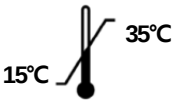
USER'S GUIDE

CE SGS C US 710219

Innovation • Value • Discovery

Exicycler™ 384

Real-Time Quantitative Thermal Block



REF A-2061

IVD

EC REP MT Promedt Consulting GmbH Altenhofstr. 80
D-66386 St. Ingbert. Germany. Tel +49 6894-58 10 20

BIONEER
Innovation • Value • Discovery

Exicycler™ 384

Real-Time Quantitative Thermal Block

User's Guide

Version No.: 3.0 (2018-10-02)

Please read all the information in booklet before using the unit

Bioneer Corporation
8-11, Munpyeongseo-ro, Daedeok-gu, Daejeon 34302, Republic of Korea
Tel: +82-42-930-8777
Fax: +82-42-930-8688
Email: sales@bioneer.co.kr
www.bioneer.com

Intended Use

This device is used for the qualitative or quantitative analysis of specific DNA/RNA in samples.

Warranty and Liability

All Bioneer products undergo extensive Quality Control testing and validation. Bioneer guarantees quality during the warranty period as specified, when following the appropriate protocol as supplied with the product. It is the responsibility of the purchaser to determine the suitability of the product for its particular use. Liability is conditional upon the customer providing full details of the problem to Bioneer within 30 days.

Trademark

Exicycler™ is a trademark of Bioneer Corporation.

Copyright

Notice

Bioneer corporation reserves the right to make corrections, modifications, improvements and other changes to its products, services, specifications or product descriptions at any time without notice. All information provided here is subject to change without notice.

CE

MANUFACTURER : Bioneer Corporation
8-11, Munpyeongseo-ro, Daedeok-gu, Daejeon, 34302
Republic of Korea

EUROPEAN REPRESENTATIVE EC REP : MT Promedt Consulting GmbH
Altenhofstr. 80
D-66386 St. Ingbert, Germany
Phone: +49 6894 581020

PRODUCT : *Exicycler*™ 384
Realtime Quantitative Thermal Block

CATALOG NO. REF A-2061

Contents

Before Starting

How to Use This Guide

This guide is written for researchers and laboratory staff responsible for installing and maintaining the *Exicycler*™ 384 Real-Time Quantitative Thermal Block.

General Requirements for Installation

This guide assumes that you:

- Are familiar with Microsoft® Windows® 7 & 10 operating system.
- Have basic techniques for handling DNA and RNA samples for PCR.
- Have basic skills for data storage, copying, and pasting data into hard drives.
- Have an experience in setting up a network. (Using any data generated by *Exicycler*™ 384 requires setting up a network.)

Word Conventions

- Bold signifies user action such as typing a text or clicking a button. For example:
  Type **Test** and click **OK** to move to the next step.
- *Italic* represents important words or sentences and is also used for emphasis. For example:
  After analysis, you must save data using Save As.
- A right arrow in bold (**>**) separates consecutive commands you select from a main menu or shortcut menu. For example: **File> Config> Scan**

How to Obtain More Information

For more information about *Exicycler*™ 384, please visit us online at www.bioneer.com.

How to Reach Customer Support

To obtain prompt customer support, please call us at +82-42-930-8777. You can also obtain technical support through www.bioneer.com (Email: exicycler-support@bioneer.com).

Product Use Limitations

- *Exicycler*™ system is designed and sold for in vitro diagnostic (IVD) applications in combination with the diagnostic kits manufactured by Bioneer and labeled for diagnostic purposes. *Exicycler*™ system can also be used for life science research and other laboratory purposes.
- Gradient function of *Exicycler*™ 384 is not available in US, EU, Japan and Germany.
- Use of *Exicycler*™ 384 is covered by U.S. patents Nos. 8,139,210, 8,427,643, and their foreign counterparts or pending patents. The purchase of these products includes a limited, non-transferable immunity from suit under the forgoing patents for using only this amount of product solely for the purchaser's own internal research. No other patent rights to use this product for any other purpose or for commercial purpose, including without limitation reporting the results of purchaser's activities for a fee or other commercial consideration, are conveyed expressly, by implication, or by estoppels. Further information on purchasing licenses may be obtained by contacting Bioneer at licensing@bioneer.com.

Safety Labels Description on *Exicycler*™ 384

English	Hot surface. Note that the surface is hot.
Francais	Surface chaude Faites attention à la surface chaude.
English	Do not work the bath at low temperature for long hours. This can cause condensation and lead to equipment malfunction.
Francais	Ne pas utiliser le bain à basses températures pendant plusieurs heures. Risque de condensation pouvant entrainer des fonctionnements défectueux de l'équipement.
English	Be aware of the door. Please pay attention to the door when it is moving.
Francais	Manipulation de la porte. Risque de blessures corporelles (pincement/écrasement) lors de. l'ouverture/fermeture de la porte.
English	CRUSH HAZARD Stay clear of this area when the door is in operation. Unawareness can cause injury.
Francais	Risque d'écrasement Ne pas rester proche de la porte durant son fonctionnement. Risque d'écrasement de doigts en cas de contact avec la porte.

Symbols on *Exicycler*™ 384

Safety Symbols on *Exicycler*™ 384

The following table describes the safety symbols that may be displayed on *Exicycler*™ 384. Each symbol may appear by itself or with text that explains the relevant hazard.

Hot surface.
Note that the surface is hot.

Do not work the bath at low temperature for long hours.
This can cause condensation and lead to equipment malfunction.

Be aware of the door.
Please pay attention to the door when it is moving.

CRUSH HAZARD
Stay clear of this area when the door is in operation.
Unawareness can cause injury.

Electrical Symbols on *Exicycler*™ 384

The following table describes the electrical symbols that may be displayed on *Exicycler*™ 384.

Indicates **On** position of the main power switch.

Indicates **Off** position of the main power switch.

Indicates a standby switch by which the instrument is switched on to the **Standby** condition.

Indicates a protective grounding terminal in the instruments that must be connected to earth ground.

Indicates a terminal that can receive or supply alternating current or voltage.

Indicates a terminal that can receive or supply direct current or voltage.

Environmental Symbols on *Exicycler*™ 384

The following symbol (WEEE) applies to *Exicycler*™ 384 placed on the European market.

Do not dispose of *Exicycler*™ 384 as unsorted municipal waste.
Follow local municipal waste ordinances for proper disposal provisions to reduce environmental impact of waste from electrical and electronic instrument.

European Union customers:
Call your local European office for Bioneer instruments pick-up and recycling.

Safety Instructions

Safety Warnings and Cautions

Safety instructions in this guide book are to cover possible dangerous conditions and situations that may occur. It is your responsibility to read this guide thoroughly when installing, maintaining, or operating *Exicycler*™ 384. The following safety alerts will be used in this section:

Hazards or dangerous actions that may result in severe injury.

Hazards or dangerous actions that may result in minor injury or damage.

Hazards or dangerous actions that may cause a burn.

Hazards or dangerous actions that may result in electric shock.

Location Consideration and Installation

General Instrument Safety

1) Check the power voltage rating before connecting to the instrument and an electrical outlet. *Exicycler*™ 384 is configured for 100–240VAC (±10%). Using AVR (Automatic Voltage Regulator) or UPS (Uninterrupted Power Supply) is recommended.
2) *Exicycler*™ 384 must be grounded for protection against electric shock. If not, it may cause serious injury and system damage.
3) Do not place any objects in front of the main door of *Exicycler*™ 384 that can interfere with door operation.
4) Leave 50 cm space between *Exicycler*™ 384 and the wall for proper ventilation.
5) Do not install *Exicycler*™ 384 in a dusty environment to prevent false operation or technical damage.
6) Keep *Exicycler*™ 384 away from heat sources.
7) *Exicycler*™ 384 must not be installed in an area where it is exposed to water or is humid. It may cause electric shock, a fire, or system damage.
8) *Exicycler*™ 384 must not be installed in an area where it is exposed to combustible or flammable vapor. In case of a gas leak, open windows and let fresh air in. Do not operate any electrical switch during a gas leak. It may cause an explosion or fire.
9) Do not disassemble or repair *Exicycler*™ 384 yourself. It may cause a fire, electric shock, and system damage. A limited warranty does not cover unauthorized alterations or damage due to abuse.

Precautions regarding the electrical environment

1) If the power cord is loose, do not use the instrument. Overheated power cord may result in shock or fire.
2) Do not operate multiple instruments out of a single wall outlet. The load may cause the instrument to overheat and lead to fire.
3) When plugging or unplugging the power cord from a wall outlet, make sure your hands are completely dry. Wet or moist hands may cause electric shock.
4) A convenient and safe power cord should be available. The power cord provided by our company should be used.
5) Socket outlets should be at least 1.5 m from a sink or wash basin.

6) Power cord should not be repaired with insulating tape. Water can still penetrate insulating tape and power cord.
7) If you use other power cord, it should be adequate for the electrical capacity for *Exicycler*™ 384 (250V, 16A, 0.75mm^2, VDE).
8) Plugs should match the socket outlets.
9) Unplug the power cord from *Exicycler*™ 384 when not in operation for a long period of time to prevent the possibility of fire by overheating.
10) *Exicycler*™ 384 is equipped with a 3-conductor AC power cord that, when connected to an appropriate AC power outlet, grounds (earths) the instrument. To preserve this protection feature, do not operate the instrument from an AC power outlet that has no ground (earth) connection.
11) Improper fuse or high-voltage supply can damage the instrument wiring system and cause a fire. Before turning on the instrument, verify that fuses are properly installed and that the instrument voltage matches the power supply in your laboratory.
12) For continued protection against risk of fire, replace fuse types (250V, F10AL, 2ea) according to rating specified for the instrument.

Installation Safety

1) Place and install *Exicycler*™ 384 away from direct sunlight.
2) It is recommended that you turn off the computer and unplug the power cable before connecting it to *Exicycler*™ 384. If the computer is turned on while connecting, the communication port connector either in the computer or *Exicycler*™ 384 may get damaged.
3) Make sure that the USB cable is firmly connected to *Exicycler*™ 384 and the computer. Unstable connection may cause damage to the communication port connector or data transfer errors.
4) Built-in camera of *Exicycler*™ 384 is a static-sensitive device. Pay particular attention to any cables connected to *Exicycler*™ 384 to avoid static damage.

Cautions

1) Ensure that the power cable is clean and connect it firmly to *Exicycler*™ 384.
2) Operate *Exicycler*™ 384 in a place where the temperature is always between 15℃ and 35℃. Poor performance is influenced by extreme temperature. High temperature can cause mis-operation and poor performance.
3) Operate the system in a place where humidity is always between 20 % and 80% with no condensation. Highly humid environment can cause corrosion of internal components and low humidity can lead to errors.
4) Do not place any objects behind or by the side of *Exicycler*™ 384 that can interfere with ventilation and cause errors.
5) The internal optical components of *Exicycler*™ 384 may get damaged when the instrument falls or is exposed to excessive physical shock.
6) Unplug the power cable from *Exicycler*™ 384 when not in operation for a long period of time to prevent the possibility of fire by overheating.

Operation and Maintenance

Warnings

1) The system can be hazardous when misused.
2) Keep the 384-well thermal block area clean to prevent damage and to generate accurate experimental data.
3) After any Real-Time PCR runs including 'Scan' or 'Melting', allow the light source lamp to cool down for at least 10 minutes. Continuous operation without a break will reduce the lamp life span and cause errors.
4) Do not place a piece of paper or a plastic cover under *Exicycler*™ 384. It could cause a fire.
5) Do not turn off *Exicycler*™ 384 right after a Real-Time PCR run is done. Wait until the cooling fan has stopped running completely. Cooling fan remains running for about 2 minutes to cool down the lamp after each Real-Time PCR run.
6) Do not cover *Exicycler*™ 384 with a piece of paper or a plastic cover. This may cause a fire or failure of the system.
7) Set 'Power Options Properties' in the Control Panel to 'Turn off monitor', 'Turn off hard disks', 'System standby', and 'System hibernates' to 'Never'. Otherwise, data transfer between *Exicycler*™ 384 and the computer will be interrupted.

IMPORTANT:
Exicycler™ 384 should be operated in a clean condition. Contaminants such as dust can cause problems and reduce the life span of *Exicycler*™ 384. Please prevent dust from entering into *Exicycler*™ 384. In order to extend life span of *Exicycler*™ 384, thoroughly remove dust periodically.

Introduction

Overview

Exicycler™ 384 Real-Time Quantitative Thermal Block combines a thermal block in a special 384-well format and is an innovative fluorescence detector. The detector monitors fluorescence emitted as an indicator for amplified nucleic acid product during each PCR cycle in real time.

Thermal block is built into the lower part of *Exicycler*™ 384, which carries out a thermal cycling. The detector is located in the upper part of *Exicycler*™ 384, which measures the fluorescence emitted from samples in the thermal block in real time and transfers data to the computer for analysis. Data transferred from the detector is analyzed with *Exicycler*™ 384 analysis program.

Exicycler™ 384 Real-Time Quantitative Thermal Block provides a maximum ramping rate of 4.5℃/sec, and features various functions such as gradient, time increment, temperature increment, and ramp rate control for myriad applications.

Exicycler™ 384 Real-Time Quantitative Thermal Block detector consists of a light source and a detector. The light source is a short arc lamp and emits a white-light as an energy source to excite fluorescent dyes. A white-light source is divided into particular wavelength groups by band path filters. The band path filters in *Exicycler*™ 384 covers within 475 to 690 nm and works as a set consisting of an excitation and an emission filter. Five filter sets are provided for various applications. Therefore, no additional band path filters is required. Also, Bioneer's imaging technique based on polarization of light enhances detection sensitivity for robust and reliable results. Patented (KR10-1089045, US8427643) polarizing optical apparatus mitigates the common problem of a reflection light, allowing precise quantification and target discrimination.

The detector is a highly sensitive 2D CCD camera and detects mulfiple fluorescence signals from a 384-well plate at once. This simultaneous detection has a great advantage over sequential detection. This innovative detector, invented by Bioneer, reduces well-to-well variation and has minimized dye-to-dye interference, therefore providing more accurate results.

Bioneer's state-of-the-art technologies applied to *Exicycler*™ 384 generate a uniform light surface over the thermal block. This allows detection of multiple fluorescent signals emitted from various dyes through the light surface and separate signals within a selective wavelength range of each dye.

Exicycler™ 384 Real-Time Quantitative Thermal Block features an auto loading function for automation reducing errors and self-diagnosis for diagnosing systematical conditions of *Exicycer*™ 384 for users convenience. *Exicycler*™ 384 software is composed of three main programs.

1) A set up program for fluorescence test, diagnosis and confirmation of the instrument's information.

2) An operation program for creating a protocol, assigning a probe and plate, saving & displaying data while operating *Exicycer*™ 384.

3) Analysis software that comes with *Exicycler*™ 384 has 4 different tools. The analysis program includes Absolute Quantification, Relative Quantification, SNP genotyping, and Existentence / Non-existentence. It is applicable to Gene expression, Quantification of cell and virus, and SNP genotyping.

System Components and Specifications

Overview

Exicycler™ 384 Real-time Quantitative Thermal Block is warranted by Bioneer against manufacturing defects in materials and workmanship for a limited warranty period of one year. Bioneer will charge for repairing products for the following conditions:

- Fault caused by the customer.
- The product is repaired after expiry of the warranty period.

Before you install *Exicycler*™ 384, check shipped materials with the system component listed below:

System Components

Component Lists		Check
Exicycler™ 384	1	☐
USB 2.0 high speed cable	1	☐
Power cord[1]	1	☐
Software	1 CD	☐
User Guide	1	☐
Optical tape sealing applicator	1	☐

[1] Provided according to country standard.

Specifications

Physical specifications	
Dimension	35.5cm(W) x 54cm(D) x 47cm(H) 13.98in(W) x 21.26in(D) x 18.50in(H)
Weight	41 ㎏ (90.39lbs)
Sample capacity/ size	384-well plate
Sample volume	5~20 ㎕ (10 ㎕ recommended)
Power consumption	100-240VAC, 50/60㎐, Max 800VA
Operating temperature	15~35℃ (59~95°F)
Operating humidity	20~80%, no condensation
Thermo module specifications	
Method of heating / cooling	Peltier
Temperature range	4.0℃ ~ 99.9℃ (39.2~211.82°F)
Max ramp rate	4.5°C/sec (8.1°F/sec)

Temperature accuracy	± 0.3℃ (± 0.54°F)
Temperature uniformity	± 0.3℃ (± 0.54°F)
Lid temperature	40 ~ 120℃ (104~248°F)
Gradient range	20 ~ 95℃ (36 ~ 203°F)
Temperature differential range	1 ~ 20℃ (1.8 ~ 36°F)
Temperature increment range	0.1~2.0℃ (0.18~3.6°F)
Time increment range	1 sec ~ 60 sec
Ramp rate control range	1~100%
Computer specifications	
Operating system	Window7 & 10(32/64 bit OS)
Processor speed	Intel Dual Core (1.8GHz) or higher
Memory	1GB or higher
Communication port	USB 2.0 high speed
Screen resolution	1280 x 1024 or higher
Optical Part	
Light source	Short arc lamp(120W)*
Sensor	16 bit 2D CCD
Excitation Filter / Emission Filter**	5 Set

* Continuously turning on and off the lamp reduces the lamp life span.
* The lamp generates heat when it starts. Make sure that you turn off *Exicycler*™ when cooling fan stops running. It usually takes about 2 minutes for the cooling fan to stop.
**Please refer to a chart below for more information about filter sets.

※The specifications of this product may change without notification for performance enhancement.

† Filter sets

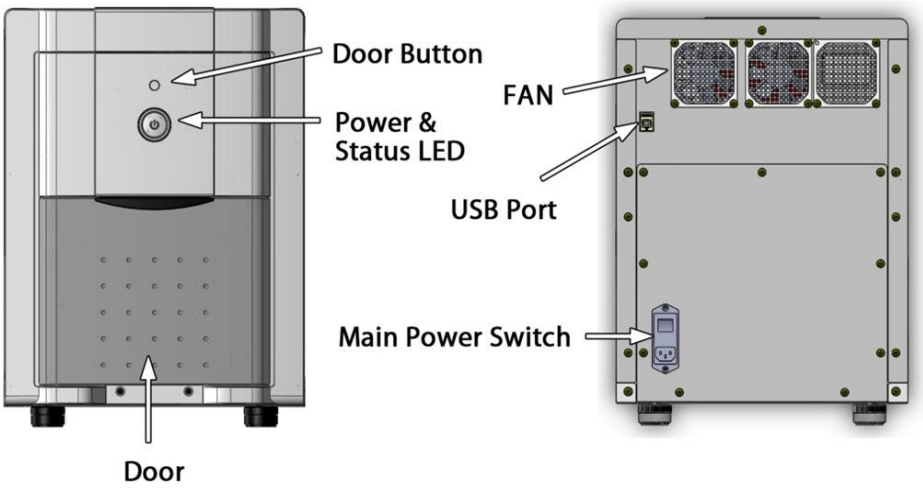
Position	Excitation (㎚)	Emission (㎚)	Set	Fluorescent dye
1	Blank	Blank	1	–
2	475	530	1	FAM, SYBR Green I
3	520	560	1	JOE, TET
4	550	590	1	TAMRA, CY3
5	570	630	1	Texas Red, ROX
6	630	690	1	CY5

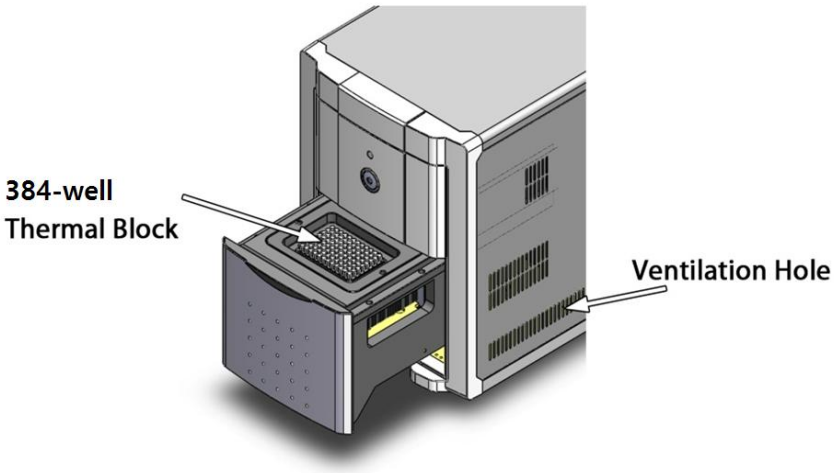
⚠ WARNING

Chemical hazard: Exposure to following florescent dyes may cause eye and skin and respiratory tract

irritation. Read the MSDS before use and follow the instructions if swallowed or inhaled. Wear appropriate protective eyewear, clothing, and gloves.

System Views





Installation and System Operation

Overview

To run *Exicycler*™ 384 Real-Time Quantitative Thermal Block, camera driver and operation software should be installed in a computer. When first installing the camera driver and operation software, or re-installing these components, please see below for instructions on how to install them.

Site Requirements

Exicycler™ 384 Real-Time Quantitative Thermal Block is for an indoor use. Ensure that the installation site:

- Meets the spatial and weight requirements.
- Meets environmental requirements.
- (The instrument) Is within 1.5 m (4.92 ft) of a 800VA power receptacle.
- (The computer) Is within 1.5 m (4.92 ft) of a 500VA power receptacle.
- Is away from water.

Required Materials

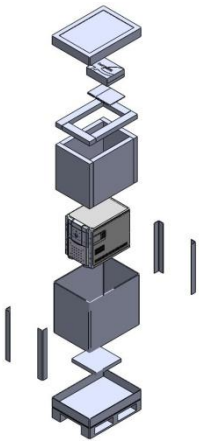
Scissors, pocket knife, or box cutter

Unpacking *Exicycler*™ 384

CAUTION

Save the packing materials and box in case you need to ship the instrument to Bioneer for service.

1) To unpack *Exicycler*™ 384:



a) Cut the straps securing the instrument box.

b) Cut the tape securing the top flaps of the instrument crate, then open the flaps.

c) Remove *Exicycler*™ 384 accessories from the instrument and set them aside.

d) Lift and remove the cover from the instrument crate.

e) Remove the packing material from *Exicycler*™ 384, and then inspect the instrument for shipping damage.

CAUTION

If *Exicycler*™ 384 is damaged, note location and appearance of the damage, and then contact Bioneer Technical Support or your service representative.

2) Move *Exicycler*™ 384 to the desired installation site. Follow these guidelines for lifting and moving:
   - Make sure that you have a secure, comfortable grip.
   - Keep your spine in a neutral position.

- Bend at the knees and lift with your legs.
- Do not lift and twist your torso at the same time.

3) Open the box containing *Exicycler*™ 384 accessories, then verify that it contains all the necessary components:
   - Refer to Component Lists.

Setting Up the Computer

Exicycler™ 384 Real-Time Quantitative Thermal Block operates through a computer. Therefore, to install *Exicycler*™ 384, a computer or a laptop is necessary. Computer must meet the following requirements listed below. Please contact Bioneer Customer Service Center if you have any questions regarding installation.

Computer Requirements

- Intel Dual Core (1.8GHz) or higher
- Windows 7 & 10 Operating System for Korean or English Version (32/64 bit)
- 1.0GB RAM or higher
- 1280 * 1024 screen resolution
- USB 2.0 high speed port
- 20GB Hard disk drive minimum
- Microsoft Excel (Option)

For questions regarding problems with the computer or operating system, please contact the computer manufacturer.

1) The computer must have at least one communication port for USB data transfer.
2) It is recommended to install an anti-virus software in order for *Exicycler*™ 384 to operate safely. A firewall should also be setup to prevent unwanted information coming in from external networks. Please contact your IT department to setup anti-virus software and firewall.
3) It is not recommended to insert a flash drive into USB port *Exicycler*™ 384 is running. It may cause a technical problem between *Exicycler*™ 384 and the computer.

Connecting the Computer and *Exicycler*™ 384

There are main power connector and a communication port connector (USB) in the rear of *Exicycler*™ 384. The computer should be turned off when connecting to *Exicycler*™ 384 to prevent any damages to the communication port. Please see below for instructions on how to connect *Exicycler*™ 384 to the computer.

1) Unpack *Exicycler*™ 384 and make sure all components are included.
2) *Exicycler*™ 384 must be installed in an area where it is not exposed to sunlight and must be set on a stable and level surface.
3) Set up the computer at the installation site.
4) Place *Exicycler*™ 384 in the installation site carefully.
5) Connect USB 2.0 cable to USB connector on the back of *Exicycler*™ 384, then to the computer.
6) Connect power cable to *Exicycler*™ 384, then to the receptable wall circuit.

Installing the Operation and Analysis Program

To run *Exicycler*™ 384 Real-Time Quantitative Thermal Block, the operation software and the camera driver must be installed in the computer. When installing the operation software and the camera driver for the first time, or re-installing these components, please see instructions below on how to install them yourself. In case of running the software for only data analysis purpose, please refer to this section for software installation.

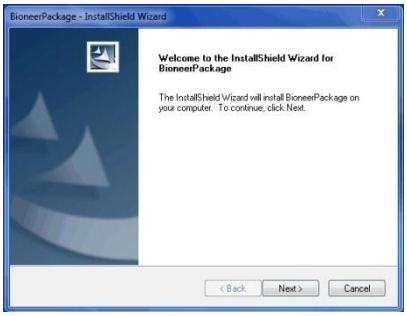
IMPORTANT:
Do not turn on *Exicycler*™384 unless you have installed the Operation and Analysis Program and a camera driver. First install the Operation Software and then the camera driver.

To install Bioneer Package and Exicycler384 softwares for an *Exicycler*™ 384 device under Windows 7, follow the instructions below:

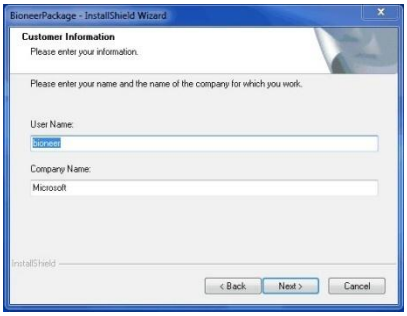
- If a previous version of software has been installed on your computer, please remove the programs and folders from the computer (Refer to 'Troubleshooting, 4. The Bioneer Package is not installed correctly'). Remaining program folders may cause errors during installation. This section is for the first time installation or re-installation of the software.
- The Bioneer Package has to be installed prior to installing Exicycler384 software. The procedures of Bioneer Package installation is described below.

Installing Bioneer Package

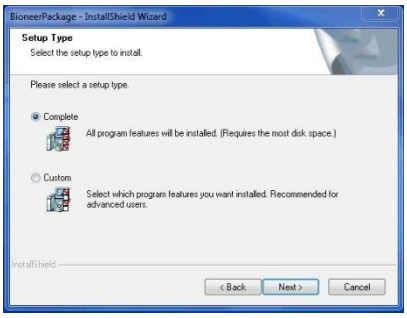
1) Turn on the computer, but not *Exicycler*™ 384 Real-Time PCR machine. Place *Exicycler*™ 384 installation CD into the CD-ROM. Go to 'Bioneer Package' folder in the CD drive and run **Setup.exe.**



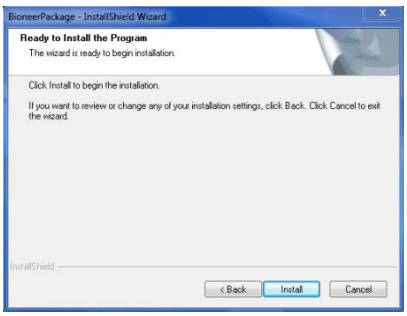
2) To install Bioneer Package, click **Next**. The installation will proceed in **C:₩Bioneer_Package.**



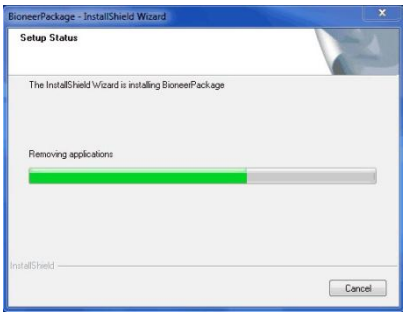
3) Enter desired name in the **User Name** box and **Company Name** box.



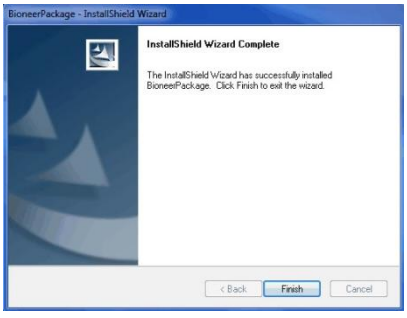
4) Select a setup type to install. **Complete** is recommended for installing all program features. **Custom** installation is recommended only for advanced users. Click **Next**.



5) InstallWizard is ready for installation. If you want to review or modify your settings, click **Back**. Click **Install** to start installation.

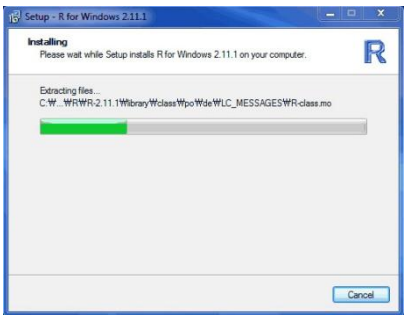


6) This process takes several minutes. Please do not cancel until installation is complete.

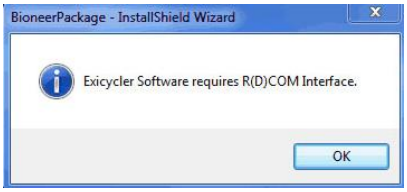


7) When installation is complete, please click **Finish** to finish installation Shield Wizard.

8) R program is also needed for Exicycler software. If you click OK, installation will begin.

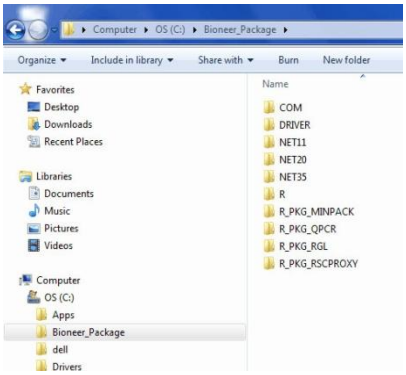


9) R program will be installed automatically. Please wait until installation is complete.



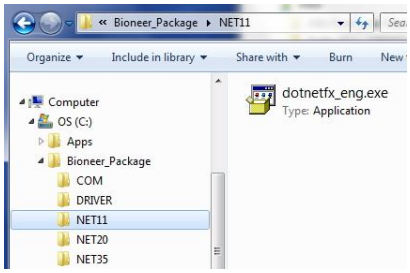
10) Click OK for installing R(D)COM interface.

11) R icon will be shown on the Desktop when installation is successful.

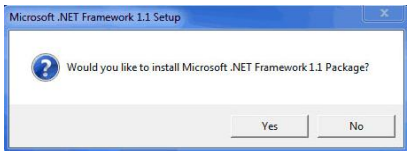


12) The folders shown here are created after installation has finished. Please confirm 'Bioneer_Package' and sub folders.

Installing .NET Framework 1.1 Package

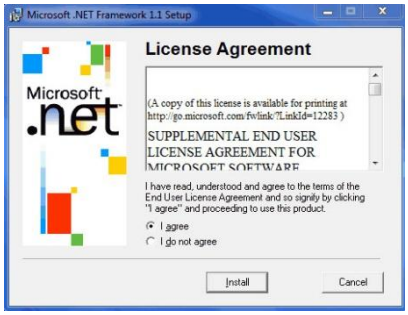


1) In order to install '.NET Framework 1.1 Package', run **dotnetfx_eng.exe** file of the **C:₩Bioneer_Package₩Net11** folder.

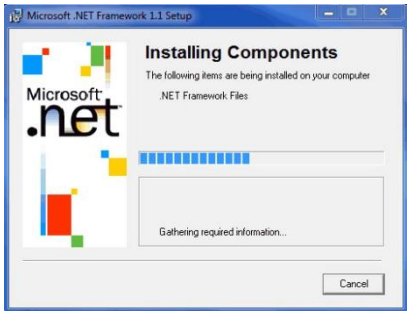


2) Click **Yes** for installing '.NET Framework 1.1 Package'.

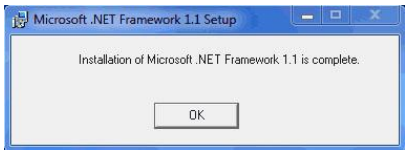
3) A setup window asking about the license agreement is displayed.



4) Select **I agree** to activate install button, Click **Install** to begin installation.



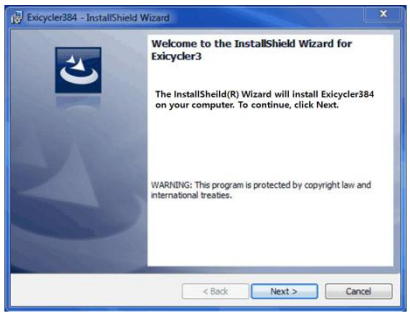
5) All components related to .Net Framework are being installed.



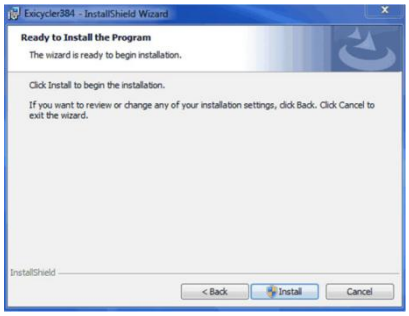
6) Installation of .NET Framework 1.1 is complete. Click **OK** to finish installation.

Installing Exicycler384 Software

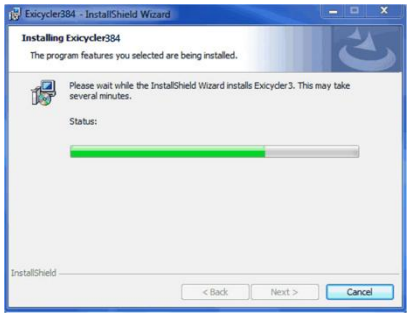
1) In Exicycler384 folder of the provided CD, you can see **setup.exe** file and double click InstallSheild Wizard to install required items automatically.



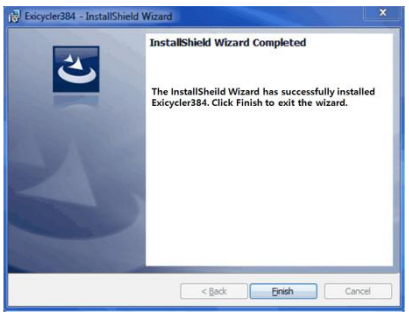
2) Click **Next** and go to the next step.



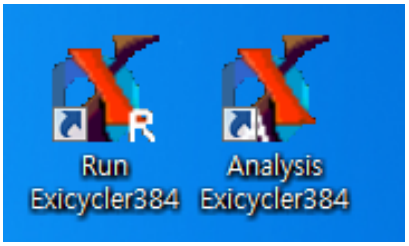
3) Click **Install** to begin installation. If you want to change any settings or cancel installing, click **Back** or **Cancel**.



4) Please wait while Install-Shield Wizard installs Exicycler384.

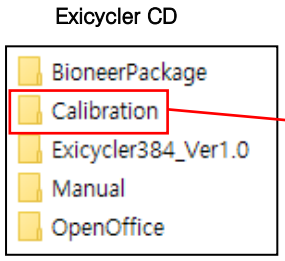


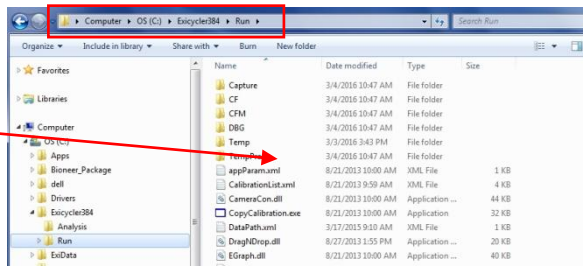
5) Message indicating "Install-Shield has successfully installed Exicycler384." is shown. Click **Finish** to complete installation.

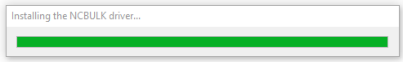


6) On the Desktop, these icons are created when installation is completed successfully.

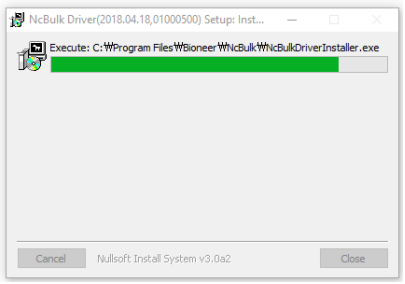
7) In the **C:₩Exicycler384₩Run** folder, folders and files that are shown on the image (below) are created. Copy '**Calibration**' folder from *Exicycler*™ 384 CD into **C:₩Exicycler384₩Run** folder.



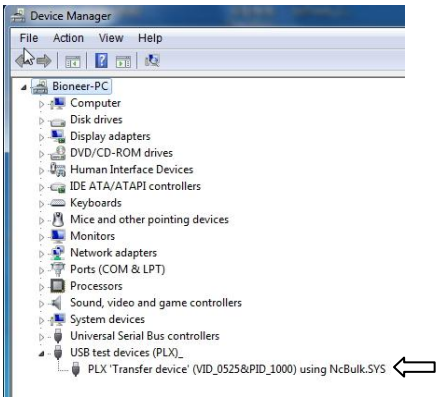




8) Run **'NcBulkDriverSetup.exe'** to install the camera driver of the *Exicycler*™ 384.



9) When you run the file, the driver will be installed automatically.

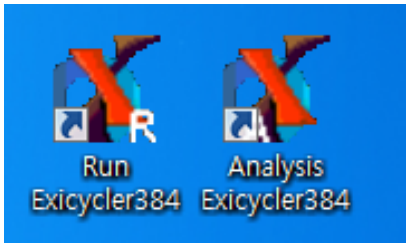


10) Open '**Device Manager**' and confirm that device driver has been installed. If **USB test devices (PLX)_₩PLX'Transfer device' VID_0525&PID_1000) using Ncbulk.SYS** has been installed properly, connected USB will be indicated.

11) Now you should connect *Exicycler*™ 384 system to a computer using provided USB cable.

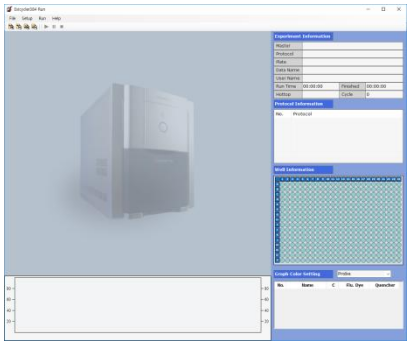
12) Turn on *Exicycler*™ 384. The status LED will be blue when the power is supplied properly.

13) Press the power button for a second to start self-diagnosis. The status LED will start blinking in green after two short beeps when the self-diagnosis is complete.



14) Click **Run Exicycler384** icon to start *Exicycler*™ 384 software.

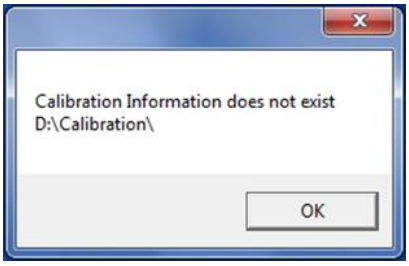
15) You can see this screen when installation is completed successfully. The software version is also indicated.



16) Now you are ready to operate *Exicycler*™ 384 system.

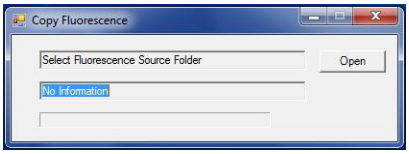
Troubleshooting

1. Appropriate calibration data does not exist

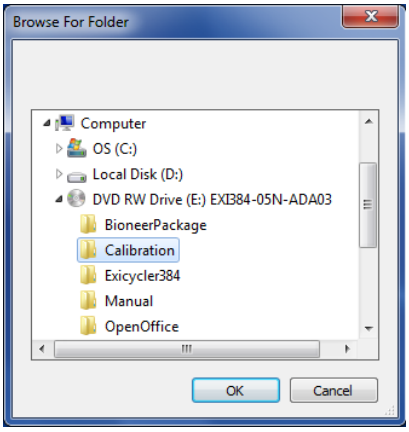


1) CD provided with *Exicycler*™ 384 has Calibration data for each device. Calibration data have to be copied into C:₩Exicycler384₩Run folder in order for a normal function. If Calibration folder does not exist on this path, following message will be displayed.

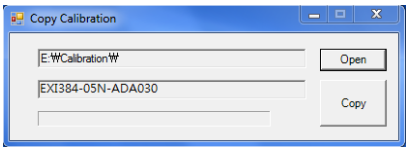
2) Click OK to change the path.



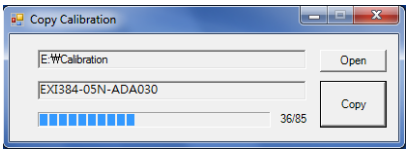
3) You should assign the path of the Calibration folder to be copied automatically. Click Open and you will see the following browser.



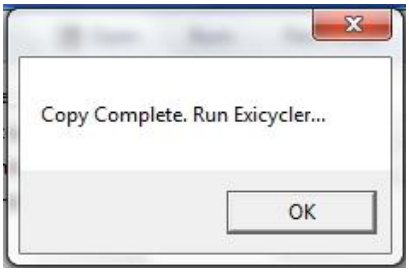
4) After assigning the Calibration folder (ex. E: ₩Calibration), click OK.



5) Copy Calibration window displays the path of Calibration folder and serial number of your device. If the serial number matches with the connected *Exicycler*™ 384 system, click Copy button on the right hand side of window.



6) Please wait for a few minutes to complete.

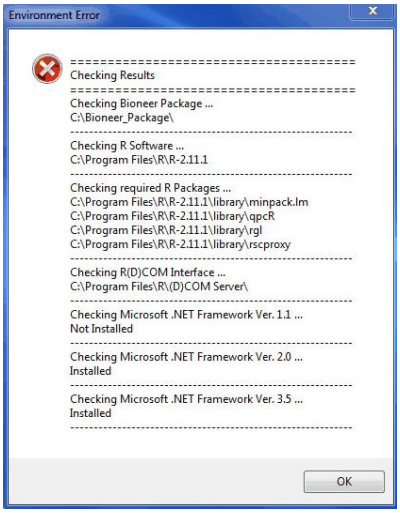


7) When copying calibration folder is complete, click **OK** to finish the process. Please confirm the connection between device and computer, and restart Exicycler384 software.

2. The computer does not register *Exicycler*™ 384 when launching 'Run Exicycler384' software.

This message means that the device is not turned on or the appropriate device driver has not yet been installed on the computer. If you see this message despite the instrument is turned on, then you should re-install the device driver to solve this problem. The driver installation steps are described in '**Installing Exicycler384 Software**' section.

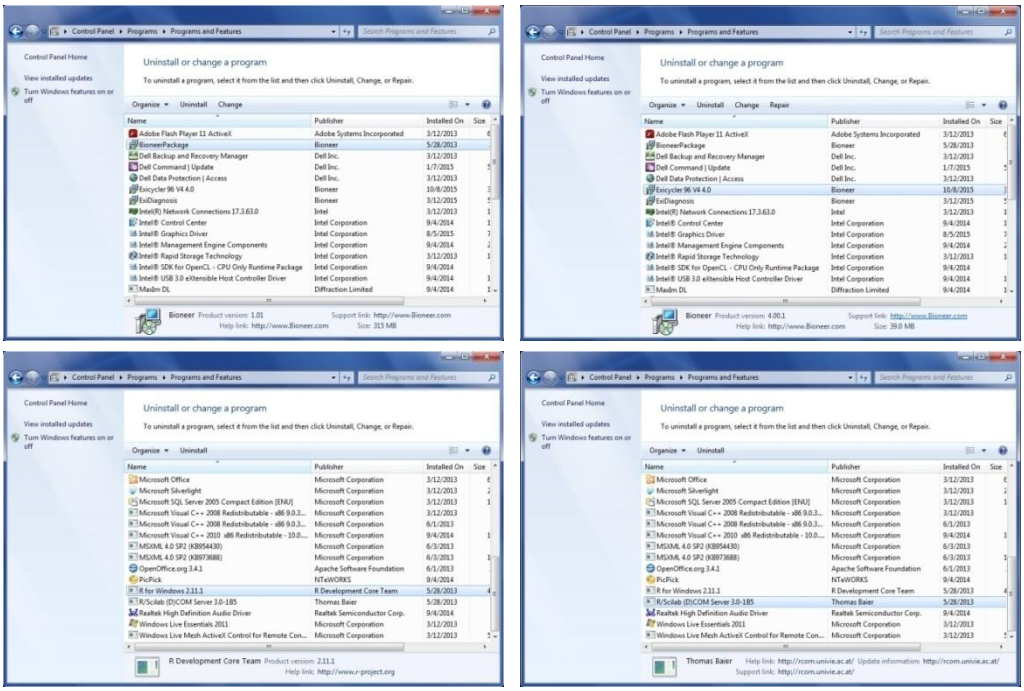
3. An Error message is displayed when lunching 'Run Exicycler384' software.



This message indicates that '.NET Framework 1.1' has not yet been installed. Go to '**Installing.NET Framework 1.1 Package**' section and re-install as described.

4. The Bioneer Package is not installed correctly.

If an incorrect installation was performed previously, 'Bioneer Package' may not be installed correctly. Go to **Control Panel–Programs Add/Remove** and remove 4 programs from the computer – **'Exicycler 384 V1 1.0', 'Bioneer package', 'R for windows 2.11.1' and 'R/Scilab(D)COM Server3.01–B5'**. After removing them from the computer, repeat the entire installation process.



Turning On and Self-diagnosis

1) Turn on *Exicycler*™ 384 by switching on the main power switch in the rear of the instrument. When power is supplied properly, a status LED turns blue.

2) Press the **POWER** button for a second to start self-diagnosis.

3) When the status LED is blinking green, you can either open or close a main door by pressing a **DOOR** button for a second. The status LED is blinking in purple when the door is open and is back to green when the door is closed.

Power Off

Power On

Door Open

Standby

IMPORTANT:
The status LED starts blinking green after two short beeps when the self-diagnosis is complete. *Exicycler*™ 384 is now ready to operate. The status LED will turn red when the self-diagnosis fails or excessive physical shock is applied from outside. (e.g. Make sure that there are no objects placed in front of the door) Make sure that the status LED is blinking green before operating *Exicycler*™ 384 to prevent mis-operation or damage. A result of the self-diagnosis is provided under 'ExiConfig'.

※ Status LED color

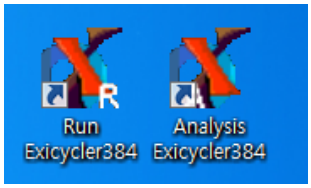
a. Red: Error
b. Blue: Standby
c. Green: Ready for running
d. Yellow: Self test
e. Cyan: Running
f. Purple: Door open
g. White: Pause

NOTE:
You can stop the operation here and turn off the *Exicycler*™ 384 by pressing the POWER button for a second when it is blinking in green like Step 3.

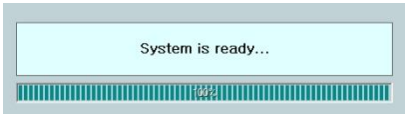
CAUTION
Do not place any obstacle in front of the system and do not load a plate or take it out when the door is moving. It can cause serious damage and lead to severe injury.

Running Real-Time PCR Using *Exicycler*™ 384 Software

Starting 'Run Exicycler384' program



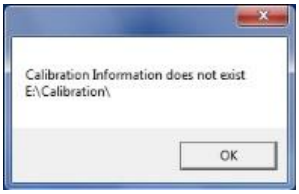
1) Ensure that *Exicycler*™ 384 is in the "Standby" mode. The status LED must be blinking green. Double click **Run Exicycler384** icon on the desk top to start **Run Exicycler384**.



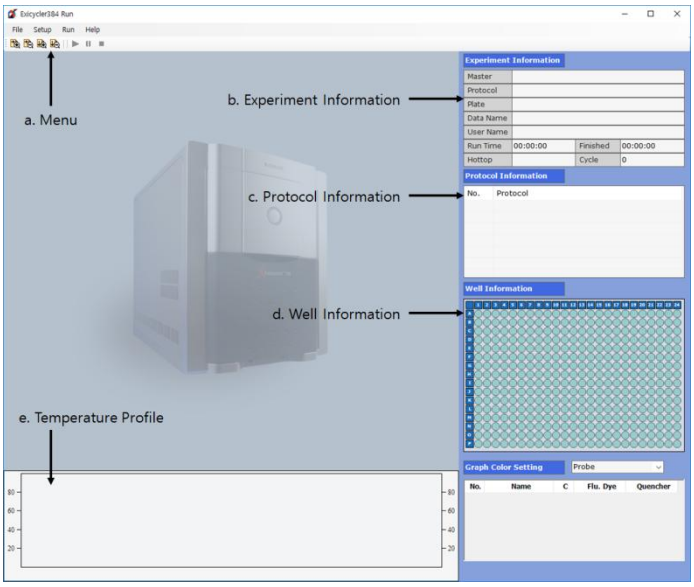
2) **System Check** window will pop up.

IMPORTANT:
The following error message will appear if *Exicycler*™ 384 is not turned on. Turn on *Exicycler*™ 384 by pressing the power switch at the back of *Exicycler*™ 384 and the power button in front of *Exicycler*™ 384. Then double click **Run Exicycler384** icon on the desktop again.

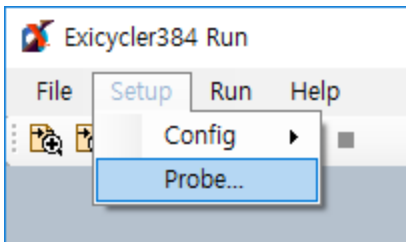
IMPORTANT:
The following error message will appear when the calibration has not been performed for *Exicycler*™ 384 or the calibration data is not present. Close Exicycler384 program and copy calibration folder into **C:₩Exicycler384₩Run**. (The CD provided with *Exicycler*™ 384 has calibration data for each device.)



3) The following window will appear when System Check is complete.

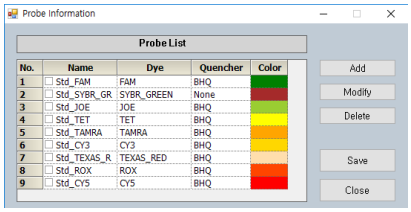


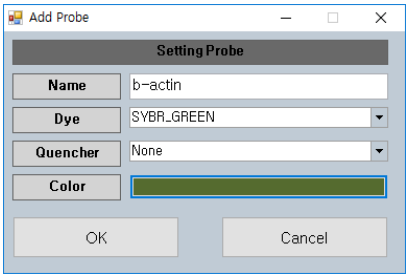
a. **Menu** consists of File, Setup, Run, Window, and Help.
b. **Experiment Information** displays file name, user name, and elapsed and estimated finish time.
c. **Protocol Information** displays a cycling protocol in detail.
d. **Well information** displays sample and probe specification for each well.
e. **Temperature Profile** displays a temperature curve of the cycling protocol.



4) Go to **Setup > Probe** from the top menu.

IMPORTANT:
9 different probe options are available in *Exicycler*™ 384. Each probe option includes specifications for a fluorescence dye and a quencher. You can either select one of existing probe options in Probe List or add your own. Ensure you specify an appropriate probe option for accurate data analysis.

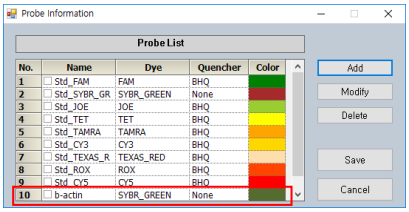




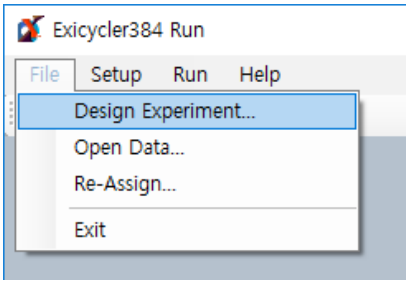
5) Probe Information window will appear. Select one or click **Add** to add additional probe.

*** Additional Probe Setting:**
ex) Enter 'b-actin' in the field.

NOTE:
In the **Dye** drop-down list of the **Add Probe** window, select **SYBR_Green**. Select **None** in the Quencher drop-down list. Select your desirable color for display and then click **OK**.

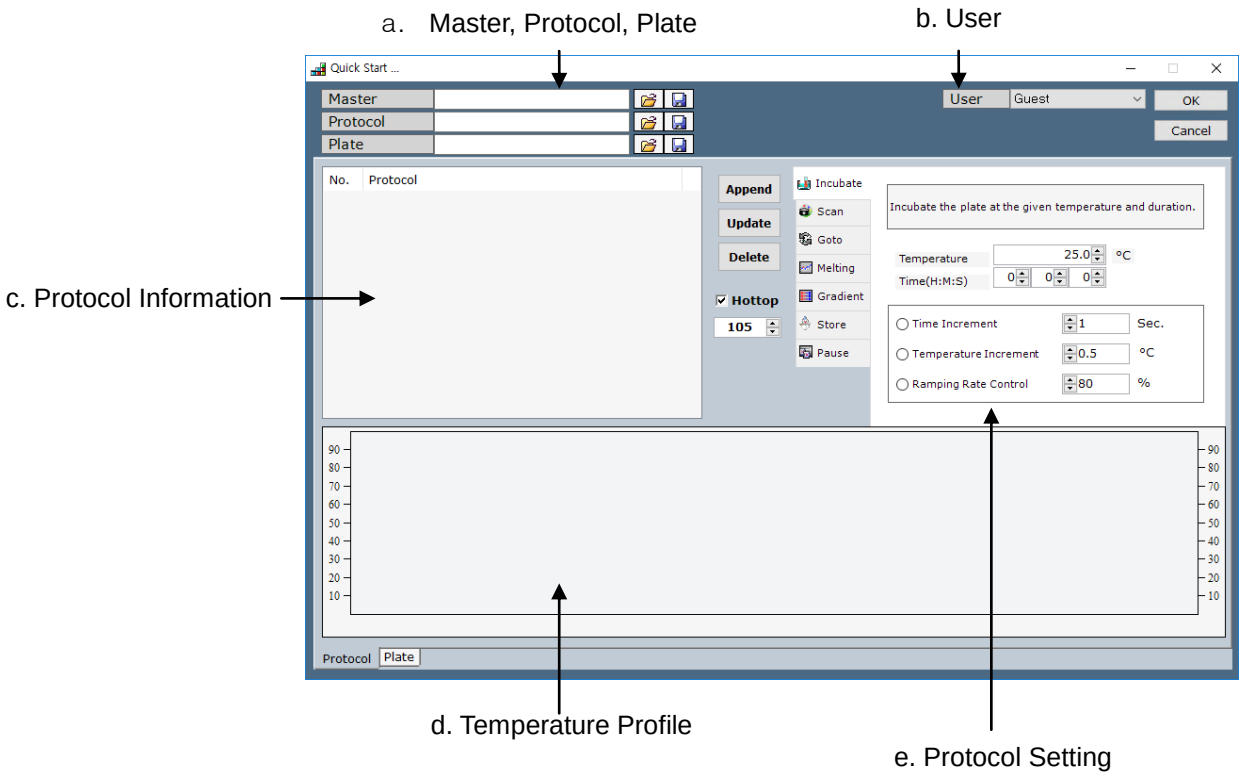


6) Check if your new probe is shown under the list, and then click **Save**.



7) Go to **File > Design Experiment** to design a new experiment.

8) **Quick Start** window will appear as shown below:



a. **Master, Protocol, and Plate:** You can create, save, or open a file.
b. **User:** Select or create the user name for personal account management.
c. **Protocol Information:** Displays a cycling protocol in detail.
d. **Temperature Profile:** Displays a temperate curve of the cycling protocol.
e. **Protocol Setting:** Specifies protocol specifications such as temperature, time, and a number of cycles.

NOTE: Example of qPCR protocol

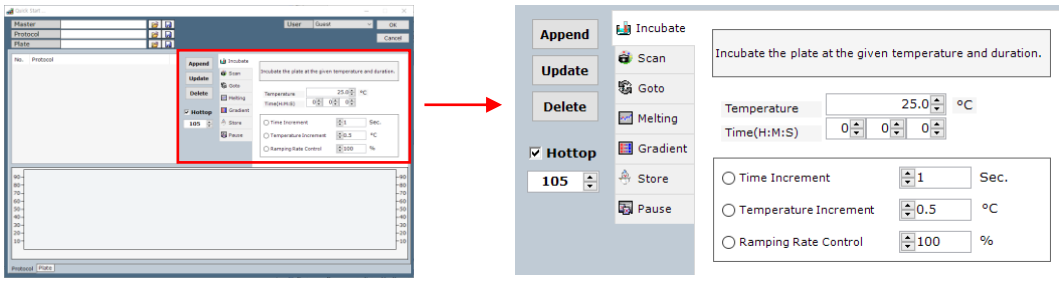
SYBR Green I

No.	Protocol
1	Incubate at 95.00°C, for 0:5:0
2	Incubate at 95.00°C, for 0:0:10
3	Incubate at 60.00°C, for 0:0:20
4	Scan
5	Goto Line : 2, Cycle : 40
6	Melting 60°C to 94°C, Every 1.0°C, 1 Sec.
7	Incubate at 25.00°C, for 0:1:30

TaqMan probe

No.	Protocol
1	Incubate at 95.00°C, for 0:5:0
2	Incubate at 95.00°C, for 0:0:10
3	Incubate at 60.00°C, for 0:0:20
4	Scan
5	Goto Line : 2, Cycle : 40
6	Incubate at 25.00°C, for 0:1:30

9) Click **Incubate** tab and enter a temperature in the **Temperature** field and then a time in the **Time** field.



a. **Incubate** sets up a temperature and a time for the thermal block.
   Time Increment sets up time increment per cycle.
   Temperature Increment sets up temperature increment per cycle.
   Ramping Rate sets up a ramping rate.
b. **Scan** measures fluorescence signals emitted from samples.
c. **Goto** specifies a starting step of a thermal cycling and a number of cycles.
d. **Melting** sets up starting temperature and ending temperature for melting curve analysis to distinguish specific and nonspecific amplification products when SYBR Green is used for Real-Time PCR.
e. **Gradient** is used to evaluate an optimum annealing temperature condition for amplification.
f. **Store** keeps the 384-well thermal block at a set temperature until you stop *Exicycler*™ 384. It is not recommended to use **Store** step when it is excessively humid.
g. **Pause** is used to pause *Exicycler*™ 384 when it is necessary to check samples during experimentation. The door of *Exicycler*™ 384 will open when the **Pause** step is inserted in the protocol file. Click **Run** to resume the experiment.
h. **Hottop** sets up a temperature for the heating lid.

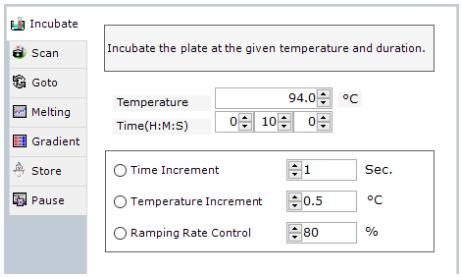
NOTE:
Scan is used to detect fluorescence signals from samples. The fluorescence signals are measured for about 25 seconds at set temperature of the previous incubation step. If the **Scan** step is not included in the protocol file, a conventional PCR will be carried out without scanning fluorescence signals.

Melting sets up starting and ending temperature for melting curve analysis to distinguish specific and non-specific amplification products when SYBR Green is used for Real-Time PCR. Enter starting temperature in the **From** field, ending temperature in the **To** field, a temperature interval within a range of 0.1°C to 1.0°C in the **Between** field, and a hold time within a range of 1 second to 255 seconds in the **Hold Time** field.

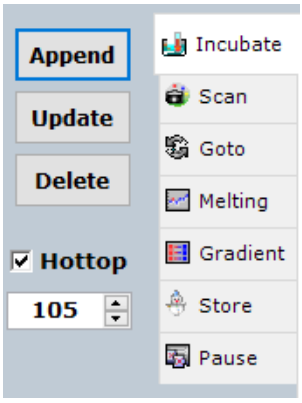
Gradient is used to select an optimum annealing temperature for amplification and programs a temperature gradient up to 20°C across the rows of a sample block. Enter starting temperature in the **From** field, ending temperature in the **To** field, and time within a range of 1 second to 5 hours 59 minutes and 59 seconds in the **Time** field.

Store keeps the 384-well thermal block at a set temperature when thermal cycling is complete. The heated lid will start cooling down automatically. *Exicycler*™ 384 will keep the main door closed and will maintain thermal block at the set temperature until you stop *Exicycler*™ 384. If the **Store** step is not inserted into the protocol file, the main door will open automatically when the thermal cycling is done.

Hottop Check Box sets up a temperature for the heating lid within a range of 40°C to 120°C. The default temperature is 105°C. When the check box is unselected, the heating lid will not be heated.



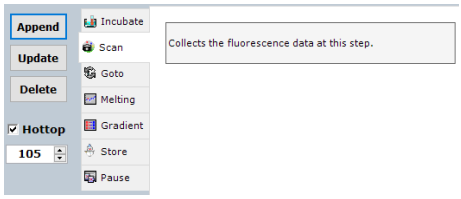
10) Click **Incubate** tab to add Incubation step and set Temperature/Time (ex, **94**℃ **10** min).



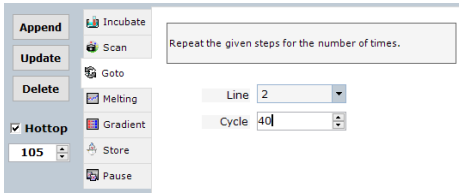
11) Click **Append** to add a new step into the protocol.
   a. **Append** button is used when you want to add new steps into the protocol.
   b. **Update** button is used when you want to make changes to a selected step.
   c. **Delete** button is used when you want to delete a selected step from the protocol.

12) Repeat steps 10 and 11 if needed (i.e. 94°C 30 sec / 55°C 30 sec / 72°C 30 sec).

NOTE:
To edit **Incubate** steps, click one of the steps in the **Protocol Information** window and edit. Click **Update** to change the protocol.



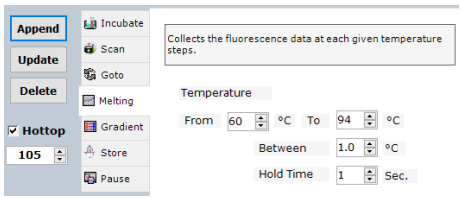
13) Click **Scan** tab and then **Append**.



14) Click **Goto** tab and select a starting step (i.e. "2") in the **Line** drop-down list. Enter a number of cycles (i.e. "40") in the **Cycle** field, and then click **Append**.

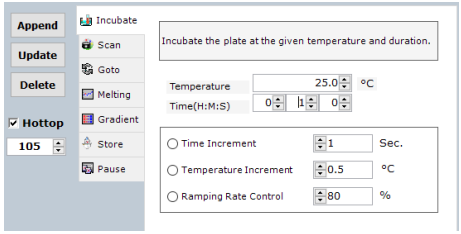
NOTE: Melting curve analysis

Melting curve analysis is an assessment of the dissociation-characteristics of double-stranded DNA during heating.



15) Click **Melting** tab to perform melting analysis. Enter starting temperature in the **From** filed (i.e. "60"), ending temperature in the **To** field (i.e. "94"), a temperature interval in the **Between** field (i.e. "1"), and a hold time in the **Hold Time** field (i.e. "1").

16) Click **Append.**



17) Click **Incubate** to add finish step (i.e. 25°C 1 min) and then click **Append.**

NOTE:

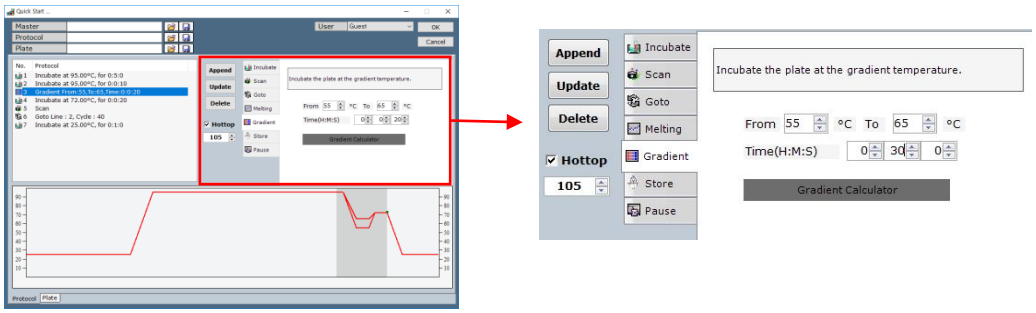
The door of *Exicycler*™ 384 will open automatically when a Real-Time PCR protocol does not include **Store** step. If **Store** step is included in the protocol, you must stop *Exicycler*™ 384 by clicking **Stop** button in order to open the door. Otherwise, *Exicycler*™ 384 will continue to store samples in the thermal block at the set temperature.

IMPORTANT:

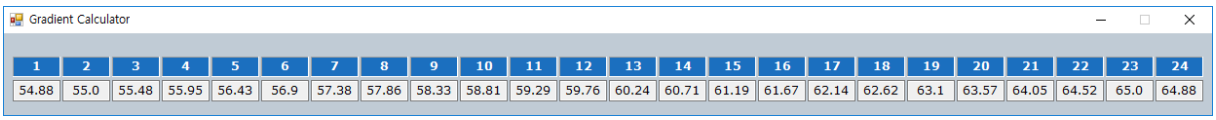
To set the temperature on the Hot top Make sure that the Hottop check box is selected. The default value is set to 105 ℃. If you turn off the Check box hot top is not heated.

Additional: Setting the temperature gradient mode

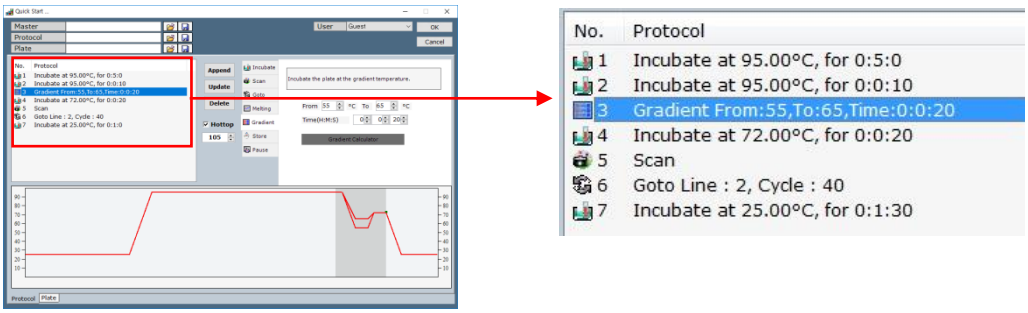
① Click **Gradient** tap, enter starting temperature in the **From** field, ending temperature in the **To** field.

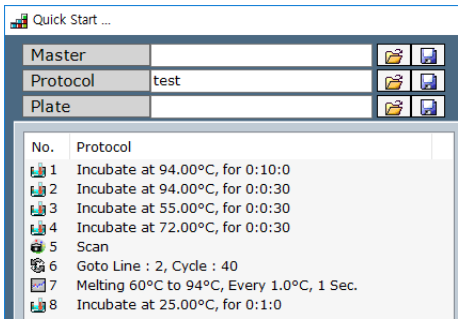


② Press 'Gradient Calculator' button below the time setting window, and 'Gradient Calculator' window will appear. It will show predicted temperatures on each well.



③ Click **Append**.

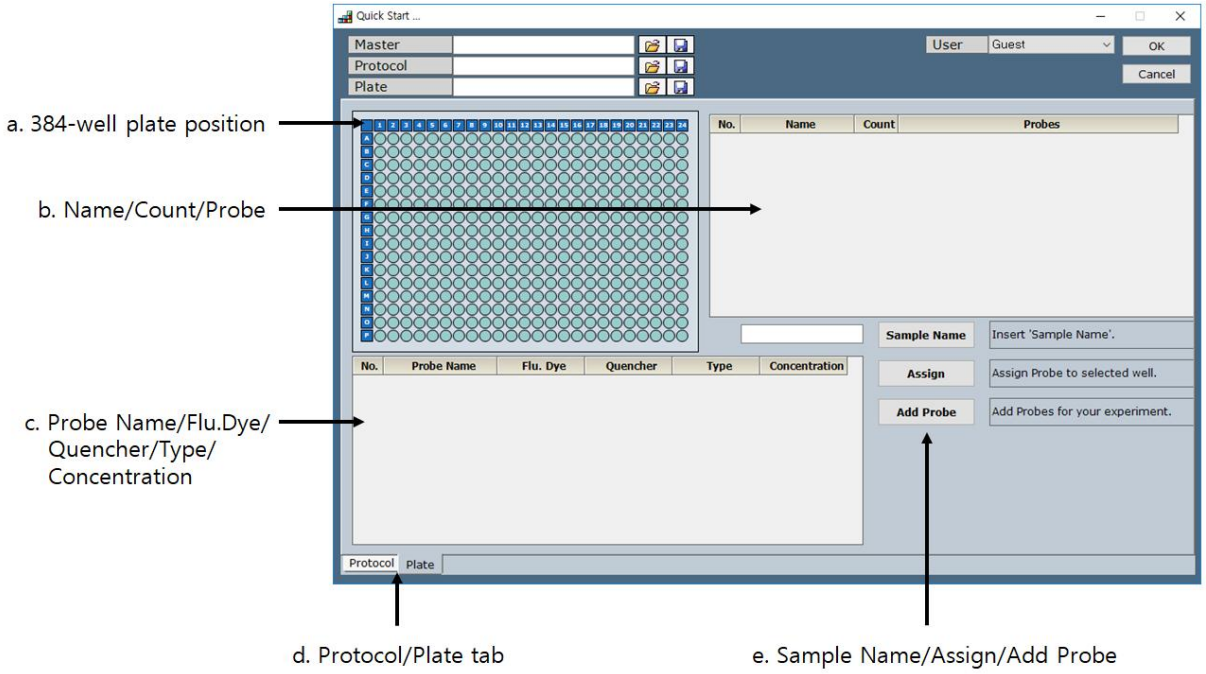




18) Enter a protocol file name in the **Protocol** field at the top left (i.e. "Test") and then click button to save the protocol.

NOTE:
Click button from the Quick Start to open the saved Master, Protocol and Plate files when needed.

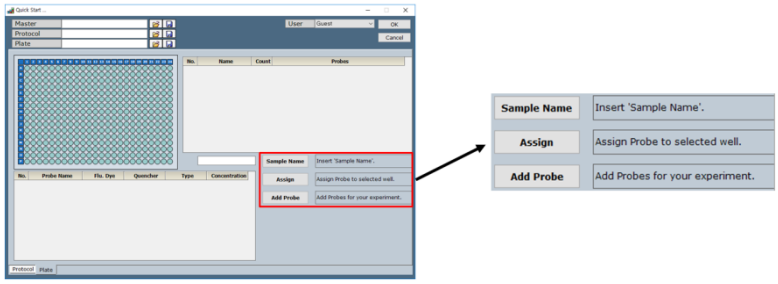
19) Once the protocol is saved, click **Plate** tab to create a plate file. Plate window will appear as follows:



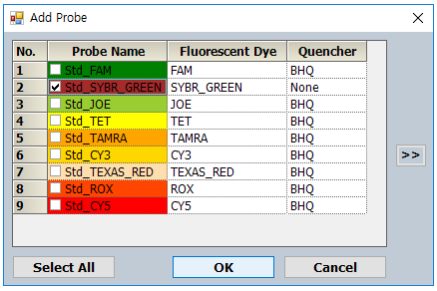
a. **384-well plate position** specifies well locations for samples.
b. **Name/Count/Probe** displays a name and probe information for each sample.
c. **Probe Name/Flu. Dye/Quencher/Type/Concentration** specifies a name, a fluorescence dye, a quencher dye, a type, and a concentration for each sample.
d. **Protocol/Plate tab** switches between the **Protocol Information** and the **Plate Information** windows.
e. **Sample Name** enters a sample name.
Assign specifies the information of each well such as probe and type.
Add Probe adds the probe to be used in experiment.

NOTE:
Ensure that you specify appropriate information (i.e. probe set up and sample type) for each well before running experiment in order to generate accurate data.

20) Click **Add Probe**, the Add Probe window will appear.

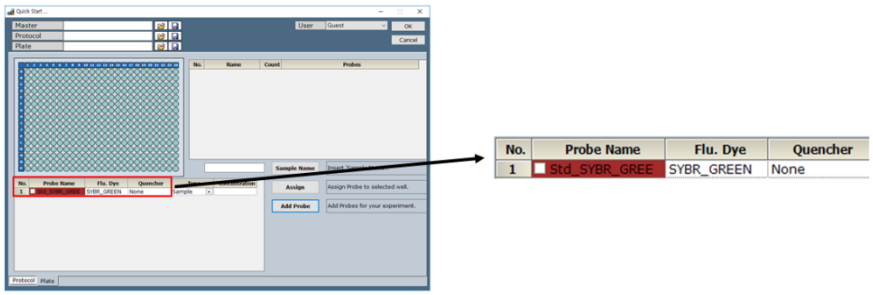


21) Select appropriate probes, and then click **OK**.

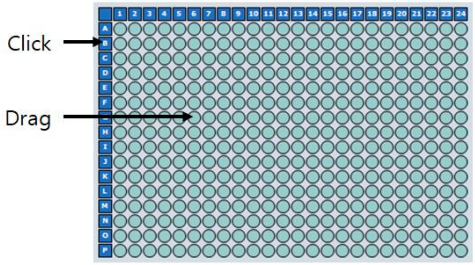


a. **Probe Name**: displays probe name assigned by the user.
b. **Fluorescent Dye**: displays name of a fluorescence dye assigned by the user.
c. **Quencher**: displays name of a quencher assigned by the user.

22) The probe is loaded and shown as follows:

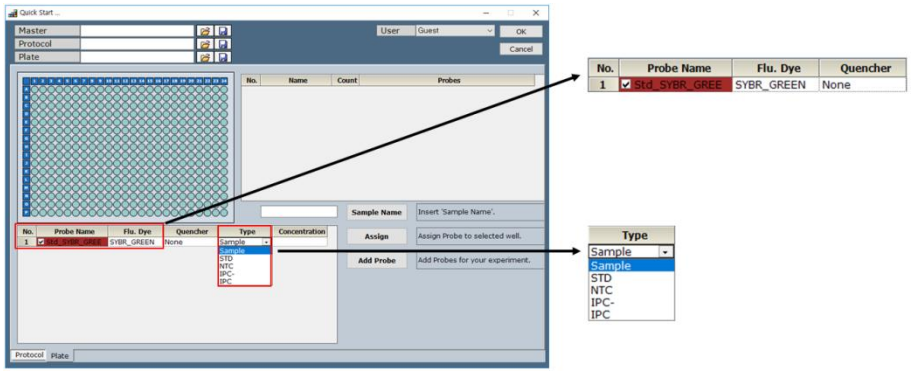


23) Select wells from the 384-well plate diagram.



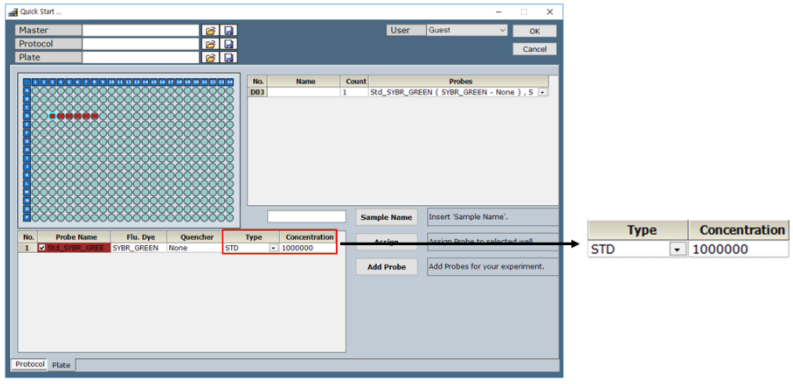
NOTE:
Use the left mouse button when selecting a single well. When selecting a range of cells, click the first cell in the range, and then drag to the last cell. You can also select cells in a row or column by pressing CTRL and clicking the row or column heading. To select all cells in 384-well plate, click the cell at the top left corner of the diagram.

24) Click the probe option cell and then select **STD** in the **Type** drop-down list. Select the check box of **Probe Name** and then click **Assign**.

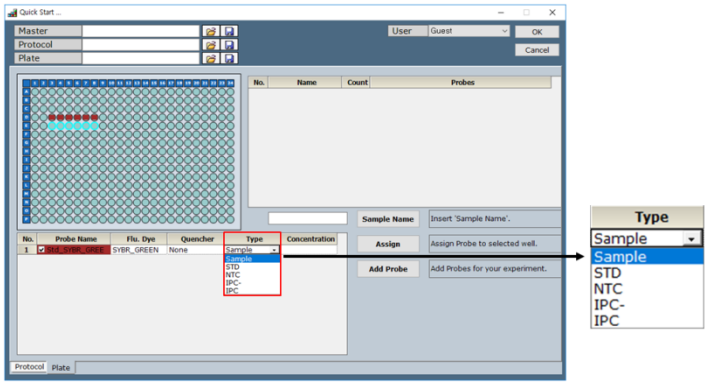


a. **Sample** represents unknown samples.
b. **STD** stands for a standard sample of a known concentration.
c. **NTC** stands for **N**o **T**emplate **C**ontrol and is a sample without a template.
d. **IPC** stands for **I**nternal **P**ositive **C**ontrol and is a sample that monitors the PCR run during Existence / Non-existence reaction. It will also diagnose cause of the negative result from PCR.
e. **IPC-** is a sample that does not contain a template of IPC.

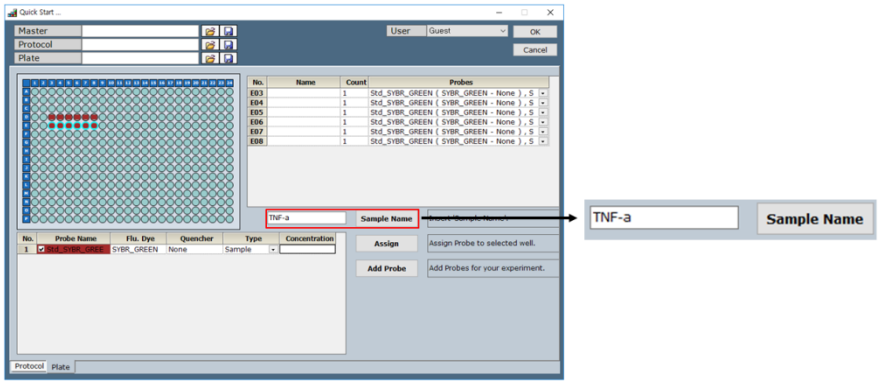
25) For example, Click well D3 and enter '1000000' for a concentration of 10^6-copy and then click **Assign**. Repeat this for wells D4 through D8 with concentrations of serial diluted standards.



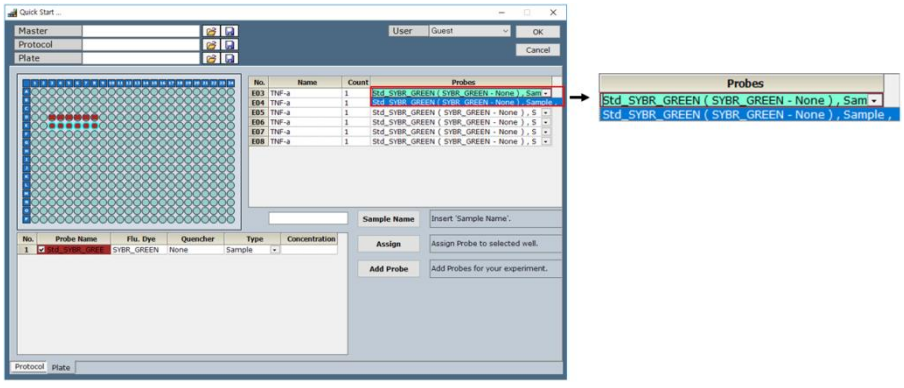
26) Select wells E3 through E8 in the plate diagram. Select **Sample** in the **Type** drop-down list, and the click **Assign**.



27) Select cells D3 through E8 and enter a sample name in the **Sample Name** field. Click **Sample Name** to save sample name.

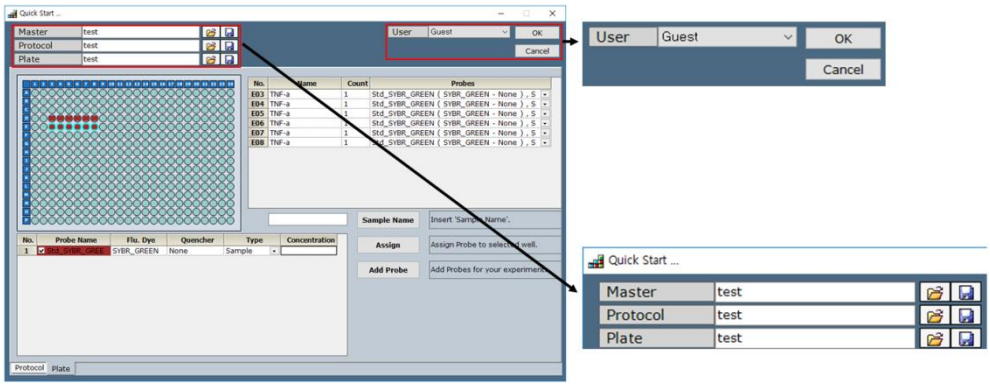


28) Verify information on each well in the **Name/Count/Probe** window.



29) Enter a plate name in the **Plate** field (i.e. Test) and then click button to save the plate file.

30) The protocol file and plate file are saved. Enter a master file name in the **Master** field (i.e. Test) and then click button to save the master file.



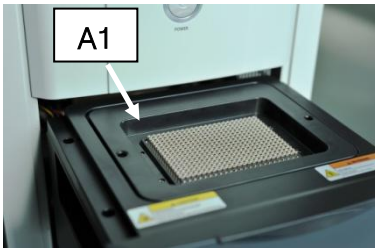
31) Click **OK** at the top right corner, Go back to the main window.

NOTE:
Save the master file, the protocol file, and the plate file by clicking button. You can open the saved master file by clicking button from Quick Start menu. The master file includes both protocol and plate file.

Power off

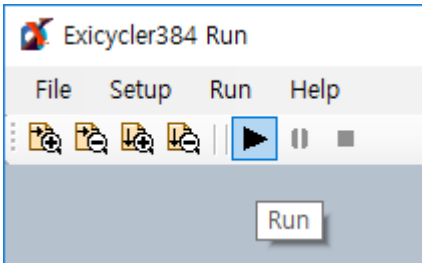
Standby

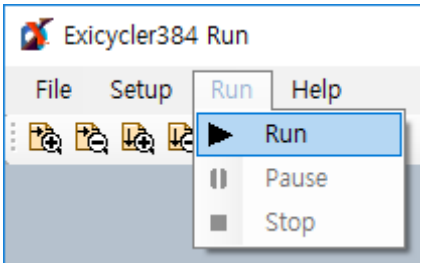
32) Ensure the status LED in front of the *Exicycler*™ 384 is blinking green.

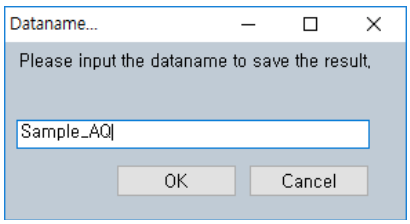


33) Press the **DOOR** button for a full second to open the door. Load the 384-well plate with A1 positioned at top-left corner of the block.

34) Verify the master file and go to **Run > Run** from the top menu bar or click the **Run** button.

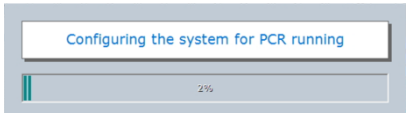




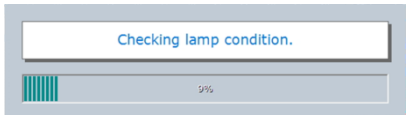


35) Enter a data name (i.e.Sample_AQ) in the **Dataname** window and click **Ok**. If you do not enter the data name, default name will be used.

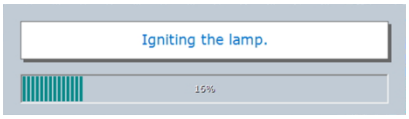
36) The following three message boxes will appear in the following order.



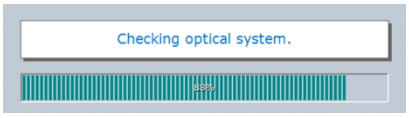
a. This message will appear when closing the door of the *Exicycler*™ 384.



b. This message will appear when the lamp condition is being checked.

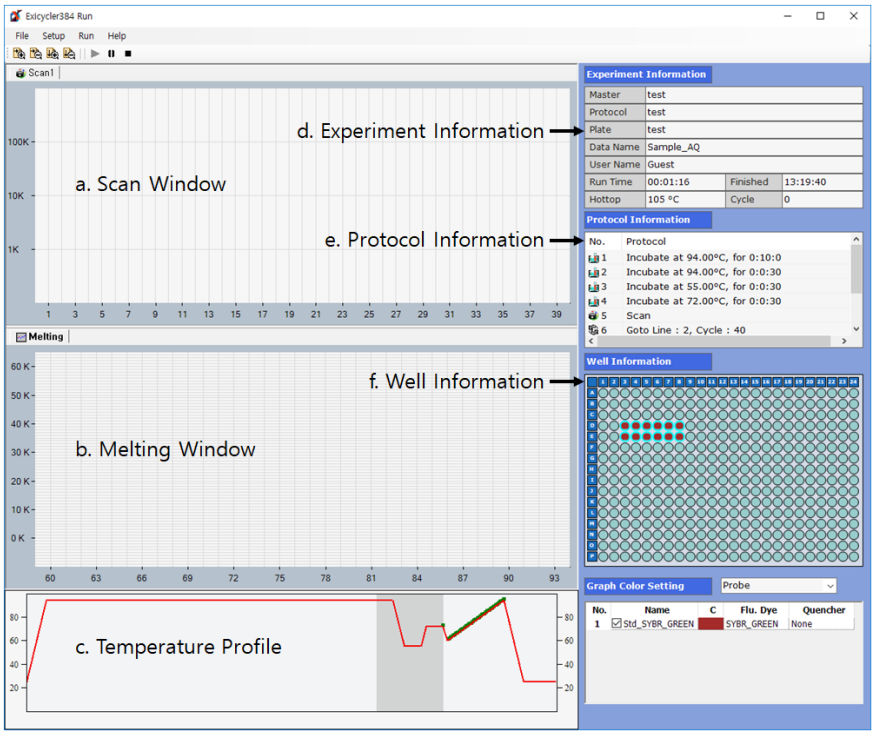


c. This message will appear during lamp ignition.

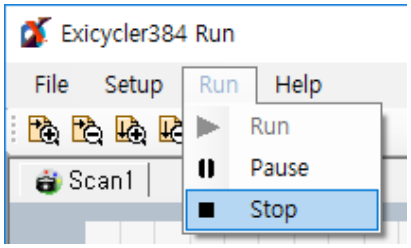


d. This message will appear when lamp is turned on and initialization of *Exicycler*™ 384.

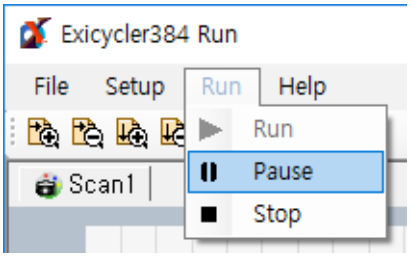
37) The main window will appear when the progress bar finishes without any problems.



a. **Scan** window displays a curve of amplification in real time during PCR.
b. **Melting** windows displays a melting curve in real time when the melting step is included in the protocol file.
c. **Temperature Profile** displays a temperature profile and in real time.
d. **Experiment Information** displays information for the current experiment.
e. **Protocol Information** indicates current progress of the experiment.
f. **Well Information** displays details of each well when selecting either **Probe** or **Type** in the **Graph Color Setting** drop-down list. To display the curve of amplification, select a well from 384-well plate diagram and click the check box from the probe option.



38) Go to **Run > Stop** to stop *Exicycler*™ 384 or click **Stop** button.



39) Go to **Run > Pause** to pause *Exicycler*™ 384 or click **Pause** button.

IMPORTANT:
If the pop up window remains more than 5 minutes after running *Exicycler*™ 384 or if the temperature profile does not display anything in the main window, turn off *Exicycler*™ 384 and turn it on again. Start **Run Exicycler384** again and run *Exicycler*™ 384 again. If the same error keeps occurring, please contact us for customer service.

IMPORTANT:
It is recommended to re-start *Exicycler*™ 384 at least 10 minutes after the previous Real-Time PCR run has finished. Continuous operation without a break will reduce the lamp life span and cause errors.

NOTE:
Do not turn off *Exicycler*™ 384 while the heating lid is warming up. While the lid is heated to the set temperature, 384-well block maintains at 25°C. The thermal cycling will start when the heating lid reaches set temperature and the lamp is stabilized.

NOTE:
*.ex8 file is created under a folder designated by the user after the experiment is complete. The user can analyze the *.ex8 file using *Exicycler*™ 384 Analysis Program to generate analysis data. Please refer to **Analyzing Data using Analysis Program** for data analysis.

Reagents and Consumable Products

Various reagents and kits are required to perform a Real-Time PCR in *Exicycler™* 384 Real-Time Quantitative Thermal Block. Please refer to this section to obtain detailed information about the appropriate reagents and kits for your experiments. For ordering, please go to the Ordering Information.

PCR Premix kit

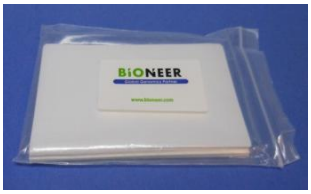
AccuPower® Greenstar™ qPCR Master Mix kit allows an easy and fast amplification in *Exicycler™* 384. *AccuPower® Greenstar™* qPCR Master Mix kit consists of *Greenstar®* fluorescent dye, HotStart Taq DNA polymerase, and necessary qPCR components. Therefore, only template, primers, and DW need to be added to start your amplification.

Optical 384-well plate

You must use a plate when performing Real-Time Quantitative PCR in *Exicycler™* 384. A standard plate can be also used, but only for a conventional PCR without fluorescent dyes.

Optical sealing tape

Seal up the optical plates tightly with the optical sealing tape for fluorescence detection. The sealing tape is available in the size of 384-well plate.



CAUTION

You must cut up the sealing tape with a box cutter, not scissors. The tape adhesive can be carried over to the surface of sealing tape when cutting up the tape with scissors. This will make the plate stick to the heating lid of *Exicycler™* 384. Plate stuck to the lid will drop inside *Exicylcer™* 384 when the lid is cooled down.

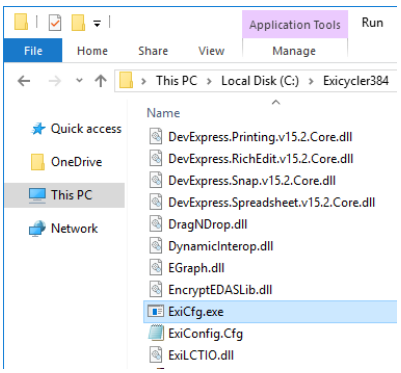
WARNING Chemical hazard:

AccuPower™ Greenstar™ qPCR Mater Mix may cause eye and skin irritation, and respiratory tract irritation. Read MSDS before use and follow the instructions if swallowed or inhaled. Wear appropriate protective eyewear, clothing, and gloves.

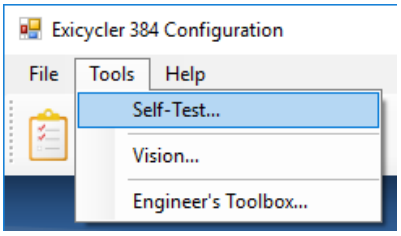
Performance Maintenance

Performing System Diagnosis using ExiCfg

1) Turn on *Exicycler*™ 384 by pressing the **POWER** button.

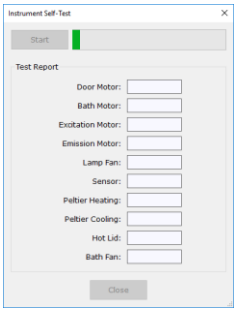


2) Go to 'C:₩Exicycler384₩Run' and double click **ExiCfg.exe** to start the program.



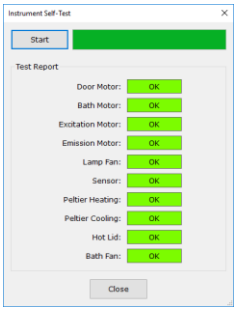
3) Go to **Tools > Self Test.**

NOTE:
'USB Communication is NOT initialized' message box will pop up if *Exicycler*™ 384 is turned off or if the cable connection between *Exicycler*™ 384 and the computer is not firmly connected.



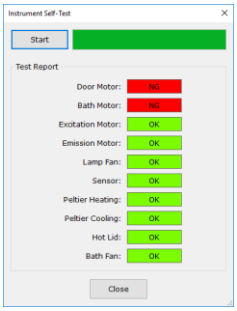
4) Click **Start** in the **Self Test** window.

IMPORTANT:
Make sure that there are no objects placed in front of the door while performing Self Test.



5) **OK** signs will show up in green, when all of the self-diagnosis tests pass completely without any problems.

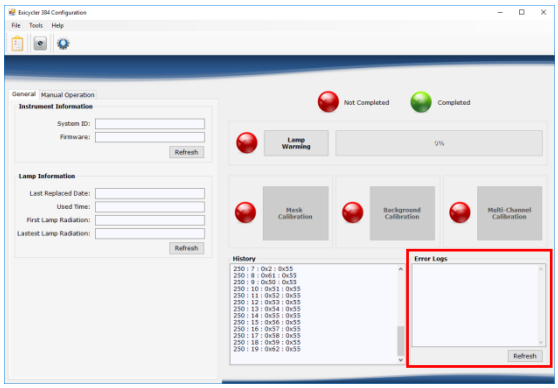
NOTE:
Self-diagnosis is complete with two short beeps.

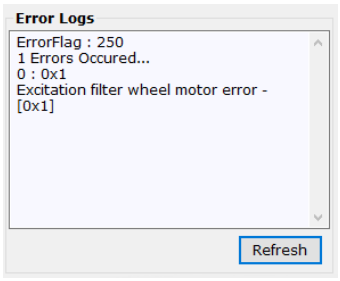


6) When any of the self-diagnosis tests fail, **NG** signs will appear in red.

NOTE:
Any problems caused during self-diagonsis are recorded in 'Error log'.

7) Check the **Error Log** screen at the bottom right of the program to see the problems. Click the 'refresh' button to display the error that occurred.





Guide to *AccuPower*® Fluorescence test kit for calibration

Prepare to perform the Calibration with a Fluorescence test kit. The Fluorescence test kit is designed for *Exicycler*™ 384 Real-Time Quantitative Thermal Block, Therefore, it is not applicable to other Real-Time instruments. The Fluorescence test kit is comprised of 11 plates, a mask plate, a background plate, multi-channel plates (9 different optical Fluorescence test plates of filter sets) present in *Exicycler*™ 384. The Fluorescence test kit is available for reuse up to 3 times. For more information about the Fluorescence test kit, please see the following:

Before Starting Calibration with the Fluorescence test kit

Centrifuge, Disposable plastic gloves, *AccuPower*® Fluorescence test kit for *Exicycler*™ 384

Fluorescence test plate preparation

The Fluorescence test kit is shipped refrigerated or frozen and must be stored in the freezer upon receipt. Retrieve one Fluorescence test kit right before starting the calibration.

1) Take out the Fluorescence test kit from the freezer.

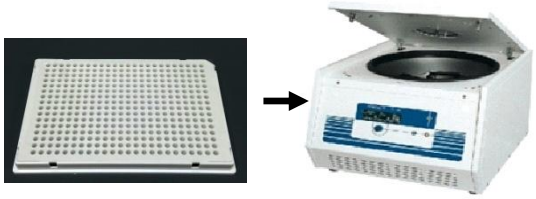
2) Take out a sealed packs in the Fluorescence test kit.

3) Allow the pack to thaw in room temperature for at least 10 minutes.

4) Take out a Fluorescence test plate from its packaging.

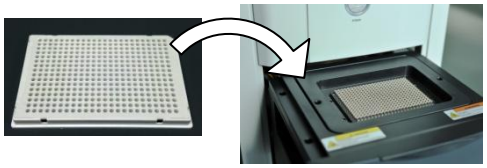
IMPORTANT:
Pay particular attention to the Fluorescence test kit when handling. Wear disposable plastic gloves to prevent contamination on the sealing tape covering 384-well plate. Wipe the surface of the plate with 70% ethanol when it needs to be cleaned.

NOTE:
Do not vortex the Fluorescence test plate. A small amount of Fluorescence test solution is pre-aliquoted in each well, and it may be lost while vortexing.



5) Place the Fluorescence test plate in a rack and centrifuge for 5 minutes.

NOTE:
It is important to centrifuge the Fluorescence test kit prior to use. This will spin down remains of fluorescence dye on the side of the well and to remove air bubbles at the bottom of the well.



6) Protect the Fluorescence test plate from direct sunlight after centrifugation. Place the plate directly into *Exicycler*™ 384.

7) Put the Fluorescence test kit back into its packaging and return it to the freezer when the calibration is done.

IMPORTANT:
The Fluorescence test kit contains photosensitive components. Keep the Fluorescence test kit away from light during calibration. Put the Fluorescence test plate back into its packaging right away for reuse.

Calibration using ExiCfg

The calibration must be performed prior to operating *Exicycler*™ 384 Real-Time Quantitative Thermal Block. You can either transfer proper calibration data associated with a serial number from the installation CD to the computer or perform the calibration by an authorized engineer of Bioneer. Install the Operation and Analysis Software for the calibration data transfer to the computer. For a reliable operation of the *Exicycler*™ 384, calibrating *Exicycler*™ 384 with a Fluorescence test kit is recommended. Calibration must be done when moving *Exicycler*™ 384 to a different location or when changing a light source lamp. A periodical calibration of every 6 months helps to maintain the optimum condition of *Exicycler*™ 384.

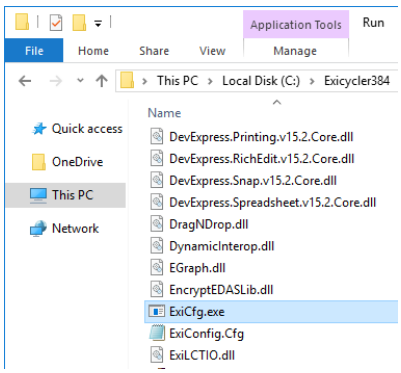
Use the ExiCfg program to calibrate *Exicycler*™ 384. The ExiCfg calibrates as well as diagnoses systemical condition of *Exicycler*™ 384. Make sure that you are fully trained for handling the ExiCfg before starting calibration. Otherwise, authorized staff is only allowed to manage the ExiCfg. Mis-use of the ExiCfg by unauthorized staff may cause serious damage to *Exicycler*™ 384.

The calibration is carried out in 4 steps, Warming up the lamp > Mask calibration > Background calibration > Multi-Channel calibration.

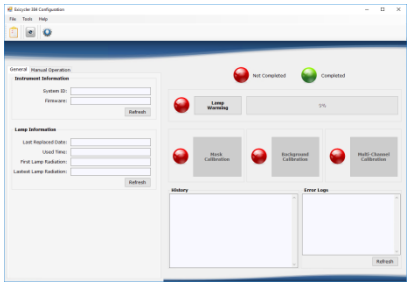
Mask Calibration

1) Turn on *Exicycler*™ 384 by pressing the **POWER** button.

NOTE:
Make sure the status LED is blinking in green after the self-diagnosis prior to starting the Fluorescence Check.



2) Go to '**C:₩Exicycler384₩Run**' and double click **ExiCfg.exe**.



3) Click **Lamp warming** to turn on the lamp. A progress bar of the **Lamp warming** will start right after 'The lamp is turned on' message appears on the 'History' box. Do not click any buttons until the progress bar finishes.

IMPORTANT:
It usually takes about 30 seconds for the message 'The lamp is turned on' to appear on the 'History' box. If the message does not appear in 5 minutes, turn off *Exicycler*™ 384 and turn it on again after 5 seconds. In case of an abnormal shutdown like this, wait about 5 minutes, then click **Lamp warming** to turn on the lamp again. It takes approximately 10 minutes to reach the maximum light intensity there after. Therefore, wait until the progress bar of the **Lamp warming** finishes before proceeding further.

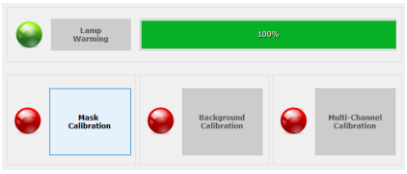
4) Press the **DOOR** button to open the door.

5) Place the Mask Fluorescence test plate on the block.

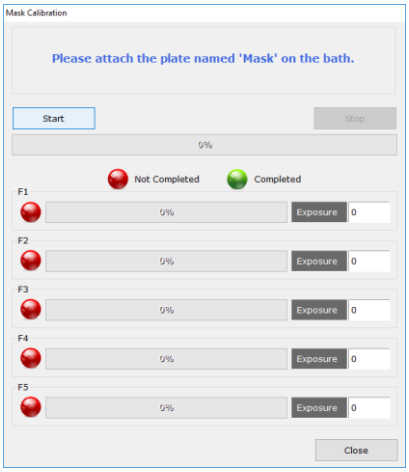
NOTE:
You must perform the Mask Calbration test prior to any Fluorescence tests.

IMPORTANT:
It is important to load the Fluorescence test plate in the right position. A misplaced plate may cause damage or technical problems.

6) Press the DOOR button again to close the door.

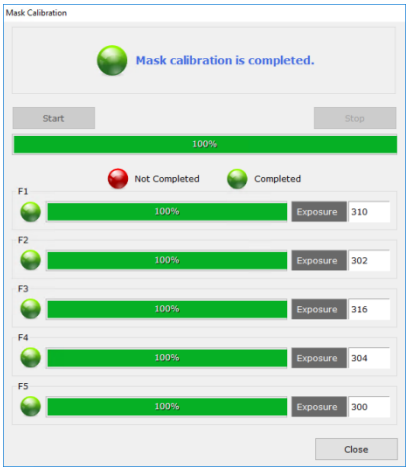


7) Click Mask Calibration Check button.



8) Click **Start** button in the 'Mask Calibration' window.

NOTE:
The Calibration time is approximately 10 minutes depending on the condition of *Exicycler*™ 384 and the computer.

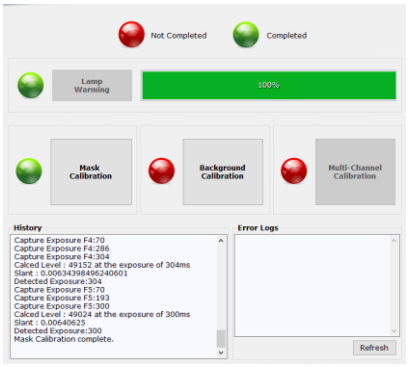


9) When the Mask Calibration is complete, click **Close**.

NOTE:
The door will open when the Mask Calibration is successfully completed. Put the plate back into its packaging and return it to the freezer.

WARNING High temperature

Wait at least 5 seconds before taking out the Fluorescence test plate from the 384-well block. The Fluorescence test plate is heated to a temperature of 100°C and may cause a burn.

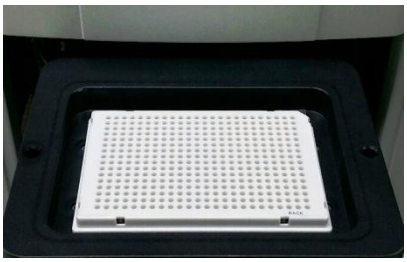


10) The 'Mask Calibration' part will turn green(completed) when the Mask Calibration is completed.

Background Calibration

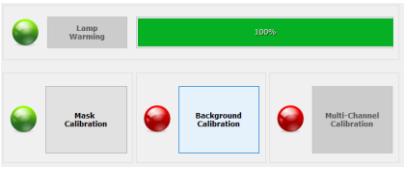
1) Press the **DOOR** button to open the door.

NOTE:
This step is omissible when performing the Background Calibration followed by the Mask Calibration.

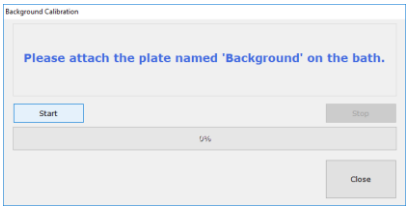


2) Place the Background Fluorescence test plate on the block.

IMPORTANT:
It is important to load the Fluorescence test plate in the right position. A misplaced plate may cause damage or technical problems.

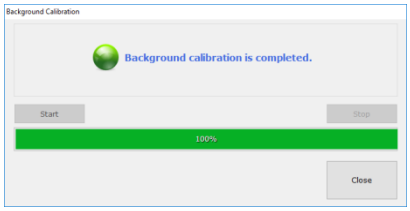


3) Click Background Calibration button.



4) Click **Start** in the 'Background Calibration' window.

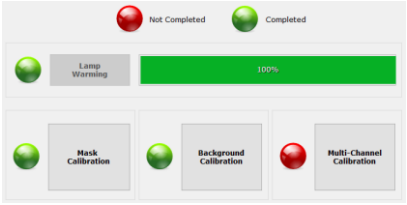
NOTE:
The Background Calibration takes approximately 10 minutes depending on the condition of *Exicycler*™ 384.



5) When the Background Calibration is complete, click Close.

NOTE:
The door will open when the Background Calibration is successfully complete. Put the plate back into its packaging and return it to the freezer.

WARNING High temperature
Wait at least 5 seconds before taking out the Fluorescence test plate from the 384-well block. The Fluorescence test plate is heated to a temperature of 100°C and may cause a burn.

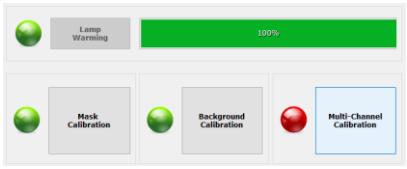


6) The 'Background Calibration' part will turn green(completed) when the Background calibration is completed.

Multi-channel Calibration

1) Press the **DOOR** button to open the door.

NOTE:
This step is omissible when performing the Background Calibration followed by the Mask Calibration.



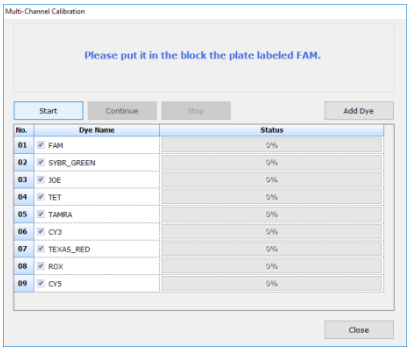
2) Click Multi-Channel Calibration button.

No.	Dye Name
01	FAM
02	SYBR_GREEN
03	JOE
04	TET
05	TAMRA
06	CY3
07	TEXAS_RED
08	ROX
09	CY5

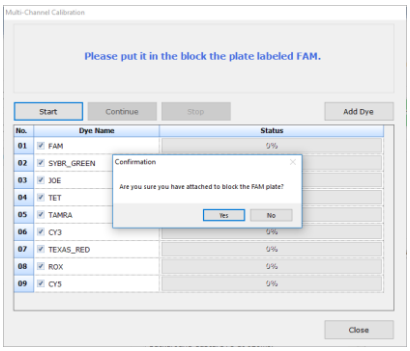
3) Select fluorescence dyes in the 'Multi-Channel Calibration' window for the Calibration.

NOTE:
9 dyes are available for calibration.

NOTE:
For a selective multi-channel calibration of your desirable dyes, select check boxes of the dyes.



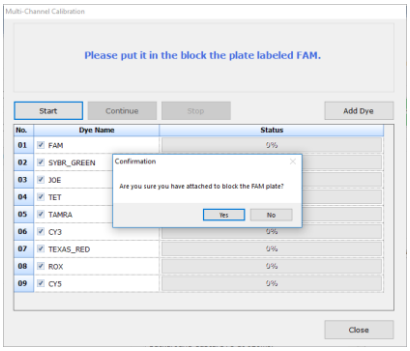
4) Click **Start.**



5) Prepare the Fluorescence Test plate with a dye **X** (i.e. "FAM") when 'Attach the plate named '**X**' (i.e. "FAM") on the bath' message appears.

6) Place the **Multi-Channel Fluorescence Test plate** on the block.

IMPORTANT:
It is important to load the Fluorescence Test plate in the right position. A misplaced plate may cause damage or technical problems.

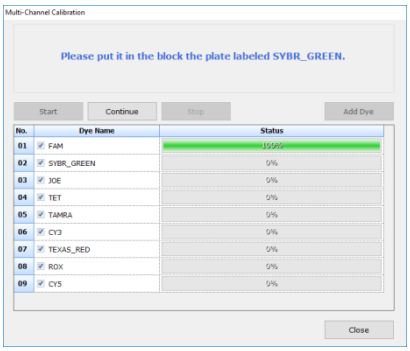


7) Click **Yes** in the 'Confirm' window.

8) The door will open when the Calibration is successfully complete.

NOTE:
An approximate calibration time for each dye is 5 minutes. Put the plate back into its packaging and return it to the freezer.

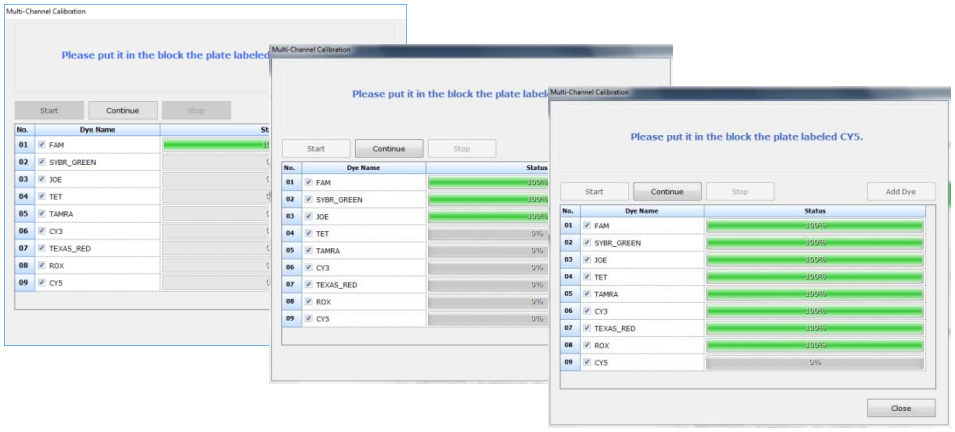
WARNING High temperature
Wait at least 5 seconds before taking out the Fluorescence test plate from the 384-well block. The Fluorescence test plate is heated to a temperature of 100°C and may cause a burn.

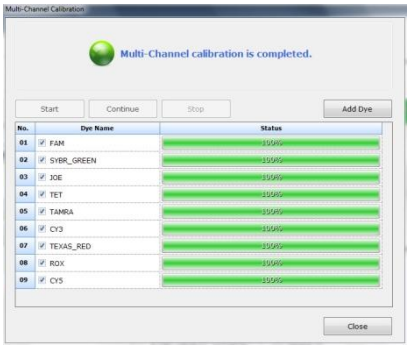


9) Prepare the Fluorescence Test plate with a dye **X** (i.e. "SYBR Green") when 'Attach the plate named '**X**' (i.e. "SYBR Green") on the bath' message appears.

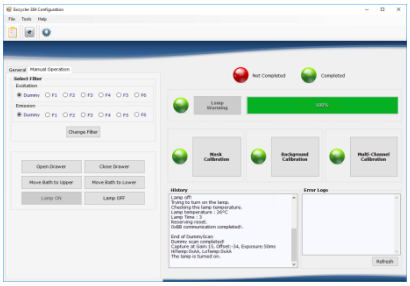
10) Place the **Multi-Channel Fluorescence Test plate** on the block.

11) Click **Continue** and repeat step 9 through 10.

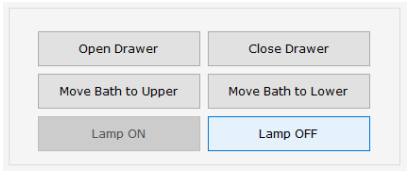




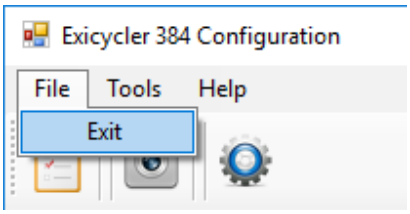
12) When the Multi-channel Calibration is complete, click **Close** in the '**Multi-Channel Calibration**' window.



13) The '**Multi-Channel Calibration**' part will turn green when the Calibration is completed.



14) Go to **Manual Operation>Lamp Off** on the bottom left of the main screen.



15) Go to **File>Exit** from the top menu bar to close ExiCfg program.

IMPORTANT:
Wait at least 5 minutes to switch off main power of *Exicycler*™ 384. The cooling fan will continue to run to cool down the lamp. Shutting down *Exicycler*™ 384 before cooling down the lamp will reduce the lamp life span.

16) Turn off *Exicycler*™ 384 by pressing the **POWER** button.

17) Turn off *Exicycler*™ 384 using the main power switch located at the back of *Exicycler*™ 384.

Cleaning the main body

Note:
- Turn off *Exicycler*™ 384 Real-time Quantitative Thermal Block, then remove the power cord.
- Allow the instrument to cool until the bath block reach room temperature.

Cleaning the Block Wells

If you use any cleaning or decontamination method, except those recommended in the manual, you risk damaging the Instrument. Clean the block wells once a week or as needed.

Always wear protective glasses and gloves when servicing the instrument. Also, make sure you disconnect the instrument from power cord before you begin any service procedure.

During instrument operation, the temperature of the bath block can be as high as 100°C. Before performing the procedure, keep hands away until the bath block reach room temperature.

To clean the block wells:

1) Open the Front door.

2) Remove the sample from the bath block and set it aside.

3) Use a cotton swab soaked in pure isopropyl alcohol or DNA Zap to clean the wells thoroughly. Make certain that the isopropyl alcohol or DNA Zap has evaporated completely before reloading a sample tray.

CHEMICAL HAZARD

Isopropyl alcohol is a flammable liquid and vapor. Exposure may cause eye, skin, and upper respiratory tract irritation. Prolonged or repeated contact may dry skin and cause irritation.
Exposure may cause central nervous system effects such as drowsiness, dizziness, and headache. Read the MSDS, and follow the handling instructions.
Remove any remaining isopropyl alcohol from the cover.

Note:
If the platen becomes contaminated with amplified DNA, raise the hot-top cover to a cleaning position, wipe the platen with a cloth or cotton swab soaked in bleach, then rinse with water.

1) Clean the heated platen once a month or as needed.
2) If the hot-top cover become contaminated with amplified DNA, raise the hot-top cover to a cleaning position. Wipe the cover with a cloth swab soaked in isopropyl alcohol or DNA zap, then wipe the cover with a damp cloth.

Replacing Fuses

The instrument has two 250V, F10AL and 5×20mm fuses.

Required Materials

- Two 250V, F10AL and 5×20mm fuses
- Small screw-driver(-)

Procedure

1) Turn off the instrument and disconnect the power cord from the back of *Exicycler*™ 384.

DANGER ELECTRICAL SHOCK HAZARD

Severe electrical shock, which could cause physical injury or death, can result from working on an instrument with high voltage power supply. To avoid electrical shock, disconnect the power supply to the instrument, unplug the power cord, and wait at least 1 minute before working on the instrument.

2) Insert a small screw-driver(-) into the slot in the upper portion of the power entry module, and open the fuse door.

3) Pull out the fuse compartment to inspect the two fuses in the fuse compartment.

4) Pull out the blown fuse(s) from the fuse compartment and replace with new 250V, F10AL, 5×20mm fuses.

5) Place the fuse compartment back into the power entry module and close the fuse door.

6) Press the fuse door until it locks in place.

7) Connect the instrument power cord.

Returning *Exicycler*™ 384 for Service

1) Decontaminate the instrument.

CAUTION

If the bath block became contaminated with radioactivity, use a commercially available decontaminant to remove the contamination. If the block is not decontaminated, the instrument cannot be returned to Bioneer for service.

2) Complete certification for instrument decontamination.

3) Fax or e-mail(exicycler-support@bioneer.com) the service request form to the customer center.

4) Pack the instrument in the provided packaging, without any accessories or power cords. Include service request form in the box.

Note:
Request for instruments repair without the service request form can lead to delayed in service.

5) Affix the provided postage to the box, then ship the instrument to the designated facility. The repair process requires 1 to 3 weeks.

Troubleshooting

Please take recommend action for each observation. Contact Bioneer Customer Service Center if problem continues after the recommended action.

Problems	Recommended Action
Exicycler™ 384 does not start.	1. Make sure the power cable is firmly connected to the wall circuit, and then switch on the power supply button at the rear of *Exicycler*™384. 2. Check a fuse box located at the rear of *Exicycler™* 384. Change the fuse if it has blown.
Errors occur either after or during self-diagnosis (Status LED in red).	1. The lamp may still be cooling down or the self-diagnosis is still running. If the status LED does not turn back to green in 5 minutes, turn off *Exicycler™* 384 and turn it on again. If the same error occurs, please contact Bioneer Customer Service Center for help. 2. Turn off *Exicycler™* 384 if unexpected errors occur. Turn on *Exicycler™* 384 again to cool down the lamp and wait for 3 minutes. Press the POWER button to start self-diagnosis. Please contact Bioneer Customer Service Center if the same error keeps occurring.
No communication detected between the computer and *Exicycler™* 384.	1. Make sure that the USB cable is firmly connected to the computer. 2. Ensure you use the USB cable provided with *Exicycler™* 384 to connect the computer and *Exicycler™* 384. 3. Check if *Exicycler™* 384 is in the "Standby" mode. The status LED must blink in green. Turn off *Exicycler™* 384 and back on again if the status LED still blinks in red.
The POWER button or the DOOR button does not work.	1. Make sure that the power cable is firmly connected to the wall circuit, and then switch on the power supply button at the rear of *Exicycler™* 384. 2. The self-diagnosis may be still running. Wait until the self-diagnosis is complete and the POWER button blinks in green.
Exicycler™ 384 has stopped running.	1. Check if the electricity is supplied properly. Operate an AVR or UPS if needed. 2. Disable Screen Saver and Monitor Power in the Control Panel. 3. Ensure you use the USB cable provided with *Exicycler™* 384 to connect the computer and the *Exicycler™* 384. 4. Check if the USB driver is installed properly. If not, reinstall it again. 5. Do not plug in any other USB cables in the computer while *Exicycler™* 384 is running to prevent friction between *Exicycler™* 384 and the computer.

Exicycler™ 384 does not start a thermal cycling.	1. *Exicycler™* 384 may still be in the "Standby" mode in order to warm up the lamp. Warming up the lamp takes about 10 minutes. The thermal cycling will start automatically after the lamp warm up has finished.
Intensity of fluorescence signal is low.	1. Either examine fluorescence signal of the probe used for Real-Time PCR or use a new kit. 2. The PCR product amplified may be too long or there may be non specific products. Perform gel electrophoresis to determine the presence of amplification product or adjust an annealing temperature or $Mg2^{+}$ concentration if needed. 3. Perform the calibrations again to adjust the light intensity of the lamp.
Excessively high intensity of fluorescence signal.	1. Adjust concentration of the probe used for Real-Time PCR. 2. Make sure you use an appropriate probe system. 3. Adjust and optimize the PCR sample conditions.
The software shuts down abnormally.	1. The USB driver is not been installed properly. Reinstall the USB driver.
PCR sample has evaporated.	1. Check if the heating lid works or if the sample tubes or plates are completely sealed with the sealing tape. 2. Ensure the heating lid option is set up correctly. The Hottop Check Box must be selected and correct temperature must be set up. 3. A small amount of evaporation is not critical. 4. It is recommended that you use Bioneer kits and reagents to prevent PCR sample evaporation.
No PCR products are amplified.	1. Perform gel electrophoresis to determine the presence of amplification products. Adjust an annealing temperature or $Mg2^{+}$ concentration if needed.
No fluorescent signal is detected although PCR product is amplified.	1. Either examine fluorescence signal of the probe used for Real-Time PCR or use a new intercalating dye. 2. Perform the calibrations again to adjust the light intensity of the lamp.
Pop-up message during calibration process.	1. **Background Calibration finished** will appear if the Background Calibration is successful. 2. **Can't Read Data from EEPROM** will appear if the computer cannot read data from *Exicycler™* 384. Check if the USB cable is firmly connected between the computer and *Exicycler™* 384. 3. **Can't close program during PCR** will appear if you attempt to close *Exicycler™* 384 software while PCR is still running. 4. **Do you want to stop PCR?** will appear if you click **Stop** button during the Multi-channel Calibration.

Pop-up message during calibration process. (continued)	5. **Did you add plate on the bath?** will appear to make sure that the calibration plate is loaded in the thermal block prior to PCR. 6. **Mask Calibration finished** will appear if the Mask Calibration is complete. 7. **Multi-Channel Calibration finished** will appear if the Multi-channel Calibration is successful. 8. **Need to set up Machine ID** will appear if an instrument ID has not been set up for *Exicycler™* 384. Assign the ID using *Exicycler™* 384 software. 9. **No matching calibration data with machine** will appear if the instrument ID does not match the ID from *Exicycler™* 384 software. Set up the instrument ID using *Exicycler™* 384 software. 10. **Turn off the lamp** will appear if you attempt to end ExiCfg while the lamp is still on. To close ExiCfg, turn off the lamp first and then close the ExiCfg. 11. **Please load Mask Calibration Information first** will appear if you attempt to perform the Uniform Calibration before the Mask Calibration. 12. **USB Communication is NOT Initialized** will appear if the computer and *Exicycler™* 384 are not communicating with each other. Check if the USB cable between the computer and *Exicycler™* 384 is firmly connected. 13. **You must insert New Dye Name** will appear if a new name is not assigned to a custom dye. 14. **You must select at least one dye** will appear if you start the Multi-channel Calibration without selecting dyes. You must select at least one fluorescent dye for the calibration.
Error message when running *Exicycler™* 384 software.	1. **Can't exit program during PCR** will appear if you click **Stop** button in *Exicycler™* 384 software while PCR is still running. 2. **Can't open data during PCR** will appear if you select File > Open Data from the top menu while PCR is still running. 3. **Can't Pause during Melting Protocol** will appear if you click **Pause** button while the melting step is still running. 4. **Can't read data from EEPROM** will appear if errors occur during PCR and the computer cannot read data from *Exicycler™* 384. 5. **Can't read Plate Information** will appear if you attempt to open an outdated plate file. 6. **Can't Stop during Melting Protocol** will display if you attempt to stop the *Exicycler™* 384 software while the melting step is running. 7. **Communication between machine and S/W was not initialized** will display if you click **Run** button when *Exicycler™* 384 is not ready. 8. **Do you want to stop PCR?** will display if you click **Stop** button while PCR is still running.

Error message when running *Exicycler™* 384 software (continued).	9. **Fail to load calibration information** will appear if the computer fails to read the calibration data from *Exicycler™* 384. Go to C:₩Exicycler384 to check if the calibration folder and file exist. 10. **Melting Protocol can't be in Cycle Region** will appear if the melting step is inserted within a thermal cycling. 11. **Need to replace the Lamp** will appear if the lamp intensity drops below 60% of the normal lamp intensity. Replace the lamp. 12. **No matching calibration data with Machine. Can't Run!** will appear if you click **Run** button when *Exicycler™* 384 ID and the calibration data ID do not match. 13. **No matching calibration data with machine** will appear if the *Exicycler™* 384 ID and the calibration data ID do not match. 14. **Please insert any protocol before inserting Goto Protocol** will appear if the Goto step is inserted in the first line of a protocol file. 15. **Please insert Data name** will appear if you have not entered a name for data prior to PCR running. 16. **Probe Information was changed, do you want to cancel this?** will appear if you click the **Cancel** button without saving the edited probe Information. 17. **Probe using same filter is assigned** will appear if you assign more than 2 probe options within the same wavelength range. 18. **Ramping rate option Must be inserted between Incubation steps** will appear if you insert a ramping rate option in a wrong position of the protocol file. 19. **Set Probe Name** will appear if you have not entered a new name for the custom probe in the Add Probe window. 20. **UserName was already in User List** will appear when entering an existing name for User set-up. 21. **You must select Dye and Quencher** will display if the dye and quencher set up has not been selected in the **Add Probe** window.

Ordering Information and Warranty

Ordering Information

Product	
A-2061	*Exicycler*™ 384 Real Time Quantitative Thermal Block
Plastic consumables	
3111-41	Adhesive Optical Sealing Film, 100 sheets
Premix & Reagent	
K-6253	*AccuPower*® 2X *Greenstar* qPCR Master Mix / 100Rxn, 50 µL reaction
K-6254	*AccuPower*® 2X *Greenstar* qPCR Master Mix / 200Rxn, 50 µL reaction
K-6603	*AccuPower*® *Plus DualStar*™ qPCR Master Mix(2X), 2.5ml, 100 rxn

Warranty

This instrument is warranted by Bioneer against manufacturing defect in materials and workmanship for a limited warranty period of one (1) year from the date you received your product. Bioneer will either (1) repair the product at no charge if a hardware defect is found or (2) exchange the product if the same hardware defect arises more than three times during the limited warranty period. Any other accessories other than the instrument itself are considered as consumables and warranted for three months. Spare parts for the instrument will be available for five years from the initial instrument release date. If a defect arises after the limited warranty period, shipping and handling charges may apply to any repairs or exchanges of the product undertaken by Bioneer.

Exclusions and limitations

This warranty does not apply: (a) to cosmetic damage, including but not limited to scratches, dents, and broken plastic on ports: (b) to damage caused by accident, abuse, misuse, flood, fire, earthquake or other external causes: (c) to a product or part that has been modified in any way without explicit written consents of Bioneer; or (d) to damage cause by any services performed by unauthorized engineers or service providers.

Obtaining Warranty Service

Please review this User Manual and access the online support referred to in the manual accompanying this product before requesting warranty service.

BIONEER Worldwide

Bioneer Corporation

Address 8-11 Munpyeongseo-ro, Daedeok-gu, Daejeon, 34302, Republic of Korea
Tel +82-42-930-8777 (Korea: 1588-9788)
Fax +82-42-930-8688
E-mail sales@bioneer.com
Web www.bioneer.com

Bioneer Inc.

Address 155 Filbert St. Suite 216 Oakland, CA 94607, USA
Tel +1-877-264-4300 (Toll-free)
Fax +1-510-865-0350
E-mail order.usa@bioneer.com
Web us.bioneer.com

Bioneer R&D Center

Address Korea Bio Park BLDG #B-702, 700 Daewangpangyo-ro, Bundang-gu, Seongnam-si Gyeonggi-do, 13488, Republic of Korea
Tel +82-31-628-0500
Fax +82-31-628-0555
E-mail sales@bioneer.co.kr
Web www.bioneer.co.kr