

## Measurement of Tritium Activity in Plants by Ice Extraction Method

O.Pelled, S.Ovad, Y.Tubul, S.Tsroya, R.Gonen, A.Abraham, M.Weinstein, U.German

*Nuclear Research Center, Negev, P.O.B 9001 Beer Sheva, 84190, Israel.*

### INTRODUCTION

Tritium is produced primarily by interactions of cosmic rays with the atmosphere. However, nuclear installations may add significantly tritium to the surroundings, increasing its concentration. The main sources of tritium released by man are linked to the nuclear power cycle: nuclear power stations, nuclear fuel reprocessing plants or tritium production plants<sup>(1)</sup>. Tritium is found in the environment mainly as tritiated water, in gaseous or liquid form (HTO, T<sub>2</sub>O), in the surrounding air and in soil. It accumulates in plants, which may use as a measure to the level of tritium concentration in the environment.

The most common routes of tritium uptake from the environment in plants are from atmospheric humidity and by precipitation water which entered the soil<sup>(2)</sup>. The fraction of tritium bound to the plant tissue is small compared to that present as tritiated water in the plant (from 0.06% to 0.3% for growing crops). The tritiated water uptake is through the roots, as tritiated water from the soil follows a pathway similar to that of ordinary water. As most tritium in plants consists of tritiated water, the measurements of only the tissue free water tritium concentration (as HTO or T<sub>2</sub>O) gives an accurate estimate of the tritium content in the plant.

Analyzing free tritium in biological matrices usually requires using the freeze-drying method to extract the water from the sample, and then measure the water collected in a cold trap with a Liquid Scintillation Counter (LSC). The "freeze-drying" occurs because of the sublimation of the frozen water inside the plant, that takes place when the temperature is beneath the triple point and the vapour pressure is low. In the temperature range of -5° to -10° C the mechanism that plants use to avoid freezing is drawing of water from the [cell protoplasm](#) into the [intercellular](#) spaces<sup>(3,4,5)</sup>. Changes in [cell membrane permeability](#) allow water to leave the cell and enter the spaces between the cells where it freezes instead of freezing within the cell and causing its death. This process continues until the cells are almost totally dehydrated. In the temperature range of -20° to -60° C the intra-cellular water freeze forms "sharp" ice crystals that cause the [death](#) of the cells. Water (H<sub>2</sub>O) and tritiated water (HTO) behave nearly identically in both liquid and vapour phases<sup>(5)</sup>.

The freeze-drying method, although relatively simple, requires the use of dedicated systems and is time consuming. When a plant is frozen in a closed bag, ice is accumulated on the exterior surface of the plant and in the plastic bag that contained the sample, producing a "self-freeze drying" effect. This ice may be directly used for tritium evaluation if the tritium measurement results are compatible with the generally accepted freeze-drying (lyophilization) method. The present work presents a comparison of this simple Ice Extraction Method (IEM) for tritiated water analysis with the standard lyophilization method.

## MATERIALS AND METHODS

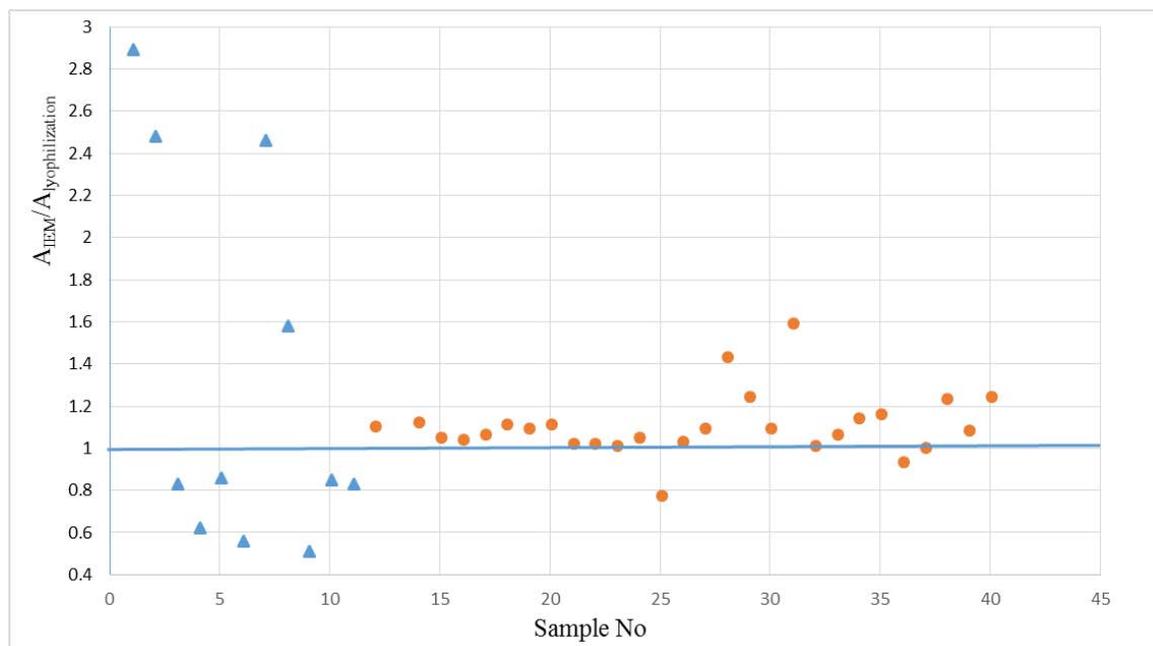
More than 100 desert plants samples containing stem and foliage were collected, put in a double plastic bags and were frozen to  $-20^{\circ}\text{C}$  for more than 3 months, allowing formation of enough ice (to produce more than 15 cc of water).

The ice from each sample was collected, melted and filtrated through filter paper (Whatman No-41), to separate solids. In some cases, when the final water sample was turbid, a distillation process was employed. After collecting the ice from the bag, the remaining plant was lyophilized and a second sample was obtained. This was done by placing the plant in a glass container, which was part of a vacuum system intended to maintain the sublimation condition and a cold trap for the water vapour collection.

The two liquid samples obtained by both methods, were counted in a liquid scintillation (LSC) system for 20 minutes. Quenching correction was applied as usual in liquid scintillation counting.

## RESULTS

The vegetation used for the investigation was random plants in the Negev area. From 100 samples, only 40 results were above the Minimal Detection Activity (MDA) of the LSC system and were suitable for comparison. Fig.1 shows the tritium activity ratio  $A_{\text{IEM}}/A_{\text{lyophilization}}$  of the samples prepared by the two methods. The blue (triangle) marks indicate results with a large standard deviation ( $>50\%$ ) caused by a very low tritium activity, near the MDA of the LSC system ( $0.01\text{Bq/cc}$ ). The red (circle) marks are related to results with a standard deviation lower than  $30\%$ .



**Fig.1:** The tritium activity ratio  $A_{\text{IEM}}/A_{\text{lyophilization}}$  for samples containing tritium activities higher than the MDA of the LSC system.

It can be seen that most results, except the values for very low activities with very high associated statistical uncertainties, show an agreement of up to about 20% between the IEM and the standard lyophilization methods. The average ratio is  $1.14 \pm 0.26$ , which indicates a slightly bias of approximately 15% in favour of the IEM method. However, this bias is within the standard error of the average.

## CONCLUSIONS

A very simple method named Ice Extraction Method (IEM) for tritiated water analysis in plants was checked in comparison to the standard lyophilization method. No preparation system is needed, as the ice which is formed in the bags containing the plants during their stay in the freezer is directly used. Plants from the natural environment in the Negev were used for this check, which contained very small tritium activities (mostly below MDA), therefore the statistical uncertainties are high. A comparison to the standard lyophilization method indicates a good agreement, within the statistical constraints. More in-depth investigation is needed to validate the method with plants containing higher tritium activities, as growing them in a controlled tritium environment.

## REFERENCES

1. Okada, S., Momoshima, N., Overview of tritium: characteristics, sources and problems. *Health Phys.* 65, (1993) 595–608.
2. C. Boyer, L. Vichot, M. Fromm, Y. Losset, F. Tatin-Froux, P. Guetat, P.M. Badot. Tritium in plants: A review of current knowledge. *Environmental and Experimental Botany* 67, (2009) 34–51.
3. J. S. McKenzie, C. J. Weiser, E. J. Stadelmann, M. J. Burke. Water Permeability and Cold Hardiness of Cortex Cells in *Cornus stolonifera* Michx. *Plant Physiol.* 54, (1974) 173-176.
4. R. S. Pearce. Plant Freezing and Damage, *Annals of Botany* 87, (2001) 417-424.
5. B. J. Andraski, M. W. Sandstrom, R. L. Michel, J. C. Radyk, D. A. Stonestrom, M. J. Johnson, and C. J. Mayers. Simplified Method for Detecting Tritium Contamination in Plants and Soil. *J. Environ. Qual.* 32, (2003) 988–995.