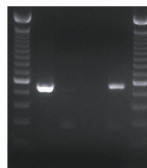


**Ple186 Promoter PCR (pEMS1385)**

**MiniPromoter:** Ple186  
**pEMS#:** 1385  
**Expected product size (bp):** 558

L 1 2 3 4 L



L - 100 bp Ladder  
 1 - Control Plasmid DNA (pEMS1385)  
 2 - WT mouse DNA  
 3 - No Template  
 4 - Transgenic Mouse DNA  
 Expected band size = 558 bp

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 oEMS2981	1.25
10 µM oEMS2487	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
2981	5'- GGCCGGTGAAGGATTATGA -3'	56.1	Sense primer for Ple186 in region "3".
2487	5'- CCGTGAAAGGGTTCCACACA -3'	56.9	Anti-Sense primer for Ple186 in region "Prom".