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# Structural studies of self-assembling ferritin-based protein complexes

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Ferritin is a protein complex responsible for storing iron in various organisms. One of the most important properties of ferritin is the spontaneous formation of a spherical hollow protein globule consisting of 24 subunits, this process is called self-assembly [1]. The protein globule is very stable and can sustain significant temperature and pH changes [2]. The self-assembly process plays a crucial role in the functioning of such proteins; however, its molecular mechanism has not been fully understood [3]. Closer look on the self-assembly mechanism might be useful for studying different oligomeric states of ferritin, which may help in the development of recombinant ferritin-based vaccines.

In this work, recombinant protein complexes based on ferritin from H. pylori were obtained. The constructs contained apoferritin with different modifications of the N-terminal region. The resulting protein complex models were made using high-resolution structures from Protein Data Bank and their Dmax are 12 nm and 20 nm, respectively.

The expression and the purification were performed under similar conditions for each of the obtained protein constructs. Briefly, the expression was carried out using culture media at 37℃ for 3 hours after reaching OD of 0.6 (approximately 5 hours after autoinduction). The purification was performed using buffer containing 20 mM Tris base at pH 8.1, with immobilized metal chelate affinity chromatography (IMAC) and SEC. Recombinant protein complexes were obtained, consisting of 24 subunits according to negative staining transmission electron microscopy (NS-TEM) and small-angle X-ray scattering (SAXS).

The experiments, studying the possibility of exchange of subunits between two protein complexes under various conditions, were carried out. Briefly, the samples of first and second constructs, and their equimolar mixture were transferred to conditions with different pH. To control the results SEC and Blue native electrophoresis ([4]) were used. As a result, the stability of globules of recombinant protein complexes based on apoferritin was confirmed in a wide range of conditions (up to pH 12). Moreover, conditions for the disassembly of protein globules of modified apoferritin have been found; the resulting oligomeric states require further study.

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Literature list

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