

Bauer Core Standard Protocol		
Title: Operating the MJ Tetrad or DNA Engine Thermal Cyclers		
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Author(s): Claire Reardon	Reviewers: Christian Daly	
Contact: claire@cgr.harvard.edu	Comment:	

1. Purpose

This protocol describes how to use MJ Research Tetrad or MJ DNA Engine thermal cyclers. The protocol is designed as a reference and is not a substitute for training. Users must complete a training session before using any of the Bauer Core's instrumentation.

2. Materials

- 2.1. PCR reaction in appropriately sized reaction vessel
 - 2.1.1. The Bauer Core has Alpha Units (thermal blocks) that are compatible with:
 - 2.1.1.1. 96 well plates (e.g. VWR # 82006-636) or 0.2 ml tubes (e.g. VWR # 20170-010)
 - 2.1.1.2. 384 well plates (e.g. VWR # 82006-678)
 - 2.1.1.3. 0.5ml tubes (e.g. VWR # 62111-500)
- 2.2. Caps (e.g. VWR # 20170-000) or sealers (eg. VWR # 82006-692).

3. Instrumentation

- 3.1. MJ Research thermal cycler (Tetrad or DNA Engine)
- 3.2. Alpha Unit thermal block (96-well, 384-well, or 30/30-well dual block)

4. Reagent preparation

- 4.1 Prepare PCR reaction
(see "PCR protocol" for an example of a PCR reaction for amplifying DNA for microarrays).

5. Procedure

- 5.1. Startup
 - 5.1.1. Determine if there is a block available using the display on the instrument.
 - 5.1.1.1. If a block is in use, the light next to the block number will be on.
 - 5.1.1.2. For 30/30 dual units, check to see if the A or B side is in use.
 - 5.1.2. Install a block if the right type of block is not already installed.
 - 5.1.2.1. Make sure there are no programs currently running on the unit.
 - 5.1.2.2. Turn off the power.
 - 5.1.2.3. Pull up and out on the handle on the back of the alpha unit to remove it.

- 5.1.2.4. Slide the new unit in and clamp the handle down.
- 5.2. Creating a new Method
 - 5.2.1. Hit the “Block” button until the main menu comes up.
 - 5.2.2. Hit the “Select” button until the “Enter” function is highlighted then hit “Proceed”.
 - 5.2.3. Create a name for the protocol.
 - 5.2.3.1. Use the “Select” button to chose a letter and “Proceed” to advance.
 - 5.2.3.2. Hit “Proceed” again once the name is entered.
 - 5.2.4. Choose the method type.
 - 5.2.4.1. Calculated methods take the sample volume and reaction vessel type into account to accurately set the sample temperature.
 - 5.2.4.2. Block methods simply set the temperature of the block as programmed. Block methods require longer cycle times since the sample temperature lags behind the block temperature.
 - 5.2.4.3. Probe methods are not available in the Bauer Core.
 - 5.2.5. Enter the program’s steps.
 - 5.2.5.1. Use “Select” to highlight the type of step you want to add
“Temperature” incubates the sample at the set temperature for the set time
“Gradient” allows the user to test a range of temperatures for a step. The user sets one temperature on the right-most column of the block and another on the left. The instrument fills in the inner columns with a range of temperatures between the two extremes, Use the “Gradient Calculator” to determine the actual temperature for each column of wells. The “Gradient Calculator” is found under the List function in the main menu.
“GoTo” tells the program to go back to a previous step and repeat the following steps a specified number of times to create a cycle.
“Ramp” sets a heating/cooling rate lower than the maximum rate (3°C/s).
“End” must always be the last step in a protocol.
 - 5.2.5.2. Hit “Proceed” to add the step.
Use the number pad to specify the step’s parameters and hit “Proceed”.
 - 5.2.5.3. Select “Yes” to add the step or “Options” to add one of the following:
“Increment” progressively increases/decreases the temperature each cycle.
“Extend” progressively lengthens/shortens a temperature step each cycle.
“Beep” makes the machine beep when the temperature is reached.
- 5.3. Running an existing program.
 - 5.3.1. Hit the “Block” button until the main menu is displayed.
 - 5.3.2. Use the “Select” button to highlight the “Run” function and hit “Proceed”.
 - 5.3.3. Use the “Select” button to choose your program from the list.
You may first have to select the folder containing your program.
 - 5.3.4. Hit “Block” to select the block on which to run the program and hit “Proceed”.
 - 5.3.5. If you have selected a “Calculated” method (see 5.2.4.1.) enter the vessel type and volume and hit “Proceed”.
 - 5.3.6. Select whether or not to use a heated lid and hit “Proceed” to start the program.
- 5.4. Stepping through a program.
 - 5.4.1. While a program is running, hit “Proceed” to skip to the next step.
- 5.5. Pausing a running program.

- 5.5.1. Hit the “Pause” button to temporarily stop a program
- 5.5.2. The samples will be held at the current temperature.
- 5.5.3. If you hit “Pause” while the machine is ramping, it will stop once the target temperature is reached.
- 5.5.4. Hit “Pause” or “Proceed” to continue.
- 5.6. Stopping a running program.
 - 5.6.1. Hit the “Cancel” button to stop a run.
 - 5.6.2. Select “Yes” to confirm the cancellation.
- 5.7. Notes for efficient usage.
 - 5.7.1. To decrease wear and tear on the tetrads, do not program long 4°C holds.
 - 5.7.1.1. Consider holding at 10°C or retrieve your samples quickly.
 - 5.7.3. Immediately report any instrument problems/malfunctions to Claire Reardon (claire@cgr.harvard.edu).