COYOTE

MD-Box-Lab-B

Version：EN / 20160819

Coyote, highly specialized in research & development, offers completed molecular diagnostic solutions by providing devices from sample pre to gene amplification / detection.

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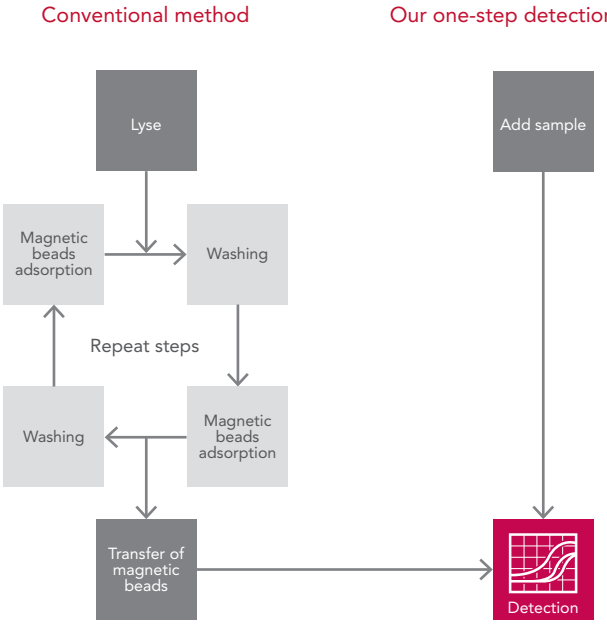
COYOTE

"One Step MD-Box-Lab", is aimed to provide portable molecular diagnostic solution. This one step MD-box-lab can be claimed as a regular bag on the flights. Batt3 is contained in this box to provide power for Mini8 Real-Time PCR System. This lab is suggested to be used with COYOTE one-step qPCR detection kit. COYOTE One-step qPCR detection kit innovate qPCR detection by eliminating nucleic acid extraction and purification steps, which will greatly simply operation and decrease operation time. Below is the comparison with conventional qPCR kit:

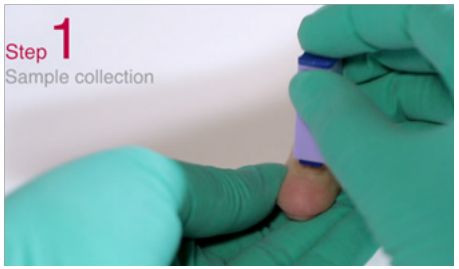


Currently COYOTE one-step qPCR detection system can be applied in several fields, e.g. field test, infection diseases, cancer screening etc. The integrated solution of "Lab in a box" is an ideal choice for the diagnosis of Ebola and Dengue fever, etc.

Features & Benefits

- Portable molecular diagnostic solution
- Extremely simple operation
- Compatible with COYOTE one step qPCR detection kit
- User friendly
- Field test

Workflow



Step 1:
Sample collection

Step 2:
Mix the sample with COYOTE one-step qPCR kit

Step 3:
Transfer the reaction mix into device

Step 4:
Set up program on Mini8 Software

Step 5:
Click RUN to start the assay, using Batt3 to supply power for Mini8

Packing List

Part	Quantity
Mini8 Real-Time PCR system	1 set
BATT-3	1 set
Eppendorf Pipettor (0.1-2.5 ul)	1
Eppendorf Pipettor (10-100 ul)	1
Mix4	1 set

Appendix

Setup

Unpack the Mini8 Plus System

1. Lift the Mini8 Plus instrument out of the package. Place it on a flat surface and remove the packaging materials (Keep the box and packaging in case of a return).

2. Check the packing list as follows to ensure that all components are present and intact. Your system comes with:

A. Mini8 Plus instrument

B. Power adapter

C. Power cable

D. USB cable

E. DVD

F. User's manual

A B C D E F

Place Mini8 Plus on the Bench

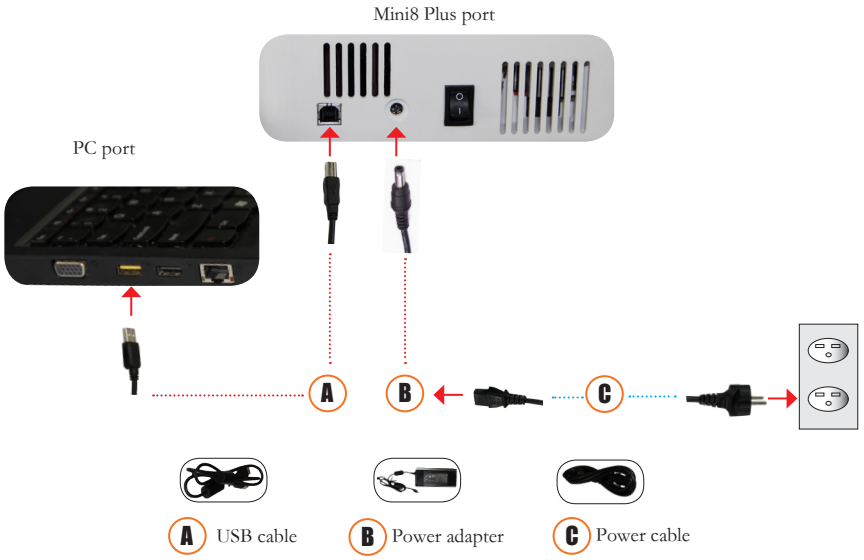
Benefit from its mini size, two distances of only 10cm (4 inches) for each sides of left and right are enough for ventilation, and also a distance of 5cm (2 inches) above is necessary for opening the lid.

Connect Mini8 Plus

1. Connect one connector of the USB cable to the port on the computer. Connect the other connector to the port on the side panel of the Mini8 Plus.

2. Connect the Mini8 Plus power cable to the DC power inlet on the side panel, and then to the wall outlet.

3. Plug the computer power cable to the wall outlet.



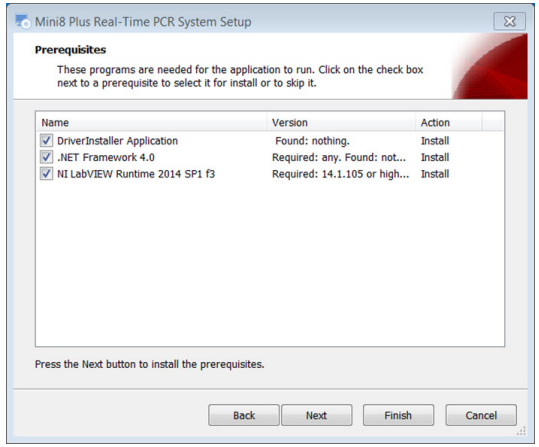
Install Mini8 Plus software

- Connect Mini8 Plus to computer and switch on the device;
- Load CD or open the software file, click setup program.

1) Before installation the Prerequisites Wizard must be started running to set up the environment as follow:

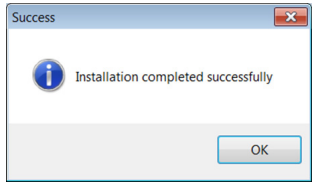
Click Next

Select ‘DriverInstaller Application’, ‘.NET Framework 4.0’, ‘NI LabVIEW Runtime 2014 SP1 f3’ and Click next. Click‘next’ per dialog box continuously to install ‘DriverInstaller Application’, ‘.NET Framework 4.0’, ‘NI LabVIEW Runtime 2014 SP1 f3’.

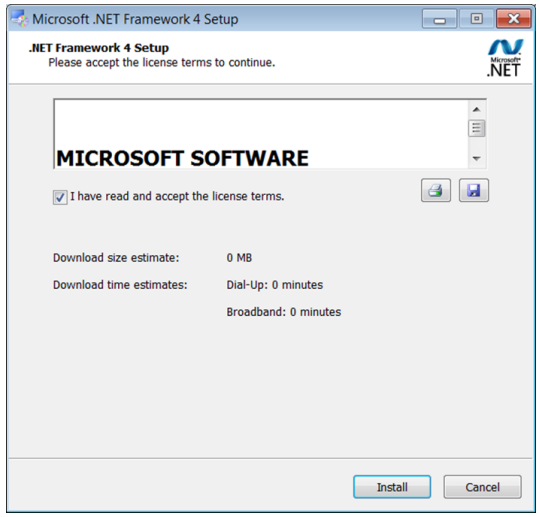


Click Next

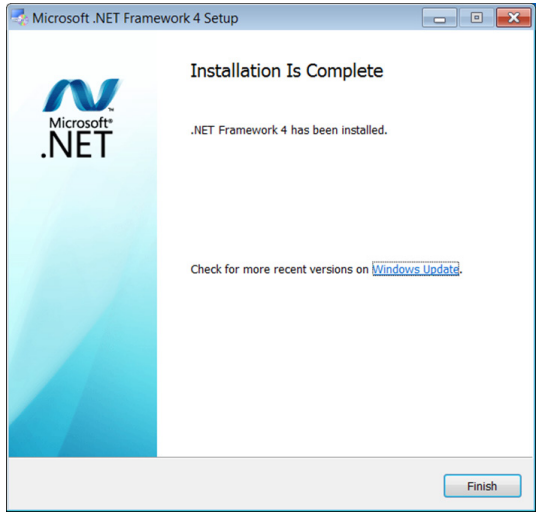
Click Install



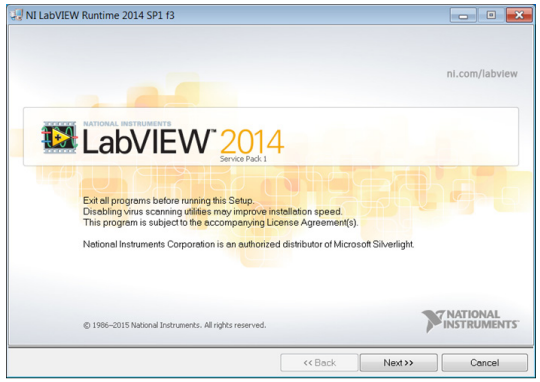
Click OK



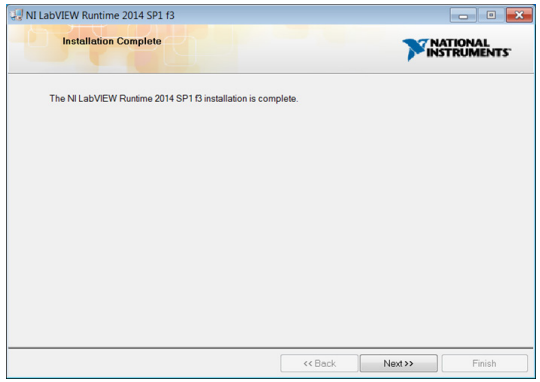
Click Install



Click Finish



Click Next



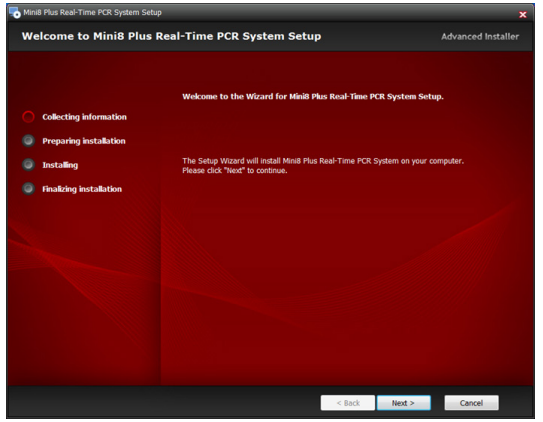
Click Next

• The System will automatically detect the operating environment, and check the installation options, If a project has been installed, you can manually cancel.

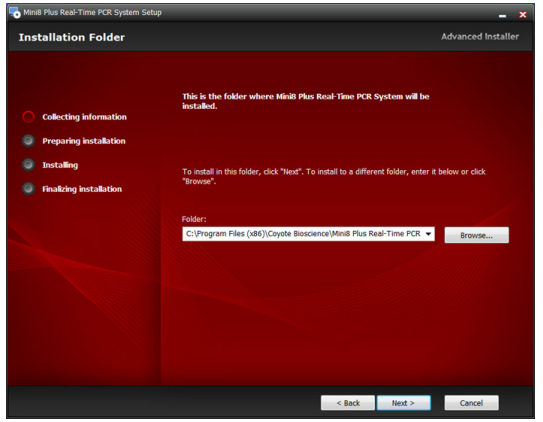
• Click next to start the installation.

Note: During the installation, the system may be requested to restart, you can choose to restart later.

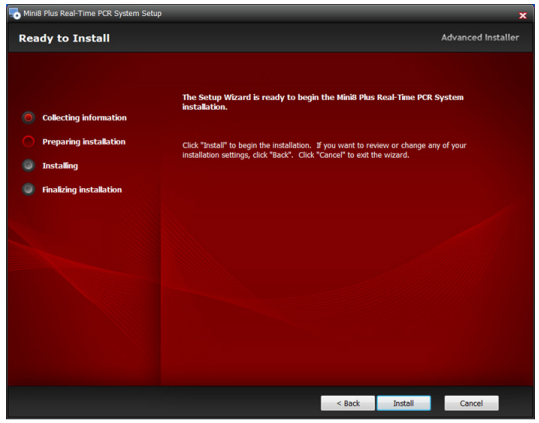
2) The software installation will be boot up once the environment detection completed.



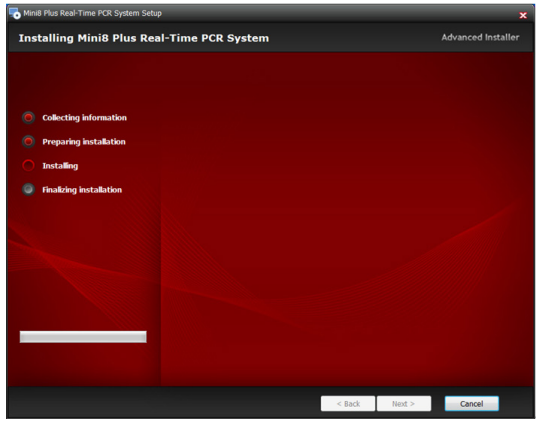
Click Next



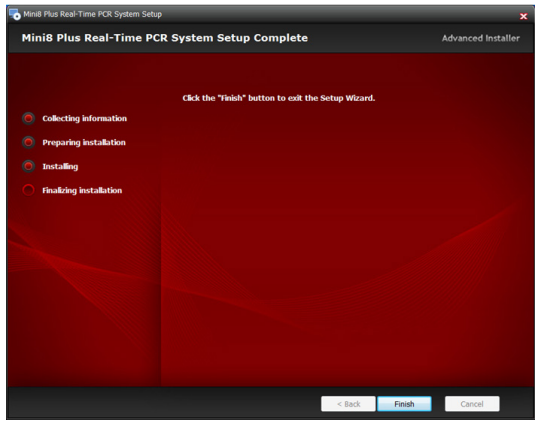
Select installation path and click Next to continue



Click Install to start the installation



The Mini8 Plus software is installing



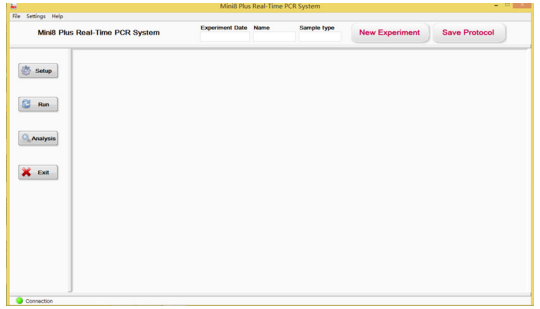
After the installation click Finish to close the setup program

Turn on the Mini8 Plus System

Turn on the Mini8 Plus instrument, then double-click the Mini8 Plus icon 

to start the software.

*When the Power indicator lights on the front panel stop flashing and remain solid, the instrument is ready.



Workflow

Mini8 Plus System Workflow

1. Prepare the sample strip, load it into the Mini8 Plus, and close the lid.

2. Double-click the Mini8 Plus icon 

on desktop to open the software.

3. Define and name the experiment, save the experiment.

Tip: to use a pre-defined thermal profile and plate layout for your experiment, click 

and select one of the template experiments saved in your computer.

4. Review the thermal profile and adapt it if needed.

5. Set up the plate layout by defining assays, samples, and standards and assigning them to wells.

6. Start the run. The Monitor Run tab opens.(Do not open the lid while a run is in progress.This will corrupt the data.)

7. When the run is completed, open the Mini8 Plus lid. Remove the strip from the block. Dispose of any hazardous materials into appropricate containers for biohazard, caustic materials, according to your local safety regulations.

Load reaction tubes

1. Thaw all necessary reagents (templates, primers, probes, and master mix).

2. Turn on the computer, then the Mini8 Plus, and wait until the Mini8 Plus "Power" light is flashing  (Standby mode)

3. Confirm that the block and optical path are clear of visible contaminants and there is no physical damage to the system.

4. Pipette samples and qPCR reagents into the strip according to your protocol. (Warning: Wear protective gloves and eyewear when operating with any material that might be considered caustic or hazardous.)

5. Open the Mini8 Plus lid and place the strip on the dock.

6. Close the Mini8 Plus lid.

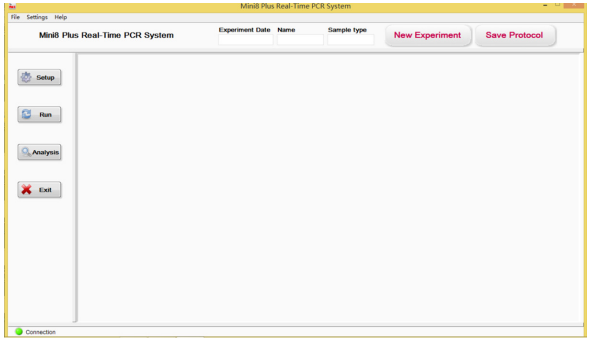
7. Proceed to define a new experiment.

Warning

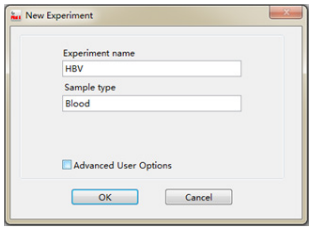
Do not touch hot lid. The hot lid temperature would be up to 105°C (221°F) when the device is working .

Define a New Experiment

1. Double-click the Mini8 Plus icon on the desktop to open the software.

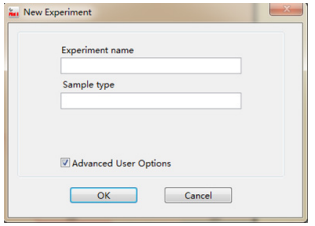


2. Click "New Experiment", the New Experiment tab opens, enter an experiment name and sample type.



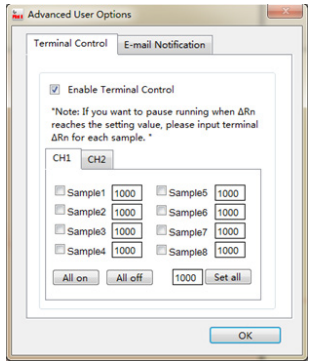
Experiment Date	Name	Sample type
2016/3/22 1:20	HBV	Blood

Advanced user options (optional functions)



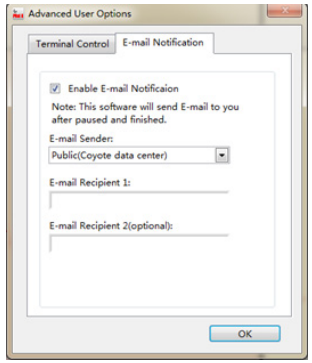
Select advanced user options and click OK

1) If you want to pause running when ΔRn reaches the setting value, please input terminal ΔRn for each sample. This function can help you to get the PCR amplification product in the status you want.

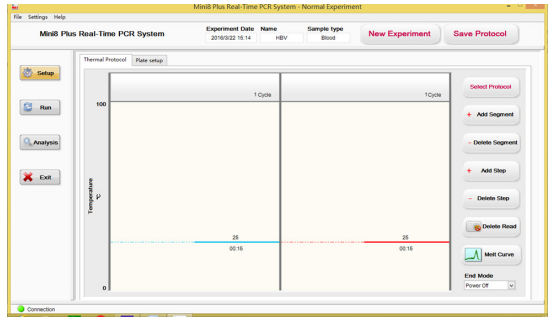


2) Fill in your Email, when the running is over, the result report (Excel file) will be sent to your email.

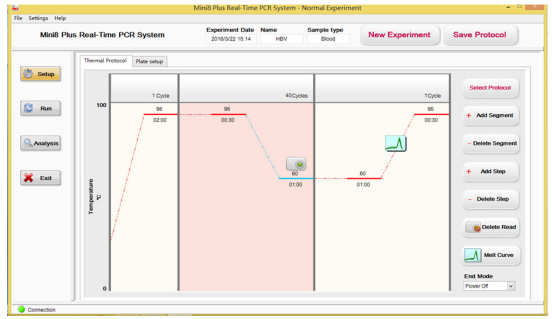
PS. The computer need to be connected to the network.



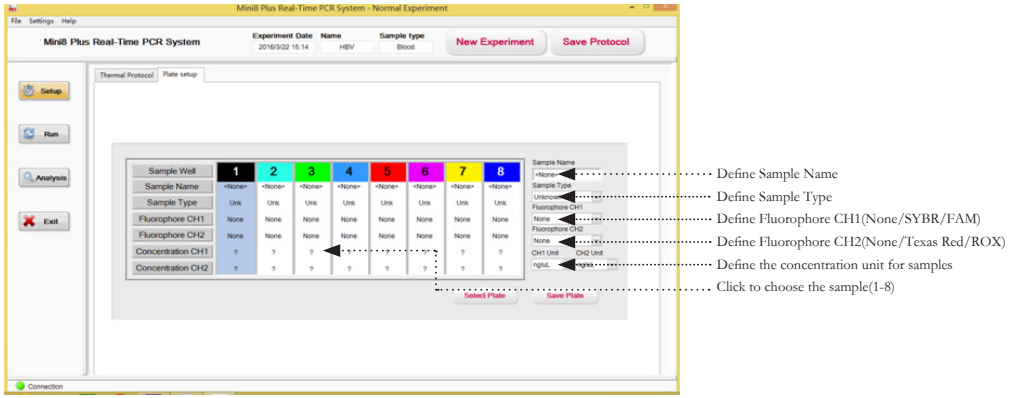
Set up the Thermal Profile



1. Click Setup , the Setup window opens, with the Thermal Protocol tab visible.
2. Click + Add Segment / - Delete Segment to add/delete segment.
3. Click + Add Step / - Delete Step to add/delete step.
4. The camera icon indicates that the fluorescence is being read.
5. Click Melt Curve to add the Melt Curve profile.
6. Click Save Protocol to save Protocol Protocol data (*.pdt)
7. Click Select Protocol to choose and use the saved Protocol (*.pdt)



Define the Plate Layout



The "plate setup" involves the following steps:

1. Click Plate setup , the Plate Setup window opens.

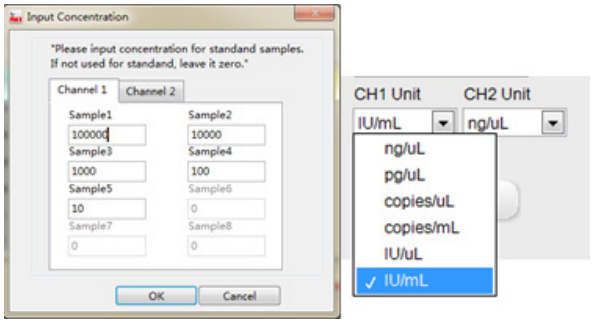
2. Click 1 2 3 4 5 6 7 8 to choose the sample.

3. Set up sample name.

4. Set up sample type: unknown, positive control, negative control, standard.

When the sample type is defined as "Standard" Sample Type: Standard

the concentration setting interface will be promoted for user to set the "Standard" concentration in the following interfaces:



5. Set up fluorophore CH1(None/SYBR/FAM)

6. Set up fluorophore CH2(None/Texas Red/ROX)*

7. Click Save Plate to save plate layout

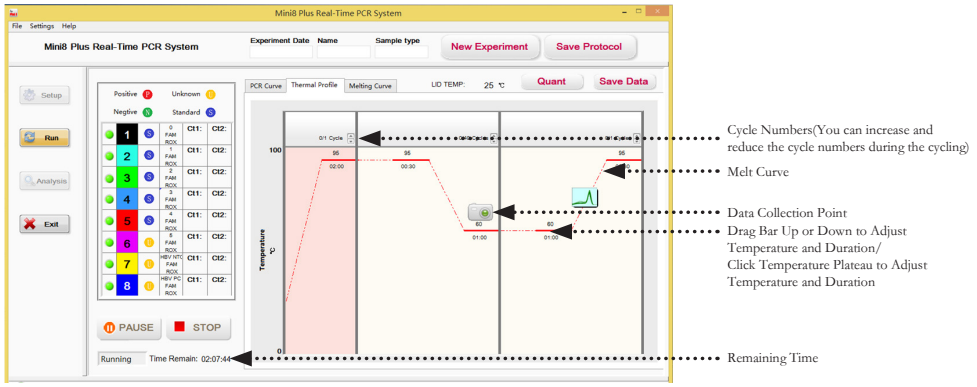
8. Click Select Plate to choose and use the saved plate

Monitor Run

Click Run to enter the running interface, and click START to start the running.

Click PAUSE to pause the running.

Click STOP to stop the running.



The Real time PCR curve will be showed during the amplification.

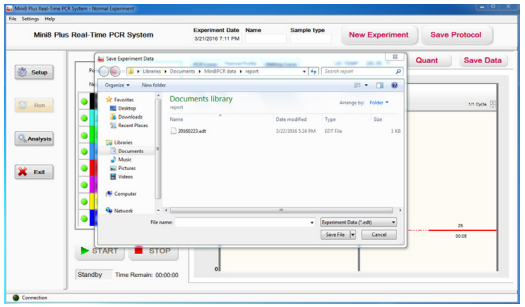
Warning

Do not open the lid while a run is in progress. It may allow extraneous light enter the system so the data will be corrupted.

Note

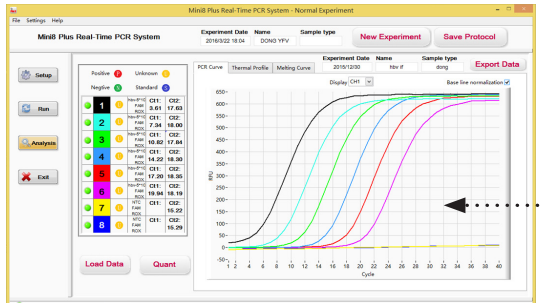
If you do not set up the fluorophore channel when set the thermal profile, there will be not any curve showing neither during nor after the process of amplification, for the fluorescence will not be read.

When the running is over, click **Save Data** to save data(*.edt).

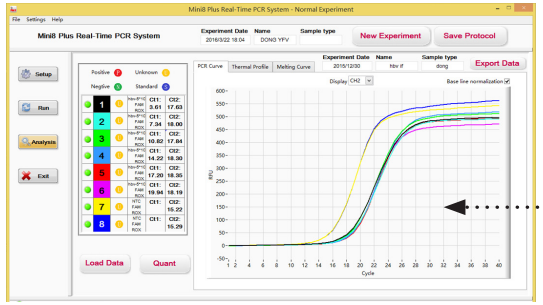


Data Analysis

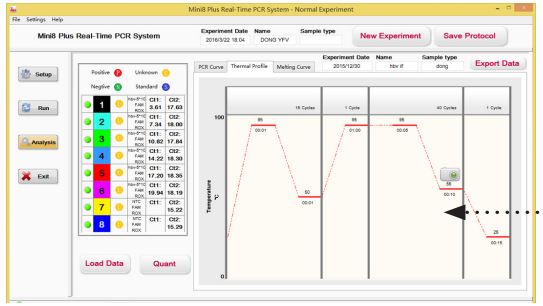
Click **Analysis** to show the saved data.

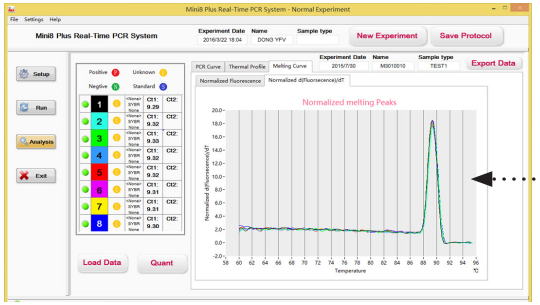


CH1 FAM

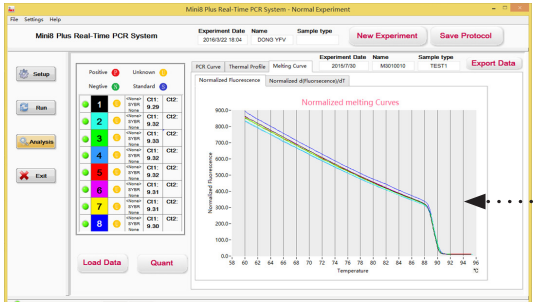


CH2 ROX



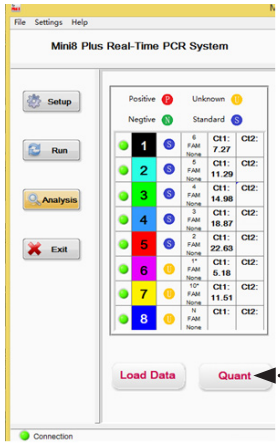


Melt Peak

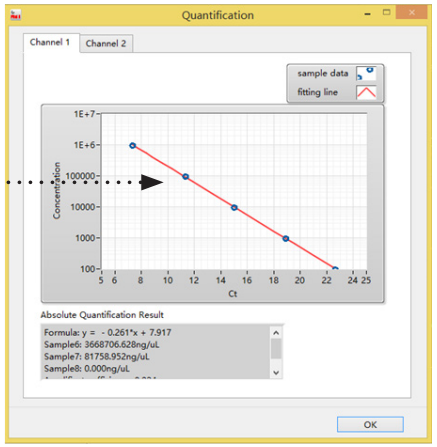


Melt Curve

Click **Quant** to show the quantification result.

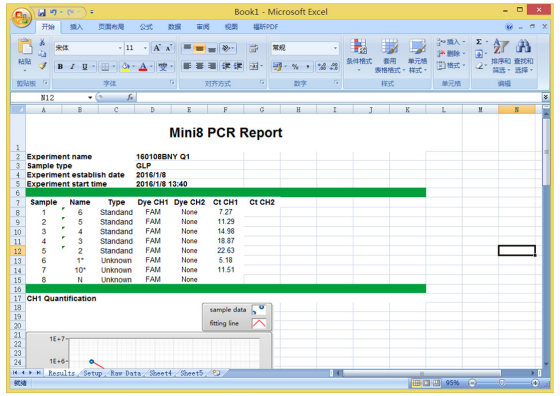


Quantification interface



Quantification

Click **Export Data** to export the result report (*.Excel).



And then click **Exit** to exit the software. Turn off the Mini8 Plus instrument.

System Information

Lights

The Mini8 Plus System has four indicator lights on the bottom right corner of the top panel: Power, Error, Status, and Scaning . The following table shows the meaning of each combination of off ,on,and flashing lights.

Lights	Description
○ ○ ○ ○	Power off
☼ ○ ○ ○	Standby mode
○ ○ ○ ○	Power on
○ ○ ● ○	PCR is running
○ ○ ● ○	PCR is running & scanning
○ ● ○ ○	Fatal Error (instrument might be overheated)

System Information

Specifications and Environmental Requirements

Optical	Light Source	High Power LED
	Detector	Photodiodes
Thermal	Heating/cooling module	Peltier
	Ramping Rate (Max.)	3°C/s
	Thermal Uniformity	±0.2°C
	Thermal Accuracy	±0.2°C
	Temperature Range	4-100°C
Operational	Sample Capacity	8 wells
	Reaction Volume	15-150μL
	Warm Up Time	1min
	Sensitivity of Detection	1 copy
	Melt Curve Resolution	Supported Resolution to 0.5°C
	Multiplexing	Detect up to 2 dyes simultaneously, (FAM/ROX)
Physical	Dimensions	205×190×98 mm (L×W×H)
	Weight	2.1 kg
	Power	12V, 10A
Computer Requirement	System	WIN 7; WIN 8.1; WIN 10
Environmental	Environmental Temperature Range	Operating: 15°C to 30°C Storage: 10°C to 60°C
	Environmental Humidity Range	Operating: 15-90% relative Humidity Storage: 5-95% relative Humidity

Electromagnetic Compatibility

• To confirm proper operation: The electromagnetic environment should be evaluated prior to operation of the system.

• Do not use this system in close proximity to sources of strong electromagnetic radiation (e.g. unshielded intentional RF sources), as these may interfere with proper operation.

• If you notice any interference, discontinue using the system until all issues are resolved. Resolution may include moving cords from other equipment away from the system, plugging the system into an outlet on a different circuit from other equipment, or moving the system away from other equipment. If you still have difficulties, contact COYOTE.

Cleaning and Maintenance

Clean the block and housing as needed, following these directions.

Caution: If hazardous or biohazardous materials are spilled onto or into the equipment, clean it immediately.

1. Turn the system off and allow the block to cool completely.

2. Using a lint-free cloth slightly dampened with clean water, gently wipe

the surfaces of the equipment. If a stronger cleaning agent is needed, use a lint-free cloth slightly dampened with 95% isopropyl alcohol.

Follow these practices for regular maintenance of your Mini8 Plus system.

1. Every time before using the system, visually check it to confirm there is no obvious physical damage such as dents, frayed cords, or damaged levers. If you see any damage, discontinue using and contact COYOTE Technical Support.

2. Once a year, run a known test sample to confirm accurate analysis.

Concepts

• The weight of one genome (g) = (size of genome in bp) x (618 g/mol/bp)/Avogadro's number

One human genome (g) = ($3x10^{9}$bp) x (618g/mol/bp) / ($6.02x10^{23}$) = $3.08x10^{-12}$g

One haploid cell (sperm/egg) = 3.08 pg of DNA

One diploid cell = 6.16 pg of DNA

• RNA concentration (μg/μl) = (A260×40×D)/1000, where D = dilution factor and A260= absorbance at 260 nm.

• DNA concentration (μg/μl) = (A260×50×D)/1000, where D = dilution factor and A260= absorbance at 260 nm.

BATT-3 is an universal battery pack, and can be used with our Mini8 Real-Time PCR System, Dry Baths, Slim PCR Cyclers and G50 motor-driven tissue grinders. It is an ideal accessary for the field applications. Also, it can be used as an alternative power source for a lab when the electricity is not stable. The power input goes from the power adapter through the battery pack to the equipment. When the power is shut off for some time, the battery pack can keep the equipment working for several hours.

Caution

Battery pack are not allowed to charge and discharge together.

Fig. 1, Batt-3 Battery
1 Output On/Off
2 Output port
3 Charging port
4 Power indicator switch
5 Power indicator
6 Charging indicator

Operation

Take the Battery Pack out, place it on a flat surface.
1) Charging: Plug a 12V DC power adaptor into the "Power Input" port
* Automatically stop charging while the battery pack is fully charged
* Charging indicator light off shows the fully charged status
2) Power Indicator: Press the power indicator button (Fig1, No.4) to check the remaining capacity of the Battery Pack. When the power indicator off, please charge the battery before using.
3) Supplying the power to Coyote's devices: Turn on the switch, connect to the Battery Pack (Fig 1, No. 2).

Specifications

Model: Batt-3
Battery: Lithium
Capacity: 130Wh
Power input: 12V, 5A DC
Power output: 12V DC, 10A max
Charging time: ~5hr
Working time: ~4hr (for dry bath)
Dimension: 152*44*200mm
Weight: ~2kg

Packing List

Main Body 1
12V 5A DC power adaptor 1
Power output Line 1(choose one below according to your need)
• Power output line (grey，for Theater Slim PCR, Mini8 Real-Time PCR System)
• Power output line (black, for G50, Dry Bath)