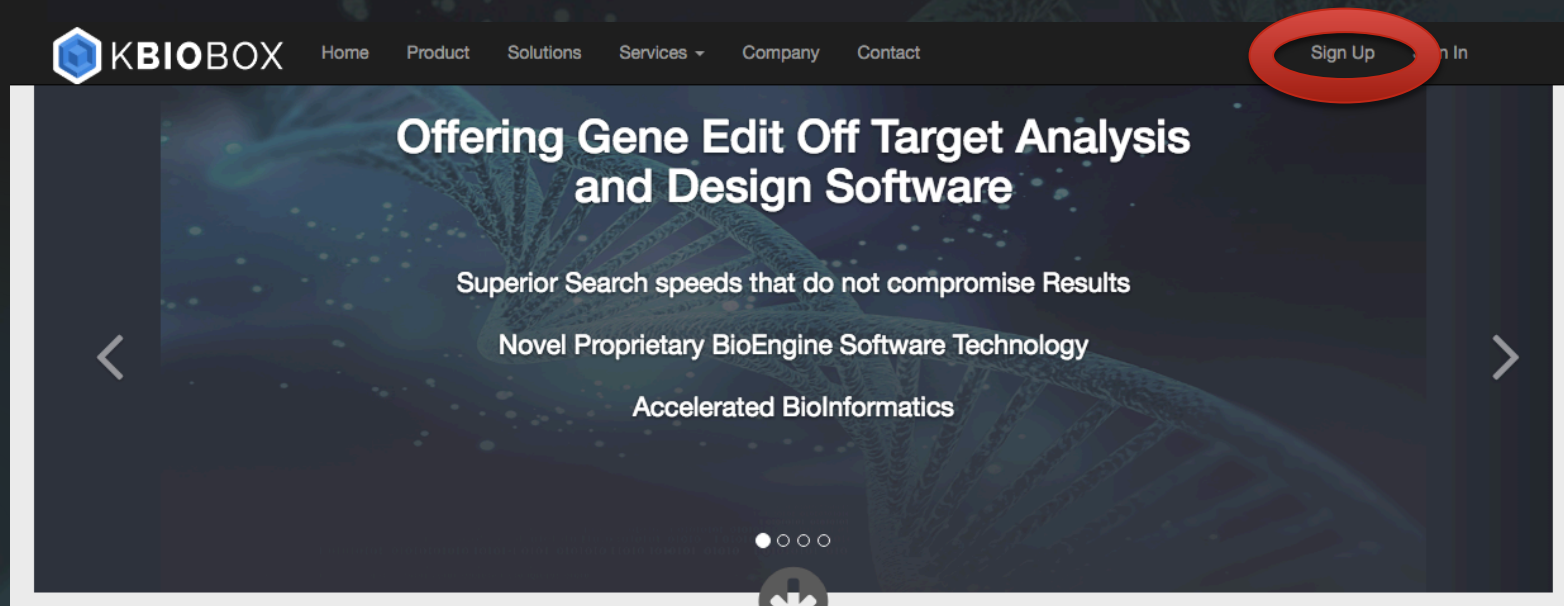




Create an Account

On the KBioBox home page click “Sign Up” in the upper right hand corner

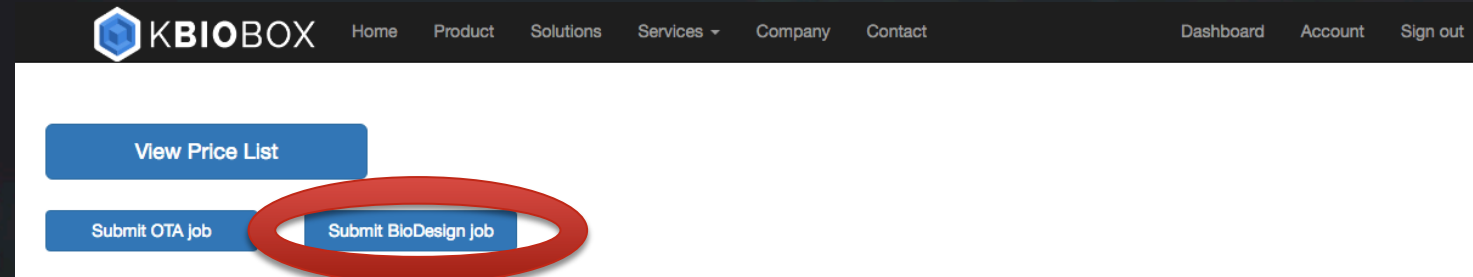


Or navigate to https://www.kbiobox.com/users/sign_up

Then use the form to create an account, and follow the instructions in the verification email

Submitting a BioDesign Job

- Once logged in, on the dashboard click *Submit BioDesign Job*
- If desired, give the job a name (optional)
- Leave the *Reference* box set to GRCh38p7
- In the *Gene Edit Location* box, type "INS". A check mark will appear and the "Submit Job" button will turn blue
- Change *Select Search type* from "Whole Gene" to "Exons". Now select "2" in the drop down menu that appears to the right
- Leave *Select Gene Edit Types* as "wtCRISPR"
- Click "Submit Job"



Create a new BioDesign job

Job Properties

Job Name New BioDesign Job	Reference Human GRCh38p7	Gene Edit Location <input type="text"/> Example location: SPRY3
--------------------------------------	------------------------------------	--

Select Search Type

☒ Whole Gene
☐ Exons
☐ 5' UTR
☐ CDS
☐ Introns
☐ 3' UTR

For access to additional search types please [contact us](#)

Select Gene Edit Types

☒ wtCRISPR
☐ TALEN
☐ StCas9
☐ SaCas9
☐ TdCas9
☐ NmCas9

For access to additional gene edit types please [contact us](#)

Submit job

Viewing a BioDesign job report

Status	Job Name	Type	Reference Genome		Target
Complete	New OTA Job	wtCRISPR Off Target Analysis	Human	GRCh38p7	TGCAACTAGACGCAGCCCGCAGG
Complete	New BioDesign Job	wtCRISPR BioDesign	Human	GRCh38p7	INS



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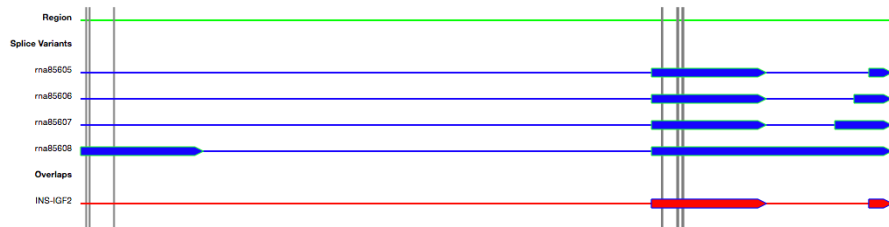
[Dashboard](#)

[Account](#)

[Sign out](#)

Job Information

Name	New BioDesign Job
Gene Edit Location	INS
Reference Genome	GRCH38P7
Search Type	First 2 exon(s)
Gene Edit Type	wtCRISPR



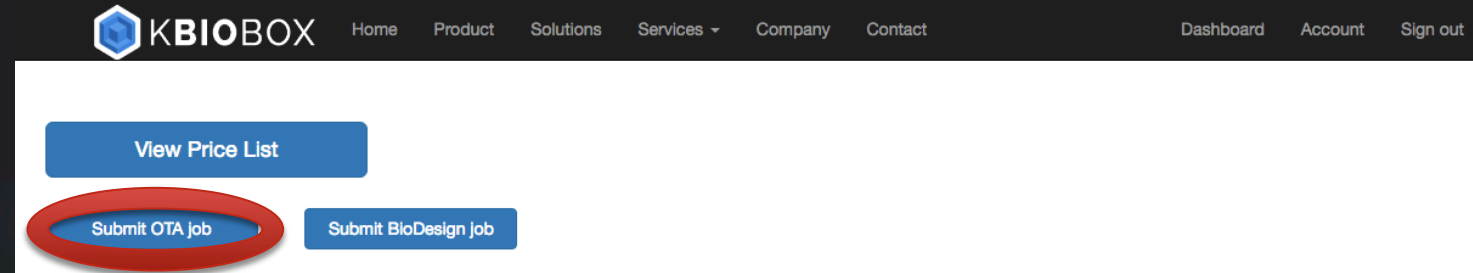
Top 10 Gene Edit Designs

Number	Sequence(s)	Offset	Direction	Additional Design Info
1	AGCCTCGTTCCCGCACACTAGG	2160830	forward	View Off-Target Analysis
2	TGCAACTAGACGCAGCCCGCAGG	2159838	reverse	View Off-Target Analysis
3	TACCTAGTGTGCGGGGACGAGG	2160832	reverse	View Off-Target Analysis
4	TCTACACACCCAAGACCGCCGG	2160804	reverse	View Off-Target Analysis
5	AGCTCTCTACCTAGTGTGCGGG	2160839	reverse	View Off-Target Analysis
6	AAGCTCTCTACCTAGTGTGCGG	2160840	reverse	Generating off-target analysis for this design. Please wait...
7	GAAGCTCTCTACCTAGTGTGCGG	2160841	reverse	Generating off-target analysis for this design. Please wait...
8	CTACACACCAAGACCGCCGGG	2160803	reverse	Generating off-target analysis for this design. Please wait...
9	GGGCTTTATCCATCTCTCGG	2159789	forward	Generating off-target analysis for this design. Please wait...
10	TATTCATCTCTCTCGGTGCAGG	2159795	forward	Generating off-target analysis for this design. Please wait...

- After submission you will be returned to the dashboard and the status of submitted jobs can be seen in the first column of the table
- Once a job status is "Complete" clicking on the job name in the second column will bring you to the job report
- For each of the Top 10 gene edit designs an Off Target Analysis (OTA) is automatically submitted, these take about a minute to run and once complete can be viewed by clicking "View Off-Target Analysis" in the fifth column of the table
- The BioDesign report is interactive, the grey lines that indicate gene edit position can be moused over, resulting in the highlighting of the corresponding row in the table
- Similarly mousing over a row in the table results in the corresponding grey line turning blue and enlarging to indicate position in the visualization

Submitting an Off-Target Analysis (OTA)

- From the Dashboard click *Submit OTA job*
- If desired, give the job a name in the *Job Name* box (optional)
- Leave the *Reference Sequence* box set to “GRCh38p7”
- Leave the *Gene Edit Type* set to “wtCRISPR”
- Enter “TGCAACTAGACGCAGCCCGCAGG”
- Click *Submit Job*



Create a new Off-Target Analysis job

Job Properties

Job Name

Reference Sequence

Gene Edit Type

For access to additional references and gene edit types please [contact us](#)

Target

Example Target: GAGCGAAGGGATCGAAACCCGGG

Viewing an Off Target Analysis Report


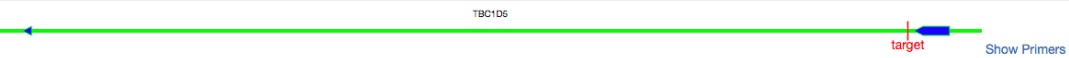
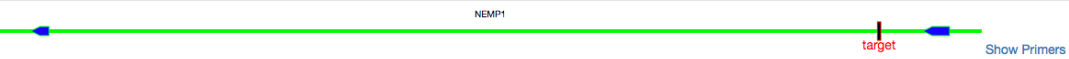
Status	Job Name	Type	Reference Genome		Target
Complete	New OTA Job	wtCRISPR Off Target Analysis	Human	GRCh38p7	TGCAACTAGACGCAGCCCGCAGG
Complete	New BioDesign Job	wtCRISPR BioDesign	Human	GRCh38p7	INS

Job Information

Name	New OTA Job
Status	Complete
Gene Edit Type	wtcrispr
Reference Sequence	GRCH38P7
Design	TGCAACTAGACGCAGCCCGCAGG

Off-Target Analysis Report

[Download OTA report as spreadsheet](#)

#	Matches	Off-Target Sequence	Chromosome	Chromosome Position	Direction
1	23	TGCAACTAGACGCAGCCCGCAGG	11	2159838 - 2159860	R
					
2	20	TGCAACTAGACGCAGCCCGCAGG	8	137400582 - 137400604	R
					
3	20	TGCAACTAGACGCAGCCCGCAGG	3	17739034 - 17739056	F
					
4	20	TGCAACTAGACGCAGCCCGCAGG	12	57078319 - 57078341	F
					
5	20	TGCAACTAGACGCAGCCCGCAGG	7	77993087 - 77993109	F

- When the Off Target Analysis has completed click the job name to view the report
- Each site includes an annotated region visualization
 - The black box labeled *Target* represents the gRNA match location
 - Horizontal black lines indicate a non-coding region
 - Horizontal green lines indicated a coding region
 - A blue trapezoidal represents an exon

Viewing Off Target Primers

Status	Job Name	Type	Reference Genome		Target
Complete	New OTA Job	wtCRISPR Off Target Analysis	Human	GRCh38p7	TGCAACTAGACGCAGCCCGCAGG
Complete	New BioDesign Job	wtCRISPR BioDesign	Human	GRCh38p7	INS

Job Information

Name	New OTA Job
Status	Complete
Gene Edit Type	wtcrispr
Reference Sequence	GRCH38P7
Design	TGCAACTAGACGCAGCCCGCAGG

Off-Target Analysis Report

[Download OTA report as spreadsheet](#)

#	Matches	Off-Target Sequence	Chromosome	Chromosome Position	Direction
1	23	TGCAACTAGACGCAGCCCGCAGG	11	2159838 - 2159860	R
2	20	TGCAACTAGACGCAGCCCGCAGG	8	137400582 - 137400604	R
3	20	TGCTACTAGACGCAGCCCGCAGG	3	17739034 - 17739056	F
4	20	TGCAACGAGACGCAGCCCGCAGG	12	57078319 - 57078341	F
5	20	TGCAACTAGACGCAGCCCGCAGG	7	77993087 - 77993109	F

- When the Off Target Analysis has completed click the job name to view the report
- Each site includes an annotated region visualization
 - The black box labeled *Target* represents the gRNA match location
 - Horizontal black lines indicate a non-coding region
 - Horizontal green lines indicated a coding region
 - A blue trapezoidal represents an exon
- To download report as an excel spreadsheet click *Download OTA report as spreadsheet*, located above the first result
- Primer information for the off target location is calculated with Primer 3 and shown by clicking *Show Primers* in the bottom right of each report