<u>SOP – Thermal Cyclers (Bio-Rad)</u>

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Equipment Overview

The Bio-Rad thermal cyclers are touchscreen devices used for PCR experiments to amplify RNA or DNA sequences. However, other experiments can be programmed as long they can fit into the typical 0.2 mL tubes (T100) or plates (96-/384-well plates). There are 3 types of instruments at the facility:

Instrument	Sample types	Features	Max Ramp Rate
CFX96	96-well plate	Incubation, real-time PCR with 6 filtered LEDs (450-730 nm)	5°C/sec
CFX384	384-well plate	Incubation, real-time PCR with 5 filtered LEDs (450-690 nm)	2.5°C/sec
T100	0.2 uL tubes, 96 well-plate	Incubation, PCR	4°C/sec

Notes before use:

- Ensure plate is clean and dry before placing it into the thermocycler
- Only use 70% ethanol for cleaning to prevent corrosion from chemicals.
- The temperature range 4–100°C.
- Book the instruments ahead of time on LMACs.
- Warning: the heating block and lid can be hot during operation.
- Below is the list of fluorophores available for the CFX96/CFX384:



Select Fluoropho	res		
Channel	Fluor	Selected	Color
1	FAM	Г	
	SYBR	7	
	Cal FL Gold		
	Cal FL Red		
	SBG1		
2	HEX		
1	TET		
	Cal Gold 540		
	VIC		1
	TexasRed		
3	ROX	Г	
	Texas Red		
	Cal Red 610		
4	Cy5		
	Quasar 670		
5	Quasar 705		1
			OK Cancel

Supplies and Reagents

Provided by CRAFT:

- 70% Ethanol
- Kimwipes
- PPE (lab coat, gloves)
- Plastic insert (for tubes on the T100) one side is for tubes with <u>dome</u> caps, the other is for <u>flat</u> caps.





Method

Setup:

- 1. Log into LMACs to turn on the thermal cycler.
- 2. On the home screen, select **Saved Files**:



3. Create a folder for your lab (le. Professor's surname, PI) under CT003109:





User: ADMIN	Saved Files		Jan	Jan 2, 2010 00:12	
Locations	Folders	Files	Preview		
Recent	<root></root>	TEST	Method: C Lid: 105°C	Calc C	
стооз109	Bio-Rad PCR	*	Volume: 30µl		
Real-Time Data	Bio-Rad gPCR		1.95°C, 3 2.95°C, 0	1. 95°C, 3:00 2. 95°C, 0:10 3. 55°C, 0:30 4. Plateread 5. Goto 2, 39X	
	CRAFT		3. 55°C, 0 4. Platere		
	LAB PI		5. Goto 2,		
	РК				
Constanting of the Constant		Anteren	A.C.		
	+ 🔅	2°°	<u>∧í</u>		
Home New	Folder Folder Options	File Options	Edit	Run	

4. Once a folder has been created, click the **Home** button, and select **New Protocol**.



5. Create a protocol for your assay.

6. Click **Save** and select the designated folder (Ie. Professor's surname, PI):





7. Click **Run** when ready to start the experiment.

8. After the experiment is finished, ensure to <u>cancel</u> or <u>stop</u> the run before shutting down the system.

9. Export the data by selecting **Export** and plugging an external drive to the USB A port:



10. Remove your sample, and log out of LMACs.

Cleanup:

1. Close the software.





2. Wipe the workstation with RTU or 70% ethanol.

Troubleshooting:

- 1. If the touchscreen is frozen, restart the instrument.
- 2. If issues cannot be resolved, contact a CRAFT staff member.

Resources:

T100 specifications: <u>https://www.bio-</u> rad.com/webroot/web/pdf/lsr/literature/Bulletin 6060.pdf

CFX96 specifications: <u>https://www.bio-</u> rad.com/webroot/web/pdf/lsr/literature/Bulletin_6093.pdf

CFX384 specifications: <u>https://www.bio-</u> rad.com/webroot/web/pdf/lsr/literature/Bulletin_6096.pdf

User Manual (CFX96/CFX384): https://www.bio-rad.com/en-ca/node/39981



