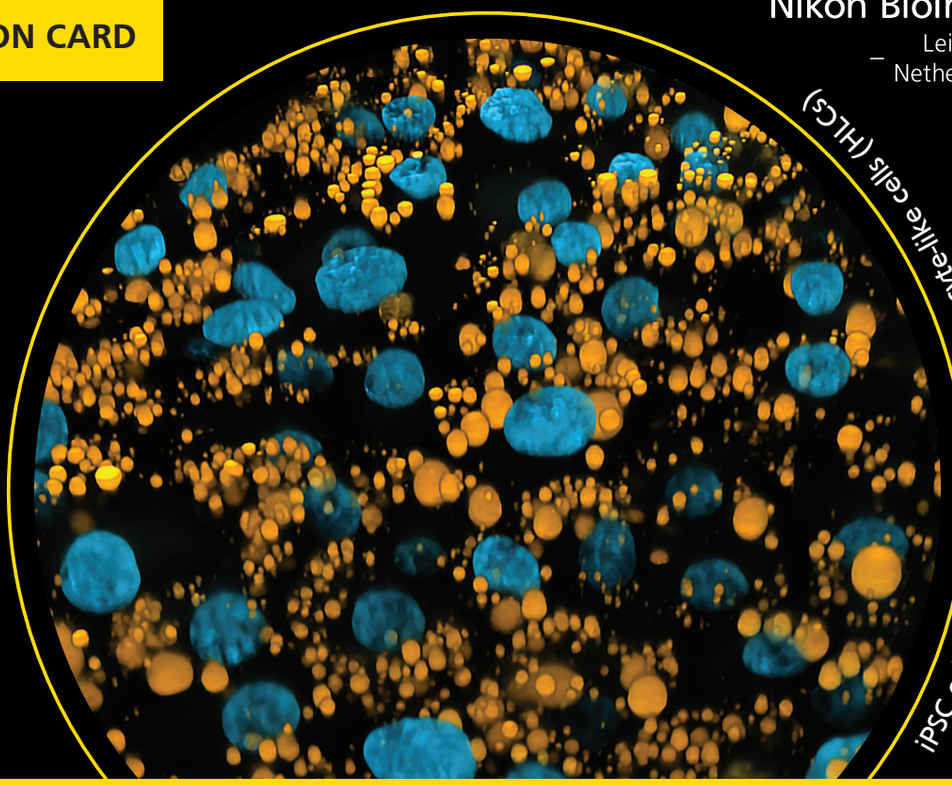
APPLICATION CARD

Nikon BioImaging Lab
Leiden
Netherlands



Complex 3D imaging

Until recently, life science research relied on 2D in vitro imaging of a single layer of cells on a flat surface. However, these systems have profound limitations that are recognized throughout the scientific community, especially for translational research. Increasingly, more physiologically-relevant models such as spheroids, organoids, and organ-on-a-chip systems are being used. Even monolayer cells can be imaged in 3D to study internal spatial changes of these cells over time.

Challenges

3D acquisition takes significantly more time, the data files become dramatically larger, and the analysis is more complicated as most existing methods have been developed and tested in 2D. While some methods can be applied to a Z-stack plane by plane, others require huge amounts of memory or lack the ability to connect a series of two-dimensional segmented objects into a single three-dimensional one.

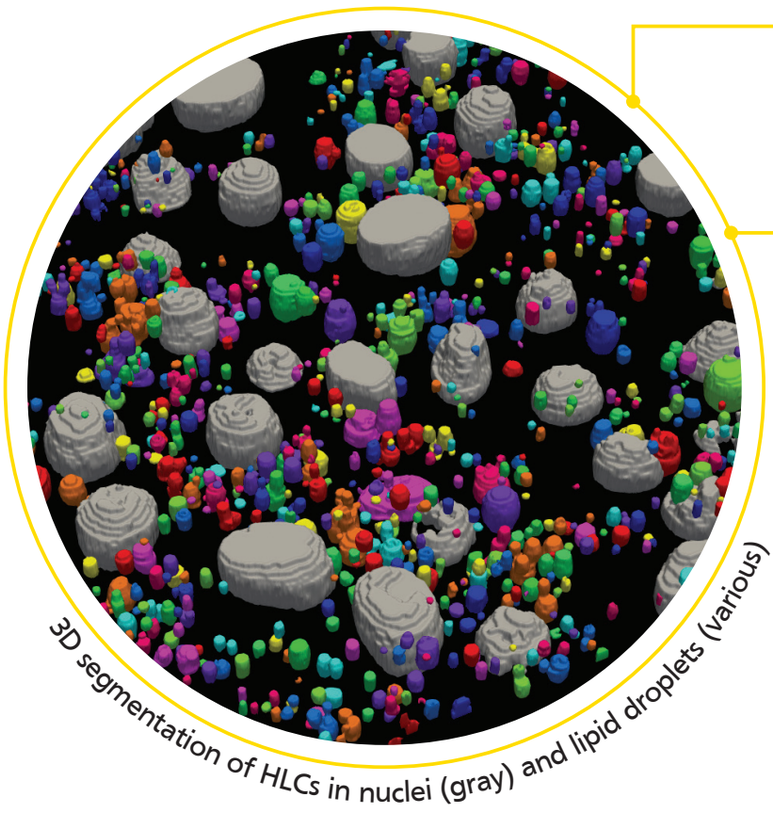
Visualizing 3D models in high definition

At NBIL we have state-of-the-art confocal microscopes along with the latest software which can perform automated acquisition in combination with on-the-fly analysis, including the use of AI. For complicated analysis questions, **our in-house experts can find the answer to any research question.**

Talk to an expert nbil.eu@nikon.com

Imaging lipid droplets inside cells

For this project, we imaged lipid droplets of various sizes in induced pluripotent stem cells (iPSC) differentiated into hepatocyte-like cells that were cultured in media with different properties. 3D imaging was used to view all the lipid droplets within these cells. This method ensures that we capture the entire cell with all the lipid droplets within, while using a high magnification objective.



3D segmentation of HLCs in nuclei (gray) and lipid droplets (various)

METHODS

Differentiated HLCs provided by trenzyme GmbH, stained with Nile Red (lipid droplets) and Hoechst (nucleus)

IMAGING

Nikon AX/AX R confocal

Denoising imaging data extends the dynamic range of object size segmentation

Powered by AI, the confocal imaging system offers high-speed capabilities and flexible configurations.

We implemented automated acquisition protocols to image multiple 96-well plates automatically and accurately. After acquisition, the images were analyzed by our experts to remove noise and segment the lipid droplets for counting and size measurements. Shot noise inherent to confocal imaging was removed with Denoise.ai, a convolutional neural network to remove Poisson shot noise from resonant confocal images.

Nikon BioImaging Lab's services:

01 Assay development › **02** Imaging › **03** Analysis › **04** Reporting