KnE Energy & Physics
PhysBioSymp17
The 2nd International Symposium on
Physics, Engineering and Technologies for Biomedicine
Volume 2018

Knowledge E
Engaging minds

Conference Paper

Development of Aquatic Bioassay with *Lemna minor* and *Spirodela polirhiza* for Screening of Waters Contaminated with Tritium

Momot O. A., Mirzeabasov O. A., and Synzynys B. I.

National Research Nuclear University MEPhI (Moscow Engineering Physics Institute), Kashirskoe shosse 31, Moscow, 115409, Russia

Abstract

The work is aimed at studying morphometric parameters of the aquatic plants *Lemna minor and Spirodela polyrhiza* to develop a bioassay for water bodies contaminated with tritium. A considerable increase is noted of *Lemna minor* plants with the greatest biomass and area at high specific activity of tritium (3700 Bq/l) in natural water objects. A reliable *Spirodela polyrhiza* species dependence on specific tritium activity is not observed.

Keywords: Tritium, bioindication, freshwater body, *Lemna minor*, *Spirodela polyrhiza*

Corresponding Author:
O. A. Momot
momotulya@gmail.com

Received: 17 January 2018
Accepted: 25 March 2018
Published: 17 April 2018

Publishing services provided by Knowledge E

© Momot O. A. et al. This article is distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use and redistribution provided that the original author and source are credited.

Selection and Peer-review under the responsibility of the PhysBioSymp17 Conference Committee.

OPEN ACCESS

1. Introduction

Tritium (H-3) is the unique radionuclide widely evidenced in the biosphere constituents, atmosphere, water reservoirs and soil, in particular. Being actively introduced into living organisms, it can violate the structure of biologically important molecules in cells not only via internal beta-radiation but also as a result of H-3 transmutation into He-3. It induces disruptions in DNA chemical bindings and the following cell death and violations in the organism activity [1]. Such discovered disturbances allowed the ISO specialists in 2005 and Russian hygienists in 2010 to develop a bioassay with *Lemna minor* for recording harmful compounds in the chemical production wastewater [2]. Since H-3 is a chemical agent as well as a radiator, it was decided to use *Lemna minor* as the bioassay for detecting natural and technical water contaminated with tritium as a part of tritium oxide (HTO) and organically bounded tritium (OBT) [1].

The research is aimed at studying the morphometric features of *Lemna minor* and (for comparison) the allied species *Spirodela polyrhiza* to develop the radiation (or combined chemical and radiation) bioassay for screening surface and groundwater contaminated with tritium compounds. In future the data will allow to interpret the results of ecological and health risk assessment when drinking water with small amounts of tritium.

2. Materials and methods

The suggested method of bioassay is based on the route chart of risk assessment for chemical substances (all at once radiators) developed by Momot O.A [3]. The specific activity of tritium in water after special sample preparation was determined by a scintillation spectrometer Quantulus-1220 with internal standards and reference water samples. Morphological indices of algae and polyrhiza were estimated by a special camera microscope; the statistical data processing has been realized with the information space R software. 100 algae and polyrhiza plants were used for any sampling site. Distribution functions were plotted from the corresponding parameter values, for which estimated were the indices corresponding to median values, the first and the third quartiles of the distribution function.

3. Results

Tritium in aquatic plants is present in a form of tritium oxide and organically bounded tritium. The structure of biologically significant molecule, for example DNA, changes as a result of beta-radiation and He-3 transmutation. Finally, these changes lead to violations in the morphometric features of plants and it is not possible to trace them in bioassay. Two *Lemnaceae* plants have been chosen: *Lemna minor* and *Spirodela polyrhiza* found in the river Protva water bodies, i.e. in the mainstream and oxbow areas which are most of the time separated by natural barriers.

3.1. Tritium determination in water bodies

Water and *Lemna minor* samples have been taken in the river Protva (site p4) and some oxbow areas (sites p1, p2, p3, p5). In July 2017 the specific activity of tritium in water was the following: site p1 – 691 Bq/l, site p2 – 654 Bq/l, site p3 – 650 Bq/l, site p4 (Protva mainstream) – 21,5 Bq/l and site p5 – 3693 Bq/l.

DOI 10.18502/ken.v3i2.1821

3.2. Tritium effect on *Lemna minor* and Spirodela polyrhiza

Leaf mass and area as well as root length have been chosen as morphometric end-points. Figure 1 presents the experimentally determined leaf mass distribution of *Lemna minor* plants: X axis – leaf mass and root mass of some plants in grams, Y axis – a fraction of species with such a mass. Figure 2 shows the root length distribution for *Lemna minor* plants from the same water bodies.

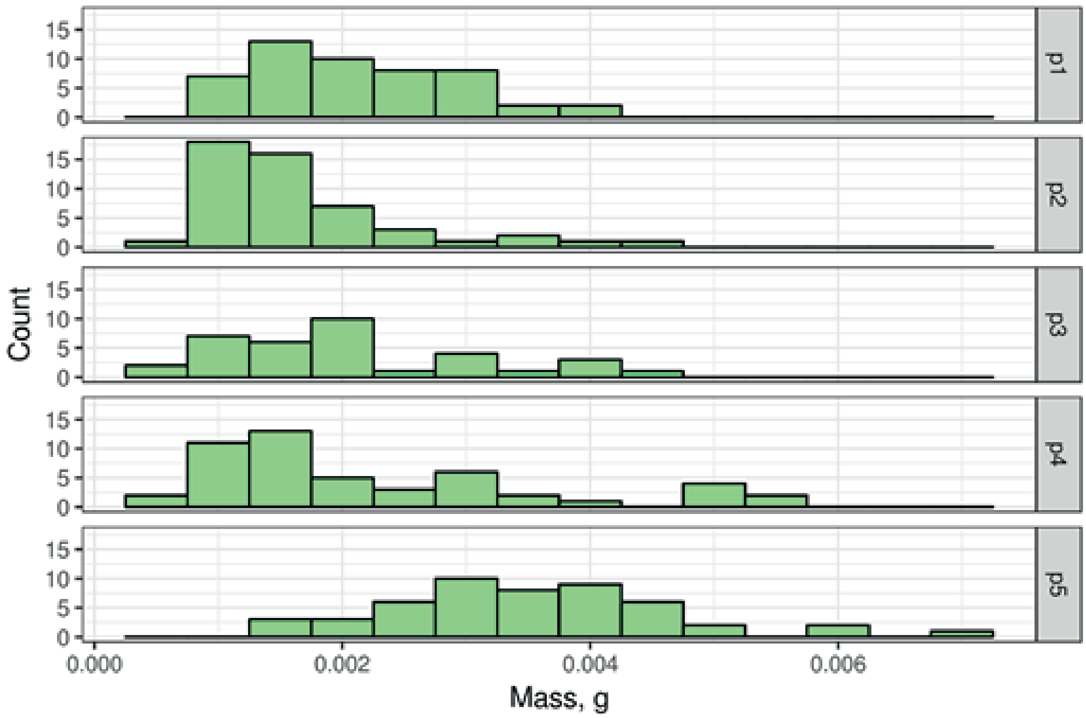

Figure 1: Mass distribution of *Lemna minor* plants from different water bodies of the river Protva basin.

4. Discussion

Mass distribution of plants from water bodies noted as sites p1–p4 does not practically differ (Figure 1) and the corresponding median values for these distribution functions are the following: 2.00; 1.50; 1.90 и 1.70, respectively. A considerable increase in a fraction of plants with the highest biomass is noted for those constantly growing at tritium radiation of about 3700 Bq/l: the corresponding median value for mass distribution is 3.35. In terms of their growth and development, the plants are supposed to undergo the stimulating effect of tritium. It is confirmed by laboratory studies and experiments in the river Yenisei (in the coverage of Zhelesnogorsk Ore Mining and Smelting Plant) (L.G. Bondareva and M.A. Subbotin as well as I.S. Bondar et.al.) [4, 5].

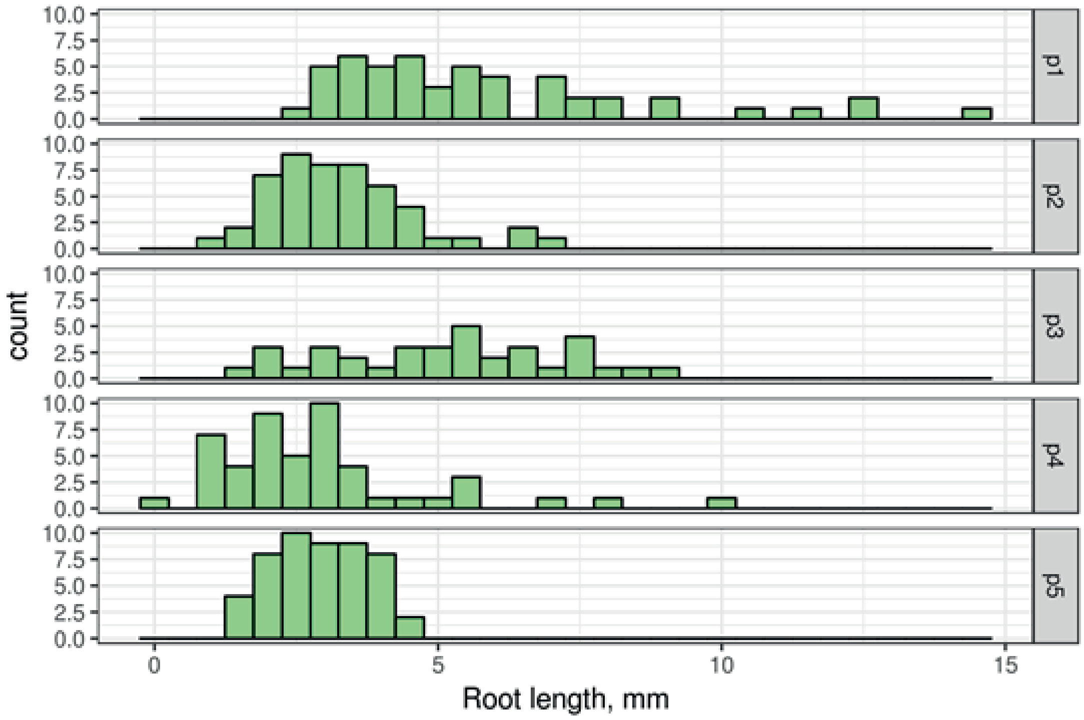

Figure 2: Root length distribution of *Lemna minor* plants from water bodies with different tritium concentration.

There is no special regularity in the *Lemna minor* root length distribution (Figure 2). The pair comparison of mean root length distributions using the pairwise.wilcox.test program (on the assumption that these distributions are not normal) shows that they range significantly in this parameter at all sampling sites, although tritium radioactivity in these water bodies is comparable. This index, in case of its application as a bioindicator, requires more studies. This conclusion is confirmed by compared median values for *Lemna minor* root lengths. For sampling sites p1, p2, p3, p4 and p5 the corresponding median values (mm) are 5.20; 3.15; 5.30; 2.60 and 2.80.

The normality of *Lemna minor* leaf area distribution has been also examined. This bioassay or bioindicator is obvious, however, its exact determination requires appropriate computer aligned devices. Nevertheless, it is naturally to expect a direct correlation between the leaf mass and its surface area. It was really observed in experiments: medians of the corresponding distributions for *Lemna minor* upper surface leaf areas differ widely. For plants growing in the water body with maximum tritium radioactivity (3690 Bq/l) the area is 4.25 мм^2, and for those in the “testing” water body, i.e. the river Protva mainstream with the specific activity of 21.5 Bq/l during measurements it is 2.56 mm^2.

Unfortunately, *Spirodela polyrhiza* did not grow in the water body with the highest tritium activity. Therefore, distributions in the mass, mean root length and leaf areas are compared only for plants growing in water bodies 1-4. There is no reliable dependence of plant mass on the specific tritium activity. The conclusion on determining such parameters as the mean root length and leaf area is similar. The expected reason of missing regularities in this plant reaction to radiation and H-3 transmutation is its relatively high radio resistance which should be laboratory and experimentally substantiated. It may be the part of more extended investigations of ecological properties of *Spirodela polyrhiza*.

5. Conclusion

Large-scale research of radiobiological regularities of *Lemna minor* has been initiated in the last ten years. The long-run objective of it is the development of an adequate bioassay to detect the stimulating as well as suppressing effect of radiation [4-6].

Technogenic radionuclides, except tritium, are not found near Obninsk. Last year the concentration of this beta-emitter in tritium oxide(HTO) in the public waterworks system was 10.5 Bq/l. Tritium activity in some water bodies does not exceed 3600 Бк/л, and it is much lower than the intervention level 7600 Bq/l according to the domestic standards NRB 99/2009 [7]. The corresponding value of individual life risk in case of drinking water containing 10.5 Bq/l for Obninsk inhabitant is 10^{-8} [3]. It should be suggested, however, that some amounts of tritium, even in trace amounts, involved in DNA, may be dangerous for stochastic reactions (endpoints) in quickly dividing cell populations and radiosensitive tissues and organs due to soft beta-radiation n He transmutations. Now we are at the next stage of our activities: finding of the specific activity of tritium in DNA from *Lemna minor* and, on this basis, developing of a microdosimetric model to assess the local irradiation dose; the mechanism of ionizing radiation which stimulates *Lemna minor* growth is also studied.

References

[1] O.A. Momot, «Application of biotesting methods in the methodology of health risk assessment,» PhD Diss., Kaluga, 180, pp.2007.

[2] ISO/DIS 20079. Water quality – Determination of the toxic effect of water constituents and waste water to duckweed (*Lemna minor*) – Duckweed growth inhibition test. Reference number ISO 20079:2005.

[3] O.A. Momot, B.I. Synzynys, A.A. Oudalova,«Health risk assessment of potable water containing small amounts of tritium oxide,» *Journal of Physics: Conference Series*, vol.

784, 2017012040 Doi: 10.1088/1742-6596/784/1/012040, 2017.

[4] L.G. Bondareva, M.A. Subbotin, «Tritium assimilation by aquatic plants *Elodea canadencis and Lemna minor*,» *Radiatsionnaia biologiia, radioecologiia*, vol.56, no. 4, pp. 440-446, 2016.

[5] I.S. Bondar, E.A. Yushkova, V.G. Zainullin «Effect of gamma-radiation on morphometric properties of *Lemna minor,» Radiatsionnaia biologiia, radioecologiia*, vol. 56, no. 6, pp. 617-622, 2016.

[6] A. Van Hoeck, N. Horemans, R. Nauts, M. Van Hees, H. Vandenhove, R. Blust, «*Lemna minor* plants chronically exposed to ionizing radiation: RNA-seq analysis indicated a dose rate dependent shift from assimilation to survival strategies,» *Plant Science*, vol. 257, pp. 84-95, 2017.

[7] Russian Radiation Safety Standards RSS-99/2009: SRS 2.6.1.2.2523-09, Moscow, 100 pp., 2009.