Fast Physical-Optics Modeling of Microscopy System with Structured Illumination

Rui Shi\textsuperscript{1,2}, Norik Janunts\textsuperscript{4}, Rainer Heintzmann\textsuperscript{4}, Christian Hellmann\textsuperscript{3}, and Frank Wyrowski\textsuperscript{1}

\textsuperscript{1} Applied Computational Optics Group, Friedrich Schiller University Jena, Jena, Germany,
\textsuperscript{2} LightTrans International UG, Jena, Germany,
\textsuperscript{3} Wyrowski Photonics UG, Jena, Germany,
\textsuperscript{4} Biomedical Imaging Group, Leibniz Institute of Photonic Technology (IPHT), Jena, Germany
Background

- One-photon fluorescence microscopy
Background: Higher Resolution

**STED**
[Hell et al., *Opt. Lett.* (1994)]
- Excitation + STED
- Resolution: ~20 nm
- High power

**STORM**
[Betzig et al., *Science* (2006)]
- Diffraction-limited:
- Localized
- Resolution: ~20 nm
- Low speed

**SIM**
[Heintzmann et al., *Proc. SPIE* 1998]
[Heintzmann, *J. Microsc.* (2000)]
- Resolution: ~80 nm
- Low power and high speed
- Sample
- Living cell
Motivation and Configuration

- Electric energy density
\[ w_e \propto \| E \|^2 \]

- Contrast:
\[ c = \frac{w^{\text{max}}_{e,\text{ave}} - w^{\text{min}}_{e,\text{ave}}}{w^{\text{max}}_{e,\text{ave}} + w^{\text{min}}_{e,\text{ave}}} \]
Best: \( c = 1 \)

- Homogeneity:
\[ \sigma = \frac{w^{\text{max}}_{e,\text{max}} - w^{\text{max}}_{e,\text{min}}}{w^{\text{max}}_{e,\text{max}} + w^{\text{max}}_{e,\text{min}}} \]
Best: \( \sigma = 0 \)

- Polarization
- Diffraction from aperture
- Inclined illumination on blazed grating
Theory: Field Tracing

\[ \mathbf{\mu} = (k_x, k_y) \]

\[ \mathbf{\alpha} = (x, y) \]

Space domain \((\rho, \omega)\)

Fourier domain \((\kappa, \omega)\)

Free space propagation

Fourier Modal Method (FMM)

Local Plane Interface Approximation (LPIA)

The concept of **bidirectional operators** and its application to the modelling of microstructures Paper 10694-15, **Prof. Frank Wyrowski**
Simulation Results via VirtualLab Fusion
Polarization
Modeling Task

\[ \phi = \arctan \frac{E_y}{E_x} \]

- energy density: \( w_e \)
- contrast: \( c \)

### Lens Property

<table>
<thead>
<tr>
<th>Lens</th>
<th>Property</th>
</tr>
</thead>
<tbody>
<tr>
<td>lens 1, 2</td>
<td>Thorlab AC254 double achromat</td>
</tr>
<tr>
<td>Tube lens</td>
<td>Nikon 200 mm</td>
</tr>
<tr>
<td>Objectives</td>
<td>Nikon 60X, NA=1.4, Effective NA: ~1.12 apochromatic</td>
</tr>
</tbody>
</table>

Result: Energy Density

\[ \phi = \arctan \frac{E_y}{E_x} \]

\[ \phi = 90^\circ \]

Simulation time: ~5 s
Diffraction from Aperture
Modeling Task

- energy density: $w_e$
- homogeneity: $\sigma$

Diagram showing a setup with a grating, relay lenses, tube lens, and objective lens, with a sample at the end.
Result: Energy Density

\[ D = 3 \text{ mm} \]

\[ D = 4 \text{ mm} \]

\[ D = 5 \text{ mm} \]

\[ D = 6 \text{ mm} \]
Result: Energy Density and Homogeneity

Simulation time: ~27s

- $D = 3 \text{ mm}$
  - $\sigma = 0.37$

- $D = 4 \text{ mm}$
  - $\sigma = 0.21$

- $D = 5 \text{ mm}$
  - $\sigma = 0.15$

- $D = 6 \text{ mm}$
  - $\sigma = 0.08$
Inclined Illumination on Blazed Grating
Modeling Task

- energy density: $w_e$
- contrast: $c$

Blazed grating

$\theta^{in}$

$\theta^{di}$

$12^\circ$
Result: Diffraction Angle and Efficiency

Calculated by FMM
Result: Diffraction Angle and Efficiency

\[ \theta_{\text{in}}^{\text{di}} [-1], [0.57^\circ], [24.55^\circ] \]

\[ \theta_{\text{in}}^{\text{di}} [+1] \]
Result: Diffraction Angle and Efficiency

\[ \theta_{\text{gra}} - \theta_{\text{in}} \pm 1 \]

\[ \theta_{\text{di}} \]

\[ 24.55^\circ, 0.57^\circ \]

\[ [24.55^\circ, 0.57^\circ] \]
Results: Energy Density

$\theta_{\text{in}} = 24^\circ$

~30%

[+1 Order]  [-1 Order]
Results: Energy Density

$\theta^{in}=24^\circ$

$[\theta^{in}, \theta^{gra}] = [24.55^\circ, 0.57^\circ]$
Conclusion and Outlook

• The complexity of microscopy system with structured illumination makes it vulnerable to the undesired effects which causes the inhomogeneity and low contract of the interference pattern.

• These effects should be analyzed and taken into account in the image reconstruction algorithm.

• In case of deep tissue imaging, adaptive optics can be applied further to compensate the undesired effects.
Thanks!