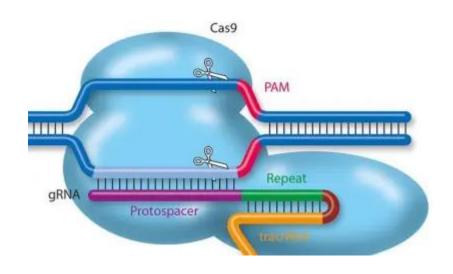
sgRNA 在线设计工具合集

CRISPR/Cas9 基因编辑系统自 2012 年问世以来就风靡全球,已然成为最主流的基因编辑系统。和前两代基因编辑系统 ZFN 和 TALEN 相比, CRISPR/Cas9 更高效、更简便且成本更低。在科研领域,CRISPR/Cas9 被广泛应用于基因功能研究及基因功能大规模筛选;而在生物医药领域,多个基于 CRISPR/Cas9 的基因治疗药物已获批迈入临床阶段。毋庸置疑,CRISPR/Cas9 将引领本世纪的生物技术与生物医学的革命。

作为一只科研汪,俺更关注如何高效使用 CRISPR/Cas9。简单来说,CRISPR/Cas9 需要三个原件/组分同时存在才能够进行精确的基因敲除或者插入。分别是 Cas 核酸酶、PAM 序列、sgRNA,其中 Cas 核酸酶与PAM 序列基本固定,而 sgRNA 的设计则成为提高 CRISPR/Cas9 基因编辑效率的关键所在。



1、GPP Web Portal



GPP Web Portal

Home | Search by Gene | Search by Clone

GPP sgRNA Designer

Soon to be relaunched as CRISPick. Please check this space for updates.

CRISPRko CRISPRa CRISPRi	
CRISPR Enzyme: SpyoCas9 (NGG) V	
Target Genome: Human GRCh38 (NCBI RefSeq v.109.20200815) ✔	
Specify Target(s):	
Input Transcript IDs, Gene IDs/Symbols, or raw DNA sequence:	Enter up to 200 Transcript IDs (e.g., NM_014911.3, ENST00000456328, etc.), Gene IDs or Symbols (e.g., 988, CDC5L, ENSG00000223972, etc.), or a single DNA sequence. File inputs must be smaller than 20kb in size, and any sequences submitted via file must be in FASTA format.
-OR- Upload a list of Transcript IDs, Gene IDs/Symbols, or a FASTA file of DNA sequences: 选择文件 未选择任何文件	Please refer to our sgRNA Designer Help Page for details on how a transcript is chosen for a gene input.
Pick Quota: 5 ✓ Report Unpicked Sequences?	

https://portals.broadinstitute.org/gppx/crispick/public

博德研究所(Broad Institute)和张锋实验室发布的 GPP Web Portal 囊括了基因敲除(CRISPRko)、激活(CRISPRa)和沉默(CRISPRi)模块,可以根据实验目的及实验需要进行相应的 sgRNA 设计。该工具根据大规模的实验室筛选结果结合深度学习,对 sgRNA 的切割效率及脱靶情况进行估计,从而给出预测结果。鉴于张锋大牛在基因编辑领域的地位和知名度较高,GPP Web Portal 可能是目前最为常用的 sgRNA 设计工具。

2 E-CRISP

E-CRISP



Design of CRISPR constructs

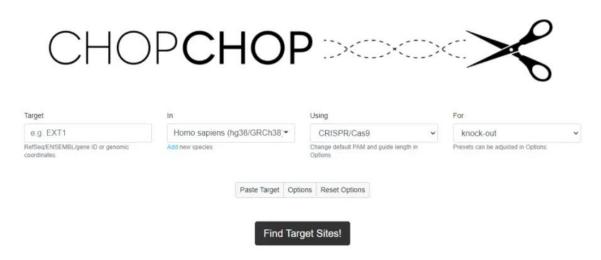
Design	Evaluation	MultiCRISP	CLD	GenomeCRISPR	Help	Links
	w CRISPR Library Des kerized version now a	signer (CLD): batch de t CLD on Github	esign of sgRNA	libraries		
1. Select organism	n:					
Homo sapiens G	RCh38	▼ [HELP]				
2. Select target re	gion by gene symbo	or sequence:				
Input is Gene	Symbol Search and in	mport ENSEMBLID				
O Input is FASTA	A sequence					
TP53						
FASTA example G	ieneSymbol example	Clear [HELP]				
3. Start application	n:					
relaxed (any PAM (NAG/NG	GG), any 5' base (A,C,	G,T,), off-targets need	d fu <mark>ll l</mark> ength per	fect match, introns are allo	wed)	
medium (any PAM (NAG/NG)	iG), any 5' base (A,C,	G,T,), off-targets tole	rate mismatche	s, introns/CPG islands are e	excluded)	
	lly G as 5' base, off-tar llowed) and UTRs are	_	smatches and i	gnores non-seed region, int	rons, purpose i	s knockout (only first 3
Single design V	Start sgRNA searc	Reset form Disp	lay <mark>ad</mark> vanced o	otions		

The older version of E-CRISP can be reached Here

http://www.e-crisp.org/E-CRISP

德国癌症研究中心(DKFZ) Michael Boutros 实验室发布的 E-CRISP 可以评估多种物种中 sgRNA 的切割与脱靶情况。

3、CHOPCHOP



http://chopchop.cbu.uib.no

挪威卑尔根大学(University of Bergen) Eivind Valen 实验室开发的在 线工具 CHOPCHOP 目前已更新至版本 3,该工具不仅可以用于设计基因 敲除用 sgRNA,还可以用来设计基于 CRISPR 的基因激活、沉默等,同时也是针对多种物种的。

4. Other sites: http://crispr.mit.edu/ and http://www.e-crisp.org/E-CRISP/. 其他

除了这几种常用在线 sgRNA 设计工具以外,还有特殊应用的工具,比如专门用于果蝇、线虫等物种的 FlyCRISPR

New:

- Drosophila vasa-cas9 on III (BDSC_51324) genome sequence generated in the Furlong lab, EMBL, Heidelberg
- Drosophila nanos-cas9 on II and III (BDSC_78781, BDSC_78782) genome sequence generated in the White lab, NIMH, National Institutes of Health

Select relevant cas9 below to search. 3

Enter genomic DNA sequence to find CRISPR target sites:

(Omit header lines and special characters.)

Enter DNA sequence here...

Select	genome:	Drosophila melanogaster (reference genome, r_6)	
Select	guide leng	th (nt) 3 20 🕶	
Find:	O CRIS	RISPR targets PR targets with 5' G	
Find	CRISPR 1	PR targets with 5' GG 1	

Already have CRISPR targets? Skip to next step

http://targetfinder.flycrispr.neuro.brown.edu

还有一些生物技术公司自己设计的小工具, 比如 SYNTHEGO 的 Knockout Guide Design

	≯SYNTHEGO	
· ·	Cnockout Guide Design	
Design	n top-scoring guide RNAs for gene knockout	
Genome	Gene	Nuclease
Type a genome name or ID	Select a genome first	SpCas9
Popular Homo Sapiens Drosophilia Mus Musculus	Search	
Searching 113,934,776,281 targets ac	cross 123,769 genomes for CRISPR knockout gui	ides in less than 10 seconds.

https://design.synthego.com/#

但是无论如何,在线工具都无法替代实验经验,毕竟这些在线工具都是计算机模型预测的结果,而在实际操作中 sgRNA 设计经验的积累来源于实践。

艾迪基因正是依赖于上百例基因编辑成功案例的丰富经验,支撑了向客户承诺 100%成功的保证,不成功不收费!

如果您有基因编辑相关问题,欢迎您随时和我们联系!