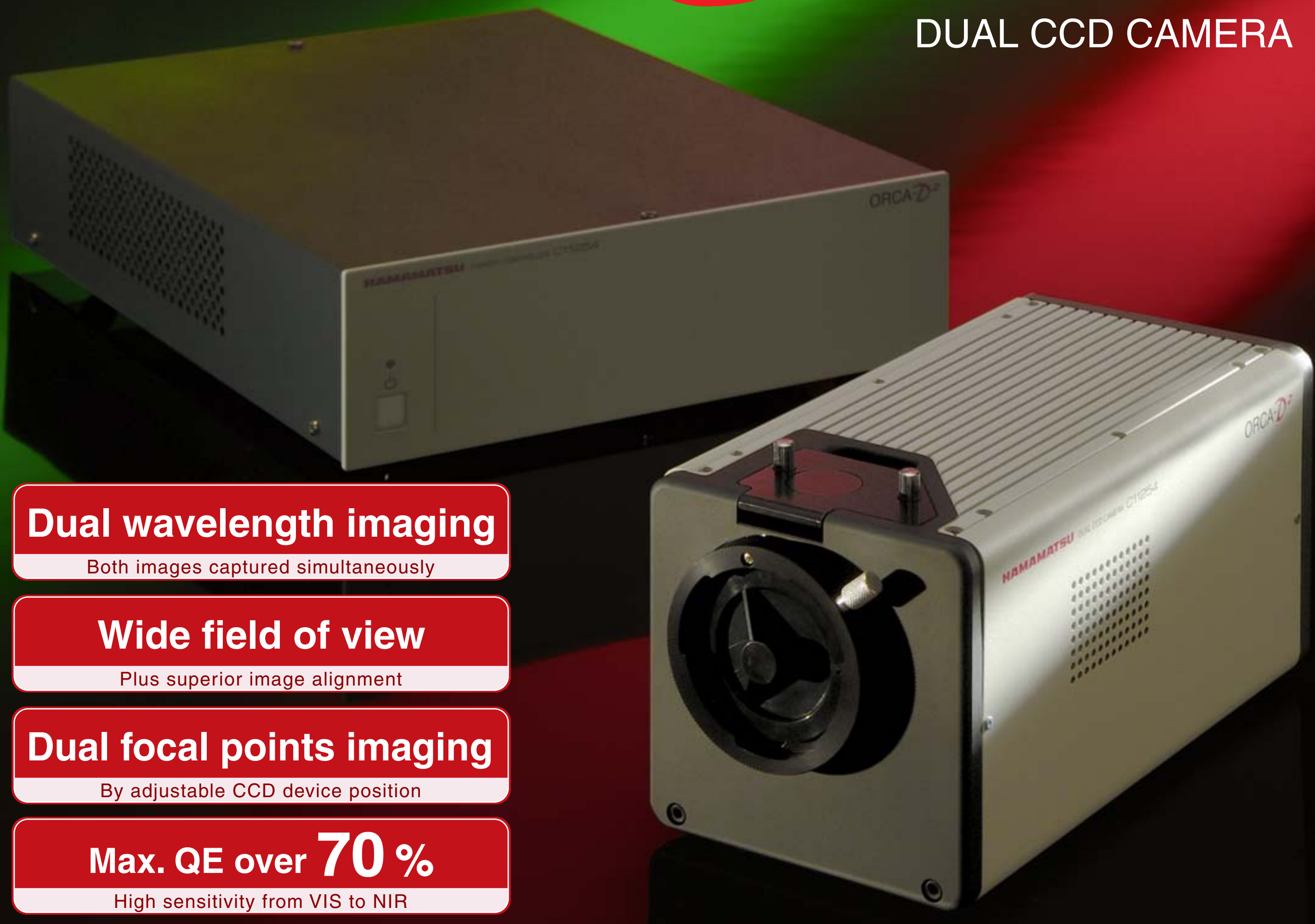
ORCA®-D²

New from Hamamatsu

DUAL CCD CAMERA



Dual wavelength imaging
Both images captured simultaneously

Wide field of view
Plus superior image alignment

Dual focal points imaging
By adjustable CCD device position

Max. QE over 70 %
High sensitivity from VIS to NIR

A One-of-a-kind Solution for Dual Wavelength Imaging

With the ORCA-D2, you can now capture dual wavelength images simultaneously, at wavelength ranges of your choice, and with a wide field of view – a unique combination that simplifies the challenges of conventional FRET and ratio imaging techniques. The ORCA-D2 contains two CCD devices. Interchangeable optical blocks are used to set the wavelength ranges for each CCD. During image capture, the camera automatically corrects focus, alignment and color shifting to produce high-quality images.
Other benefits of the ORCA-D2 include high sensitivity and low noise, courtesy of Hamamatsu's advanced CCD cooling technology; wide dynamic range; standard IEEE1394b interface; and Hamamatsu's proprietary vacuum-sealed chamber technology, which promotes long-term maintenance-free operation with even the most demanding applications.

APPLICATIONS

- Ratio imaging
- Single and dual wavelength fluorescence microscopy, FRET
- Blue to NIR fluorescence applications
- Colocalization and FISH applications
- Dual wavelength TIRF microscopy, real-time confocal microscopy
- Combined transmission and fluorescence imaging
- Multi-focal point imaging microscopy

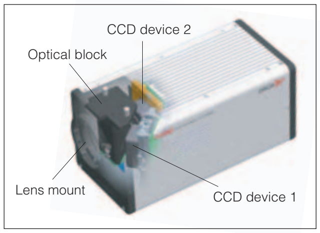
HAMAMATSU

FEATURES

Simultaneous capture of wide-field dual wavelength images

Dual wavelength imaging

The unique camera design makes it possible to capture simultaneous dual wavelength images with a full field of view, even when the intensities differ significantly. Such images are difficult to obtain with conventional FRET or ratio imaging techniques.



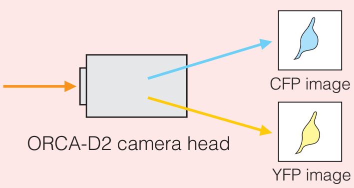
▲Internal structure of camera head

Selectable wavelengths

By changing optical blocks, you can select your wavelengths of interest. Hamamatsu offers three types of optical blocks for different wavelength ranges.

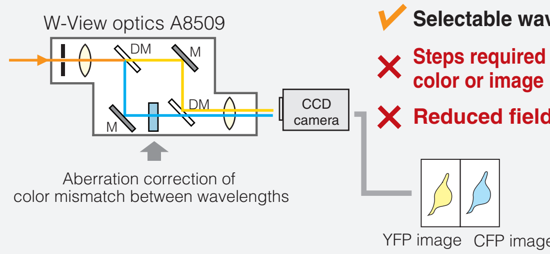
Comparison with conventional methods

ORCA-D2



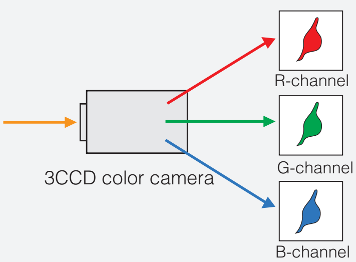
- ✔ Simultaneous capture at dual wavelengths
- ✔ Selectable wavelengths
- ✔ Auto-correction of color or image shifting
- ✔ Wide field of view

W-view optics



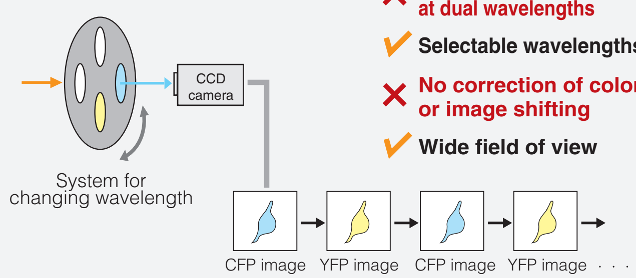
- ✔ Simultaneous capture at dual wavelengths
- ✔ Selectable wavelengths
- ✘ Steps required to correct color or image
- ✘ Reduced field of view

3CCD color camera



- ✔ Simultaneous capture at dual wavelengths
- ✘ No selection of wavelengths
- ✘ No correction of color or image shifting
- ✔ Wide field of view

Filter wheel

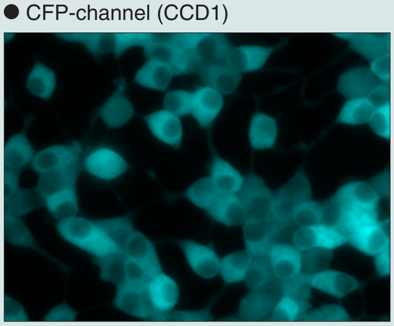


- ✘ No simultaneous capture at dual wavelengths
- ✔ Selectable wavelengths
- ✘ No correction of color or image shifting
- ✔ Wide field of view

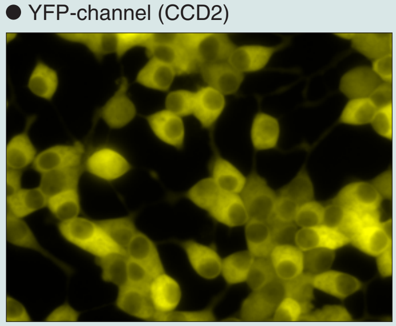
Application example 1 Ca^{2+} measurement using YellowCameleon 3.6 (optical block: DM 510 nm)

The following images are an example of ratio imaging. Separated CFP and YFP (FRET) are measured with dual CCD devices. This sequence observed the YellowCameleon 3.6 (Ca^{2+} sensor based on CFP-YFP FRET) expressed Ins-1 cell response with a depolarizing stimulus.

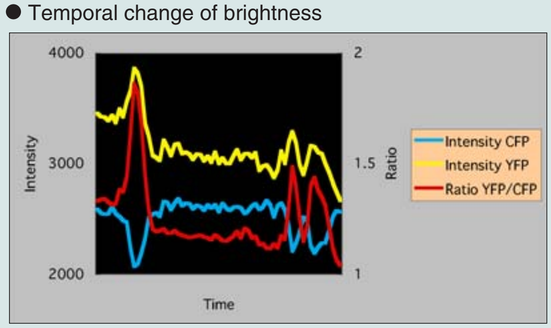
● CFP-channel (CCD1)



● YFP-channel (CCD2)



● Temporal change of brightness



Samples courtesy of Hideo Mogami, Ph.D.
Dept. of Physiology, Hamamatsu University School of Medicine
Dept. of Environmental Biology, Okazaki Inst. for Integrative Bioscience

Sample: Ins-1 cell (insulin-producing cell)
ORCA-D2 optical block: A11400-03 (DM 510 nm, Em1 483 nm/32 nm, Em2 542 nm/27 nm)
Microscope: Olympus IX71
Objective lens: Olympus LUCPlanFLN 60x, NA 0.70

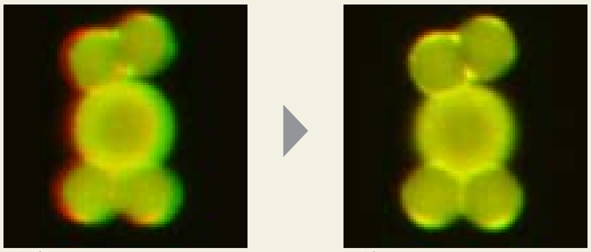
Easy setup

Auto-correction of focus and alignment

During dual wavelength image acquisition, the camera will automatically correct image alignment, color mismatch, etc. Hardware and special software work together to adjust focus* and alignment to your experimental setup. Calibration results are saved in the software, eliminating the need to readjust for the same setup.

*Please note that auto-correction may not be possible in some experimental setups.

Example of correction of focus and alignment



▲Before ▲After

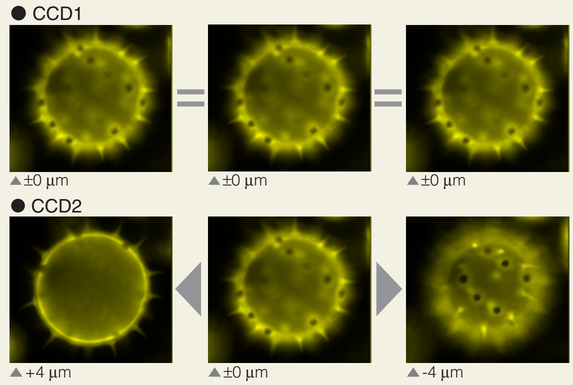
Sample: Multi fluorescent beads (6.0 μm to 7.9 μm)
ORCA-D2 optical block: A11400-04 (DM 550 nm, Em1 520 nm/35 nm, Em2 593 nm/40 nm)

Focus adjustment

Images with different focal points can be acquired at the same time through the use of the camera's special software and adjustable CCD device position.

Pollen with a 10 μm diameter

The following images are examples of using different focal points imaging at the same time.



High-sensitivity imaging from visible to near-infrared

Dual light mode

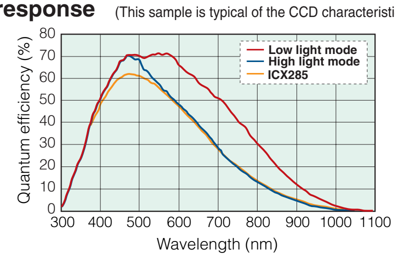
Two modes (high light, low light) are available for different situations. Low light mode provides high sensitivity over a wide spectral range from visible to near-infrared.

Max. quantum efficiency over 70 %

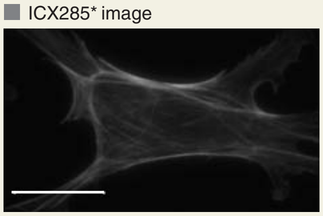
The ORCA-D2 offers high sensitivity from visible to near-infrared with its ER-150 interline CCD. In low light mode, sensitivity at 700 nm is nearly twice that of the ICX285* device used in most other scientific CCD cameras.

* ICX285 is a conventional high-sensitivity CCD device which is used for scientific instrumentation.

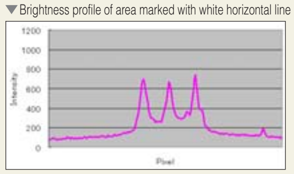
Spectral response (This sample is typical of the CCD characteristics, not guaranteed.)



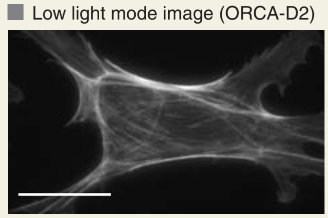
■ ICX285* image



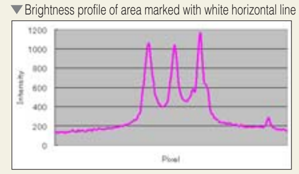
▼Brightness profile of area marked with white horizontal line



■ Low light mode image (ORCA-D2)



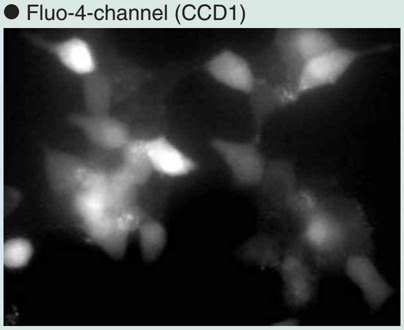
▼Brightness profile of area marked with white horizontal line



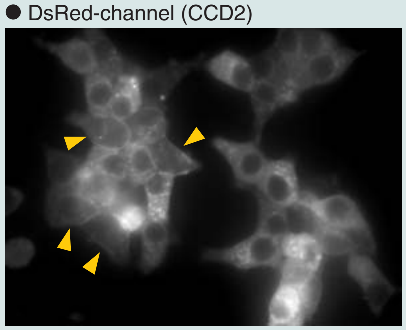
Application example 2 Ca^{2+} and PKC dual imaging using Fluo-4 and DsRed (optical block: DM 550 nm)

The images below show fluorescence signal from Fluo-4 AM loaded cells expressing PKC-DsRed when excited by 480 nm light. Observable in this time sequence is the translocation of PKC-DsRed to the cell membrane concurrent with free Ca^{2+} elevation in response to a depolarizing stimulus.

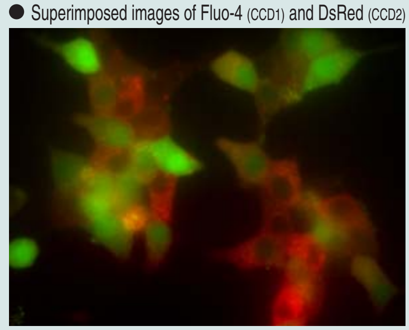
● Fluo-4-channel (CCD1)



● DsRed-channel (CCD2)



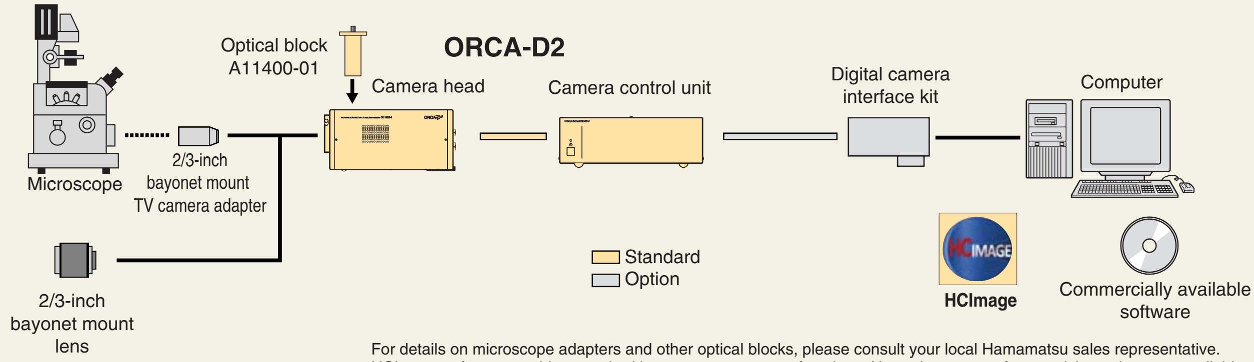
● Superimposed images of Fluo-4 (CCD1) and DsRed (CCD2)



Images courtesy of Hideo Mogami, Ph.D.
Dept. of Physiology, Hamamatsu University School of Medicine
Dept. of Environmental Biology, Okazaki Inst. for Integrative Bioscience

Sample: Ins-1 cell (insulin-producing cell)
ORCA-D2 optical block: A11400-04 (DM 550 nm, Em1 520 nm/35 nm, Em2 593 nm/40 nm)
Microscope: Olympus IX71
Objective lens: Olympus UPlanAPO 60x / 1.20 W

SYSTEM CONFIGURATION



For details on microscope adapters and other optical blocks, please consult your local Hamamatsu sales representative.
HCImage software provides standard image measurements functions. Upgrades to more feature-rich versions are available.

SPECIFICATIONS

Type number		C11254-10B (ORCA-D2)
Camera head type		Hermetic vacuum-sealed head
Imaging device		ER-150 progressive scan interline CCD
Effective number of pixels		1280 (H) × 960 (V)
Cell size		6.45 µm (H) × 6.45 µm (V)
Effective area		8.26 mm (H) × 6.19 mm (V)
Pixel clock rate		20.00 MHz/pixel
Readout speed	1 × 1	11.2 frames/s
	Binning 2 × 2	18.8 frames/s
	3 × 3	27.7 frames/s
	4 × 4	33.5 frames/s
Readout noise (r.m.s.) typ.		8 electrons
Full well capacity typ.	1 × 1	18000 electrons
	Binning 2 × 2	36000 electrons
Dynamic range typ. ①		2250 : 1 typ. (1 × 1)
Cooling method		Peltier device + Forced-air cooled
Cooling temperature		- 10 °C (Ambient temperature: + 20 °C)
Dark current		0.1 electrons/pixel/s (- 10 °C)
A/D converter		12 bit
Exposure time		117 µs to 60 s
Sub-array		Yes
Contrast intensification		Analog gain (10 times max.), offset
External trigger mode		Edge trigger, Level trigger, Synchronous readout trigger, Start trigger
Trigger output		Programmable timing output, Trigger ready output
Lens mount		2/3-inch bayonet mount (flange back focus 48 mm)
Interface		IEEE1394b-2002
External control		IIDC 1394-Based Digital Camera Specification Ver.1.32
Power requirements		AC 100 V to AC 240 V, 50 Hz / 60 Hz
Power consumption		Approx. 70 V·A
Ambient storage temperature		- 10 °C to + 50 °C
Ambient operating temperature		0 °C to + 35 °C
Ambient storage / operating humidity		70 % max. (no condensation)

① Calculated from the ratio of the full well capacity and the readout noise

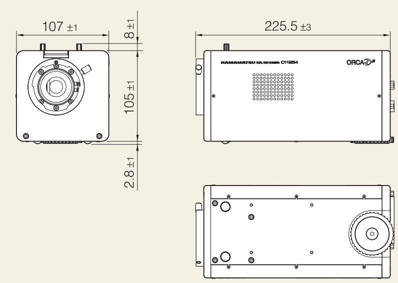
● Optical blocks

Type number	Optical block			Applications
	Dichroic	Em1	Em2	
A11400-01	None	0 %	100 %	1CCD camera
A11400-02	Half	50 %	50 %	Beam splitter
A11400-03	510 nm	483 nm / 32 nm	542 nm / 27 nm	CFP / YFP
A11400-04	550 nm	520 nm / 35 nm	593 nm / 40 nm	FITC / TRITC
A11400-05	630 nm	593 nm / 40 nm	692 nm / 40 nm	Cy3 / Cy5

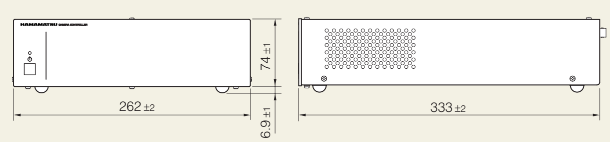
A11400-01 is included with camera. Other optical blocks are optional.

DIMENSIONAL OUTLINES (Unit: mm)

■ **Camera head** (Approx. 3.0 kg)



■ **Camera control unit** (Approx. 3.5 kg)



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