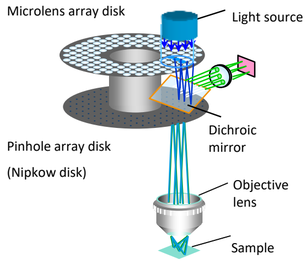
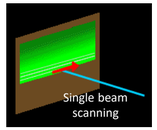
■Yokogawa Technology

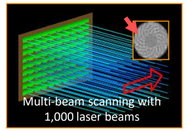
Microlens enhanced dual Nipkow disk
→ high-speed, low photo-toxicity and low photo-bleaching

Multi-beam scanning by the microlens-enhanced Nipkow disc enables high-speed image acquisition. Furthermore, photo-toxicity and photo-bleaching caused by multiplexed micro-beam scanning with moderate power lasers is remarkably lower than that caused by conventional single beam scanning.





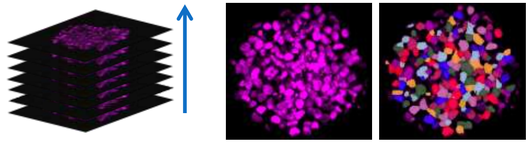
Conventional confocal



YOKOGAWA

3D analysis

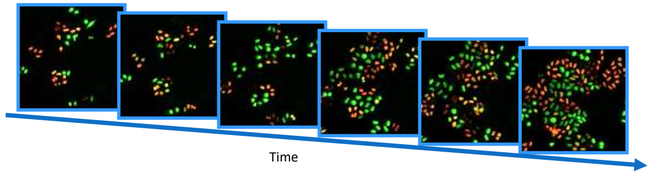
・Analysis of Z-stack images in three-dimensional space.
・The volume and the location of objects in 3D space can be quantified.



Recognition of the cells in a spheroid

Live cell imaging

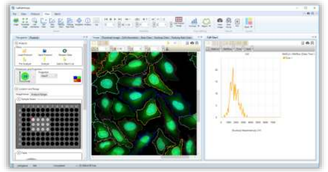
・Dynamic behaviors of live samples can be tracked by long-term time-lapse imaging.
・Built-in stage incubator maintains ideal culture conditions throughout the recording session.



■System configuration

CQ1

Import image data



High Content Analysis Software
CellPathfinder™

- ✓ Easy quantification of feature data
- ✓ 3D and live cell imaging
- ✓ Cell-friendly image acquisition
- ✓ Bench-top size and no need for darkroom

- ✓ Simple workflow with user-friendly interface
- ✓ Large collection of ready-to-use image analysis templates
- ✓ Various output options including CSV tables, graphs and movies
- ✓ Sophisticated analysis functions
  - Machine learning
  - Texture analysis
  - Gating
  - Digital phase contrast
  - Object tracking
  - Label-free analysis

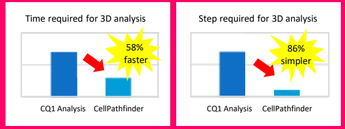
	2 laser model	2 laser with incubator model	4 laser with incubator model
Optics	Microlens enhanced dual wide Nipkow disk confocal / Bright field		
Laser	405,488nm		405,488,561,640nm
EM Filter	Max. 10 filters		
Camera	sCMOS 2560x2160 pixel,16.6x14.0mm		
Objective lens	Max.6 lenses Dry : 2x, 4x, 10x, 20x, 40x , 60x Long working distance : 20x, 40x Phase contrast : 10x, 20x		
Sample vessel	Microplate（6,12,24,48,96,384,1536well） Option（glass slide, cover glass chamber, 35mm dish, 60mm dish）		
Stage incubator	-	Controllable temperature range : Room temperature +5 – +17 °C, Max.40 °C CO_2 concentration : Atmospheric concentration – 7 % O_2 concentration : 3 % – Atmospheric concentration	
Workstation	Measurement workstation and analysis workstation		
Analysis software	High content analysis software CellPathfinder™		

understanding

Enhanced

High throughput and easy-to-use

Analysis is much faster and simpler in CellPathfinder.



◆Contact Information

Yokogawa Electric Corporation
Bio Solution Center, Life Innovation Business HQ
Web site https://www.yokogawa.com/solutions/products-platforms/life-science/
E-mail csu_livecell_imaging@cs.jp.yokogawa.com

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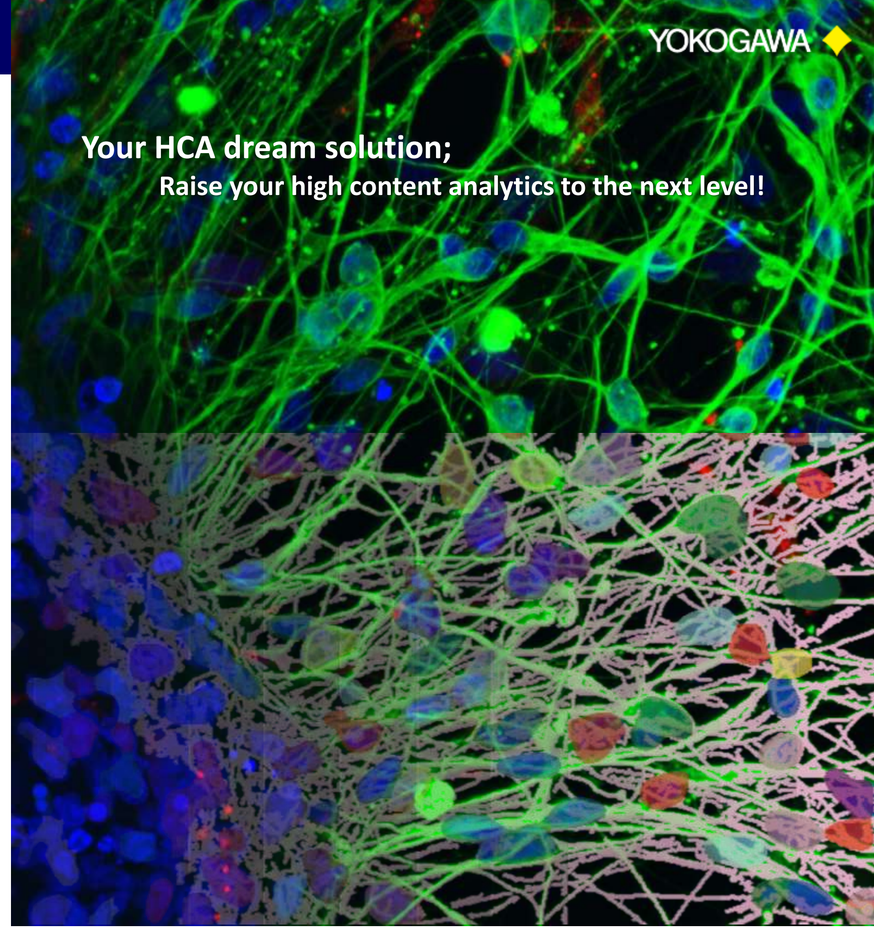


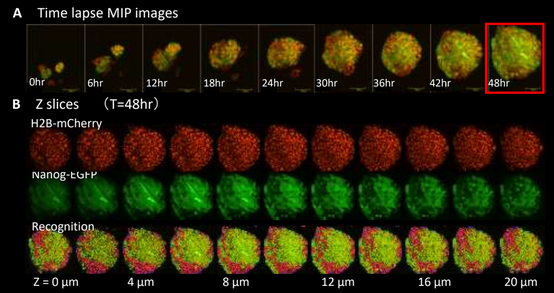
High content imaging system
Confocal Quantitative Image Cytometer

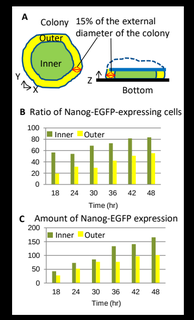
Cell Voyager

CQ1

Data provided by Ecole Normale Supérieure de Lyon（ENS Lyon）

4D imaging of embryonic stem cell colony
- Time-lapse and 3D imaging of live cell-



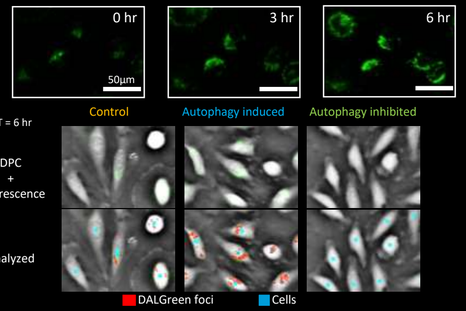


The expression of a nondifferentiation marker in the outer and the inner region of a ES cell colony was tracked for 48 hrs. Confocal imaging provides the precise location of objects in 3D space, so that the position-dependence of biological phenomena can be examined in detail.

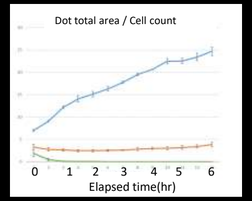
Data provided by Dr. Horie, Nara Medical University

Autophagy - Analysis using DPC images-

DALGreen: Time-lapse recording after medium change



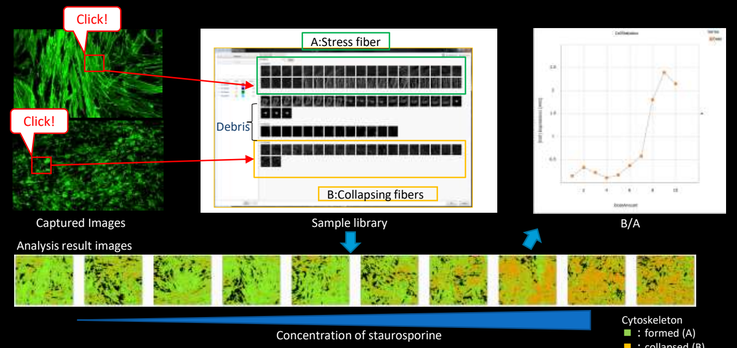
Cell: Hela cell
Objective: 20x
Wavelength: Ex 405nm Em 525/50
Bright field
Time-lapse: Interval 30min
Duration 6hrs
Autophagy detection :
DALGreen (DOJINDO LOBORATORIES)



Autophagy was detected by the increase of fluorescence of DALGreen reagent and the total number of cells was counted using the DPC (Digital Phase Contrast) images. The DPC images enabled the recognition of cells without specific labeling for nuclei or cell bodies.

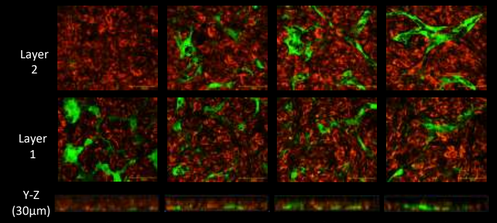
Collaboration with DOJINDO LABORATORIES

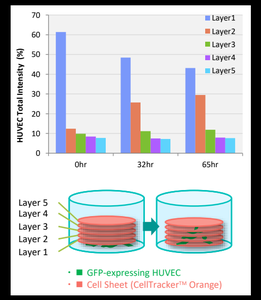
Stress fiber collapse - Quantification by using machine learning -



The collapse of stress fibers induced by staurosporine was quantified. The analysis using the machine learning function clearlly visualized dose-dependent effect of the drug.

Multi-layered cell sheet - Live imaging of 3D migration-

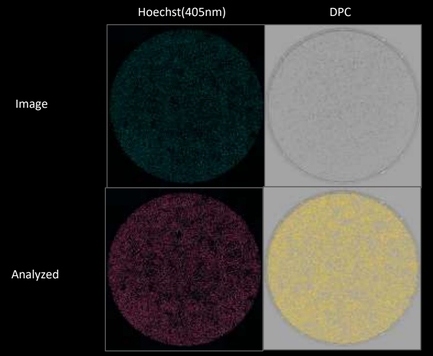




Single slice images showing the migration of HUVECs into upper layers. (*Rows, from top to bottom*) Single slice images of layers 2, 1 and corresponding Y-Z plane images of the cell sheet. (*Columns, from left to right*) Images acquired at 0, 17, 34 and 51 hr incubation. The image filed is the same.

Data provided by Dr. Nagamori, Osaka University
Reference: Nagamori E. et al., Biomaterials, 34, 662-668. (2013)

Counting cells in a whole well - Label-free Analysis-



The result of cell counting in the DPC images almost perfectly matched the result obtained with a conventional method counting fluorescently-labeled nuclei.

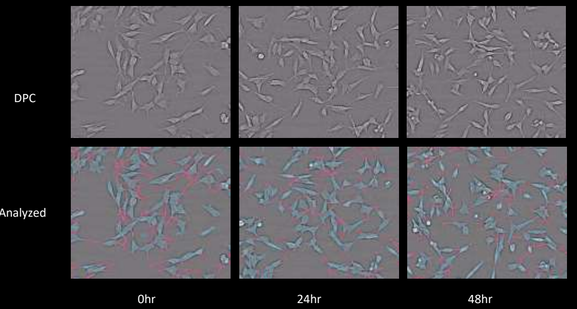
Cell: Hela cell
Objective: 10x
Plate: Greiner 96well plate
Tile image of DPC (phase type) and 405nm

CellCount

Hoechst (405nm)	14351.7	10867.7	6944.3
DPC	14316.3	10847.7	6942.0
Ratio_DPC/405 (%)	**99.75**	**99.82**	**99.97**

(n=3wells)

Live cell analysis of neurite outgrowth
- Combined use of DPC and machine learning for label-free analysis -



Cell:SH-SY5Y cell
Objective: 10x
Wavelength: Bright field
Time-lapse: Interval 1.5hrs;
Duration 67.5hrs
Pink: Neurite
Blue: Cell body

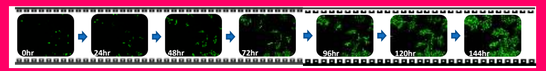
The DPC images were created from bright field images of unstained neuronal cultures and analyzed using the machine learning function to recognize cell bodies and neurites.

Enhanced

4D Live cell analysis

- Long-term time-lapse live imaging was performed for 6 days.
- The cultures were maintained in a healthy and proliferative state until the end of the experiment.

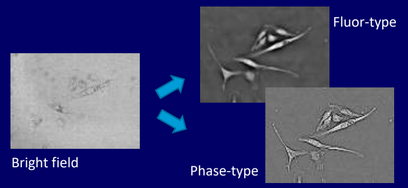
Reach a new level of



New

DPC for Label-free analysis

High contrast DPC (Digital Phase Contrast) images are created from unstained bright field images. The DPC images are suitable for label-free analysis.

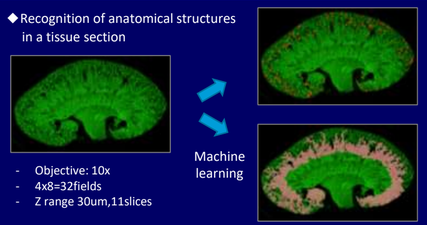


New

Machine learning

The machine learning function enables the recognition of complex structures that cannot be readily distinguished by conventional intensity threshold-based object recognition methods.

◆Recognition of anatomical structures in a tissue section



- Objective: 10x
- 4x8=32fields
- Z range 30um,11slices

Object count: 120
Average Area: 7331.4

Total Area: 10470637.6
Average Intensity: 7774.5