

Ucp1-2A-tdTomato Genotyping Protocol



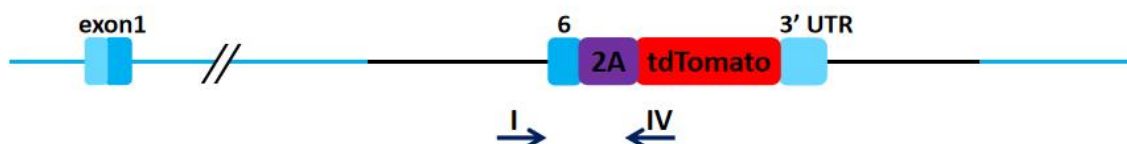
Common Name	Ucp1-2A-tdTomato	Cat. NO.	NM-KI-234734
Strain of Origin	C57BL/6J	Version	V1

Genotyping strategy

Wild type allele



Targeted allele



→ : primer location

Primers

Primer	Sequence (5' → 3')	Primer type
P1	CACTTAGCAGCCCATCACCA	Forward
P2	TCGTGGTCTCCAGCATAGA	Reverse
P4	CTTTGATGACCTCCTCGCCC	Reverse

Expected results

Results	P1+P2+P4 HE HE WT H₂O M DL2000
Genotype	Wild type: P1P2 =243 bp; Heterozygote: P1P2 =243 bp; P1P4 =280 bp Homozygote: P1P4 =280 bp

Reaction &Cycling

PCR Reaction System	Reaction Component			Volume (μl)
	ddH ₂ O			7.5
	2×Rapid Taq Master Mix			10.0
	P1 (10 pmol/μl)			0.5
	P2 (10 pmol/μl)			0.5
	P4 (10 pmol/μl)			0.5
	Genomic DNA			1.0
	Total			20
	2×Rapid Taq Master Mix from Vazyme (Code Number: P222-01)			
Cycling	Step	Temp	Time	Note
Reaction	1	95° C	5 min	

	2	95° C	20 sec	
	3	60° C	20 sec	
	4	72° C	20 sec	35 repeats to 2
	5	72° C	5 min	
	6	12° C	Hold	