High-speed, large field-of-view and deep imaging with an adaptive excitation source

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Motivation

1. Neurons occupy <10% of the brain.
2. Conventional raster scan illuminates the whole field-of-view (FOV).

→ leads to low efficiency, brain damage.

Region-of-interest (ROI) imaging is a solution.

Nadella, Nature Methods, 2016

Concept: adaptive excitation

Laser scanning microscopy

Neuronal activity

Two-photon microscopy

Mouse: GCaMP6s, 7 month old. Power: 18 mW. Speed: 30 Frames/second

Depth: 680 µm

Three-photon microscopy

Mouse: jRGECO1a, 7 month old. Power: 35 mW. Speed: 30 Frames/second

Comparison

Same depth: 750 µm, same power: 35 mW

Conventional raster scan

Adaptive excitation

Compatibility

Commercial microscope

The adaptive excitation source can be integrated with any existing laser scanning MPM as long as the pixel clock of the microscope is accessible.

Future work

Blood cell flow in hippocampus

>200 frames/s with polygon scanner

Future work

Soliton fiber source

What could it do?

1. 775 nm - 1260 nm for "2P + Green, yellow, red dye" or "3P + green dye"
2. 1550 nm - 2520 nm for "2P + NIR dye" or "3P + red dye"

Table 1 Comparison of state-of-the-art excitations for 2PM at 920 nm

<table>
<thead>
<tr>
<th>Excitations</th>
<th>Pulse energy</th>
<th>Repetition rate on ROI</th>
<th>Cons</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ti:Sapphire laser</td>
<td>1-17 nJ</td>
<td>80 MHz</td>
<td>Low pulse energy</td>
</tr>
<tr>
<td>Optical parametric oscillator (OPO)</td>
<td>2-3 nJ</td>
<td>80 MHz</td>
<td>Low pulse energy</td>
</tr>
<tr>
<td>Optical parametric amplifier (OPA)</td>
<td>250-1000 nJ</td>
<td>1-4 MHz</td>
<td>Low repetition rate</td>
</tr>
<tr>
<td>Our adaptive excitation source</td>
<td>~60 nJ</td>
<td>32 MHz</td>
<td></td>
</tr>
</tbody>
</table>

Table 2 Comparison of state-of-the-art excitations for 3PM at 1700 nm

<table>
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Acknowledgements

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