

Cultured human cells exposed to GFP-positive EVs



Detecting Low-Intensity Signals Using NSPARC

To acquire an optimal image in microscopy, it is essential to fine-tune fluorescent labeling and acquisition parameters to achieve an optimal signal-to-noise ratio and robust intensity levels. This is particularly true for widefield and confocal microscopy. However, low fluorescent signal may present substantial challenges in this regard.

Challenges

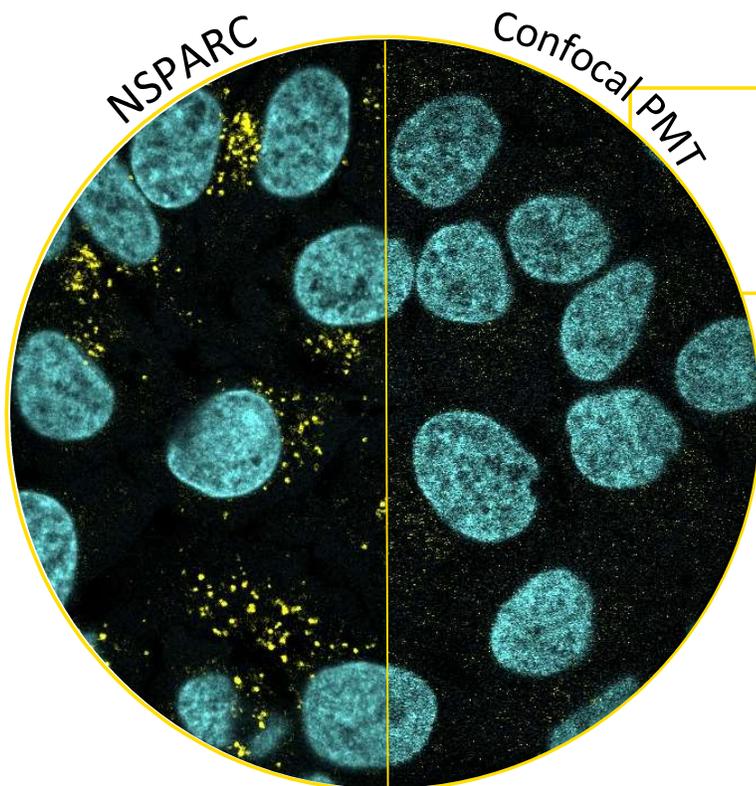
When dealing with faint fluorescent signals, detecting the resulting signal can be exceptionally challenging, even when employing highly sensitive cameras or photomultiplier tubes (PMTs). NBIL's AX-R confocal microscope tackles this issue by incorporating the NSPARC (Nikon Spatial Array Confocal) detector. The NSPARC detector not only enables super-resolution capabilities but also features a single-photon counting detector array that delivers exceptionally low noise levels. This technology therefore enables the capture of dim signals, providing high-contrast images at reduced excitation power.

Applications

- Detection of faint intensity signals in live or fixed cells
- Minimally invasive live-cell imaging
- Super-resolution imaging of live or fixed samples

Using NSPARC to detect fluorescently-labelled vesicles in human cells.

Extracellular vesicles (EVs) have gained significant attention in biomedical research due to their potential as efficient delivery vehicles for drugs, proteins, or nucleic acids. Cells naturally produce EVs are endogenous vesicles shed by cells and can carry mRNA or proteins to recipient cells. Given their cellular origin, EVs are associated with minimal toxicity and are readily internalized by recipient cells, making them highly suitable for drug delivery applications. However, due to their small, nanometer range size, EVs can be challenging to specifically stain and detect.



GFP-labeled EVs

METHODS

Human cells were exposed to GFP-labeled EVs.

IMAGING

Nikon AX-R NSPARC and AX-R PMT detector.

The NSPARC's ultra-low-noise detector can detect even faint signals, just a few photons above autofluorescence

In this application, EVs were labelled with a GFP-fluorescent tag. Despite this labelling, the EVs remained undetectable using traditional camera-based or PMT-based detection methods. In addition, the background autofluorescence of cells in the GFP channel obscured the faint signal of GFP-labeled EVs. In contrast, NSPARC can easily detect the GFP-labeled EVs over background at identical laser power.

Nikon BioImaging Lab's services:

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