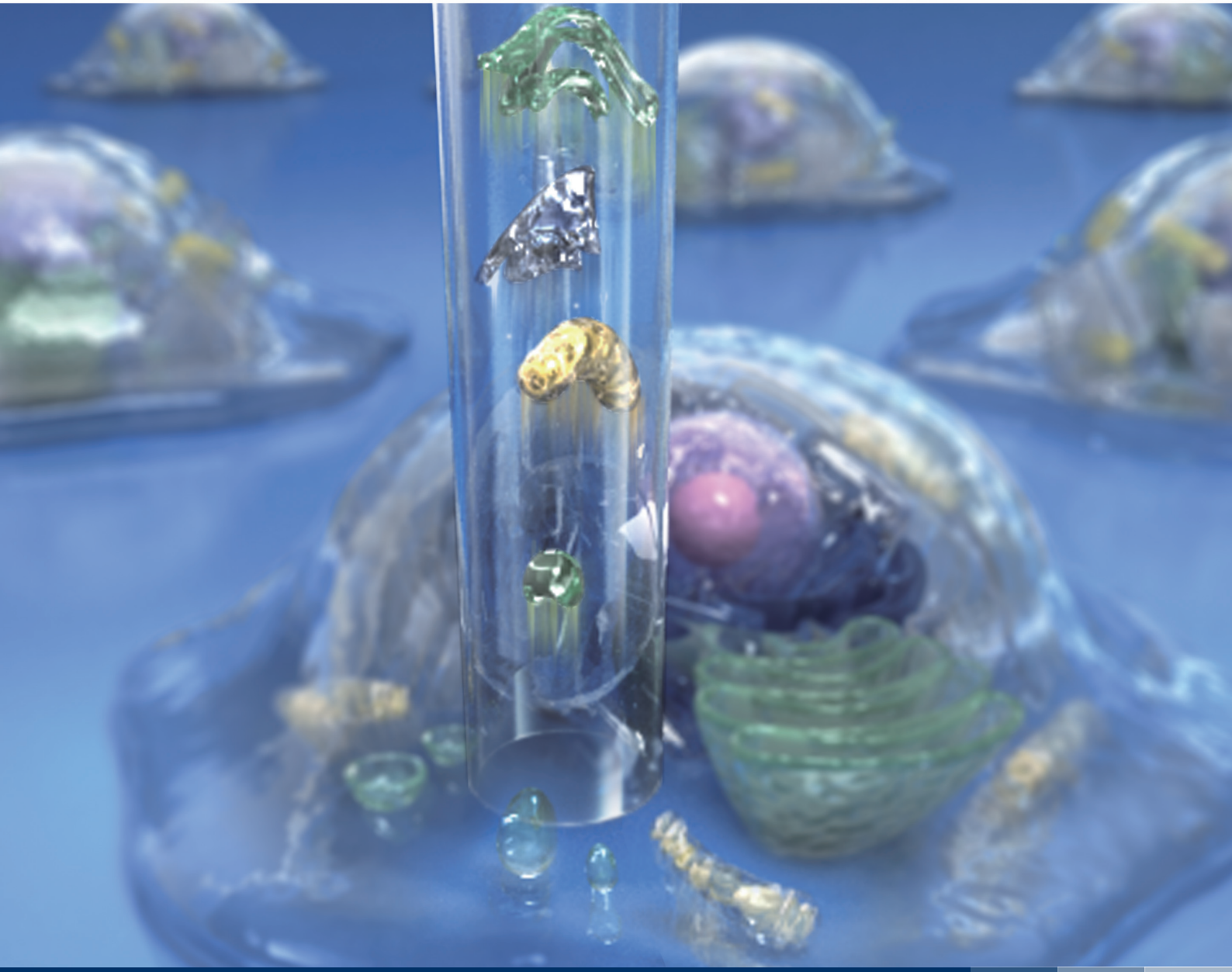
Co-innovating tomorrow™

YOKOGAWA



Single-cell solution

Single Cellome™ System
SS2000

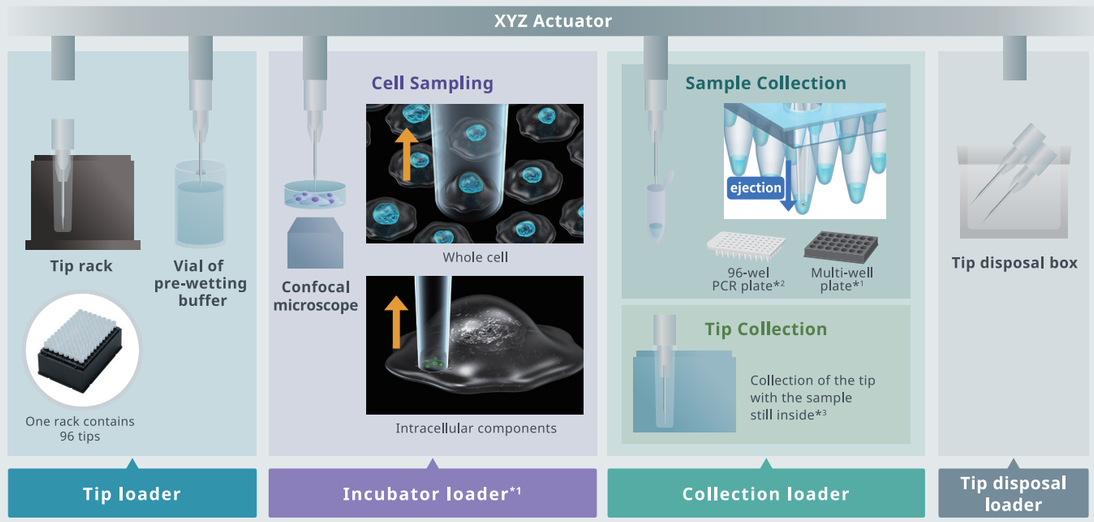
SS2000 is an automated subcellular sampling system.

Bulletin 80S01B01-01EN

Subcellular Sampling System

SS2000 is an automated direct sampling system
for specific intracellular sites at the single cell level
while simultaneously imaging cells with a confocal microscope.

Product Schematic



*1 The culture environment can be preserved through control of temperature, humidity, and CO_2 concentration.
*2 A temperature of 4°C can be maintained using a special cooling block. (Option)
*3 Can be used for single-cell mass spectrometry, etc. (Option)

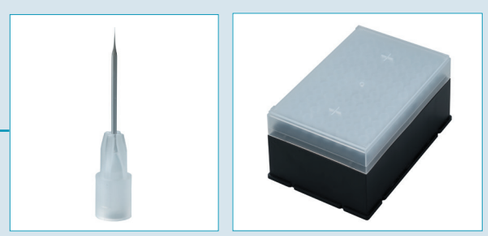
Special Glass Tips

- Four inner diameter sizes

Size

Inner diameter of tips	3µm	5µm	8µm	10µm

- One rack contains 96 tips.
- The number,position,size of the tips are automatically readed and reflected on the software.
- Tips indivisually wrapped with hermetic sealing.



Glass tip

Tip Rack

Single Cellome™ System **SS2000**

Tip loader

The Tip loader loads racks holding the glass tips.The positions and types of the inserted tips are automatically read and reflected to software. To prevent accidents and contamination, the tips are for single use. A buffer can be loaded to prevent adsorption of samples onto the tips.

Tip disposal box

The disposal box is for discarding used glass tips to prevent injuries and contaminations. The entire box can be taken from the disposal box door so that the glass tips can be disposed without being touched. Once a certain amount has accumulated,an alarm is displayed in the software to indicate when to dispose of the tips.

Incubator loader

The loader loads cells from each culture vessel in an incubated environment while sampling and taking confocal images. 35mm dishes and (6-to96-well) microwell plates can be used.*1

Collection loader

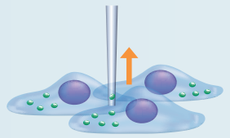
The collection loader collect samples that it ejects onto PCR plates and multiwell culture plates (96well) Cooling or incubating functions can be selected. Samples for Direct-MS can also be taked out by leaving them in the tip instead of ejecting them.

*1: Restricted to imaging vessels with a bottom thickness of 0.2 mm or less. *2: Not restricted to imaging plates.

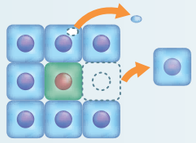
Single Cellome™ System **SS2000**

Application Examples

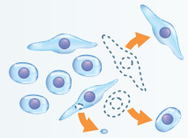
1 **Direct sampling of intracellular components such as organelles and parts of cytoplasm**



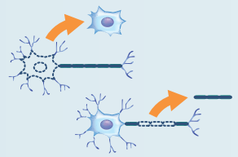
2 **Sampling cells next to a target cell**



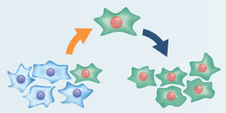
3 **Sampling cells with different morphologies**



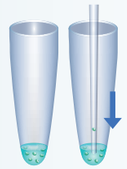
4 **Sampling of specific cell regions**



5 **Single-cell cloning from cells expressing unique behavior**

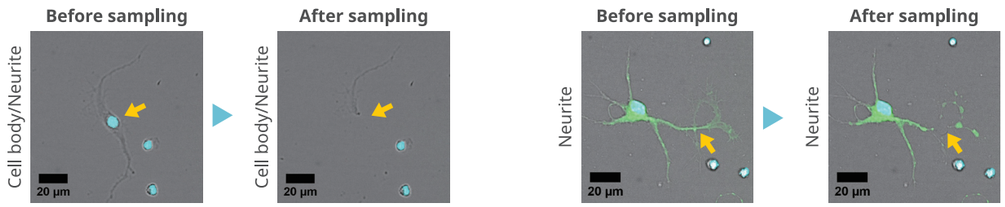


6 **Collecting of multiple samples into the same well**



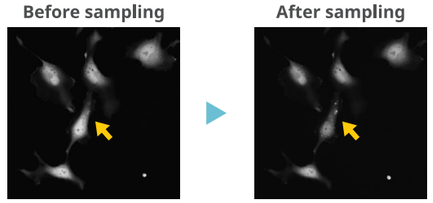
Sampling Examples

Cell region-specific collection from first-generation mouse neurons



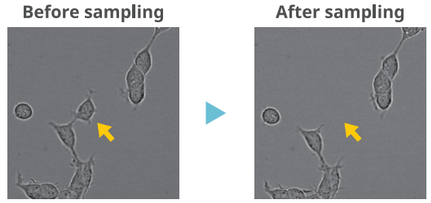
Sampling of the cell body with the neurites (10µm inner diameter tips) or only neurites (3µm inner diameter tips) after staining the nucleus (blue) or cell body (green) (images merged from bright-field and fluorescence images).

Collection of cytoplasm from HeLa cells



Sampling with a 3µm inner diameter tip after staining the cytoplasm (The sampling reduced the fluorescent region / fluorescence images).

Collection of a specific HEK293 cell

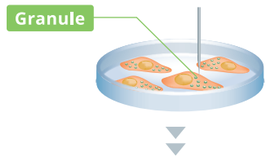


Sampling of an entire cell with a 10µm inner diameter tip (The sampling removed only the single, targeted cell / bright-field images).

Genetic Analysis of intracellular components

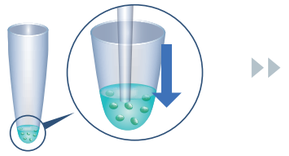
Sampling of intracellular components

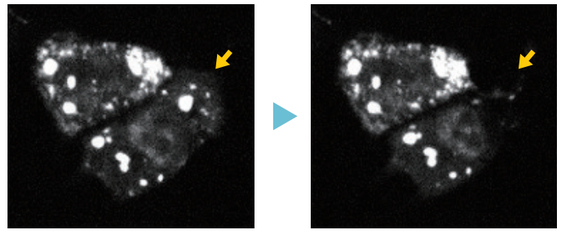
Sampling with a 3µm inner diameter tip after staining the granules.

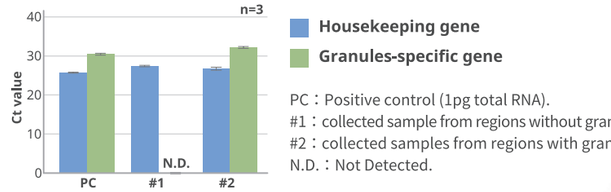


Sample collection

Collecting of granules sampled from multiple cells into the same well.





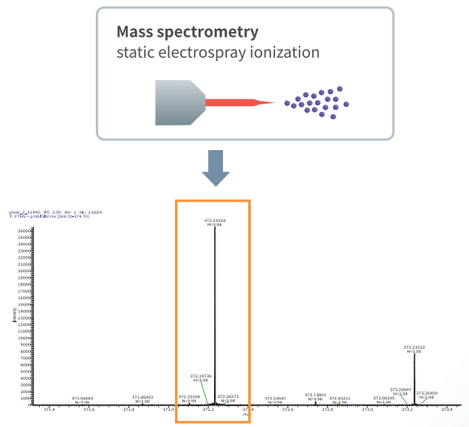


PC：Positive control (1pg total RNA).
#1：collected sample from regions without granules.
#2：collected samples from regions with granules.
N.D.：Not Detected.

Results Subcellular sampling by SS2000 enables down-stream analysis of region-specific gene detection.

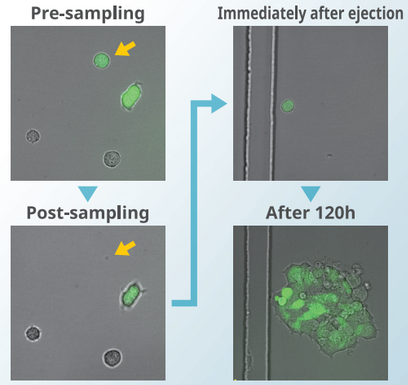
Mass Spectrometry of intracellular components

- The ESI-MS method developed by Prof. Masujima of the RIKEN Institute (where the sample is used for mass spectrometry after being collected and retained in the tip), was used.
- Drug metabolites were detected from the cytoplasm of cultured cells exposed to a drug for 24 hours.



Single-cell Cloning

- It is possible to culture cells that were sampled.
- Single-cell cloning can be achieved using cells designated from microscopic imaging or either cells with an unique behavior or single cells after transfection can be selected.
- Accurate and efficient cloning is possible by combining various imaging analyses.



GFP-expressing HEK293 cells and normal HEK293 cells were co-cultured, and a single GFP-expressing HEK293 cell was collected and cultured. It was confirmed that only HEK293 cells expressing GFP proliferated.

Sampling Technology

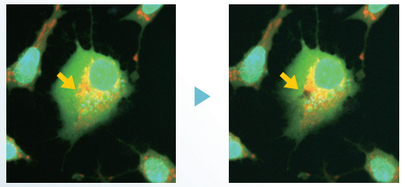
Sampling features

- Automated operation
- Precise pipetting control of location works
- Sampling with location and morphology information
- High resolution images and imaging analysis using confocal microscopy
- Incubator function for maintaining cell activity

Core technology

Sampling of intracellular components

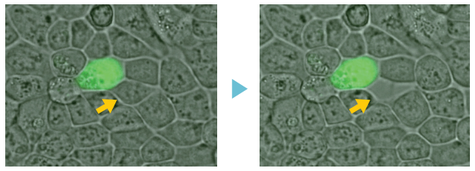
Isolated target regions within a cell can be automatically aspirated. Regions including the targeted organelles and cytoplasm can be selectively sampled.



After staining of HeLa cell nuclei (blue), cytoplasm (green), and mitochondria (red), a mitochondria-rich region (arrow) of the cytoplasm was sampled.

Maintaining of cell location and morphology information

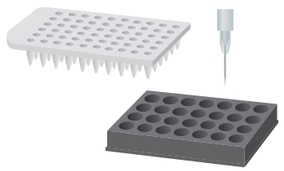
Since it is possible to sample just the target cells without detaching cells during culture, sampling can be achieved while maintaining location and morphology information.



Normal MDCK cells and target cells—abnormal MDCK cells with a green fluorescent label—were cultured at a 50:1 ratio, and then a normal cell (arrow) adjacent to an abnormal cell expressing a fluorescent signal was sampled.

High usability samples

Collected samples can be transferred to PCR plates and microwell plates. It is also possible to accumulate multiple samples into the same well or retain it in the glass tip for removal without ejecting. The sample collection site is equipped with a cooling function to prevent sample degradation and has an incubating function to maintain culture conditions. Collected samples can be used for genetic analysis, mass spectrometry, single-cell cloning, and more.

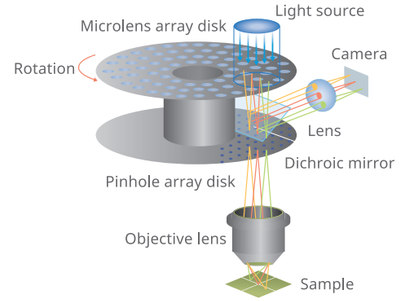


Live Cell Imaging Technology

Live Cell Imaging with Confocal Microscopy

It is based on the technology of the live cell imaging product developed by our company. High-speed, high-resolution 3D imaging is possible using our unique confocal microscope technology. Samples can be taken from targeted cells under a confocal microscope in an incubator environment. Time-lapse photography is also possible, allowing dynamic changes in the target cell to be captured. Since it is possible to record moving images during sampling and images before and after sampling, it is possible to compare the results of analysis of collected samples with cell imaging data.

Microlens enhanced dual wide Nipkow disk confocal



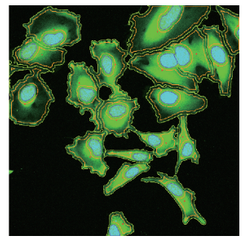
A Yokogawa proprietary multi-scan method utilizing approximately 1,000 laser beams on the observation region and tandem disks rotating at high speed. The disks comprise a pinhole array disk with approximately 20,000 pinholes arranged in an equal pitch spiral pattern, and a microlens array disk that focuses the excitation light laser into individual pinholes. Not only does this allow high speed imaging, but it also largely prevents phototoxicity and fluorescence photobleaching.

Microlens enhanced dual wide Nipkow disk confocal

The stage heater controls the temperature and humidity of the environment around the sample, and the gas mixer is connected to control the concentration of CO_2, enabling sampling while maintaining cell activity.

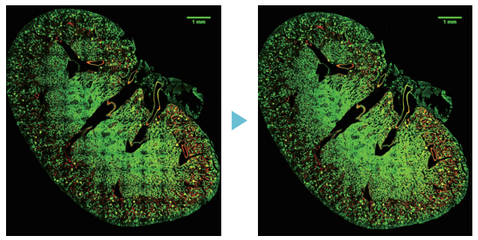
Automatic target selection

Target cells and sampling positions can be automatically selected by imaging analysis. (Targets can be automatically selected as shape of cells, size of nuclei, density of organelles etc.)



Tile imaging & Illumination uniform tool Uniformizer

Tile imaging can efficiently capture images of the entire wide field of view. Illumination uniform tool Uniformizer is installed as standard equipment, and image seams can be captured uniformly.



Specification

Automatic sampling functions	Tip diameter	3μm, 5μm, 8μm, 10μm	
	Incubator loader environment	37°C, 5%CO_2, humidified	
	Collection loader environment	37°C, 5%CO_2, humidified (for culture)/ 4°C (for cooling)	
	Collection loader compatible vessels	96-well PCR plate (0.1mL, 0.2mL)　Multiwell culture plate (96well)	
	Positioning precision of sampling	XYZ axial designated resolution: 0.1μm	
Imaging functions	Confocal scanning method	Microlens enhanced dual wide Nipkow disk confocal	
	Incubator loader compatible vessels	When sampling cell	φ35mm dishes*1 Microplate (6well, 24well, 96well)
		When observing cell	φ35mm dishes*1 Microplate (6well, 12well, 24well, 48well, 96well, 384well, 1536well) Slideglass*2
	Excitation laser wavelength	405, 488, 561, 640 nm(Uniformizer installed)	
	Emission filter	Filter size: φ25mm, Maximum slot number: 10 (Electric switching), Adjacent switching speed: 100msec	
	Transmission illumination	Bright-field, LED source	
	Objective lens	Dry lens: 4x, 10x, 20x, 40x　Long-working distance lens: 20x, 40x Note that only the 40x dry lens can be used for cell sampling.	
	Electric stage	XYZ axial designated resolution: 0.1μm	
	Z focus	Electric Z motor, designated resolution: 0.1 μm	
	Autofocus	Laser autofocus	
	Camera	sCMOS camera 2,000 x 2,000 pixel　Pixel size: 6.5 x 6.5μm	
	Workstation	Workstation for sampling, measurement, analysis, 24 inch display x2	
Other	Measurement software	Measurement functions (2D, 3D, Time-lapse, Map imaging), Viewing measurement and sampling data, Reporting functions (Image data, Video data) Whole cell sampling, Intracellular component sampling	
	Analysis software	Analysis functions (3D, Tile, Label-free, Texture analysis, Deep Learning, Gating), 3D viewer, Graphing functions, Reporting functions(Image data, Video data, EC50, IC50, Z'-factor)	
	External dimensions , Weight	Main unit: W1,217 x D643 x H595 mm/145kg Utility box: W275 x D432 x H298 mm/18kg Gas mixer: W275 x D432 x H298 mm/10kg Special purpose workstation: W172 × D471 × H414 mm/14kg Display: W531 x D500 x H166 mm/5.6kg	
	Operating environment	Temperature: 15 to 30°C　Humidity: 30 to 70%RH　no condensation (Recommended condition: 23±2℃, 40 to 70%RH)	
	Power consumption	Main unit, Utility box and Gas mixer: 1,200VAmax Workstation: 950VAmax　Display: 42VAmax ×2	
	Data formats (Measurement software)	Captured images: 16bit TIFF (OME-TIFF, TIFF) Output image data: TIFF, PNG, JPEG　Output video data: WMV, MPEG4	
	Data formats (Analysis software)	Numeric data: CSV　Output image data: TIFF, PNG, JPEG Output video data: WMV, MPEG4	

*1 A sample holder is required, and with it, up to 3 samples can be installed.　*2 A sample holder is required, and with it, up to 4 samples can be installed.

Installation example

Safety Precautions

* Read the user's manual carefully in order to use the instrument correctly and safely.
* This product falls under the category of class 1 laser products.

Contact us for more information and demonstration requests

Yokogawa Electric Corporation
Sales & Solution Center, Life Business HQ

2-9-32 Nakacho, Musashino-shi, Tokyo, 180-8750 Japan
Phone: (81)-422-52-5550
E-mail: SingleCell@cs.jp.yokogawa.com
https://www.yokogawa.com/solutions/products-platforms/life-science/

Represented by:

Printed in Japan, 111(VC) [Ed:01/b]

YOKOGAWA Co-innovating tomorrow™