## Preparations:

- 1. Get ready with an optical spanner wrench (Thorlab SPW602).
- 2. Get ready with a soldering station.
- 3. Gather all the parts on Fluorescence Module Parts List.xls in the Parts repository.
- 4. Order a PCB board of Motherboard.fzz (two layers) from a PCB manufacturer.

Assembling the optics: For a single colour, you need four assembled optical tubes, two containing only aspheric condenser lenses, and another two containing both aspheric lenses and emission/excitation filters. In addition a PMT and fibres/coupling. For two colours this means you need six assembled optical tubes, two containing only aspheric condenser lenses, and four containing corresponding excitation/emission filters in addition to two PMTs and fibres/coupling. See the blueprint for this two colour configuration.

- Use the optical spanner wrench and the retaining rings to fix the aspheric condenser lens
  (ACL2520U-A) and the filter (25mm diagonal) in the SM1 Lens tubes. The distance of the flat side of the
  aspheric condenser lens to the end of the tubes needs to be set as described (12 mm, back focal
  length) in the blueprint to ensure the lens is focused properly.
  - o The filters can be placed anywhere in the tube beyond the round side of the condenser lens.
  - Use the thick retaining rings (SM1RRC) on the curved side of the lens.
  - Use the thin retaining rings (SM1RR) on the flat side of the lens.
  - Use the thin retaining rings (SM1RR) for both sides of the filters.
- 2. Thread the SM1 Lens tubes containing the excitation filters and aspheric condenser lenses into the LEDs as shown in the blueprint.
  - Make sure that the tube containing the corresponding wavelength of the filter goes to the corresponding LED.
- Use the CMount adaptors (Hamamatsu A9865) to connect the PMT to the SM1 Lens tubes containing the emission filters and aspheric condenser lens according to the blueprint.
  - Make sure that the tube containing the corresponding wavelength of the filter goes to the corresponding PMT.
- 4. Install the dichroic filters (25x36mm) onto the dichroic filter mount cages (CM1-DCH).
  - o Don't thread too tight you might break the filters.
- 5. Install the lens tubes mounted with LED or PMT onto the filter mount cages.
  - Make sure each side of the dichroic filter are facing to the corresponding LED/PMT tubes (the transmission/reflection wavelength facing to the tube with corresponding wavelength of the filters)
- 6. Fix the SMA filber to SM1 adapters (SM1L20) to the tubes containing only the aspheric condenser lens with the thin retaining rings (SM1RR) on both sides of the adapters.
- 7. Connect the above tubes onto the dichroic filter mount cages.
- 8. Connect the fiber optical probe (RP26) to the SMA filber to SM1 adapters on both tubes.
  - Make sure to connect the legs of the fiber bundle to the corresponding tube as shown on the blueprint.
- 9. Connect the rest of the fibre bundle legs of the optical probe onto the LED reference senor (DET24A2)
- 10. Connect the LEDs to the LED drivers (LEDD1B)

## Assembling the PCB:

- 1. Solder the corresponding elements onto the PCB
  - make sure the directions of the elements are solder correctly onto the PCB as shown on the PCB blueprint.
- 2. Solder the power supply onto the PCB
  - Make sure the positive/ground wire goes to the correct PCB holes.
- 3. Solder the PCB mount headers onto the holes for Arduino Mega 2560 on PCB
- 4. Install the Arduino Mega 2560 onto the PCB board with the PCB mount headers.

## Interface optics with PCB:

 Connect the LED reference sensor, LED Driver cubes and PMTs onto the corresponding port on the PCB with proper connectors.

- 1. Connect the Arduino to a PC or a Raspberry PI via a USB cable
- 2. Upload the Arduino firmware TwoColors.ino to Arduino via the Arduino software.
- B. Run Twocolors.py to start an experiment.
  - a. The code was developed on Python 2.7.16, tested ok on Python 3.6
  - b. Remember to modify the I/O port in the Twocolor.py to the corresponding USB port which connected to the Arduino
- 4. The data collected and the operating log will be generated as a .txt file
- 5. Run the Analyze.py for preliminary analysis and data visualization
  - a. The code was developed on Python 2.7.16, tested ok on Python 3.6
  - b. Remember to modify the file path of the .txt file in the Analyze.py code

