

IonWizard 6x Experiment Setup

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1 Overview

The basic recording element in IonWizard is called a Recording Source. An experiment is basically a group of recording sources along with rate and duration information. Recording Sources provide the connection between one or more elements in the hardware tree and a specific acquisition method.

There are simple recording sources which read a device and store the result

- analog inputs
- digital inputs
- video inputs
- pmt counters
- etc...

In addition there are complex recording sources which provide addition functionality beyond basic data collection

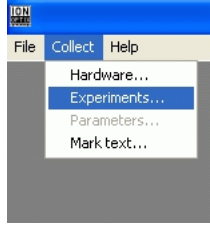
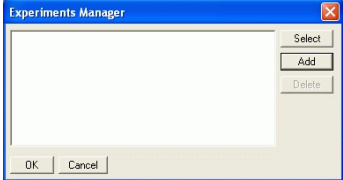
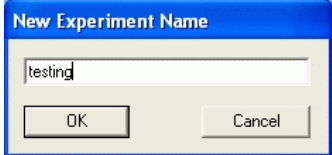
- single excitation (controls light source to select specific excitation wavelength)
- dual excitation (controls light source to sample alternating excitation wavelengths)
- edge detection (process video images to record cell left & right edge locations)
- sarcomere length (process video images to record sarcomere spacing)
- and more

2 First Time Setup

After installing the IonWizard 6x hardware, interface card(s), and driver(s) and configuring the IonWizard hardware tree the final step is to setup your experiments. At this point your experiment settings will be completely blank. To create your first experiments you will have to follow these steps.

2.1 Create New Experiment Name

Before you can access the recording source definitions you must create a new (blank) experiment.

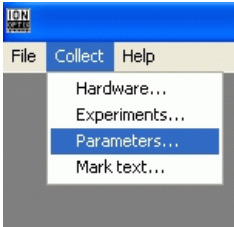
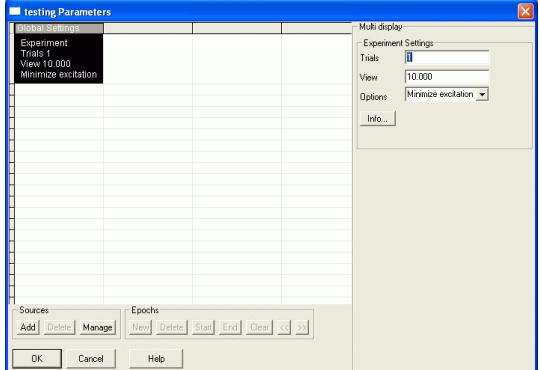
<p>1. Start IonWizard, select a user, then pick "New" from the file menu. In the "Collect" Menu pick "Experiments"</p>		
<p>2. This will bring up the Experiments Manger. Click the "Add" button</p>		
<p>3. Enter the name of your new experiment then click Ok. Click Ok again to close the Experiments Manager.</p>		

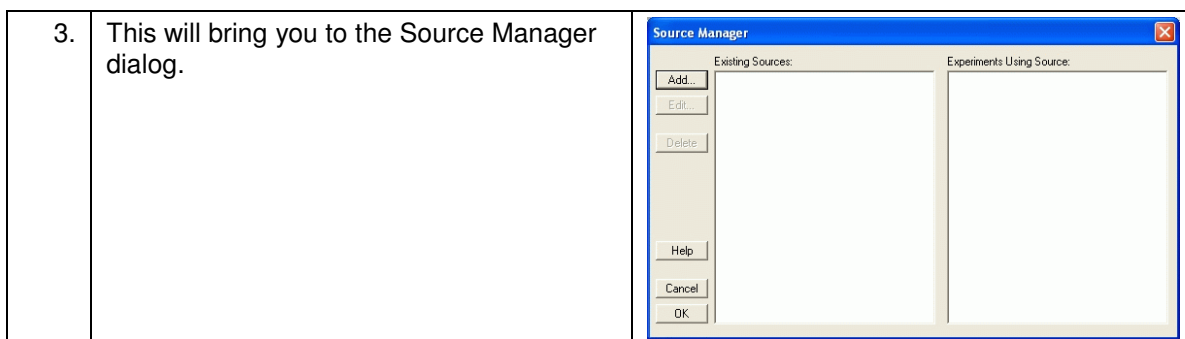
2.2 Create Initial Recording Sources

Now that you have defined an experiment you can access the experiment parameters which will allow you to access the recording source manager.

2.2.1 Access Recording Sources Manger

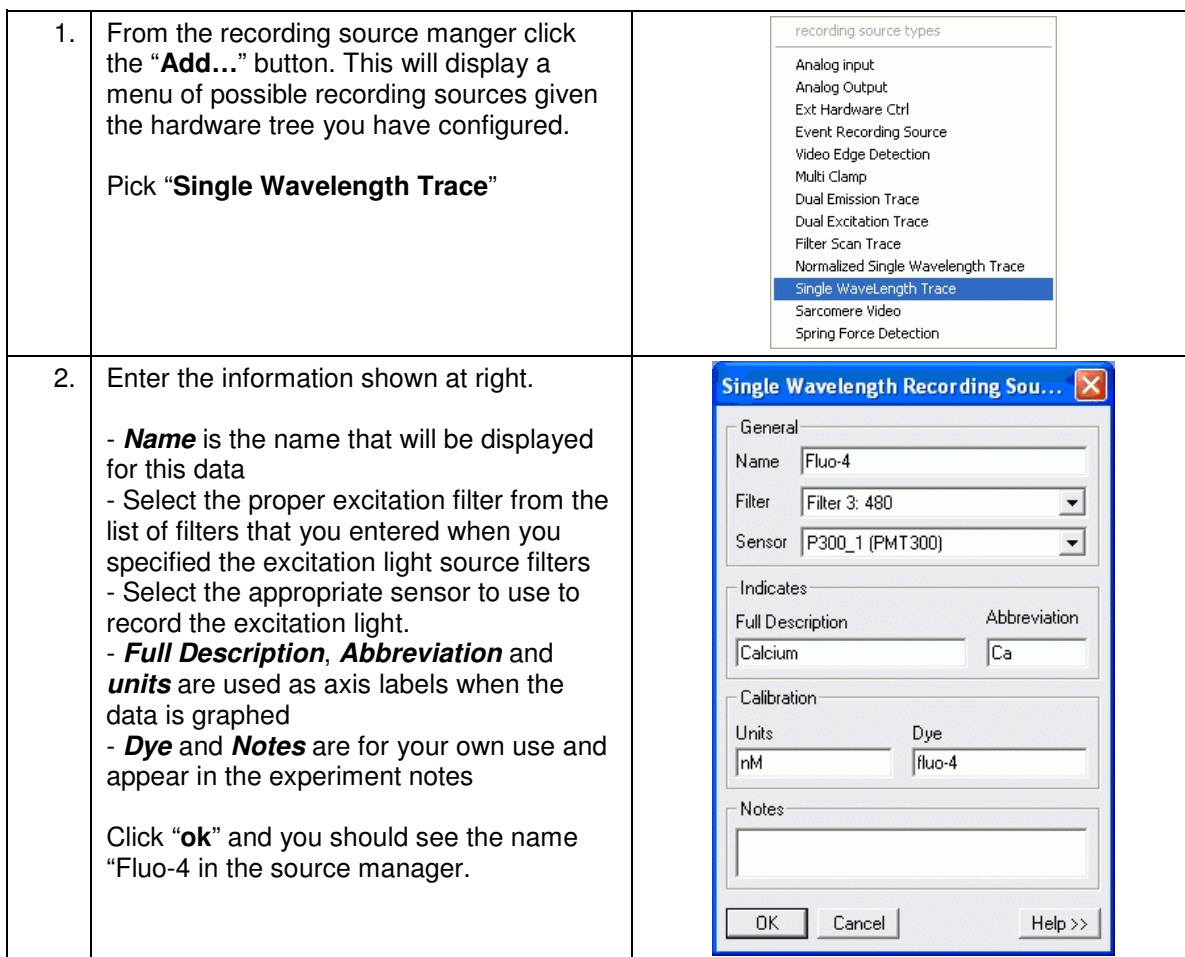
The recording source manage is accessed from within the experiment parameters.

<p>1. In the "Collect" Menu pick "Parameters"</p>		
<p>2. When you first edit a new experiment you will see this empty parameters dialog. At this point we need to create the recording sources that will be using for this example so click on the "Manage" button in the "Sources" section (above the ok button)</p>		




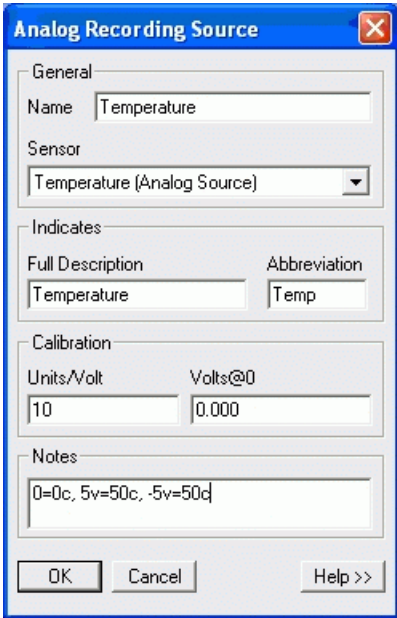
2.2.2 Add Single Excitation Recording Source

The following steps will create a recording source that will position the proper excitation filter and sample emission data using a photomultiplier tube. In this example we will say we are creating the recording source for use with cells loaded with Fluo-4. This example can be adapted to any other single excitation dye by making the appropriate edits to the example text.




2.2.3 Add Analog Input Recording Source

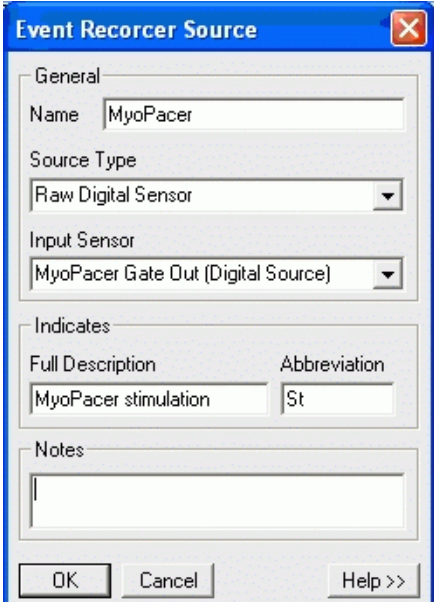
For this example we are going to configure an analog input that is connected to our cell temperature controller. You may use this example to record the analog output of any device or skip it if you do not have any

<p>1.</p>	<p>From the recording source manger click the “Add...” button then pick “Analog Input”</p>	
<p>2.</p>	<p>This dialog is similar to the Single Excitation Recording Source in the previous example. All recording sources have a similar setup. All have Name, Full Description, Abbreviation and Notes fields which function as described above.</p> <p>In this case of the Analog Recording Source the choices for Sensor are different. However as before the list of sensors is defined by how you configured your hardware tree.</p> <p>This recording source also introduces a new concept of unit scaling. By entering the correct values for Units/Volt and Volts@0 you can choose to display the data collected by this recording source as either raw data (Volts) or scaled units (Temperature)</p> <p>Click “Ok” when done</p>	

2.2.4 Add Digital Input Recording Source

In order to use digital inputs you need to create a recording source. For this example we are going to configure an analog input that is connected to our cell temperature controller. You may use this example to record the analog output of any device or skip it if you do not have any

<p>1.</p>	<p>From the recording source manger click the “Add...” button then pick “Event Recording Source”</p>	
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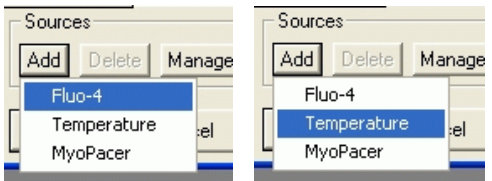
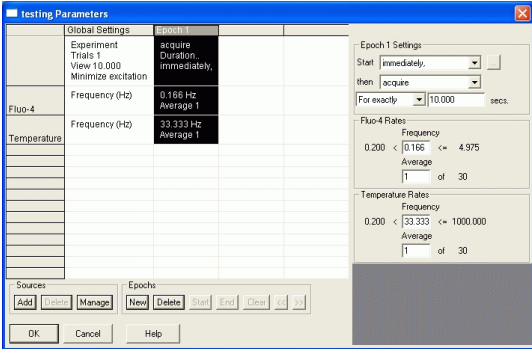
<p>2. The Name, Full Description, Abbreviation and Notes fields are the same as all other recording sources, see the Single Excitation Recording source description for notes.</p> <p>The Source Type field determines what event this recording source triggers. Select “Raw Digital Sensor”</p> <p>The Input Sensor field determines which digital signal is monitored. As with all sensors the list depends on what was defined in the hardware tree configuration.</p> <p>Click Ok when done</p>	
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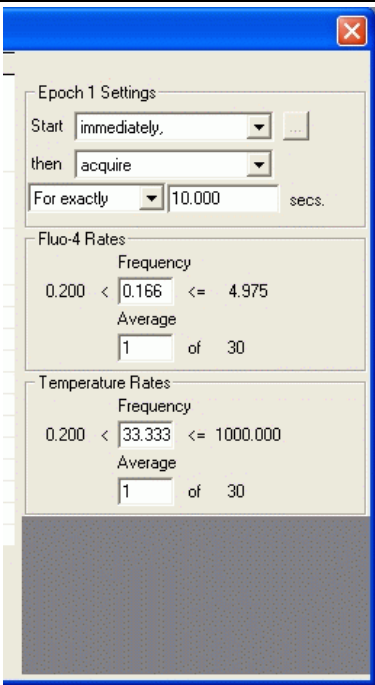

2.2.5 Recording Source Summary

As shown in the examples above all recording sources have a common setup. How you define your recording sources provides the basic configuration information about how each type of data is collected. In the recording sources provide the place for you to customize how data are named and labeled.

2.3 Define Initial Experiment

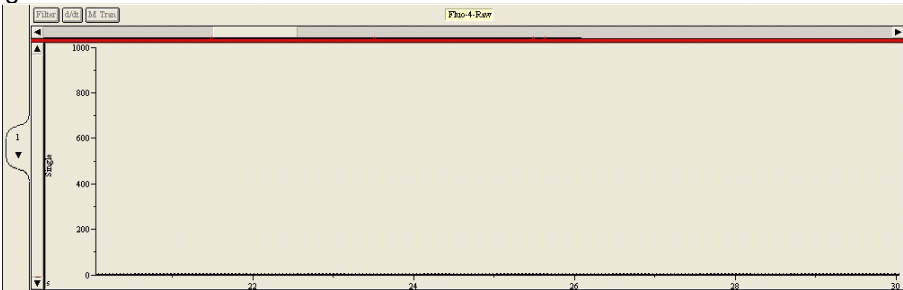
Now that you have created a basic set of recording sources you can create your first actual experiment.




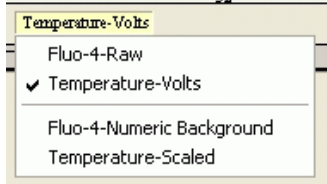
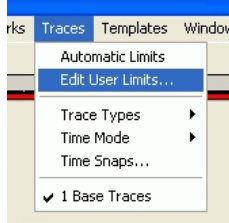
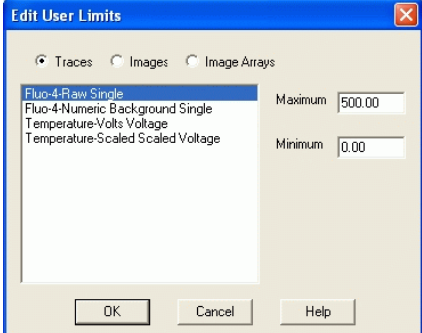
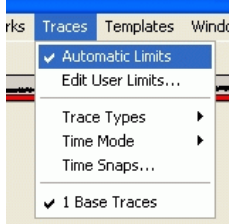
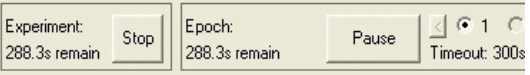
<p>1. The first thing to do is to add the recording sources that you created in step 2.2 that you want to use for this experiment.</p> <p>In this example add both “Fluo-4” and “Temperature”</p>	
<p>2. Next click the “New” button in the Epochs group at the bottom of the page. This will add a 2nd column to the parameter table.</p> <p>The experiment parameters should look similar to this figure at right</p>	

<p>3. When you highlight the “epoch 1” column the area to the right will show you the edit controls for the items in that column. Each group of controls (Epoch 1 Settings, Fluo-4 Rates and Temperature Rate) corresponds to a row in the main experiment table.</p> <p>The first group (epoch 1 settings) is for general epoch settings such as start condition and duration while the remaining groups depend on the recording sources that have been added. For now leave start set to “immediately”, then set to “acquire”, “for exactly” and change to 600 seconds (not shown)</p> <p>For the rates enter any value between the indicated ranges (the maximum rate is determined by the timer settings and the recording source definition). For now enter “1” for the average parameter</p>	
<p>4. Click “Ok” to save your changes to the experiment.</p>	
<p>5. At this point you should see a new element at the bottom of your screen. The experiment control bar provides information about the experiment and gives you quick access to major experiment functions. As the experiment has not been started all elements except “Start” are grayed out</p>	

3 Run Initial Experiment

Now that you have a valid experiment you may start it to collect data.

<p>1. Pick “Start” from the collect menu or click on the “Start” button in the experiment control bar.</p>	
<p>2. Once started you will see one or more trace viewers. Each trace viewer consists of a small full length view (above the red line) and a large selected view. The full view is actually an enhanced horizontal scroll bar that allows you to pick what point in time you want to display in the selected view. In addition to moving the full view “thumb” you can resize it by dragging the ends.</p>	 <p>The vertical scale of the viewer is controlled by the vertical scrollbar on the left edge of the viewer. It can be stretched by dragging the ends. Note that the maximum vertical range for each type of data is determined by settings in the traces menu (see steps 5-7 below)</p>

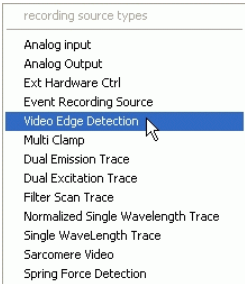
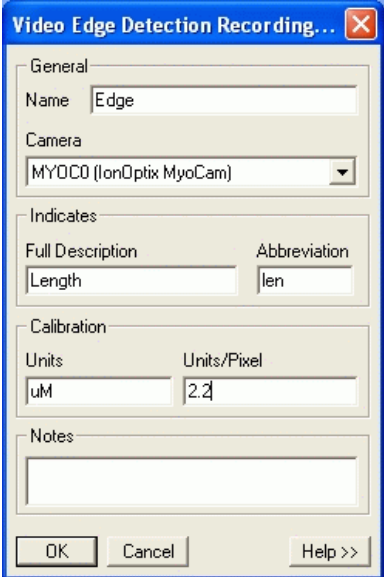
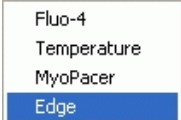
<p>3.</p>	<p>You may have up to 4 trace viewers open at a time. To close a trace viewer click on its tab (on the left) and it will minimize to a button.</p> <p>To open a closed trace viewer click on the button.</p> <p>Some functions work on the current trace which is indicated by a highlight.</p>	 <p>Open tab, click to close</p>  <p>closed tab, click to open</p>  <p>Current open tab</p>
<p>4.</p>	<p>You select the data to view in a trace viewer by clicking on the yellow boxes. The first yellow box selects the type of data that you want to view. This list depends on the recording sources that you have selected in the experiment. You may choose to look at raw data (above the line) or calculated data from any recording source.</p> <p>If your recording source created more than one channel of data (for example the numerator and denominator for dual excitation) there would be a 2nd yellow button to select the channel to display</p>	
<p>5.</p>	<p>To set the limits of the vertical scale for a trace view select “Edit User Limits...” in the “Traces” menu.</p>	
<p>6.</p>	<p>For each of the trace types you can enter the minimum and maximum display values. Click Ok when done.</p>	
<p>7.</p>	<p>For a quick display of existing data you can also select “Automatic limits”. When this is selected the data will be scanned to determine the minimum and maximum values on all trace viewers.</p>	
<p>8</p>	<p>You may stop the experiment by picking “Stop” from the collect menu or by clicking “Stop” in the experiment control bar.</p>	

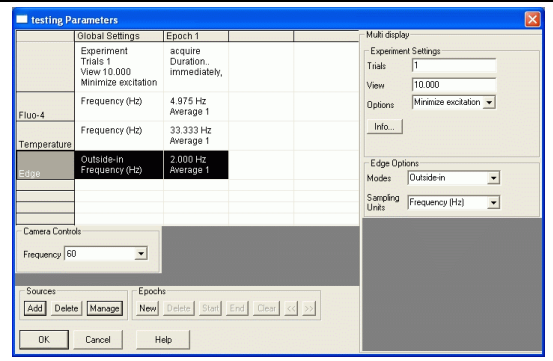
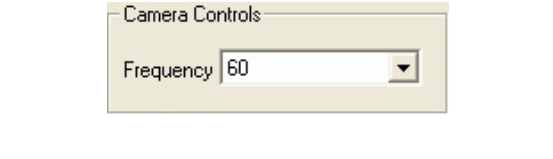
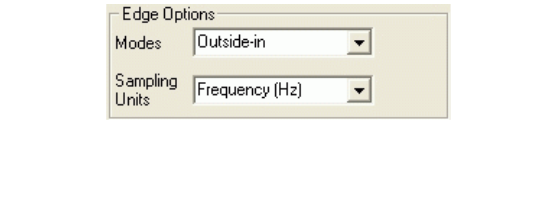
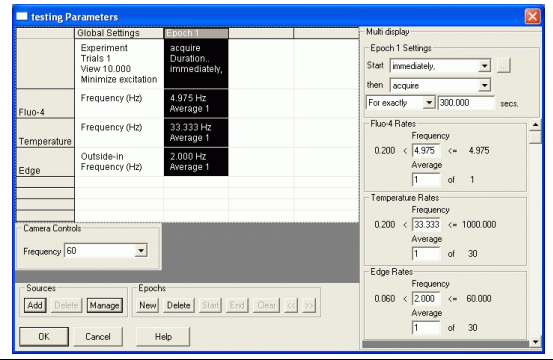
9.	To save you data, pick “Save” from the “File” menu.	
10.	When you are done pick “Close” from the “File” menu.	

4 Editing an Existing Experiment

Now that we have an existing experiment lets edit it to add cell length measurement. We will be adding Video Edge detection to our previous experiment.

4.1 Add new Recording Source

1.	Open IonWizard then pick “New” from the file menu. Next pick “Parameters” from the “Collect” menu. The previously created experiment should be displayed	
2.	Click on the “Manage” button in the “Sources” section (above the ok button). In the Source manager click the “Add” button then select “Video Edge Detection”	
3.	<p>The Video Edge Detection Recording source is similar to the Analog Input Recording section in section 2.2.3.</p> <p>The Video Detection Recording source uses the images from the camera selected in the Camera field. If you have multiple cameras you will have to select the correct camera to use.</p> <p>The Units/Pixel field defines the scaling factor that is used to convert from camera pixels to actual units. This conversion factor is determined by measuring the distance between marks on stage micrometer slide in pixels. The exact procedure is described in a separate document.</p> <p>Press “Ok” when done</p>	
4.	Press “Ok” to close the Sources Manager dialog and return to the main Parameters dialog	
5.	Now you can click on the “Add” button in the sources section then pick the “Edge” source that you just added.	

<p>6.</p>	<p>After adding there anew row will be added to the parameters table for the edge source.</p> <p>In addition to the normal recording source row and “Camera control” control is added below the main experiment table</p>	
<p>7.</p>	<p>For the IonOptix MyoCam the camera control must be set to match the switch on the camera controller (60/120/240). Other camera may have different software controls</p>	
<p>8.</p>	<p>When you highlight the edge row or the global settings column you will be able to set the global edge options (shown right) in the right hand column. The edge detection algorithm operates in two modes “Outside-in” and “Inside-out” which are selected here.</p>	
<p>9.</p>	<p>When an Epoch column is selected you will get the same type of controls for setting rate and average parameters as other recording sources.</p>	
<p>10.</p>	<p>Click “OK” to close the parameters dialog.</p>	

4.2 Edge-Detection On-Screen Controls

When you close the parameters dialog after adding a Video Edge Detection recording source you will see the Video Edge Detection control bar on the bottom of the screen.

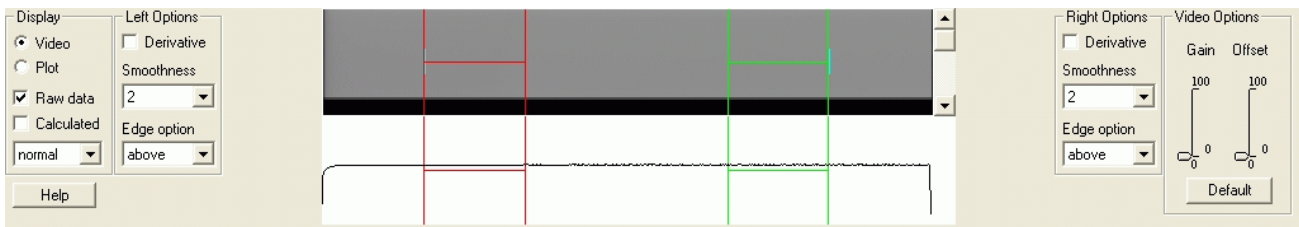
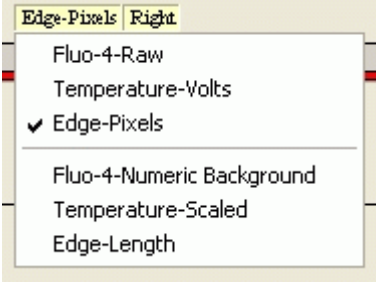
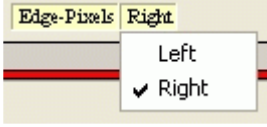
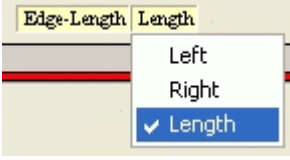


Figure 1 - Video Edge Detection control bar

This bar allows you to interactively tune the visually related parameters of the video edge detection control before and during an experiment. The top window (gray) will display a picture of the video being captured by the camera (which is blank in this figure). The bottom window (white) will show the graphs of the raw and calculated traces that are used to determine the edges. The red and green controls are moved by the mouse to select the active are for the left (red) and right (green edges). The vertical lines in the top window selects the video line that is scanned and the vertical line in the bottom window selects the threshold for the select edge option. More details of these controls are provided in separate document.

4.3 Running the Edited Experiment

Once you have completed editing the experiment you can run it as before.

<p>1.</p>	<p>As mentioned in Section 3, step #4, the choices in the yellow button menus depend on the recording source in the experiment. The choices for the first yellow button will now be:</p>	
<p>2.</p>	<p>Since the raw data for video detection is the location of the left and right edges there is a 2nd yellow box that lets you pick the left or right channel</p>	
<p>3.</p>	<p>Also note that the calculated length is only available when you pick "Edge-Length" in the first yellow box (type).</p>	

5 Appendix A – Advanced MyoCam-S Parameters

When using the MyoCam-S you can program the camera to go at faster rates by sampling fewer lines. This is done by selecting the advanced camera options in the experiment parameters. The maximum rate is determined by the interaction of a number of these parameters. Some example combinations of values are listed below; please refer to other documentation for more specific information.

<p>To collect 80 line (1/3 height) image at 250hz use these parameters:</p> <p>Y Size = 80 Field Integration time = 350 Frame rate period = 400</p>	<p>Camera Controls</p> <p><input checked="" type="checkbox"/> Show Advanced Options</p> <p>Image Format</p> <p>Bits/Pixel: 8 bpp</p> <p>Mode: Progressive</p> <p>Field Size: Manual</p> <p>X Start: 1 [0...134]</p> <p>X Size: 640 [1...774]</p> <p>Y Start: 0</p> <p>Y Size: 80 [1...245]</p> <p>Frame Information</p> <p>Size: 640 x 80</p> <p>Pixels: 51200</p> <p>Bytes: 51200</p> <p>Timing</p> <p>Pixel Clock: 24 MHz</p> <p>Serial BW: 192 Mbps (full rate)</p> <p>Field Integration: Maximize</p> <p>Delay: 0 10 uS [0...65535]</p> <p>Time: 350 10 uS [0...65535]</p> <p>Frame Rate: Manual</p> <p>Period: 400 10 uS [387...65535]</p> <p>Freq: 250 Hz</p> <p>Mbps: 104.45</p> <p>Output Sync. Pulse</p> <p>Delay: 0 10 uS [0...65535]</p> <p>Width: 1 10 uS [0...65535]</p>
<p>To collect a 34 line (1/7 height) image at 500Hz use these parameters:</p> <p>Y Size = 34 Field Integration time = 160 Frame rate period = 200</p>	<p>Camera Controls</p> <p><input checked="" type="checkbox"/> Show Advanced Options</p> <p>Image Format</p> <p>Bits/Pixel: 8 bpp</p> <p>Mode: Progressive</p> <p>Field Size: Manual</p> <p>X Start: 1 [0...134]</p> <p>X Size: 640 [1...774]</p> <p>Y Start: 0</p> <p>Y Size: 34 [1...245]</p> <p>Frame Information</p> <p>Size: 640 x 34</p> <p>Pixels: 21760</p> <p>Bytes: 21760</p> <p>Timing</p> <p>Pixel Clock: 24 MHz</p> <p>Serial BW: 192 Mbps (full rate)</p> <p>Field Integration: Maximize</p> <p>Delay: 0 10 uS [0...65535]</p> <p>Time: 160 10 uS [0...65535]</p> <p>Frame Rate: Manual</p> <p>Period: 200 10 uS [197...65535]</p> <p>Freq: 500 Hz</p> <p>Mbps: 98.30</p> <p>Output Sync. Pulse</p> <p>Delay: 0 10 uS [0...65535]</p> <p>Width: 1 10 uS [0...65535]</p>
<p>To collect a 8 line tall image at 1000Hz use these parameters:</p> <p>Y Size = 8 Field Integration time = 350 Frame rate period = 100</p>	<p>Camera Controls</p> <p><input checked="" type="checkbox"/> Show Advanced Options</p> <p>Image Format</p> <p>Bits/Pixel: 8 bpp</p> <p>Mode: Progressive</p> <p>Field Size: Manual</p> <p>X Start: 1 [0...134]</p> <p>X Size: 640 [1...774]</p> <p>Y Start: 0</p> <p>Y Size: 8 [1...245]</p> <p>Frame Information</p> <p>Size: 640 x 8</p> <p>Pixels: 5120</p> <p>Bytes: 5120</p> <p>Timing</p> <p>Pixel Clock: 24 MHz</p> <p>Serial BW: 192 Mbps (full rate)</p> <p>Field Integration: Maximize</p> <p>Delay: 0 10 uS [0...65535]</p> <p>Time: 60 10 uS [0...65535]</p> <p>Frame Rate: Manual</p> <p>Period: 100 10 uS [97...65535]</p> <p>Freq: 1 kHz</p> <p>Mbps: 49.15</p> <p>Output Sync. Pulse</p> <p>Delay: 0 10 uS [0...65535]</p> <p>Width: 1 10 uS [0...65535]</p>