Fluorescent Quantitative Detection PCR system

User's Manual

Attention

Users are recommended to read the contents of this manual thoroughly before operating the Bioer Fluorescent Quantitative PCR Detection System. To carefully observe all special Warnings and Cautions outlined in this manual. This manual should be maintained properly in good condition for reference. **Caution:** Copyright reserved. The Bioer Co. reserves the right to modify this manual at any time without notice.

The manual contains copyright protected and patented material. Without prior written consent from Hangzhou Bioer Technology Co., Ltd., any part of the manual shall not be duplicated, reproduced or translated into any other language.

Thank you for your purchase of this product. Before initial use of this instrument, please read this manual thoroughly !

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Important Notes

1 Practice

| Note: | Very important information is contained within this manual and it should be carefully |
|----------|---|
| | read before first use of the instrument. Failure to operate instrument according to the |
| | instruction could result in damage or abnormal functioning of the instrument. |
| | |
| Warning: | The warning message requires extremely careful operation of a certain step. If the |
| 8 | instrument is not used in the manner prescribed by the manufacturer, the protection |
| | provided may be compromised. |

2 Safety

During operation, maintenance and repair of this instrument, the following basic safety notes must be observed. In case of failure to follow these measures or the warnings or notes indicated herein, the basic protection provided by the instrument, its safety criteria of design and manufacture, and its predicted use range would be impaired.

Hangzhou Bioer Technology Co., Ltd. shall not be held responsible for any consequences resulting from the user's failure to observe the following requirements.

| Note: | The instrument, complying with the Standard GB4793.1/IEC61010-1, is a general instrument of class I, the protection degree is IP20. It is intended for indoor use |
|-------|---|
| | |
| Note: | The instrument complying with the Standard YY0648/IEC61010-2-101 is used for IVD Medical Equipment. |

a) Instrument earth

In order to avoid an electric shock, the input power cable of the instrument must be properly earthed. This instrument uses a 10A 3-core earthed plug, which is provided with a third (earth) pin. It is for use with an earth type power socket and is a safety unit. If the plug cannot be inserted into the socket, the socket must be fixed by a qualified electrician, to maintain the safety function of the plug and the protection it provides.

b) Keeping apart from the live circuit

Operators are not allowed to disassemble instrument protection, replace components or make internal adjustment without authorization. If necessary, it must be completed by certified professional maintenance personnel. It is forbidden to replace components when power supply is connected.

c) Use of power supply

Before connecting to the mains and switching the instrument on, make sure the voltage is consistent with the instruments requirements (220V~,50Hz). The rated load for the power socket must not be less than the instruments maximum load of 1000VA

d) Power wire

The instrument is supplied with a power cable which should be used at all times when operating the instrument. If the power cable is damaged it should be replaced with a new one of the same specification. When using this instrument, do not press anything on the power cord and do not put the power cord in the traffic area. If the power cord comes in contact with the hot surface, add protection to prevent the insulation from being damaged.

e) Insertion and withdrawal of power cable

At insertion and withdrawal of power cable, the back of the plug shall be firmly held with the hand. The plug must be completely and tightly inserted into the socket and must not be removed by pulling the cable.

f) Placement of instrument

This instrument should not be positioned in a place where it is difficult to cut off the power supply. This instrument should be placed in a low relative humidity (RH) and low dust environment well away from any water (e.g. sinks and pipes). The room should be well ventilated, and free from corrosive gas, or interference from a strong magnetic field. The instrument should not be placed in a wet or dusty location, but should be positioned on a sturdy, level and secure table appropriate to its weight.

The openings on this instrument are for ventilation purposes and in order to avoid over-heating of the instrument they shall not be blocked or covered. When a single set or several sets of instruments are used, the space between its ventilation openings and the nearest object should not be less than 30cm. When multiple instruments are used at the same time, the distance between each instrument should not be less than 50cm.

Excessive environmental temperature would impair the test performance and could result in failure of the instrument. This instrument should not be used in locations subjected to direct sunlight or strong radiation or light source, as this could impair the fluorescence detection. The instrument should be kept away from hot gas, furnaces, stoves and all other sources of heat.

When switched off, the power should also be switched off. If the instrument is not going to be used for a long time, the power should be switched off, the power plug withdrawn and the instrument covered with soft cloth or plastic film to prevent dust or foreign bodies entering the machine.

g) Notes during operation

During test, cares shall be taken to prevent liquid from dropping onto the instrument. The castoff used in test, such as consumables, reagent, and so on, should be treated as require, and should not be thrown away or poured.

During test, if there is hazardous substances, user must be trained before using.

Hazardous substances, which has been used, should be coped with and saved according to derection for use.

User, who operates the instrument, must be trained and has relevant quantification.

Caution: If any of the following should occur, you should immediately switch off the power supply, withdraw the power plug from the power socket, and contact the supplier to effect a repair: Repairs can only be carried out by suitably qualified engineers.
Liquid gets inside the instrument.
The instrument is rained upon or water is spilled over it.
The instrument works abnormally, or generates an abnormal sound/s or generates a strange odour.
The instrument is dropped or its casing is damaged.
There is an obvious change in the function of the instrument.

Caution: When you deal with potential contagious matter such as body's tissue sample or reagent, which is likely to touch skin, protecting glove or other protecting measures are need to be used.

h) Re-transport

If the instrument needs to be transported again, the detection hole position and the instrument should be thoroughly cleaned and sterilized with ultraviolet light before transportation.

i) Warning Sign

• Warning identification

| DANGER! | Area with the mark pasted on the instrument shall avoid improper use and be careful of danger. |
|----------------------------------|---|
| SCALDING! | Area with the mark pasted on the instrument causes high temperature and is scalding during use. |
| BIOHAZARD | Area with the mark pasted on the instrument will caused biohazard during use. |
| PROTECT CONDUCTOR TERMINAL | PROTECT CONDUCTOR TERMINAL is near to the area with the mark pasted on the instrument |

• Warning mark



Warning! When "HOT SURFACE!" is pasted in the instrument, it means that the metal part (module) near this sign shall not be touched with any part of the body during the operation of the instrument or a period of time immediately after the operation of the program to avoid burns !

Warning! The operator may come into contact with or remain substances harmful to the organism or infectious substances during the use of the instrument. The operator should be aware of its hazards and strictly comply with the relevant provisions of the national PCR laboratory in accordance with the use environment of the instrument. Operators need to be trained and qualified.

| Up | <u><u><u></u></u></u> | Indicates that the correct position of the transport package is vertical upward. | | | | |
|-----------------------------|-----------------------|--|--|--|--|--|
| Fragile | Ţ | The transport packages contain fragile goods, so they should be handled with care. | | | | |
| Keep dry | Ť | The package should be rain-proof. | | | | |
| The limit of stacking layer | | Maximum stacking layer of the same package is 2. | | | | |
| Temperature limit | -370m | Indicates that the temperature limit of transportation package should be - 20 °C to 55 °C. | | | | |

j) Signs on the external packaging

3 Maintenance of Instrument

If there is any stain on the surface of the instrument, it can be cleaned with soft cloth and cleaning paste. Heat conducting oil medium is not allowed in the module hole of this instrument.

The drawer should be closed in time after the normal storage and use of the instrument to prevent dust accumulation.

| Warning! | When cleaning the instrument, the power should be turned off. |
|----------|--|
| 8 | The instrument surface should not be cleaned with corrosive cleaning agents. |
| | should be avoided. |

4 After-sales Services

The warranty content and scope are shown in the warranty sheet.

- **Note:** After unpacking, immediately check the goods against the packing list. If any parts are damaged or missing, please contact the supplier immediately.
 - After qualification of acceptance, complete the product acceptance sheet and send (or fax) the copied sheet to the supplier for filing and maintenance.
 - Before first use of the product, the user shall complete the instrument registration form and send to Hangzhou Bioer Technology Co., Ltd. for product registration.
 - After unpacking, the packing box and packing materials should all be kept in case it is required for transportation or service in the future.
 - In the event that a repair is required, the instrument must be disinfected before being sent to the repair department.
 - It is recommended that service personnel disinfect the instrument on receipt in the service department, before commencing any scheduled work.
 - Hangzhou Bioer Technology Co., Ltd. shall bear no liability in the event of any damage to the instrument occurring during transportation to the service department due to improper packaging.

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Chapter 1 General description

This chapter mainly describes the uses, characteristics, specifications, performance parameters and software functions of 96-well real-time quantitative PCR analyzer.

1. Scope of Application

The product is based on the principle of fluorescence quantitative polymerase chain reaction (PCR) and is used together with the supporting detection reagent. It is used for qualitative and quantitative detection of the analyzers in DNA/RNA samples from human nucleic acid samples, including pathogens and human genes.

2. Features

- New, user-friendly operation, operation interface, smooth operation
- Fluorescence real-time detection method is adopted to realize simultaneous amplification and detection in the same tube without post-processing
- Advanced thermoelectric refrigeration technology ensures super high speed heat cycle system heating, fast and stable refrigeration
- Multi-point temperature control ensures higher temperature uniformity of 96 sample wells
- 6 partition temperature control function
- Stable and accurate gradient functions of $1 \sim 36 \,^{\circ}\text{C}$ ensure optimized PCR conditions
- the thermostatic function of SOAK allows the PCR reagent to be stored at low temperature
- Long life LED excitation light source requires no maintenance
- Advanced fiber conduction technology makes photoelectric detection system more sensitive and reliable
- Real-time dynamic monitoring of the whole process of PCR amplification was carried out Real-time dynamic monitoring of the whole process of PCR amplification was carried out
- Wide linear range, initial DNA copy Numbers up to 10 orders of magnitude do not require gradient dilution
- There is no need to turn on the PCR reaction tube, which can avoid product contamination during and after PCR and ensure the accuracy of the results
- Multi-color fluorescence detection in a single reaction obtains more information
- The application of thermal cover technology has realized the oil-free operation of PCR
- Chinese language interface, flexible program setting, comprehensive analysis and reporting functions, all parameters can be stored
- Multiple or single sample reports can be printed
- The automatic, accurate and timely service of remote network provides the most advanced technical support for the 96-well quantitative PCR analyzer

3. Product Structure and Composition

This product is mainly composed of control parts, thermal cover parts, thermal cycle parts, photoelectric parts, transmission parts, power parts and software (V1).

4. Specification and Model Description

| The specification and wroach Description | |
|---|--|
| $\mathbf{Model}: \underline{FQD} - \underline{96} \mathbf{C}$ | |
| Design serial number | |
| Specification code, 96 means the maximum number of samples is 96 | |
| Fluorescence quantitative polymerase chain reaction (PCR) detection | |
| system | |
| Abbreviation of Fluorescence Quantitative Detection system | |
| Specification: Quant Gene 96 ×× | |
| Associated with the filter pack | |
| 40 (Equipped with F1, F2, F3, F4 filters) | |
| 50 (Equipped with F1,F2,F3,F4,F5 filters) | |
| 60 (Equipped with F1,F2,F3,F4,F5,F6 filters) | |
| Specification code, 96 means the maximum number of samples is 96 | |
| The English name of the series of real-time fluorescence quantitative | |
| PCR analyzer | |

5. Performance Parameters

| | Model | | FQD-%C | | | | | | |
|---|--|-------------------------|--|-----------------------|------------------------|-------------------------|----------------|------------|---|
| | Sample size | 96×0.2ml | 96×0.2ml (Suitable for single tube, 8 row tube and 96-well plate (no skirt board, half skirt board)) | | | | | oard, half | |
| | Detection channel | F1 | F2 | F3 | F4 | F5 | F6 | | |
| | Applicable dye | FAM, SYBR Green I | VIC, HEX, TET, JOE, | ROX, TEXAS -RED | Cy5 Quasar - 670 | Cy5.5 Quasar -705 | Optional | | |
| | Module operating temperature range | | 4°C~99.9°C(Minimum setting scale:0.1°C) | | | | | | |
| | Average heating rate | | When ris | ing from 50 |)℃ to 90℃, | it should b | e no less that | n 3.5°C/s | |
| | Average cooling rate | | From 90°C to 50°C, should not be less than 3.0°C/s | | | | | | |
| | Module temperature control accuracy | | Should be no greater than 0.1 °C | | | | | | |
| - | Temperature uniformity | | The temperature difference is within ± 0.3 °C | | | | | | |
| | Temperature control accuracy of hot cover | 105℃±5℃ | | | | | | | |
| | Fluorescence intensity test repeatability | CV≤3% | | | | | | | |
| | | | | | | | | | 2 |

| Mode of operation | Continuous operation | |
|---------------------|--|--|
| Operating system | Windows XP/Windows Vista/Windows 7/Windows 8 | |
| Input power | $100-240$ V \sim 50Hz 1000VA | |
| Overall dimensions | 490mm×290mm×391mm | |
| Weight | 28kg | |

6. Production Date and Service Life

Production date: see label for details.

Product life: 5 years

7. Function Overview of Supporting Software

- a) Parameter setting function (including temperature, time, cycle number, rise and drop rate, detection channel selection);
- b) Note function of text content;
- c) Sample data recording function (sample number, sample name, sample data);
- d) File operation display function (PCR thermal cycle data display, fluorescence detection data display, real-time display of various data during the operation of the instrument);
- e) Test data analysis function (analysis function can be used alone without instrument connection);
- f) Analysis results output function (one can output the analysis results to other types of files, such as :EXCEL, TXT files; be able to query and print the analysis results; one can change the print format and select the print item);
- g) File storage function (setting data, running data, analysis results);
- h) Fault protection and alarm function.

Caution: The above software functions are for reference only, without prior notice to the change of software functions.

8. Product Software Version

Release version of this product software: V 1

3

Chapter 2 Preparations

This chapter mainly introduces the use, transportation and storage conditions, structure composition, software installation/unloading, and preparation before starting up the Quant Gene 96xx series fluorescence quantitative PCR analyzer.

1. Transportation and Storage Conditions of the Instrument

Ambient temperature: −20°C ~55°C Relative humidity:≤80% Atmospheric pressure:75kPa~106kPa.

2. Normal Working Condition

Ambient temperature: $10^{\circ}C \sim 30^{\circ}C$ Relative humidity: $\leq 70\%$ Atmospheric pressure: 100-240V ~ 50 Hz 1000VA

Caution: Before using the instrument, please confirm whether the Working Conditions meet the above requirements. Note that the power socket is a 3-hole socket with reliable grounding.

3. Preparation before the Instrument is Switched on

3.1 Power Cord Connection

Power cord connection: the power cord attached to the instrument should be used. When connected, the instrument power switch should be in the closed state; After connecting, check whether the power cord and the instrument socket are too loose, if too loose, it should be replaced.

Caution: The attached power cord is reliable, but may cause the connection to be too loose after several unplugging. In this case, the power cord should be replaced. The power cord should be replaced with the same specification.

4. Installation of Supporting Software

4.1 Selection of a Computer System

System environment

Operating system: Windows XP/Windows Vista/Windows7/Windows8 Operating environment:.Net Framework 4.0 Other software: PDF reader **Minimum configuration:** Processor: Intel Core i3 Memory: 2GB Hard disk: 10GB

4.2 LineGene96xx Software Installation

Double click PcrServer installation file (PcrServerSetup.exe) ► Display the installation interface (select the installation language) ► Set installation path ► install

Double click LineGene96xx installation file (LineGene96xxDiagnosisSetup.exe) \blacktriangleright Display the installation interface (select the installation language) \blacktriangleright Set installation path \blacktriangleright install

4.3 LineGene96xx Software Uninstall

Control panel ► Add/remove programs ► PcrServer ► uninstall Control panel ► Add/remove programs ► LineGene96xx ► uninstall

Chapter 3 Start

1. Check before Starting

Before putting in the power plug and powering up the detection system, the following contents should be confirmed:

- Whether the power supply is consistent with the voltage required by the system;
- Make sure the power cord plug is correctly and reliably plugged into the power socket;
- Whether the surrounding working environment and equipment placement conditions meet the requirements.

2. Boot

Step 1: turn on the power switch of the backboard of the instrument;

Step 2: after entering the operating system, start the Quant Gene 96xx series real-time fluorescence quantitative PCR analyzer.

To start the software, click "LineGene 96xx" on the start/program menu or double click on the shortcut icon on the desktop.

3. Software Startup Interface

Double click any software shortcut icon of the LineGene 9600 series real-time quantitative PCR analyzer on the desktop, the corresponding startup screen will appear.



The system window consists of the menu bar, the toolbar and the main page.

Chapter 4 Absolute Quantification

1. Design Experiment



This section describes how to design a new absolute quantification experiment and covers inspection item setting, sample information setting, reaction plate setting and programme setting.

1.1 Create New Absolute Quantitative Experiment

1. Click build **Absolute** on the **Home** interface and this will open the absolute quantitative experiment window.

NOTE: The Absolute quantitative experiment can be also created by:

a. Clicking **File New Absolute** on the menu bar

b. Clicking New 🕨 Absolute on the toolbar



1.2 Detector Setting

1. Click Setup **Detector**



2. Input experiment properties

Input the experiment name, user name and any comments in the experiment properties column.

| Experiment Prop | | | | |
|------------------|---------------------|----------|--------|--|
| Experiment Name: | 20111117_Experiment |] | remark | |
| User Name: | user | Comment. | | |

3. Detector Setting

Set up the Detector, Assay, Dye and Colour.

If necessary, the user can also:

- a. Add detector
- b. Add assay
- c. Delete detector
- d. Delete assay

e. Add the detector in the Detector Library: click Add Detector From Library > the

Detector Library window will pop up **b** select the Detector in the window to be added

The user can also conduct Add, Modify and Delete operations in the item library.

| C Detector | Library | | | | | | | |
|------------|----------|--------|------------|--------|-------|----------|--------------|----------|
| Add | Modify | Delete | | | | | | |
| Detector | Reporter | Color | Master Mix | Primer | Probe | Supplies | Batch Number | |
| Target1 | FAM | | | | | | | |
| Target2 | FAM | | | | | | | |
| | | | | | | | Selec | ct Close |

f. Set up the detector, set up the assay, set up the dye name and set up the colour

| Detector | Reporter | Color | Master Mix | Primer | Probe | Supplies | Batch Number |
|----------|----------|-------|------------|--------|-------|----------|--------------|
| Target1 | FAM | - | | | | | |
| Target2 | FAM | | | | | | |

1.3 Sample Information Setting

1. Click Setup **>** Sample

VIC

-

| Setup | • |
|----------|---|
| Detector | 0 |
| Sample | 0 |
| Plate | 0 |
| Program | 0 |

2. Add sample information

a. Itemized addition: input ID in **Sample ID** > press **Enter** > add information for one sample

b. Batch addition: click **Batch Add** I the Batch Add window will pop up

| R Batch Add | |
|-------------------|------------------|
| Start Sample Id a | Sample Count 5 💌 |
| | Add Cancel |

3. Delete sample information

a. Itemized deletion: select one sample ► click **Delete** ► delete the selected sample information

b. Delete all: click **Clear All** ► delete all sample information

4. Import/Export sample information

a. Click **Import Sample Info** the File Import window will pop up import sample information file in CSV format

b. Click **Export Sample Info** ► the Save As window will pop up ► the sample information will be exported in CSV file format

5. Set up sample information

| Sample Id | Color | Sample Name | Sampling Time | Submitting Date | |
|-----------|-------|-------------|---------------|-----------------|--|
| a1 | | Sample1 | 2013-12-06 | 2013-12-06 | |
| a2 | | Sample2 | 2013-12-06 | 2013-12-06 | |
| a3 | | Sample3 | 2013-12-06 | 2013-12-06 | |
| a4 | | Sample4 | 2013-12-06 | 2013-12-06 | |
| a5 | | Sample5 | 2013-12-06 | 2013-12-06 | |

1.4 Reaction Plate Setting

| 1. | Click | Setup | | Plate |
|----|-------|-------|--|-------|
|----|-------|-------|--|-------|



- 2. Set up the inspection criteria of the reaction plate
- a. Select reaction plate well site: click Reaction Plate well Site

The user can also right click the reaction plate well site to Copy, Paste and Add New Detector. Adding a new detector will open the **Edit Detector Library** window.

| PUR Edit Det | ector Li | ibrary | | | | | |
|---------------|------------|------------|--------|-------|----------|--------------|--------|
| Detector Name | e: Target3 | | | | | | |
| Add | Delete | | | | | | |
| Reporter | Color | Master Mix | Primer | Probe | Supplies | Batch Number | |
| FAM | - | | | | | | |
| | | | | | | | |
| | | | | | | | |
| | | | | | | | |
| | | | | | | | |
| | | | | | | | |
| | | | | | | | |
| | | | | | | 1 1 2 | |
| | | | | | | ок | Cancel |
| | | | | | | 20240 N | |

b. Select Assay item and modify the property, concentration and concentration unit.

| Property | Name | Concentration | Concentration unit |
|----------|----------|---------------|--------------------|
| U | Unknown | NO | Copies/ml |
| S | Standard | YES | IU/ml |
| R | Negative | NO | Fg/ml |
| P | Positive | NO | Pg/ml |

c. Select a sample and the list displayed will change

d. Zoom-In, Zoom-Out and reset the reaction plate.

e. Sample Auto Arrange

f. Check Well Table



| - | 100 | Targett | Ulikilowi | 1 Pun | |
|----|-----|---------|-----------|-------|--|
| 2 | AU2 | | | | |
| 2 | AUS | | | | |
| 4 | A04 | | | | |
| 5 | A05 | | | | |
| 6 | A06 | | | | |
| 7 | A07 | | | | |
| 8 | A08 | | | | |
| 9 | A09 | | | | |
| 10 | A10 | | | | |
| 11 | A11 | | | | |
| 12 | A12 | | | | |

1.5 Programme Setting

1. Click Setup **>** Programme

| Setup | 0 |
|----------|---|
| Detector | 0 |
| Sample | 0 |
| Plate | 0 |
| Program | 0 |

2. Run Programme Setup

a. Create new stage: the user can create a new Hold Stage, Cycling Stage or Melting Stage

The user can also click **Add Stage** directly and the default will be creating a new **Cycling Stage**.

b. Create new step: the user can create a new step **Before** or **After** the currently selected step

The user can also click **Add Step** and the default will be adding a new Step at the end of the currently selected stage or after the currently selected step.

c. Delete: the user can delete the currently selected step or stage

d. Display form: click **Display With Table** ► new window will pop up ► the details of the current experiment will be displayed in a table.

e. Set up the experimental data of the hold stage, cycling stage and melting stage melting section

f. Set up the hot-lid temperature and liquid volume



2. Prepare for Reaction

| Start |
|---------------------|
| ▼ |
| Design experiment |
| ▼ |
| Prepare for the |
| reaction |
| \blacksquare |
| Run the experiment |
| \blacksquare |
| Experiment analysis |
| ▼ |
| Experiment report |
| \blacksquare |
| Data export |
| \blacksquare |
| End |

The user should make full preparations prior to the experiment:

- Ensure appropriate materials are used.
- Ensure the arrangement of the PCR reaction plate is consistent with the setting layout of the reaction plate in Section 1.4.

3. Run the Experiment Start Start Design experiment Prepare for the reaction Prepare for the reaction Run the experiment Experiment analysis Experiment report Experiment report Data export End

This section describes how to run/operate the experiment after loading the reaction plate and includes how to operate the fluorescence curve, the temperature curve and programming

Caution: Before starting the machine, please confirm that you have completed the inspection before starting the machine, and carry out the correct operation according to the starting steps. Turn on the system, and the system is in running state.

3.1 Preparation for reagent sample

- Prepare reagent: Quant Gene 9600 series real-time fluorescence quantitative PCR analyzer adopts 0.2ml centrifuge tube to place reagent samples, and 10 μ l \sim 50 μ l is recommended for the best reaction system for samples.
- The instrument allows the use of standard single tube, rack tube, skirt-free plate and other types of top optical transparent tube.
- Centrifugal operation: Before placing reactions into the instrument, it is recommended that a short centrifugal spin is used to ensure that the reagent is at the bottom of the reaction tube and the reagent/sample mix is free from bubbles.
- Placement of test tubes: if the number of samples is less than the number of holes in the module, try to distribute the sample tubes evenly in the holes of the module during the placement of test tubes, so as to ensure the smooth pressure of hot cover on the top of the tube during operation. Meanwhile, the load of the module is uniform, and the temperature change of each test tube is uniform.



Correct. The sample is at the bottom of the PCR tube



Incorrect

- 1. Requires a greater spin speed
 - Requires a longer spin time

3.2 Run Fluorescence Curve

1. Click Run **Fluorescence Curve**



2. Click Start Run



- 3. Operating confirmation
 - a. Modify hot-lid temperature and liquid quantity (sample volume).
- 4. After it starts operating, the user can: a. Skip the current stage

 - b. Add a cycle
 - c. Delete a cycle
 - d. Stop run

5. Plot display setting

- a. Assay item
- b. Plot colour



3.3 Run Temperature Curve 1. Click Run ► Temperature Curve



2. Click Start Run



- 3. Operating confirmation
 - a. Modify hot-lid temperature and liquid quantity (sample volume).
- 4. After it starts running, the user can:
 - a. Skip the current stage
 - b. Add a cycle
 - c. Delete a cycle
 - d. Stop run



3.4 Programme Setting

The user can only check the programme setting but cannot make modifications.

3.5 Prompts which may occur during running

- Hot-lid temperature sensor alarm prompt
- Sink temperature sensor alarm prompt
- Environmental temperature sensor alarm prompt
- Module temperature sensor alarm prompt
- Module sensor short-circuit or short-circuit alarm prompt

Caution: In case the temperature alarm displays during the running of a programme, the PCR detection system will terminate the current programme. The instrument should be switched off and then re-started.

4. Experiment Analysis

| Start | | | | |
|--------------------------|--|--|--|--|
| ▼ | | | | |
| Design experiment | | | | |
| ▼ | | | | |
| Prepare for the reaction | | | | |
| ▼ | | | | |
| Run the experiment | | | | |
| V | | | | |
| Experiment analysis | | | | |
| ▼ | | | | |
| Experiment report | | | | |
| ▼ | | | | |
| Data export | | | | |
| ▼ | | | | |
| End | | | | |

This section describes how to view the experiment analysis results after running an experiment and adjusting parameters for re-analysis.

This section covers the analysis of amplification curves and standard curves, adjusting parameters for re-analysis and importing parameters.

4.1 Check Results

4.1.1 Check the Amplification Plot 1. Click Analysis ► Amplification Plot

| Analysis | ۲ |
|-----------------------|---|
| Amplification Plot | 0 |
| Standard Curve | 0 |
| Melting Curve | 0 |

2. Check the amplification curve

- a. Set up colour
- b. Set up plot type
- c. Set up show dye

When the background colour of a dye name is blue, it will be displayed; while white indicates it will not be displayed.



3. Check the reaction plate

a. Select reaction plate well site and check corresponding well site curve The default is all wells are selected

- b. Zoom-In, Zoom-Out and reset the reaction plate
- c. Check well table
- d. Check results summary



- 4. Set up assay
 - a. Set up assay
 - b. Set up threshold
 - c. Set up automatic baseline
 - When the threshold value is not automatic, the user cannot set up the automatic

baseline Assay

Assay Targeti-FAM 🔽 Threshold 🛄 Auto 🛛 267 🚔 🔲 Auto Baseline

4.1.2 Check Standard Curve

1. Click Analysis Standard Curve



2. Check standard curve a. Set up assay



3. Check the reaction plate

a. Select reaction plate well site and check corresponding well site curve The default is all wells are selected

- b. Zoom-In, Zoom-Out and reset the reaction plate
- c. Check well table information
- d. Check results summary



4.1.3 Check Melting Curve 1. Click Analysis ► Melting Curve



2. Check the melting curve



b. Check the derivative curve

c. Set up colour



3. Check the reaction plate

a. Select reaction plate well site and check corresponding well site curve The default is all wells are selected

- b. Zoom-In, Zoom-Out and reset the reaction plate c. Check well table information



target 1

🕶 Color Well

4.2 Adjusting Parameters and Re-analysis

1. Click **Analysis Settings** > the Analysis Settings dialog box will pop up

- a. Adjust the start cycle and end cycle of the baseline b. Adjust Ct analysis algorithm
- c. Set up the use of S fitting
- d. Set up the stage to use for Ct analysis
- e. Set up the automatic threshold value
- f. Advanced setting
- g. Standard curve setting

| Analysis Set | tings | | | |
|-----------------|----------------|--------------|----------------|----------------------------------|
| Ct Settings Ad | dvanced Settin | gs Standard | Curve Settings | |
| he stage to use | for Ct analys | is: Stage 2 | | |
| he algorithm to | calculate Ct: | Baseline Thr | eshold 🔽 🔝 S F | litting |
| ssay Item | Threshold | Start Cycle | End Cycle | target1 - SYBR |
| arget1 - SYBR | Auto | Auto | Auto | Auto Threshold |
| | | | | Threshold: 293.41 |
| | | | | V Auto Baseline |
| | | | | Start Cycle: 3 🗣 End Cycle: 15 🗣 |
| | | | | |
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5. Experiment Report



This section describes how to print an experiment report and covers designing of a report template and print settings.

5.1 Designing a Report Template

1. Click **Report > Report Template Editor >** the Report Designer window will pop up

The report consists of controls and the user can add, modify and delete controls. Available controls include Static Text, Dynamic Text, Line, Static Image, Amplification curve and Quantification Analysis Results.



5.2 Print Setting

1. Click **Report > Print Template Setting >** the Print Template Setting window will open

The user can set up the laboratory name, report name, reference value, tester, checker, amplification plot, default report template and paper size.

| Template Setu | ip | |
|--|---|-----|
| Hospital | | |
| Report | | - i |
| Reference | 100 | _ |
| Tester | | |
| Chasher | | |
| Unecker | | |
| | | |
| | | |
| | | |
| | | |
| | | |
| Amplification | Plot Setup | |
| Amplification Legend: () Co | Plot Setup Door 🔘 LineStyle | |
| Amplification Legend: Co | Plot Setup Door 🔘 LineStyle | |
| Amplification Legend: () Co Print Setup | :Plot Setup olor ◯ LineStyle | _ |
| Amplification Legend: © Co Print Setup - Default Repor | : Flot Setup lor 🔘 LineStyle rt Template [default | |
| Amplification Legend: O Co Print Setup Default Repor Paper Size A | r Flot Setup lor C LineStyle rt Template default 4 | |
| Amplification Legend: () Co Print Setup - Default Repor Paper Size (A Printer | t Plot Setup lor DineStyle rt Template default 4 | |
| Amplification Legend: Co Print Setup - Default Repoi Paper Size A Printer © Use Defau | Plot Setup olor DineStyle rt Template default 4 v ult Printer | |
| Amplification Legend: © Co Print Setup - Default Repor Paper Size A Printer © Use Defau © Use Custo | Plot Setup olor LineStyle rt Template default 4 ult Printer xm Printer | |
| Amplification Legend: © Co Print Setup - Default Repor Paper Size A Printer © Use Defau Use Custo | Plot Setup lor C LineStyle rt Template default 4 ult Printer m Frinter | |

5.3 Comprehensive Report

1. Click **Report** ► **Consolidated Reports** ► the Consolidated Report window will pop up The Consolidated Report includes the basic information, sample information, amplification curve, standard curve, plate information, etc.

| | | | | | | | | | | Basic Informatio Run Program |
|------------|-----------------|---------|-------------|------------|------------|------------|------------|----------------|----------|---------------------------------|
| | | | | | | | | | 1 / 0 | / Detectors |
| | | | LineGe | ne9600 C | onsolida | ated Rep | ort | | 1/0 | Plot Plate |
| Evnerim | ant Name: 2011 | 1104 1 | F1-600183 | | | | | | | Table Plate |
| Experim | ent Type: Abso | lute | | | | | | | | Amp. Curve(Linear |
| File Nam | me: F:\L | ineGen | e9600\201 | 11104_1F1- | 600183.fqd | | | | | 🖌 Quan. Analysis Re |
| Run Tim | e: 2011 | /11/04 | 15:22:05 | - 2011/11/ | 04 16:22:1 | 8 | | | | 🚺 Standard Curve |
| Gain. | 11.1 | 5, 12.0 | , 10.2, 14. | 4,10.1 | | Ζ. | | | | |
| R | un Program — | | | | | | | | | Create Report |
| Hold Stag | e | | | | | | | | | |
| Target | Incubation Time | Rate | Sampling | | | | | | | |
| 94 | 120 | 4 | | | | | | | | |
| PCR Stage | Cycles:40 | | | | | | | | | |
| Target | Incubation Time | Rate | 2nd Temp. | Step Size | Step Delay | Grad Temp. | Grad Range | Sampling | | |
| 95 | 5 | 4 | | | | | | | | |
| 60 | 30 | 4 | | | | | | V | | |
| Detector | etectors | As | say | | Dye | | Color | r | | |
| Target 1 | | | | | FAM | | | | | |
| | | | | | HEX | | | | | |
| | | | | | ROX | | | | | |
| | | | | | Cy5 | | - | | | |
| P | lot Plate | | | | | | | | | |
| 1 | 1 | | 4 | 8 6 | 7 | 8 | 9 1/ | 0 11 | 12 | |
| A U Tarr | cet 🚺 1 | arget | | Target | U Targe | | J Target | U Target | 11000 | |
| | THE R. LEWIS | | | 100 - | | | | | | |
| | - target | | arget | | 224 | target | | | target | |
| C U Targ | set 🚺 1 | arget | U | Target | U Targe | t [| Target | U Target | | |
| | U Target | 1 | Target | U Ta | rget | U Target | U T | arget | U Target | |
| : U Targ | get 🛄 1 | arget | U | Target | U Targe | t | Target | U Target | | |
| | | | | | | | | | | |
| 10000 0000 | et d larget | | a larget S | larget | 1arge | t Marget | S | arget 🔯 larget | | |
| | | | | | | | | | 1000 | |
| | | | | | | | | | | |
| C B | | | | | | | | | | |
| C R | | | | | | | | | | |

5.4 Report Printing 1. Click Report ► Report Print 2. Report print setting a. Set up report template b. Print setting (please refer to Section 5.2) c. Select items to print d. Print preview e. Print the report

Bed No

Outpatient No.

5.5 QC Summary

1. Click Report **C** Summary

mple Id Sample Name | Test Item | Name | Sex | Age | Case No

Assay Item:SYBR Detect Concentration:7.82e+06 Conclusion:Positive



2. Check the QC summary

| Amp | olificatio | on Plo | ì | | | | | | | | | | QC Summary | | | _ | | |
|------|------------|--------|-----|--------|------|-----------------|-------------|------|-------|------|-----|---|---------------------------------|------------------|-------|-----|--------|---|
| | Well | | P | otType | Line | ar | | Show | FI F | 2 F3 | F4 | [| Description | | Value | Use | Result | |
| | 3 | | | | | | | | | | | 2 | Vegative contro: than | l with a Ct less | 38 | 1 | | |
| 5 | 5000- | | | | | | | | 1 | A | | H | Positive contro greater than | l with a Ct | 30 | ~ | | |
| e 4 | 1000- | | | | | | | -/ | | F | 1 | l | Jnknown without | a Ct | N/A | 1 | | |
| cenc | 000 | | _ | _ | | | | | | | | 5 | Standard withou | t a Ct | N/A | | | |
| 1 | .000 | 0 | 4 | 18 | 12 1 | 1 16 2 Cy | 0 24 cle | 1 28 |) | 1 36 | 40 | | | | | | | |
| A01 | A02 | A03 | A04 | A05 | A06 | A07 | A08 | A09 | A10 | A11 | A12 | | | | | | | |
| BUI | B02 | B03 | B04 | B05 | 506 | B07 | 508 | B09 | 510 | BII | B12 | | | | | | | |
| C01 | 002 | 003 | C04 | C05 | 006 | D07 | 008 | 009 | D10 | DII | D12 | | | | | | | |
| F01 | E02 | E03 | E04 | E05 | FOR | E07 | E08 | E09 | E10 | F11 | E12 | | | | | | | |
| F01 | E02 | E03 | E04 | E05 | E06 | E07 | E08 | E09 | E10 | F11 | F12 | | | | | | | |
| G01 | G02 | G03 | G04 | G05 | G06 | G07 | G08 | G09 | G10 | G11 | G12 | | | | | | | |
| H01 | H02 | H03 | H04 | H05 | H06 | H07 | H08 | H09 | H10 | H11 | H12 | | | | | | | - |

6. Data Export



This section describes how to export data and covers exporting to a database, experiment filing and exporting the experiment data to EXCEL.

6.1 Export to Database

Click **Data Summary** ► **Export to Database** ► the Save File dialog box will pop up ► save the exported database file

6.2 Experiment Filing

1. Click **Data Summary** ► **Archived Experiment Directory** ► the Experimental archive storage directory window will pop up ► set up the storage path of file.

| 🐮 Experimenta | al archive storage directory 🛛 🛛 🔀 |
|------------------|------------------------------------|
| Experiments in t | he following archive directory: |
| D:\LineGene9600 | |
| Change | |
| | OK Cancel |
| 4 | |

2. Experiment filing

Click **Data Summary** Archived Experiment export the filed experiment file The suffix of the filed experiment file is .fqh

6.3 Export Experiment Data to EXCEL

Click **Data Summary** ► **Export Experiment** ► **Export Experiment to Excle** ► the exported experiment data will generate EXCEL file

6.4 Export Experiment Data to TEXT

Click **Data Summary ► Export Experiment ► Export Experiment to Text ►** the exported experiment data will generate TEXT file

Chapter 5 Relative Quantitative 1. Design Experiment



This section describes how to design a relative quantitative experiment and covers creating new relative quantitative experiment, inspection item setting, sample information setting, reaction plate setting and programme setting.

1.1 Create New Relative Quantitative Experiment

1. Click **Relative** on **Home** interface and create Relative Quantitative Experiment window. Relative quantitative experiment can be also created by:

- a. Clicking **New > Relative** on the toolbar
- b. Clicking **File New Relative** on the menu bar



2. Input Experiment Properties

Input the Experiment name, User name and Comment in the basic information column.

3. Inspection Item Setting

a. Set up the Detector, Assay, Dye and Colour.

- b. Add detector
- c. Delete detector
- d. Add detector from library

The user can also conduct Add, Modify and Delete operations in the item library.

| Detecto | r Library | | | | | | | |
|----------|-----------|--------|------------|--------|-------|----------|--------------|---------|
| Add | Modify | Delete | | | | | | |
| Detector | Reporter | Color | Master Mix | Primer | Probe | Supplies | Batch Number | |
| arget1 | FAM | | | | | | | |
| arget2 | FAM | | | | | | | |
| | | | | | | | Sele | st Cose |

| Add Detector | Delete Deter | ctor Add Detector | From Library | | | | |
|--------------|--|--|---|--|--|--|--|
| Reporter | Color | Endogenous Contro | Master Mix | Primer | Probe | Supplies | Batch Num |
| FAM | - | | | | | | |
| HEX | | | | | | | |
| | Add Detector Reporter FAM HEX | Add Detector Delete Dete Reporter Color FAM T HEX T | Add Detector Delete Detector Add Detector Reporter Color Endogenous Contro FAM HEX | Add Detector Delete Detector Add Detector From Library Reporter Color Endogenous Contro Master Mix FAM Image: Color Image: Color Image: Color HEX Image: Color Image: Color Image: Color | Add Detector Delete Detector Add Detector From Library Reporter Color Endogenous Contrc Master Mix Primer FAM Image: Color Image: Color Image: Color Image: Color Image: Color HEX Image: Color Image: Color Image: Color Image: Color Image: Color Image: Color | Add Detector Delete Detector Add Detector From Library Reporter Color Endogenous Contrc Master Mix Primer Probe FAM Image: Color Image: Colo | Add Detector Delete Detector Add Detector From Library Reporter Color Endogenous Contrc Master Mix Primer Probe Supplies FAM Image: Color Image: Color Image: Color Image: Color Image: Color Supplies HEX Image: Color Image: Color Image: Color Image: Color Image: Color Image: Color |

4. Set up reference dye

Reference Dye
VIC

1.3 Sample Information Setting

1. Click Setup 🕨 Sample



2. Add sample information

a. Itemized addition: input ID in **Sample ID** > press **Enter** > add information for one sample.

b. Batch addition: click **Batch Add** > the Batch Add window will pop up



3. Delete sample information

a. Itemized deletion: select one sample click **Delete** delete the selected sample information

- b. Delete all: click Clear All > delete all sample information
- 4. Import/Export sample information

a. Click **Import Sample Info** ► the File Import window will pop up ► import sample information file in CSV format

b. Click **Export Sample Info** ► the Save As window will pop up ► the sample information will be exported in CSV file format

| Sample ID | Batch Add | Delete | Clear All | Import Samples Info | Export Samples Info |
|-----------|-----------|--------|-----------|---------------------|---------------------|
|-----------|-----------|--------|-----------|---------------------|---------------------|

5. Set up sample information

| Comolo Id | Color | Cample Name | Compling Time | Cubmitting Date |
|-----------|-------|-------------|---------------|-----------------|
| Sample Iu | COIOI | Sample Name | Sampling time | Submitting Date |
| al | | | 2013-12-06 | 2013-12-06 |
| a2 | | | 2013-12-06 | 2013-12-06 |
| a3 | | | 2013-12-06 | 2013-12-06 |
| a4 | | | 2013-12-06 | 2013-12-06 |
| a5 | | | 2013-12-06 | 2013-12-06 |

1.4 Reaction Plate Setting

1. Click Setup **>** Plate

| Setup | • |
|----------|---|
| Detector | 0 |
| Sample | 0 |
| Plate | 0 |
| Program | 0 |

2. Set up the inspection criteria of the reaction plate

a. Select reaction plate well site: click Reaction Plate well Site

The user can also right click the reaction plate well site to Copy, Paste and Add New Detector. Adding a new detector will open the **Edit Detector Library** window.

| | 1.020.200 | 1222 N 2 1 1 1 2 2 1 1 | 1.2.2 | | 1002000000000 | | |
|----------|-----------|------------------------|--------|-------|---------------|--------------|--|
| leporter | Color | Master Mix | Primer | Probe | Supplies | Batch Number | |
| AM | | | | | | | |
| | | | | | | | |
| | | | | | | | |
| | | | | | | | |
| | | | | | | | |
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| | | | | | | | |
| | | | | | | | |
| | | | | | | | |
| | | | | | | | |
| | | | | | | | |

b. Select inspection item and modify the property, concentration and concentration unit.

| Property | Name | Concentration | Concentration unit |
|----------|----------|---------------|--------------------|
| U | Unknown | NO | Copies/ml |
| S | Standard | YES | IU/ml |
| | Negative | NO | Fg/m |
| | | | Pg/ml |

c. Select a sample and the list displayed will change

- d. Zoom-In, Zoom-Out and reset the reaction plate.
- e. Sample Auto Arrange
- f. Check Well Table

| Dete | ctors | | _ | _ | | Pla | | p We | ll Table | - | | | | | | | | |
|--------------------------|---------------|-------|------------|----------|--------------|-----|-------|------|----------|---------|----------|-------|---|----------|-----------|----|----|----|
| Assay 1 | [tem | Prop | erty Co | n. | | | | | | Zoom In | Zoom Out | Reset | | Sample A | uto Arran | ge | | |
| Target | I - FAM(GOI) | U | | | | | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 |
| Target: | 1 - HEX (HKG) | U | - | | 1 | | | | | | | | | | | | | |
| | | | | | | A | Targe | | | | | | | | | | | |
| | | | | | | | | | | | | | | | | | | |
| | | | | | | | | 2 | | | | | | | | | | |
| | | | | | | в | | | | | | | | | | | | |
| | | | | | | | | | | | | | | | | | | |
| Conc | entration U | nit e | copies/ml | | | | | | | | | | | | | | | |
| Samp | les Sh | ow Co | lumns: Sam | mple Nam | - | | | | | | | | | | | | | |
| S | ample ID | S | ample Name | • | | Ľ | | | | | | | | | | | | |
| a | 1 | S | ample 1 | | \mathbf{V} | | | | | | | | | | | | | |
| a | 2 | S | ample 2 | | | | | | | | | | | | | | | |
| a | 3 | S | ample 3 | | | D | | | | | | | | | | | | |
| a | 1 | S | ample 4 | | | | | | | | | | | | | | | |
| a | 5 | S | ample 5 | | | | | | | | | | | | | | | |
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| | | | | | | | | | | | | | | 1.1 | | | | |

| Plate | Setup | Well Ta | ble | | | |
|-------|-------|-----------|------------|----------|-----|------|
| # | Well | Sample Id | Assay Item | Property | Dye | Con. |
| 1 | A01 | | Target1 | Unknown | FAM | |
| 1 | A01 | | Target2 | Unknown | HEX | |
| 2 | A02 | | | | | |
| 3 | A03 | | | | | |
| 4 | A04 | | | | | |
| 5 | A05 | | | | | |
| 6 | A06 | | | | | |
| 7 | A07 | | | | | |
| 8 | A08 | | | | | |
| 9 | A09 | | | | | |
| 10 | A10 | | | | | |
| 11 | A11 | | | | | |
| 12 | A12 | | | | | |

1.5 Programme Setting

1. Click Setup **Programme**



2. Run Programme Setup

a. Create new stage: the user can create a new Hold Stage, Cycling Stage or Melting Stage

The user can also click **Add Stage** directly and the default will be creating a new **Cycling Stage**.

b. Create new step: the user can create a new step **Before** or **After** the currently selected step

The user can also click **Add Step** and the default will be adding a new Step at the end of the currently selected stage or after the currently selected step.

c. Delete: the user can delete the currently selected step or stage

d. Display form: click **Display With Table** ► new window will pop up ► the details of the current experiment will be displayed in a table.

e. Set up the experimental data of the hold stage, cycling stage and melting stage melting section

| Run Programs Setup Add Stage Add Step | Delete | | | D | isplay With Tab |
|--|---------------------------------------|-----------------------------------|--|---------------------------------|-----------------|
| Hold Stage | | PCR S | tage | | |
| | | Cycles | 40 | | |
| | 2nd Temp. Step Delay Grad Temp. | - Step Size - - -Grad Range | 2nd Temp St Step Delay - Grad Temp Gra | ep Size – d Range | |
| - 300 500 - 500 - | 95 °C 4 °C | 95°C 00:00:15 v Sampling | 4 °C/s | 60 ℃ 0:00:20 🔷 ¥ Sampling | |

f. Set up the hot-lid temperature and liquid volume

2. Prepare for Reaction

| Start |
|---------------------|
| ▼ |
| Design experiment |
| ▼ |
| Prepare for the |
| reaction |
| ▼ |
| Run the experiment |
| ▼ |
| Experiment analysis |
| ▼ |
| Experiment report |
| ▼ |
| Data export |
| V |
| End |

The user should make full preparations prior to the experiment

- Ensure appropriate materials are used.
- Ensure the arrangement of the PCR reaction plate is consistent with the setting layout of the reaction plate in Section 1.4.

3. Run the Experiment



This section describes how to run/operate the experiment after loading the reaction plate and includes how to operate the fluorescence curve, the temperature curve and programming

3.1 Run Fluorescence Curve 1. Click Run ► Fluorescence Curve



2. Click Start Run



- 3. Operating confirmation
 - a. Modify hot-lid temperature and liquid quantity (sample volume)
- 4. After it starts running, the user can:
 - a. Skip the current stage
 - b. Add a cycle
 - c. Delete a cycle
 - d. Stop run
- 5. Plot display setting
 - a. Assay item
 - b. Plot colour



3.2 Run Temperature Curve

1. Click Run 🕨 Temperature Curve



2. Click Run 🕨 Start



- 3. Operating confirmation
 - a. Modify hot-lid temperature and liquid quantity (sample volume)
- 4. After it starts running, the user can:
 - a. Skip the current stage
 - b. Add a cycle
 - c. Delete a cycle
 - d. Stop run



3.3 Programme Setting

The user can only check the programme setting but cannot make modifications.

4. Experiment Analysis



This section describes how to view the experiment analysis results after running an experiment and adjusting parameters for re-analysis.

This section covers the analysis of amplification curves and standard curves, the analysis of relative quantification, adjusting parameters for re-analysis and importing parameters.

4.1 Check Results

4.1.1 Check the Amplification Plot

1. Click Analysis > Amplification Plot



2. Check the amplification curve

- a. Set up colour
- b. Set up plot type
- c. Set up show dye

When the background colour of a dye name is blue, it will be displayed; while white indicates it will not be displayed.



- 3. Check the reaction plate
 - a. Select reaction plate well site and check corresponding well site curve
 - The default is all wells are selected
 - b. Zoom-In, Zoom-Out and reset the reaction plate
 - c. Check well table





4. Set up assay

- a. Set up assay
- b. Set up threshold

c. Set up automatic baseline

When the threshold value is not automatic, the user cannot set up the automatic line

Baseline Assay

Assay Target1-FAM 💽 Threshold 🛄 Auto 267 💽 🔝 Auto Baseline

4.1.2 Check Standard Curve

1. Click Analysis > Standard Curve



2. Check standard curve a. Set up assay



3. Check the reaction plate

- a. Select reaction plate well site and check corresponding well site curve
- The default is all wells are selected
- b. Zoom-In, Zoom-Out and reset the reaction plate
- c. Check well table
- d. Check results summary



4.2 Check Relative Quantification 1. Click Analysis ► Relative Quantification

| Analysis | ۲ |
|----------------------------|---|
| Amplification Plot | 0 |
| Standard Curve | 0 |
| Relative Quantification | 0 |

2. Check relative quantitative a. Set up the show type



b. Check the analysis results

| Result | | | | | | | | | |
|-----------|------------|------------|----------------|-------------|----------------|-------------|--------|-------|--------|
| Sample Id | Assay Item | Property | GOI Aver. Con. | GOI Con. SD | HKG Aver. Con. | HKG Con. SD | Max | Min | Aver. |
| | target1 | Comparison | 7.99e+03 | 0.00e+00 | 1.37e+04 | 0.00e+00 | 1 | 1 | 1 |
| 01 | target1 | Unknown | 1.10e+07 | 1.05e+06 | 1.93e+05 | 1.48e+04 | 63.92 | 49.95 | 56.94 |
| 02 | target1 | Unknown | 8.48e+05 | 1.31e+05 | 6.14e+04 | 9.61e+03 | 16.84 | 10.78 | 13.81 |
| 03 | target1 | Unknown | 9.40e+04 | 1.40e+04 | 3.67e+04 | 2.06e+03 | 2.97 | 2.15 | 2.56 |
| 04 | target1 | Unknown | 3.72e+03 | 2.66e+01 | 3.08e+04 | 8.82e+02 | 0.12 | 0.12 | 0.12 |
| 06 | target1 | Unknown | 9.44e+05 | 1.43e+05 | 3.95e+04 | 6.33e+03 | 29.18 | 18.63 | 23.9 |
| 07 | target1 | Unknown | 9.33e+04 | 3.53e+04 | 2.73e+04 | 5.86e+03 | 4.9 | 1.93 | 3. 41 |
| 08 | target1 | Unknown | 4.14e+03 | 2.62e+03 | 2.33e+04 | 8.42e+02 | 0.29 | 0.07 | 0.18 |
| 09 | target1 | Unknown | 8.44e+06 | 5.34e+05 | 9.71e+04 | 3.93e+04 | 122.5 | 51.28 | 86.89 |
| 11 | target1 | Unknown | 7.21e+04 | 1.20e+03 | 1.57e+04 | 2.97e+02 | 4.7 | 4.47 | 4.58 |
| 12 | target1 | Unknown | 1.10e+04 | 0.00e+00 | 1.51e+04 | 0.00e+00 | 0.73 | 0.73 | 0.73 |
| 13 | target1 | Unknown | 8.12e+06 | 8.33e+05 | 8.05e+04 | 1.74e+04 | 125.02 | 76.77 | 100.89 |
| 14 | target1 | Unknown | 8.25e+05 | 6.25e+04 | 2.50e+04 | 2.87e+03 | 37.59 | 28.5 | 33.05 |
| 16 | target1 | Unknown | 6.87e+03 | 3.28e+03 | 8.01e+03 | 4.28e+02 | 1.27 | 0.45 | 0.86 |

4.3 Adjust Parameter Reanalysis

1. Click **Analysis Settings** ► the Analysis Settings dialog box will pop up a. Adjust the start cycle and end cycle of the baseline

- b. Adjust Ct analysis algorithm
- c. Set up the use of S fitting
- d. Set up the stage to use for Ct analysis
- e. Set up the automatic threshold value
- f. Advanced setting
- g. Standard curve setting
- h. Relative quantification setting

| t Settings Ad | vanced Setting | gs Standard | Curve Settings | Relative Quantification Settings |
|--------------------|----------------|--------------|----------------|----------------------------------|
| The stage to use i | for Ct analysi | is: Stage 2 | | |
| The algorithm to o | calculate Ct: | Baseline Thr | eshold | S Fitting |
| ssay Item | Threshold | Start Cycle | End Cycle | target1 - SYBR |
| arget1 - SYBR | 230.6 | Auto | Auto | Auto Threshold |
| arget1 - HEX | Auto | Auto | Auto | Threshold: 230.6 |
| | | | | |
| | | | | |
| | | | | |

5. Experiment Report

This section describes how to print experiment report and covers report template designing and print setting.



This section describes how to print an experiment report and covers designing of a report template and print settings.

5.1 Comprehensive Report

1. Click **Report** ► **Consolidated Reports** ► the Consolidated Report window will pop up The Consolidated Report includes the basic information, sample information, amplification curve, standard curve, plate information, etc..

| (1) | ou mopore | | | | | | | | Y | | |
|---|---|---------------------------|---|---|---|--|---|--|--|----|--|
| | | | | | | | | | | Re | port Items |
| Experim | ment Name: 20111 ment Type: Relat | 1123 tive | LineGer | ne9600 (| Consolida | ated Rep | ort | | 1 / 12 | | Basic Information Run Program Detectors Plot Plate Table Plate Samples Arm Curve(Linear) |
| File Na Run Tim Gain: | me: F:\Li ie: 2011/ F1:6, | ineGene /11/23 F2:7 | e9600\2011 09:47:00 | 11123.fqd - 2011/11 | /23 10:45:4 | 19 | | | | | Amp. Curve(Log) Quan. Analysis Res Standard Curve |
| P | | | | | | | | | | | Relative Plot(Line |
| Hold Star | un riogram | | | | | | | | | - | Relative Plot(Log) |
| Target | Incubation Time | Rate | Sampling | 1 | | | | | | | Relative Analysis |
| 94 | 120 | 4 | | | | | | | | | |
| PCR Stars | e Cucles:40 | | 1000 | | | | | | | | treate Keport |
| Target | Incubation Time | Rate | 2nd Temp. | Step Size | Step Delay | Grad Tenp. | Grad Range | Sampling | | | |
| 95 | 5 | 4 | | | | | tran tange | | | | |
| 60 | 30 | 4 | | | | | | | | | |
| Detector 项目1 | | GOI GOI | I/HKG I | | Dye FAM | | Colo | | - | | |
| P | Plot Plate | HKG | 3 | | HEX | | | | | | |
| 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 | | | | • • | U Marsenau U Marsenau | 8. 201 | 1 :: 1 :: 1 :: 1 :: 1 :: | 4 11 | 13 | | |
| | 11 \$ \$ 1 - 740 (200 1 \$ 2 1 - 400 (200) 1 \$ 2 1 - 400 (200) | | | 21 Ban(602) 21 Ban (602) 21 Ban (822) | 10 (44) | 11, 21 (2 = - 7 xx (ccc)) 10, 22 (2 = - x 22) (ccc) | | II (∦iji) − nam(sac) U (\$ 2 1 − naz nam | | | |
| ¢ | | | Sitt Past(stat) | | | [] ==] | | 748 (902) | U (2 = 1 - ann (and) | | |
| • 11 30 41 1 144 | 1.25 | | Cont. of Stations | | U. 2 51 - 100 | | | - 123 (200) | | | |
| | | | | 21 - AAN(422) 21 - AZ2 (822) 21 - AZ2 (822) | 11 (1 - a a a a a a a a a a a a a a a a a a | | 1 (() - AAE (()) | | 11 - 2 (23 - 2 mil (2033) | | |
| | | | 9 = 1 = 100 (400) | = 1 - FAR(403) 3 : • • • • • • • • • • • • • • • • • • | U State mark | | 15((2) = Aut(COD) 2(2) = 223 (20) (1) (2) = 1 (2) (2) = 1 (2) (2) = 1 | - 22 (23) - 22 (24) - 22 (24) - 22 (25) - 22 (25) | 11) - 25 (d. 1) - 25 (d. 1000) 10 - 5 | | |
| | | - 7x((400)) | S = 1 = 744 (40) U = 1 S = 1 = 744 (40) S = 1 = 15 (40) S = 1 | 21 - 440(42) | Li de | C (1 - 740 (CC) C (1 - 740 (CC)) C (1 - 740 (CC)) | 9 193 - 206(50) U 1981 U | - 202 202 - 202 - 202 - 202 - 202 - 202 - 202 - 202 - 2 | 1) (Refer - Paul (nor) 1) (Refer - Paul (nor) 1) (Refer - Paul (nor) 1) (Refer - Paul (nor) | | |
| | able Plate | | | | | State = xxx (xx) | 1 (2) - FALLOO) U (2) - U | - 74 - 74 - 74 - 74 - 74 - 74 - 74 - 74 | 1) ∰(2) = Parl(cor) 1) ∰(2) = = Parl(cor) 1) ∯(2) = = Parl(cor) 1) ∯(2) = = Parl(cor) | | |

5.2 QC Summary

1. Click Report **C** Summary



2. Check the QC summary



6. Data Export



This section describes how to export data and covers exporting to a database, experiment filing and exporting the experiment data to EXCEL.

6.1 Export to Database

Click **Data Summary** ► **Export to Database** ► the Save File dialog box will pop up ► save the exported database file

6.2 Experiment Filing

1.Click **Data Summary** ► **Archived Experiment Directory** ► the Experimental archive storage directory window will pop up ► set up the storage path of file

| 🐮 Experimental | archive storage directory | |
|--|------------------------------|---|
| Experiments in the | following archive directory: | |
| D:\LineGene9600 | | |
| Change | | |
| and the second sec | OK Cancel | |
| | | 6 |

2. Experiment filing

Click **Data Summary** Archived Experiment export the filed experiment file The suffix of the filed experiment file is .fqh

6.3 Export Experiment Data to EXCEL

Click **Data Summary ► Export Experiment ► Export Experiment to Excle ►** the exported experiment data will generate EXCEL file

6.4 Export Experiment Data to TEXT

Click **Data Summary ► Export Experiment ► Export Experiment to Text ►** the exported experiment data will generate TEXT file

Chapter 6 SNP

1. Design Experiment



This section describes how to design an SNP experiment and covers creating a new SNP experiment, inspection item setting, sample information setting, reaction plate setting and programme setting.

1.1 Create SNP Experiment

1. Click **SNP** on **Home** interface and create SNP Experiment window.

An SNP experiment can be also created by:

- a. Clicking **New SNP** on the toolbar
- b. Clicking File > New > SNP on the menu bar



1.2 Detector Setting





2. Input basic information

Input the experiment name, user name and any comments in the experiment properties column.

| Experiment Prope | erties | | |
|------------------|---------------------|----------|--|
| Experiment Name: | 20111117_Experiment | remark | |
| User Name: | user | Comment. | |

3. Inspection Item Setting

Set up the Detector, Allele, Dye and Colour.

If necessary, the user can also:

- a. Add Detector
 - b. Delete Detector

c. Add the Detector in the Detector library: click **Add Detector From Library** ► the **Detector Library** window will pop up ► select the Detector on the window to be added

| The use | er can also co | nduct Add, I | Modify and | Delete oper | ations i | n the it | tem | library. |
|---------------------------|----------------|--------------|------------|-------------|----------|----------|-----|----------|
| Contraction of the second | | | | | | | | - |

| Detector | Allele | Reporter | Color | Master Mix | Primer | Probe | Supplies | Batch Number |
|----------|---------|----------|-------|------------|--------|-------|----------|--------------|
| Target1 | Allele1 | FAM | | | | | | |
| | Allele2 | HEX | | | | | | |
| Target2 | Allele1 | FAM | | | | | | |
| | Allele2 | HEX | | | | | | |
| | | | | | | | | |
| | | | | | | | | |

d. Set up the item name, set up the dye name and set up the colour

| Detectors | Add Detector | Delete Detector | Add I | Detector From Libra | IRY . | | | |
|-----------|--------------|-----------------|-------|---------------------|--------|-------|----------|--------------|
| Detector | Allele | Reporter | Color | Master Mix | Primer | Probe | Supplies | Batch Number |
| Target1 | Allele1 | FAM | | | | | | |
| | Allele2 | HEX | | | | | | |
| | | | | | | | | |

4. Set up reference dye



1.3 Sample Information Setting

1. Click Setup **Sample**

| Setup | • |
|----------|---|
| Detector | 0 |
| Sample | 0 |
| Plate | 0 |
| Program | O |

- 2. Add sample information
 - a. Itemized addition: input ID in **Sample ID** ► press **Enter** ► add information for one sample
 - b. Batch addition: click **Batch Add** I the Batch Add window will pop up

| R Batch Add | × |
|-------------------|----------------|
| Start Sample Id a | Sample Count 5 |
| | Add Cancel |

3. Delete sample information

a. Itemized deletion: select one sample ► click **Delete** ► delete the selected sample information

b. Delete all: click **Clear All**

delete all sample information

4. Import/Export sample information

a. Click **Import Sample Info** the File Import window will pop up import sample information file in CSV format

b. Click **Export Sample Info** ► the Save As window will pop up ► the sample information will be exported in CSV file format

| Sample ID Batch Add Delete Clear | All Import Samples Info Export Samples Info |
|----------------------------------|---|
|----------------------------------|---|

5. Set up sample information

| Samples | _ | | | | _ |
|-----------|-------|-------------|---------------|-----------------|---|
| Sample Id | Color | Sample Name | Sampling Time | Submitting Date | |
| al | | Sample1 | 2013-12-06 | 2013-12-06 | |
| a2 | | Sample2 | 2013-12-06 | 2013-12-06 | |
| a3 | | Sample3 | 2013-12-06 | 2013-12-06 | |
| a4 | | Sample4 | 2013-12-06 | 2013-12-06 | |
| a5 | | Sample5 | 2013-12-06 | 2013-12-06 | |

1.4 Reaction Plate Setting

1. Click Setup 🕨 Plate

| Setup | • |
|----------|---|
| Detector | 0 |
| Sample | 0 |
| Plate | 0 |
| Program | 0 |

- 2. Set up the inspection criteria of the reaction plate
 - a. Select reaction plate well site: click Reaction Plate well Site

The user can also right click the reaction plate well site to Copy, Paste and Add New Detector. Adding a new detector will open the **Edit Detector Library** window.

| Edit De | tector Libr | rary | | | | | |
|--------------|-------------|-------|------------|--------|-------|----------|--------------|
| Detector Nan | me: Target2 | | | | | | |
| Allele | Reporter | Color | Master Mix | Primer | Probe | Supplies | Batch Number |
| Allele1 | FAM | - | | | | | |
| Allele2 | HEX | - | | | | | |
| | | | | | | | 2 |
| 4 | | | | | | | |

b. Select inspection item and modify the property, concentration and concentration unit.

| Property | Name | Concentration | Concentration unit |
|------------|----------------------------|---------------|--------------------|
| | Unknown | NO | |
| z | Negative | NO | Copies/ml |
| 1 | Positive Allelic gene 1 | NO | Ea/ml |
| <u>1</u> 2 | Positive Heterozygous | NO | Pg/ml |
| 22 | Positive Allelic gene 2 | NO | |

- c. Select a sample and the list displayed will change
- d. Zoom-In, Zoom-Out and reset the reaction plate.
- e. Sample Auto Arrange
- f. Check Well Table

| ▲ De | tectors | | | | Plate Se | tup Well | Table | | | | | | | | | |
|------|-------------|--------------------------|------------|--------|----------|----------|-------|---------|----------|-------|---|--------------|---------|----|----|----|
| Dete | ctor | Property | Con. | - (- J | | | | Zoom In | Zoom Out | Reset | S | ample Auto A | Arrange | | | |
| Targ | et1 | U | - | | 1 | | | | | | 7 | 8 | | 10 | 11 | 12 |
| 1 | am 📝 hex | | | | | etl | | | | | | | | | | |
| C | oncentratio | n Unit <mark>copi</mark> | es/ml | - | в | | | | | | | | | | | |
| ▲ Sa | amples | Show Column | us: Sample | • Name | с | | | | | | | | | | | |
| | al | Samp | le1 | | - | | | | | | | | | | | |
| | a2 | Samp | le2 | | D | | | | | | | | | | | |
| | a3 a4 | Samp Samp | le3 le4 | | | | | | | | | | | | | |
| | а5 | Samp | le5 | | E | | | | | | | | | | | |
| | | | | | F | | | | | | | | | | | |
| | | | | | G | | | | | | | | | | | |
| | | | | | н | | | | | | | | | | | |

| Plate | e Setup | Well Ta | ble | | _ | |
|-------|---------|-----------|------------|----------|-----|---|
| # | Well | Sample Id | Assay Item | Property | Dye | 4 |
| 1 | A01 | al | Target1 | Unknown | FAM | 3 |
| 1 | A01 | al | Target1 | Unknown | HEX | 4 |
| 2 | A02 | | | | | |
| 3 | A03 | | | | | |
| 4 | A04 | | | | | |
| 5 | A05 | | | | | |
| 6 | A06 | | | | | |
| 7 | A07 | | | | | |
| 8 | A08 | | | | | |

1.5 Programme Setting

1. Click Setup > Programme



2. Run Programme Setup

a. Create new stage: the user can create a new Hold Stage, Cycling Stage or Melting Stage

The user can also click **Add Stage** directly and the default will be creating a new **Cycling Stage**.

b. Create new step: the user can create a new step **Before** or **After** the currently selected step

The user can also click **Add Step** and the default will be adding a new Step at the end of the currently selected stage or after the currently selected step.

c. Delete: the user can delete the currently selected step or stage

d. Display form: click **Display With Table** ► new window will pop up ► the details of the current experiment will be displayed in a table.

e. Set up the experimental data of the hold stage, cycling stage and melting stage melting section

| | 🔽 Hot. | lid(C) 105 🛉 Liquid Quant. (ul) 40 🔦 | |
|----------|---------------------------------|--|-------|
| Run Prog | rams Setup Stage Add Step De | Display With | fable |
| | Hold Stage | PCR Stage | |
| | | Cycles 40 | |
| | | 2nd Temp Step Size - <mark>2nd Temp Step Size -</mark> | |
| | | Step Delay - Step Delay - | |
| | | Grad Temp Grad Range Grad Temp Grad Range | |
| 100°C — | | | |
| | 95 °C | 4 °C/s 95 °C | |
| | 00:00:20 | 00:00:15 | |
| | Sampling | Sampling | |
| | | 4 °C/s | |
| | | | |
| | | | |
| | 4 °C/s | | |
| | | | |
| | | Sampling | |
| | | | |
| | / | | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |

f. Set up the hot-lid temperature and liquid volume

2. Prepare for Reaction

| Start |
|---------------------|
| ▼ |
| Design experiment |
| ▼ |
| Prepare for the |
| reaction |
| ▼ |
| Run the experiment |
| ▼ |
| Experiment analysis |
| ▼ |
| Experiment report |
| ▼ |
| Data export |
| ▼ |
| End |

The user should make full preparations prior to the experiment:

• Ensure appropriate materials are used.

• Ensure the arrangement of the PCR reaction plate is consistent with the setting layout of reaction plate in Section 1.4.

3. Run the Experiment

This section describes how to run the experiment after loading the reaction plate and covers the operating of fluorescence curve, the operating of temperature curve and programme setting.



This section describes how to run/operate the experiment after loading the reaction plate and includes how to operate the fluorescence curve, the temperature curve and programming

3.1. Run Fluorescence Curve 1. Click Run ► Fluorescence Curve

| Run | • |
|-----------------------|---|
| Fluorescence Curve | 0 |
| Temperature Curve | 0 |
| Program | 0 |

2. Click Start Run



3. Operating confirmation

a. Modify hot-lid temperature and liquid quantity (sample volume).

- 4. After it starts running, the user can:
 - a. Skip the current stage
 - b. Add a cycle
 - c. Delete a cycle
 - d. Stop run
- 5. Plot display setting
 - a. Assay item
 - b. Plot colour



3.2 Run Temperature Curve 1. Click Run ► Temperature Curve

| Run | • |
|-----------------------|---|
| Fluorescence Curve | 0 |
| Temperature Curve | 0 |
| Program | 0 |

2. Click Start Run



- 3. Operating confirmation
- a. Modify hot-lid temperature and liquid quantity (sample volume).
- 4. After it starts running, the user can:
 - a. Skip the current stage
 - b. Add a cycle
 - c. Delete a cycle
 - d. Stop run



3.3 Programme Setting

The user can only check the programme setting but cannot make modifications.

4. Experiment Analysis



This section describes how to view the experiment analysis results after running an experiment and adjusting parameters for re-analysis.

This section covers the analysis of amplification curves and standard curves, adjusting parameters for re-analysis and importing parameters.

4.1 Check Results 4.1.1 Check the Amplification Plot 1. Click Analysis ► Amplification Plot



2. Check the amplification curve

- a. Set up colour
- b. Set up plot type
- c. Set up show dye

When the background colour of a dye name is blue, it will be displayed; while white indicates it will not be displayed.



3. Check the reaction plate

a. Select reaction plate well site and check corresponding well site curve The default is all wells are selected

- b. Zoom-In, Zoom-Out and reset the reaction plate
- c. Check well table
- d. Check results summary



4. Set up inspection item

- a. Set up assay
 - b. Set up threshold
 - c. Set up automatic baseline

When the threshold value is not automatic, the user cannot set up the automatic baseline

Assay Target1-FAX 🔽 Threshold 🔝 Auto 🔽 267 🚔 🛄 Auto Baseline

4.1.2 Check SNP

Assay

1. Click Analysis > SNP



2. Check SNP

a. Select well site

The user can select well site by dragging a rectangle with the mouse around the wells of interest or select wells one by one.

- b. Set up Assay
- c. Set up manual calls



3. Check the reaction plate

a. Select reaction plate well site and check corresponding well site curve The default is all wells are selected

- b. Zoom-In, Zoom-Out and reset the reaction plate
- c. Check well table information
- d. Check results summary

4.2 Adjust Parameter Re-analysis

1. Click **Analysis Settings** I the Analysis Settings dialog box will pop up

a. Adjust analysis data

b. Adjust whether the inspection item will retain manual recognition genotype



5. Experiment Report



This section describes how to print an experiment report and covers designing of a report template and print setting.

5.1 Designing a Report Template

1. Click **Report > Report Template Editor >** the Report Designer window will pop up

The report consists of controls and the user can add, modify and delete controls. Available controls include Static Text, Dynamic Text, Line, Static Image and SNP Typing Curve.

| 🛃 Report De | signer - D:\LineG | Gene9600\Scientific\config\report\Snp\default.rpt | | | | | | | |
|---|-------------------------------|--|---|--|--|--|--|--|--|
| 🗄 🗋 New 🔗 Ope | n 🔚 Save 🛛 🗟 Preview | / 📓 View 👻 🗙 Delete Selected Controls 🖹 🗐 🕂 🔟 📴 | () Settings | | | | | | |
| Available controls | Used controls | | | | | | | | |
| Common Controls Static Text Static Text Static Image Line ShP Typing Curve Known Controls Static Text Controls Dynamic Text Controls | | [Hospital] [Report] | | | | | | | |
| | | Name: [Name] Sex: [Sex] Age: [Age] HospitalNo.: [HospitalNo.] | | | | | | | |
| | | Test Item: [Test Item] Gene Typing: [Gene Typing] SNP Typing Result Plot SNP Typing Result Plot SNP Typing Result Plot 15 15 10 | | | | | | | |
| 2↓ □ | | | | | | | | | |
| | | 0 5 10 15 20 25 | 30 | | | | | | |
| Alignment | MiddleLeft | E | *************************************** | | | | | | |
| BackColor | White | and a second | Suturne | | | | | | |
| 🕀 Border | Solid, 1, False, False, False | Submitting Date: Submitting Date: Report Date: Report Date: Report Date: Submitting Date: S | ckerj | | | | | | |
| Color | Black | | | | | | | | |
| Font | Tahoma, 8.25pt | | | | | | | | |
| 🗆 Data | | | | | | | | | |
| DataField | Sex | | | | | | | | |
| Tag | | | | | | | | | |
| 🗄 Design | | | | | | | | | |
| DesignVisible | True | | | | | | | | |
| Name | DataField6 | | | | | | | | |
| Layout | | | | | | | | | |
| Location | 266, 109 | | | | | | | | |
| | 0, 0, 0, 0 | | | | | | | | |
| 🛨 Size | 49, 20 | | | | | | | | |
| Туре | DataField | | | | | | | | |
| DataField data field of the ele | ement | | | | | | | | |
| | | | | | | | | | |

5.2 Print Setting

1. Click **Report** > **Print Template Setting** > the Print Template Setting window will pop up

The user can set up the laboratory name, report name, reference value, tester, checker, amplification plot set up, default report template and paper size.

| R Print Template Settings(SMP) | |
|---------------------------------|-----------|
| Template Setup | |
| Hospital | |
| Report | |
| Tastar | |
| lester | |
| Checker | |
| | |
| | |
| | |
| | |
| | |
| Print Setup | |
| Default Report Template default | |
| Paper Size A4 | - |
| Printer | |
| Use Default Printer | |
| 🔘 Use Custom Printer | - |
| | |
| | OK Cancel |
| | |

5.3 Comprehensive Report

1. Click **Report > Consolidated Reports >** the Consolidated Report window will pop up.

The Consolidated Report includes the basic information, sample information, amplification curve, SNP, plate information, etc..

| solidate | d Report | | | | | | | | | | |
|---|---|-------------------------|-----------------------|--------------------------|--------------------|--------------------------|--------------------------------|-------------|--------|-----|---|
| ⊕ ⊖ | | | | | | | | | | | Report Items |
| Experime | ent Name: 20111 | .125 | LineGe | ne9600 C | Consolida | ated Rep | ort | | 1 / | 11 | Basic information Run Program Detectors Plot Plate Table Plate Samples |
| Experime File Nar Run Time Gain: | ent Type: SNP me: F:\Li e: 2011/ F1:6, | neGen (11/25 F2:7 | e9600\201 13:42:45 | 11125_SNP. - 2011/11, | fqd /25 14:41:2 | 29 | | | | | Amp. Curve(Linear) Amp. Curve(Log) Quan. Analysis Rest |
| | | | | | | | | | | | SNP Analysis Resul |
| Hold Stag | in Program —— e | | | | | | | | | _ | |
| Target | Incubation Time | Rate | Sampling | | | | | | | | Create Report |
| 94 | 120 | 4 | | | | | | | | | |
| PCR Stage | Cycles:40 | | | | | | | | | _ | |
| Target | Incubation Time | Rate | 2nd Temp. | Step Size | Step Delay | Grad Temp. | Grad Range | Sampling | . I. | | |
| 95 | 5 | 4 | | | | | | | | | |
| 60 | 30 | 4 | | | | | | | × | | |
| De | etectors | | | | | | | | | | |
| Detector | | Allele | | | Dye | | Color | Color | | | |
| - <u>щ</u> п1 | | A11 | lele2 | | HEX | | | | | | |
| | | 1 | | | 1.000 | | | | | - | |
| P1 | lot Plate | | | | | | | | | | |
| 1 | 2 3 | | 4 | 5 6 | 7 | 8 | 9 | 10 | 11 12 | - | |
| A | | | | | | | | | | | |
| | 181 - FAN 🖬 🏹 811 - | FAM IN 1 | EU1 - PAR | 项目1 - FAM | 22 项目1 - FAM | 2 项目1 - FAM ² | 2 项目1 - FAM 22 | 项目1 - ITAM | | | |
| | | HCA I | S H 1 - HEA | er e 1 - HEX | 22 - X E 1 - HEX | | | | | | |
| c | | | | 1000 | | | | | | | |
| | | | | - | | 75.11. 200 | | 75 11 . 941 | - | - | |
| D 2 | | 200 | ABI PAR 2 | | | | | | N JEL- | REX | |
| 1 12 3 | | and Fast at | S H 1 - DEAL | -3 C + 040 | | CALCER STORE | C3 - 2 C - 1 - 1 - 1 - 2 C - 2 | | | | |
| | | | S H I T BLALLE | | | | | | | | |
5.4 Report Printing 1. Click **Report > Report Print**



- 2. Report print setting a. Set up report template
 - b. Print setting (please refer to Section 5.2)c. Select print items

 - d. Print preview

| Э. | Pr | in | t ti | he | re | ро | rt |
|----|----|----|------|----|----|----|----|
| _ | | | | | | | |

| elect/Un | Select | Select All Sa | mples | | | | | | | | | | | | 🔜 Print One Assay PerRepo |
|----------|----------|---------------|-----------|------|-----|-----|----------|----------------|---------|--------------|-------------|---------------|-----------|-------|---------------------------|
| rint S | ample Id | Sample Name | Test Item | Name | Sex | Age | Case No. | Outpatient No. | Bed No. | Hospital No. | Nationality | Sampling Time | Diagnosis | Notes | |
| 0 | 4 | | target1 | 1 | 1 | 1 | 2 | | 0 | | 0 | 2011/12/15 | | | |

5.5 QC Summary

1. Click Report **>** QC Summary



2. Check the QC summary

| Amp | olificatio | on Plot | | | | | | | | | | QC Summary | | | | |
|-------|------------|---------|-----|-----|--------|-------------|------|-----|------|------|-----|--|-------|-----|--------|--|
| ſ | Color | Well | | - P | lotTvp | e Line | ar | | Show | EI E | 2 | Description | Value | Use | Result | |
| | 7000-[| | | | | | | | | | 4 | Negative control with a Ct less than | 38 | - | | |
| (| 5000- | | | | | _ | | | | _ | _ | Positive control with a Ct greater than | 30 | - | | |
| 8 | 5000 | | | | | | | | | | | Unknown without a Ct | N/A | 1 | | |
| cenc | 1000 | | | | | | | | | 1 | | Standard without a Ct | N/A | - | | |
| lores | 8000- | | | | | | | | _/ | | 1 | | | | | |
| Ē | 2000 | | | | | | | | | P | | | | | | |
| 1 | 1000 | | | | | | | 1 | 1 | / | _ | | | | | |
| | 1 | | | | | | - | | | _ | | | | | | |
| | | 9 1 | 2 1 | 5 1 | 8 2 | 1 24 CVC | 4 27 | 30 | 33 | 36 | 39 | | | | | |
| | 1.000 | | | | | 0,0 | | | 20.5 | | | | | | | |
| A01 | A02 | A03 | A04 | A05 | A06 | A07 | A08 | A09 | A10 | A11 | A12 | | | | | |
| B01 | 802 | B03 | B04 | B05 | 806 | B07 | 808 | B09 | B10 | B11 | 812 | | | | | |
| C01 | 002 | C03 | C04 | C05 | 006 | 007 | 008 | C09 | C10 | C11 | C12 | | | | | |
| DOI | D02 | D03 | D04 | DUS | D06 | D07 | D08 | D09 | D10 | D11 | D12 | | | | | |
| E01 | E02 | E03 | E04 | E05 | E06 | E07 | E08 | E09 | EIO | EII | E12 | | | | | |
| F01 | F02 | F03 | F04 | P05 | F06 | F07 | F08 | F09 | F10 | F11 | F12 | | | | | |
| 601 | 602 | 603 | 604 | 605 | 606 | 607 | 608 | 609 | 610 | 611 | 612 | | | | | |
| H01 | H02 | H03 | H04 | H05 | H06 | H07 | H08 | H09 | H10 | H11 | H12 | - | | | | |

6. Data Export



This section describes how to export data and covers exporting to a database, experiment filing and exporting the experiment data to EXCEL.

6.1 Export to Database

Click **Data Summary** ► **Export to Database** ► the Save File dialog box will pop up ► save the exported database file

6.2 Experiment Filing

1. Click **Data Summary** ► **Archived Experiment Directory** ► the Experimental archive storage directory window will pop up ► set up the storage path of file

| 👯 Experimenta | l archive storage directory | |
|-------------------|--------------------------------|--|
| Experiments in th | e following archive directory: | |
| D:\LineGene9600 | | |
| Change | | |
| | OK Cancel | |
| 1 | | |

2. Experiment filing

Click **Data Summary** Archived Experiment export the filed experiment file

The suffix of filed experiment file is .fqh

6.3 Export Experiment Data to EXCEL

Click **Data Summary ► Export Experiment ► Export Experiment to Excle ►** the exported experiment data will generate EXCEL file.

6.4 Export Experiment Data to TEXT

Click **Data Summary ► Export Experiment ► Export Experiment to Text ►** the exported experiment data will generate TEXT file.

Chapter 7 High Resolution Melting

1. Design Experiment



This section describes how to design an SNP experiment and covers creating a new SNP experiment, inspection item setting, sample information setting, reaction plate setting and programme setting.

1.1 Create High Resolution Melting Experiment

1. Click **HRM** on **Home** interface and create SNP Experiment window.

An SNP experiment can be also created by:

- a. Clicking **New HRM** on the toolbar
- b. Clicking File **New HRM** on the menu bar



1.2 Detector Setting 1. Click Setup ► Detector

| Setup | 0 |
|----------|---|
| Detector | 0 |
| Sample | 0 |
| Plate | 0 |
| Program | 0 |

2. Input basic information

Input the experiment name, user name and any comments in the experiment properties column.

| Experiment Prop | | | | |
|------------------|---------------------|----------|--------|--|
| Experiment Name: | 20111117_Experiment | | remark | |
| User Name: | user | Comment. | | |

3. Inspection Item Setting

Set up the Detector, Allele, Dye and Colour.

If necessary, the user can also:

- a. Add Detector
- b. Delete Detector

c. Add the Detector in the Detector library: click Add Detector From Library ► the Detector Library window will pop up ► select the Detector on the window to be added

The user can also conduct Add, Modify and Delete operations in the item library.

| | ary | | | |
|---------------|-------|-----|-------|--|
| Add Modify De | lete | | | |
| Detector | Assay | Dye | Color | |
| Target3 | GOI | FAM | | |
| | HKG | HEX | | |
| Target4 | | FAM | | |
| | | HEX | | |
| | | | | |
| | | | | |
| | | | | |
| | | | | |
| | | | | |

d. Set up the item name, set up the dye name and set up the colour

| Detectors | Add Detector | Add Assay | Delete Detector | Delete Assay | Add Detector From Library |
|-----------|--------------|-----------|-----------------|--------------|---------------------------|
| Detector | Ass | say | Dye | Color | |
| Target1 | G | DI | FAM | | |
| | | | | | |

4. Set up reference dye

Reference Dye

1.3 Sample Information Setting

1. Click Setup 🕨 Sample

| Setup | • |
|----------|---|
| Detector | 0 |
| Sample | 0 |
| Plate | 0 |
| Program | 0 |

2. Add sample information

a. Itemized addition: input ID in **Sample ID** > press **Enter** > add information for one sample

b. Batch addition: click **Batch Add** I the Batch Add window will pop up

| C Batch Add | | | <u>×</u> |
|-----------------|---|--------------|----------|
| Start Sample Id | a | Sample Count | 5 🔦 |
| | | Add | cel |

3. Delete sample information

a. Itemized deletion: select one sample ► click **Delete** ► delete the selected sample information

b. Delete all: click **Clear All**

delete all sample information

4. Import/Export sample information

a. Click **Import Sample Info** ► the File Import window will pop up ► import sample information file in CSV format

b. Click **Export Sample Info** ► the Save As window will pop up ► the sample information will be exported in CSV file format

| Sample ID | Batch Add | Delete | Clear All | Import Samples Info | Export Samples Info |
|-----------|-----------|--------|-----------|---------------------|---------------------|
| | | | | | |

5. Set up sample information

| Sample Id | Color | Sample Name | Sampling Time | Submitting Date | |
|-----------|-------|-------------|---------------|-----------------|--|
| al | | Sample1 | 2013-12-06 | 2013-12-06 | |
| a2 | | Sample2 | 2013-12-06 | 2013-12-06 | |
| a3 | | Sample3 | 2013-12-06 | 2013-12-06 | |
| a4 | | Sample4 | 2013-12-06 | 2013-12-06 | |
| a5 | | Sample5 | 2013-12-06 | 2013-12-06 | |

1.4 Reaction Plate Setting

| 1. | Click Set | up 🕨 | Plate |
|----|-----------|------|-------|
| | Setup | • | |
| | Detector | 0 | |
| | Sample | 0 | |
| | Plate | 0 | |
| | Program | 0 | |

2. Set up the inspection criteria of the reaction plate

a. Select reaction plate well site: click Reaction Plate well Site

The user can also right click the reaction plate well site to Copy, Paste and Add New Detector. Adding a new detector will open the **Edit Detector Library** window.

| Edit Detec | tor Library | | |
|----------------|-------------|-------|--|
| Detector Name: | Target4 | | |
| Assay | Dye | Color | |
| | FAM | | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |

b. Select inspection item and modify the property, concentration and concentration unit.

| Property | Name | Concentration | Concentration unit |
|----------|----------|---------------|--------------------|
| U | Unknown | NO | Copies/ml |
| S | Standard | YES | IU/ml |
| Z | Negative | NO | Fg/ml |
| P | Positive | NO | Pg/ml |

- c. Select a sample and the list displayed will change
- d. Zoom-In, Zoom-Out and reset the reaction plate.
- e. Sample Auto Arrange
- f. Check Well Table

| ▲ De | tectors | | | | Plate Se | tup Well | Table | | | | | | | | | |
|------|-------------|--------------------------|------------|--------|----------|----------|-------|---------|----------|-------|---|--------------|---------|----|----|----|
| Dete | ctor | Property | Con. | - (- J | | | | Zoom In | Zoom Out | Reset | S | ample Auto A | Arrange | | | |
| Targ | et1 | U | - | | 1 | | | | | | 7 | 8 | | 10 | 11 | 12 |
| 1 | am 📝 hex | | | | | etl | | | | | | | | | | |
| C | oncentratio | n Unit <mark>copi</mark> | es/ml | - | в | | | | | | | | | | | |
| ▲ Sa | amples | Show Column | us: Sample | • Name | с | | | | | | | | | | | |
| | al | Samp | le1 | | - | | | | | | | | | | | |
| | a2 | Samp | le2 | | D | | | | | | | | | | | |
| | a3 a4 | Samp Samp | le3 le4 | | | | | | | | | | | | | |
| | а5 | Samp | le5 | | E | | | | | | | | | | | |
| | | | | | F | | | | | | | | | | | |
| | | | | | G | | | | | | | | | | | |
| | | | | | н | | | | | | | | | | | |

| Plate | e Setup | Well Ta | ble | | _ | |
|-------|---------|-----------|------------|----------|-----|---|
| # | Well | Sample Id | Assay Item | Property | Dye | 4 |
| 1 | A01 | al | Target1 | Unknown | FAM | |
| 1 | A01 | al | Target1 | Unknown | HEX | 4 |
| 2 | A02 | | | | | |
| 3 | A03 | | | | | |
| 4 | A04 | | | | | |
| 5 | A05 | | | | | |
| 6 | A06 | | | | | |
| 7 | A07 | | | | | |
| 8 | A08 | | | | | |

1.5 Programme Setting

1. Click Setup > Programme



2. Run Programme Setup

a. Create new stage: the user can create a new Hold Stage, Cycling Stage or Melting Stage

The user can also click **Add Stage** directly and the default will be creating a new **Cycling Stage**.

b. Create new step: the user can create a new step **Before** or **After** the currently selected step

The user can also click **Add Step** and the default will be adding a new Step at the end of the currently selected stage or after the currently selected step.

c. Delete: the user can delete the currently selected step or stage

d. Display form: click **Display With Table** ► new window will pop up ► the details of the current experiment will be displayed in a table.

e. Set up the experimental data of the hold stage, cycling stage and melting stage melting section

| | | 💙 Hotlid(C) | 105 📥 Liquid Quant.(ul) | 40 💌 | |
|----------|--------------------------------|--|--|-----------------------|---|
| Run Prog | rams Setup Stage Add Step D | elete | | | Display With Table |
| | Hold Stage | PC Cyc | R Stage | | Melting Stage Step 0.1 Step Holding Sec. |
| | | 2nd Temp Step Size Step Delay - Grad Temp Grad Range | - 2nd Temp Step Size - Step Delay - Grad Temp Grad Range | | |
| | 85 °C | 95 °C 4 °C/s 00:00:15 Sampling | 437/6 | 95 °C 00:00:15 👻 🗸 | 437/6 |
| | A TC/s | J. | 60 °C | | 57 00 57 00 10:00 |
| | | | Sampling | | Sampling |
| | | | | | |
| | P1 0 | The second secon | | 0 | |

f. Set up the hot-lid temperature and liquid volume

2. Prepare for Reaction

| Start |
|---------------------|
| ▼ |
| Design experiment |
| ▼ |
| Prepare for the |
| reaction |
| ▼ |
| Run the experiment |
| ▼ |
| Experiment analysis |
| ▼ |
| Experiment report |
| ▼ |
| Data export |
| ▼ |
| End |

The user should make full preparations prior to the experiment:

• Ensure appropriate materials are used.

• Ensure the arrangement of the PCR reaction plate is consistent with the setting layout of reaction plate in Section 1.4.

3. Run the Experiment

This section describes how to run the experiment after loading the reaction plate and covers the operating of fluorescence curve, the operating of temperature curve and programme setting.



This section describes how to run/operate the experiment after loading the reaction plate and includes how to operate the fluorescence curve, the temperature curve and programming

3.1. Run Fluorescence Curve 1. Click Run ► Fluorescence Curve

| Run | • |
|-----------------------|---|
| Fluorescence Curve | 0 |
| Temperature Curve | 0 |
| Program | 0 |

2. Click Start Run



3. Operating confirmation

a. Modify hot-lid temperature and liquid quantity (sample volume).

- 4. After it starts running, the user can:
 - a. Skip the current stage
 - b. Add a cycle
 - c. Delete a cycle
 - d. Stop run

5. Plot display setting

- a. Assay item
 - b. Plot colour



3.2 Run Temperature Curve 1. Click Run ► Temperature Curve

| Run | • |
|-----------------------|---|
| Fluorescence Curve | 0 |
| Temperature Curve | 0 |
| Program | 0 |

2. Click Start Run



- 3. Operating confirmation
 - a. Modify hot-lid temperature and liquid quantity (sample volume).
- 4. After it starts running, the user can:
 - a. Skip the current stage
 - b. Add a cycle
 - c. Delete a cycle
 - d. Stop run



3.3 Programme Setting

The user can only check the programme setting but cannot make modifications.

4. Experiment Analysis



This section describes how to view the experiment analysis results after running an experiment and adjusting parameters for re-analysis.

This section covers the analysis of amplification curves and standard curves, adjusting parameters for re-analysis and importing parameters.

4.1 Check Results 4.1.1 Check the Amplification Plot 1. Click Analysis ► Amplification Plot

| Analysis | \odot |
|-----------------------|---------|
| Amplification Plot | 0 |
| Standard Curve | 0 |
| HRM Curve | 0 |

- 2. Check the amplification curve
 - a. Set up colour
 - b. Set up plot type
 - c. Set up show dye

When the background colour of a dye name is blue, it will be displayed; while white indicates it will not be displayed.



3. Check the reaction plate

a. Select reaction plate well site and check corresponding well site curve The default is all wells are selected

- b. Zoom-In, Zoom-Out and reset the reaction plate
- c. Check well table
- d. Check results summary



4. Set up inspection item

- a. Set up assay
- b. Set up threshold
- c. Set up automatic baseline

When the threshold value is not automatic, the user cannot set up the automatic baseline

Assay Assay Targeti-FAX 🔽 Threshold 🛄 Auto 🔽 267 🚔 🛄 Auto Baseline

4.1.2 Check the Standard Curve 1. Click Analysis ► Standard Curve



2. Check the Standard Curve

a. Set up array



 Check the reaction plate
 a. Select reaction plate well site and check corresponding well site curve The default is all wells are selected

- b. Zoom-In, Zoom-Out and reset the reaction plate
- c. Check well table
- d. Check results summary



4.1.3 Check HRM 1. Click Analysis ► HRM Curve



- 2. Check the fluorescence curve
 - a. Set up target
 - b. Set up color



- 3. Check the derivative curve
 - a. Set up target
 - b. Set up color



4. Check the aligned curve

74

a. Set up target

b. Set up color



79

6. Check the reaction plate

- a. Select reaction plate well site and check corresponding well site curve
- The default is all wells are selected
- b. Zoom-In, Zoom-Out and reset the reaction plate
- c. Check well table



4.2 Adjust Parameter Re-analysis

- 1. Click **Analysis Settings** ► the Analysis Settings dialog box will pop up a. Adjust analysis data
 - b. Adjust whether the inspection item will retain manual recognition genotype



5. Experiment Report



This section describes how to print an experiment report and covers designing of a report template and print setting.

5.1 Comprehensive Report

1. Click **Report > Consolidated Reports >** the Consolidated Report window will pop up.

The Consolidated Report includes the basic information, sample information, amplification curve, HRM curve, plate information, etc..

| | | | | | C | onsoli | dated R | Report | | | | | | Run Program |
|-----|------------|----------------|---------------|---------|---------------|--------------|----------------|----------------|---|-----------------|-----------|------------|--------------|-----------------------|
| | Plot Plate | | | | | | | | | | | - 11 | V Plot Plate | |
| . 1 | 2 | 3 | 4 | 5 | | 8 | 7 | 8 | 9 | 10 | 1 | 1 | | Table Plate |
| A | | | 1 | U targe | ti - FAM U ta | arget1 - FAM | U target1 - FA | | | J target1 - FAI | W U targe | 11 - FAM U | II. | Amplification Curve |
| | | | | | | | | | | | | | | Plot Type Linear |
| в | 0 | arget1 - FAM U | target1 - FAM | U targe | 1 - FAM | | | U target1 - Fa | M | | | | | Quan, Analysis Result |
| c | | | | | | | | | | | | | | Melting Curve |
| | | | | | | | | | | | | | | Melting Curve(Derivat |
| D | 8.1 | | | | 4 | | | | | | | | | Melting Analysis Resu |
| | | | | | | | | | | | | | - | HRM(Aligned) |
| E | | | | | | | | | | | | | | HRM(Difference) |
| F | | | | | | | | | | | | | | |
| | | | | | | | | | | | | | | Create Report |
| G | | | | | | | | | | | | | | Crocket Stopper |
| | | | | | | | | | | | | | | Management and P |
| н | | | | | | | | | | | | | | Print Report |
| | — Ta | hle Plate - | | | | | | | | | | | | |
| # | Woll | Accoultom | Property | Duo | Std Con | Sampla | | | | | | | - 1 | |
| # | AOF | Assay item | Fighter | EAM | Stu. Con. | Sample | | | | | | | - | |
| 6 | A05 | target1 | Unknown | EAM | | | | | | | | | - 1 | |
| 7 | A07 | target1 | Unknown | FAM | | | | | | | | | | |
| 10 | A10 | target1 | Unknown | FAM | | | | | | | | | | |
| 11 | A11 | target1 | Unknown | FAM | | | | | | | | | | |
| 12 | A12 | target1 | Unknown | FAM | | | | | | | | | | |
| 15 | B03 | target1 | Unknown | FAM | | | | | | | | | | |
| 16 | B04 | target1 | Unknown | FAM | | | | | | | | | | |
| 17 | B05 | target1 | Unknown | FAM | | | | | | | | | | |
| 20 | B08 | target1 | Unknown | FAM | | | | | | | | | | |

5.2 QC Summary

1. Click Report **>** QC Summary



2. Check the QC summary



6. Data Export



This section describes how to export data and covers exporting to a database, experiment filing and exporting the experiment data to EXCEL.

6.1 Export to Database

Click **Data Summary** ► **Export to Database** ► the Save File dialog box will pop up ► save the exported database file

6.2 Experiment Filing

1. Click **Data Summary** ► **Archived Experiment Directory** ► the Experimental archive storage directory window will pop up ► set up the storage path of file

| 👯 Experimenta | l archive storage directory | |
|-------------------|--------------------------------|--|
| Experiments in th | e following archive directory: | |
| D:\LineGene9600 | | |
| Change | | |
| | OK Cancel | |
| 1 | | |

2. Experiment filing

Click **Data Summary** Archived Experiment export the filed experiment file

The suffix of filed experiment file is .fqh

6.3 Export Experiment Data to EXCEL

Click **Data Summary ► Export Experiment ► Export Experiment to Excle ►** the exported experiment data will generate EXCEL file.

6.4 Export Experiment Data to TEXT

Click **Data Summary ► Export Experiment ► Export Experiment to Text ►** the exported experiment data will generate TEXT file.

Chapter 8 Service

1. User Management

User management is used to manage user information Click Service > User Management on the menu bar

| | User M | langement | iana | goi | | | | Torra | , bui | | |
|-------------|-----------|-----------|------------|------------------------|------------|----------------|------------|-------------|------------------|-------------|---|
| 1 | - Osci in | ungement | | | | | Delete | Update | Change P | Add | |
| | User ID | User Name | Disabled I | Locked C | CreateTime | Last Login Tim | e Last Log | gin Address | Last Locked Time | Failed Cour | nt Permission |
| | 1 | admin | NO M | NO 2 | 014-04-29 | 2014-05-06 | 127.0.0.3 | 1 | 0001-01-01 | 0 | Manage experiment, Run experiment, View experiment, Use |
| | 2 | aaa | NO NO | NO 2 | 014-05-04 | | | | | 0 | Manage experiment, Run experiment, View experiment, Use |
| | 3 | DDD | NO | NO 2 | 14-05-04 | 1 | | | | U | |
| | | | | | | | | | | | |
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| c. change password | |
|--|--|
| Change password | |
| Old password: | |
| Confirm new password: | |
| | |
| OK Reset Cancel | |
| | |
| | |
| d. add user | |
| | |
| User name: | |
| Password: | |
| | |
| Confirm password: | |
| Permission: User manage Run experiment | |
| OK Cancel | |

2. Experiment Management

Experiment Management is used to manage experiment information and deleted experiment information.

2.1 Experiment Management

Click Service Experiment management Experiment management on the menu bar the user can:

- a. clear query condition
- b. set query condition
- c. query
- d. delete experiment
- e. download experiment
- f. edit experiment



2.2 Deleted Experiment Management Click Service ► Experiment Management ► Deleted Experiment Management on the menu bar

The user can:

a. clear query condition b. set query condition c. query d. delete experiment e. recover experiment f. clear experiment - -Delete experiment 查询条件 ----Clear Co ----Delete Clear Recover Experiment type Experiment name Executor Notes Serial No Start time End time Purpose Creator name Create time Updater name Update time

3. Template Management

Template Management is used to manage template information. Click **Service ► Template Management** on the menu bar The user can:

- a. download template
- b. delete template

| Template mangen | nent | | | | | | | X |
|-------------------|--------------------|------------------|------------|---------|--------|--|--|-----|
| | | | | Downloa | Delete | | | |
| Template category | Template name | Create user name | CreateTime | | | | | |
| 48 | 20140416_135107 | admin | 2014-05-04 | | | | | - 4 |
| 48 | 参考增益测量 | admin | 2014-05-04 | | | | | |
| 96 | 2 | admin | 2014-05-04 | | | | | |
| 96 | 20140428_155955 | admin | 2014-05-04 | | | | | |
| 96 | 3 | admin | 2014-05-04 | | | | | |
| 96 | 7项_20120522_123347 | admin | 2014-05-04 | | | | | |
| | | | | | | | | |

4. User Login

Click Service ► User Login on the menu bar

| User Login | × |
|----------------|-----------|
| Upper Service: | 127.0.0.1 |
| UserName: | admin |
| Password: | |
| ок | Cancel |

5. Change Password

Click Service ► Change Password on the menu bar

| Change password | |
|-----------------------|---|
| Old password: | |
| New password: | |
| Confirm new password: | |
| OK Reset Cancel | |
| | > |
| | |

6. See Running Experiment

See Running Experiment is used to see running experiment which is running on connected instrument.





Chapter 9 Tool Usage

1. Gain Setting

Instrument is the automatic gain version, and there is no need to set the gain manually.

2. Block Scan Method

There is no need to set Block Scan Method

3. Detector Library

The **Detector Library** tool is used to set up the inspection libraries of absolute quantitative, relative quantitative and SNP analysis.

Click Tools ► Detector Library ► (Absolute /Relative/SNP) ► open the following window

The user can:

- a. Add Detector
- b. Modify Detector
- c. Delete Detector

| R Detecto | r Library | | | | | | | |
|-----------|-----------|--------|------------|--------|-------|----------|--------------|-------|
| Add | Modify | Delete | | | | | | |
| Detector | Reporter | Color | Master Mix | Primer | Probe | Supplies | Batch Number | |
| Target1 | FAM | | | | | | | |
| Target2 | FAM | | | | | | | |
| | | | | | | | | |
| | | | | | | | | |
| | | | | | | | | Close |

4. Customized Dyes

The **Customized Dyes** tool is used to set up existing dyes and newly added dyes. Click **Tools** ► **Customize Dyes** ► open the following window The user can:

- e user can: a. Create dye
 - b. Modify dye name and channel
 - c. Delete dye
 - d. Move dye upward
 - e. Move dye downward

After adding new dyes or modifying dyes, the user should conduct crosstalk parameter measurements.

| AM 1 YYER 1 HEX 2 FET 2 TOE 2 TOE 2 YYS3 3 AMRA 3 FED 3 TOX 4 fexRed 4 Type MoveUp MoveUp MoveDown |)ye | Channel | |
|--|------------|-------------------|----------------------|
| SYER 1 HEX 2 TET 2 VIC 2 JOE 2 Cy3 3 TAWRA 3 VED 3 X0X 4 Cy5 5 .CRed 6 MoveUp MoveUp MoveUp | FAM | 1 | Dye |
| HEX 2 TET 2 TET 2 VIC 2 JOE 2 Cy3 3 TAMRA 3 VED 3 ROX 4 TexRed 4 Cy5 5 .CRed 6 MoveUp MoveDown | SYBR | 1 | FAM |
| TET 2 Channel VIC 2 1 (470nm -525nm) JOE 2 Delete JAMRA 3 Exp ROX 4 ExaRed Ly5 5 CRed CRed 6 MoveUp MoveDown MoveDown | HEX | 2 | Contraction in |
| VIC 2 JDE 2 JDE 2 JTAURA 3 ED 3 KED 3 KOX 4 IexRed 4 Cy5 5 .CRed 6 MoveUp MoveUp | TET | 2 | Channel |
| JOE 2 Cy3 3 IAMRA 3 NED 3 ROX 4 TexRed 4 Cy5 5 .CRed 6 MoveUp MoveUp MoveDown MoveUp | VIC | 2 | 1 (470nm -525nm) |
| Cy3 3 Delete TAMRA 3 | JOE | 2 | AL 9996-9450-0 |
| TAMRA 3 XED 3 ROX 4 TexRed 4 Cy5 5 .CRed 6 Movelip MoveDown | Cy3 | 3 | Delete |
| NED 3 ROX 4 TexRed 4 Cy5 5 JCRed 6 MoveUp MoveUp | TAMRA | 3 | |
| ROX 4 TexRed 4 Cy5 5 .CRed 6 MoveUp MoveDown | NED | 3 | 1 |
| TexRed 4 Cy5 5 JCRed 6 MoveUp MoveUp | ROX | 4 | 1 |
| Cy5 5 LCRed 6 MoveUp MoveDown | TexRed | 4 | |
| CCRed 6 MoveUp MoveUp MoveDown | Cy5 | 5 | 1 |
| MoveUp MoveDown | LCRed | 6 | |
| MoveDown | | 2.4 | MoveUp |
| MoveDown | | | |
| | | | MoveDown |
| | asurement. | du or mourry pure | accers for crosseare |
| asurement. | | | |

5. Customize Columns

Click Tools ► Customize Columns ► the following window will pop up

The user can:

- a. Add columns
- b. Delete columns
- c. Modify column name

| New Item | | | |
|----------|---|--------|--|
| | - | | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | | |
| | | Add | |
| | | Delete | |
| | | | |

6. Column Selection

The **Select Columns** tool is used to add the new columns in above section into current existing columns, or remove existing columns in current column.

Click **Tools** ► **Select Columns** ► the following window will pop up

1. Current existing column items include sample, report, report setting, query and query condition

2. Double click column can add or remove a column

3. Column with (*) indicates it cannot be removed



7. Sample Column Library

The **Sample Column Library** tool is used in the experiment design phase. The user can select the definition of contents in the drop-down box when setting up sample information.

Click **Tools** ► **Sample Column Library** ► the following window will pop up The user can:

- a. Add columns
- b. Delete columns
- c. Edit the columns content 4

| RE Sample Column Library | |
|--|------------------------------------|
| Auto Complete Co Auto Complete Columns | lumns Management Current Values |
| Nationality(Nationality) Diagnosis() Doctor(Doctor) Dept.(Department) Tester(Tester) Notes(Notes) | China |
| Add Delete | One Value per Line |
| | OK Cancel |

8. Instrument Calibration Parameters

The Instrument Calibration Parameters tool is used to calibrate the instrument parameters.

| Sel | ect Instrument 600254 |
|-------------------------|-----------------------|
| Baseline Parameters | Measured |
| Reference Gain Parame | Cy3,Cy5,FAM,HEX,ROX |
| Proportion Parameters | Measured |
| Crosstalk Correction Pa | Cy5,FAM,HEX,ROX,TAMRA |
| Crosstalk Gain Paramet | F1,F2,F3,F4,F5 |
| | |
| | |
| | |

Click Tools ► Instrument Calibration Parameters ► the following window will pop up

9. Measure Crosstalk Calibration Parameters

The Measure Crosstalk Calibration Parameters tool is used to measure crosstalk correction parameters.

Click Tools
Measure Crosstalk Calibration Parameters
the following window will pop up

The user can add and modify the channels to be tested and dyes according to his needs; upload corresponding reaction plates and operate the experiment. When the experiment is over, the system will automatically save the crosstalk correction parameters. Experiment Type: Absolute Experiment Name: Crosstalk Parameter Measurement

| | | 18-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1- | | | | | | |
|---------------|------------------|---|-----------------|--------------|---------|-------------------|--------------|---|
| Experiment I | Properties | _ | | | | _ | | |
| Experiment Na | ame: Crosstalk P | 'arameter Meas | urement | C | omment: | | | |
| User Na | ame: | | | | | | | _ |
| Detectors | Add Detector | Add Assay | Delete Detector | Delete Assay | Add | Detector From Lit | prary | |
| Detector | Reporter | Color | Master Mix | Primer | Probe | Supplies | Batch Number | |
| F1 | FAM | | | | | | | |
| F2 | HEX | - | | | | | | |
| | | | | | | | | |
| Reference D | ye | | _ | | | | | |
| | | | | | | | | |

10. Crosstalk Gain Parameter Measurement

The Crosstalk Gain Parameter Measurement tool is used to measure crosstalk gain parameters.

Click **Tools** ► **Measure Crosstalk Gain Parameters** ► the following window will pop up. The user can add and modify the channels to be tested and dyes according to his needs; upload corresponding reaction plates and operate the experiment. When the experiment is over, the system will automatically save the crosstalk gain parameters. Experiment Type: **Absolute** Experiment Name: **Crosstalk Gain Measurement**

| operiment Nam User Nam | e: Crosstalk (| Gain Measuren | nent | Cc | omment: | | |
|---------------------------|----------------|---------------|-----------------|--------------|---------|------------------|--------------|
| Detectors | Add Detector | Add Assay | Delete Detector | Delete Assay | Add | Detector From Li | brary |
| Detector | Reporter | Color | Master Mix | Primer | Probe | Supplies | Batch Number |
| F1 | FAM | | | | | | |
| F2 | HEX | at a | | | | | |
| | | | | | | | |
| Reference Dye | | | | | | | |

11. System Maintenance

The System Maintenance tools are used for system maintenance.

Click **Tools** ► **System Maintenance** ► the Password Input box will pop up ► input the correct Password ► conduct the following settings:

- a. Y-axis commissioning
- b. X-axis origin calibration
- c. Machine serial number setting
- d. Photomultiplier setting
- e. Runtime zero clearing
- f. Background measurement
- g. Reference gain measurement
- h. Fluorescence incremental calibration
- i. Firmware Upgrades

Firmware Upgrade tools are used to upgrade the firmware.

• Click **Tools** ► **System Maintenance** ► **Firmware Upgrade** ► the following window will pop up.

The user can:

- a. Select serial ports
- b. Select the BIN file to be upgraded
- c. Upgrade



| Firmware Upgrade | Σ |
|-------------------|---------|
| Choose Fort: COM1 | |
| Upgrade File | |
| E:\PCR.bin | Select |
| | Upgrade |

12. Upgrade Experiment File Format

The Upgrade Experiment File Format tools are used to convert old files with the suffix of .fqj or .fqs into new files with the suffix of .fqd.

Click **Tools** ► **Upgrade Experiment File Format** ► the following window will pop up.

The user can:

- a. Add files to be upgraded
- b. Remove selected files
- c. Select the output directory of new files

| a | Ingrada | |
|----|---------|--|
| u. | opgrade | |

| 🔜 Upgrade fqj/fqs file | |
|--|-------------------|
| Upgrading files | |
| File | State |
| | |
| Add upgrading files Remove selected files Output directory | Browse Upgrade |

13. Ta Calculator

Click **Tools** ► **Ta Calculator** ► the following window will pop up.

Input Forward Primer and Reverse Primer, click Calculate to gain Forward Tempperature Reverse Tempperature Average Tempperature and Anneling Tempperature.

| Forward Primer Reverse Primer |
|--|
| Forward Temperature C Reverse Temperature C Average Temperature C Anneling Temperature C |
| Calculate Close |
| |
| |
| |
| 95 |
| |

Chapter 10 Other Functions

1. Instrument Operation

The Instruments operations include **Connect** instrument, **Disconnect** instrument and **Instrument Information**.

1.1 Connect

Click **Instrument** Connect select port number or select automatic port matching.



When the instrument is connected, the icon on the status bar will be status if the instrument

is disconnected, the icon on the status bar will be

1.2 Disconnect

Click Instrument Disconnect disconnect currently connected instrument

1.3 Instrument Information

When the instrument is connected, the user can check the instrument information.

Click Instrument lnformation be the following dialog box will pop up

Instrument information includes instrument serial number, runtime, currently connected ports, and whether an experiment is in operation.



2. Data Query

Data Query is used to query the data already exported to the database.

Click Data Summary ► Data Query ► the following window will pop up

The user can:

a. Select database files

| c. Query d. Clear all query conditions |
|---|
| d Clear all query conditions |
| |
| (1) Data Gaorge Carlos |
| Path: Bromar |
| Deary Condition |
| |
| |
| |
| Oury Sealt |
| File Name Sample da Sample Name Test Item Name Ser Age Case No. Outpatient No. Hed No. Notionality Sampling Time Diagnosis Doctor Dept. Test Result I |
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3. System Help Click Help ► Help Topics
Chapter 11 Maintenance

1. Regular cleaning

In order to ensure the normal operation and test use of the instrument, it is suggested to clean the instrument regularly.

Surface cleaning: use a soft cloth to clean; If necessary, dip in alcohol, distilled water or clean paste to clean;

Cleaning of module hole: clean cotton swabs with dust-free, and dip a small amount of 95% medical anhydrous ethanol or distilled water when necessary.

Warning !

1. When cleaning the instrument, the power must be cut off.

2. Corrosive cleaning agent is strictly prohibited on the surface of this instrument. If in doubt, please consult the manufacturer or its agent.

| S.N. | Fault Phenomenon | Cause Analysis | Way of Handling |
|------|--|--|---|
| 1 | The power switch behind the instrument has been set ON, but the instrument is not responsive | The RUN SWITCH in front of the instrument is not pressed. | Press RUN SWITCH. |
| 2 | System parameters menu shows that "password" needs to be entered. | The system parameters are used for internal calibration of the instrument manufacturer and need special pass word to enter. | Users do not need to use this feature. For calibration, please contact the manufacturer's service personnel. |
| | The rising and | The vent is blocked | Remove obstructions from vents |
| 3 | module obviously slows down or the temperature control is inaccurate | Loose connection Refrigeration piece is damaged Fan is damaged or doesn't work Temperature sensor is damaged | Contact with the supplier or manufacturer |
| 4 | The modules are | Internal instrument fault | Contact with the supplier or |
| | nor cooled | Refrigeration piece is damaged | manufacturer |
| | | Hot cover heating process | When the hot cover temperature of the instrument reaches the target value. The module temperature is automatically controlled at 30° C when it stops running |

2. Analysis and Troubleshooting

| 5 | Temperature or Fluorescence Curve Exception: straight line, | The Run Program was infected with a virus and the computer CPU was severely occupied. | After antivirus reinstall the application software. |
|----|---|--|---|
| 6 | The hot cover won't heat | Thermal fuse is damaged Connector is loose The heating element in the hot | Contact with the supplier or manufacturer |
| | | cover is damaged Temperature sensor in hot cover is damaged | |
| 7 | The fluorescence value of each hole increased or the background was very large without test tube | Contamination of test tube holes or hot covers; Baseline background parameters are misused. | depollution, each instrument should correspond to BaselineFile. After long-term use, the optical element is offset. Please contact the manufacturer to recalibrate the background. |
| 8 | Reagent evaporation | Tube quality problems, loose seal; Tube cover or film is not correct, not appropriate. | Select suitable consumables with better sealing performance |
| 9 | Signal crosstalk between channels | There is crosstalk between dye signals in different channels objectively. | It can be measured by "crosstalk coefficient measurement" function in the software, and the calibration parameters can be saved for correction. |
| 10 | Abnormal fluorescence | External strong light irradiation | Turn off external light source Contact with the supplier or |
| | detection values | Photovoltaic system is damaged | manufacturer |

Caution: During the warranty period, users are forbidden to open the expansion instrument shell for self-inspection. If there is a fault in the above table that requires the instrument shell to be opened for inspection,, timely contact with the supplier or manufacturer. Users are strictly forbidden to inspect or replace parts without permission. Only manufacturers or agencies can inspect or provide parts.



Note:



