

High-speed holographic fluorescence microscopy system with submicron resolution

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Overview of the developed high-speed holographic fluorescence microscopy system for scanless 3D measurement with submicron resolution. Credit: National Institute of Information and Communications Technology (NICT), Tohoku University, Toin University of Yokohama, Japan Science and Technology Agency (JST)

The National Institute of Information and Communications Technology (NICT), Tohoku University, Toin University of Yokohama, and Japan



Science and Technology Agency (JST) have succeeded in developing a scanless high-speed holographic fluorescence microscopy system with submicron resolution for a 3-D space. The system is based on digital holography.

The developed microscopy system has an algorithm to acquire 3-D information of fluorescent objects toward scanless 3-D measurement in less than 1 millisecond. Scanless 3-D sensing with submicron resolution and color-multiplexed holographic fluorescence imaging have been demonstrated using the algorithm. The microscopy system will be further developed to achieve holographic 3-D motion-picture sensing of specimens with incoherent light.

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The scanless high-speed holographic fluorescence microscopy system shown in Figure 1 was constructed based on digital holography and is applicable to the sensing of incoherent light such as fluorescence light and natural light. The developed algorithm enables the adoption of a phase modulator to generate two phase values, which is expected to increase the measurement speed. Submicron resolution for a 3-D space was successfully demonstrated using fluorescent objects with a diameter of 0.2 micron.

The <u>experimental results</u> shown in Figure 2 indicate that the developed microscopy system achieves 3-D sensing of nanoparticles and has submicron resolution quantitatively for a 3-D space. Scanless 3-D measurement in less than 1 millisecond is achievable by using the algorithm with either a ferroelectric liquid crystal on silicon (FLCOS) or an electro-optic (EO) device. Color-multiplexed holographic fluorescence imaging with the algorithm and only four exposures has also been demonstrated by combining the proposed algorithm and



computational coherent superposition (CCS). The number of exposures is reduced by the <u>algorithm</u>, and the number of photons per hologram is increased even for ultimately weak <u>light</u>.



Upper left: experimental results of 3D sensing for fluorescent particles with a diameter of 0.2 micron. Upper right: x-z image of the reconstructed particle marked by the violet arrow. Bottom left: plots of the particle marked by the violet arrow along the x- and y-axes. Bottom right: plots of the particle marked by the violet arrow along the z-axis. Credit: National Institute of Information and Communications Technology (NICT), Tohoku University, Toin University of Yokohama, Japan Science and Technology Agency (JST)

More information: Tatsuki Tahara et al, Two-step phase-shifting



interferometry for self-interference digital holography, *Optics Letters* (2021). DOI: 10.1364/OL.414083

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