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TRITIUM IN THE AQUATIC ENVIRONMENT<sup>1,2</sup>

B. G. Blaylock, F. O. Hoffman, and M. L. Frank

**MASTER**

ENVIRONMENTAL SCIENCES DIVISION  
Oak Ridge National Laboratory  
Oak Ridge, Tennessee 37831

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## ABSTRACT

Tritium is of environmental importance because it is released from nuclear facilities in relatively large quantities and because it has a half life of 12.26 y. Most of the tritium released into the atmosphere eventually reaches the aqueous environment, where it is rapidly taken up by aquatic organisms. This paper reviews the current literature on tritium in the aquatic environment. Conclusions from the review, which covered studies of algae, aquatic macrophytes, invertebrates, fish, and the food chain, were that aquatic organisms incorporate tritium into their tissue-free water very rapidly and reach concentrations near those of the external medium. The rate at which tritium from tritiated water is incorporated into the organic matter of cells is slower than the rate of its incorporation into the tissue-free water. If organisms consume tritiated food, incorporation of tritium into the organic matter is faster, and a higher tritium concentration is reached than when the organisms are exposed to only tritiated water alone. Incorporation of tritium bound to molecules into the organic matter depends on the chemical form of the "carrier" molecule. No evidence was found that biomagnification of tritium occurs at higher trophic levels. Radiation doses from tritium releases to large populations of humans will most likely come from the consumption of contaminated water rather than contaminated aquatic food products.

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### INTRODUCTION

Tritium is released from nuclear facilities in relatively large quantities; however, because it is a weak beta emitter ( $E_{\max} = 18 \text{ keV}$ ), tritium is considered one of the least hazardous of the radioisotopes. Nevertheless, there has been a continuing interest in the biological effects of tritium and in its transport in the environment. This interest is stimulated by tritium's physical half-life (12.26 y), its continued release from existing nuclear facilities, and the potential of even greater releases from thermonuclear reactors.

Tritium is of interest ecologically because it is rapidly transported through the environment and is rapidly taken up by organisms.<sup>(1)</sup> Most of the tritium that is taken up by organisms is lost just as rapidly; however, a fraction of the tritium that is bound in tissues may be incorporated into macromolecules, such as proteins and DNA, and turned over at a slower rate. While external exposure to tritium is not a major concern, tritium incorporated into the genetic material of organisms is of concern because of its ability to produce chromosome breaks and mutations.

For radioecologists, the problems associated with tritium are the uptake, concentration, and elimination of tritium in organisms, and its transport through the food chain. Tritium from nuclear facilities is released into the environment mainly as tritiated water (HTO). Most of

the tritium which is released into the atmosphere ends up in the hydrosphere. Tritiated water dispersed in rivers or oceans is rapidly taken up by biota. In many cases equilibrium conditions are reached in only a few minutes. Eventually most of the tritium entering the environment finally reaches the aqueous environment. (1)

The tissue-free-water tritium (TFWT) in aquatic organisms equilibrates rapidly with the surrounding medium, usually reaching a concentration factor ( $^3\text{H}$  concentration in the organism  $\div$   $^3\text{H}$  concentration in water) of from 0.8 to 1.0. The tissue-bound tritium (TBT) accumulates more slowly than the TFWT and reaches a concentration of about 0.5 in most organisms when the surrounding medium is HTO. (2) Organisms that are fed tritiated food, accumulate tritium more rapidly and to a higher level in the TBT than when exposed to HTO alone. It has also been established that tritiated precursors, such as tritiated thymidine, will accumulate rapidly in the DNA of organisms. Apparently, tritium ingested as complex molecules such as amino acids will accumulate more rapidly and occupy more sites in the TBT than will occur when the intake of tritium is in the form of HTO.

The ecological question related to tritium has been whether organisms at the lower trophic levels that accumulate TBT from HTO will pass the tritium on to higher trophic levels in the form of macromolecules that will be biomagnified at these higher trophic levels. An accompanying question of equal importance is at what level will biomagnification occur in the trophic levels in natural ecosystems. The purpose of this paper is to review the literature on

aquatic ecosystems that address these questions. Because of the many existing reviews<sup>(2,3,4,5,6)</sup> on tritium in the environment, this review will not be comprehensive but will emphasize the more recent literature.

## ALGAE

The kinetics of tritium uptake and release by various species of algae has probably been more intensely studied than for any other group of organisms. In aquatic environments algae are the primary producers of organic matter. Thus, one of the major concerns is that tritium will be concentrated in algae and passed along the aquatic food chain leading to man. Studies on algae can be divided into three categories: (1) the uptake and elimination of tritium from cells cultured in HTO, (2) the uptake and elimination of organically bound tritium in cells cultured in HTO, and (3) the incorporation of tritiated precursors into the organic matter of algae.

### Uptake of tritium by algae from tritiated water

The tissue-free water in an alga cell equilibrates within minutes with the external HTO. Several studies using a variety of marine and freshwater species have documented the kinetics of the uptake and elimination of tritium.<sup>(7,8,9,10,11)</sup> Typical uptake and elimination curves for algae cultured in HTO are shown in Figure 1. The uptake curve is divided into two components, and the elimination curve into three. The uptake times for two multicellular marine species and a giant unicellular alga are compared in Table 1. Almost

half of the tritium is taken up by these species in less than 0.5 min., while most of the remainder is taken up in from 1.6 to 22 min. The elimination rates resemble the uptake components, with the exception of the third component, which has a half-life of up to 500 min for about 1% of the tritium.

The incorporation of tritium into organic matter in cells cultured in HTO has been investigated by a number of researchers.<sup>(9,11,12,13)</sup> Incorporation of tritium from HTO into the total organic matter of algae is expected to occur mainly by photosynthesis, which causes the splitting of the tritiated molecule.<sup>(9)</sup> Incorporation of tritium into the organic matter of the marine alga Acetabularia showed a rapid phase (half-life 2.3 d), followed by a slower one (half-life of about 50 d), during the exponential growth phase of a culture; however, when the algae reached the stationary phase of growth, organic tritium was replaced very slowly with a half-life of 150 d.<sup>(9)</sup> Figure 2 shows a typical uptake curve for the incorporation of tritium into the organic matter of algae.

The specific activity of the total organic matter of the algae depends upon the external tritium concentration.<sup>(9)</sup> The marine alga Acetabularia attained a specific activity in its organic matter of about 6% of the external medium; by comparison, the freshwater alga Chlamydomonas reached a specific activity which was about five times higher for the same concentration of HTO.

Planktonic algae are known to utilize dissolved organic compounds present in the external medium. Tritium bound in organic compounds

behaves differently from that in HTO. The fate of organically bound tritium will depend largely upon the chemical form of the "carrier" molecule.<sup>(12)</sup> Uptake studies with the marine alga Dunaliella bioculata showed that all ten of the tritiated precursors studied were taken up by the alga.<sup>(12)</sup> The intracellular concentration of most of the tritiated precursors may reach the concentrations of the external medium; however, leucine and adenine accumulated in the algae to respective concentrations of 10 and 100 times the external medium. The concentration ratios for the intracellular incorporation of tritium are shown in Table 2. In a similar experiment<sup>(13)</sup> Acetabularia mediterranea selectively accumulated adenine [concentration ratio (CR) of 4.6] and D-glucose-6-<sup>3</sup>H (CR 5.8), but not leucine (CR 0.3). Adenine was accumulated by both algae and was present primarily in the nucleic acid fraction of the organic matter. Thus the fate of organically bound tritium depends largely upon the chemical form of the carrier molecule.

#### AQUATIC MACROPHYTES

Compared with the amount of data available on the uptake of tritium by algae, very little information is available on its uptake by aquatic macrophytes. Harrison and Koranda<sup>(14)</sup> found that the TFWT of the cattail (Typha angustifolia) that had grown for 230 d in a pool containing HTO never equilibrated with the medium. Similar results were obtained for Typha latifolia growing in a chronically contaminated lake<sup>(15)</sup>; however, the tissue free water of the submergent pond weed (Potamogeton foliosus) was at equilibrium with the lake water. For the

emergent cattails, which had their roots in HTO, this phenomenon is similar to that of terrestrial plants grown in a tritiated nutrient solution in which the lack of equilibration was attributed to the atmospheric exchange of water by the leaves.

#### AQUATIC INVERTEBRATES

Tritium concentration factors and biological half-lives for a number of marine invertebrates are summarized by Kirchmann et al.<sup>(2)</sup> Concentrations and turnover times for tritium in freshwater invertebrates have been determined in several studies.<sup>(14,15,16)</sup> Much of the information on invertebrates has been obtained either from studies using large fiberglass pools in which aquatic communities were maintained in HTO for over 200 d or from natural populations from contaminated lakes.<sup>(14,15)</sup>

Recently, investigators have examined the transfer of tritium from one feeding of HTO-labeled algae to the mussel (Mytilus edulis) and one feeding of <sup>3</sup>H-leucine-labeled algae to the mussel (Dreissena polymorpha).<sup>8,10,17</sup> In both cases the highest activity was found in the hepatopancreas and gills. The mussels that received algae labeled with HTO lost 90% of their activity after 16 d. Biological half-lives were determined for the shell and soft tissue of the mussels receiving <sup>3</sup>H-leucine-labeled algae. Loss of tritium by the shell showed two components with respective half-lives of 0.6 and 41 d. The soft tissues showed a rapid decrease in tritium for the first day followed by a slower long-component loss (T<sub>b</sub> 1/2) of 36 d.<sup>(17)</sup> The longer half-life of the tritium in the mussels that received the



<sup>3</sup>H-leucine-labeled algae was apparently the result of a higher concentration of TBT in these mussels than in those that received HTO-labeled algae.

Figure 3 shows the uptake of tritium in the saltwater clam (Mya arenaria), cultured in HTO in a fiberglass pool for more than 200 d.<sup>(14)</sup> Not only were these clams exposed to HTO, but also all their food originated in the pool containing HTO. The gills and viscera contained the highest concentrations of tritium, attaining specific activities of 6.5 and 7.0, respectively, while the specific activity of the TFWT approached that of the pool water. A biological half-life of 190 d was calculated for the shell (long component), 150 d for gills, and 110 d for viscera. Mussels living in an environment contaminated with tritium accumulated tritium from both HTO and food and, over a period of 200 d, reached an equilibrium concentration related to the concentration of tritium in the water.

## FISH

One of the principal pathways for contaminants transfer through the aquatic food chain to man is by the consumption of fish. For this reason, a number of studies<sup>(18,19,20,21)</sup> have involved the uptake, concentration, and elimination of tritium in fish. Other studies<sup>(14,15,16,22)</sup> have examined the concentration of tritium in natural and artificial ecosystems. These latter studies will be reviewed in the section on aquatic food chains. Channel catfish (Ictalurus punctatus) and bluegill (Lepomis macrochirus) were used to examine the rapid uptake and elimination of tritium from HTO.<sup>(19)</sup>

Tritium was taken up more rapidly by catfish than by bluegill. Within 30 min catfish had reached one-half their equilibrium concentration; it took bluegill 90 min to reach the same level. After 20 d of exposure to HTO, catfish lost one-half of their body burden in 30 min. Elwood<sup>21</sup> used enclosures in a chronically contaminated lake to expose goldfish (Carassius auratus) and bluegill to tritium for 36 d. The whole-body and body-water concentration factor (tritium concentration in fish/tritium concentration in water) ranged from 0.6 to 1.0. Body-water tritium was eliminated from goldfish at two exponential rates, with biological half-lives of 0.2 and 0.9 h. Tissue bound tritium was eliminated at a single exponential rate, with a biological half-life of 8.7 d. Patzer et al.<sup>20</sup> compared the concentration of tritium in fish hatched from eggs in HTO with that in fish that spent an entire generation in HTO. Channel catfish and rainbow trout (Salmo gairdnerii) eggs were cultured in HTO. After hatching, the fish were maintained in HTO and fed nontritiated food for 140 d. In a related experiment, the live-bearing mosquito fish (Gambusia affinis) and the Gila top minnow (Poeciliopsis occidentalis) were placed in outdoor plastic pools containing HTO in which an aquatic community had developed. Offspring from these fish were placed in similar pools and maintained for 200 d. Not only did these fish incorporate tritium from the HTO but also they obtained most of their food from the aquatic community that had developed in the pool. The ratio between the specific activity of the TBT and that of water was less than unity in all of these experiments. The concentration of tritium in fish that

were eating tritiated food was 50 to 90% higher than that in fish being fed nontritiated food. The biological half-life for tritium in the mosquitofish was about 5 d. Figure 4 shows the rapid uptake of tritium by the mosquitofish from HTO into the TFWT and TBT.

#### FOOD CHAIN STUDIES

In some of the early food chain studies<sup>(18)</sup> Daphnia galeata, snails (Lymnaea reflexa and Helisoma trivolvis), and tree frog larvae (Pseudacris triseriata) cultured in HTO and fed tritiated food equilibrated rapidly with the environmental water. The tritium content of the tissue-bound water was related to the concentration of the tritium in the assimilated food. In another study<sup>(22)</sup> tadpoles (Rana pipiens) and snails (Lymnaea reflexa and Helisoma trivolvis) were maintained in 15-L microcosms for up to 9 months. Rates of incorporation of tritium into tissues as well as the rates of uptake and elimination of tritiated tissue-free water were determined, and rate constants were derived to describe the dynamics of tritium movement.

Harrison and Koranda<sup>(14)</sup> introduced clams (Anodonta nuttalliana), crayfish (Astacus sp.), goldfish (Carassius auratus), cattails (Typha angustifolia), and filamentous algae (Pithophora sp.) into an 8 m<sup>3</sup> freshwater pool containing HTO. They observed the transfer of tritium to the TFWT and TBT in the organisms over a period of 240 d. The TFWT of the biota increased rapidly to 95% of the level in the environmental water and remained at that level throughout the experiment. The concentration of organically bound tritium was higher in the

photosynthetic organisms than in the animals and was higher in algae than in the cattails. In the animals, the organically bound tritium concentrations were highest in the visceral organs. The specific activity of the TBT increased to a maximum of about 60% of that of the TFWT.

Using experimental conditions similar to those described previously for a freshwater system<sup>(14)</sup>, Harrison et al.<sup>(16)</sup> investigated the transfer of tritium from water in a marine system consisting of clams (Mya arenaria), crabs (Cancer productus), and fishes (Gillichthys mirabilis). The food for the animals was the algae and detritus present in the water. Tritium concentrations in various organs of the different organisms were determined and compared with model predictions. The concentration of tritium in clams, crabs, and fishes was similar to that in freshwater systems. Biomagnification through the food chain was not indicated.

The transfer of tritium through an experimental aquatic food chain was investigated by Arapis et al.<sup>(8)</sup> The microalga (Chlamydomonas reinhardtii) were labeled with either HTO or tritiated leucine. Labeled algae were then fed to the marine mussel (Mytilus edulis) and fresh water mussels (Anodonta sp., Unio sp, and Dreissena polymorpha) as a single meal. The animals were then transferred to a fresh culture medium that was renewed periodically. Most of the tritium was present initially in the hepatopancreas of the mussels and was gradually transferred to other organs. Approximately 70% of the tritium was lost to culture water, and there was no biomagnification of tritium in the

simple aquatic food chain. Similar results were obtained by Arapis et al.<sup>(10)</sup> In another experiment when labeled Chlamydomonas cells were given to Mytilus edulis, most of the radioactivity was found initially in the hepatopancreas. Investigations<sup>(17)</sup> of extracts from the hepatopancreas suggested the presence of tritium in proteins and other macromolecules, as well as in compounds having low molecular weights.

Komatsu<sup>(23)</sup> used a model ecosystem consisting of diatoms (Chaetoceros gracilis), bacteria (Escherichia coli), brine shrimp (Artemia salina), Daphnia, and Japanese killifish (Oryzias latipes) to investigate the transfer of tritium in an aquatic food chain. Daphnia, brine shrimp, and killifish were cultured in HTO or under conditions where they were fed tritiated food. The tritium level was raised when the organisms were fed tritiated food, and the organisms retained the tritium longer. No specific site of tritium accumulation was observed among the various organs, in the DNA or in the whole body; also, no evidence of biological magnification in the model ecosystem was observed.

#### CONSIDERATIONS FOR HUMAN EXPOSURE TO TRITIUM

Although the consumption of aquatic food products from marine environments may be a viable exposure pathway to individual subgroups of the human population, collective doses to large population groups will most likely be influenced by the direct consumption of contaminated drinking water and the cycling of tritium from the marine environment to terrestrial and freshwater systems via the atmosphere.<sup>(24)</sup> The

cycling of tritium from the marine environment will be controlled by evaporation and diffusion from seawater and the subsequent accumulation of HTO in rain.<sup>(25)</sup> Within a few years after a single release of tritium to the atmosphere at the 30-50° N latitude band, the concentration of tritium in human tissues should approximate tritium concentrations in the atmosphere, surface-soil water and ocean's surface.<sup>(6)</sup>

## CONCLUSIONS

Most of the tritium released from nuclear facilities eventually enters aquatic ecosystems.

The TFWT in aquatic organisms equilibrates rapidly with the surrounding medium. Algae equilibrate in a matter of minutes, while 1 or 2 h may be required for the TFWT of fish to reach levels near the concentration of tritium in the water.

Organically bound tritium is accumulated in aquatic organisms at a much slower rate and does not attain concentrations as high as can TFWT.

Algae in the exponential growth phase will accumulate organically bound tritium more rapidly than will algae in the stationary phase.

Organisms that consume tritiated food accumulate organically bound tritium faster and attain a higher tritium concentration than do organisms exposed to only HTO.

Tritium bound in organic compounds behaves differently from that in HTO. The fate of the organically bound tritium depends upon the chemical form of the carrier molecule.

Neither experimental food chain studies or data from contaminated environments indicate that tritium is biomagnified through the food chains.

Collective doses from tritium releases to large populations will most likely come from the direct consumption of contaminated water instead of through the consumption of aquatic food products.



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## FIGURE LEGENDS

- Figure 1. (a) Uptake of tritiated water by *Laurencia obtusa* showing two components for uptake with half lives of 0.4 and 22 min. (b) Release of tritiated water from *Laurencia obtusa* showing three components with half lives of 0.2, 3.9, and  $\approx 75$  min (after Cinelli et al. 1983).
- Figure 2. Incorporation of tritium into the organic matter of the alga *Acetabularia* (after Bonotto et al. 1982).
- Figure 3. Uptake of tissue free water tritium and tissue bound tritium by the clam (*Mya arenaria*) exposed to tritiated water in a fiberglass pool containing an aquatic community exposed to tritiated water.
- Figure 4. Uptake of tissue water tritium and tissue bound tritium by the mosquitofish (*Gambusia affinis*) exposed to tritiated water.

## COMPARISON OF UPTAKE OF TRITIATED WATER BY ALGAE

SPECIES	FIRST COMPONENT		SECOND COMPONENT	
	%	T1/2 min	%	T1/2 min
ACETABULARIA ACETABULUM	60	0.065	35	1.75
CYSTOSEIRA COMPRESSA	46	0.22	24	3.31
LAURENCIA OBTUSA	40	0.40	19	22

AFTER ARAPIS et. al. 1983 and  
CINELLI et. al. 1983

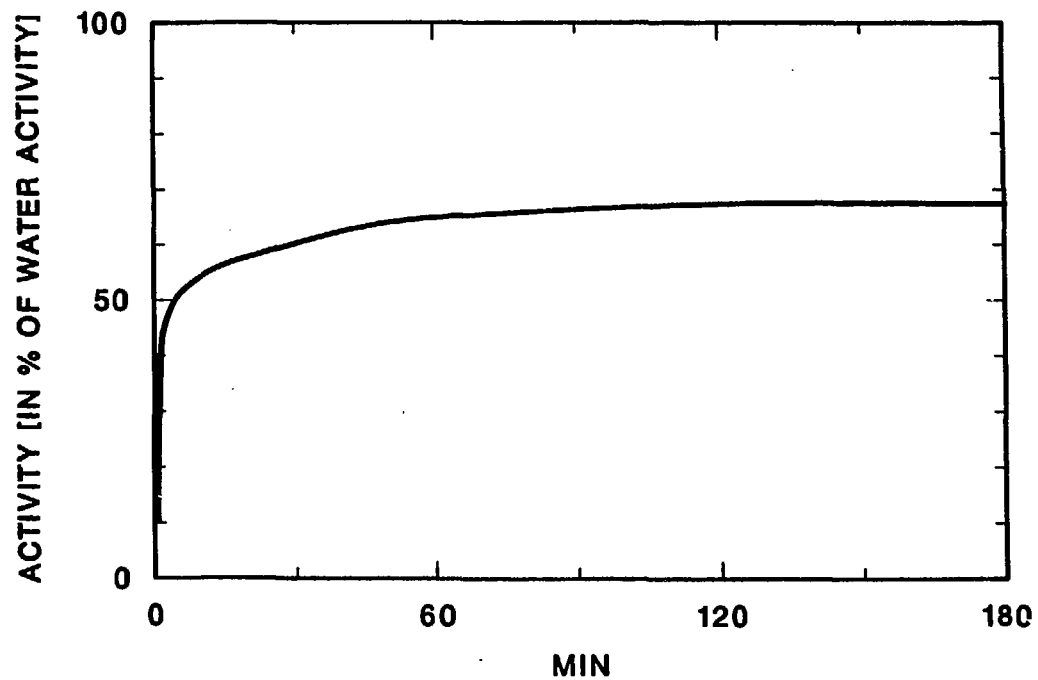
**UPTAKE OF VARIOUS TRITIATED ORGANIC  
COMPOUNDS BY *DUNALIELLA BIOCULATA*  
AND *ACETABULARIA MEDITERRANEA***

	<i>DUNALIELLA BIOCULATA</i>	<i>ACETABULARIA MEDITERRANEA</i>
	CONCENTRATION RATIO	CONCENTRATION RATIO
THYMIDINE-METHYL- <sup>3</sup> H	0.8	1.8
ADENINE-2- <sup>3</sup> H	122.7	4.6
URIDINE-5- <sup>3</sup> H	2.0	0.1
L-LEUCINE-4- <sup>3</sup> H	11.4	0.3
GLYCINE-2- <sup>3</sup> H	1.1	0.1
L-ARGININE 3.4- <sup>3</sup> H	0.6	5.1
L-ASPARTIC ACID-2.3- <sup>3</sup> H	1.0	2.3
L-PHENYLALANINE-2.3- <sup>3</sup> H	0.5	2.6
D-GLUCOSE-1- <sup>3</sup> H	0.4	5.7
D-GLUCOSE-6- <sup>3</sup> H	0.9	2.7

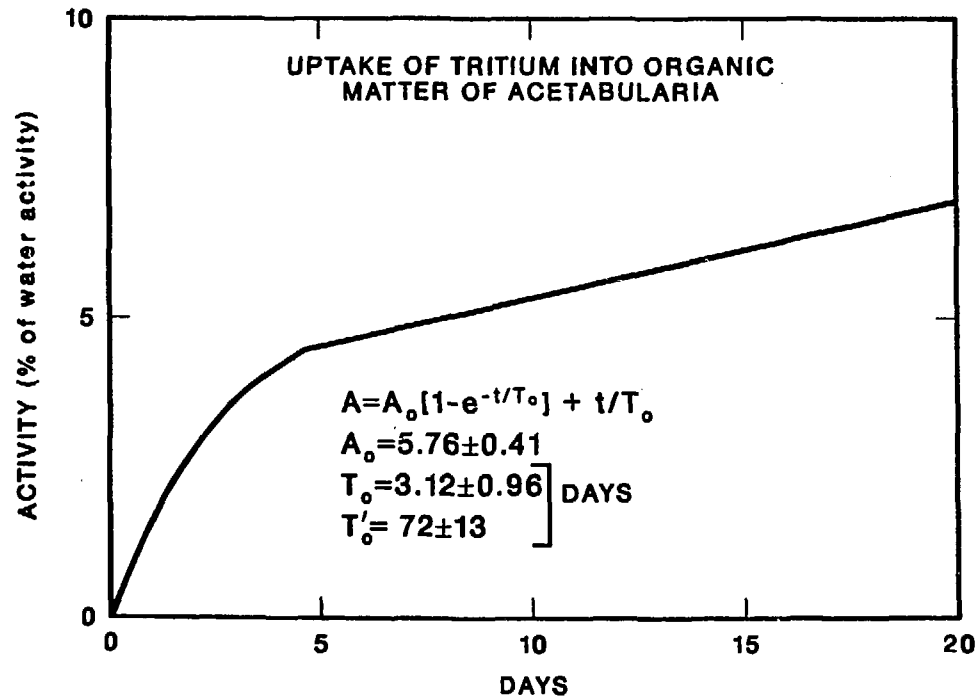
AFTER STRACK et al. 1982



**UPTAKE OF TRITIATED WATER IN LAURENCIA OBTUSA,  
SHOWING TWO COMPONENTS, WITH A HALF LIFE OF  
0.4 AND 22 MIN. (after CINELLI et al. 1983)**

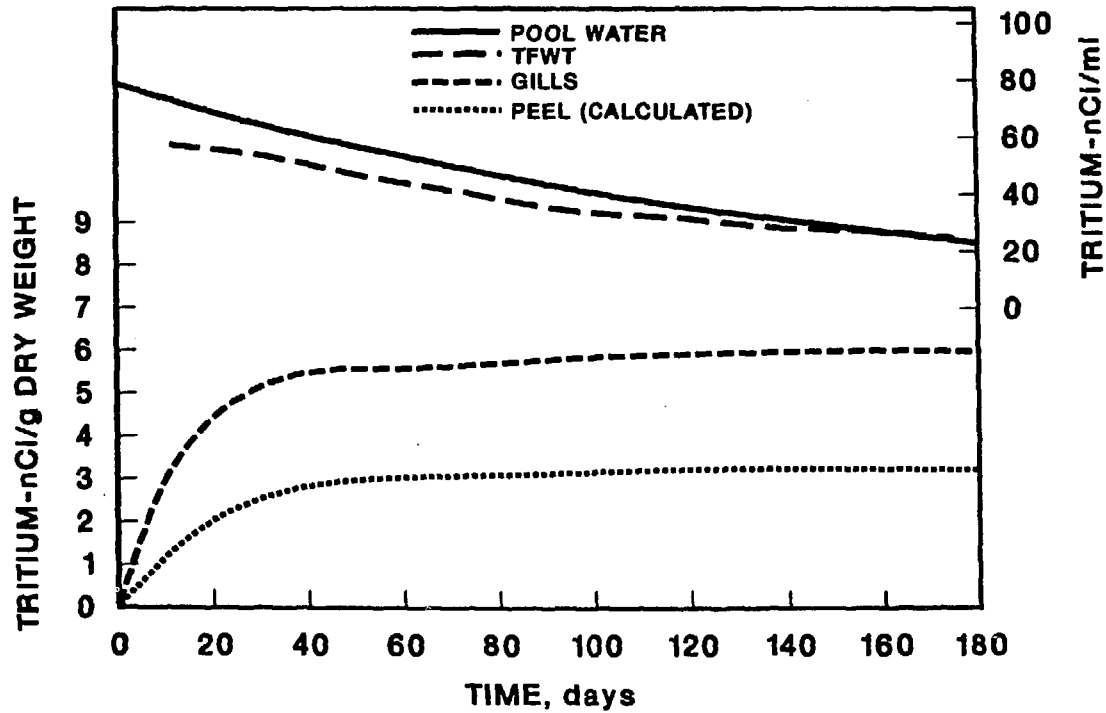


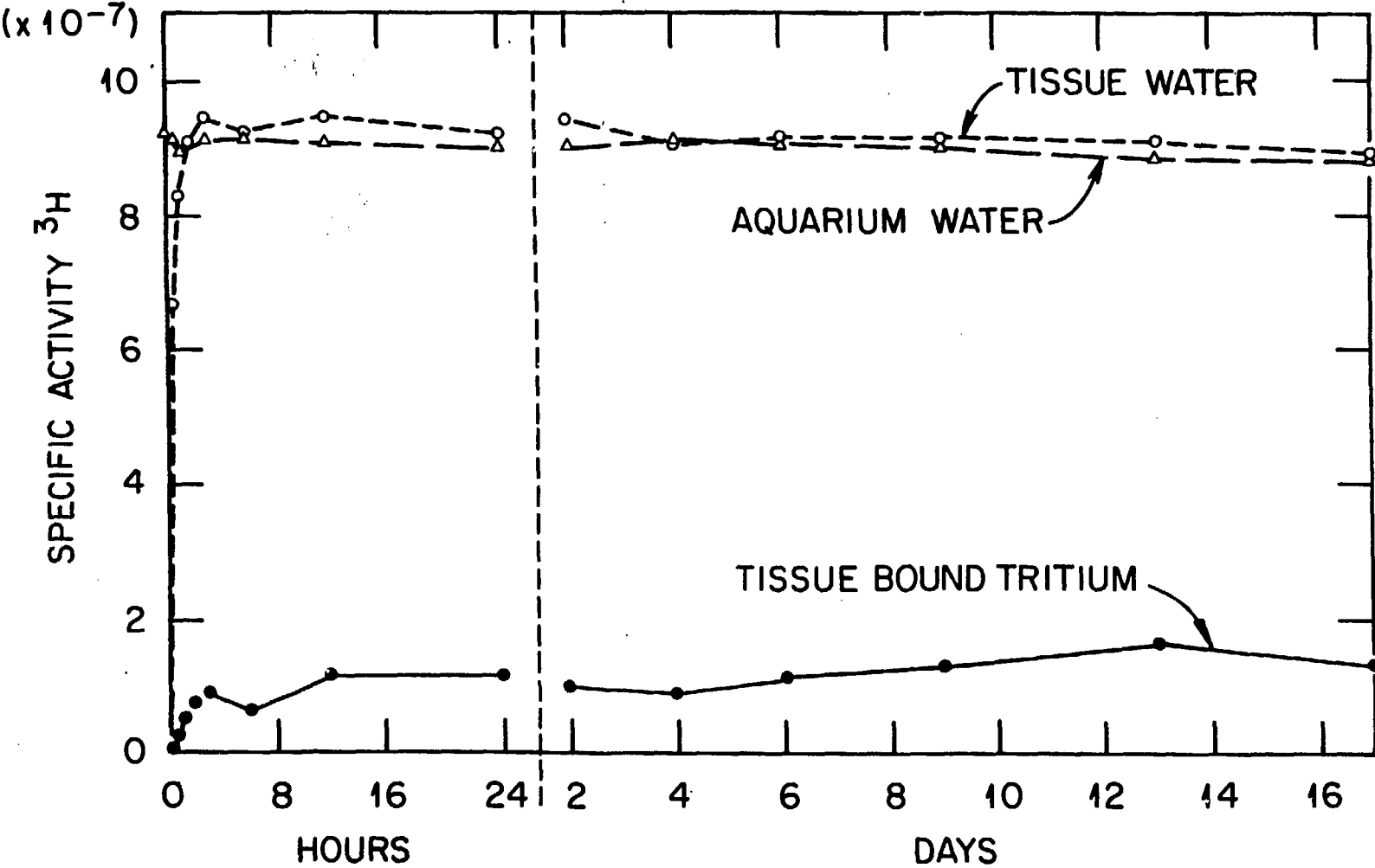
**INCORPORATION OF TRITIUM INTO THE ORGANIC MATTER OF THE  
ALGA ACETABULARIA SHOWING TWO COMPONENTS WITH HALF-LIVES  
OF 2.3 d AND 50 d**



(AFTER BONOTTO et al. 1982)

### TRITIUM UPTAKE IN CLAMS (AFTER HARRISON et al. 1973)





The Uptake of Tissue Water Tritium and Tissue Bound Tritium by *Gambusia* Exposed to Tritiated Water.