Update On 3D Orientation Reconstruction

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Spheroid Distribution

$$\kappa = 0.1 \qquad \kappa = 0.25 \qquad \kappa = 0.5 \qquad \kappa = 1 \qquad \kappa = 2 \qquad \kappa = 4 \qquad \kappa = 10$$

$$f(\vec{r}; \mathbf{A}) = \frac{r^T \mathbf{A}^{-1} r}{|\mathbf{A}|}$$

$$f(\vec{r}; \vec{r}', \kappa) = \frac{\kappa}{S(\kappa) \sqrt{1 + (\vec{r} \cdot \vec{r}')^2 \left(\frac{1}{\kappa^2} - 1\right)}}$$

$$S(\kappa) = 2\pi \left(1 + \frac{\kappa^2}{2\sqrt{1 - \kappa^2}} \log \left[\frac{1 + \sqrt{1 - \kappa^2}}{1 - \sqrt{1 - \kappa^2}}\right]\right)$$

Forward Model: Single diSPIM Frame

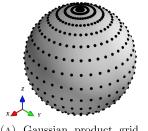
Measured Intensity:
$$I = \int_{\mathbb{S}^2} d\vec{r} \ I^s(\vec{r}) f(\vec{r}; \vec{r}', \kappa)$$

Single Fluorophore:
$$I^s(\vec{r}) = 2[A + B(1 - \cos^2\phi \sin^2\theta)] \times \sin^2\theta \cos^2(\phi - \phi_{\text{pol}})$$

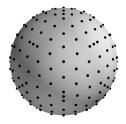
Spheroid Distribution:
$$f(\vec{r}; \vec{r}', \kappa) = \frac{\kappa}{S(\kappa)\sqrt{1 + (\vec{r} \cdot \vec{r}')^2 \left(\frac{1}{\kappa^2} - 1\right)}}$$

► No luck integrating in closed form ;(

Integrating Faster With Spherical Quadrature



(A) Gaussian product grid with N = 800.



(B) Lebedev grid with N = 266.

Beentjes, 2012

- ► Instead of evaluating integrals one after the other and using Gaussian quadrature (left), evaluate the integral all at once using Lebedev quadrature (right)
- ▶ How many grid points do I need to use to accurately integrate? To within 1%? Perfectly? \rightarrow

Harmonic Analysis On The Sphere

Measured Intensity:
$$I = \int_{\mathbb{S}^2} d\vec{r} \; I^s(\vec{r}) f(\vec{r})$$

- ▶ Rewrite $I^s(\vec{r})$ and $f(\vec{r})$ in terms of orthogonal spherical harmonics
- ► Expensive integral becomes a cheap dot product of coefficients
- ▶ With measurements under different polarization settings we get $\vec{g} = \mathcal{H}\vec{f}$ where \vec{g} is a vector of intensity measurements, \vec{f} is a vector of spherical harmonic coefficients of the fluorophore distribution, and the rows of \mathcal{H} are the spherical harmonic coefficients of $I^s(\vec{r})$ for each measurement
- \triangleright Null space of \mathcal{H} corresponds to symmetries/degeneracies

Harmonic Analysis

- ightharpoonup Harmonic analysis on $\mathbb{R}^n \to n\text{-D}$ Fourier Transform
- ▶ Harmonic analysis on \mathbb{S}^1 (circle) → Fourier Series
- ▶ Harmonic analysis on \mathbb{S}^2 (sphere) → 2D Fourier Series

Spherical Harmonics

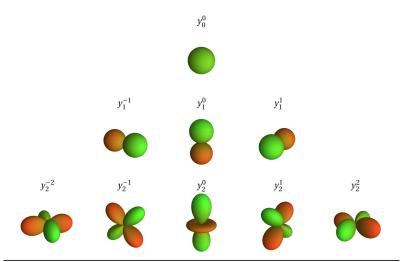
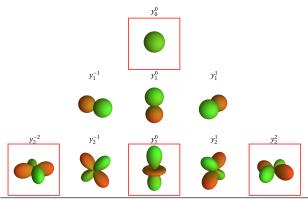


Figure B.1: Plots of the real-valued spherical harmonic basis functions. Green indicates positive values and red indicates negative values.

Jarosz, 2008

Single View diSPIM Frame



 $\overline{\text{Figure B.1: Plots of the real-valued spherical harmonic basis functions. Green indicates positive values and red indicates negative values.}$

Jarosz, 2008

▶ Non-zero coefficients for single view diSPIM frame shown in red boxes

Early Conclusions + Upcoming Work

- ▶ Odd bands aren't antipodally symmetric so they aren't present in the sample.
- ▶ Zero coefficients on $y_2^{\pm 1}$. Corresponds to degeneracy? Investigating.
- ▶ Adding polarizers on the detection side with give us access to 4th order band. Still investigating degeneracy.
- ▶ A single fluorophore has non-zero components in all even bands (sort of like FT of $\delta(x)$ is 1). We can only measure the first few bands which constrains the accuracy of our estimates.
- ▶ Now we're estimating spherical harmonic coefficients instead of parameters of a hypothesized distribution. That's all of the info we can access.
- ▶ Upcoming—reconstruction in terms of matrix inversion!