Nikon AX: Phasor-Based Structural Separation Using FLIM

Introduction

Fluorescence lifetime imaging microscopy (FLIM) is a powerful tool for analyzing biological systems by distinguishing between different populations based on their fluorescence lifetimes. The phasor plot is a graphical representation that converts the fluorescence decay of individual pixels in a sample into points on a plot. These points naturally cluster into distinct regions, each corresponding to a unique lifetime population within the sample.

In the Nikon AX-PicoQuant FLIM system, using a single spectral window enables the differentiation of fluorescent species or environments solely based on their lifetimes. This method simplifies both the experimental setup and data analysis by eliminating the need for multiple spectral channels while still providing precise identification of distinct lifetime populations. This streamlined approach enhances the efficiency of resolving lifetime components in complex biological systems.

Process

- **Software information:** NIS-Elements version 6.10.01 with PicoQuant FLIM-FCS full integration

nis_phasor_publish.mp4

- **Sample information:**
  - Daisy pollen

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- **Software information:** NIS-Elements version 5.42.06 with PicoQuant FLIM-FCS full integration

- **Sample information:**
  - Glial Fibrillary Acidic Protein (GFAP) — labeled with BDP-TMR
  - Mitochondria (via TOM20) — labeled with CY3

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