

# 计算光学成像与 光信息处理技术前沿

#### (第9讲)

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# 光学衍射层析与强度 衍射层析成像

Optical diffraction tomographic imaging and intensity diffraction tomographic imaging

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## **Computational** microscopy



#### Phase of a image



#### Phase of a object



#### 3D phase imaging ?

![](_page_5_Figure_1.jpeg)

#### QPI vs ODT (optical diffraction tomography)

![](_page_6_Picture_1.jpeg)

QPI: 2.5D optical path length Profile

C. Zuo, et. al., Opt Express, 21 24060-24075 (2013).

#### ODT: true 3D refractive index Volume

Volunne

J Li, et. al. Biomed. Opt. Express 9, 2526-2542 (2018)

### X-ray in biomedical imaging

![](_page_7_Picture_1.jpeg)

![](_page_7_Picture_2.jpeg)

1895 X-rays and Uranium Rays, wedding ring

Marie Curie and the Discovery of Radioactivity

#### Brief review of optical diffraction tomography(ODT)

X-ray computed tomography(CT) & Application

![](_page_8_Picture_2.jpeg)

## Computed tomography (CT)

http://www.merckmanuals.com/home/special-subjects/common-imaging-tests/computed-tomography-ct

### Integration projection & Radon transform

![](_page_10_Figure_1.jpeg)

![](_page_10_Picture_2.jpeg)

#### Fourier central slice theorem & back projection

![](_page_11_Figure_1.jpeg)

### Fourier central slice theorem

![](_page_12_Figure_1.jpeg)

![](_page_13_Picture_0.jpeg)

### Back projection algorithm

![](_page_14_Picture_1.jpeg)

![](_page_14_Picture_2.jpeg)

ramp filter

![](_page_14_Figure_4.jpeg)

#### Hamming

![](_page_14_Figure_6.jpeg)

Cosine

#### Optical diffraction tomography

![](_page_15_Figure_1.jpeg)

### Diffraction tomography

![](_page_16_Figure_1.jpeg)

Born approximation  $U^{(s)}(\vec{R}) \ll U^{(i)}(\vec{R})$  $U(\vec{R}) \approx U^{(i)}(\vec{R})$  Rytov approximation  $U^{(s)} \approx U^{(i)}(\vec{R}) \ln \left( \frac{U(\vec{R})}{U^{(i)}(\vec{R})} \right)$ 

 $U^{(i)}\left(\vec{R}\right) = \exp(ik_0s_0\cdot\vec{R})$  $U\left(\vec{R}\right) = U^{(i)}\left(\vec{R}\right) + U^{(s)}\left(\vec{R}\right)$ 

 $\nabla^{2} U(\vec{R}) + k_{0}^{2} n(\vec{R})^{2} U(\vec{R}) = 0$  $\nabla^{2} U^{(i)}(\vec{R}) + k_{0}^{2} n(\vec{R})^{2} U^{(i)}(\vec{R}) = 0$ 

 $(\nabla^{2} + k_{0}^{2})U^{(i)}(\vec{R}) = 0$   $(\nabla^{2} + k_{0}^{2})U^{(s)}(\vec{R}) = F(\vec{R})U(\vec{R})$   $F(\vec{R}) = -k_{0}^{2}[n(\vec{R})^{2} - 1]$  $F(\vec{R}) \text{ Scattering} \text{ potential}$ 

### Fourier diffraction theorem

$$\widehat{F}(U,V,W) = \frac{ik_z}{\pi} \widehat{U}^{(s)}(u,v;z^+=0)$$

In words eq. (15) shows that some of the *three*dimensional Fourier components of the scattering potential may be immediately determined from the knowledge of the *two-dimensional* Fourier components of the scattered field in the two planes  $z = z^+ > Z$  and  $z = z^- < 0$ . It is now

where	
	$U = u - k_{\rm o} p_{\rm o} , \qquad )$
	$V = v - k_0 q_0,$
	$W^{\pm} = \pm w - k_0 m_0,  )$
and	
	$w = (k_0^2 - u^2 - v^2)^{\frac{1}{2}} .$

![](_page_17_Figure_4.jpeg)

![](_page_17_Figure_5.jpeg)

#### 3D K-space of Scattering potential

![](_page_18_Figure_1.jpeg)

# 3D OTF for transmission microscope

![](_page_18_Figure_3.jpeg)

N. Streibl, "Three-dimensional imaging by a microscope," JOSA A 2, 121-127 (1985).

#### Optical diffraction tomography

	$\widehat{F}_{\phi_0}(k_{\mathrm{x}},k_{\mathrm{z}}) = A \cdot \sqrt{rac{1}{2\pi}} \widehat{P}_{\phi_0}(k_{\mathrm{Dx}})$		
	Fourier Slice Theorem (equation 1.10)	Fourier Diffraction Theorem (equation 4.24)	
${ m Sinogram} {\widehat P}_{\phi_0}(k_{ m Dx})$	Fourier transform of projections $\widehat{P}_{\phi_0}(k_{\mathrm{Dx}})$	Fourier transform of complex scattered waves $\widehat{U}_{\mathrm{B},\phi_0}(k_{\mathrm{Dx}})$	
$\begin{array}{c} \text{Factor} \\ A \end{array}$	A = 1	$A = -\frac{2ik_{\rm m}M}{a_0}\exp(-ik_{\rm m}Ml_{\rm D})$	
Coordinates $(k_{\rm x}, k_{\rm z})$ sliced at $\phi_0$	$egin{aligned} k_{\mathrm{x}} &= k_{\mathrm{Dx}} \ k_{\mathrm{z}} &= k_{\mathrm{t}} = 0 \ & (\mathrm{straight\ line}) \end{aligned}$	$\begin{split} k_{\rm x} &= k_{\rm Dx} \\ k_{\rm z} &= \sqrt{k_{\rm m}^2 - k_{\rm Dx}^2} - k_{\rm m} \\ (\text{semicircular arc}) \end{split}$	
Fourier space $\widehat{F}(\mathbf{k})$ coverage (180°)		R <sup>Z</sup> R	

![](_page_20_Figure_1.jpeg)

Sample rotation

#### Sample rotation

![](_page_21_Figure_2.jpeg)

![](_page_22_Figure_1.jpeg)

Beam scanning

![](_page_23_Figure_1.jpeg)

Beam scanning

### Diffraction tomography result

DHM phase measurement

Original 3D Fourier

iterative nonnegative constraint

![](_page_24_Figure_4.jpeg)

![](_page_24_Picture_5.jpeg)

## Results

![](_page_25_Figure_1.jpeg)

## Commercial product of ODT

![](_page_26_Picture_1.jpeg)

PRODUCT	APPLICATIONS	PUBLICATIONS
NEWS & EVENTS	SUPPORT	COMPANY

#### Revolutionary holotomographic (3D holographic) microscopy opens new era for label-free live cell imaging

Cellular analysis plays a crucial role in a wide variety of research and diagnostic activities in the life science. However, the information available to researchers and clinicians is limited by current microscopy techniques. An innovative new tool – Holotomographic microscopy – can overcome many of these limitations and open new vistas for researchers and clinicians to understand, diagnose and treat human diseases.

Holotomographic Microscopy – New era of microscopy Tomocube's holotomography series utilize optical diffraction tomography (ODT), which enables users to quantitatively and noninvasively investigate biological cells and thin tissues. ODT reconstructs the 3D refractive index (RI) distributions of live cells and by doing so, provides structural and chemical information about the cell including dry mass, morphology, and dynamics of the cellular membrane.

![](_page_26_Picture_6.jpeg)

## Commercial product of ODT

![](_page_28_Picture_1.jpeg)

#### New Live T Cell Assay

#### Multi-parametric, non-invasive immuno-therapy analysis

Learn more

Take a tea break with Emma and discover how to quantify cell cytotoxicity in 96 WP format label-free. Register here.

#### **Applications**

A selected overview of Nanolive's top applications

#### Live T Cell Assay

![](_page_29_Picture_3.jpeg)

#### Mitochondria & Cell Metabolism

![](_page_29_Figure_5.jpeg)

#### Cytotoxicity

![](_page_29_Figure_7.jpeg)

https://www.nanolive.ch/

## How non-interferometric?

#### Transport-of-intensity diffraction tomography (TIDT)

![](_page_31_Figure_1.jpeg)

E Wolf. Opt. Commun. 1, 153–156 (1969).

#### Transport-of-intensity diffraction tomography (TIDT)

#### Fourier diffraction theorem for a limit-aperture system

![](_page_32_Picture_2.jpeg)

Microscopic imaging: partial spherical cap bounded by the lens aperture

E Wolf. Opt. Commun. 1, 153–156 (1969).

#### 3D phase imaging ?

![](_page_33_Picture_1.jpeg)

## Lensless TIE microscopy

![](_page_34_Picture_1.jpeg)

#### Lensless TIE microscope

C. Zuo, J. Sun, J. Zhang, Y. Hu, and Q. Chen, Optics Express 23, 14314-14328 (2015).

## Lensless TIE microscopy

![](_page_35_Figure_1.jpeg)

C. Zuo, J. Sun, J. Zhang, Y. Hu, and Q. Chen, Optics Express 23, 14314-14328 (2015).
# Lensless TIE microscopy

# Digital refocusing

C. Zuo, J. Sun, J. Zhang, Y. Hu, and Q. Chen, Optics Express 23, 14314-14328 (2015).

# Lensless TIE tomography



Change Illumination angle ( ≈±45°)



Fill the 3D Fourier Space of the object

C. Zuo, J. Sun, J. Zhang, Y. Hu, and Q. Chen, Optics Express 23, 14314-14328 (2015).

# Lensless TIE tomography

The uterus of Parascaris equorum

Phase tomography

Absorption tomography







C. Zuo, J. Sun, J. Zhang, Y. Hu, and Q. Chen, Optics Express 23, 14314-14328 (2015).

Optics and Lasers in Engineering 95 (2017) 26-34

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(a) 20µm (b) 465 (g) Contents lists available at ScienceDirect Optics and Lasers in Engineering 1.46 ELSEVIER journal homepage: www.elsevier.com/locate/optlaseng 1.455 Optical diffraction tomography microscopy with transport of intensity -10µm -5µm CrossMark .45 equation using a light-emitting diode array (c) (d) Jiaji Li<sup>a,b</sup>, Qian Chen<sup>b</sup>, Jialin Zhang<sup>a,b</sup>, Zhao Zhang<sup>a,b</sup>, Yan Zhang<sup>a,b</sup>, Chao Zuo<sup>a,b,\*</sup> <sup>a</sup> Smart Computational Imaging (SCI) Laboratory, Nanjing University of Science and Technology, Nanjing, Jiangsu 210094, China
<sup>b</sup> Jiangsu Key Laboratory of Spectral Imaging & Intelligent Sense, Nanjing University of Science and Technology, Nanjing, Jiangsu 210094, China -----TIE (a) (b) (c) -2µm 2µm 1.45 (e) (f) 10µm 0 10 -10 X(um) (d) (f) -Idea 20µm 0.6 5µm 10µm 0.4 1.45 0.2 15 -10 0 5 10 X(um) Camera 11 11 ... ••• Tube Lens 1 11  $\theta_x = 0^\circ, \theta_y = 0^\circ$ ... 11  $\theta_x = 37^\circ, \theta_y = 0^\circ$ **3D** Fourier spectrum Objective  $\theta_{\gamma} = 23^{\circ}, \theta_{\gamma} = 0^{\circ}$ 벁 Pupil Phase retrieval by LED element Objective non-negative constraint Sample 10µm FT and mapping FT and mapping FT and mapping Condenser Lens LED Array

\_

#### Transport of intensity equation



# TIE Phase retrieval(noise-free)



# TIE Phase retrieval(Gauss noise 0.002)



# About Setup



#### Schematic diagram of imaging system



#### The procedure of diffraction tomography imaging



#### Positional misalignment correction for LED array



#### Comparison results of phase measurement between DHM and TIE



#### The comparison images of final 3D structure and Fourier spectrum



#### Tomographic reconstruction of polystyrene bead







Imaging results of the fixed sample of Pandorina morum

# 3D refractive index tomograms rendering of lung cancer cell and HeLa cell



# Iterative method for Diffraction tomography



Horstmeyer, Roarke, et al. Optica 3.8 (2016): 827-835.



# FPDT image formation





Free-space: semi-sphere in 3D Fourier space



Microscopic imaging: partial spherical cap bounded by the lens aperture



# On-axis bright-field illumination

Synthetic aperture using bright-field images

Synthetic aperture using both bright- and dark-field (0.9NA) images





 $\hat{f}(\mathbf{k})$ 

 $k_y$ 







 $NA_{obj} = 0.4 \ \lambda = 507 \text{nm}$ 







Full-FOV 3D quantitative RI reconstruction of unstained blood smear (~ 20,000 RBC)



Full-FOV 3D quantitative RI reconstruction of unstained HeLa cells

#### Coherent / partially coherent ODT



# Theory

- 3D Fluorescence deconvolution microscopy
- 3D Optical diffraction tomography microscopy
  3D Deconvolution phase microscopy
  - 3D Deconvolution phase microscopy

P. Sarder and A. Nehorai, "Deconvolution methods for 3-D fluorescence microscopy images," IEEE Signal Processing Magazine 23, 32-45 (2006).

E.Wolf, "Three-dimensional structure determination of semi-transparent objects from holographic data," Optics Communications 1, 153-156 (1969).

N. Streibl, "Three-dimensional imaging by a microscope," JOSA A 2, 121-127 (1985).

#### **3-D** Fluorescence Microscopy



P. Sarder and A. Nehorai, "Deconvolution methods for 3-D fluorescence microscopy images," IEEE Signal Processing Magazine **23, 32-45 (2006).** 

3D Optical diffraction tomography microscopy

Volume 1, number 4

OPTICS COMMUNICATIONS

September/October 1969

#### THREE-DIMENSIONAL STRUCTURE DETERMINATION OF SEMI-TRANSPARENT OBJECTS FROM HOLOGRAPHIC DATA \*

Emil WOLF

Department of Physics and Astronomy, University of Rochester, Rochester, N.Y. 14627, USA

Received 11 August 1969

E.Wolf, "Three-dimensional structure determination of semi-transparent objects from holographic data," Optics Communications 1, 153-156 (1969).

#### Emil Wolf



#### Emil Wolf

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Czech-American physicist

Emil Wolf was a Czech-born American physicist who made advancements in physical optics, including diffraction, coherence properties of optical fields, spectroscopy of partially coherent radiation, and the theory of direct scattering and inverse scattering. Wikipedia

Born: July 30, 1922, Prague, Czechia Died: June 2, 2018, Rochester, NY Doctoral students: Girish Agarwal; M. Suhail Zubairy Research interests: Optics, Coherence

#### Emil Wolf, pioneer of optical physics, dies at 95

June 4, 2018



Emil Wolf was one of the most recognized optical scientists of his generation and served on the Rochester faculty for more than 50 years. (University of Rochester photo / Richard Baker)





#### Research Article

Biomedical Optics EXPRESS

#### Three-dimensional tomographic microscopy technique with multi-frequency combination with partially coherent illuminations

JIAJI LI,<sup>1,2,3</sup> QIAN CHEN,<sup>1,2,4</sup> JIASONG SUN,<sup>1,2,3</sup> JIALIN ZHANG,<sup>1,2,3</sup> JUNYI DING,<sup>1,2,3</sup> AND CHAO ZUO<sup>1,2,3,\*</sup>

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3D intensity spectrum



3D phase / amplitude transfer function

Transport-of-intensity diffraction tomography (TIDT)

[Li, et. al. Biomed. Opt. Express 9, 2526-2542 (2018)

#### 3D Deconvolution phase microscopy



N. Streibl, "Three-dimensional imaging by a microscope," JOSA A 2, 121-127 (1985).





#### 3D OTF for coherent illumination source


#### Rendering result of 3D OTF





N. Streibl, "Three-dimensional imaging by a microscope," JOSA A 2, 121-127 (1985).

#### Partially coherent 3D imaging

 $WOTF(\mathbf{u}) \equiv TCC(\mathbf{u}, \mathbf{0})$  $= \iint S(\mathbf{u}') H(\mathbf{u}' + \mathbf{u}) H^*(\mathbf{u}') d\mathbf{u}'$  $H(\rho, l) = \int P(\rho) e^{jkz\sqrt{1-\lambda^2\rho^2}} e^{-j2\pi z l} dz$  $= P(\rho) \delta\left(l - \sqrt{\left(\frac{1}{\lambda}\right)^2 - \rho^2}\right)$ 

## Mapping of 3D Ewald sphere





Fig. 3. (a) Computer plot of 3-D PTF under annular illumination;
(b) perspective plot of the PTF value of ρ(= 1/r) and ζ(= 1/z).





#### 3D OTF for optical microscope



N. Streibl, "Three-dimensional imaging by a microscope," JOSA A 2, 121-127 (1985).

#### Partially coherent 3D imaging











Volume Viewer

#### **Optical transport of bacteria: Real-time reconfigurable trajectory**



# Annular-illumination ODT





#### 3D rendering of Pandorina

I.4 NAill + I.4 NAobj 100X MO; Lateral resolution 200nm; axial resolution 650 nm.

J Li, et. al. Biomed. Opt. Express 9, 2526-2542 (2018)

# Annular-illumination ODT



I.4 NAill + I.4 NAobj 100X MO; Lateral resolution 200nm; axial resolution 650 nm.

High-speed in vitro intensity diffraction tomography

Brief review of IDT (intensity diffraction tomography)



# System illumination unit





Programmable Square LED array (1.25mm) ~100\$ Bright annular LED from adafruit ~10\$

# System setup (System illustration of aIDT)









Illumination unit

Photography of setup

# Key parameters about illumination unit



24 Frames or 12 Frames or 8 Frames (More different size is available)

Maximum speed 800Kbps (33KHz for each LED also depends on Microcontroller)

Adjustable for objective with different NA

## Demonstration of aIDT



10Hz for each imaging volume

#### Experimental result of stained Algae

0.05



Absorption



- 0.05

#### LED illumination calibration and comparative result of diatom



#### **3D** Reconstruction results



Diatom region I

Diatom region2

## RI tomography of Surirella spiralis



## 3D reconstruction of diatom



## Fixed cheek cell



#### **RI stack of cheek cell**



1.33

# Fixed C. Elegant results



Refractive index stacks





Depth color coding



Morphological analysis of C. elegans

# C. Elegans RI slice



RI slice of sample at t = 0 sec

#### Depth color rendering of C. Elegans





Time lapse results of *C*. *Elegans* 

#### Transport of intensity equation



### For further details, please refer to:



Transport of intensity equation: a tutorial

C Zuo, J Li, J Sun, Y Fan, J Zhang, L Lu, R Zhang, B Wang, L Huang, Q Chen Optics and Lasers in Engineering, 106187, 2020

## Computational imaging



第 40 卷 第 1 期	光学学报	Vol. 40, No. 1
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・特邀综述・

#### 深度学习下的计算成像:现状、挑战与未来

#### 左超<sup>1,2</sup>,冯世杰<sup>1,2</sup>,张翔宇<sup>1,2</sup>,韩静<sup>2</sup>,陈钱<sup>2</sup>\*

<sup>1</sup>南京理工大学电子工程与光电技术学院,智能计算成像实验室(SCILab), 江苏南京 210094;

2南京理工大学江苏省光谱成像与智能感知重点实验室, 江苏南京 210094

摘要 近年来,光学成像技术已经由传统的强度、彩色成像发展进入计算光学成像时代。计算光学成像基于几何 光学、波动光学等理论对场景目标经光学系统成像再到探测器采样这一完整图像生成过程建立精确的正向数学模 型,再求解该正向成像模型所对应的"逆问题",以计算重构的方式来获得场景目标的高质量图像或者传统技术无 法直接获得的相位、光谱、偏振、光场、相干度、折射率、三维形貌等高维度物理信息。然而,计算成像系统的实际成 像性能也同样极大程度地受限于"正向数学模型的准确性"以及"逆向重构算法的可靠性",实际成像物理过程的不 可预见性与高维病态逆问题求解的复杂性已成为这一领域进一步发展的瓶颈问题。近年来,人工智能与深度学习 技术的飞跃式发展为计算光学成像技术开启了一扇全新的大门。不同于传统计算成像方法所依赖的物理驱动,深 度学习下的计算成像是一类由数据驱动的方法,它不但解决了许多过去计算成像领域难以解决的难题,还在信息 获取能力、成像的功能、核心性能指标(如成像空间分辨率、时间分辨率、灵敏度等)上都获得了显著提升。基于此, 首先概括性介绍深度学习技术在计算光学成像领域的研究进展与最新成果,然后分析了当前深度学习技术在计算 光学成像领域面临的主要问题与挑战,最后展望了该领域未来的发展方向与可能的研究方向。

关键词 成像系统;计算成像;深度学习;光学成像;光信息处理

**中图分类号** O436 文献标志码 A

doi: 10.3788/AOS202040.0111003

## Computational imaging



# 出国留学情况简介

- PostDoc@SCILab 智能计算成像实验室
- 2018-2019国家留学基金委(CSC)资助
- •为期12个月
- •美国波士顿大学(Boston University, BU)
- 电子工程学院-计算成像系统实验室(CISL)
美国 马萨诸塞州







Tian Lab Research People Publications Teaching Open Source News Openings

#### Welcome to Computational Imaging Systems Lab at Boston University!

computationalMicroscopy PhaseRetrieval CompressiveImaging CompressiveImaging CompressiveImaging CompressedSening CompressedSening Holography Onlice

#### What's new $\rightarrow$

Benjamin Wong wins UROP award September 27, 2019

Joe, Hao, and Jiabei join CISL for PhD, Welcome! August 30, 2019

Joe defended MS thesis

August 16, 2019

Yunzhe presents at SPIE Defense + Commercial Sensing Congress April 25, 2019

Alex, Waleed present at OSA Biophotonics Congress April 24, 2019

# Boston生活照片

## 第一次踏上踏上美利坚的土地,内心充满着对 未知的恐慌,也充满了 对未来的期待



# 无处不在的名校印记













## Boston 雪景









SCI Lab@ NJUST

- Qian Chen(Advisor)
- Chao Zuo
- CISL @ Boston University
  - Lei Tian (Advisor)

Funding











Thank you

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