

## pLenti-TDRD10-sgRNA

产品编号	产品名称	包装
L06835	pLenti-TDRD10-sgRNA	5μg

### 产品简介:

- pLenti-TDRD10-sgRNA (TDRD10基因敲除质粒)是一种在动物细胞中可以同时表达Cas9、目的基因的sgRNA和puromycin抗性基因的质粒。用于在动物细胞中直接基于CRISPR/Cas9技术敲除目的基因，或者通过包装慢病毒后基于CRISPR/Cas9技术敲除目的基因。本质粒中sgRNA的有效性已经通过T7EI法的验证。
- 本质粒在细菌中为Amp抗性，全长约13,000bp。本质粒的关键图谱信息请参考图1。本质粒可直接转染细胞用于目的基因的CRISPR/Cas9敲除，以及通过puromycin筛选稳定细胞株。也可以与pMDLg、Rev及VSV-g共转HEK293T细胞进行重组慢病毒(lentivirus)的包装，然后再用于感染细胞或组织并进行目的基因的CRISPR/Cas9敲除。



图1. 表达sgRNA、Cas9和puromycin抗性的pLenti-sgRNA质粒关键图谱信息。

- 本质粒中的sgRNA基于碧云天研发的CRISPR/Cas9 sgRNA快速筛选和验证体系获得，sgRNA的有效性已经通过T7EI法验证。
- 本质粒用于实验时，建议同时选购无任何靶向的对照质粒pLenti-Control-sgRNA (L00011)或靶向GFP的对照质粒pLenti-GFP-sgRNA (L00013)。
- 碧云天同时提供基于CRISPR/Cas9技术的TDRD10基因敲除的质粒(L06835 pLenti-TDRD10-sgRNA)、慢病毒(L06836 TDRD10 Knockout Lentivirus)、HEK293T细胞(L06837 TDRD10 Knockout HEK293T Cells)、HEK293T敲除细胞的RIPA裂解液(L06838 TDRD10 Knockout HEK293T RIPA Lysate)、HEK293T敲除细胞的Trizol裂解液(L06839 TDRD10 Knockout HEK293T Trizol Lysate)等产品，具体请在碧云天网站查询或在本产品网点击相应产品。
- TDRD10基因的基本信息如下：

Species	Gene Symbol	Gene ID	GenBank Accession	Transcript
Human	TDRD10	126668	BC044864	NM_182499

About the gene	
Official Symbol	TDRD10
Previous Symbol	-
Official Full Name	tudor domain containing 10
Synonyms	DKFZp434M202
Location	1q21.3
Gene Type	protein_coding
Uniprot ID	Q5VZ19
Pathway/Library	Epigenetic Regulators Related Genes Library
Gene Summary	Tudor domain-containing proteins (TDRDs) are characterized by various numbers of Tudor domains, which are known to recognize and bind to symmetric methylated arginine residues. These proteins affect a wide variety of processes, including differentiation, genome stability and gametogenesis. In mammals, there are 12 members (TDRD1-TDRD12) in the TDRD protein family. Porcine TDRD10 has an opening reading frame (ORF) of 1068 bp, which encodes 355 amino acids. It localizes to chromosome 4. The gene product of porcine TDRD10 contains a Tudor domain and a RNA recognition motif (RRM). Serial deletion shows that the 5'-flanking sequence of porcine TDRD10 contains several negative and positive regulatory elements and identifies a 670-bp TATA-less region as an optimal promoter. Site-directed mutagenesis reveals that the nucleotides from -451 to -445 relative to the transcriptional start site forms one of the very important positive regulatory elements. Real time PCR detects the highest expression level of porcine TDRD10 gene in heart among 12 tissues. In PK15 cells, it mainly distributed in the cell nucleus, but also exhibited localization to the cytoplasm.



<https://www.beyotime.com/product/ST551-10mg.htm>。

- c. 转染后约48小时，按照上述检测获得的puromycin筛选浓度加入puromycin，筛选阳性细胞。一般筛选2天后，阴性细胞逐渐死去。培养过程中，可以将细胞转至6孔板或10cm培养皿进行扩大培养。一周之后，puromycin浓度可减半。如果有必要后续可以通过将细胞稀释至2.5个/ml，然后按照每孔200 $\mu$ l接种到96孔板中(每孔平均0.5个细胞)，筛选单克隆细胞株。

#### 5. 基因编辑的鉴定:

- a. 对于多克隆细胞，可以通过T7 Endonuclease I (T7EI)进行鉴定，即提取细胞的基因组DNA，在sgRNA序列两边设计引物进行PCR扩增，然后进行T7EI酶切，具体请参考碧云天的T7 Endonuclease I (CRISPR等基因突变鉴定用) (D7080)或基因组编辑突变检测试剂盒(D0508); 也可以通过相应的抗体进行检测。
- b. 对于单克隆细胞，可通过PCR扩增出sgRNA靶向的基因片段后进行常规测序的方式进行验证，同时也可以使用相应的抗体进行检测。

#### 相关产品:

产品编号	产品名称	包装
L00002-5 $\mu$ g	CRISPR/Cas9 Packaging Vectors Set A	5 $\mu$ g/each
L00002-100 $\mu$ g	CRISPR/Cas9 Packaging Vectors Set A	100 $\mu$ g/each
L00011-5 $\mu$ g	pLenti-Control-sgRNA	5 $\mu$ g
L00011-100 $\mu$ g	pLenti-Control-sgRNA	100 $\mu$ g
L00013-5 $\mu$ g	pLenti-GFP-sgRNA	5 $\mu$ g
L00013-100 $\mu$ g	pLenti-GFP-sgRNA	100 $\mu$ g
C0222	青霉素-链霉素溶液(100X)	100ml
C0351-1ml	Polybrene (Hexadimethrine Bromide)	1ml
C0351-50mg	Polybrene (Hexadimethrine Bromide)	50mg
C0521	Lipo293 <sup>TM</sup> 转染试剂	0.5/1.5/7.5ml
C0526	Lipo6000 <sup>TM</sup> 转染试剂	0.5/1.5/7.5ml
C0533	Lipo8000 <sup>TM</sup> 转染试剂	0.5/1.5/7.5ml
D0378	Stbl3甘油菌	200 $\mu$ l
ST551-10mg	Puromycin Dihydrochloride (嘌呤霉素)	10mg/ml $\times$ 1ml
ST551-50mg	Puromycin Dihydrochloride (嘌呤霉素)	10mg/ml $\times$ 5ml
ST551-250mg	Puromycin Dihydrochloride (嘌呤霉素)	250mg
ST1380-500mg	Polybrene ( $\geq$ 94%, Reagent grade)	500mg
ST1380-2g	Polybrene ( $\geq$ 94%, Reagent grade)	2g
ST1380-10g	Polybrene ( $\geq$ 94%, Reagent grade)	10g
FF345-10pcs	针头滤器(0.45 $\mu$ m/28mm, PES, Sterile, Sartorius分装)	10个/袋
FF345T-10pcs	针头滤器(0.45 $\mu$ m/28mm, PES, Sterile, 进口分装)	10个/袋
FF345-50pcs	针头滤器(0.45 $\mu$ m/28mm, PES, Sterile, Sartorius原装)	50个/盒
FF365-10pcs	BeyoGold <sup>TM</sup> 针头滤器(0.45 $\mu$ m/33mm, PES, Sterile)	10个/袋
FF365-100pcs	BeyoGold <sup>TM</sup> 针头滤器(0.45 $\mu$ m/33mm, PES, Sterile)	100个/盒
FF375-10pcs	BeyoGold <sup>TM</sup> 针头滤器(0.45 $\mu$ m/13mm, PES, Sterile)	10个/袋
FF375-100pcs	BeyoGold <sup>TM</sup> 针头滤器(0.45 $\mu$ m/13mm, PES, Sterile)	100个/盒
FUF158-2pcs	超滤管(15ml, 100kDa MWCO, PES, Sartorius分装)	2个/袋
FUF158-12pcs	超滤管(15ml, 100kDa MWCO, PES, Sartorius分装)	12个/袋

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