TRITIUM TRANSFER PATHWAYS IN THE AQUATIC PLANT *HYDRILLA VERTICELLATA**

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ABSTRACT

Rates of uptake and release of tritium by the aquatic plant *Hydrilla verticellata* were studied under controlled conditions. Significance of the persistence patterns of the aqueous and organic phases of tritium in the plant was established under different periods of exposure. The mean residence time for tissue-free-water-tritium was found to be 0·36 hours while the tissue-bound-tritium showed two components having mean residence time of 2·53 hours and 15·87 days respectively.

By reversing the pathways of tritium between the plant and the medium after long exposure times, the relative significance of the aqueous and organic phases of tritium in the plant and their persistence patterns have been established. It was also found that the water turnover rates in the plant was 0·028 ml/min/g.

INTRODUCTION

TRITIUM produced by the natural and artificial processes in the atmosphere is invariably present in the oxidized state as HTO and in this form it follows the hydrologic cycle, and entering into all hydrogen-containing compounds by isotopic exchange. Tritium transfer pathways in the ecosystems *in general and in food chains in* particular have attracted the attention of several workers[1-3]. Detailed investigations on the tritium persistence in the different compartments of the ecosystem have significant applications both in the field of radioecology and from environmental *radiation point of view.*

An attempt has been made in the present work to generate data on the parameters governing the fixation of tritium in the organic phase. The mean residence times have been calculated on the basis of whole tritium values obtained from samples exposed for different periods of times under different conditions of exposure. By this method the significant fraction of tritium in the plant is gradually changed from tissue-free-water-tritium (TFWT) to tissue-bound-tritium (TBT) labile component and later to TBT non-labile component.

MATERIALS AND METHOD

About 2 kg of *Hydrilla verticellata* obtained from the Taraporewalla Aquarium, Bombay, were exposed to varying periods of time in a laboratory aquarium of approximately 25 l capacity, under controlled humidity and temperature conditions. The medium water was changed at a fixed time schedule depending on the type of investigations. The plants were constantly exposed to 40 W fluorescent lighting throughout the period of study.

The exposure time of the aquatic plants in the active media was varied in each of the experiment in order to determine the period of time required for effective tritium transfer between the two main hydrogen pools of the system, *viz.*, the plant water and the organic fraction in the plant.

The periodicity of sampling varied from 15 mts to 24 hrs depending upon the status of exposure of the plant-system. The wet sample collected at random was dried using a filter paper taking care not to break their stems while removing the adhering water droplets. The wet and lyophilised tissues were burnt separately in an oxygen atmosphere in order to extract the activity contained in HTO or HT[4]. The activity in the extracts is then counted in a Liquid Scintillation Spectrometer (Packard Model 2211) applying appropriate corrections for quenching.

Hydrogen content of the lyophillised tissue was estimated by burning the sample and absorbing the steam in $MgClO_4$, using conventional chemical apparatus.

Exposure schedule.—F. L. Harrison *et al.*[3], describe the procedures required to fix preferentially the TBT in certain marine animals on the basis of a mathematical model which takes into effect all the possible tritium pathways in a total aquatic environment. The same model was used here to a major extent with only minor modifications for application to aquatic plants. The aquatic plants are exposed to active media for a certain period and are transferred to inactive media in stages thereby eluting the different components of tritium compounds in the plant. Periodic sampling was carried out on the whole plant for total tritium content in order to identify the nature of release patterns. The investigations are divided into a number of batches of experiments.

A preliminary run indicated the fast water turnover rates of these plants. The tritium concentration

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values of the pool water was increased from 10^4 dpm/ml to 10^6 and 10^8 dpm/ml in order to have sufficient concentration in the TBT phase.

The plants were exposed to 8 hours in an active medium to determine the TFWT uptake pattern and then were transferred to inactive medium to determine the TFWT and labile TBT release patterns. These studies were continued upto 80 hours.

The next batch of experiments on the plant covered three different phases of investigations :

Phase 1.—The plants were immersed in a tritiated water medium (concentration of about 10^8 dpm/ml) for a period of 18 days during which period the uptake pattern and equilibrium behaviour under controlled conditions were studied. The medium water was also regularly monitored, in order to correlate the saturation activity levels of the plant and the medium, as well as to study the loss of tritium from the aquarium due to evaporation.

Phase 2.—The plants exposed to tritium concentrations as per the schedule in phase 1 were then transferred to an inactive pool and the investigations continued for 42 days. During this phase the TFWT release pattern as well as labile and non-labile tritium release patterns were studied. The pool water concentrations were found to have increased during this period reaching an equilibrium value. This is attributed as due to tritium releases from the aquatic plant to the medium water.

Phase 3.—The plants were transferred at this juncture to a freshwater aquarium for a second time. The TBT release patterns were found to be the most prominent one during this period.

Results and Discussion

Figure 1 presents the results of the rate of uptake and release of tritium for short intervals of time. The aquatic plants reached an equilibrium concentration corresponding to 80% of the medium water concentration, within a period of one hour, when normalised for moisture content of the plant (0·76 w/w). A least square analysis of the results showed three components of release pattern. The mean residence times corresponding to TFWT phase and TBT labile phase were found to be 0·36 h and 2·53 h respectively.

The water turnover rate of the plant was determined on the basis of the steady-state tracer kinetics[5]. Under equilibrium conditions

$$S_m \cdot R = \lambda \cdot S_s$$

where S_m and S_s are specific activity of tritium in the medium (cpm/ml) and the sample (cpm/wet g), R is the rate of uptake (ml/min.) and λ is the inverse of the tritium mean residence time in minutes.

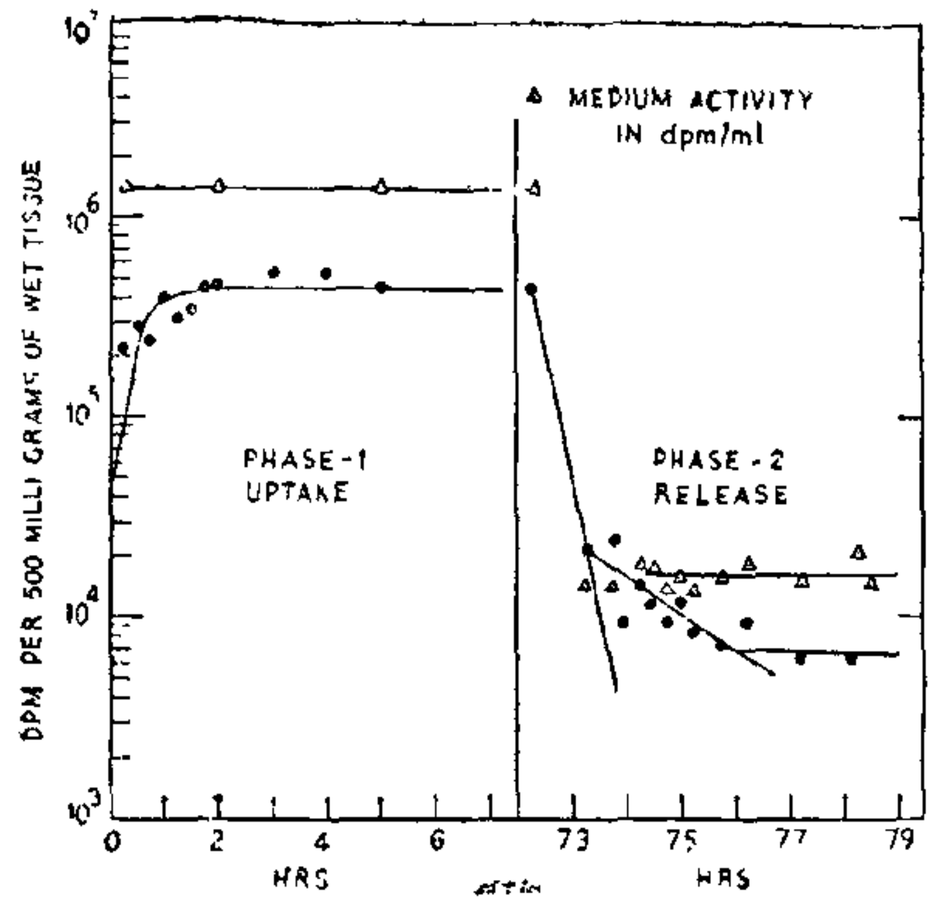

FIG. 1. Water turnover rate and TFWT release pattern

Since the mean residence time values for TBT are large compared to those of TFWT, R is calculated as 0·028 ml/min./g corresponding to a tritium mean residence value of 0·36 h.

Figure 2 shows the three phases of tritium transfer pathways observed in the experiments. Phase 1 has an uptake pattern identical to that in Fig. 1, except for the evaporation losses shown in the medium concentration values (as well as those shown by the corresponding aquatic plant samples). In phase 2, the two components of tritium releases are observed. The first one shows an extremely rapid rate of release corresponding to that of TFWT and TBT labile components. The second component shows the long-lived TBT non-labile tritium in the aquatic plants. The mean residence time for this component is found to be 15·87 days indicating that the organic phase transfer mechanism has an extremely slow rate constant compared to the other phases. Under equilibrium conditions, when the tritium concentrations of the plant tissue were normalised, the medium water showed values of specific activity lower than those for the plant tissue for a few days confirming the fact that pathways (of tritium) have been reversed due to the preferential incorporation of tritium in the organic phase of the plant when immersed in an inactive medium.

In phase 3, the above phenomenon is highly pronounced due to the fact that the only major component of tritium left behind is the non-labile

one. The mean residence time was once again found to be same, *viz.*, 15·87 days.

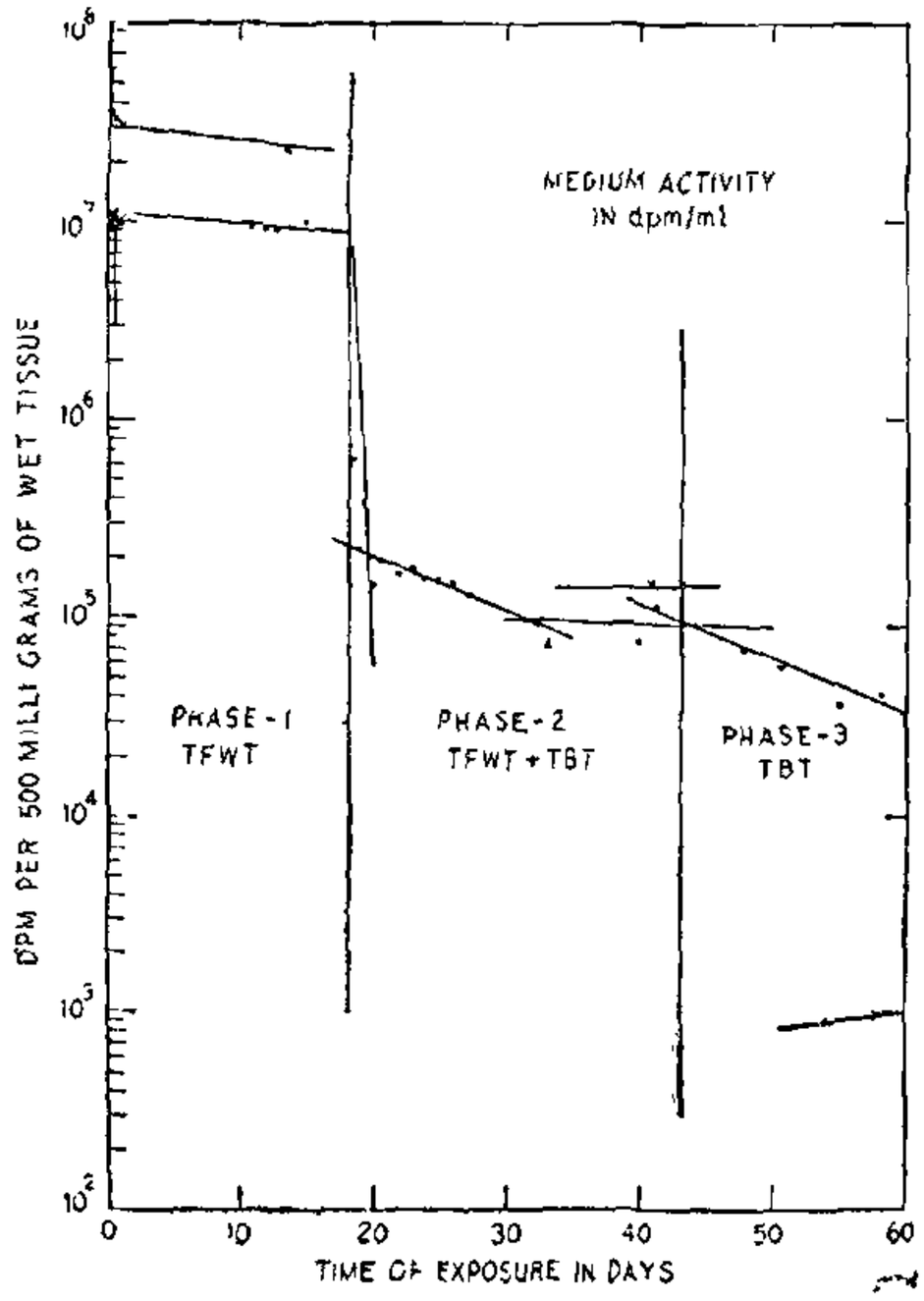

FIG. 2. Tritium uptake and release pattern or long periods of exposure.

Figure 3 shows the schematics of the tritium transfer pathways in the aquatic plant as reported in the experimental observations (*vide* Figs. 1 and 2) during different phases of the experiments.

The relative fractions of tritium incorporated in the three phases of the bound and free forms of tritium in the aquatic plant were found to be 100 : 3·27 : 0·77. This shows that 3·14% of tritium is in the labile TBT form and 0·74% is in the non-labile TBT form. This could be the reason for the necessity to have high concentrations of medium water in order to obtain a preferential fixing of TBT in the aquatic plant. The values obtained for the different fractions of tritium (bound and free) in the aquatic plant *Hydrilla verticellata* in the above study agree well with the values obtained by H. D. Bruner[6]. He has obtained similar orders of magnitude in his study of the specific activity values for different forms of bound and free tritium in phytoplankton, zooplankton and certain aquatic plants.

The hydrogen content of the lyophillised tissue of the aquatic plant was found to be 7·26%. The moisture content of the plant being 0·76 w/w, it can be seen that the available hydrogen in organic phase is 1·74%. However the total TBT fraction as determined by uptake measurements was found to be 3·85%. This variation in values can be attributed to either or both of the following reasons.

(*i*) The value of hydrogen content in organic phase can be on the lower side, due to possible losses of certain volatile substances during lyophillisation.

(*ii*) The TBT fraction as determined by the analysis of total ^3H content estimations of the aquatic plant can show a slightly higher va'ue due to possible interferences from TFWT fractions. In order to circumvent this the mathematical treatment has to be enlarged so as to determine the extent of such interferences.

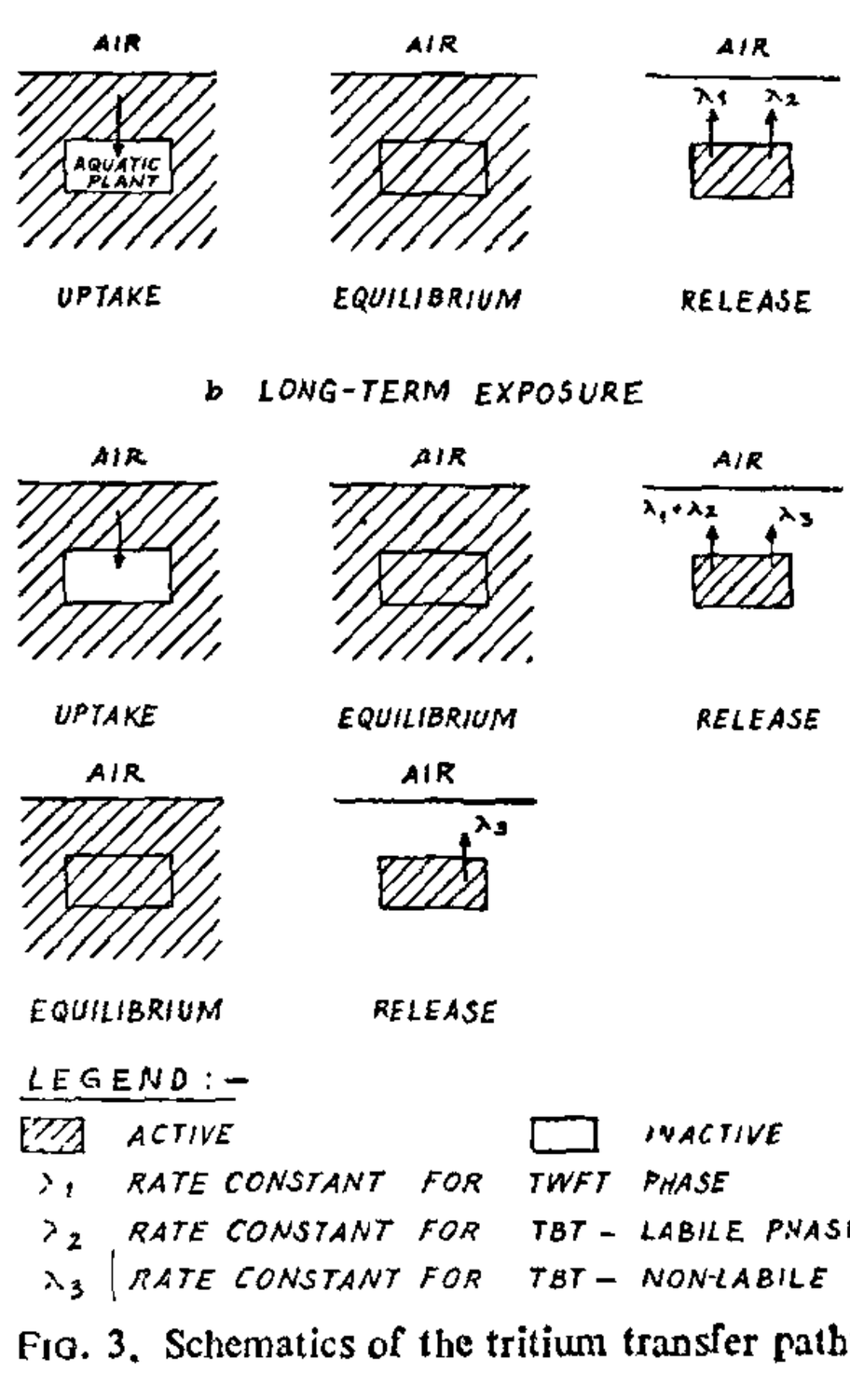

FIG. 3. Schematics of the tritium transfer pathways.

CONCLUSION

The aquatic plant, *Hydrilla verticellata*, shows three distinct mean residence time values for tritium, one relating to aqueous phase and two corresponding to the organic phase. It is possible to determine these components by estimating the total tritium content of the plant samples at different periods of time after a certain type of exposure schedule. Further, it has been observed that the tritium

pathway can be reversed between the medium and the plant, after long exposure times.

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2. Kirchmann, R. *et al., Ibid.,* 1973, p. 341.
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6. Bruner, H. D., *Tritium.* Ed. A. Alan Moghissi and Melvin W. Carter (Messenger Graphics, Phoenix, Arizona and Las Vegas, Nevada), 1973, p. 303.

ULTRASONIC RELAXATION IN ETHYL SALICYLATE

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ABSTRACT

Measurements of ultrasonic absorption have been made by pulse technique in liquid ethyl salicylate and also in its solutions with *m*-xylene and nitrobenzene as solvents. Results are analysed on the basis of single relaxation mechanism. Relaxation frequency is found to be independent of the concentration of ethyl salicylate in both the solvents. Results are explained on the basis of rotational isomerism in ethyl salicylate.

SEVERAL investigators[1-3] have reported ultrasonic relaxation effects in esters of monocarboxylic acids. Relaxation in OH substituted esters of carboxylic acids has not been thoroughly investigated. These esters from a separate class in that they exhibit intramolecular hydrogen bonding. Murthy and Rao[4] have reported the relaxation in liquid methyl salicylate which was attributed to rotational isomerism. In this note measurements of ultrasonic abosorption in pure liquid etbyl salicylate and also in solutions of ethyl salicylate with *m*-xylene and nitrobenzene are presented.

Measurements of absorption and velocity have been made by pulse technique in the frequency range 4 to 15 MHz. Liquids used are of high grade purity and have been distilled before use.

Results of absorption have been analysed on the basis of a single relaxation mechanism

$$\frac{\alpha}{f^2} = B + \frac{A}{1 + \left(\frac{f}{f_r}\right)^2}$$

where A and B are constants of relaxation and f is the characteristic frequency of relaxation.

The excess absorption per wavelength ($\alpha'\lambda$) at any frequency is calculated from

$$(\alpha'\lambda) = \left(\frac{\alpha}{f^2} - B\right) f C$$

Maximum excess absorption per wavelength μ_m is computed from

$$\mu_m = \frac{1}{2} A f_r C$$

where C is the velocity of sound.

TABLE I

Ultrasonic absorption and relaxation parameters in ethyl salicylate

Temp. °C	v m/sec	f Mc/sec	$10^{17} \times \alpha/f^2$ cm^{-1} sec	$10^2 \times \alpha'\lambda$ *db*	10^{17} A cm^{-1} sec^2	f_r Mc/sec	10^{17} B cm^{-1} sec^2
30	1360	4·44 7·20 10·10 13·00	1300 570 360 225	6·7 4·6 3·9 3·0	2500	3·9	30
40	1300	4·44 7·20 10·10 13·00	1365 680 430 320	6·5 5·0 4·2 3·8	2000	4·9	60
50	1240	4·44 7·20 10·10 13·00	1400 800 490 410	6·4 5·7 4·6 4·8	1666	6·3	65