# Lasers / Shutters Tab

The Lasers / Shutters tab is where laser power modulation and shutter control are configured along with some related hardware options such are uncaging galvos, PMT (high-speed) shutters and PMT gating. The PMT controls on this tab are here instead of on the Channels / PMTs / Preamps tab because they involve configuring analog output lines on the 6713 base card and it is nice to have all those on the same screen to avoid accidently reusing the same output for two tasks.

### *Lasers*

This section doesn't define which physical lasers are present, but rather defines which laser modulation signals are being provided to any physical lasers on the system. Some physical lasers are defined on the 2P Lasers tab to allow the software to communicate with the lasers directly.

Up to 12 lasers can be configured, any of which can be configured for imaging, uncaging, or both; note that there are only two switchable outputs on the GPIO box (see the GPIO Laser Modulation Signal Routing section) so without additional switching hardware is it only possible to have 2 lasers used for both imaging and uncaging. With additional switching hardware the Alternate Beam Routes section can be used to allow additional lasers to be used for both imaging and uncaging.

For each laser there are a number of columns to fill in:

- **UI Text** specifies the text that will be visible above the laser slider in Prairie View as well as the text used to identify a specific laser elsewhere in the UI. This name must be unique to avoid unpredictable behavior
- **OUT Min / Max** are the minimum and maximum drive signal values specified in the units used by the selected device type, more details can be found in the **Device Type Notes** section beneath the laser grid
- **Display Min / Max** are the minimum and maximum powers used in Prairie View. Historically we've used 0-100, or more recently 0-1000, but in most cases those values make no sense. For example most Pockel cells are driven with a maximum of 1.9 Volts or 390 DAQ units, meaning that there are only 390 discrete values, so having a display range with 1000 discrete values makes no sense
- **Small / Large Change** (in display units) specify how much the laser power changes when interacting with the laser power slider controls. Clicking the arrows, or using the up/down keys in the text box, makes a small change. Holding the Ctrl key while making a small change, or clicking the laser power slider itself, makes a large change
- **Lag Time [µs]** specifies how long it takes for a laser to modulate to a specific power after the software updates the drive signal. Unlike the galvo lag time this is just use to turn on the laser a bit earlier and has no effect on the off time
- **Device Type** specifies both what hardware will be used to modulate the laser power and which light path the laser will be used on (imaging and/or uncaging). The following options are available (legacy options may be encountered in the field, but should not be used for new systems):
    - **Imaging (DAQ Buffered)** specifies an imaging laser modulated using the 6713 option card (or in a pinch the 6052E card).
    - **Uncaging (Mark Points)** specifies an uncaging laser modulated using the 6713 base card (aka PVAsychronousAnalogOutput).
    - **Imaging/Uncaging (Alt. Beam Route)** specifies a laser used for both imaging and uncaging and requires that an Alternate Beam Route be configured to determine which light path the laser is currently being used with. The imaging modulation (**Line**) is done with the 6713 option card and

the uncaging modulation (**Shared Line**) is done with the 6713 base card (aka PVDAQBufferedShared).

- **SFC** specifies an imaging laser modulated using the SFC control box.
- **Sutter Lambda DG-4/5** specifies a camera imaging light source modulated using an included Sutter control box.
- **GPIO Box (Legacy)** specifies an imaging laser modulated by the GPIO box.
- **Device Control Box (Legacy)** specifies an imaging laser modulated by the device control box.
- **NI Analog Output (Legacy)** specifies an imaging laser modulated by an NI card.

- **Device** is used when multiple devices/cards are present to specify which one to use.
- **Line** is used to specify which line on a particular device/cards should be used to modulate the laser.
- **Shared Line** applies only to the **Image/Uncaging (Alt. Beam Route) Device Type** and specifies which line on the 6713 base card should be used for uncaging power modulation.
- **GPIO Box PA DAC** applies only to the **GPIO Box (Legacy) Device Type** and specifies which analog output line on the GPIO box should be used to modulate power for a photoactivation mask. It was an unnecessary complication and never really used, instead the the primary DAC was usually just used for both (see GPIO Laser Modulation Signal Routing section).
- **2P Laser** specifies which laser configured on the 2P Lasers tab, if any, is being modulated by this laser slider. It is used mainly to automate 2P laser power calibration.
- **Shutter** specifies which of the shutters defined in the Shutters section is used to block any bleed through light from the laser from reaching the microscope/sample. If a shutter number is specified, that shutter will only open when this laser slider (or another with the same shutter number selected) is greater than the minimum/zero value. Alternatively if NA is selected then no shutters will be opened for this laser, but if the selection is left blank ALL shutters will be opened when the laser slider is greater than the minimum/zero value (this is a catch all when upgrading from versions where this field didn't exist).
- **Alternate Beam Route/Shutter** (see Alternate Beam Routes section).

### *GPIO Laser Modulation Signal Routing*

This section configures which signals are routed through PC1 Out and PC2 Out on the back of the GPIO box in the following situation:

Note that the selections are for the PC_#_IN connectors on the inside of the GPIO box which in turn need to be connected to the appropriate source signals. In any cases where switching signals is undesirable simply select the same source signal in multiple places

- **Input** is used by default when none of the following cases apply, normally PC_1_IN and PC_2_IN for analog outputs 0 and 1 on the 6713 option card for imaging, and PC_5_IN for analog output 6 on the 6713 base card for uncaging
- **Mark Points / Uncaging Input** is used for systems without uncaging galvos to route a signal from the 6713 base card to modulate the laser during sequential uncaging experiments, this will be disabled when uncaging galvos (or a high-speed switch) is present
- **Alternate Beam Route Input** is used for an **Imaging/Uncaging** laser to specify which alternate source

signal to use when the selected alternate beam route is enabled (see Alternate Beam Routes section)

- **Photoactivation Input (Legacy)** can be used to route the **GPIO Box PA DAC** for photoactivation masks instead of the imaging DAC, but it is just as easy to select the imaging DAC here and ignore the PA DAC

### *Disable Interlaced Scan Patterns*

This option is used to hide the line interlacing option in Prairie View, it was requested to hide the option in the specific case where only one laser is present.

### *Photoactivation (Legacy)*

This option only applies to systems using NI breakout (2090) boxes in conjunction with a smaller photoactivation switch box to route a photoactivation mask specific blanking pattern to the device control box. The blanking signal, or Output Device/Line, is generated on analog output 1 (Line 1) on the 6052E card (Device 1), and the switch/relay signal uses digital output 1 on the same card. Since these settings are now hardcoded rather than configurable it may be necessary to update the wiring/hardware prior to updating Prairie View on some older systems.

### *Shutters*

This section is used to define any hard shutters preventing bleed through laser power from entering the microscope and hitting the sample. Up to 6 shutters can be defined by filling in values for the following columns:

- **UI Text** is currently only displayed in the Maintenance dialog next to the controls to manually fore the shutter to stay open or closed
- **Default State** (normally true) determines whether a logic high (True, 5V) or logic low (False, 0V) is used to open the shutter
- **Device Type** determines which piece of hardware is generating the signal to open and closed the shutter:
    - **PVDigitalOutputDeviceControlBoxShutter** (Legacy) is used when a device control box is controlling the shutter
    - **PVDigitalOutputDAQINTShutter** (Legacy) is used when an NI card is controlling the shutter
    - **PVDigitalOutputPrairieGPIOBoxShutter** is used when a GPIO box is controlling the shutter
- **Device** is used to specify which card/device is used to drive the shutter when more than one exist
- **Line** is use to specify which output/line is used to drive the shutter based on the selected hardware
- **Delay [ms]** is used to specify how long it takes the shutter to open and any vibrations to subside before imaging or uncaging can begin
- **Light Path** determines if the shutter is on the imaging or uncaging light path, in turn the light path with be assumed for any lasers with this shutter selected
- **Alternate Beam Route / Light Path** (see Alternate Beam Routes section)
- **High Speed Shutter Box Input** is used to specify which input of the high speed shutter control box the shutter's drive signal is plugged into. Prairie View will then automatically program the high speed shutter controller logic based on how the shutter is configured

### *PMT Shutters*

This section is used to specify that high speed shutters are installed on the system. Check the **High-Speed**

**Shutter Box Present** checkbox if one is installed then continue to set the following:

- **Uncaging Line on 6713 Base Card** specifies which analog output on the 6713 base card will be plugged into the Uncaging input on the back of the high-speed shutter box. Mark Points automatically generates this signal to make sure the PMT shutters are closed during uncaging laser/LED pulses
- **Shutter Type** specifies which type of physical shutters are installed on the PMTs. If they are mixed select the largest shutter present. Displayed underneath are the DIP switch settings for the shutter driver board inside the high-speed shutter box for the selected shutter type
- **Additional Close Delay (ms)** specifies how much earlier to close the PMT shutters before an uncaging pulse compared to the hardware specifications hardcoded into Prairie View. By default this is 0 meaning just use the hardware specifications, that delay can be extended (shutter closes earlier) with a positive number or shortened (shutter stays open longer) with a negative number
- **Additional Open Delay (ms)** specifies how much later to open the PMT shutters after an uncaging pulse compared to the hardware specifications hardcoded into Prairie View. By default this is 0 meaning just use the hardware specifications, that delay can be extended (shutter stays closed longer) with a positive number or shortened (shutter opens sooner) with a negative number

Generally only the open delay is extended to prevent any phosphorescence from tripping the PMT safety circuits.

Also remember to set the **High Speed Shutter Box Input c**olumn in the Shutters grid, and the **High Speed Shutter Index** column in the PMTs grid on the Channels / PMTs / Preamps tab.

### *PMT Gating*

This section is used to specify that gated PMTs are present so that a gating signal line can be generated on an analog output line of the 6713 base card. The gating signal will be automatically generated by Mark Points to make sure the PMTs are gated during an uncaging pulse. If the uncaging pulse is longer than the maximum gating time a warning will be displayed in the Mark Points grid.

### *Alternate Beam Routes*

This section specifies what abilities a system has to flip lasers to be used for either imaging and uncaging, and in conjunction with the laser and shutter grids, how the system is set up physically. The first step is to fill in the columns for the alternate beam route, each alternate beam route can be thought of as a specialized utility button which can be used to toggle a subset of defined lasers between imaging and uncaging. Those basic columns are as follows:

- **UI Text** is the text that will be displayed under the laser sliders in the Alternate Beam Routes section next to the button which will toggle the alternate beam route. The affected laser name, or names, would be good to have here
- **Disabled Text** is the text that will be displayed on the alternate beam route button to the right of the UI Text when the alternate beam route is disabled. This would normally say something imaging or uncaging depending on the default use of the affected laser(s)
- **Enabled Text** is the text that will be displayed on the alternate beam route button to the right of the UI Text when the alternate beam route is enabled. This would normally say something imaging or uncaging depending on the alternate use of the affected laser(s)

  For example, UI Text: "1064 nm laser is" Disabled Text: "Uncaging" Enabled Text: "Imaging"

- **Device Type** specifies whether or not there is a digital output driving some automated optical/electrical

elements, the options are:

- **PVDigitalOutputDAQInt** specifies that an NI card will be used to drive a digital output high/low when the alternate beam route is toggled on/off. The **Device** specifies which card will be used and the **Line** specifies which digital output line will be used
- **PVDigitalOutputPrairieGPIOBox** specifies that the GPIO box will be used to drive a digital output high/low when the alternate beam route is toggled on/off. The **Line** specifies which digital output line will be used
- **Manual** specifies there is no automation and anything needing to be moved will be handled manually

The second step once an alternate beam route has been defined is to set the affected laser(s) to switch shutters when the alternate beam route is enabled, or set the affected shutter(s) to change light paths:

- If the laser beam is being physically routed to go through a different shutter the **Alternate Beam Route / Shutter** columns in the laser grid are used to specify which shutter will be used for the alternate beam route.
- If the laser is using the same shutter then the shutter light path will need to be changed using the **Alternate Beam Route / Light Path** columns in the shutters grid.

Finally in the GPIO Laser Modulation Signal Routing section in the upper right the affected PC # Out source signals will need to be set appropriately under **Alternate Beam Route Input**. If more than two lasers are being switch between imaging and uncaging, and the 2 laser modulation outputs on the GPIO box are insufficient, an additional switch block can be used any driven by a digital output configured in the Alternate Beam Routes grid.

### *Uncaging*

This section specifies what hardware is present on a system to facilitate uncaging experiments. Three options for basic uncaging hardware are mutually exclusive:

- **Uncaging Galvos Present / Simultaneous Uncaging** specifies that there are uncaging galvos present on the system and an uncaging light path independent from the imaging galvos and light path. In this configuration uncaging can happen while imaging.
- **No Uncaging Galvos / "Simultaneous" Uncaging** specifies that there are no uncaging galvos, but instead a switch will be used to take control of the imaging galvos (and optionally lasers) to perform the uncaging during the imaging (aka high-speed switch), but will interrupt the imaging. The **Uncaging Switch Line on 6713 Base Card** specifies an analog output to drive the switch, and the **Switch Delay (ms)** specifies how long it takes the switch to swap between the two inputs.
- **No Uncaging Galvos / Sequential Uncaging Switch** (legacy) specifies a switch will be used to uncage with the imaging galvos sequentially (aka low-speed switch). This option is only relevant for older systems using NI/2090 breakout boxes without a GPIO box. The GPIO box handles this switching itself for the galvos and the laser switching is handled in the GPIO Laser Modulation Signal Routing section.

Additionally there is an **SLM Present** option, which only applies when a dedicated uncaging path is present with uncaging galvos, which should be checked when an SLM is installed on the system. The **Trigger Line on 6713 Base Card** specifies which analog output will be connected to Input B on the SLM control box. Mark Points will then automatically generate that signal to trigger mask changes at the appropriate times. In order to test some SLM functionality without any SLM hardware the hardware can be emulated by prefixing the trigger line with one or more 9's (i.e. 91 would emulate an SLM and use analog output 1 to generate the trigger signal).

## Ultima / Dual Scan / Interlock Tab

The Ultima / Dual Scan / Interlock tab is where 2P confocal based acquisition modes are configured.

### *Ultima Enabled*

Checked when galvo mode is available on a system.

If **Include park sample in galvo drive waveforms** is checked the very last sample of each frame will park the galvos to minimize bleed through power at the sample.

### *Mode Servo Present*

Checked if servos are used on the system to move optics for AOD/resonant mode or SLM. All new systems will use a quad servo board.

**Disable Servo After Move** should normally be checked to avoid noise/vibration caused by the servos straining to maintain a preset position, the only case it makes sense to not disable the servos is for the SLM dropper which the servo cannot hold up without being powered, in most other cases magnets are used to maintain positions.

### *Interlock Present*

Checked to indicate there is a safety interlock installed and wired to the quad servo board. When enabled Prairie View will monitor this signal and pop up a message box when the interlock is tripped letting the user know the scan head cover had been opened.

### *AOD Power*

Checked for AOD systems to specify an analog output to provide 0.8 Volts to the VCO/PA AM input on the back of the AOD control box.

### *Dual Scan System*

Checked if an AOD or resonant scanning is installed on the system. Additionally the **High Speed Device Type** should be selected.

As neither high speed scanner is capable of panning the conventional X galvo is used to pan in these scan modes instead of scan. By default line 0 on the 6110 card is used (**Device Type** PVanalogOutputDAQINT, **Device** 3, **Line** 0). For Signal Core systems this setting is hardcoded in Prairie View to use the imaging X galvo output and any pan device/line settings in the configuration utility are ignored.

Depending on where the shutters are in the imaging light path it is possible that resonant/AOD mode will have it's own shutter. When that is the case check the **Use shutter # instead of shutter # option** and fill in the shutter indices appropriately.

**DAQ Buffered Update Clock (Legacy)** defaults to PFI Line 3 on the 6713 option card and is only used for systems with NI hardware. Note that Signal Core systems will assume this is wired to PFI3 in the GPIO box and changing this setting will have no effect.

## Channels / PMTs / Preamps Tab

The Channels / PMTs / Preamps tab is where PMTs, imaging channels, and preamplifiers are configured.

### *PMTs*

This section can define up to 8 PMTs, or more accurately up to 8 analog output signals can be defined which are each used to modulate the high voltage level of a PMT. For each PMT the following columns need to be filled in:

- **UI Text** specifies the text that will appear above the PMT slider. Usually this is the channel name and/or color

- **OUT Min / Max** specifies the minimum and maximum drive signal level in raw units used by the specific Device Type, see the **Device Type Notes** to the right of the PMT grid for more information

- **Display Min / Max** specifies the minimum and maximum display values for the PMT gain, usually in Volts to match the high voltage display on the PMT control box

- **Small / Large Change** (in display units) specify how much the PMT gain changes when interacting with the PMT gain slider controls. Clicking the arrows, or using the up/down keys in the text box, makes a small change. Holding the Ctrl key while making a small change, or clicking the PMT gain slider itself, makes a large change

- **Device Type** specifies what hardware will be providing the analog output drive signal to the PMT control box:

    - **PVAnalogOutputDeviceControlBox** means the analog output will be provided by the device control box, the **Device** specifies the analog output index used

    - **PVAnalogOutputDAQINT** means the analog output will be provided by an NI card, the **Device** specifies the card ID and the **Line** specifies the analog output index on that card

    - **PVAnalogOutputPrairieGPIOBox** means the analog output will be provided by the GPIO box, the **Device** specifies the analog output index used

    - **Quad GaAsP Controller** means the analog output will be provided internally by the Quad GaAsP Controller box, the **Device** is the index of the controller box (in case there is more than 1) and the **Line** specifies the PMT index on that box. If there is a second Quad GaAsP Controller the **2nd Quad GaAsP Controller** checkbox must be checked and the serial number filled in so that Prairie View can tell them apart

- **High Speed Shutter Index** specifies the index of the PMT shutter control output going to the PMT from the High Speed Shutter Control box. This is used by Prairie View to keep unused (zero gain) PMTs shuttered all the time, otherwise all the PMT shutters with the imaging shutter and unused PMTs are exposed unnecessarily

- **Gated?** should be checked if the PMT is a gated PMT. This is used by Prairie View to properly initialize the Quad GaAsP Controller box.

### *Channels*

Up to 4 imaging/display channel can be enabled and named. Even if a system doesn't have 4 discrete imaging channels, additional channels can be used for interlaced scanning as well was display of multi-track data in playback mode, so for future flexibility it is probably best to keep all four enabled in most cases.

The **UI Text** in the text that displays on the channel buttons on the image windows as well as a few other places in the UI, it's best to keep the names short so they fit in those smallish buttons. As described underneath the channel controls the **UI Text** for each blade in the preamplifiers section will be used in cases where input signal routing hardware is present in the GPIO box to allow for up to 8 discrete channels (four of which can be imaged simultaneously).

### *Preamplifiers*

This section specifies what preamplifiers are present on the system (up to 2) and which blades are populated in

each preampflifier. Simply check the **Preamplifier Present**, and **Second Preamplifier Present**, checkboxes appropriately and then check **Blade # Present** for each blade populated in the preamp(s). If there are two preamps the only way to tell them apart is their name when connected over USB with a utility like FTermDI, unplugging one and seeing what name appears will help distinguish the two.

The **Do not reset at startup** checkbox should be checked for all new systems, but will be left unchecked on some older systems where the preamplifier doesn't reliably connect at startup otherwise. Having this checked significantly speeds up the Prairie View load time.

The **Legacy** preamp checkbox should be checked if the preamplifier(s) contain older model blades. Older blades will have white plastic at the base of the BNC connectors and the current blades will have black plastic there. We do not support mixing both types of blades on the same system.

For blade settings the only fields which should be modified are the **UI Text** and **GPIO Input** when input signal routing hardware is present in the GPIO box to allow for up to all 8 image channel inputs to be used on the back of the GPIO box. The **UI text** is the text that will be displayed on the channel buttons in the image windows for the 4 active image channels, and the **GPIO Input** is the index of the input the channel source is connected to on the back of the GPIO box.

If any of the preamp blade settings should get changed accidentally the correct values are listed in the **Preamp Configuration Notes** at the bottom of the section. This is only an issue for legacy preamp blades, for our currently blades those settings are hardcoded and cannot be modified.

## Stage (XY) Device Tab

The Stage (XY) Device tab is where an XY stage is configured. A secondary/auxiliary stage is currently partially supported with the MAMC, knob control only, no software control, but configured on this tab.

Check **XY Stage** if a supported stage is present and configure the device as instructed in the **XY Configuration Notes** text box at the bottom of the tab.

## Focus (Z) Devices Tab

The Focus (Z) Devices tab is where one or more Z focus devices are configured.

Use the **Add Device** button to add one or more supported focus devices and fill in the fields as instructed by the **Z Configuration Notes** text box in the upper right-hand corner of the tab.

By default the name of each device will be the selected device type, but that can be changed by clicking on the name of the selected focus device in the list of configured Z Devices.

The focus devices can be reordered using the up/down buttons to the right of the list, or removed by selecting a focus device and clicking the **Remove Device** button.

Focus devices can be enabled/disable independently by toggling the checkbox next to the name of each focus device, or selecting the focus device and toggling the **Focus Assembly** checkbox.

## Misc Tab

The Misc tab is where many different aspects of the system are configured which don't fit well with an existing tab and don't warrant creation of a new tab.

### *Device Control Box*

This section specifies the serial port and communication settings for communicating with the device control box. These settings are also duplicated when configuring a stage or focus device controlled by a device control box (which can potentially be different boxes). To emulate a device control box in order to enable software features which require a stage and/or focus device enter 999 for the serial port.

### *Multi-Axis Motor Controller (MAMC)*

This section specifies how fast the motor control knobs move the stage and/or focus devices.

### *Orbital Nosepiece*

This section specifies if a motorized nosepiece is installed, how many axes of motion it supports and whether it is motorized or the axes are moved manually.

### *GPIO Box*

This section specifies how the GPIO box is wired/configured internally. For the first section specifying where the imaging and uncaging source signals are connected it would be unusual to deviate from the defaults.

The **Disable external frame trigger for voltage output/recording/mark points frame counting** will use the internally generated frame trigger signal when checked. The internally generated frame triggers are only supported for galvo and AOD modes, so this must be unchecked to work with camera or resonant galvo acquisitions.

The **PMT signal routing hardware present** option is checked when the additional relay card is added to use up to 8 imaging channel inputs on the back of the GPIO box instead of the default 4. See the Channels / PMTs / Preamps Tab page for more information.

The **Laser routing hardware present** option is checked when the additional relay card is added to switch laser modulation signals between two sources. In addition to checking that the addition relay card is present an option must be selected to determine when the switching should occur:

- **SFC and Ultima** will share control of a 1P laser launch between an Ultima and an SFC, automatically switching control when the acquisition mode changes. Usually this is used for a launch with two fiber outputs which would to the SFC and a 1P confocal module.
- **Imaging and Uncaging** will share control of one or more lasers for imaging and sequential uncaging with one set of galvos. This would only need needed if more than 2 lasers were being used for both imaging and uncaging.
- **Alternate Beam Route** will switch control of lasers when the selected alternate beam route changes. This is used for simultaneous uncaging.

### *Dodt Detector*

This section specifies whether or not there is a Dodt detector on the system. Assuming there are filters installed they can be named and all new Dodt detectors use a quad servo board. The old style checkbox is used to differentiate between units that were wired slightly differently as described next to the check box.

### *Microscope Base*

This section specifies what microscope base is on the system. This is used both to determine which default scan settings are allowed to be applied in the scan settings dialog as well as defaulting the tube lens focal length which is required for calibrating the ETL when present.

### *Utility Buttons*

This section specifies up to 8 utility buttons, or digital outputs which can be toggled between high and low using a button on the MIsc tab within Prairie View. For each utility button values need to be entered for the following

columns:

- **Caption** specifies the text next to the button. Normally this describes what the button controls
- **'Off' Text** specifies what the utility button will read when toggled off/low
- **'On' Text** specifies what the utility button will read when toggled on/high
- **Device Type** specifies which piece of hardware will be providing the digital output for the utility button
- **Digital Output Line** specifies the index of the digital output to be used by the utility button on the type of device selected
- **Trigger Pulse** when checked will cause the utility button to go high and then low again right away to generate a trigger pulse. When this is checked only the **'Off' Text** is used for the utility button

### *Patch Clamp Device*

This section specifies which patch clamp device is being used by the system so that it can be controlled using the Seal Test feature within Prairie VIew.

### *XZ/YZ*

This section specifies whether or not the system should support XZ/YZ scanning. There is an option trigger routing signal which goes high during an XZ/YZ scan to provide line triggers to a focus device instead of frame triggers.

### *Plane Scanning*

This section specifies whether or not the system should support plane scanning. Similar to XZ/YZ scanning, but support oblique planes. Currently the line triggers must be connected manually or use a utility button to switch since there is no automated signal like XZ/YZ scanning.

## SFC / Camera / Filters Tab

The SFC / Camera / Filters camera based acquisition modes are configured.

## 2P Lasers Tab

The 2P Lasers tab is where 2P lasers are configured so that Prairie View can control their wavelengths, internal shutters, and in some cases their dispersion values and powering them on or off.

Enable one or more 2P Lasers and configure then according to the **Default Configuration Notes** on the right side of the tab. In the case of the Mai Tai laser entering 999 for the serial port will emulate a laser so that Prairie View functionality requiring a laser can be explored without having an actual 2P laser.

The **Name** of the laser is what will be displayed in the laser selection dropdown on the 2P Laser tab in Prairie View if more than one 2P laser is present. Additionally this laser name will be written into the metadata to be able to differentiate which values were for this device, which is mostly helpful when there is more than one 2P laser configured.

The **Wavelength Presets** for each laser determine which buttons are visible on the 2P Lasers tab to quickly change the wavelength to the preset value.

## Custom Outputs / Dual Galvo Scanning Tab

The Custom Outputs / Dual Galvo Scanning tab is where additional analog outputs synchronized with imaging are configured.

### *Custom Outputs / BOT Drive Signals*

Up to 8 unused analog outputs can be reserved on the 6713 option card to generate user defined signals synchronized with image collection.

Optionally any of these outputs can be used to synchronize an analog output with the relative brightness of a BOT region. Currently technical limitations of NI's DAQmx implementation make these signals a bit noisy (they bounce around a lot to previous values) so they are not terribly useful at the moment.

### *Dual Galvo Scanning*

Checked to provide alternate imaging drive signals to the uncaging galvos to use then to image simultaneously with the imaging galvos. This type of scanning is done using Dual Galvo ROIs in Prairie View where two different regions are selected to be scanned simultaneously. One limitation of scanning simultaneously is that the florescence signal from the two regions needs to be able to be separated using filters otherwise there will be a lot of cross-talk between the two regions.

### *Additional Line/Frame Triggers*

Additional unused analog output lines on the 6713 option card can be used to generate line and/or frame triggers. These triggers will not have extra pulses at the end of an acquisition as compared to the free running counters. The additional frame triggers are also useful in cases where it is desirable to generate both start and end of frame triggers and also count frame triggers.

## 1P Confocal Tab

The 1P Confocal tab is where a 1P confocal module is configured when present.

The legacy options are for our original scanning confocal which predated the current 3 channel confocal.

For the 3 channel confocal check the 2 dichroic options, the three filter options, the pinhole option, and then make sure to check **Read Device Labels from Confocal Controller Module** to avoid clobbering them with other settings if they had already been set. If the labels had not been set previously uncheck that checkbox and enter the available options in the fields proviced.

## FLIM Tab

The FLIM tab is where fluorescence/phosphorescence lifetime imaging is configured if the hardware is present.

Check **Enable FLIM** when FLIM hardware is present.

The router remapping options are there because the connectors on the B&H router were too close together to thread two of the provided SMA cables on next to each other, so instead channel 3 gets remapped to channel 2 for 2 channel FLIM to avoid skipping over a channel. Additionally in cases where FLIM uses it's own PMTs rather than switching control, the remapping can be used to map the FLIM PMTs with the channels of the same color/filter.