

# Effects of Salinity on Uptake of DDT, DDE and DDD by Fish

by PHILIP G. MURPHY

*Hopkins Marine Station, Stanford University  
Pacific Grove, California*

DDE constitutes a smaller proportion of the DDT residues (DDT, DDD and DDE) in fresh water fish than in marine fish (1, 2, 3, 4), and marine fish from estuaries can have DDT residues with 24 to 35% DDE compared with 61 to 73% DDE for the same species from coastal waters (4). Although a greater conversion of DDT to DDE in the marine environment has been postulated to account for these differences, this study was undertaken to assess the effect of salinity on the uptake of DDT residues by fish from pesticide concentrations similar to those of environmental waters. Mosquito fish (*Gambusia affinis*) were used because of their tolerance of salinity and convenient size.

## MATERIAL AND METHODS

Young, unsexed, pond-raised fish were exposed to the experimental salinities for 24 hours before use. Prior to the experiments, the DDT residues in six fish were extracted (5) and estimated by gas liquid chromatography to be 0.34 parts per million (ppm) DDE with a standard deviation of  $\pm 0.073$  ppm,  $0.088 \pm 0.024$  ppm DDD and  $0.080 \pm 0.028$  ppm DDT.

Ring labelled p,p'-DDT-C<sup>14</sup> with a specific activity of 19.1 millicuries per millimole was obtained from Amersham/Searle Corporation, Des Plaines, Illinois. DDE was prepared from the DDT-C<sup>14</sup> by the method of Gunther and Blinn (6). DDD was prepared by a modification of the method of Singh and Malaiyandi (7). 300 micrograms of DDT-C<sup>14</sup> dissolved in 1 ml of 30% 1-4 dioxane in water was heated in a sealed glass ampule for 24 hours at 100°C. The products were partitioned into hexane, concentrated to 5 ml and chromatographed on an 11 by 1.2 cm diameter floridil column using hexane as an eluent. The 60 to 150 ml fraction contained only DDD. The purity (>99%) of the pesticide was ascertained by gas chromatography.

Artificial sea salts were dissolved in distilled water to give salinities of 0.15, 10 and 15 parts per thousand and the solutions passed through 0.45 micron Millipore filters immediately before use.

## EXPERIMENTAL PROCEDURES

Boiling flasks of 12 liters capacity, each containing 8 liters of water, served as aquaria. 200 microliters of ethanol solution of DDT, DDE or DDD were pipetted into each aquarium and resulted in concentrations of 41 parts per trillion (ppt) for DDT, 32 ppt for DDE and 55 ppt for DDD. The fish were added 5 min after the pesticide and the aquaria mouths were stoppered to prevent evaporation. The experiments were conducted at room temperature; slight stirring was provided by magnetic stirrers, and the fish were not fed for the duration of the experiments.

At the end of an experiment the fish were each weighed to the nearest milligram, dissolved in NCS solubilizer (Amersham/Searle Corporation) and the radio-activity measured by liquid scintillation counting using the channels ratio method of quench correction. The amount of residue taken up per gram of fish was divided by the amount available per gram of fish and expressed as percent uptake.

## RESULTS

The results of these experiments are presented in Table I. It is evident that salinity of 15 parts per thousand reduced the amount of DDT, DDE and DDD accumulated by the fish, and that salinity had a greater effect on the uptake of DDT than on the uptake of DDE or DDD.

## DISCUSSION

The gills are the primary route of DDT uptake by fish from water (8). Salinity could conceivably affect residue uptake by changing the water metabolism since both the amount and the pathways of water and salt metabolism are determined by the salinity of a fish's environment (9), or salinity could change the absorption and/or the water-lipid partition characteristics of residue molecules in aqueous solution.

Regardless of the mechanism, the differential effects of salinity on the uptake of DDT residues found in these experiments would result in a larger proportion of DDE in the residues in fish from brackish water than in fish from fresh water, even though both environments contained identical proportions of residues. However, in order to determine the extent to which salinity is responsible for the proportions of residues mentioned in the introduction,

TABLE I

DDT, DDE and DDD Taken up from Waters of Different Salinity by Gambusia affinis

SALINITY (parts per 1000)	PERIOD OF EXPOSURE (hr)	NO. FISH	FISH WEIGHT (mg)	TOTAL PESTICIDE* (ng)	% UPTAKE	p**
0.15	24	20***	101±27	658±3	22.2±3.1	-----
10.0	24	10	104±24	329±1.5 (4)	17.7±3.3	0.003
15.0	24	11	98±22	" "	16.6±2.0	<0.001
0.15	84	10	88±29	" "	37.1±6.7	-----
15.0	84	10	97±35	" "	21.6±4.3	<0.001
<hr/>						
0.15	24	10	120±33	253±3 (3)	20.2±4.6	-----
15.0	24	10	116±35	" "	19.0±4.1	0.573
0.15	84	10	151±36	" "	51.0±8.6	-----
15.0	84	10	149±39	" "	42.3±6.1	0.015
<hr/>						
0.15	24	11	120±39	439±3 (3)	19.2±2.3	-----
15.0	24	11	130±43	" "	20.2±5.1	0.603
0.15	48	9	81±24	" "	27.9±5.2	-----
15.0	48	10	96±21	" "	21.9±3.9	0.017
0.15	84	10	89±15	" "	38.5±6.7	-----
15.0	84	10	97±23	" "	32.8±4.9	0.060

\* Mean, standard deviation and number of observations in parentheses.  
ng denotes 10<sup>-9</sup> grams.

\*\* Probability that the observed distribution occurred by chance.

\*\*\* The 20 fish were exposed in groups of 10 to 329 ng DDT.

additional information is needed concerning the effects of salinity on DDT residue uptake by aquatic plants and invertebrates as well as the effects on uptake from food by fish.

#### ACKNOWLEDGEMENT

The author thanks the Salinas Valley Mosquito Abatement District, Salinas, California for the fish used in this study.

#### LITERATURE CITED

1. C. L. HOPKINS, S. R. B. SOLLY and A. R. RITCHIE, New Zealand J. Mar. Freshwat. Res. 3, 220-229 (1969)
2. J. O. KEITH and E. G. HUNT, Transactions of the Thirty-First North American Wildlife and Natural Resources Conference, Wildlife Management Institute, pp. 150-177 (1966)
3. M. W. ODEMAR, P. W. WILD and K. C. WILSON, California Dept. Fish and Game, MRO Ref. No. 68-12 (1968)
4. R. W. RISEBROUGH, pp. 5-23. IN M. W. Miller and G. G. Berg, (ed.), Chemical Fallout, Charles C. Thomas, Springfield, Illinois (1969)
5. A. M. KADOUM, Bull. Environ. Contam. Toxicol. 2, 264-273 (1967)
6. F. A. GUNTHER and R. C. BLINN, J. Chem. Education 27, 654-658 (1950)
7. J. SINGH and M. MALAIYANDI, Bull. Environ. Contam. Toxicol. 4, 337-342 (1969)
8. A. V. HOLDEN, Ann. Appl. Biol. 50, 467-477 (1962)
9. C. L. PROSSER and F. A. BROWN, Jr., Comparative Animal Physiology. W. G. Saunders Company, Philadelphia (1961)