Imaging with high spatial resolution is crucial in the development of science and technology nowadays. Manipulation of matter at nanometer scale and the ability to later or "in situ" verification of the results of this manipulation are very important in recent times, in which the trend in the development of science and technology is determined by the semiconductor industry to pursue smaller and smaller structures, more efficient, consuming less electricity ... the computer industry efforts translate into other areas of science such as biology or materials science. It turns out for some time now that the world seems to us quite simple and well known, until we go deeper, using imaging tools with increasingly higher magnification to understand the basics of its operation. Then it often turns out that the effect is not at all easy, nor always predictable and then we discover a whole new field of science. Particularly it is evident in biology, where the cells, formerly regarded as the basic building blocks of life, are today virtually whole "energy factories" in a complex organism, and themselves are made up of a large number of smaller biological structures. The objective of the project, entitled “"Water window" radiation for nanoimaging of biological objects and three-dimensional electron density reconstruction in bioengineering and material science applications” is to perform two- and three dimensional (2-D and 3-D) imaging in the “water window” spectral range of various objects, especially biological cells, with sub-micron to nanometer spatial resolution for variety of applications, especially biology, bioengineering, medicine, material science and nanotechnology. This 3-D imaging system will be based on recently developed “water window” soft X-ray (SXR) microscope, capable of resolving features with spatial resolution of approximately 60 nm, relatively short exposure times and extremely compact design. Long term goal of our project is to encourage scientists from various fields of science and technology to apply such 2-D and 3-D SXR imaging methods in their pursuits, by imaging of various objects, including biological samples, to gain new knowledge and insights about them. The results of the project allow development of high spatial resolution, compact, desk-top imaging tool, operating in the water window spectral range for applications in science and technology, to acquire more complete and complementary information about objects studied. The project will also help to expand our knowledge and experience in the field of biology, bioengineering, materials science and nanotechnology.

The work plan requires accomplishing a few basic research tasks listed below:
1. Numerical simulations of Fresnel zone plate – objective (lens) used in SXR "water window" imaging system for 2-D and 3-D image acquisition.
2. Development and optimization of the SXR imaging system to perform 2-D and 3-D imaging of various objects in the SXR water window spectral range to reconstruct the electron density of the objects and acquire additional information about the object available in the 3-D mode of operation.
3. SXR imaging of various, including biological samples to acquire additional, complementary information about samples, especially about internal structure of cells, which should be visible with a very good contrast due to natural contrast in the water window for biological material.
4. Acquiring projections for test objects and biological object, performing the numerical reconstruction of the 3-D electron density in the reconstruction volume from previously acquired projections and testing the reconstruction resolution.

These tasks allow achieving goals of this project and perform 2-D and 3-D imaging of various objects in the water window spectral range to gain a more complete and complementary information about the objects studied with high resolution. This plan is based on our experience with SXR microscopy and EUV tomography of transient gaseous objects.

In the proposed project we are planning to develop very compact water window imaging system and employ it to acquire 2-D and 3-D images of micro and nanoscale objects to gain complementary information about them, often impossible to obtain using other imaging techniques. Our system does not use large scale facilities, but will be based on compact, laser plasma SXR source employing a double stream gas puff target.

Imaging in the “water window” spectral range, which is the subject of this proposal, allows obtaining spatial resolution better than 100nm, superior to the visible light microscopes, fast exposure times and optical contrast in the short wavelength spectral region, which allows assessing additional information about object. Moreover, water window radiation is particularly suitable for biological imaging due to natural optical contrast. High contrast in this range is obtained due to a significant difference in absorption (attenuation of radiation transmitted through the objects) of biological specimen constituents: water (oxygen) and carbon. Water has relatively small attenuation/absorption coefficient, while carbon has much higher value, which provides very good optical contrast in this specific wavelength range.

Water window imaging uses radiation instead of high energy particles, allows the observation of very delicate samples, which in the case of the use of electron microscopy may be destroyed or disrupted by high-energy electron beam. Water window imaging also allows observation of the samples coated with photoresist sensitive to electron beam exposure, which cannot be observed by electron microscopy without affecting the properties of the photoresist. Another advantage is the lack of need for special preparation of the samples for water window, which is necessary in the case of other imaging techniques such as electron microscopy, to dissipate surface charges.

Short wavelength and high energy photons are generated often using synchrotrons or free electron lasers. These, although state-of-the-art systems are large-scale installations (facilities), expensive to maintain, difficult to access and, unfortunately, impossible to commercialize, making impossible in the future for the direct transfer of the technology to science, industry and everyday life. Thus, we focus our effort to employ compact sources in imaging experiments and our goal in this project is to use a compact source in our imaging experiments to overcome above limitations. Such system, dedicated for 3-D imaging in the water window spectral range, will allow for acquisition of additional, complementary information about the objects, which cannot be obtained directly from the microscopic images. This may allow discovering new phenomena, new effects, which in turn will allow for the development of variety of scientific disciplines, such as biology, material science and nanotechnology.