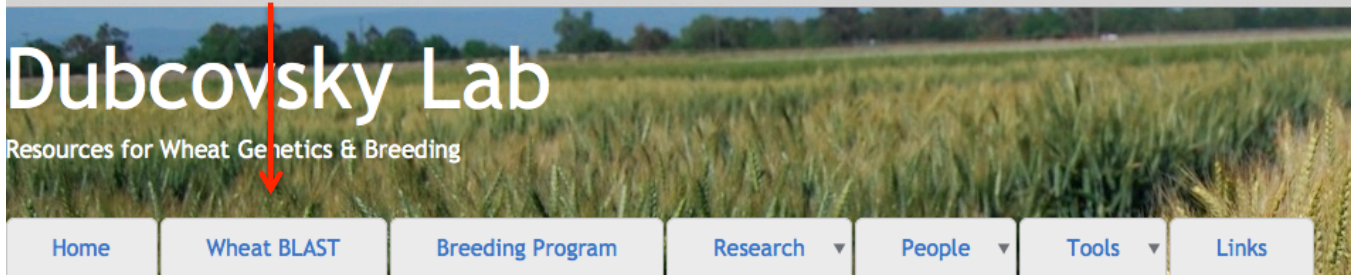


## Tutorial: How to use the Wheat TILLING database

1. Visit [http://dubcovskylab.ucdavis.edu/wheat\\_blast](http://dubcovskylab.ucdavis.edu/wheat_blast) to go to the BLAST page or click on the 'Wheat BLAST' button on the homepage.



### Navigation

- [Sign In](#)

## Home

Welcome to the Dubcovsky Lab's website.

[Jorge Dubcovsky Bio & Publications](#)

## Research projects

2. **A)** Paste in your nucleotide sequence, **B)** select the "TILLING Genomic Reference (4x and 6x)" database, and then **C)** hit the "Basic search" button.

**Basic Search - using default BLAST parameter settings**

Enter query sequences here in [Fasta format](#)  
or [paste an example](#)

**A)** →

Or upload sequence fasta file:  No file chosen

Program:  Database(s): **B)**   
 IWGSC AB + UCW (4x TILLING)  
 IWGSC ABD Genome  
 IWGSC ABD + U (6X TILLING)  
 TILLING Capture Design Padded Exons  
 IWGSCv2.2 Psuedomolecules  
 T. turgidum (Kronos) Transcriptome  
 T. urartu Transcriptome  
 URGI A chromosome  
 URGI B chromosome

**C)** →

And/or upload sequence fasta file:  No file chosen

3. Wait 5-20 seconds while the BLAST search runs. The time is dependent on number of query sequences and the length of the query.

4. When finished, you will be presented with a table of BLAST hit results. Here, **A)** you can view the BLAST HTML output, **B)** download tabular BLAST TSV output, **C)** check the box for contig sequence you want to download, **D)** click on Scores to view the BLAST HTML alignment for a specific hit, and **E)** click a link to view the annotation with possible mutant lines in the same region.

From this page, you can also **download full contig sequences** based on your results.

## BLAST Result

Home About BLAST Help

**A)** [Inspect BLAST HTML output](#)  
**B)** [Download BLAST TSV output](#)

Filter current page by score:  
 Show  for each query sequence

Re-parse current blast results (please select cutoff criterion):  
 Similarity percentage Cutoff %:   
 Blast score **C)** Cutoff score:   **D)**

Retrieve and download subject sequences in FASTA format:  
 Check here to download All sequences... OR select particular sequences of interest below  
 your selection of sequences to download  
 Select/Deselect all

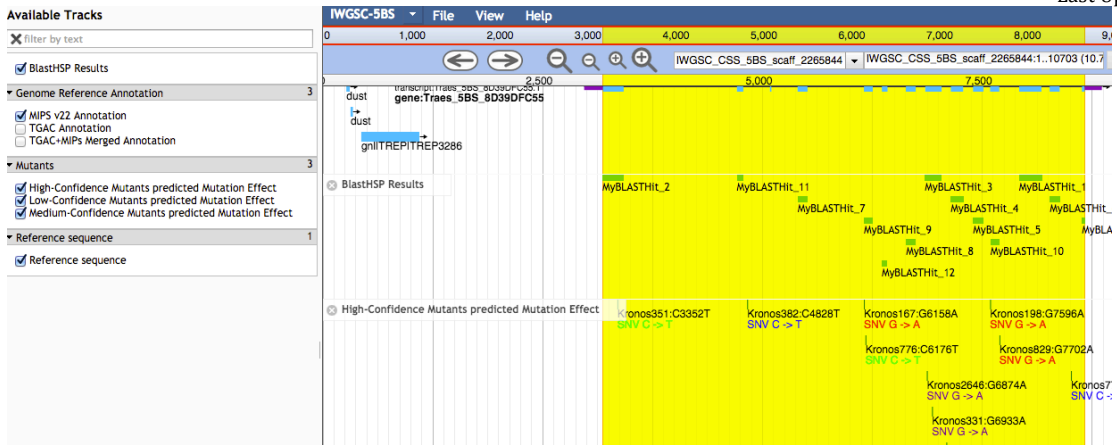
**E)**

Query	Subject	Score	Identities (Query length)	Percentage	Expect	VisualizationLink (If available)	Kronos Mutant Line (confidence)	Cadenza Mutant Line (confidence)
g 33333044 gb AF543316.1	<input type="checkbox"/> IWGSC_CSS_5DL_scaff_1357463	488	270/270 (961)	100.00	1e-134	<a href="#">IWGSC_CSS_5DL_scaff_1357463</a>	0 (high) 0 (intermediate) 0 (low)	56 (high) 5 (intermediate) 16 (low)
g 33333044 gb AF543316.1	<input type="checkbox"/> IWGSC_CSS_5BL_scaff_10869581	426	258/270 (961)	95.56	3e-116	<a href="#">IWGSC_CSS_5BL_scaff_10869581</a>	103 (high) 13 (intermediate) 8 (low)	121 (high) 18 (intermediate) 33 (low)
							37 (high)	64 (high)

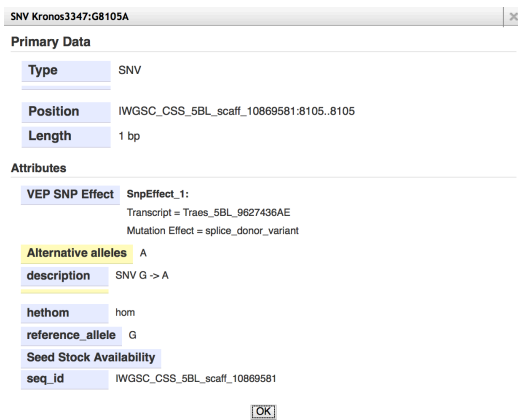
5. After clicking on a visualization link, allow JBrowse to load the sequence and annotation data. This may take anywhere from 5 seconds to 3 minutes depending on connection speed.

Once finished loading, you will have your hit region highlighted in yellow, with your BLAST HSPs depicted by green bars in the BlastHSP Results track.

Below the Blast HSP Results track, we can see the High Confidence Mutants track where Single Nucleotide Variants (SNVs) are color-coded depending on the severity of the variant effect. SNVs that are most severe are colored in **RED** (stop\_gained, stop\_lost, splice\_site changes), medium severity SNVs are colored in **PURPLE** (missense/amino acid change), the least severe SNVs are colored in **GREEN** (synonymous/no amino acid change), and lastly SNVs are colored in **BLUE** for intron\_variants or when the effect is unknown (I.E. in the case of all UCW chromosome assembled contigs or anything that is not annotated in IWGSC).

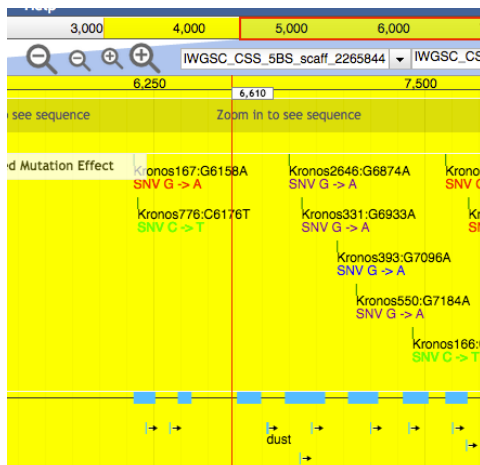


6. When clicking on a particular SNV, an information window will be displayed with details.

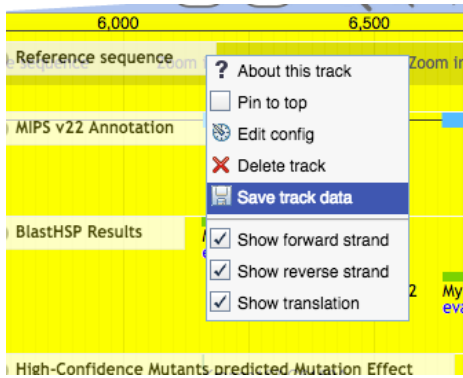


7. In order to **download nucleotide sequence data** for only a particular region, you can use the Reference Sequence track to export the sequence in FASTA format. First, zoom into the region of interest so that it is displayed in the visualization window.

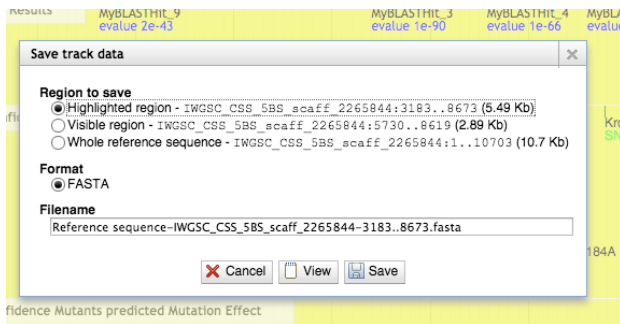
To zoom, hold the <SHIFT> key and use your mouse to click and drag over the exact region you want. You should see a **RED** line with the positional start information after pressing SHIFT.



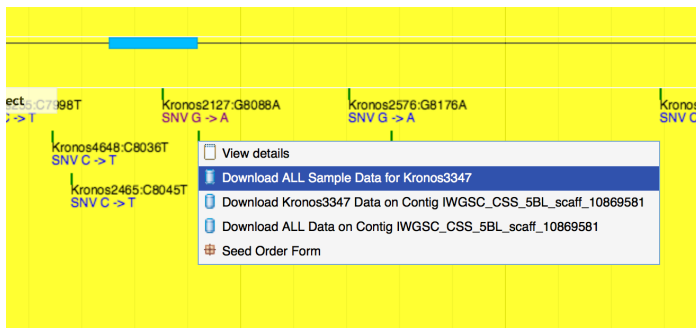
- Then, click on the reference track down arrow and choose 'Save track data'. This track may be at the bottom of the interface.



- You will then be presented with a dialog window where you can choose from a couple of options: Highlighted region, visible region, or whole reference sequence. In this case, since we are trying to grab only a particular sub-region of the highlighted region, we will choose 'Visible region' and then click the 'Save' button



- To download SNP data in TSV format (can be opened in excel), right-click on a SNP for different export options.



## Alternate download option:

- Downloading mutation data can also be searched based on IDs from the Sample Search page: <http://dubcovskylab.ucdavis.edu/wheat-tiling/sample-search>

Filter by Kronos or Cadenza and then click result to download data.

Kronos  Cadenza  Both

Line Name:

Example Search:

Gene Name:

Example Search: Traes\_1AS\_BF353B963.3, Traes\_1

Scaffold Name:

Example Search: IWGSC, IWGSC\_CSS\_1AL\_scaff\_1091068

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Search by any combination of line, gene, or scaffold

Example Search: Traes\_1AS\_BF353B963.3,Kronos2101,Kronos2106, IWGSC\_CSS\_1AL\_scaff\_1091068

Kronos  Cadenza  Both

- The first 3 options (Line Name, Gene Name, or Scaffold Name) will search automatically and present you with a list of options. Clicking on one in the dropdown box will automatically download the mutation data for the search term. Please note the search is CASE SENSITIVE.
- Additionally, the bottom box can be used to paste in mixed ID list to automatically generate one file for all the searched mutation data.