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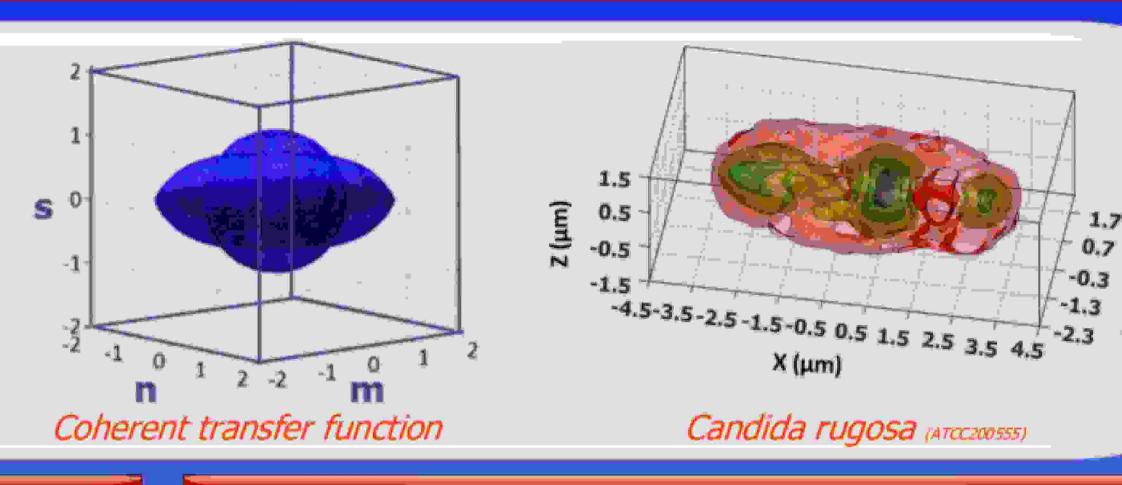
Development of integrated dual-mode tomography for free-floating single live cell label-free analysis

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- > This study focused on the development of integrated dual-mode tomography (IDT) system, which combines both beam rotation and full-angle sample rotation tomographic methods.
- The spatial frequencies corresponding to the beam rotation and the full-angle sample rotation methods are collected and combined. Consequently an 'UFO' (unidentified flying object) like shaped enlarged coherent transfer function is obtained.
- Label-free three-dimensional refractive index profile of live single cell is generated at subcellular level without using complex algorithms.

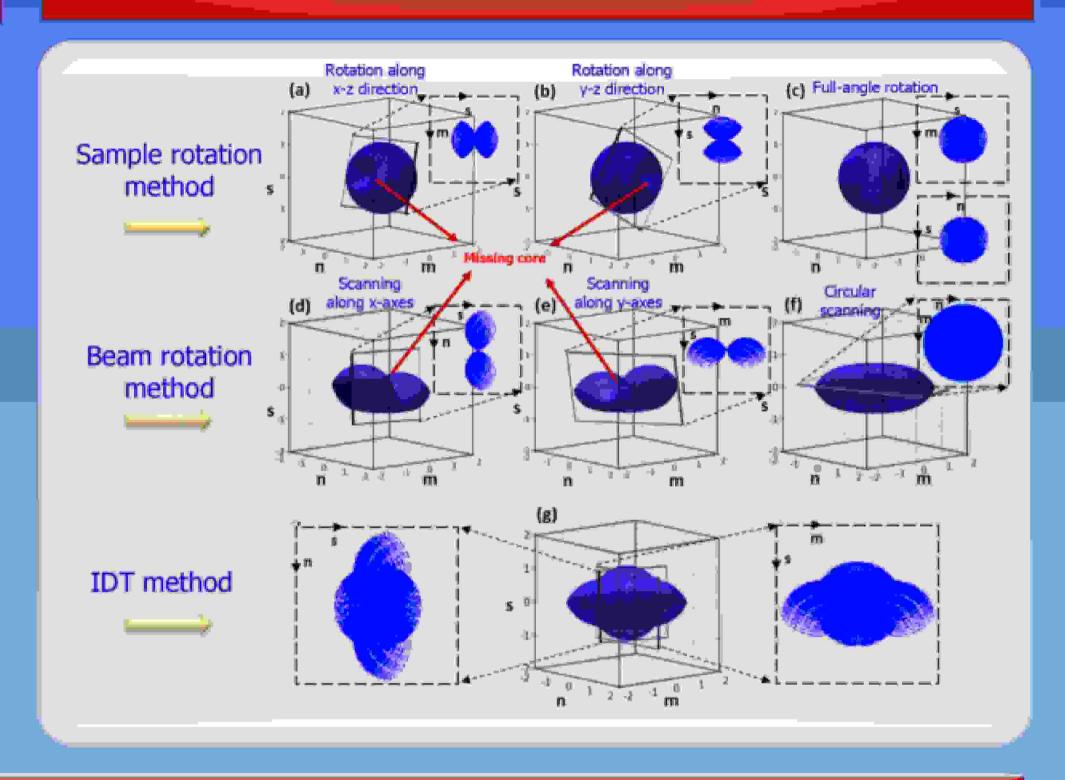


Conceptual Representation

(b) Full-angle sample rotation

- > IDT system is developed by combining holographic optical tweezer (HOT) with synthetic aperture-digital holographic microscopy (SA-DHM) imaging system
- ➤In HOT system, a series of computer-generated holograms are designed based on Gerchberg-Saxton algorithm to generate two trapping points namely P₁ (fixed trap) and P₂ (rotation trap) respectively.
- Using the HOT system, the sample is rotated in fullangle directions and the corresponding holograms are recorded using DHM.
- The probe beam of the imaging system is rotated to collect the beam rotation holograms.
- The recorded holograms are reconstructed and mapped to Ewald's sphere to generate the threedimensional RI profile of the sample

Coherent transfer functions (CTF)



Beam rotation results:

Reconstructed phase of Candida rugosa at

different illumination angles

(根)建设

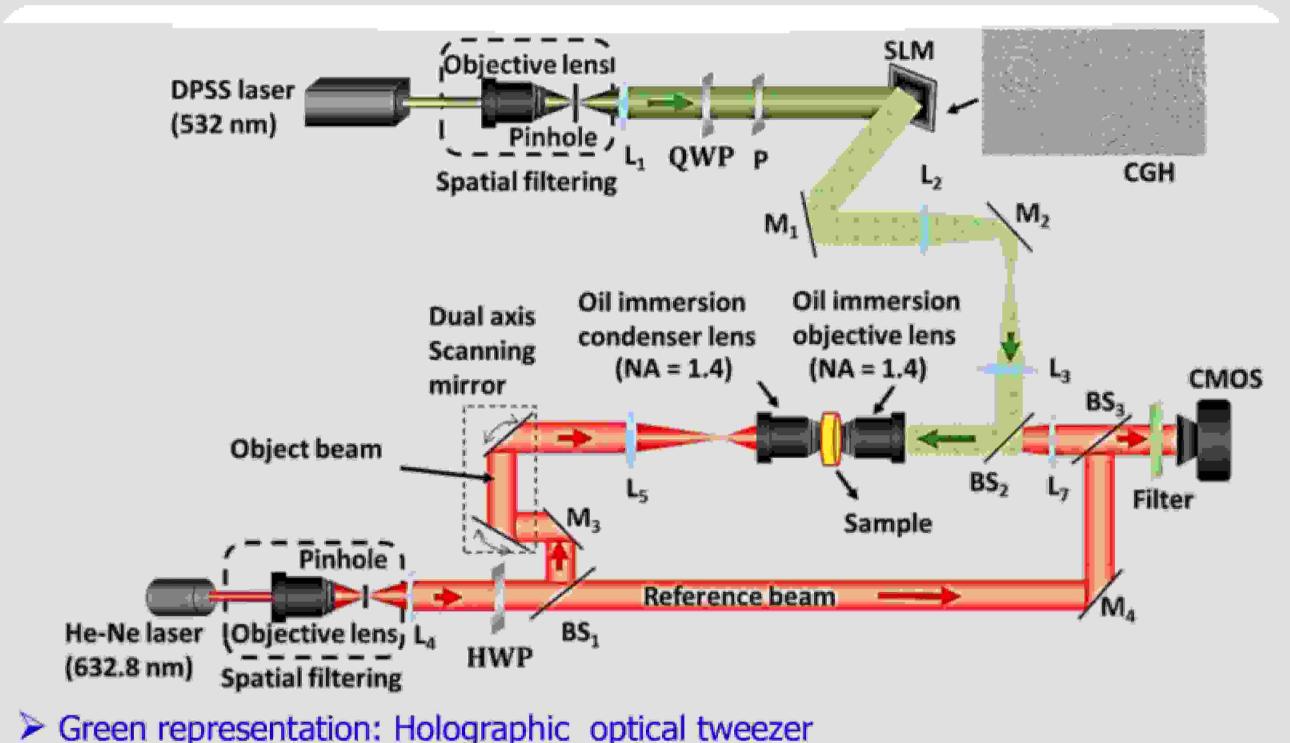
 $\theta_{BR-x} = -60^{\circ}$ $\theta_{BR-x} = 60^{\circ}$ $\theta_{BR-y} = -60^{\circ}$ $\theta_{BR-y} = 60^{\circ}$

Scale bar: 3 µm

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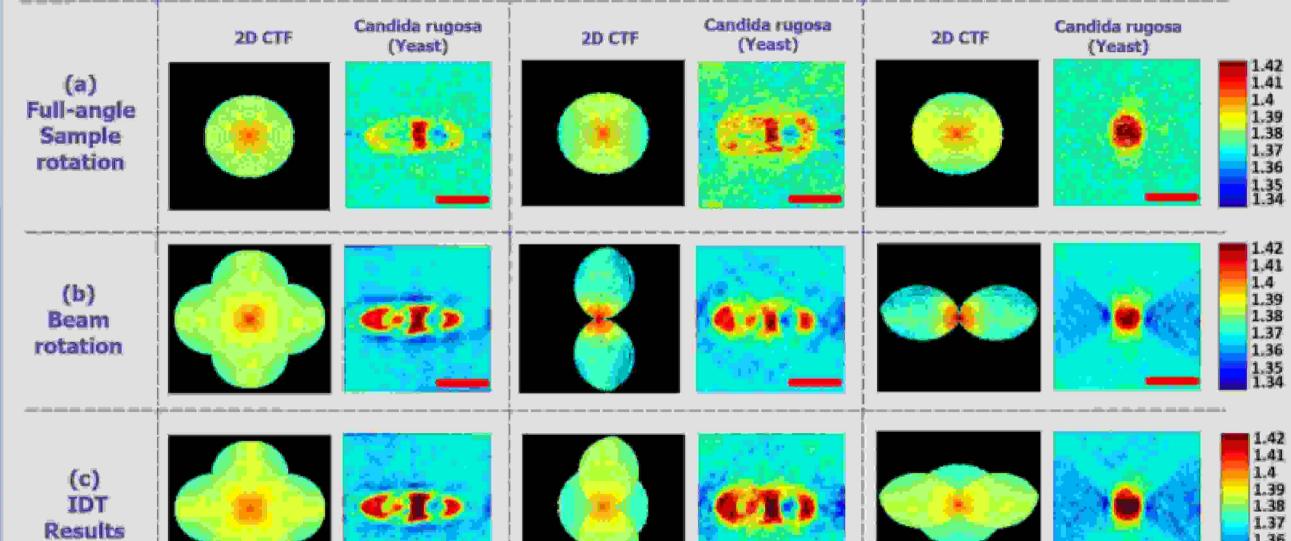
➤ Axial resolution= 310 nm

Integrated dual-mode tomography – Experimental sysytem



- Green representation: Holographic optical tweezer
- Red representation: Synthetic-aperture holographic microscopy

2D CTF 2D CTF (Yeast)



Experimental results

Reconstructed tomography comparisons

XY view $(K_x, K_y, K_z = 0)$ YZ view $(K_x = 0, K_y, K_z)$ XZ view $(K_x, K_y = 0, K_z)$ XZ view $(K_x, K_y = 0, K_z)$

IDT system resolution

Sample rotation results:

Reconstructed phase of Candida rugosa at

different rotation angles

 $\theta_{SR-x}=60^{\circ}$

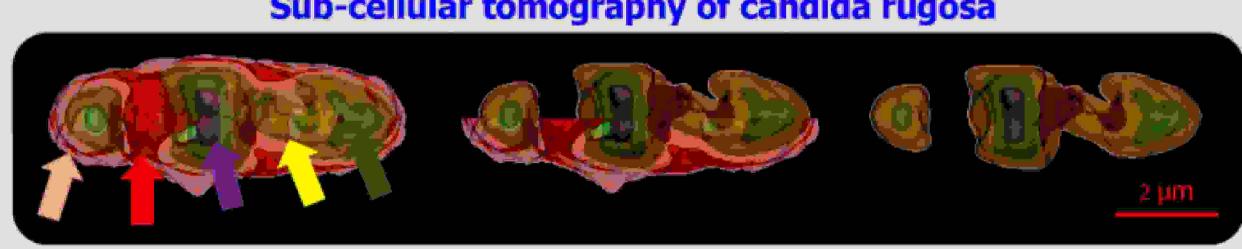
 $\theta_{SR-x}=90^{\circ}$

 θ_{SR-x} =30°

Lateral fitting Axial fitting

Lateral resolution = 150 nm

Sub-cellular tomography of candida rugosa



Arrows indicates the subcellular organelles of candida rugosa pink : cell wall, violet: nucleus, green: mitochondria, red: vacuole, yellow: cytoplasm

-Conclusions

- A novel IDT system is developed and demonstrated for free-floating single living cell analysis.
- ➤ A novel experimental coherent transfer function ('UFO') is reported.
- Free-floating single live candida rugosa is imaged by the developed IDT imaging system with isotropic superresolution
- >IDT imaging system is a potential tool for free-floating single live sample analysis at subcellular level.

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