Thank you for using JAX Synteny Browser. The most common use cases for this tool involve uploading genomes of two or more species and explore the relationships of conserved synteny between the two at different levels of depth and detail. Based on a selected reference (aka source) and comparison (aka target or destination) species, users have the ability to search for conserved features by name, function, or phenotype in the reference genome and can investigate the corresponding matched features within the comparison genome. This manual will explain how to use each of the features in Synteny Browser and then provide a few user scenarios.

2 : Tool Panels and Views

2.1 : View Settings

On the right-hand side of the browser window, you'll see a blue sidebar with a gear icon at the top. Upon clicking anywhere on the sidebar, it will open a panel with some settings:

**The reference species (Figure 1, #1):** (default *Mus musculus*); if there are only two genomes uploaded, when the reference species is changed from the dropdown menu, the comparison species will automatically become the other species. The Genome View will automatically reflect the current state of the two selections.

**The comparison species (Figure 1, #2):** (default *Homo sapiens*); if only two species are uploaded, the only way of changing the comparison is to change the reference. If there are more than two species loaded, the comparison dropdown menu will have options to choose from.

**The reference genome interval (Figure 1, #3):** allows the user to focus on a specific region on a particular chromosome. In order to use it, enter a genomic range with the following format: "Chr<chromosome>:<starting bp>-<ending bp>" or, if you'd like, you can leave off the leading "Chr". Clicking the "Update View" button will automatically scroll the page to the Block Detail View and will load your specified range.

**True orientation (Figure 1, #4):** (applies to Block Detail View only) if checked, will show the reference and comparison syntenic blocks with accurate orientation. Some syntenic regions are located on different strands. By default, you will see the syntenic regions in the orientation that lines up genes as closely as possible. If a comparison block is not truly oriented, it will be indicated with red crossed lines between the reference and comparison blocks in the Block Detail View.

**Gene symbols (Figure 1, #5):** (applies to Block Detail View only) if checked, will show gene symbols regardless of the visible interval within the Block Detail View. This can add a lot of visual clutter if the interval is large and by default, gene symbols will only appear if you are viewing an interval of 5 Mb or less unless you have highlighted features selected, in which case, the features' symbols will always be visible in red.

**Anchors (Figure 1, #6):** (applies to Block Detail View only) if checked, will show dotted lines at the edges of the homologs. This can help create a more visual map of where genes are in the reference and comparison if not exactly lined up, but can also create some visual clutter so by default, anchors are hidden.

2.2 : Feature Search

The Feature Search panel allows you to search for genomic features such as QTLs and individual (or groups of) genes, by gene symbol, or ontologies containing function and phenotypic annotations. Users can select the desired search type (Figure 2, #1) from the drop down in the upper left-hand side of the panel, which will also specify which species the search will browse from. The Feature Search will always search and display results for the reference species, which can be changed from the View Settings (see 2.1) at any time. The search term (Figure 2, #2) input will display suggestions as the user types. For some search types, like ontologies, where hundreds to thousands of results may apply, if the search term is to broad, there will be a message displayed that a more specific search is needed in order to display results. The results table (Figure 2, #3) will be where the results of the search appear. You can filter this table with the input on right, above the table and may select as many of the results as you wish. Once you have some selections you wish to view, click one of the view buttons to see where your searches map on the genome view.

You may perform as many searches as you wish and continue to select them, however if you make selections for a search but do not click view before performing another search, your unviewed selections will be lost.

2.3 : Genome View
The Genome View is a way for you to visualize the entire genome of the reference species. The outer ring of the circos plot (Figure 3, #1) represents the reference genome and is made up of a number of segments, one for each of the autosomes and sex chromosomes.

Each of the colored bands within these segments indicates a syntenic region between the two species with the mapped chromosome region of the comparison with that color. White bands within these segments marks a lack of conserved synteny in that area on the reference. The inner ring of the circos plot (Figure 3, #2) represents the comparison species genome using a color legend. Similar to the outer ring, there is a segment for each of the comparison autosomes and sex chromosomes.

If you click on one of the reference chromosomes in the outer ring, you will see how the regions on that chromosome map to those in the comparison genome with arching threads (Figure 3, #3) of scaled width and accurate location, both in the reference and comparison. If you've chosen to view specific features from the feature search in the Genome View, a red circle (Figure 3, #4) will indicate the reference chromosome where one or more of your selected features are located and a colored band will be protruding from the chromosome (sometimes these bands are quite thin). Hovering your cursor over the red circle will show you feature information such as the gene symbol(s) or QTL(s) that you've selected that are located in that chromosome. Clicking on the red circle will show mapping of the respective chromosome in the inner circle, though will only show the mapping of syntenic regions that contain at least one selected feature. If you've selected to view QTLs, the regions displayed will be all regions the QTL spans.

2.3.1 : Viewing a selection

Once either a chromosome or set of features are selected (selections/chromosome is mapped with arching threads across the center of the plot), clicking the view button will trigger the rendering of the associated region of the reference genome in the Block Detail View.

2.3.2 : Downloading the Genome View

If you have a set of features and/or a mapping you wish to save as an image, the download button will initiate two downloads, the first being the actual image and the second, a legend, will depict the information conveyed through the hover tooltips so it will break down the features (and the type, gene or QTL) under their respective chromosome in the reference. Since two images start downloading simultaneously, your browser may ask you to allow the site to download multiple files at once. **You must give the site permission to do this in order to get any images.**

2.3.3 : Clearing the plot

If you have selections and wish to clear them, clicking the clear button at the bottom left-hand side of the panel will remove any features you had previously selected to visualize. Clearing the plot will not clear the selections that are shown in the current feature results table (e.g. if you had previously selected (and clicked 'View') several genes using Mouse Gene Ontology identified with 'BMP signaling pathway', but then selected a subset of the results from a search by gene symbol 'Trappc' and wanted to clear the genome plot of your selections, your ontology-related selections will be lost in the feature search table but your gene symbol-related selections will remain).

2.4 : Block Detail View

The Block Detail View displays and allows you to navigate the syntenic regions on a selected chromosome and features in an interactive way. Once a region is rendered, you'll see three major elements, a series of colored blocks above a linear scale, a set of two sequences of blocks stacked on top of each other and a color key at the bottom.

The blocks on the scale is the chromosome-wide view of the selected chromosome (Figure 4, #1). These blocks will remain static as they show blocks that may not be visible in the actual browser. You may notice that there's a rectangle that covers a span of the chromosome; this rectangle is the view box and represents the section of the chromosome that is visible in the browser. Red ticks above the blocks in the chromosome view represent locations of selected genes and hovering over the line will show you a few details about that gene. Dark purple lines above the blocks indicate QTL spans and hovering over the line will show details about that QTL. Red ticks below the blocks indicate locations of homologs of the selected features in the reference. The linear scale below the blocks help approximate locations of features and syntenic regions. The chromosome view is interactive; you can click and drag the view box across the chromosome, draw a new view box by clicking and dragging in a different location from the existing box, drag box edges to expand or contract one of the sides. If you have scrolling ability, you can also scroll over the chromosome view to zoom the view box in and out. The browser will update whenever you change the size or location of the view box for quick navigation. Above the chromosome view is a set of navigation buttons (Figure 4, #2) which can also be used to move the view. From left to right: move left, zoom in, go back to view when initially loaded (the whole chromosome if no features were selected or a view of all selected features), zoom out, move right.

The color key at the bottom of the panel (Figure 4, #3) is the color key for the comparison genome. Comparison chromosomes that have at least one syntenic region mapping to the selected reference chromosome will have a vibrant color and label, while chromosomes that are not represented in the reference chromosome will be faded. Additionally, if you hover over a present chromosome in the key, all syntenic regions except for those of that chromosome will become grey, making it more obvious where the syntenic regions of that chromosome are located over the entire chromosome or in the browser view.

The actual browser (Figure 4, #4) is where you'll be able to see sizes and locations of features. At each of the corners of the browser are coordinates that indicate where on the reference chromosome the view is and the coordinates on the comparison regions. Note that, unless you have the true orientation checkbox checked from the settings panel, comparison coordinates of some regions may increase right to left rather than left to right. Labels for the species being compared are on the far left-hand side of the browser, lining
up with two tracks. Regions that are large enough to display them have start and stop coordinates and those that are too small can be hovered over to display the coordinates. The top track (Figure 4, #5) represents the reference species. This track is a full representation of the reference chromosome, including features that may not occur in syntenic regions. The track below the reference (Figure 4, #6) represents the comparison species genome. The syntenic regions of the comparison track are colored to map to the chromosomes in the comparison species genome in which they occur.

Genes in the browser have their symbols hidden by default (this can be changed in the settings panel) until the visible region in the browser is less than 5Mb to assist with visual clutter, in which case, exons will also become visible. QTL regions will be a translucent purple and any selected genes will be highlighted in red and their symbols will be always be visible. Hovering over a QTL or a gene will show more information about the feature in a tooltip (Figure 4, #7). Clicking on a gene will open a "sticky" tooltip (Figure 4, #8) that contains link(s) to other resources such as NCBI or MGI. Moving the browser or clicking elsewhere will dismiss this tooltip. Hovering over a gene with at least one homolog in the comparison genome will also highlight those homologs.

Unless you have the true orientation option checked from the settings panel, you may notice that there are some syntenic regions that have a red cross (Figure 4, #9) between the reference and comparison. This indicates that the comparison block is reversed to visually match up better to the reference block. If you wish to view conserved synteny with true orientation, be sure to check the appropriate box in the collapsible settings panel on the right of the window.

In the upper right of the panel is a colored status bar (Figure 4, #10). If an interval is rendering, it will appear a yellow-orange with a loading message. Once done, it should turn green with a "done" message. If an error occurs in loading or while interacting with the browser, it may turn red, indicating an error. If this happens and affects your interactions and/or navigation, clicking on the bar will open a separate window with an error log. If you wish to report a problem or bug, you can click the "Download Log" button at the top of the window which will download a text file which can be attached to a bug report or email (it will help a software engineer more easily diagnose the issue).

If you wish to download the current view in the browser, the download button in the upper left of the panel will download a snapshot of the browser (the image will contain the chromosome view as well), which could take a several seconds, and a second image that contains the color key. Since two images start downloading simultaneously, your browser may ask you to allow the site to download multiple files at once. You must give the site permission to do this in order to get any images.

2.5 : Feature Display Filters

Synteny Browser has the ability to filter/highlight features by gene ID/symbol, type, or ontology using Feature Display Filters. If you're looking to search for genes individually, using the first filter criteria (Figure 5, #1) will be the most helpful. You can start typing a gene symbol or id, and hint menu will appear to give you suggestions. At this time, you can only search for one gene using this criteria. If you wish to filter an entire gene type, you can use the second filter criteria (Figure 5, #2) which contains a list of gene types and the number of genes within this list. You may select one or more of these types by shift clicking on the types you want. The types can also be applied to only in the reference chromosome, only in the comparison genome, or in both. The third filter criteria (Figure 5, #3) can be used to filter by ontology. The ontology filter contains a dropdown menu where you can select which ontology you wish to search, a term you want to search for, and whether you want to look for genes that match your ontology term in just the reference, just the comparison or both. Finally, if you're using more than one filter criteria at a time, you can apply different combinations of these filters using the selection options (Figure 5, #4). With this tool, you can decide whether you want a union of the filters you've entered or perhaps an intersection.

Once you're ready to see the results of the filter, clicking the run button will run the search and the appropriate filters and display the results in the table on the right (Figure 5, #5). This results table can be filtered and downloaded, if desired. Once the operation is complete and if there was at least 1 result in your filter search, if you scroll back up to view the browser, you should see blue tick marks along the chromosome view (Figure 5, #6), each one representing one of the filtered features. Hovering over any one of these tick marks will show the details for that particular feature. Additionally, if the feature you've chosen is in your current view within the browser, the filtered genes will be highlighted in blue (Figure 5, #7). Back in the Feature Display Filter panel, you have the option to hide all features that don't match the filter (Figure 5, #8). This option will hide all non-colored features in the browser (any selected features in red will remain as well as any QTLs). If you want to clear the filtered features from the browser, the clear button in the selection options section of the filter tools.