



# CFX Real Time PCR System

## Instrument Guide



**正茂生物科技股份有限公司**  
**Genmall Biotechnology Co., Ltd.**

TEL: (02) 2796-0803 FAX: (02)2796-0833

免費諮詢服務及訂貨專線：0800-045-168

# CFX Maestro software user guide

The CFX series is operated with CFX Maestro software.

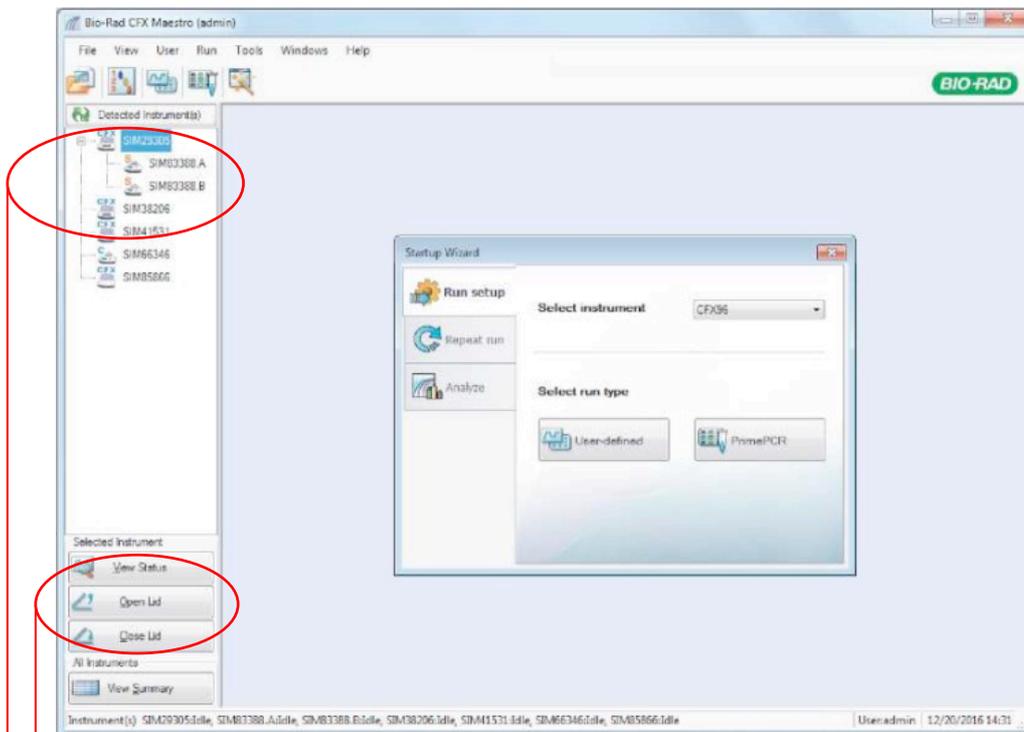
It is recommended to install on Windows 10 (64-bit) for operation.



## Software operation

- ✓ After connecting to the instrument, switch on the computer at first and power on the instrument. Then, click and initiate software. **Notification:** After the detection was finished, turn off the instrument in reverse Maestro order.
- ✓ **Confirmation of the software connection**
  1. Checking the connection status
  2. Instrument can be operated via 「 open lid 」 as well as 「 close lid. 」 ( Fig.1 )

Fig.1



→ Confirm that the instrument status is Idle, and you can click Open lid and Close lid to control the instrument to open and close the lid.

→ The instrument serial number will be displayed after the software is connected.

- ✓ Click **User-defined** in **Startup Wizard** to set the experimental protocol (Fig. 1)

## Three step for setting program <Protocol→Plate→Start Run>

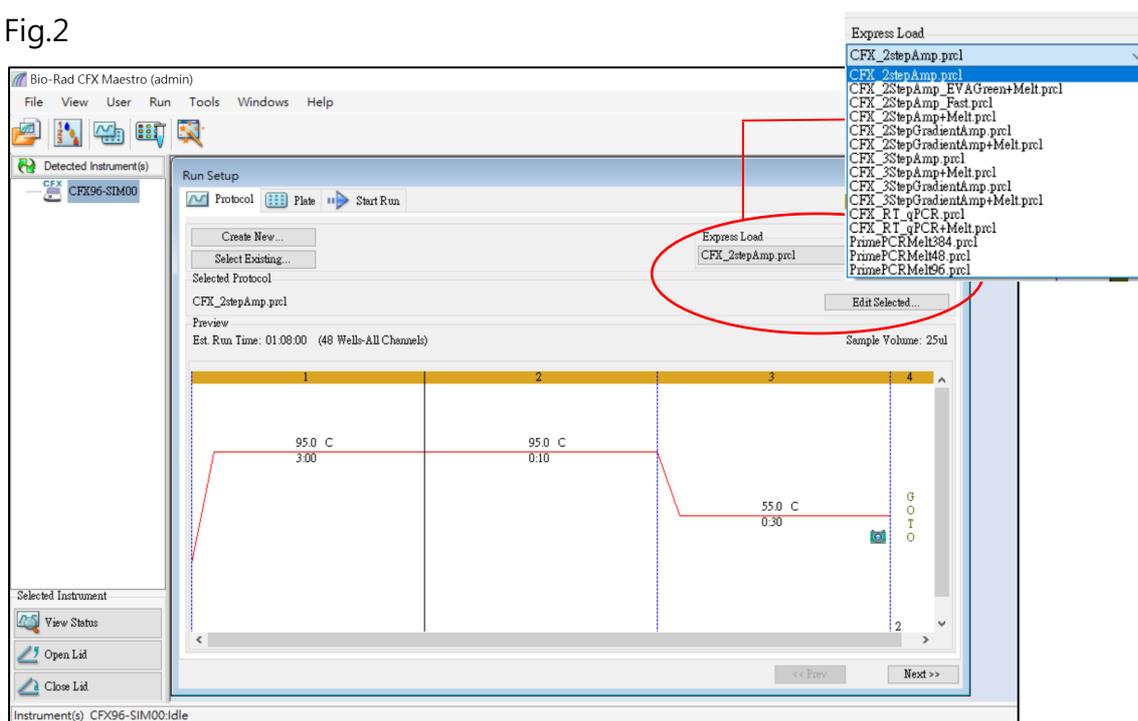
- ✓ Step1. Set up the protocol · Adjust reaction temperature and time base on experimental design.

1. Select 「 Express Load 」

Notification: Commonly used protocols have been built in the software (Fig.2)

2. Select a built-in protocol template, then click 「 Edit Selected 」 at below to perform edition or modification (Fig.2)

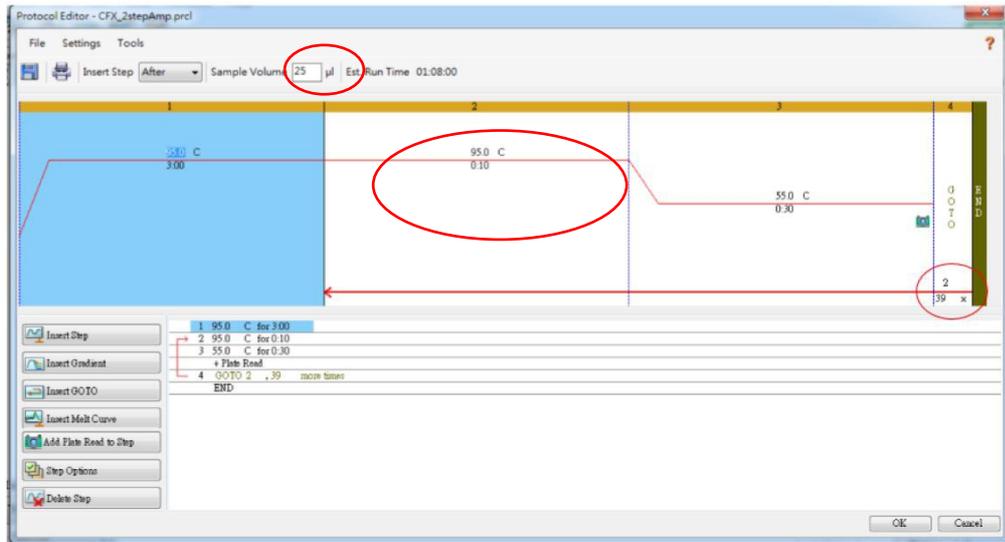
Fig.2



3. Protocol edition and modification

- Reaction temperature, reaction time and sample volume can be directly edited and modified (Fig.3)
- Cycle numbers are able to be modified through 「 GOTO 」 , which indicates anticipatable cycle numbers must to be 「 - 1 」 . (Fig. 3)
- (Ex: 40 cycles will key in 39)

Fig.3



Protocol setting can be adjusted on left toolbar menu at the bottom of window (Fig.4).

Fig.4

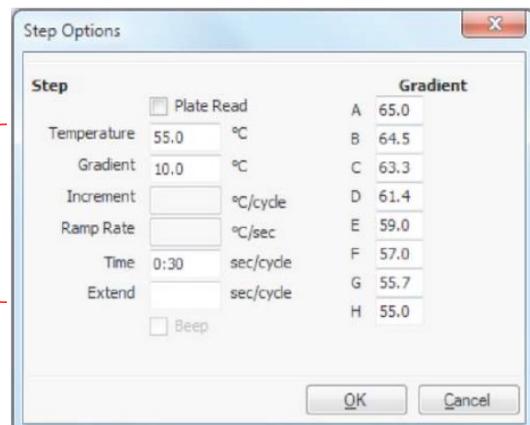


- Insert Step: inserts a step before or after the selected
- Insert Gradient: inserts a gradient step based on the type of well block
- Insert GOTO : inserts a cycling (loop) step
- Insert Melt Curve : inserts a melt curve read
- Add Plate Read to Step : adds plate read command to the selected step. \*\*
- Step Options : displays the options available for the selected step
- Delete Step: deletes the selected step from the protocol.

\*\* Tip: After you add a plate read command to a step, the button changes to Remove PlateRead when you select the step.

Fig.5

- Sets the target temperature
- Sets the gradient range (1–24°C)
- To increase (or decrease) the temperature
- The ramp rate for the selected step

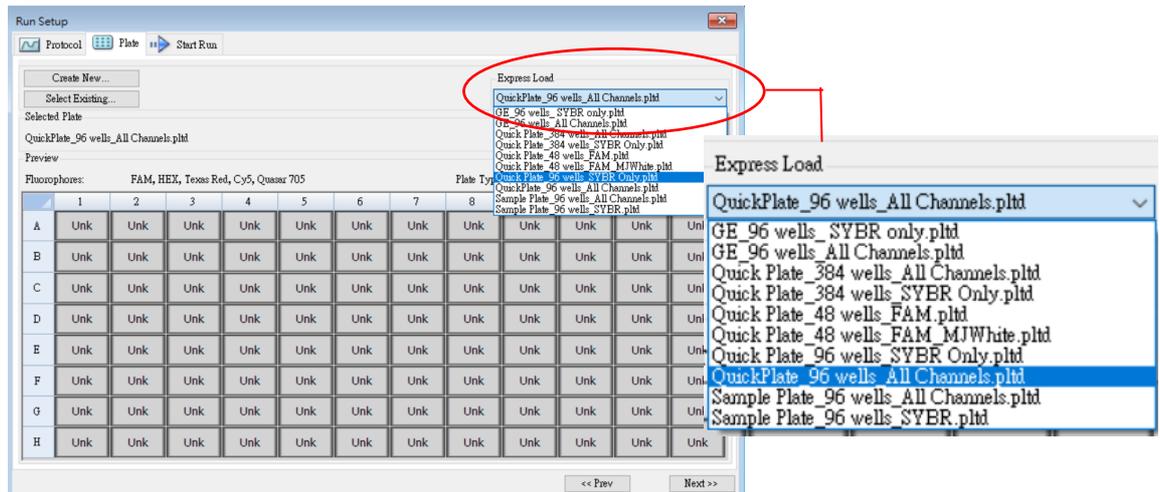


- After modification of the temperature time step, save it and click 「 Next 」 .

✓ Step2. Select fluorescent channel module

On the 「 Plate 」 tab site , click “Express Load” , choose a built-in protocol and select the corresponding fluorescent channel module for the experiment. (Fig.6)

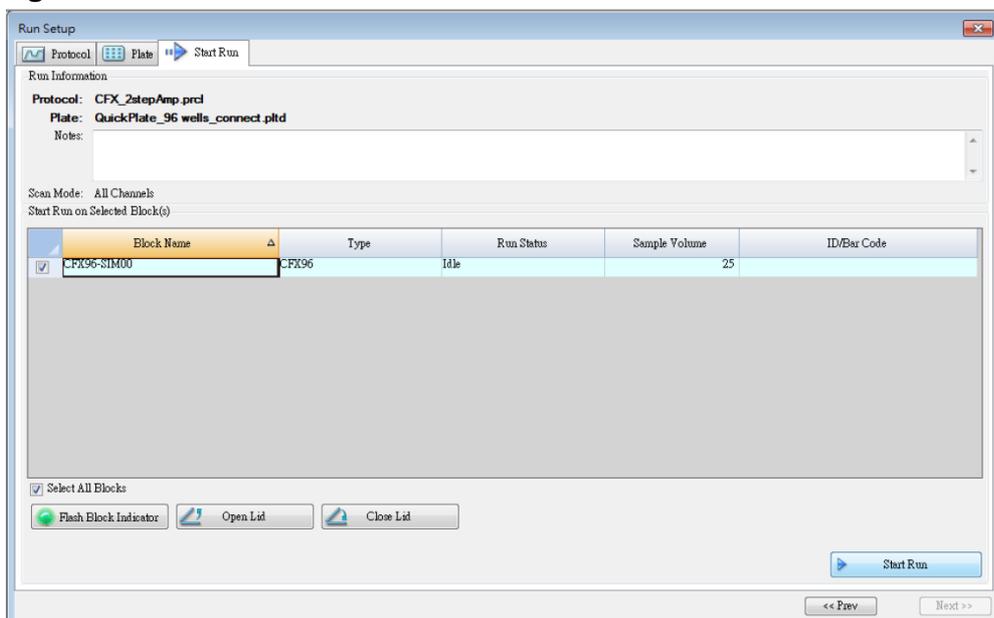
Fig.6



✓ Step2. : Start Run

- After confirming the serial number, status and volume of the instrument. Then, you can click 「 Start Run 」 to start the experiment (Fig.7)
- Save the experimental data in personal folder and the analysis data will be label with “pcrd” file.

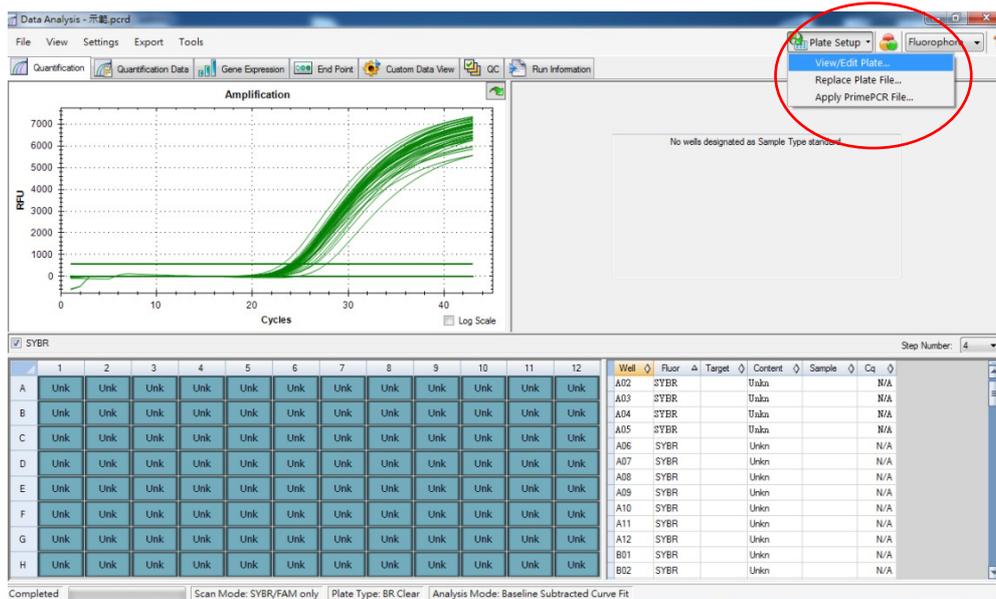
Fig.7



## Data analysis

- ✓ After the experiment was finished, Click on 「Plate Setup」 on the upper right page and select 「View Edit Plate」 (Fig. 8) to enter into the Plate Editor. On the page of the plate, select the area at first and input or edit the sample name stepwisely from right to left.

Fig.8



圖九

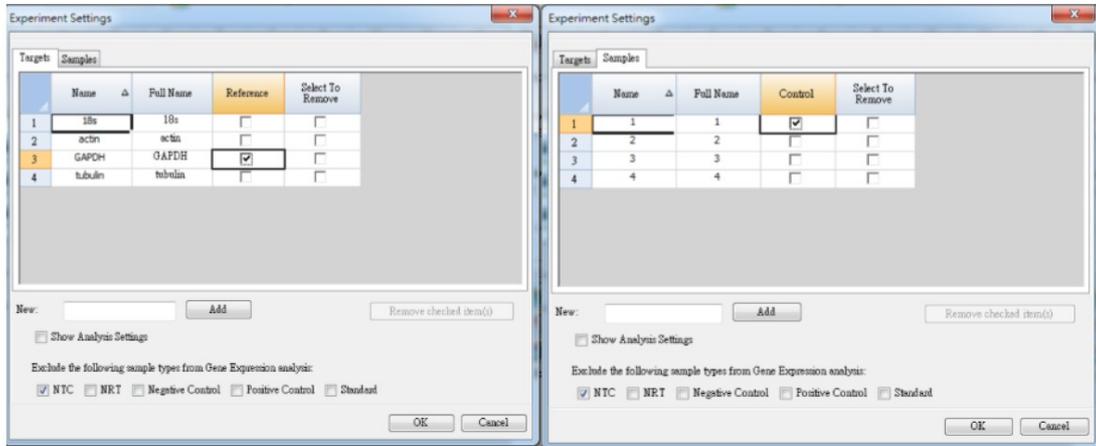
The 'Plate Loading Guide' dialog box is shown. It has several sections: 'Select Fluorophores...' with a button; 'Sample Type' set to 'Unknown'; 'Target Names' with a list of targets (FAM, HEX, Texas, Cy5, Quasar) and their corresponding wells; 'Sample Names' with a field for sample identifiers; 'Biological Group' with a field for group identifiers; 'Replicate #' with a field for the number of replicates; and 'Experiment Settings...' with buttons for 'Clear Replicate #' and 'Clear Wells'. At the bottom are 'OK' and 'Cancel' buttons.

- Select the requisite fluorophores for the plate:
- Select sample type (unknown, standard, NTC, PC,
- Targets of interest (genes or sequences) in each loaded well.
- The identifier or condition that corresponds to the sample.
- The identifier or condition that corresponds to a group.
- Enter repeatable sample settings (Horizontal / Vertical)
- Setting reference gene and control sample (Fig.10)
- Remove all of Replicate
- Clear all well

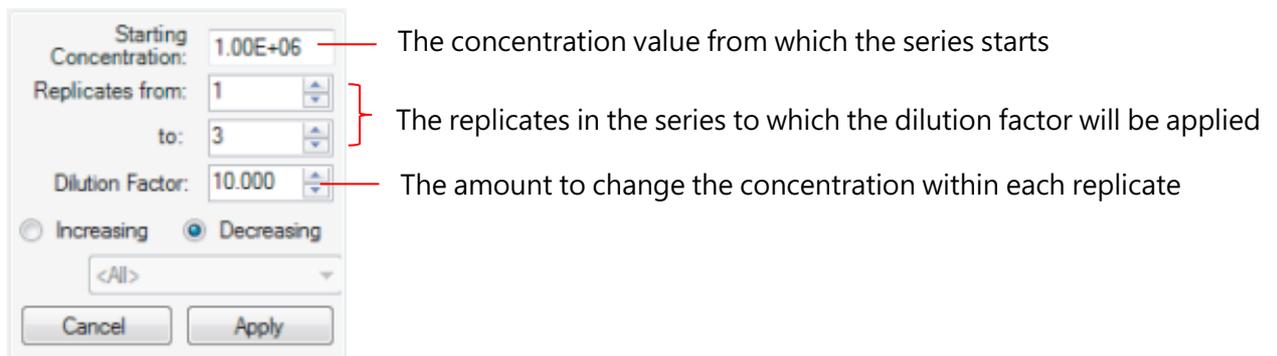
Fig.10

選擇 Reference gene

選擇 Control Sample



\*\* When the sample is spiked with a standard of known concentration, enter the reaction concentration

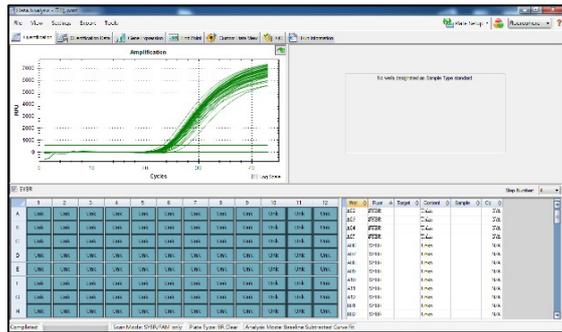


✓ Press 「ok」 to process the data analysis page after setting the Plate.

✓ Data Analysis and Application tool

Quantification

Amplification curves can be observed along with their Cq values



Quantification Data

The raw data for this experiment is presented, including thresholds and mean values for each sample and standard deviation of replicates.

Sample	Cq	Mean	SD
1	25.17	28.21	0.443
2	25.29	28.20	0.449
3	25.41	28.21	0.446
4	24.76	27.25	0.279
5	24.66	27.1	0.279
6	25.29	28.21	0.443
7	25.41	28.21	0.446
8	25.29	28.21	0.443
9	25.41	28.21	0.446
10	25.29	28.21	0.443
11	25.41	28.21	0.446
12	25.29	28.21	0.443
13	25.41	28.21	0.446
14	25.29	28.21	0.443
15	25.41	28.21	0.446
16	25.29	28.21	0.443
17	25.41	28.21	0.446
18	25.29	28.21	0.443
19	25.41	28.21	0.446
20	25.29	28.21	0.443
21	25.41	28.21	0.446
22	25.29	28.21	0.443
23	25.41	28.21	0.446
24	25.29	28.21	0.443
25	25.41	28.21	0.446
26	25.29	28.21	0.443
27	25.41	28.21	0.446
28	25.29	28.21	0.443
29	25.41	28.21	0.446
30	25.29	28.21	0.443

Melt Curve Data

Observe the melting curve and its Tm value



Gene Expression

Observe the relative expression of genes



End Point Analysis

Present data analysis data of the end point

Well	Target	Content	Sample	End Point	Cq
A01	Actin	Uln-01	M1	7890	
A02	Actin	Uln-02	M1	7294	
A03	Actin	Uln-03	M1	7058	
B01	Actin	Uln-10	M2	7465	
B02	Actin	Uln-11	M2	8122	
B03	Actin	Uln-12	M2	7955	
C01	Actin	Uln-19	M3	6903	
C02	Actin	Uln-20	M3	6674	
C03	Actin	Uln-21	M3	8406	
D01	Actin	Std-01	db-1	2274	
D02	Actin	Std-02	db-2	3056	
D03	Actin	Std-03	db-3	5841	
D04	Actin	Std-04	db-4	8516	
D05	Actin	Std-05	db-5	10142	
D06	Actin	Std-06	db-6	11469	
E01	Actin	Std-01	db-1	2456	
E02	Actin	Std-02	db-2	4232	
E03	Actin	Std-03	db-3	6571	
E04	Actin	Std-04	db-4	8068	
E05	Actin	Std-05	db-5	11215	
E06	Actin	Std-06	db-6	12542	
F01	Actin	Std-01	db-1	2269	
F02	Actin	Std-02	db-2	4037	
F03	Actin	Std-03	db-3	6313	
F04	Actin	Std-04	db-4	8468	

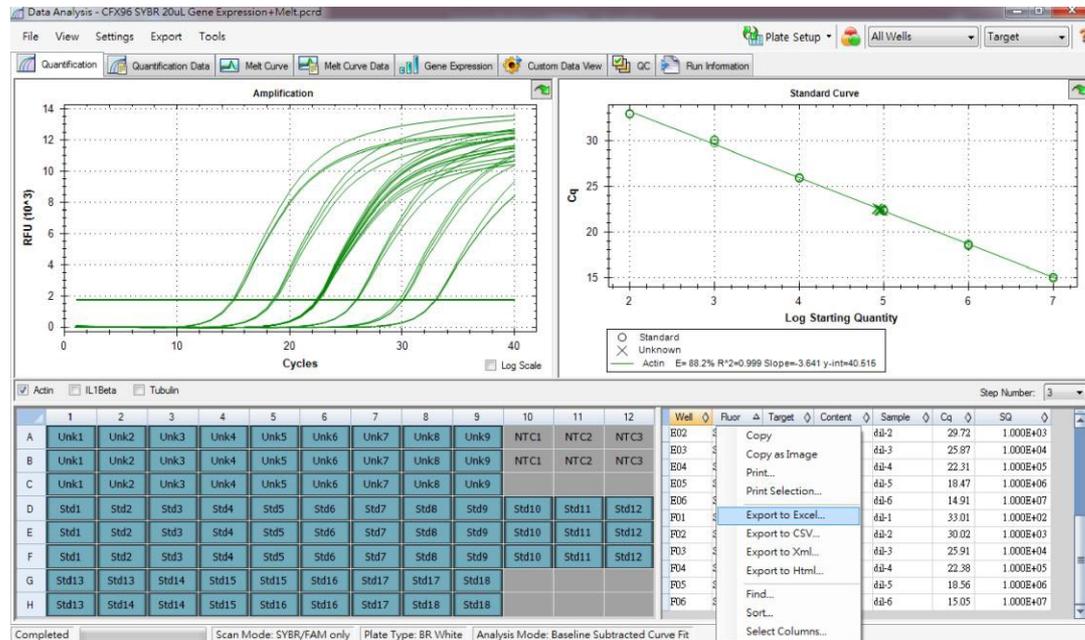
Quality Control

Provide evaluation information

Description	Value	Use	Results	Exclude Wells
Negative control with a Cq less than	38	<input checked="" type="checkbox"/>		<input type="checkbox"/>
NTC with a Cq less than	38	<input checked="" type="checkbox"/>		<input type="checkbox"/>
NRT with a Cq less than	38	<input checked="" type="checkbox"/>		<input type="checkbox"/>
Positive control with a Cq greater than	30	<input checked="" type="checkbox"/>		<input type="checkbox"/>
Unknown without a Cq	N/A	<input checked="" type="checkbox"/>		<input type="checkbox"/>
Standard without a Cq	N/A	<input checked="" type="checkbox"/>		<input type="checkbox"/>
Efficiency greater than	110.0	<input checked="" type="checkbox"/>		<input type="checkbox"/>
Efficiency less than	90.0	<input checked="" type="checkbox"/>	Actin	<input type="checkbox"/>
Std Curve R <sup>2</sup> less than	0.980	<input checked="" type="checkbox"/>		<input type="checkbox"/>
Replicate group Cq Std Dev gre	0.20	<input checked="" type="checkbox"/>	Actin.D2, E2, F2, IL11B	<input type="checkbox"/>

## Save file

To save Excel, right-click and select 「Export」 for Excel form. If you want to save image file, you can choose 「Save Image」 as to save it as a JPG file.



## Output as PDF

Select 「Report」 in Tools on the upper toolbar. You can customize the report and save as a PDF file.

