

**Ple53 Promoter PCR (pEMS1199)**

**MiniPromoter:** Ple53  
**pEMS#:** 1199  
**Expected product size (bp):** 735

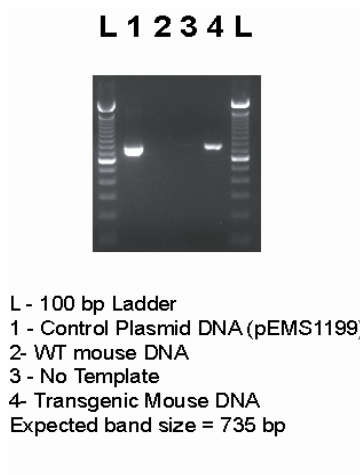
Reaction components	Vol/Rxn (µl)
H <sub>2</sub> O	15.15
10X PCR buffer*	2.5
50 mM MgCl <sub>2</sub> *	0.75
2.5 mM dNTPs**	2
10 µM oEMS2364	1.25
10 µM oEMS2753	1.25
Taq Pol. (5 U/µl)*	0.1
DNA***	2
Total Volume of Rxn:	25

\* Taq Polymerase set from Invitrogen (Cat no.18038-042)

\*\* dNTPs from Invitrogen (Cat no.10297-018)

\*\*\*Approximately 100 ng DNA used for samples, approximately 5 ng used for plasmid control

Samples run on a 2% agarose gel (containing SYBRsafe (Invitrogen Cat no. S33102))



Cycling conditions:	Step	Temp	Time	Note
	1	94°C	3 min	
	2	94°C	1 min	
	3	61°C	1 min	
	4	72°C	45 sec	repeat steps 2-4 34 times
	5	72°C	5 min	
	6	4°C	hold	

**Primers:**

Name	Sequence	T <sub>m</sub> (°C)	Notes
2364	5'- GCGTATCACGAGGCCCTTTC -3'	56.0	Sense primer located in Vector backbone 5' to MCS
2753	5'- GCTGCCACCAAGTGGGATAAA -3'	56.3	Antisense primer for Ple53 in region "LongProm".