

# Automated Parfocality Correction with the CELENA® S

#### Introduction

Parfocal distance refers to the distance between the mounting position of an objective and the focal plane of the specimen. In theory, if objectives of different magnifications have the same parfocal distance, focus is maintained when switching from one magnification to the next. Most microscope companies, including Olympus and Zeiss, manufacture objectives with the parfocal distance of 45 mm. Nikon recently introduced CFI60 objectives that have a longer parfocal distance of 60 mm. Although manufacturers try to produce objectives with the same parfocal distance across different magnifications, it is difficult to obtain perfect parfocality. High magnification objectives (above 40X) have a depth of field that is less than 5 µm, meaning that parfocal distance accuracy within 5 µm for perfect parfocality between objectives. This is a challenging feat when considering the precision required to manufacture an objective lens, and parfocality deviations across different objective lenses are normal.





Automated microscope systems with a motorized Z drive offer the convenience of automated parfocality correction. The CELENA® S Digital Imaging System from Logos Biosystems has a parfocality correction feature that automatically adjusts for the parfocal deviations that normally occur across different objectives. The CELENA® S has a motorized Z stage fitted with a precision ball screw and stepper motor system for repetitive positioning accuracy and the intuitive user interface makes setting up parfocality correction quick and simple.



OBJECTIVES panel in the CELENA® S user interface.



### **Setting Parfocality Correction**

The CELENA® S is precalibrated for the objectives installed at the time of order. The objectives should be calibrated when you add or change objectives.

#### To calibrate:

- 1. Click LIVE.
- 2. Turn off Parfocality correction in the OBJECTIVES panel.
- 3. Place a slide onto the stage.
- 4. Select an objective.
- 5. Focus as sharply as possible on the sample and click Set.
- 6. Repeat steps 4-5 with all objectives.

#### **Using Parfocality Correction**

- 1. Turn on Parfocality correction.
- 2. Place a sample onto the stage.
- 3. Focus on the sample.
- Select a different objective and wait 1 second. The Z-stage will adjust automatically to maintain focus.

#### **Notes**

- ! The parfocality correction feature calculates and adjusts for the relative distance between different objectives. When used properly, this helps you maintain focus when switching to a different objective. If the focal position of the currently selected objective is not correct, switching to a different objective will not correct the focus. Focus correctly with the selected objective prior to switching objectives.
- ! This feature can be used with objectives that are corrected for the same vessel bottom thickness.

#### **Specifications**

Imaging methods	Epifluorescence & transmitted light (brightfield & phase contrast)		
Illumination	LED filter cubes with adjustable intensity (>50,000 hr. lifespan)		
Channels	3 fluorescence channels and 1 transmitted light channel		
Objectives	Long working distance (LWD) and coverslip-corrected; 1.25X-100X		
Turret	5 positions		
Condenser	47 mm LWD condenser; 3-positions		
Stage	Mechanical X/Y stage & motorized Z stage; accommodates a onstage incubator		
Computer	Built-in dual core CPU, 128 GB SSD internal storage		
Camera	1.3 MP monochrome CMOS with 1280 x 1024 pixels		
Images	8 or 16-bit TIFF, JPG, BMP, or PNG		

## **Ordering Information**

CELENA®	) S Digital Imaging System	
CS20001	CELENA® S Digital Imaging System	1 set
CS20002	CELENA® S Digital Imaging System Starter Kit	1 set
Onstage I	ncubation System	
110501	Universal Heating System	1 set
110502	Gas Incubation System for CO <sub>2</sub>	1 set
110503	Gas Incubation System for CO <sub>2</sub> /O <sub>2</sub>	1 set
Objective	5	
110001	TC PlanAchro 4X Ph (NA 0.13, WD 17.3)	1 unit
110002	TC PlanAchro 10X Ph (NA 0.25, WD 7.6)	1 unit
110003	TC PlanAchro 20X Ph (NA 0.4, WD 7.0)	1 unit
110004	TC PlanAchro 40X Ph (NA 0.65, WD 2.8)	1 unit
110005	TC PlanFluor 4X (NA 0.13, WD 17.5)	1 unit
110006	TC PlanFluor 10X (NA 0.3, WD 7.5)	1 unit
110007	TC PlanFluor 20X (NA 0.4, WD 7.5)	1 unit
110008	TC PlanFluor 40X (NA 0.6, WD 2.9)	1 unit
110013	Plan Apochromat Fluor 1.25X (NA 0.04, WD 3.7)	1 unit
110014	Plan Apochromat Fluor 4X (NA 0.13, WD 17.2)	1 unit
110009	Plan Apochromat Fluor 10X (Coverslip-Corrected, NA 0.3, WD 8.6)	1 unit
110010	Plan Apochromat Fluor 20X (Coverslip-Corrected, NA 0.65, WD 0.7)	1 unit
110011	Plan Apochromat Fluor 40X (Coverslip-Corrected, NA 0.8, WD 0.2)	1 unit
110015	Plan Apochromat Fluor Oil 40X (Coverslip-Corrected, NA 0.85, WD 0 .2)	1 unit
I10012	Plan Apochromat Fluor Oil 100X (Coverslip-Corrected, NA 1.25, WD 0.19)	1 unit
LED Filter		
110101	DAPI (Ex375/28, Em460/50)	1 unit
110102	EGFP (Ex470/30, Em530/50)	1 unit
110103	RFP (Ex530/40, Em605/55)	1 unit
110104	mCherry (Ex580/25, Em645/75)	1 unit
110105	ECFP (Ex436/20, Em480/40)	1 unit
110106	EYFP (Ex500/20, Em535/30)	1 unit
110107	DSRed (Ex530/40, Em620/60)	1 unit
110108	Cy5 (Ex620/60, Em700/75)	1 unit
110109	Cy7 (Ex710/75, Em810/90)	1 unit
110110	Cy3/TRITC Long Pass (Ex530/40, Em570lp)	1 unit
110111	GFP Long Pass (Ex470/40, Em500lp)	1 unit
110112	Cy5 Long Pass (Ex620/60, Em665lp)	1 unit
110113	Custom Filters	1 unit
	lder Frames	4
110201	Universal Holder	1 unit
110202	25 mm x 75 mm Slide Holder, Two Positions	1 unit
110203	35 mm Cell Culture Dish Holder, Four Positions	1 unit
110204	60 mm Cell Culture Dish Holder, Two Positions	1 unit
110205	100 mm Cell Culture Dish Holder, One Position	1 unit
110206	25 cm <sup>2</sup> Nunc T-25 Flask Holder, Two Positions	1 unit
110207	75 cm <sup>2</sup> Nunc T-75 Flask Holder, One Position	1 unit
110208	25 cm <sup>2</sup> BD/Greiner T-25 Flask Holder, Two Positions	1 unit
110209	75 cm <sup>2</sup> BD/Greiner T-75 Flask Holder, One Position	1 unit
110210	Glass Hemocytometer Holder, One Position	1 unit



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