MiniOne® PCR System Validation Kit

Cat #M6000 Version 100125

The MiniOne®PCR System Validation Kit provides enough reagents to run two rounds of four PCR reactions in the MiniOne PCR System (Cat #M4000). With the three primer sets you can amplify three different fragments from the lambda phage genome, plus a negative control.
GreenGel™ cups and buffer concentrate are provided for you to analyze your results on your MiniOne Electrophoresis System.

Materials included in the Validation Kit(Qty)		Materials required but not included
FastTaq™ polymerase PCR MasterMix (2X)	100 µL	MiniOne Electrophoresis System (Cat #M1000)
Lambda phage genomic DNA template	40 µL	MiniOne Casting System (Cat #M2002)
Three Primer Sets – PS1, PS2, PS3	40 µL	2-20 µL adjustable volume micropipette (Cat #M2008)
Sterile dH_2O (negative control reaction; -C)	40 µL	2-200 µL micropipette tips
Sample Loading Dye (5X)	50 µL	Benchtop centrifuge (optional, Cat #M2031)
MiniOne DNA Marker (M1M)	30 µL	Fine tip permanent marker
0.2 mL thin-wall PCR tubes	10 tubes	Microwave oven
Buffer concentrate bottle	20 mL (TAE) or 25 mL (TBE)	Distilled or deionized water for diluting running buffer
2% agarose GreenGel™ Cups	2 cups	Digital camera or cell phone camera

Instructions:

1. Label **four PCR tubes** with the sample name and add the following reagents to each tube:

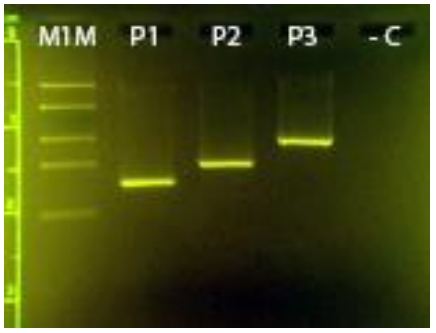
	Sample Name			
	P1	P2	P3	-C
FastTaq PCR MasterMix (2X)	10 µL	10 µL	10 µL	10 µL
Primer Set	5 µL (PS1)	5 µL (PS2)	5 µL (PS3)	5 µL (dH_2O)
Lambda genomic DNA	5 µL	5 µL	5 µL	5 µL

2. Cap the tubes and gently flick with your finger to mix the contents. Briefly spin in a centrifuge or tap on the bench top if reagents are stuck to the sides of the tube. Place the tubes in the wells of the MiniOne PCR machine.
3. Open the MiniOne PCR App on your iPad, tablet, or cell phone, connect to your PCR machine through Bluetooth. Program the following PCR Thermocycling Conditions:

	Temp	Time	Cycles
Denaturation	94°C	5 seconds	20
Annealing	54°C	5 seconds	
Extension	72°C	5 seconds	

Estimated Duration: 16:43 min

4. During the PCR, use your MiniOne Casting System to prepare a 2% agarose GreenGel. (takes ~15 minutes)
5. Prepare running buffer by diluting 1 part buffer concentrate into 19 parts distilled water.
6. When the PCR protocol is complete, remove the tubes and add 5 µL Sample Loading Dye (5X LD) to each tube.
7. Set up your MiniOne Electrophoresis chamber with your gel and 135 mL running buffer.
8. Load 10 µL of each sample, along with 10 µL of the MiniOne DNA Marker (M1M) into the wells.
9. Allow your gel to run for approximately 20 minutes or until the bands and the marker have clearly separated.
10. Photograph your gel by placing your smartphone or camera directly on top of the amber photohood.



Example Results:
P1 = 202 bp fragment
P2 = 304 bp fragment
P3 = 501 bp fragment
-C = no fragment