

**PTi QuantMaster 40
Fluorescence Anisotropy
Standard Operating Procedure**

- Install the appropriate filters. (Please see Tony Adamo)
- Load a standard Polarizing Method

- Click on Setup and choose one of the following modes of operation:
 - Time-based
 - Emission

- Select the 'Polarizers' tab and check off 'Use Polarizers'
- follow the instructions below

Normal polarizer operation

G Factor

Click the G-Factor radio button.

Acquire background values if desired.

If a **Timebased scan** is used, an average value of the G-Factor will be calculated and saved as a G-Factor Global Value. If an **emission scan** is to be done, the G-Factor will vary with wavelength. In this case, a derived trace on the Traces tab should be created with **Name** = G-Factor, **Source 1** = HV trace, **Function** = Gfactor, **Source 2** = HH trace. After the scan is done, right-click on the G-Factor trace, **Create Lookup Table, Name** = Gfactor, **Type** = Gfactor, **OK**.

Polarization/Anisotropy

Click the Polarization/Anisotropy radio button.

Acquire background values if desired.

Choose the G-Factor to use by the radio buttons.

Either Use:

1) Use Last Acquired Global Value: This refers to the most recent G-Factor Global Value (a scalar value) acquired by a timebased scan.

2) Use Value: Enter a value into the text box.

3) Use Lookup Table: Use a Lookup Table that is saved as G-Factor values vs. emission wavelength.

Choose: Opens a list of saved G-Factor Lookup Tables. Choose one from the list.

Configure: Opens a Lookup Table editor where you can modify individual X and Y values of the Lookup Table.

On the Traces tab, create a derived trace with **Name** = Polarization or Anisotropy, **Source 1** = VV trace, **Function** = Polarization or Anisotropy, **Source 2** = VH trace.