

RBRC 05146

Strain name B6;129S6-Gt(ROSA)26Sor<tm1(CAG-mTFP1)Imayo>

Primer	primer_name	primer_seq	length	
1	R26F2	AAAGTCGCTCTGAGTTGTTAT	21	Depositor's primer
2	oIMR0316	GGAGCGGGAGAAATGGATATG	21	JAX Genotyping Protocol
3	oIMR0315	GCGAAGAGTTTGTCTCAACC	21	JAX Genotyping Protocol
4	LFcheck-common-fw	AATTCCTCGACGGGGAATTCGGG	23	Depositor's primer
5	LFcheck-mTFP1-rv	CAGCGTAATCTGGTACGTCGTATG	24	Depositor's primer
6	mTFP1-400bp-fw	GAATGGCCACGCCTTCGTGATCGAG	25	Depositor's primer
7	mTFP1-400bp-rv	TTCAGCACGCCGTCGCGCACGTACATC	27	Depositor's primer

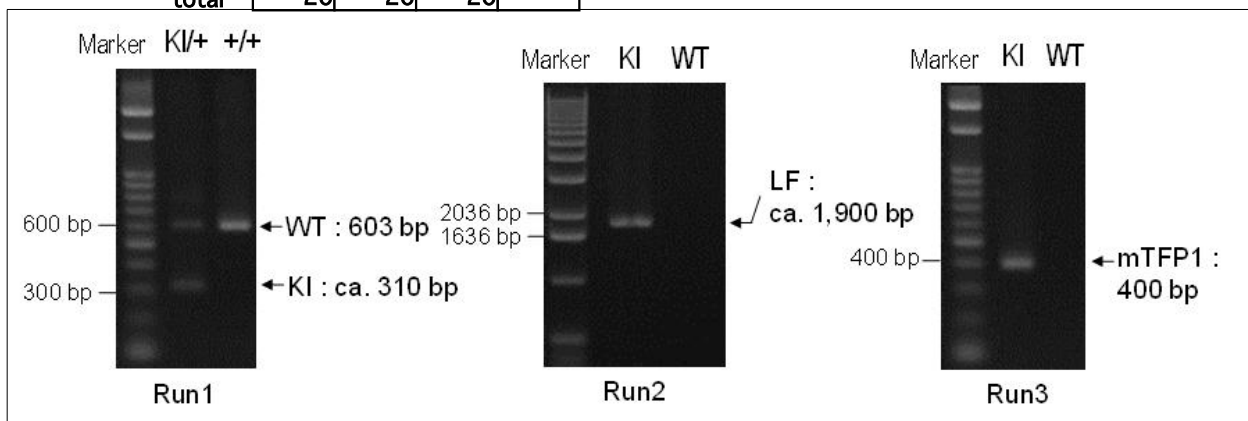
### Reaction Components

Enzyme TOYOBO KOD FX (Code.No : KFX-101)

### Reaction Conditions

step	temp (°C)	time (sec)	note
1	94	120	
2	98	10*	
3	63	30*	
4	68	120*	*Repeat for 30 cycles

	Conc	Run 1	Run 2	Run 3	Run 4
H2O		1.8	2.8	2.8	
PCR buffer	2 X	10.0	10.0	10.0	
dNTP	2.0 mM	4.0	4.0	4.0	
primer 1	10 pmol/μl	1.0	0	0	
primer 2	10 pmol/μl	1.0	0	0	
primer 3	10 pmol/μl	1.0	0	0	
primer 4	10 pmol/μl	0	1.0	0	
primer 5	10 pmol/μl	0	1.0	0	
primer 6	10 pmol/μl	0	0	1.0	
primer 7	10 pmol/μl	0	0	1.0	
DNA		1.0	1.0	1.0	
Taq	1 U/μl	0.2	0.2	0.2	
<b>total</b>		<b>20</b>	<b>20</b>	<b>20</b>	



**Marker**  
Gene Ladder 100 (Wako, Cat. 316-06951)

**Gel**  
2% agarose in 1 X TAE (100 V, 30 min)

PCR products			
	primer_set	product_size	product
	primer 1	primer 2	603 bp WT
	primer 1	primer 3	ca. 310 bp KI
	primer 4	primer 5	ca. 1900 bp LF
	primer 6	primer 7	400 bp mTFP1

**Comment** Novagen KOD Xtreme™ Hot Start DNA Polymerase can be purchased from EMD.

