

TRITIUM LABEL IN STUDYING PROTEIN-LIGAND INTERACTION: SELF-ORGANIZATION AT THE INTERFACES

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Tritium is the only radioactive isotope of hydrogen and it is an appropriate radiolabel for organic molecules including surfactants, peptides, proteins and etc. Moreover, tritium as a tracer can be also used for analysis of protein-ligand complex formation at the interfaces, including liquid-liquid system that is a model of cellular membrane. In the present study we developed a novel approach to the study complexes formation between protein and organic ligand at the interfaces as well as in the bulk of the solution. We used lysozyme as a model protein and as an enzyme of medical purpose. Surfactants, including ionic and non-ionic, low and high molecular weight substances and even humic substances, were considered as a ligand.

The procedure of analysis of protein-ligand intermolecular interactions includes two important radiochemical stages. First, studying adsorption of both protein and ligand at the liquid-liquid interface by means of tritium labeled compounds and liquid scintillation spectrometry as a scintillation phase technique [1]. To this end both protein and organic ligand required to be labeled with tritium and we used tritium thermal activation method for radiolabeling [2].

The second radiochemical study includes the bombardment of the protein-ligand mixed adsorption layer with tritium atoms following by the analysis of tritium distribution in the components of the target. In the case of protein, it was subjected to total hydrolysis and determination of the specific radioactivity of each amino acid. On the bases of lysozyme structure phenylalanine and proline were chosen as reference amino acid residues for determining protein orientation in the mixed adsorption layer that was done for the adsorption layers with ionic and non-ionic surfactants.

The developed approach allowed us to reveal the encapsulation of lysozyme with humic substances and in conjunction with classical methods of protein structure analysis to describe completely lysozyme-surfactant complexes. The main results obtained with tritium labeled lysozyme, cationic, anionic and non-ionic surfactants, and humic substances will be discussed in the presentation.

1. M.G. Chernysheva, G.A. Badun, Langmuir. 27, 2188 (2011).
2. G.A. Badun, M.G. Chernysheva, A.L. Ksenofontov, Radiochimica Acta. 100, 401 (2012).

The speaker is a student or young scientist

No

Section

1. Applications of nuclear methods in science and technology

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