

GeneArt

CRISPR

Search and Design Tool



Quick reference guide

Our Invitrogen™ GeneArt™ CRISPR Search and Design Tool allows you to search our database of >600,000 predesigned CRISPR guide RNA (gRNA) sequences or analyze your sequence of interest for *de novo* gRNA designs using our proprietary algorithms. Once you've selected the optimal gRNA designs, you may purchase your gRNAs and other recommended products for genome editing directly from the web tool.

Accessing the GeneArt CRISPR Search and Design Tool

1. Go to thermofisher.com/crisprdesign to access the GeneArt CRISPR Search and Design Tool. If you are using an unsupported browser, the following message will appear:

Unsupported Browser

Thermo Fisher Cloud includes advanced data security and display features that require the use of one of the following browsers/versions:

[Internet Explorer 10 and newer](#)

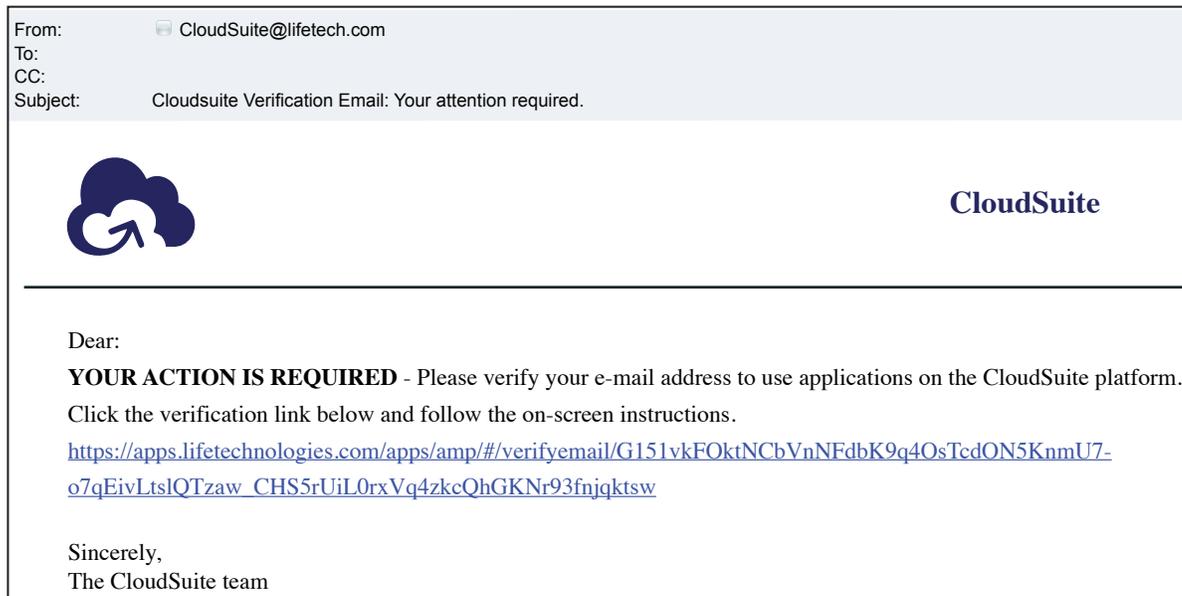
[Google Chrome 25 and newer](#)

[Apple Safari 5 and newer](#)

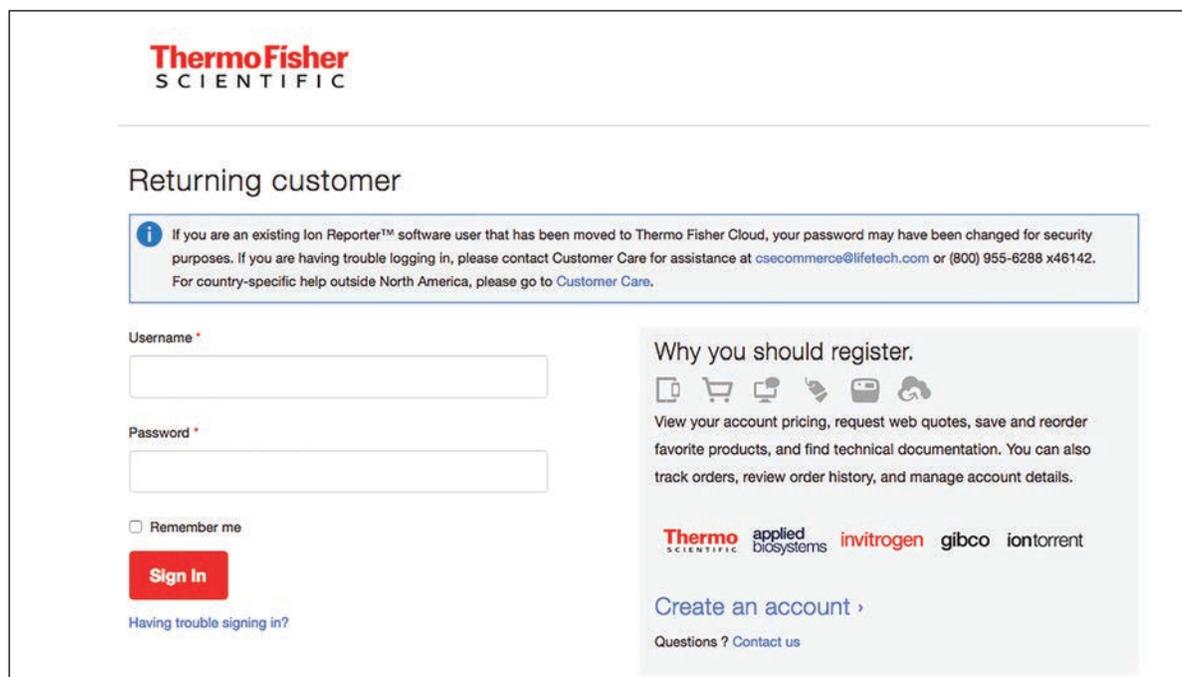
[Firefox 16 and newer](#)

Click on the link above to download and install a compatible browser, and thank you for using Thermo Fisher Cloud.

- Once you are in a supported browser, you will need to log into Thermo Fisher Cloud using your existing username and password. You will be asked to verify your email address the first time you log in. A verification request email, like the one pictured below, will be sent to your email address. Check your spam mailbox if you don't immediately receive this verification request. Future visits to the GeneArt CRISPR Search and Design Tool will not require a Thermo Fisher Cloud login.



- Log into your account using your existing username and password or follow the prompts to create your own account. You will be directed to the GeneArt CRISPR Search and Design Tool after you sign in.

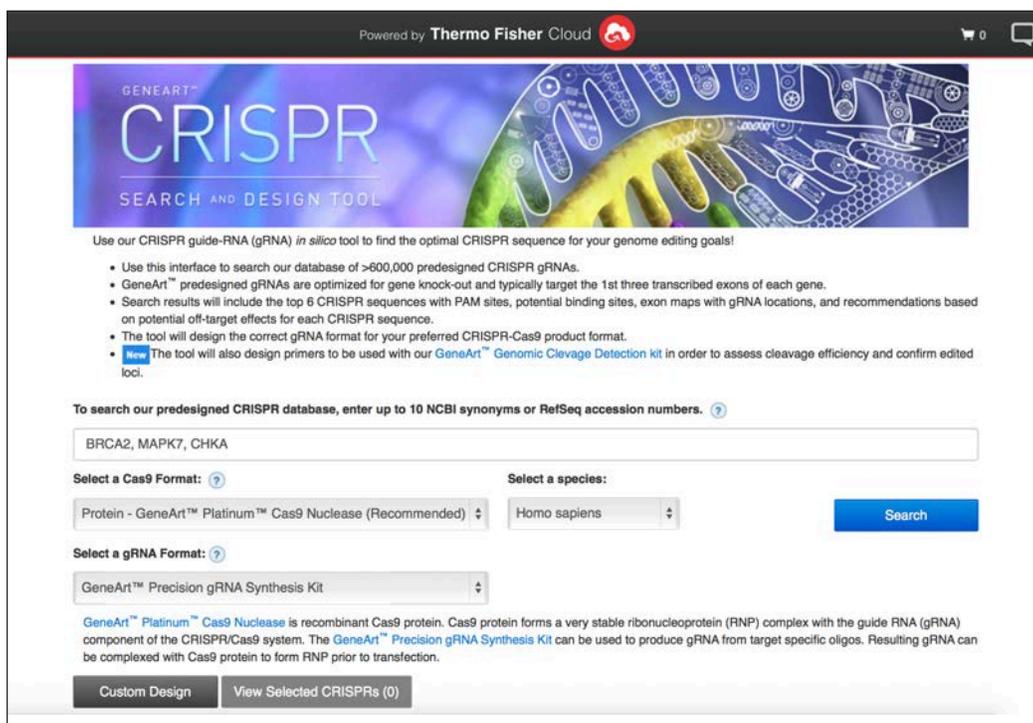


Using the GeneArt CRISPR Search and Design Tool

Use this tool to search our database of >600,000 predesigned CRISPR gRNA sequences. These sequences are optimized for gene knockout and target the first three transcribed exons for each gene. Search results include the top 6 CRISPR target sequences with PAM sites, exon maps with gRNA binding sites, and potential off-target binding sites for each CRISPR sequence. The tool will design the correct gRNA format for your preferred Invitrogen™ CRISPR-Cas9 product. To search for predesigned CRISPR gRNAs:

1. In the search box, enter up to 10 gene IDs, gene symbols, or RefSeq accession numbers, separated by commas. An example is shown below with the search terms BRCA2, MAPK7, and CHKA.
2. Select a CRISPR-Cas9 expression format: Invitrogen™ GeneArt™ CRISPR Nuclease Vector, GeneArt™ CRISPR Nuclease mRNA, or GeneArt™ Platinum Cas9 Nuclease.
 - Our GeneArt CRISPR Nuclease Vector Kits are used to express the gRNA and the Cas9 protein necessary for genome editing. The vectors can be purchased with or without competent cells, and include options for enrichment and monitoring of transfected cells:
 - The Invitrogen™ GeneArt™ CRISPR Nuclease Vector with OFP Reporter Kit is used for fluorescence-based sorting of cells by flow cytometry
 - The Invitrogen™ GeneArt™ CRISPR Nuclease Vector with CD4 Enrichment Kit is used for enrichment of cells using beads coupled to anti-CD4 antibody
 - GeneArt CRISPR Nuclease mRNA is ready-to-transfect, wild-type Cas9 mRNA. The mRNA is used with our custom-made Invitrogen™ GeneArt™ CRISPR Strings™ DNA, available in two formats:
 - GeneArt™ CRISPR U6 Strings™ DNA contains a U6 promoter for direct transcription of gRNA after cotransfection with Cas9 mRNA
 - GeneArt™ CRISPR T7 Strings™ DNA contains a T7 promoter for *in vitro* transcription of gRNA, which is followed by cotransfection of the *in vitro*-transcribed gRNA with Cas9 mRNA
 - GeneArt Platinum Cas9 Nuclease is purified, ready-to-transfect recombinant Cas9 protein containing a nuclear localization signal (NLS) for targeting to the nucleus
 - In our example shown below, GeneArt Platinum Cas9 Nuclease has been selected with the Invitrogen™ GeneArt™ Precision gRNA Synthesis Kit for *in vitro* transcription of gRNA
3. Select a species, either *Homo sapiens* or *Mus musculus*.
4. Select a gRNA format based on your Cas9 format selection.
5. Click “Search” to search the database for predesigned CRISPR gRNAs.

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GENEART™ CRISPR SEARCH AND DESIGN TOOL

Use our CRISPR guide-RNA (gRNA) *in silico* tool to find the optimal CRISPR sequence for your genome editing goals!

- Use this interface to search our database of >600,000 predesigned CRISPR gRNAs.
- GeneArt™ predesigned gRNAs are optimized for gene knock-out and typically target the 1st three transcribed exons of each gene.
- Search results will include the top 6 CRISPR sequences with PAM sites, potential binding sites, exon maps with gRNA locations, and recommendations based on potential off-target effects for each CRISPR sequence.
- The tool will design the correct gRNA format for your preferred CRISPR-Cas9 product format.
- **New** The tool will also design primers to be used with our GeneArt™ Genomic Cleavage Detection kit in order to assess cleavage efficiency and confirm edited loci.

To search our predesigned CRISPR database, enter up to 10 NCBI synonyms or RefSeq accession numbers. ?

BRCA2, MAPK7, CHKA

Select a Cas9 Format: ? Protein - GeneArt™ Platinum™ Cas9 Nuclease (Recommended) ▾

Select a species: Homo sapiens ▾

Select a gRNA Format: ? GeneArt™ Precision gRNA Synthesis Kit ▾

Search

GeneArt™ Platinum™ Cas9 Nuclease is recombinant Cas9 protein. Cas9 protein forms a very stable ribonucleoprotein (RNP) complex with the guide RNA (gRNA) component of the CRISPR/Cas9 system. The GeneArt™ Precision gRNA Synthesis Kit can be used to produce gRNA from target specific oligos. Resulting gRNA can be complexed with Cas9 protein to form RNP prior to transfection.

Custom Design View Selected CRISPRs (0)

Using the CRISPR designer

To access the CRISPR designer, click on “Custom Design” in the GeneArt CRISPR Search and Design Tool. Use this tool to analyze your sequence of interest and design *de novo* CRISPR gRNA sequences using our unique design algorithms. As with the predesigned gRNA search option, search results include the top 6 CRISPR target sequences with PAM sites, exon maps with gRNA binding sites, and potential off-target binding sites for each CRISPR sequence. To design your custom gRNA sequences:

1. In the search text box, enter the gene name and up to 500 bp in FASTA format. An example is provided below.
2. Select a CRISPR-Cas9 format: GeneArt CRISPR Nuclease Vector, GeneArt CRISPR Nuclease mRNA, or GeneArt Platinum Cas9 Nuclease. Next, select a gRNA format based on your Cas9 format selection.
3. Select a species, either *Homo sapiens* or *Mus musculus*.
4. Click “Search” to initiate *de novo* design of CRISPR gRNAs.

Search | CRISPR Powered by **Thermo Fisher Cloud**



Use our CRISPR guide-RNA (gRNA) *in silico* tool to find the optimal CRISPR sequence for your genome editing goals.

- Use this interface to analyze your sequence of interest and design CRISPR sequences using our unique design algorithm.
- Search results will include the top 6 CRISPR sequences with PAM sites, potential binding sites, exon maps with gRNA locations, and recommendations based on potential off-target effects for each CRISPR sequence.
- The tool will design the correct gRNA format for your preferred CRISPR-Cas9 product format.
- **New** The tool will also design primers to be used with our **GeneArt™ Genomic Cleavage Detection kit** in order to assess cleavage efficiency and confirm edited loci.

For CRISPR designs for sequences not included in our predesigned CRISPR database, enter up to 500 bp into the text box, below. Sequence must be in FASTA format. [?](#)

```
>brca2
ATGCCTATTGGATCCAAAGAGAGGCCAACATTTTTGAAATTTTAAAGACACGCTGCAACAAGCAGATTTAGGACCAATAAGTCTTAATTGGTTTGAAGAAGCTTCTTCAGAAGCTCC
ACCCATAAATCTCGAACCTGCAGAGAATCTGAACATAAAAACAACATTACGAACCAACCTATTAAAACT
```

Select a Cas9 Format: [?](#) Select a species:

Protein - GeneArt™ Platinum™ Cas9 Nuclease (Recommended) Homo sapiens Search

Select a gRNA Format: [?](#)

GeneArt™ Precision gRNA Synthesis Kit

GeneArt™ Platinum™ Cas9 Nuclease is recombinant Cas9 protein. Cas9 protein forms a very stable ribonucleoprotein (RNP) complex with the guide RNA (gRNA) component of the CRISPR/Cas9 system. The GeneArt™ Precision gRNA Synthesis Kit can be used to produce gRNA from target specific oligos. Resulting gRNA can be complexed with Cas9 protein to form RNP prior to transfection.

Search for Predesigns View Selected CRISPRs (0)

Search results

The results from the predesigned gRNA search example are shown below.

- Highlight a row in the “Search Summary” table on the left. The information on the right will update with results for that gene:
 - Predesigned CRISPR sequences target the first three transcribed exons, which are shown in the gene representation at the top
 - Hover over each sequence to see where it falls in the gene representation; an arrow will appear for each CRISPR sequence in the table
 - The top 6 CRISPR sequences are shown with the top 4 recommendations; we recommend analyzing at least 3 for optimal results
 - Predicted off-target binding sites are calculated based on the number and position of mismatches to the target

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Your search has returned gRNAs for 3 genes. Search Again

Search Summary

Gene	Targeting Exons	Chr Location
BRCA2	2,3,4	chr13
CHKA	1,2	chr11
MAPK7	2,3	chr17

Gene BRCA2
mRNA : NM_000059 Total No. Exons: 27 Targeted Exons: 3 (2,3,4)

Detailed view of CRISPRs for BRCA2

- GeneArt™ predesigned CRISPR sequences target the 1st three transcribed exons.
- Top 6 CRISPR sequences are shown with top 4 recommendations. We recommend screening at least 3.
- Predicted off-target cleavage sites are calculated based on the number and position of mismatches to the target.
- Click on "Binding Sites" number to view potential off-target binding sites and mismatches!
- Click on to select forward and reverse primers for Genomic Cleavage Detection (GCD) assay (this may take a few seconds to complete)
- GCD primers will not be added. GCD primers will be added.

CRISPR Sequence	Direction	PAM	Binding Sit.	Recommen.	GCD Primers
TTTACAGGAGATTGGTACAG	(-)	CGG	4	✓	<input type="checkbox"/>
TGCGACAGTGAAAACATAAA	(+)	TGG	8	✓	<input type="checkbox"/>
TGCCTATTGGATCCAAAGAG	(+)	AGG	7	✓	<input type="checkbox"/>
GCAGGTTCAGAATTATAGGG	(-)	TGG	6	✓	<input type="checkbox"/>
GGCCTCTCTTGGATCCAAT	(-)	AGG	12		<input type="checkbox"/>
GAAACCATCTTATAATCAGC	(+)	TGG	13		<input type="checkbox"/>

Add CRISPRs View Selected CRISPRs (0)

- Click on a number in the “Binding Sites” column to view potential off-target binding sites and mismatches. Binding sites for the first CRISPR sequence are shown below.

Binding Sites for TTTACAGGAGATTGGTACAG (BRCA2) (89.57%) ✕

Binding Sequence	PAM	Mismatches	Genomic Location
TTTACAGGAGATTGGTACAG	CGG	0	chr13 [32319274]
AA TACAGTAGATTGGTACAG	GGG	3 [1, 2, 8]	chr1 [196955479]
TTT ACT GGAG CT GGGTACAG	TGG	3 [6, 11, 13]	chr18 [422981]
TTT AGAG AAGATGGGTACAG	TGG	3 [5, 8, 13]	chr18 [28085937]

- Return to the “Search Summary” page and check the box for each gRNA design desired, and click “Add CRISPRs”.
- Select another row in the “Search Summary” table and repeat analysis of the gRNA sequences.
- Once all gRNAs have been analyzed for each gene and added to the project, click “View Selected CRISPRs” for a summary of your order.
- A summary of your project, like the one pictured below, will appear.
- Carefully review gRNA designs before adding them to your cart. Once gRNA designs have been added to your cart, edits cannot be applied. Should you require different gRNA designs, simply remove your project from the cart and return to the web tool for additional designs.
- Confirm gRNA sequences and select any recommended products you would like to include in your order.
- Click “Add Items To Cart” to place your order. Reminder: If your pop-up blocker is set to “on”, the cart will not open. Please turn “off” the pop-up blocker to add your items to the cart for purchase.

Summary

GeneArt™ Platinum™ Cas9 Nuclease

<input type="checkbox"/>	<input checked="" type="checkbox"/>	Added In Cart	Gene	CRISPR Sequence	Direction	PAM	Binding Sites	gRNA Format
<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>		BRCA2	TTTACAGGAGATTGGTACAG	(-)	CGG	4	GeneArt™ Pr...
<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>		BRCA2	TCGCACAGTGAAAATAAAA	(+)	TGG	8	GeneArt™ Pr...
<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>		BRCA2	TGCCTATTGGATCCAAAGAG	(+)	AGG	7	GeneArt™ Pr...

Selected CRISPRs: 3
Selected GCD Primers: 0

Click on to view the designed forward and reverse primers for Genomic Cleavage Detection (GCD) assay

Required Products:

- GeneArt™ Precision gRNA Synthesis Kit (A29377)
- GeneArt™ Platinum™ Cas9 Nuclease 25µg @ 1µg/µL (B25640)

Recommended Products:

- GeneArt™ Platinum™ Cas9 Nuclease 75µg @ 3µg/µL (B25641)
- GeneArt™ Genomic Cleavage Detection Kit (A24372)

Find out more at thermofisher.com/crisprdesign