

qpod 2e™ Temperature-Controlled Sample Compartment

DNA melting and annealing, protein thermodynamics, fluorophore characterization, enzyme kinetics and on-line thermocycling of biological particles... The qpod 2e provides rapid and precise temperature control for fiber optic spectroscopy from -30 °C to +105 °C.

Fully integrated electronics • Bluetooth capable



The **qpod 2e™** is a complete sample compartment for fiber optic spectroscopy, including a Peltier-controlled cuvette holder with magnetic stirring, and fused silica lens systems with SMA fiber optic connectors. Combine the qpod 2e with a fiber optic-based light source and a detector, and get a powerful spectroscopy system for a fraction of the cost of a traditional bench top spectrometer.

All of the electronics are built into the qpod 2e. Control temperature from your own computer via USB or Bluetooth.

Figure 1 - qpod 2e set up for absorbance measurements

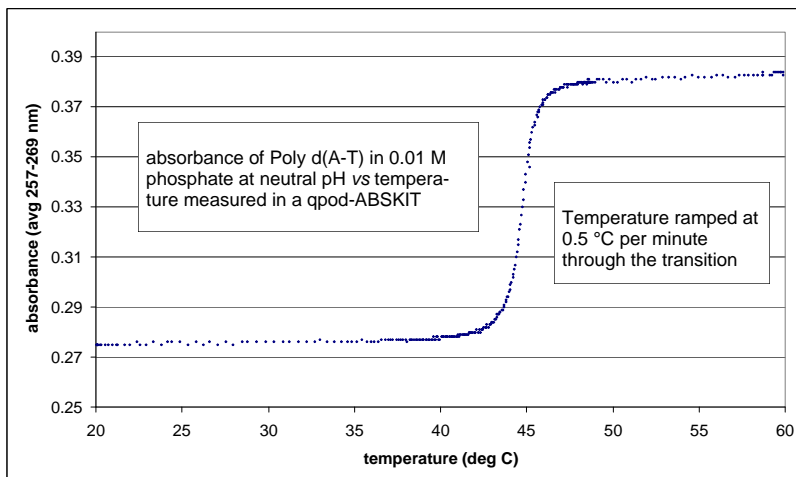
Three standard configurations:

- **qpod 2e-ABSKIT** - for absorbance, transmittance or turbidity measurements
- **qpod 2e-FLKIT** - for excitation and detection of fluorescence emission at right angles
- **qpod 2e-MPKIT** - a multipurpose kit for either absorbance or fluorescence measurements

Figure 2 - Thermal melting of a synthetic DNA performed with three linear temperature ramps using a qpod

If you do not control the temperature of your sample, will you get the same result the next time?

If you do not know how your sample responds to temperature, what have you missed?



Description

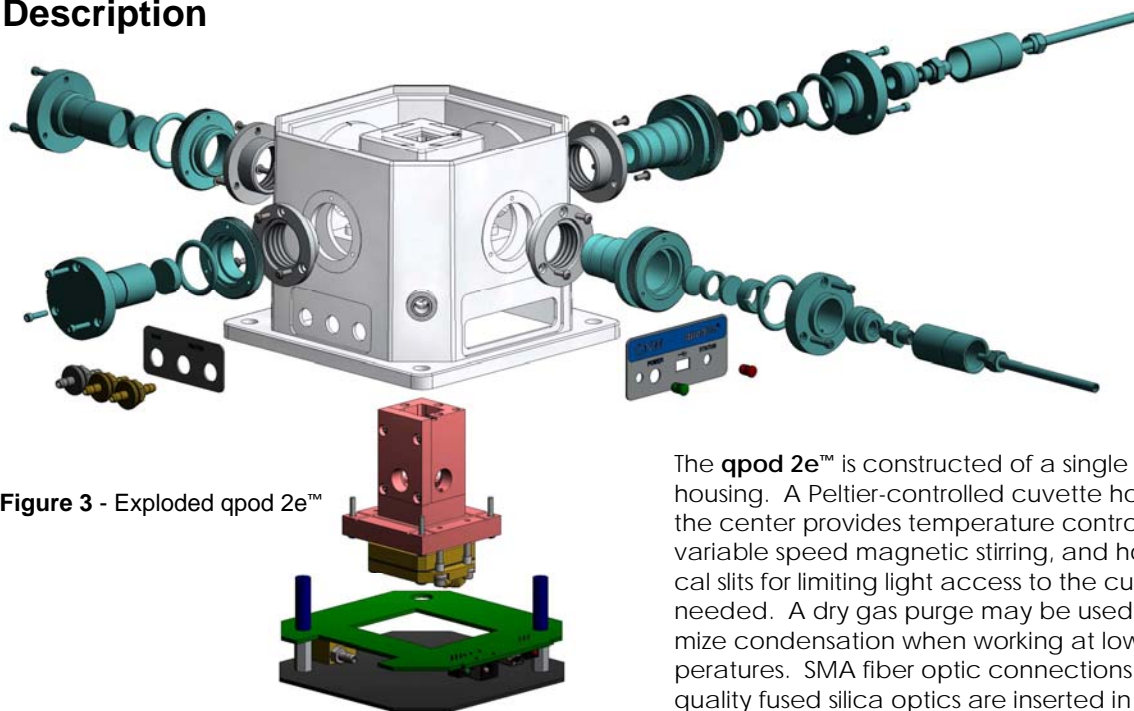


Figure 3 - Exploded qpod 2e™

The **qpod 2e™** is constructed of a single molded housing. A Peltier-controlled cuvette holder in the center provides temperature control and variable speed magnetic stirring, and holds optical slits for limiting light access to the cuvette as needed. A dry gas purge may be used to minimize condensation when working at low temperatures. SMA fiber optic connections and high quality fused silica optics are inserted in different combinations into optical ports in the walls surrounding the cuvette holder. Accessories such

as polarizers and filter holders may be inserted in the light paths. Spherical mirrors may be used to enhance excitation or emitted light. All optics provide focusing and position adjustments for maximizing signal throughput. Electronics are inserted below the cuvette holder.

For **absorbance** or transmittance measurements, insert two collimating lenses on opposite sides of the **qpod 2e™**. The first takes the excitation light from the source and collimates it into a parallel beam about 4 mm in diameter. The beam passes through the sample to be collected by the second collimating lens and focused on the end of the collection fiber, which passes the light to the spectrometer.

To measure **fluorescence** at 90°, insert imaging lenses on adjacent sides of the **qpod 2e™**. The first takes the excitation light and focuses it down to form an image (at about 1:1 magnification) of the end of the fiber into the center of the cuvette. The second imaging lens collects fluorescence by imaging the center of the illuminated volume onto the end of the collection fiber. Front surface spherical mirrors may be placed opposite the excitation and emission to enhance the signal.

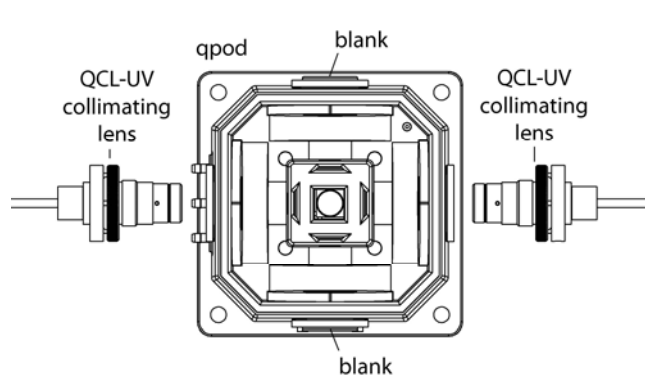


Figure 4 - setting the qpod 2e™ up for absorbance measurements (see the qpod 2e-ABSKIT)

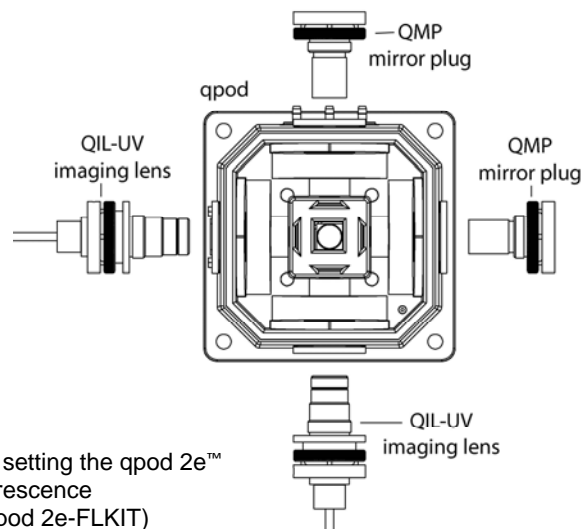


Figure 5 - setting the qpod 2e™ up for fluorescence (see the qpod 2e-FLKIT)

Software Control

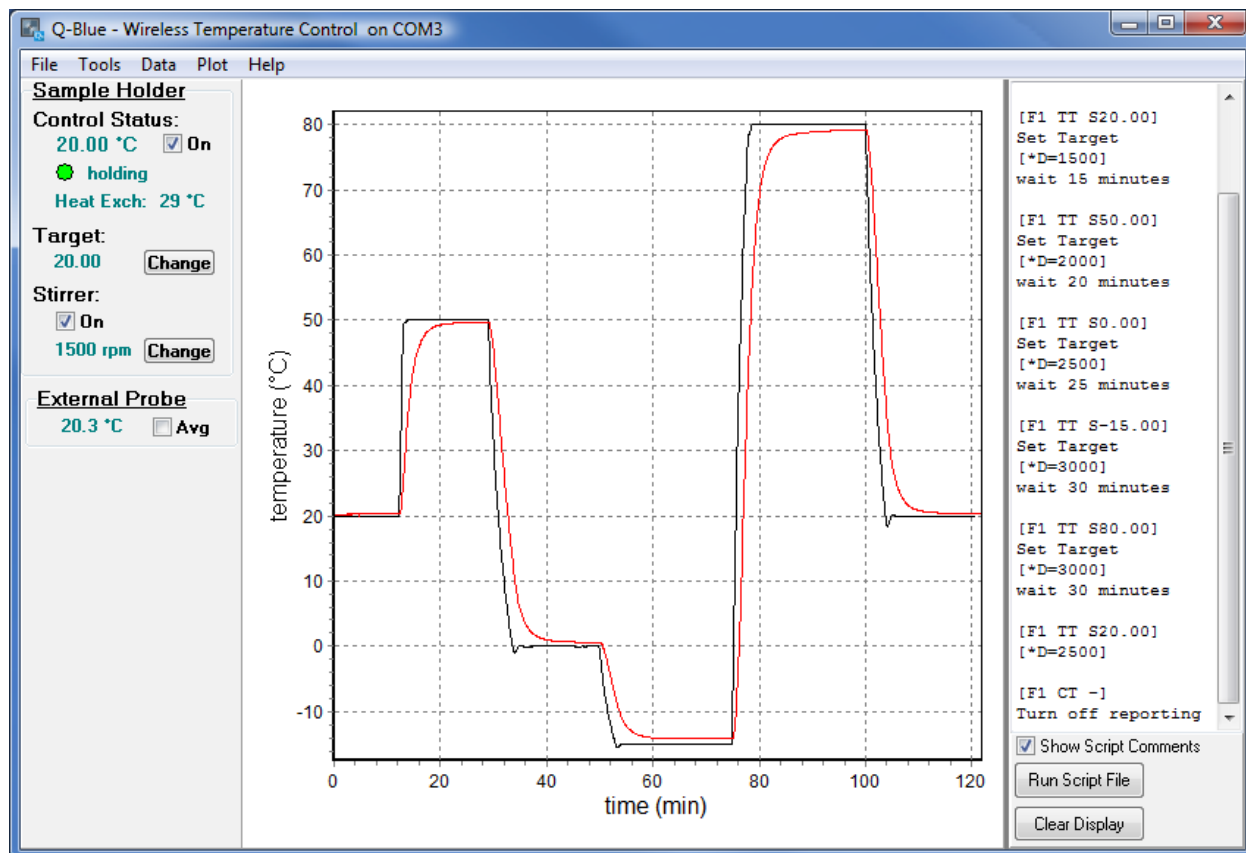


Figure 6 - Q-Blue, the program that controls the **qpod 2e™**

When opened, the program, Q-Blue, connects to the **qpod 2e™** via Bluetooth or USB. Under "Control Status" in the panel on the left of figure 6, the current cuvette holder temperature is displayed. Below this under Target, you are given the opportunity to set the temperature you would like to achieve. Under Stirrer, you may set the stirring speed and turn the magnetic stirrer on or off. When the correct target temperature is set, turn on temperature control. As the target temperature is approached, the flashing green light mimics the status button on qpod 2e, showing that you are "seeking." When the sample holder stabilizes at the target, the green light stops blinking and displays "holding." Plug a standard series 400 or 500 thermistor probe into the qpod, insert the probe in the sample and the sample temperature will display as well.

The main Q-Blue window displays the time dependence of your choice of the sample holder temperature, and/or the sample temperature.

Options under the pull-down menu let you:

- open a script window (shown in the figure) to allow you to automate the function of the qpod by following a set of text commands that may be easily written or edited
- choose data to be displayed
- capture temperature data to be copied into a spreadsheet
- capture publication quality plots of temperatures vs time
- execute a temperature ramp by entering a rate of change in °C and the final temperature

qpod Temperature Performance

After the temperature calibration procedure is completed, each qpod is put through a performance run and the data included in the manual. **Figure 17** shows an example of such a run. Using script control, the **qpod** is instructed to set and hold the temperature sequence 20.00, 50.00, 0.00, -15.00, 80.00, and 20.00 °C. Note the rapid changes in temperature, stable holding temperatures and very little overshoot. These are the temperatures of the sample holder. Temperatures inside a cuvette would be expected to lag behind as the cuvette comes to equilibrium.

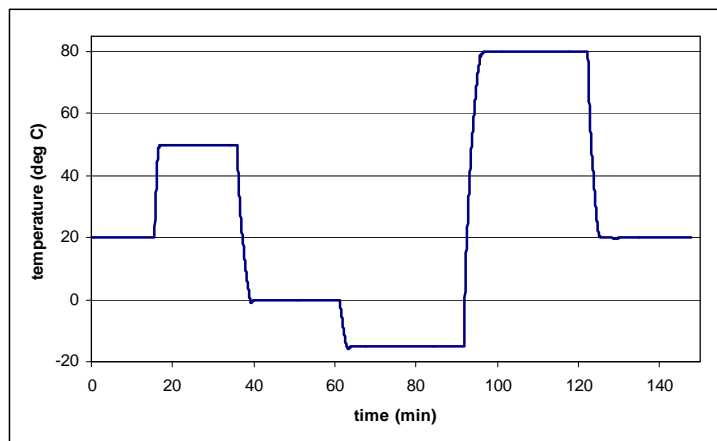


Figure 7- qpod temperature performance plot

To examine the low temperature performance of a **qpod**, ice water was used in the circulating bath and the temperature set to the lowest possible setting, -40.00 °C. Note in **Figure 8** the rapid drop in temperature, falling well below the -30 °C specified minimum temperature. To work at even lower temperatures, use a refrigerated circulating bath containing ethylene glycol- or methanol-water mixtures, cooled to within 25 or 30 °C of the lowest temperature you wish to achieve. A small amount of added foam or bubble wrap inside the qpod 2e around the cuvette holder may help for very low temperature work.

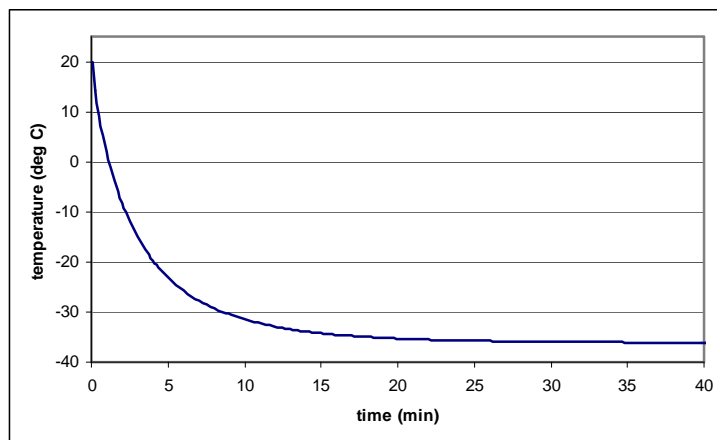


Figure 8 - qpod low temperature performance

DNA Melting Exercise

Poly d(A-T), a simple model double-stranded DNA, was purchased from the Midland Certified Reagent Company and prepared at a concentration of 16 μ M base pairs in 0.01 M phosphate at neutral pH. Ethidium bromide was from Sigma Chemical Company.

Due to base stacking, double-stranded DNA has a lower absorbance than single-stranded DNA, a property known as Hypochromism. Melting of the double-stranded structure thus results in an increase in absorbance. Hypochromism provides a simple absorbance test using the **qpod** for the melting or breakup of the DNA double-helical structure.

The dye, ethidium, intercalates between base pairs of double stranded DNA. When intercalated, the ethidium has a strong fluorescence, whereas free in solution it fluoresces much less. Thus, melting of DNA with intercalated ethidium will eliminate binding sites and result in a large decrease in fluorescence. Thus, ethidium fluorescence provides us with a simple fluorescence test of DNA melting.

DNA Melting Exercise (continued)

Absorbance

Excitation light for the **qpod** was provided with an Ocean Optics Mini-D2T UV Source and a 300 μm diameter fused silica fiber to a **qpod 2e-ABSKIT**. Transmitted light was passed through a second 300 μm fiber to an Ocean Optics USB 2000 spectrograph. Using script control, the temperature of the **qpod** was ramped from 35 to 50 $^{\circ}\text{C}$ at a rate of 1 $^{\circ}\text{C}/\text{minute}$. Absorbance data (blue points) were averaged over the wavelength range of 255 to 272 nm. Note in **Figure 9** the sharp melting transition with a midpoint just under 45 $^{\circ}\text{C}$. Ethidium was added to a poly d(A-T) solution to a concentration of 0.45 μM , or about one ethidium to 36 base pairs. Note also that the ethidium stabilized the DNA double helix, shifting the curve to a higher temperatures.

Fluorescence

Ethidium fluorescence was excited using an LS-1 Ocean Optics Visible Source and a 300 μmeter fiber connected to a **qpod 2e-FLKIT**. Visible excitation was isolated to a 450 to 550 nm band using 450 long pass filter and 550 short pass filters mounted in a **QFH** filter holder in the **qpod**. Emitted fluorescence was conducted via a second fiber to an Ocean Optics HR 4000 High Resolution Spectrometer, and averaged from 560 to 700 nm.

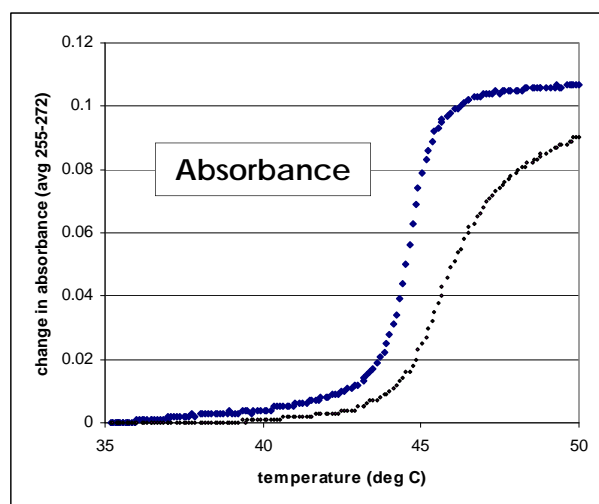


Figure 9 - Hypochromism changes in poly d(A-T) on melting without (blue) and with (black) ethidium

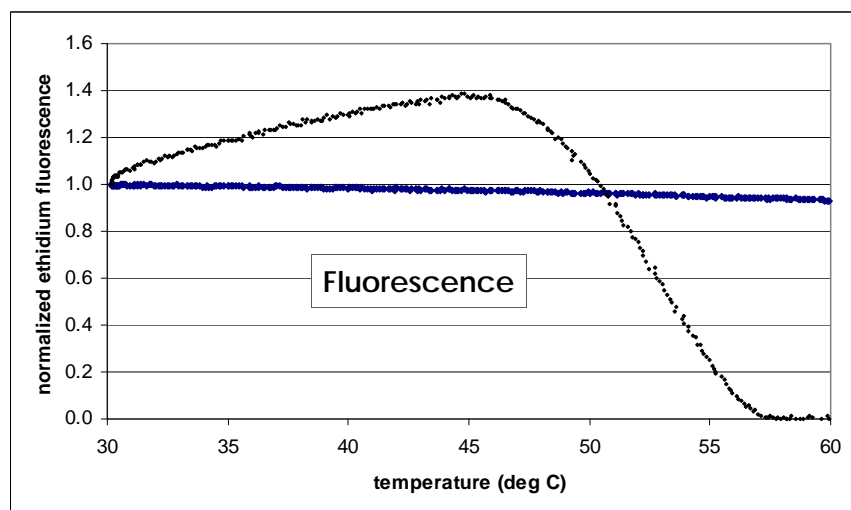





Figure 10 - Fluorescence of ethidium alone (blue) and ethidium in the presence of poly d(A-T)

Packages Available for Specific Applications


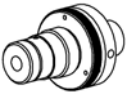





product	components	image
<p>qpod 2e-ABSKIT Absorbance</p>	<ul style="list-style-type: none"> • qpod 2e™ Sample Compartment • two QCL-UV Collimating Lenses 	
<p>qpod 2e-FLKIT Fluorescence</p>	<ul style="list-style-type: none"> • qpod 2e™ Sample Compartment • two QIL-UV imaging lenses • two QMP mirror plugs 	
<p>qpod 2e-MPKIT Absorbance and Fluorescence</p>	<ul style="list-style-type: none"> • qpod 2e™ Sample Compartment • two QCL-UV collimating lenses • two QIL-UV imaging lenses • two QMP mirror plugs 	

Each **qpod 2e™** sample compartment is provided with:

- **Q-Blue**, computer program to control temperature and magnetic stirring
- **AC power adapter**, to provide power to the qpod 2e
- **USB cable**, to be used if Bluetooth is unavailable
- **circulating pump**, bucket and fittings to provide circulating water to the Peltier unit
- **temperature calibration certificate**
- **tool kit** containing a hex screw driver for optical adjustments, a stir bar for use in the cuvette, a set assorted optical slits to limit optic access to the cuvette, and plastic blanks to cover unused optical ports on the qpod 2e

Components Available Individually

Use the following components to add to an existing package, or to assemble a system that has exactly the components you wish.

model number	description	image
qpod 2e™	qpod 2e sample compartment and temperature controller without optics	
QCL-UV	AR-coated fused-silica collimating lens with SMA 905 fiber optic connector and fiber optic steering plate	
QIL-UV	AR-coated fused-silica imaging lens doublet with SMA 905 fiber optic connector and fiber optic steering plate	
QMP	spherical mirror plug with steering plate to enhance excitation or emitted light	
QFH	filter holder for 12.5 mm diameter filter, mounts to a lens assembly	
QPOL-47-215	polarizer for mounting on the QIL-UV lens system, includes an Edmund Optics 47-215 Linear Glass Polarizer	
Q-Blue	computer program for external computer control—required for complex functions such as ramping and script control	

qpod 2e™ Feature Summary

- Rapid and precise temperature control from -30 °C to +105 °C ± 0.05 °C
- Designed for standard 1 x 1 cm square cuvettes or standard microcuvettes with a z-height of 8.5 mm (distance between the height of the optical center line and the bottom of the cuvette)
- Variable speed magnetic stirring to maintain uniform temperature in the cuvette
- Dry gas purge to reduce condensation when working at low temperatures
- Light tight cover with access cap providing a means of holding a thermistor probe in the cuvette
- Collimating optics available for straight through absorbance measurements
- Imaging optics available for efficient excitation and light collection for fluorescent samples
- Spherical mirror plugs available for enhancing excitation or emitted light
- All optical components with focusing and position adjustments to maximize light throughput
- Optical slits provided for limiting light access to the cuvette as needed
- Optional filter holders for 12.5 mm diameter filters (25 mm filters may be used without holders.)
- Optional polarizer
- Input jack on temperature controller to accept a thermistor probe for measuring sample temperature
- Window-based software control software provided with each unit
- Control via Bluetooth or USB
- **Two years' parts and labor warranty** as described on www.qnw.com



Inexpensive classroom solution

If you use spectroscopy in your laboratory course, equip your class with qpod 2e's, run them all via Bluetooth. Ask us about volume discounts.



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