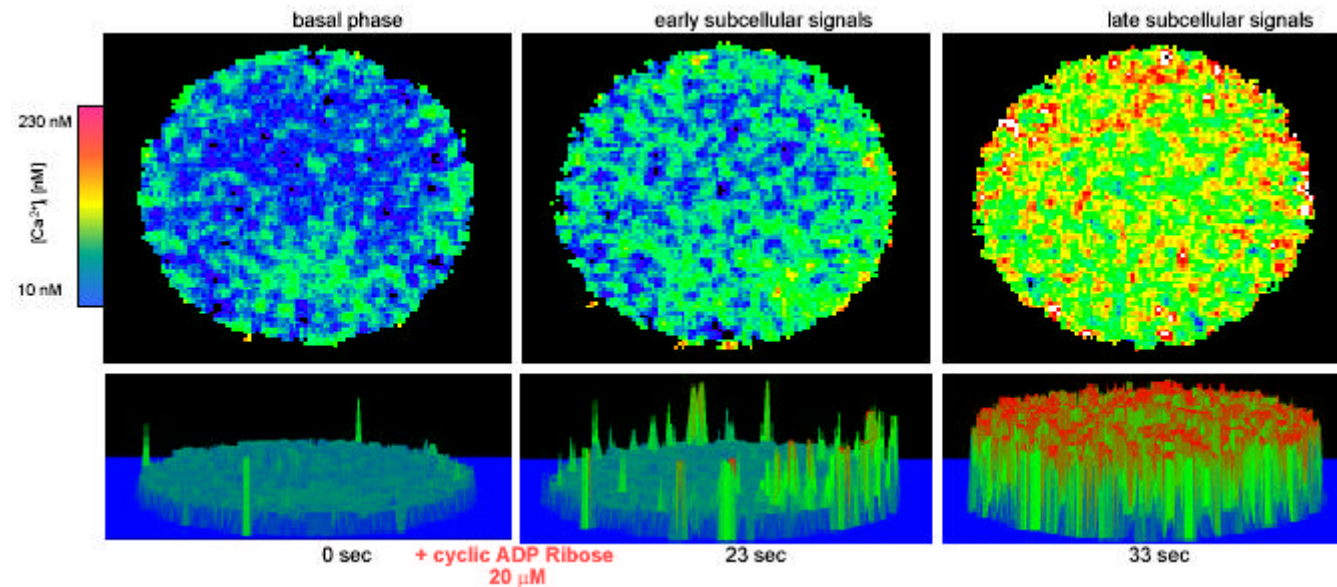


Subcellular calcium signals in T-lymphocytes



Prof Andreas Guse and Svenja Kunerth of the University Hospital Hamburg-Eppendorf are studying intracellular Ca²⁺ signaling. In many cell types, intracellular Ca²⁺ signaling starts with highly localized subcellular events, which are often followed by global Ca²⁺ signals. Prof Guse is using an Openlab cell imaging system and established a technique to allow visualization of subcellular Ca²⁺ signals in living cells using a light microscope acquisition system. Activation of such subcellular Ca²⁺ signals was accomplished here by microinjection of the second messenger cyclic ADP-ribose.

To produce the T-lymphocyte images shown here, a Z series of three planes, 0.5 mm apart, was acquired by excitation at 340 and 380 nm at each time point. The images were deconvolved using the Openlab Nearest Neighbor Deconvolution module, then ratioed and a Rainbow cLUT was applied for display. Using the Openlab Profiling module, it is possible to visualize both the position and the intensity of the subcellular Ca²⁺ signals across the cell at each time point.

Module configuration

Module Families

Camera

Snapper Video

Hardware

Filters & Shutters

Polychrome

File Filter

Quick Time File Filter

TIFF file filter

PICT file filter

Application

Ratio

Nearest Neighbor DCi

Profiling

Automation

Automator

Critical Points

- Orca ER, high resolution, high dynamic range, monochrome cooled camera used to acquire high quality images
- Monochromatic light source allows fast switching (1.5ms) between two excitation wavelengths to minimise the time between A and B images
- Image sequences can be saved in other formats, individual PICTs or TIFFs or QuickTime movies
- Images can be annotated for publication
- Rapid ratioing of image sequences
- Deconvolution for rapid improvement of XY resolution
- Easy to use and powerful icon based programming language for automated imaging experiments
- Ideal for fast acquisition and on-or off-line post capture analysis