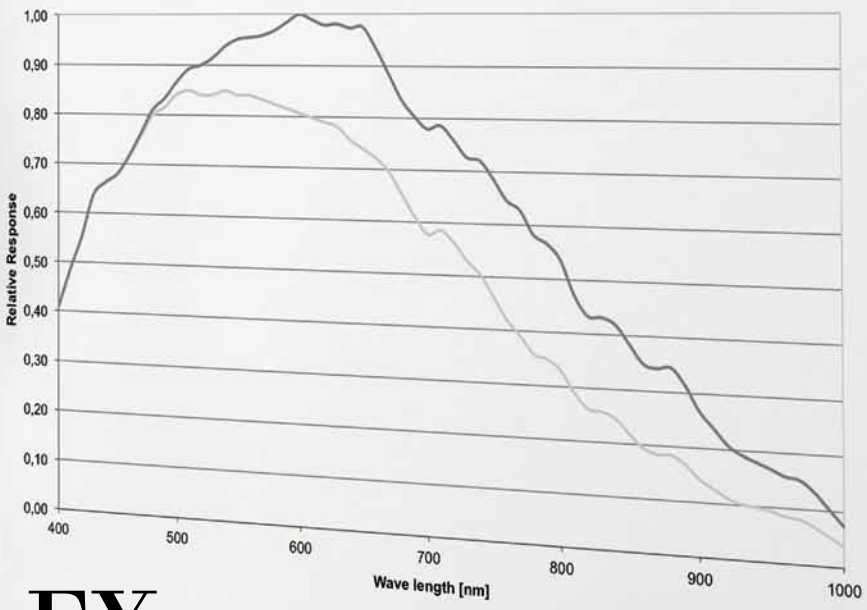
Living up to Life

Leica
MICROSYSTEMS



Leica DFC365 FX

High-end Scientific Fluorescence CCD Camera

Leica DFC365 FX: High-end Scientific Fluorescence CCD Camera

The Leica DFC365 FX offers excellent fluorescence documentation and fast time-lapse recordings under low light conditions. Based on the state-of-the-art Sony EXview HAD ICX285 sensor®, this cooled camera is exceptionally well suited for a wide range of fluorescence applications from basic fluorescence imaging to demanding applications such as TIRF, FRET, and Structured Illumination.

SUPERIOR IMAGE QUALITY

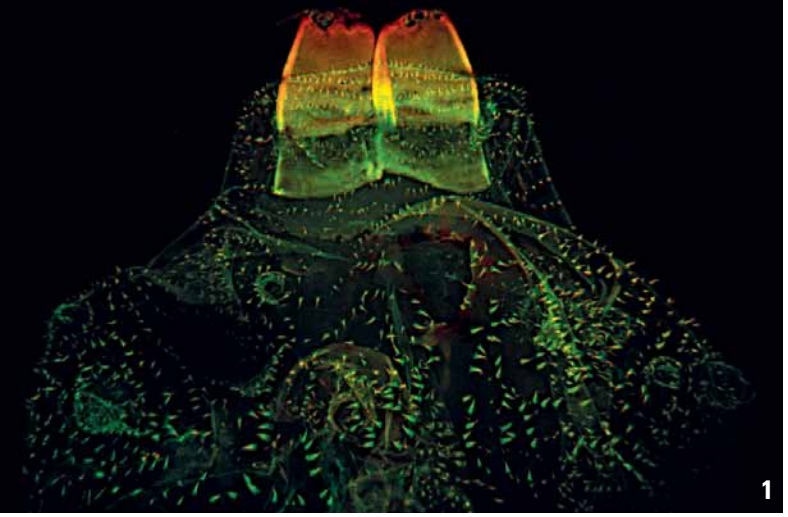
The new Leica DFC365 FX provides an excellent signal-to-noise ratio due to the regulated one-stage Peltier cooling of the sensor, which is highly effective even under live cell temperature conditions of 37°C. The read-out speed of the sensor can be down-regulated to 1.6 MHz for further noise reduction if needed. Any residual noise within the unlabeled background of the specimen is easily eliminated using the camera's "Black Balance" that resets a definable gray value to zero. The settings are absolutely reproducible and can be fine-tuned by the user.

EXTENDED VERSATILITY

The Leica DFC365 FX offers two acquisition modes: Standard and NIR (near infrared). The NIR mode is the better choice when increased quantum efficiency is needed in the spectral range starting at a wavelength of 700 nm. This widens the selection of fluorescence markers to include some that may not be suitable for imaging with standard CCD cameras. Also, this optional imaging mode protects live cells from photo-damage by offering reduced exposure times.

MAXIMUM DEPTH OF DETAIL

Fluorescence cameras generally offer a broad dynamic range to ensure that dark and bright signals with all details are captured within the same image. This means that tiny differences in fluorescence signal can be detected without time-consuming HDR (High Dynamic Range) multiple image acquisition. With a dynamic range of more than 69 dB, the Leica DFC365 FX covers all application requirements. No image information is lost, and very bright signals as well as black background can be monitored without missing details.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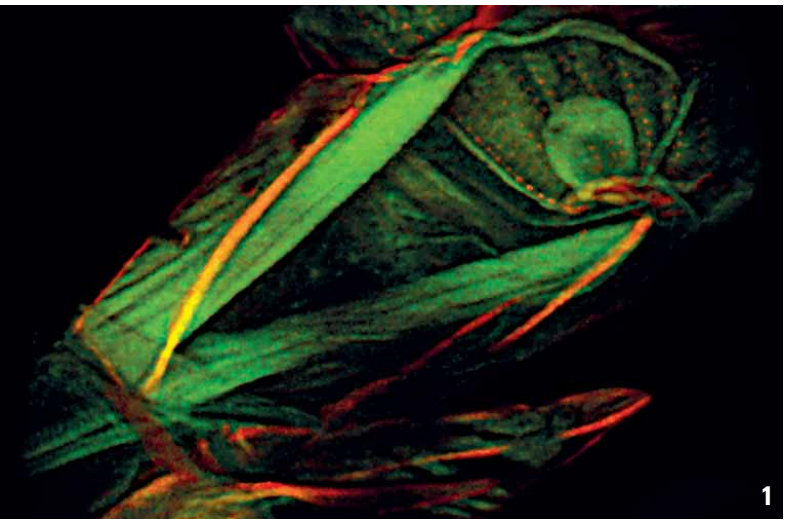
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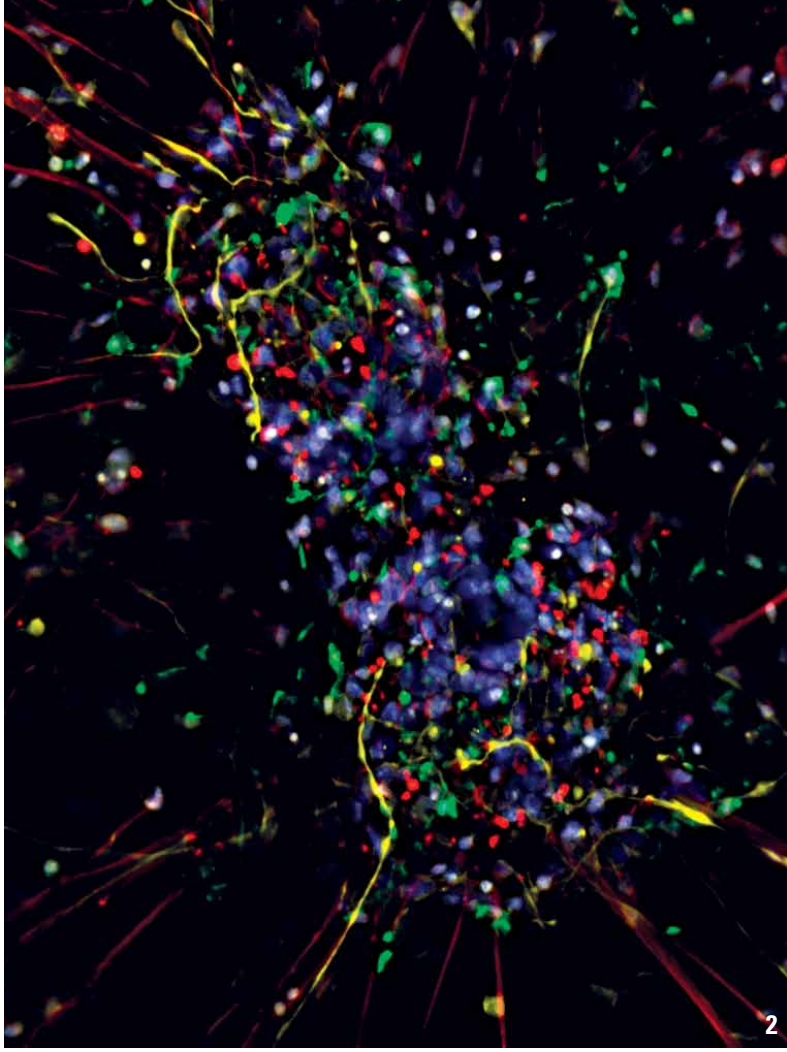
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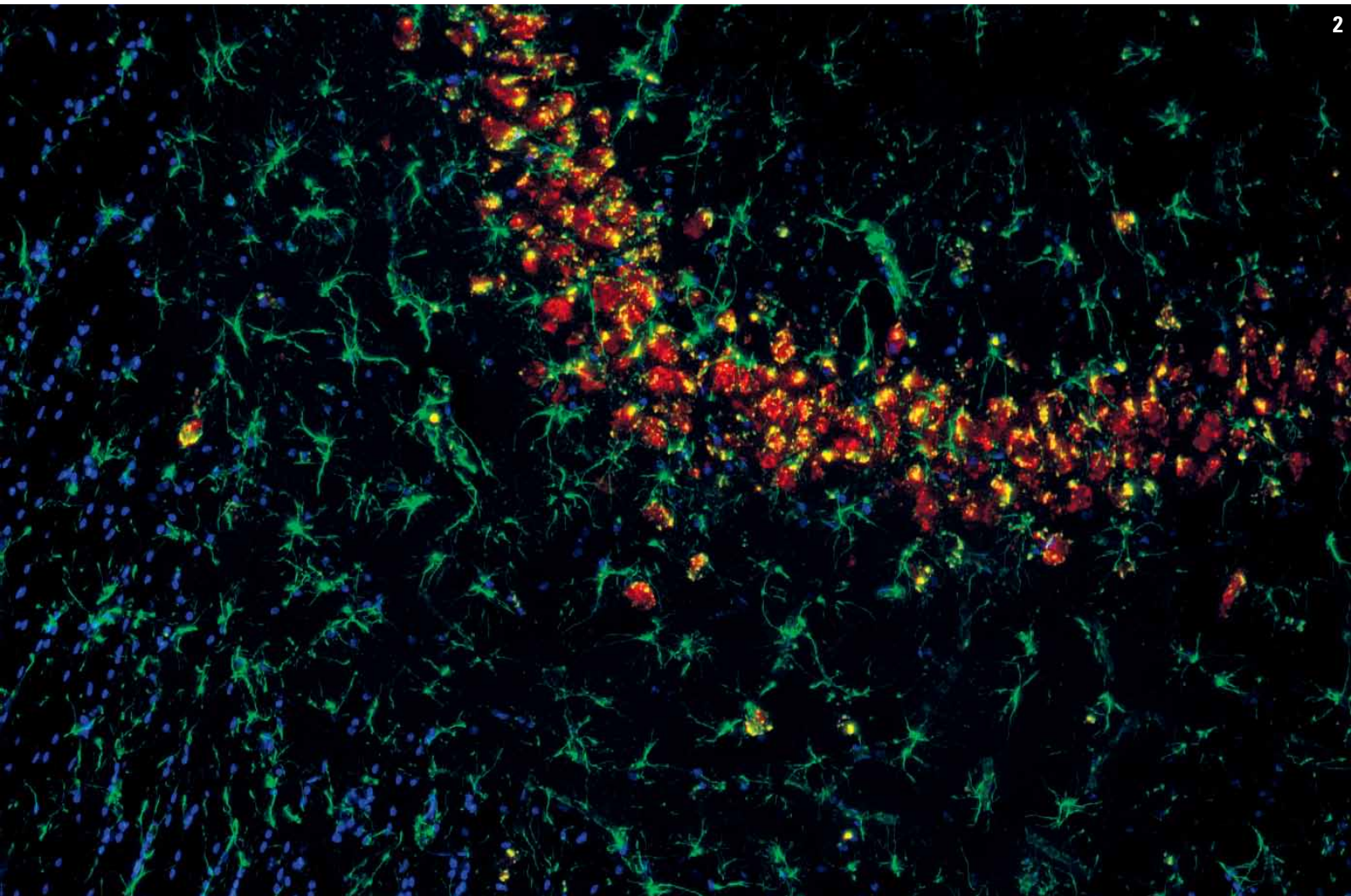
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Top left: Posterior spiracles of a 3rd instar *Drosophila melanogaster* larva. Maximum projection of 45 optical planes (z distance 1.5 µm) using 20x Plan Apo objective (NA = 0.6). Autofluorescence after excitation of light with the wavelengths 488 nm and 545 nm. Sample: Courtesy of Prof. Stephan Sigrist, Free University Berlin, Germany.

Middle left: Labium of an adult *Drosophila melanogaster*. 3D reconstruction of 48 optical planes (z distance 1.5 µm) using 20x Plan Apo objective (NA = 0.6). Green: Titin-GFP, a muscle protein. Red: autofluorescence of Chitin. Sample: Courtesy of Prof. Dr. Ralf Jacob, University Marburg, Germany.

1 APPLICATION EXAMPLE: STRUCTURED ILLUMINATION

For structured illumination techniques based on Optigrid® and related devices, the calculation algorithms can increase the noise of the acquired images. The Leica DFC365 FX noticeably reduces these effects by its significantly low dark and read noise, and provides high-quality images at an attractive price.

Top right: Cortical neuronal cells. Maximum projection of 21 optical planes (z distance 0.5 µm) acquired with 40x Plan Fluotar L objective (NA = 0.7). Deconvolved using the Blind Deconvolution algorithm. Red: Cy5 staining of βIII-tubulin. Green: Cy2 staining of Nestin, a marker of neuronal stem cells. Purple: Cy3 staining of DCX, a marker of immature neurons. Blue: DAPI, stains the nuclei.

Bottom: Section of rat brain. Maximum projection of 26 optical planes (z distance 1.5 µm) acquired with 40x Plan Fluotar L objective (NA = 0.7), "Black Balance" applied. Deconvolved using the Blind Deconvolution algorithm. Red: Cy2 staining of GFAP (glial fibrillary acidic protein). Green: Cy5 staining of NeuN, a marker of mature neurons. Purple: Cy3 staining of DCX, a marker of immature neurons. Blue: DAPI, nuclei. Both samples provided by FAN GmbH, Magdeburg, Germany.

2 APPLICATION EXAMPLE: FLUORESCENCE LABELING

Especially in neurobiology, there is a trend toward multiple simultaneous labeling in immuno-fluorescence and live cell imaging. This entails a greater use of fluorescence markers that emit light in the long-wavelength spectrum. Using the optional NIR mode, the Leica DFC365 FX is ideally suited to document these fluorochromes.

HIGH-SPEED LIVE CELL IMAGING

Unique in this class of fluorescence cameras is the flexibility of three pixel clocking rates to define readout speed of the sensor. In addition to the 1.6 MHz-mode, the Leica DFC365 FX offers 20 MHz and 40 MHz. The 40 MHz mode in particular is dedicated to fast, real-time time-lapse recording.
In combination with an overlapping mode, where the signal is read out while the next image is acquired, the user can easily execute high-speed experiments. With a maximum of 21 frames per second in full frame mode (1392 x 1040) or over 76 fps with 4 x 4 binning, the Leica DFC365 FX is the high-performer in this class of scientific fluorescence CCD cameras.

ULTIMATE CELL PROTECTION

Minimizing exposure times during time-lapse recordings is crucial to avoid photo-damaging the specimen. The sensor's high sensitivity and the careful selection of glass interfaces within the camera – in combination with NIR mode – are ideal requisites to protect cells and ensure optimal acquisition conditions during live cell imaging.

All the valuable data generated is easily and reliably transferred to the PC via the proven FireWire B interface with maximum 800 Mbit/sec transfer rate. No additional controllers or power supplies are needed – just "plug and play." The image buffer in the camera head ensures that no image is lost, even during fast time-lapse recordings.

EFFICIENT SYSTEM SOLUTIONS

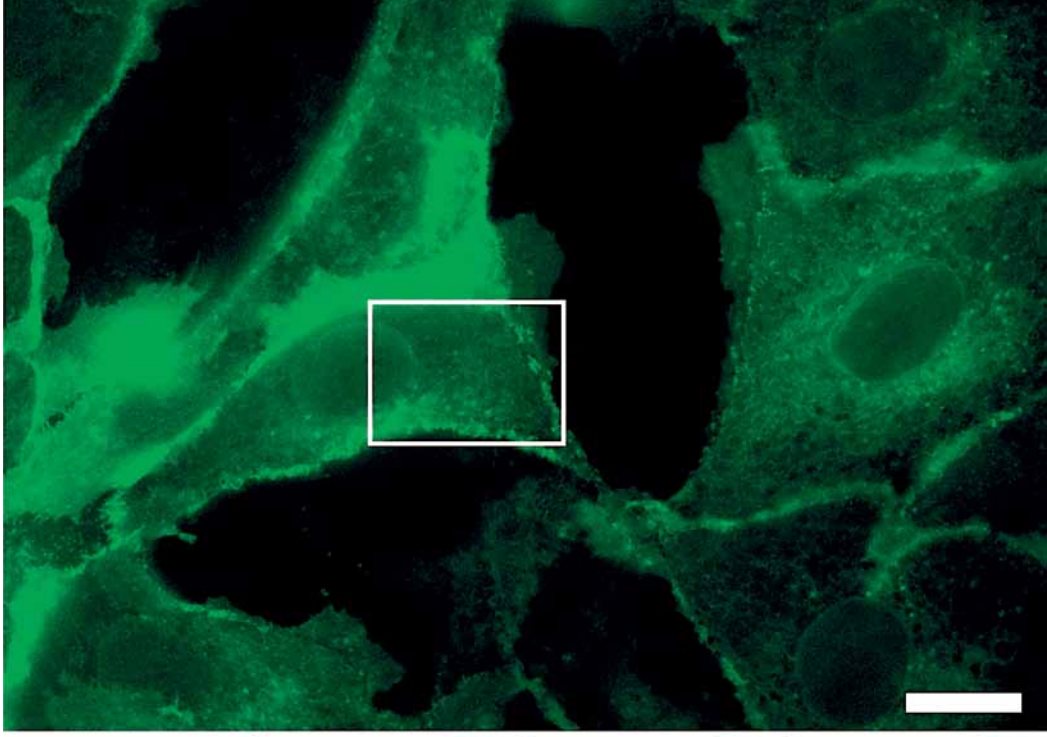
Synergy based on perfect integration: the Leica DFC365 FX works seamlessly with Leica Microsystems' three life science research software platforms to support an unlimited number of applications:

- Leica Application Suite (LAS) for basic image acquisition and analysis
- Leica LAS AF – the highly flexible and modular software platform dedicated to Advanced Fluorescence applications
- Leica MM AF (NX) based on MetaMorph® (NX) for sophisticated analysis applications

With the camera's leading-edge trigger capability, the complete microscope system is perfectly harmonized – including ultra-fast filter wheels, shutters, and precise attenuators.

BENEFIT FROM THESE HIGHLIGHTS EVERY DAY:

- Highly sensitive 1.4 megapixel Sony EXview HAD ICX285 sensor®
- Regulated Peltier cooling for excellent signal-to-noise ratio
- Two imaging modes: standard and NIR mode with at least 1.5 times enhanced sensitivity in the near infrared range of the spectrum
- Three pixel clocking rates (1.6 MHz, 20 MHz, 40 MHz) for full control of image quality and acquisition speed
- Groundbreaking acquisition speed with up to 21 fps in full frame and 122 fps in 8x8 binning mode



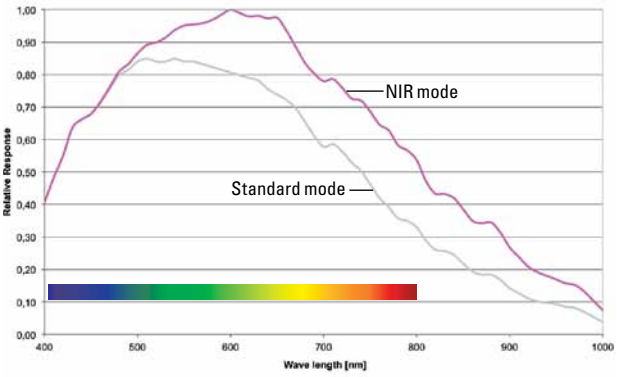
MDCK (Madin-Darby canine kidney) cells expressing p75-GFP. p75 is a common receptor for neurotrophins and is known to be transported to the apical membrane domain in MDCK cells. These images depict a region of one transfected cell where one single transport vesicle is shown (red arrow). Images were acquired using the camera's 40 MHz pixel clocking rate. Bar, 10 µm. Sample courtesy of: Prof. Dr. Ralf Jacob, University of Marburg, Germany.

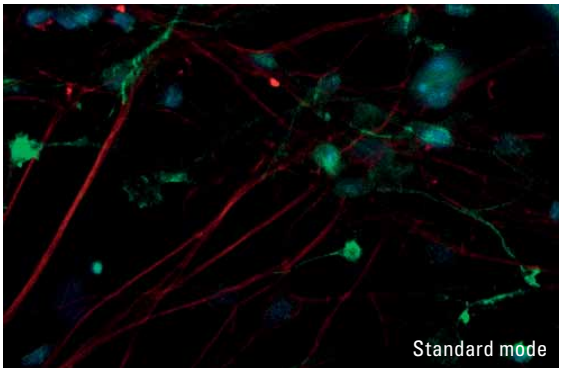
Leica Design by Christophe Apothéloz

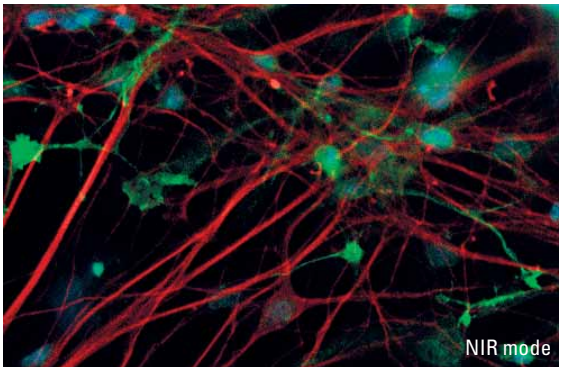
The Leica DFC365 FX is the perfect complement to the Leica DMi8 with AFC (Adaptive Focus Control) live cell imaging system and fits well on a wide variety of compound and stereo microscopes.

www.leica-microsystems.com

Quantum efficiency – relative response (curves are typical, not guaranteed)







Primary cultured cortical neuronal cells. Comparison of standard acquisition mode and NIR mode with enhanced sensitivity. Maximum projection of 35 optical planes (z distance 0.5 µm) acquired with 40x Plan Fluotar L objective (NA = 0.7). Red: Cy5 staining of βIII-tubulin, a neuron specific marker. Green: Cy2 staining of Nestin, a marker of neuronal stem cells. Purple: Cy3 staining of DCX, a marker of immature neurons. Blue: DAPI, stains the nuclei of the cells. Sample: FAN GmbH, Magdeburg, Germany.

LEICA DFC365 FX TECHNICAL DATA

Camera type	Digital monochrome, high-sensitivity cooled camera for fluorescence microscopy
Housing	Aluminum, Size (L × W × H) 132 mm × 74 mm × 71 mm
Weight	572 g

SENSOR

CCD sensor	Sony ICX285, EXview HAD CCD technology, 2/3" interline transfer progressive scan CCD
Number of pixels	1392 x 1040
Pixel size	6.45 µm × 6.45 µm
Sensitive surface	9.00 mm × 6.70 mm
Full well capacity	18.000 electrons typical
Exposure time	7 µseconds – 10 minutes *
Cooling	One-stage Peltier cooling
Dark noise	6 electrons typical (1.6 MHz, 12 bit)
Dark current	< 0.5 electrons/pixel/second
Dynamic range	> 69 dB typical (1.6 MHz)
Operating modes	Standard mode and NIR mode
A/D converter	14 bit
Bit depth	8 bit/12 bit
Partial scan	Freely definable ROI (region of interest), combination with binning possible
Analog gain	1× – 10×

IMAGE FORMATS	PIXEL	PIXEL CLOCK MHZ	FPS*
Full frame	1392 x 1040	1.6	1
		20	11
		40	21
2x2 binning	696 x 520	1.6	2
		20	21
		40	42
3x3 binning	464 x 346	1.6	3
		20	31
		40	63
4x4 binning	348 x 260	1.6	4
		20	38
		40	77
8x8 binning	172 x 130	1.6	8
		20	61
		40	122

SOFTWARE

Supported operating systems	Windows 7, Windows XP, Vista (32 and 64 bit)
PC Software	DFC Twain, Leica LAS, Leica LAS AF, MetaMorph (NX)®

INTERFACES

C-mount	0.7 x
Data	IEEE 1394b FireWire 9-pin single cable

MISCELLANEOUS

Power supply	12 V via FireWire cable
Power consumption	6 W (with cooling)
Operating temperature range	+5°C to +50°C
Air humidity	max 80%, non condensing

MICROSCOPES AND STEREO MICROSCOPE SYSTEMS (EXAMPLES)

Upright	Leica DM1000–DM6000 B
Inverted	Leica DMI4000 B–DMI6000 B
Systems	Leica SD AF (Spinning disk system), Structured Illumination, AM TIRF MC
Stereos	Leica MacroFluo, MZ10 F, M165 FC, M205 FA
Confocal	Leica TCS SP5, TCS LSI

*depends on software