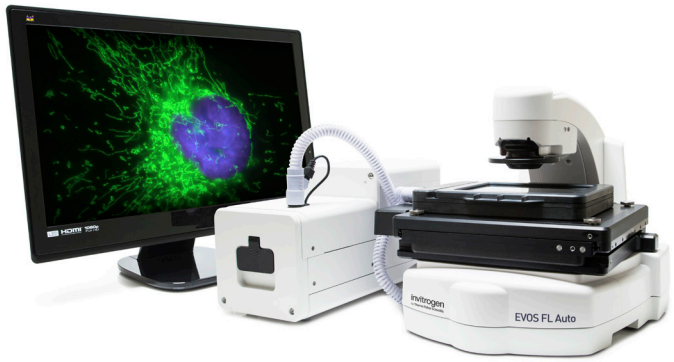
Using time-lapse imaging with the EVOS FL Auto Imaging System

Introduction

The Invitrogen™ EVOS™ FL Auto Imaging System is a fully automated, digital, inverted multi-channel fluorescence and transmitted-light imaging system with outstanding workflow efficiency. Designed to meet demanding requirements over a broad range of applications, it supports high-resolution mosaic tiling, multiple-position well scanning, cell counting with thresholding, and time-lapse studies.

Time-lapse imaging can allow scientists to interrogate temporal and spatial changes in a cell or group of cells when using fluorescence microscopy. When combined with fluorescent proteins and reagents compatible with live cells, time-lapse imaging can provide more in-depth interrogation of cellular events and protein interactions.

In this application note, we show how Invitrogen™ Molecular Probes™ reagents were used in combination with the EVOS FL Auto Imaging System to observe cellular division in HeLa cells over a period of 18 hours. During this time period, asymmetric cell division was observed in the HeLa cell culture.



Materials

- Invitrogen™ Molecular Probes™ CellLight™ Histone 2B-GFP, BacMam 2.0 (Cat. No. C10594)
- Invitrogen™ Molecular Probes™ CellLight™ Mitochondria-RFP, BacMam 2.0 (Cat. No. C10601)
- Invitrogen™ EVOS™ Light Cube, GFP (Cat. No. AMEP4651)
- Invitrogen™ EVOS™ Light Cube, RFP (Cat. No. AMEP4652)
- Thermo Scientific™ Nunclon™ Delta Surface (Cat. No. 140675)

invitrogen

Methods

HeLa cells grown in 6-well plates were transduced with CellLight Histone 2B-GFP and CellLight Mitochondria-RFP overnight. Cells were then washed once with DPBS and fresh medium was added. Time-lapse imaging was performed over a period of 24 hours with a 20x objective using the time-lapse function of the EVOS FL Auto Imaging System with the Invitrogen™ EVOS™ Onstage Incubator.

Results and discussion

Time-lapse imaging is a powerful technique that allows researchers to visualize cellular events and morphology over an extended period of time. Combining fluorescent reagents and time-lapse imaging provides even greater information to analyze cellular processes in real time. In this experiment, cell division in HeLa cells was monitored over a period of 18 hours to identify potential anomalies in the process. Cells were transduced with CellLight Histone 2B-GFP and CellLight Mitochondria-RFP to visualize cellular processes during division. Images were captured every 12 minutes in the GFP and RFP channels on the EVOS FL Auto Cell Imaging System with all underlying images saved as well as the merged images. A movie was then generated from the merged images at rate of 1 frame/sec. During this experiment, the time lapse movie captured HeLa cells dividing in two different ways. In one particular cell (from intervals 15–29), multipolar mitosis can be observed (Figure 1). Multipolar mitosis is an aberrant cell division event where chromosomes are pulled to more than two poles in the cell [1]. In an adjacent cell, from intervals 42–56, normal mitosis can be seen (Figure 2).

Reference

1. Vitale I et al. (2010) Multipolar mitosis of tetraploid cells: inhibition by p53 dependency on Mos. *EMBO J* 29:1272–1284.

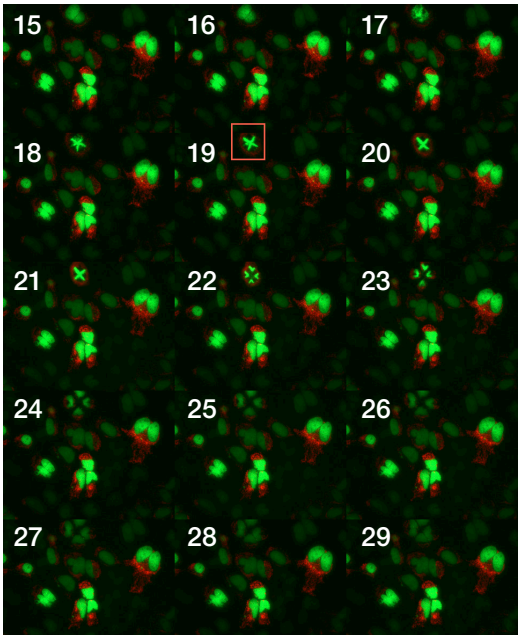


Figure 1. HeLa cell undergoing multipolar mitosis (red box) following transduction with CellLight Histone 2B-GFP (green) and CellLight Mitochondria-RFP (red). To see the full time-lapse movie, visit **thermofisher.com/evosflautogallery**.

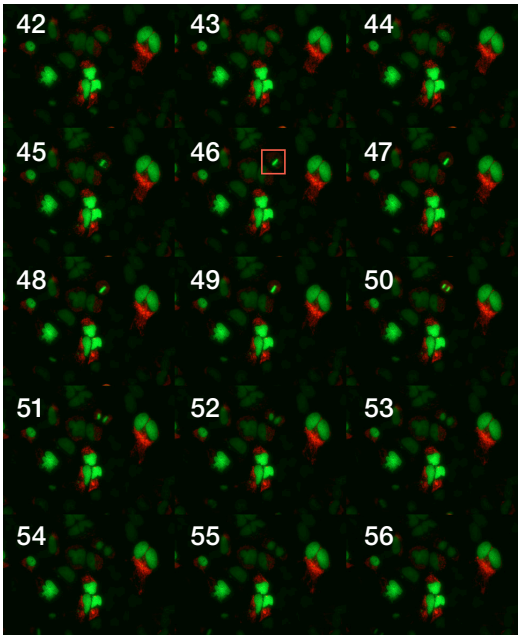


Figure 2. HeLa cell undergoing normal cell division (red box) following transduction with CellLight Histone 2B-GFP (green) and CellLight Mitochondria-RFP (red). To see the full time lapse movie, visit **thermofisher.com/evosflautogallery**.

Find out more at **thermofisher.com/evosflauto**

ThermoFisher SCIENTIFIC