Micro-Manager's CRISP Control Plugin

Accessing the Plugin

The Plugin comes default in current versions Micro-Manager; if you don't have it in **Plugins** > **Beta**, update Micro-Manager to the latest nightly build.
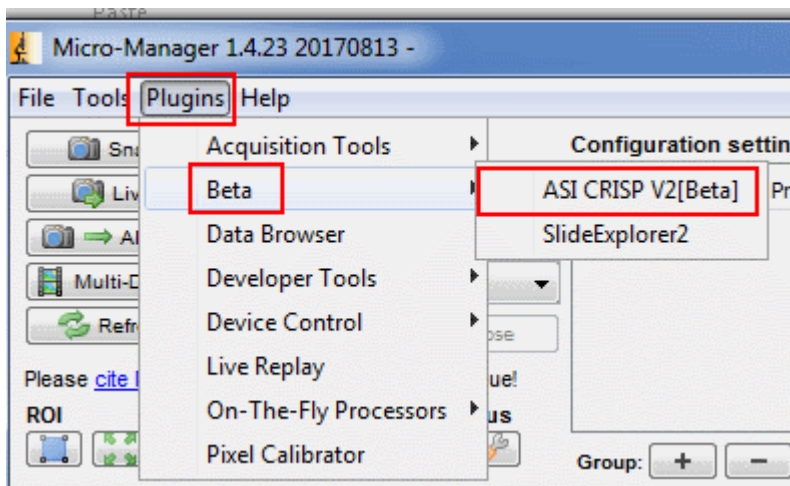

Fig. 1
Access the Plugin window
Micro-Manager Window > **Plugins** > **Beta**
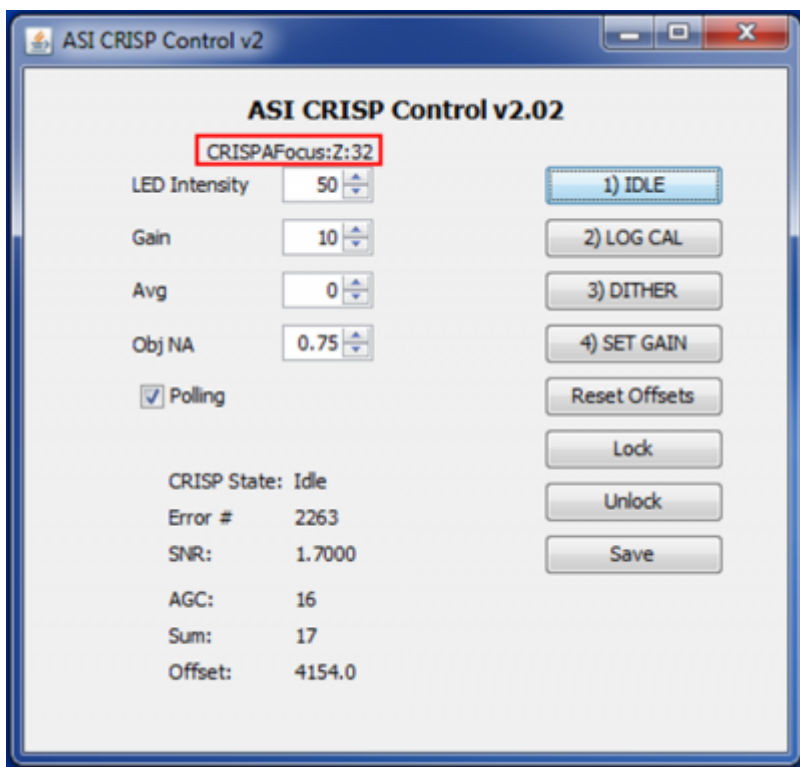
Plugin Controls Explained

Fig. 2
CRISP Plugin's control window

The red rectangle indicates which axis the Plugin is controlling.

Buttons

- **1) IDLE**: turns off the IR LED and stops position adjustments
- **2) LOG CAL**: puts the CRISP in Log Amp Calibration state and measures SNR
- **3) DITHER**: causes CRISP to dither in order to measure the Error #
- **4) SET GAIN**: puts the CRISP in Set Gain state
- **Reset Offsets**: resets the focus offset to zero for the present position
- **Lock**: sets the focal position
- **Unlock**: unlocks the focal position
- **Save**: saves the settings (for all of the spin boxes) onto the controller

Spin Boxes

- **LED Intensity**: controls CRISP's infrared (IR) LED from 0-100%
- **Gain**: controls the gain multiplier or loop gain; decrease the value if CRISP seems to oscillate or jitter
- **Avg**: the number of samples to be averaged
- **Obj NA**: the objective's numerical aperture

Check Box

- **Polling**: directs the Plugin to poll or query the controller at frequent intervals for CRISP state, error number, and SNR; uncheck when done with the 4-step calibration

Reported Values

- **CRISP State**: such as Idle, Calibrating, Ready, In Focus, etc.
- **Error #**: during DITHER higher error values are better; after SET GAIN the Error # will be close to zero
- **SNR**: (in decibels) should be at least 2 dB else CRISP performance may be unstable and lock will be lost easily; increasing LED intensity and performing the 4-step calibration again to increase the SNR
- **AGC**: minimum of ~20 is acceptable for average samples; may be much higher for very reflective samples
- **Sum**: indicates the amount of light hitting the photodiode; if outside 50-80, redo the 4-step calibration
- **Offset**: the difference between the locked-in focal position and the current position

Quick Setup Guide

- Start the 4-step calibration[1] by clicking the **1) Idle** button.
- Enter the objective's NA (which affects the dither).
- Click the **2) LOG CAL** button.
- Check the SNR display. If <2 dB, click through the rest of the 4-step calibration, increase the

LED Intensity, and begin the calibration again. If SNR is still low after the LED is at maximum intensity, proceed to the next step anyway.

- Click the **3) DITHER Button**.
- Check the Error #. It must be at least +/- 100; if it isn't, move the Lateral Adjustment Screw. The farther from zero the error number, the stronger the lock.
- Press the **4) Set Gain** button.
- Press the **Lock** button to preserve the focal position. (Press the **Unlock** button to release CRISP's focus lock.)
- If CRISP loses lock, repeat the calibration steps and try to get a higher error number after dithering.

Fig. 3
Lateral Adjustment Knob
Used to increase error number during dither

Additional Tips

If you would like CRISP to maintain focus at a certain position, but the error isn't 0, use the **Reset Offsets** button to make your desired focal position have "0" error.

Further Reading Refer to CRISP : Continuous Autofocus System for a description of CRISP operation and troubleshooting guides.

crisp, tech note, MicroManager, Micro-Manager
1)
Note that the 4-step calibration should always go through all 4 steps before re-starting; Log Amp Calibration has a countdown in the CRISP State display but otherwise you can quickly click through them to get back to the beginning.