Image from jax.org

Mouse Strain Datasheet

Strain Name

Pkd2fl/fl

MGI Gene ID MGI:4843126

Full Allele Name Pkd2<tm1.1Tjwt>

Type of Allele conditional allele

Human Gene (HGNC) PKD2

Genetic Background C57BL/6J

Commercial Source Jax mice

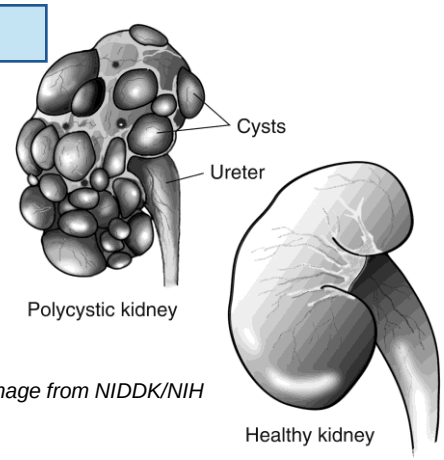

Image from NIDDK/NIH

Stock Number 17292

Link https://www.jax.org/strain/017292

Genotyping Protocol https://www.jax.org/Protocol?stockNumber=017292&protocolID=24847

Strain Details

A loxP site was inserted upstream of exon 11 and an FRT-flanked neo cassette with a 5' loxP site was inserted downstream of exon 13. Flp-mediated recombination removed the neo cassette leaving exons 11 through 13 floxed. These Pkd2 conditional mutant mice possess loxP sites flanking exons 11-13 of the polycystic kidney disease 2 (Pkd2) gene. Expression of Cre recombinase results in removal of the intervening exons generating a null allele. This strain may be useful for studying renal development in autosomal dominant polycystic kidney disease or the finction of pkd2 in various tissues and throughout adult life.

Validation or publication

https://pubmed.ncbi.nlm.nih.gov/20862291/

Contact Name Patricia Outeda

Email pgarcia@som.umaryland.edu

PCR Protocol for Genotyping: Pkd2fl/fl

A. Digestion of mouse tail or ear notch, and embryo tail (in red):

1. Add 100µL of Tissue Digestion Buffer and 2µL of Proteinase K per tail (~1-2mm length). For embryos tail add 50µL of Tissue Digestion Buffer and 1µL of Proteinase K. Make sure tail is immersed in the buffer.
2. In a thermocycler incubate at 55°C for 1 h followed by 95°C for 8 min to inactivate the enzyme and hold at 10°C. For embryos incubate at 55°C for 30 min followed by 95°C for 8 min and hold at 10°C.
3. Vortex and store at 4°C (-20°C for long storage) or use immediately to set up the PCR.

B. PCR Genotyping Protocol

Primers			
flox11-13(A)-F	5'-	CCT TTC CTC TGT GTT CTG GGG AG	-3'
"flox11-13(B)-R	5'-	GTT TGA TGC TTA GCA GAT GAT GGC	-3'
	5'-		-3'
	5'-		-3'

PCR Reaction			PCR Conditions		
BioMix (Bioline)	10.0 µL		Heated Lid		105°C
Primers (@10 µM each)	0.8 µL		Initial Denaturation	94°C	5 min
			Number of Cycles	x35	
$_{dd}H_2O$	7.2 µL			94°C	20 sec
				56 °C	35 sec
DNA template	2.0 µL			72°C	35 sec
Total Volume	20.0 µL		Final Extension	72°C	10 min
			Final Hold	10°C	

PCR Product Size (bp)	
Wild type band	232 bp
Pkd2fl band	318 bp

C. Reagents

Reagent	Cat #	Final Concentration	Working Concentration
Tissue Digestion Buffer for ear notch or tail			
Tris pH8.5		50mM	
EDTA		1mM	
Tween20		0.5%	
Proteinase K (Invitrogen)	25530-015	20mg/mL	
BioMix (Bioline)	BIO-25012		

Image from jax.org

Mouse Strain Datasheet

Strain Name

mTmG

MGI Gene ID n/a

Full Allele Name Gt(ROSA)26Sor<tm4(ACTB-tdTomato,-EGFP)Luo>

Type of Allele reporter

Human Gene (HGNC) n/a

Genetic Background C57BL/6J

Commercial Source Jax mice

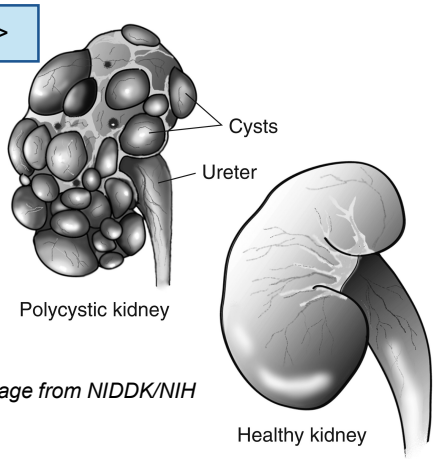

Image from NIDDK/NIH

Stock Number 007676

Link https://www.jax.org/strain/007676

Genotyping Protocol https://www.jax.org/Protocol?stockNumber=007676&protocolID=20368

Strain Details

ROSAmT/mG is a cell membrane-targeted, two-color fluorescent Cre-reporter allele. Prior to Cre recombination, cell membrane-localized tdTomato (mT) fluorescence expression is widespread in cells/tissues. Cre recombinase expressing cells (and future cell lineages derived from these cells) have cell membrane-localized EGFP (mG) fluorescence expression replacing the red fluorescence.

Validation or publication

Contact Name Stephen Parnell

Email sparnell@kumc.edu

PCR Protocol for Genotyping: mTmG

A. Digestion of mouse tail or ear notch, and embryo tail (in red):

1. Add 100µL of Tissue Digestion Buffer and 2µL of Proteinase K per tail (~1-2mm length). For embryos tail add 50µL of Tissue Digestion Buffer and 1µL of Proteinase K. Make sure tail is immersed in the buffer.
2. In a thermocycler incubate at 55°C for 1 h followed by 95°C for 8 min to inactivate the enzyme and hold at 10°C. For embryos incubate at 55°C for 30 min followed by 95°C for 8 min and hold at 10°C.
3. Vortex and store at 4°C (-20°C for long storage) or use immediately to set up the PCR.

B. PCR Genotyping Protocol

Primers			
wildtype forward	5'-	CTC TGC TGC CTC CTG GCT TCT	-3'
wildtype reverse	5'-	CGA GGC GGA TCA CAA GCA ATA	-3'
mutant reverse	5'-	TCA ATG GGC GGG GGT CGT T	-3'
	5'-		-3'

PCR Reaction			PCR Conditions		
BioMix (Bioline)	10.0 µL		Heated Lid		105°C
Primers (@10 µM each)	0.8 µL		Initial Denaturation	94°C	5 min
			Number of Cycles	x35	
$_{dd}H_2O$	7.2 µL			94°C	20 sec
				60 °C	35 sec
DNA template	2.0 µL			72°C	35 sec
Total Volume	20.0 µL		Final Extension	72°C	10 min
			Final Hold	10°C	

PCR Product Size (bp)	
Wild type band	330 bp
transgene	250 bp

C. Reagents

Reagent	Cat #	Final Concentration	Working Concentration
Tissue Digestion Buffer for ear notch or tail			
Tris pH8.5		50mM	
EDTA		1mM	
Tween20		0.5%	
Proteinase K (Invitrogen)	25530-015	20mg/mL	
BioMix (Bioline)	BIO-25012		