The goal of the GaboScope project is to enable experimental and numerical development of holographic Gabor microscopy (HGM) for dynamic imaging of live cells in uniquely large volumes without implementation of lenses and fluorescent markers. Lensless HGM has advantages in terms of (1) compact setups composed of light source, sample and camera, (2) in-line Gabor hologram recording with effective utilization of detector bandwidth and numerical back-propagation to the sample plane, (3) very large depth of focus not limited by numerical aperture of microscopic objective. Due to attractive features lens-free HGM methods found a vast number of applications in various branches of science and technology, e.g., digital histopathology, sperm motility analysis and holographic liquid biopsy for parasite detection in bodily fluids. Interestingly, all beneficial attributes of HGM are the sources of its limitations.

1. Distances between light source, sample and detector determine HGM system magnification and field of view – desirable compact layouts utilize strongly spherical illumination, which is not accounted for in classical plane wave reconstruction of Gabor holograms. Additionally, minimization of sample-camera separation distance results in advantageous field of view augmentation (thus enlarging the holographically rendered volume of interest), however it causes undesired low magnification and undersampling (too big pixel size). Moreover, the quality of hologram numerical propagation suffers from too severe simplification of diffraction integral (so-called weak object assumption) and is susceptible to densely packed objects. High depth of focus is very favorable in dynamic imaging of diversified objects in large volumes, however strong scattering and overlapping of the information coming from various planes is a problematic issue. Specialized algorithms for autofocusing, segmentation and 4D object tracking are thus indispensable. In the GaboScope project we plan to incorporate novel data analysis solutions to advance HGM through developing new numerical propagation engine which accounts for spherical wave-front, multiple-scattering and allows accurate dynamic 4D tracking of high number of diversified objects in large volumes of interest (thousands of cubic millimeters with micrometer resolution).

2. Numerical reconstruction of Gabor hologram, implemented upon back-propagation of the recorded optical field from hologram plane to the object plane, comes with the downside of twin-image effect, which is a consequence of in-line recording of interference between object scattered and reference unscattered wavefronts. What is more, back-propagation distance to the object plane of focus is not known, thus automatic numerical focusing methods are of great interest. Coherent illumination calls for semitransparent objects and generates speckle noise and other artifacts which decrease the hologram quality and jeopardize its effective numerical reconstruction. Robust noise removal, twin-image suppression and autofocusing constitute very interesting open problems in lensless HGM. Within the GaboScope project we plan to develop new single-shot advanced algorithmic solutions with increased signal to noise ratio (in contrast to popular time-consuming multi-frame techniques with limited temporal resolution). Exemplifying results of novel two-wavelength twin image removal scheme are presented in Fig. 1.

3. Innovative numerical approaches originally developed under the GaboScope project will aid two new experimental setups of lensless HGM: (1) two-sourced system with enhanced reconstruction quality and resolution and (2) spherical illumination based optical tomography with translation of the object (e.g., enabling crucial 3D structural tomographic imaging of live spermatozoa or cardiomyocytes translated by optical tweezers).

Project methodology considers utilizing numerical simulation to gain full control over the process of hologram formation, recording and reconstruction in designed lensless HGM setups. Computational models will enable developing both specialized and versatile algorithms for holographic data analysis in full volume of interest with minimized errors and augmented accuracy of dynamic 4D object imaging and characterization. Final verification of implemented numerical methods and developed HGM systems will be conducted in experimental conditions in cooperation with partner institutions of excellent track record in biomedical imaging research (Mossakowski Medical Research Center PAS, The Arctic University of Norway, University of Valencia, Harvard University, Nanjing University of Science and Technology).

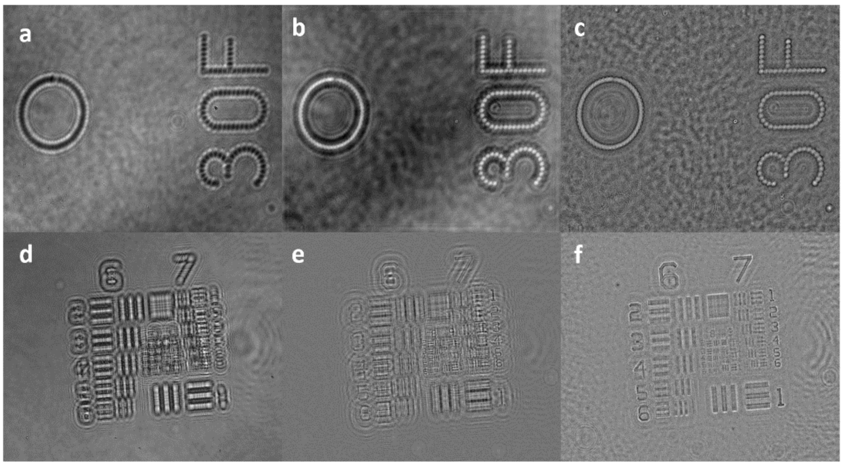

Fig. 1. Preliminary results: (a) lensless Gabor hologram of engravings made on progressive lens, (b) its numerical reconstruction to the object plane spoiled with twin-image and background errors, (c) preliminary results of our innovative reconstruction with background and twin-image suppression; (d) lensless Gabor hologram of USAF resolution test target, (e) its reconstruction with twin-image errors, (f) preliminary reduction of twin-image errors by our new single-shot two-wavelength technique.