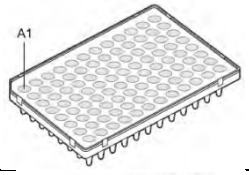
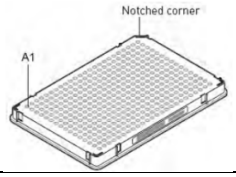
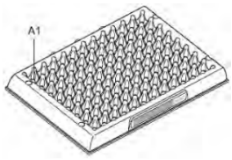
QUANTSTUDIO 7 FLEX REAL-TIME PCR SYSTEM GUIDELINES FOR TRAINED USERS

Access only by authorized and trained persons with a booked slot time in iLab [OncPat Core Facility QS7-PCR]

INSTRUMENT CONSUMABLES

Use only the consumables appropriate for the sample block for which you have received the introduction.

Table.1 - Authorized Consumables

Sample Block	Consumables	
96-well plate, **0.2 mL**		▪ MicroAmp™ Optical Adhesive Film ▪ MicroAmp™ Optical 96-Well Reaction Plate ▪ MicroAmp™ Optical 96-Well Reaction Plate with Bar Code
384-well plate		▪ MicroAmp™ Optical Adhesive Film ▪ MicroAmp™ Optical 384-Well Reaction Plate
TaqMan™ Array Card		▪ TaqMan™ Array Card

IMPORTANT! The Fast 96-Well (0.1mL) PCR Plate: Do NOT use this on the QS7.
Please do not use rounded caps for 0.2 mL tubes, as they can cause damage to the heated cover.

IMPORTANT! Properly seal the reaction plate.
Adhesive side facing the plate: Make sure that the film completely covers all wells of the reaction plate.

Apply pressure to the optical film during application to ensure a tight, evaporation-free seal.

Cleanly remove each tab along the precut dotted line.

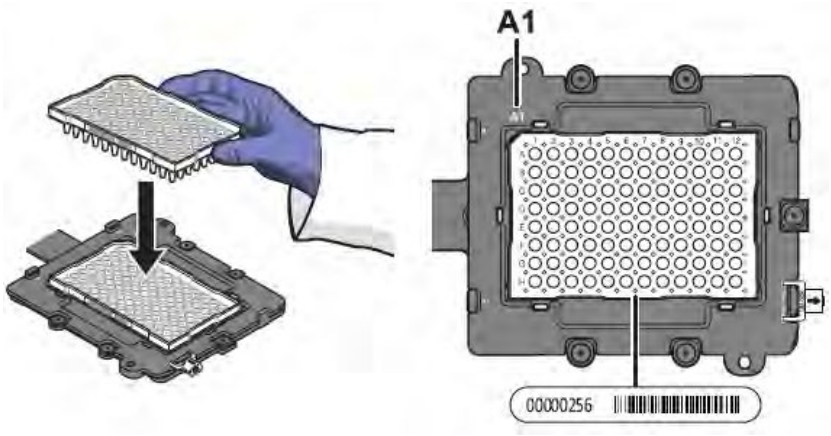
You must remove any excess glue from the plate to avoid the plate being stuck inside the Machine.

If necessary, use a lint-free wipe to remove all excess glue from around the perimeter of the adhesive film.

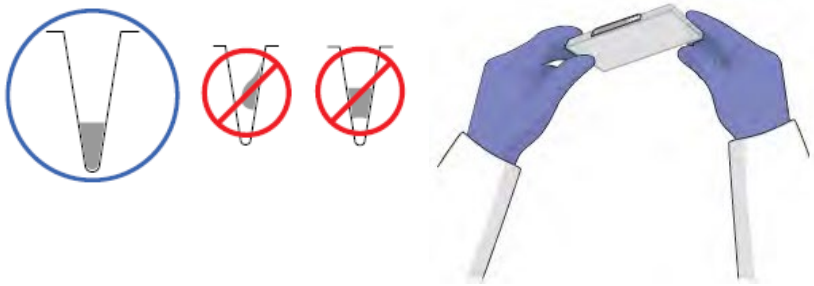
LOADING THE PLATE INTO THE PLATE HOLDER

Instrument in standby, tap the QuantStudio™ 7 Flex Real-Time PCR System touchscreen to activate it, then tap In the right lower corner of the main menu, tap to open the side door and load the appropriate plate.

- Ensure that the consumable is properly aligned in the holder:
  - Well A1 of the plate or array card is in the top-left corner of the adapter.
  - The barcode faces the front of the instrument



IMPORTANT! The reaction plate must be well mixed and centrifuged.
Centrifuge the reaction plate for 2 minutes at < 1500 rpm.
Confirm that the liquid is at the bottom of each well of the reaction plate.
If not, centrifuge the reaction plate again at a greater rpm and/or for a longer period of time.



Do not allow the bottom of the plate to become dirty.
Fluids and other contaminants that adhere to the bottom of the reaction plate can contaminate the sample block, causing an abnormally high background.

For any Questions & Inquiries
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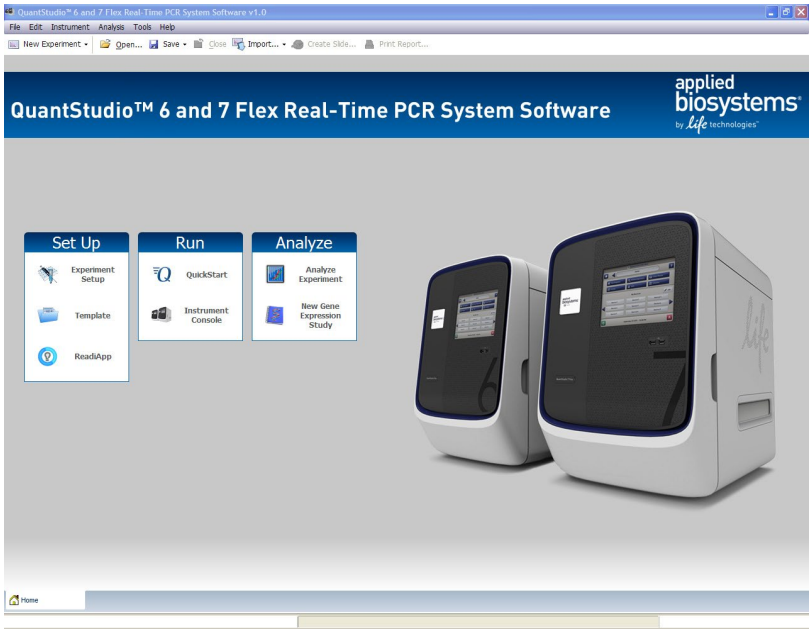
Double-click  (QuantStudio™ 6 and 7 Flex Real-Time PCR System Software shortcut) to access the home screen, shown in the following image.



EXPERIMENT SETUP

Experiment: QS6_QuantStudio_384-Wel... **Type:** Standard Curve **Reagents:** TaqMan® Reagents

How do you want to identify this experiment?

* Experiment Name:	QS6_QuantStudio_384-Well_Standard_Curve_Example	Comments:	Standard Curve Example
Barcode:			
User Name:	Example User		

*** Which instrument type are you using to run the experiment?**

✓ QuantStudio™ 6 Flex System | QuantStudio™ 7 Flex System

*** Which block are you using to run the experiment?**

✓ 384-Well | 96-Well (0.2mL) | Fast 96-Well (0.1mL)

*** What type of experiment do you want to set up?**

✓ Standard Curve | Relative Standard Curve | Comparative CT (ΔΔCT) | Melt Curve

High Resolution Melt | Genotyping | Presence/Absence

*** Which reagents do you want to use to detect the target sequence?**

✓ TaqMan® Reagents | SYBR® Green Reagents | Other

*** What properties do you want for the instrument run?**

Standard | ✓ Fast

What is the reagent information?

New Delete

Type	Name	Part Number	Lot Number	Expiration Date
Master Mix	TaqMan Fast Universal PCR Master Mix	4984571	1206155	12-31-2013

SET EXPERIMENT

From the Home tab, select **Experiment Setup**, then complete the setup screens.

Define the experiment properties

1. From the Experiment Properties screen, enter information identifying your experiment:

a. In the **Experiment Name** field, enter up to 100 characters to uniquely identify the experiment.

b. (Optional) In the **Barcode** field, enter the barcode of the plate or array card you are using to run the experiment.

c. In the **Username** field, enter your **First and Last Name**
(To be able to be easily contacted when needed)

d. (Optional) In the **Comments** field, enter up to 2000 characters to associate with the experiment.

2. Select the instrument type you are using to run the experiment: **QuantStudio™ 7 Flex System**

3. Select the block type you are using to run the experiment: **384-Well**, **96-Well (0.2mL)**

IMPORTANT! Fast 96-Well (0.1mL) not available (Do not select)

4. Select the type of experiment to set up: **Standard Curve**, **Relative Standard Curve**, **Comparative CT (ΔΔCT)**, **Melt Curve**, **High Resolution Melt**, **Genotyping**, or **Presence/ Absence**.

5. Select the reagent you are using to detect the target sequence: **TaqMan® Reagents**, **SYBR® Green Reagents**, or **Other**.

6. Select the run properties:

- Select the ramp speed for the experiment: **Standard** or **Fast**.
- (Optional) If you selected:

– Melt Curve or High-Resolution Melt as the experiment type, then you have the option of including a PCR stage for that experiment.

– Genotyping or Presence/Absence as the experiment type, then you have the option of including a pre-PCR Read and Amplification stage for that experiment.

– SYBR® Green as the reagent, then you have the option of including a melt curve for that experiment.

7. (Optional) In the reagent information panel, click **New** to Add a row for data entry, then enter the detailed information (including the **Part Number**, **Lot Number**, and **Expiration Date**) of the reagents you will use in your experiment.

8. Define the targets, samples, biological replicates, passive reference dye, and controls.

9. Assign the targets, samples, biological replicates, and Controls.

Note: Select the detection task for the target from the **Task** drop-down menu. Available tasks include **Unknown**, **Standard**, **Negative Control**, **Positive Control**, **Custom**, and others, depending on the experiment type.

10. Run method

From the Experiment Menu, select **Run**.

-Enter the **Reaction Volume per Well** to use to run the experiment:

- 96-Well plate: **1-200 µL**
- 384-Well plate: **1-30 µL**
- Array Card: **1 µL**

-Review the information in the **Graphical View** tab and edit the run method as needed:

- Add and delete steps or stages
- Edit the time, temperature, or ramp rate for a step

Start running by selecting the machine code:

278872441

IMPORTANT!

After running your plate, wait for 15 minutes for the machine to cool down, then remove and discard the reaction plate in the yellow bin below the tabletop centrifuge

Data Storage and Export

Make sure to save the experiment and export the data to the Folder of your Research Group.

Always check the Folder path before exporting data.

Note: Export the data as an **.xlsx** file.

For any Questions & Inquiries
E-mail Us!
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Adaptation from the following sources

- *QuantStudio™ 6 and 7 Flex Real-Time PCR Systems (v1.3) Maintenance and Administration Guide*
- *QuantStudio™ 6 and 7 Flex Real-Time PCR Systems Quick Reference*
- *Getting Started with QuantStudio™ 6 and 7 Flex Real-Time PCR System Software Experiments*