations	Total volume extracted (liters)	DDT concentrations in water-parts per 10^{12}
1-2	2.6	2.3
3-8	4.1	2.7
9-10	2.8	2.3
11-14	4.3	2.3

TABLE 2.—DDT residue concentrations in seawater obtained by liquid-liquid whole water extracts from transects shown in Figure 2.

Stations	Total volume extracted (liters)	DDT concentrations in water-parts per 10^{12}
4-7	2.8	4.1
10-13	4.0	3.0
14-15	1.6	5.6
16-19	3.3	3.4

sources is not known, but it is quite likely that sources (1) and (2) account for the higher DDT residue concentration in the whole seawater samples taken off southern California.

PARTICULATE MATERIAL

Results of the analyses of the particulate material are shown in Tables 3 and 4. Transect 10-11 (Table 4) yielded an abnormally high value when compared to the other values for particulate material. During transect 10-11, visual observations were made of oil globules at the sea surface. The abnormally high value may have been caused by inclusion of a small globule of this material in the particulate material for transect 10-11, after entrainment in the seawater system of the vessel. This value has been deleted from further data presentations.

TABLE 3.—DDT residue concentrations in organic particulate material collected by continuous-flow centrifugation and collection of the centrifugal pellet on GFC-glass-fiber filter papers. Transects shown in Figure 1.

Transect stations	Total volume filtered (liters)	Wt. of carbon in centrifugal pellet (g × 10⁻⁶)	DDT concentration μg DDT residues/g carbon (ppm)
1-2	48	4,500	1.4
3-4	48	2,980	2.2
5-6	29	2,550	1.8
7-8	17	490	5.7
9-10	39	2,680	2.1
11-12	24	1,780	2.3
13-14	28	600	8.3

TABLE 4.—DDT residue concentrations in organic particulate material collected by continuous-flow centrifugation and collection of the centrifugal pellet on GFC-glass-fiber filter papers. Transects shown in Figure 2.

Transect stations	Total volume filtered (liters)	Wt. of carbon in centrifugal pellet (g × 10⁻⁶)	DDT concentration μg DDT residues/g carbon (ppm)
1	(1)	1725	27.0
2-3	44	4700	1.6
4-5	36	2130	2.4
6-7	37	2750	2.4
8-9	23	2140	2.5
10-11	36	3320	16.0
12-13	19	2330	1.5
14-15	32	2690	1.4
16-17	50	3280	1.6
18-19	60	3030	1.2
20-21	42	4010	1.3
22	(1)	3770	1.2

[1] These samples were obtained by using a net; see text for details.

On the May cruise to southern California (Figure 2), two phytoplankton ¼-m net tows (35-μ effective aperture) were taken at stations 1 and 22, and analyzed along with the particulate material samples. These tows consisted of 10 successive vertical hauls from 15 m to the surface at station 1 and one oblique haul from 10 m to the surface at station 22. The station 1 value is in approximate agreement with earlier published DDT residue concentrations for net phytoplankton samples (27 ppm per unit of carbon converts to 0.27 ppm wet weight; compare to values given by Cox, 1970a). This value is considerably higher than the values listed in Tables 3 and 4 for particulate material. At station 22, the ship was stopped for an investigation of a dense phytoplankton bloom, which consisted principally of *Rhizosolenia* spp. No measurements of chlorophyll were made, but the water was visibly discolored due to the high concentration of algal cells in parallel streaks at the surface. The concentration of DDT residues in net-tow material from this bloom was considerably lower than in the sample taken at station 1 (0.012 ppm wet weight compared to 0.27 ppm). This may be explained by the fact that the standing crop density was much higher at station 22 than at station 1.

The generally lower values in the particulate material compared to net-tow material (except in the case of station 22 as discussed above) could result from at least three causes: (1) loss of materials by cells bursting during the centrifugation (filtration as a cause of bursting of cells is well known, but cannot account for a difference in this case since the net-tow samples [Cox, 1970a and this report] were vacuum filtered through GFC papers as well), (2) inclusion of smaller particulate material having a lower intrinsic DDT residue concentration, or (3) exclusion from the centrifuge of larger zooplankters which would be trapped by the phytoplankton net.

Cause 1 represents one reasonable source of loss of DDT residues from the particulate material, if in fact they should have higher DDT residue concentrations than those reported herein. However, experiments with the same cen-

trifuge showed that at least 98% of the particulate chlorophyll *a* in the incurrent water is recoverable from the centrifugal pellet in whole particulate form (trappable on GFC filters). This indicates that breakage of cells must be minimal.

Cause 2 is also a possible explanation. Pfister, Dugan, and Frea (1969) pointed out that chlorinated hydrocarbons showed quantitative differences of distribution among particles greater than 0.15 μ which were separable by density gradient centrifugation. Although they found no recurrent patterns of distribution among the DDT metabolites they were able to detect, their results suggest large differences in the pesticide concentrations in the four different density classes of particles analyzed. The form in which their data are presented, however, does not allow any conclusions about lower or higher DDT residue concentrations in the material which was collected in the centrifuge, but not included in the net-tow material.

Odum, Woodwell, and Wurster (1969) found lower DDT residue concentrations associated with smaller detrital particles in a core taken from a sprayed marsh, but it is uncertain if these results may be applied to oceanic seston.

Cause 3 is a possible explanation on the basis of the mesh size of the zooplankton exclusion filter used in the centrifugation/filtration procedure (0.176 mm) compared to the one used in the processing of the net-tow material both in this report and the earlier published data (0.33 mm).

EFFECT OF STANDING CROP DENSITY

The effect of standing crop density, alluded to above, was observed in the analyses of the particulate material. Standing crop densities were calculated for the transects using estimates of the volume of water filtered during the centrifuge running time and the carbon analyses of the centrifugal pellet. The values for DDT residue concentration are plotted vs. the standing crop density in Figure 3. The slope of the regression line fitted to the data points from both cruises is approximately —1, indicating that equal amounts of DDT residues were taken up by the algal materials within a given volume of water over the range of standing crop densities encountered. This is essentially the same conclusion mentioned earlier (Cox, 1970a).

PARTICULATE MATERIAL AS A PART OF WHOLE SEAWATER

Data points from the Vancouver to San Francisco cruise seem to fit the empirical linear relationship detailed in Figure 3 much more closely ($r = -0.99$) than the data points from the Monterey Bay to southern California cruise ($r = -0.54$). This variability is undoubtedly due to the greater variability of the DDT residue concentrations of the whole seawater from the southern California region, where most of the samples were taken. There would be no need to impute causal relationships between the whole seawater concentration and the concentration of DDT residues in the particulate material, if the particulate material represented a major portion

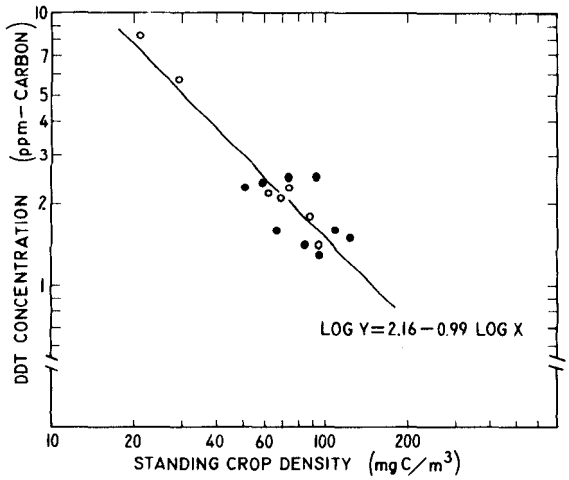

FIGURE 3.—DDT concentrations in the particulate samples as a function of particulate carbon standing crop density. Stations 1 and 22 (Table 4) are not included, since the density of the standing crops could not be computed because there were no measurements of the volume of water filtered in these net-tow samples. Also, for reasons outlined in the text, they may not be comparable to the samples collected by the centrifuge. Transect station 10-11 (Table 4) was omitted because of the possible interference of oil, as described in the text. The remaining 16 values from Tables 3 and 4 appear in this figure. Open circles refer to data from Table 3; solid circles refer to data from Table 4.

of the DDT residues in whole seawater. In fact, the particulate material accounted for less than 10% of the DDT residues in the corresponding whole water extracts (range: 1.8% to 9.9%). Unless the remaining amount of DDT residues (<90% of the total present) is in soluble form, it must be fixed to particles not collected in the centrifugation/filtration procedure. Typical natural distributions of particulate matter in seawater (Bader, 1970; Beardsley, Pak, and Carder, 1970) suggest that most of the particulate volume and almost all of the particulate surface area is accounted for by particles of less than 2 μ in diameter. Thus it is quite likely that the balance of the DDT residues in whole seawater are fixed to these smaller particles, in view of the hydrophobicity and affinity for interfaces characteristic of the different metabolites of DDT. The possibility also exists that it may occur as micelles or aggregates which cannot be taken up by the particulate matter.

EXPERIMENTAL EVIDENCE

Two experiments were performed to examine the distribution of DDT residues between seawater and phytoplankton. In both experiments, ^{14}C-DDT in a 1-ml ethanol carrier was added to GFC filtered oceanic seawater in a 4-liter glass carboy which was stirred by a magnetic stirrer. Repeated subsamples of 25 ml each were taken from the system until successive samples gave a constant ^{14}C activity. All counts were made on a Nuclear-Chicago Unilux II scintillation counter.

Aliquots of a dense suspension of *Dunaliella salina* culture were added to the carboy from a large separatory funnel with a 25-ml dispensing chamber, via a tube connected to the carboy. Sampled and added amounts were such that a constant volume was maintained. After addition of an aliquot of culture, one or two aliquots of 25 ml each were removed from a tap at the bottom of the carboy. This amount was vacuum filtered onto a GFC-glass-fiber filter paper, and counts of ^{14}C-DDT were made of the filter and of a petroleum ether extract of the filtrate. Cumulative ^{14}C activity in the filter and filtrate equalled amounts present in the 25 ml aliquots (both filter and filtrate) before addition of the algal suspension, when the net amounts of ^{14}C-DDT removed from the system by sampling were taken into account. A correction was made for adsorption or possible trapping of small particles of ^{14}C-DDT on the filter. The correction factor, expressed as percent of total activity per 25-ml aliquot which was on the filter before addition of the algal suspension, was constant in the five replicates taken just before the algal cells were added. This correction factor may have changed during the course of addition of the algal cells, but the techniques used did not allow a distinction between ^{14}C activity on the filter which adsorbed, associated with trapped small particles, or associated with the algal cells themselves. I believe that this change was small and did not materially affect the outcome of the experiments.

Figure 4 shows the results of the two experiments. In Experiment 1, the seawater used in the carboy was not altered; in Experiment 2, the seawater was specially prepared to increase the load of small (<1-2 μ) inorganic particles, to see what effect this might have on the uptake function. Nuchar-attaclay, a mixture of finely divided charcoal and clay particles (attapulgite), was added to 2 liters of GFC-filtered seawater. After shaking, the mixture was refiltered through a GFC filter. It is estimated that only a tiny fraction of the initially added Nuchar-attaclay (initially added amount was 0.1 g) actually got through the filter. The 2 liters of water produced in this way were mixed with another 2 liters of GFC-filtered seawater and put into the carboy. Two other conditions were different in Experiment 2. The culture of *Dunaliella salina* used was denser (note that the arrow in Figure 4 indicates that 750 μg C/liter is reached at a lower volume of culture added). The initial concentration of ^{14}C-DDT in Experiment 2 was approximately 15 ppt.

The first part of the uptake functions in Experiments 1 and 2 appeared to be linear, indicating that under the conditions prevailing at the beginning of each experiment, each *Dunaliella* cell took up a constant amount of the ^{14}C-

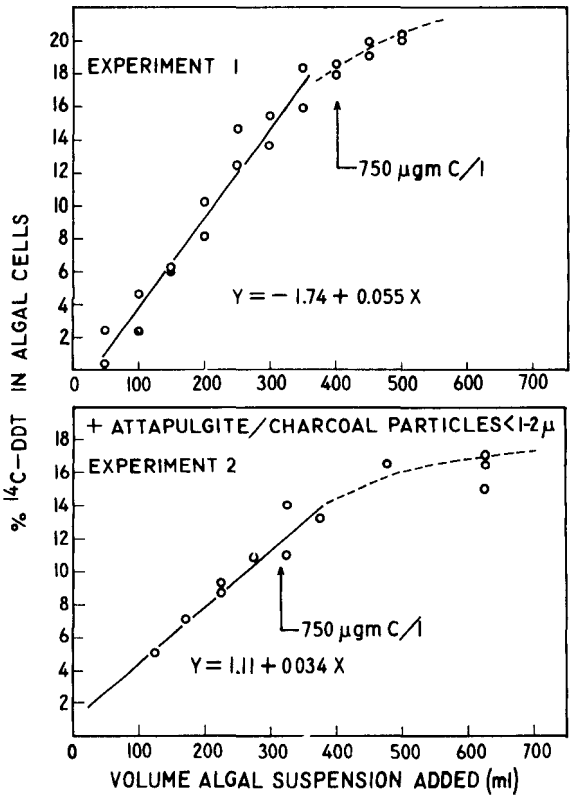

FIGURE 4.—Percentage of total ^{14}C-DDT in sample aliquots recovered on GFC filters, plotted as a function of total volume of *Dunaliella salina* culture added to a constant volume system. See text for detailed discussion.

DDT which was available. Both curves show inflection points after the density of cells increases beyond 750 μg C/1. The fact that the uptake per cell was constant over the linear range indicates that each cell has a saturation value for uptake of DDT, which is independent of the ambient concentration of DDT available. The curves presumably begin to level off when the ^{14}C-DDT which was available for uptake is mostly associated with the algal mass already added.

If algal cells exhibit a saturation value for uptake of DDT, then adsorption of DDT to the cell surface is a more likely explanation for DDT uptake than phase partitioning of DDT between seawater and the lipid component of the algal cell, as has been previously hypothesized (Cox, 1970b). Each *Dunaliella salina* cell probably had a total cell surface area of 240 μ^2 (Mullin, Sloan, and Eppley, 1966). The cells in Experiments 1 and 2 took up a mean of 5×10^{-5} picograms ^{14}C-DDT/μ^2. This value may be near the asymptotic saturation value for *Dunaliella salina* for the experimental conditions described above. The validity of a saturation value of this kind needs to be tested with other phytoplankton species over a wide range of ambient DDT concentrations.

A quantitative solution to simultaneous Freundlich adsorption equations for the algal cells and the <1-2 μ particles could explain the uptake curves if the adsorption energy coefficient were known in each case. Studies such as those of Weber and Gould (1966) should therefore be applied to uptake of DDT residues by phytoplankton and smaller particles to elucidate the relationships discussed here.

No measurements were made of the concentration of the <1-2 μ particles in the untreated seawater of Experiment 1 or the treated seawater of Experiment 2. Thus the differences can only be explained qualitatively. The higher concentration of ^{14}C-DDT in Experiment 2 (30 ppt) was apparently reflected in the uptake of ^{14}C-DDT per unit of cell surface area; Experiment 2 yielded a value of about 6×10^{-5} picograms ^{14}C-DDT/μ^2, which is higher than the mean for both experiments quoted above. The uptake of ^{14}C-DDT per unit of cell surface area in the case of Experiment 2 is probably closer to the asymptotic saturation value because of the higher concentration of ^{14}C-DDT in the medium. The main difference between the curves for Experiments 1 and 2 is the position of the inflection point. Experiment 2 shows an apparent inflection point which is lower than the apparent inflection point of Experiment 1, indicating a lowering of the available percentage of total ^{14}C-DDT in the system. The total small particle concentration of the system was not measured, so this apparent change must be regarded as a presumptive effect of the Nuchar-attaclay addition.

If a large percentage of the DDT added to aqueous systems is fixed to a particle fraction

less than 1-2 μ in diameter, then the experimental DDT uptake results that have been interpreted in terms of a partition coefficient or a concentration factor using the nominal concentration of DDT in the aqueous medium become relatively meaningless without knowledge of the fraction of the initial amount of DDT present which is fixed to these small particles and hence unavailable to the test organism. In the oceanic environment, it is quite likely that the amounts of "available" DDT residues are exceedingly small. Uptake of DDT by the seston is probably closely coupled with input pulses which would be largely determined by fallout conditions at the surface and seasonal runoff of DDT residues from land areas. "Available" DDT residues which may rise during these periods will be taken up by plankton. A complete picture of the processes involved in DDT transport to the pelagic environment has yet to be drawn and will require further experimental and analytical work.

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