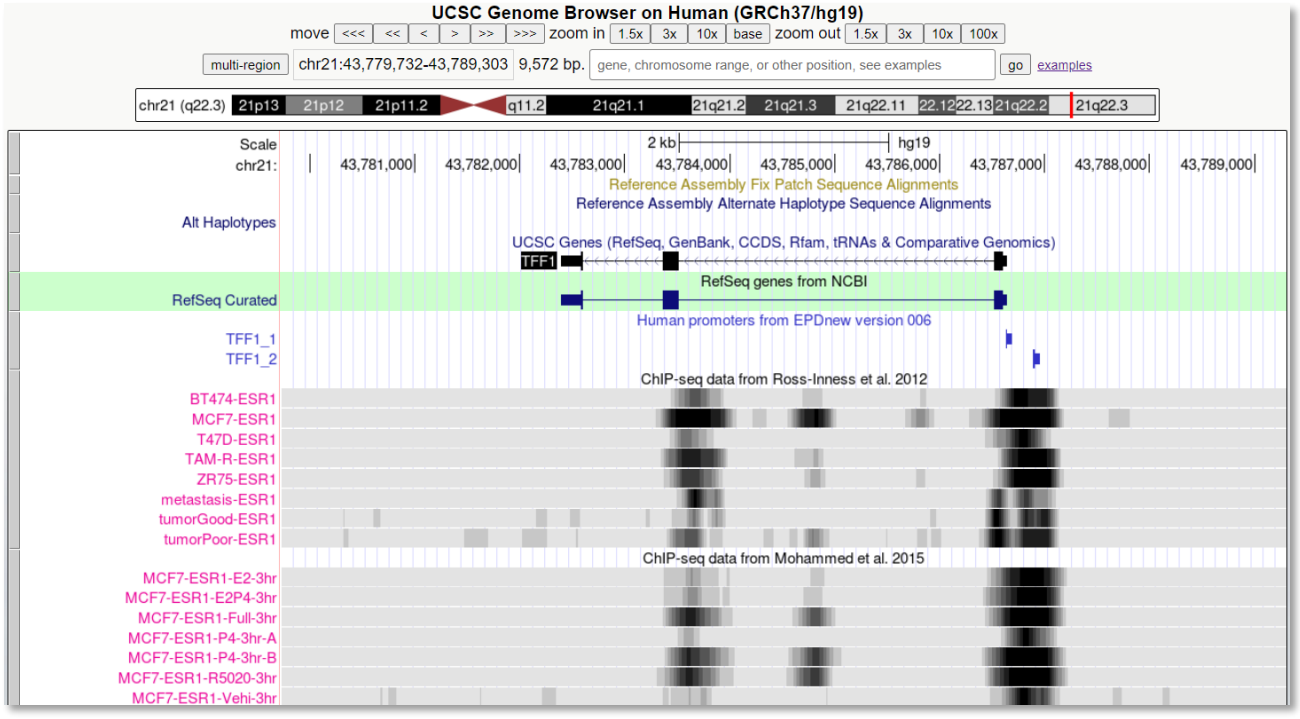
BREAST CANCER HUB USER MANUAL (v1.3)

The Breast Cancer Hub (BC Hub, https://ccg.epfl.ch/bchub/) is embedded in the UCSC genome browser (https://genome.ucsc.edu/) and integrates multiple breast cancer-related datasets for visualization of transcription factors (TFs) cistromes and epigenetic datasets with a standardized pipeline that allows direct comparison. Datasets displayed in the BC Hub are gathered from published studies and include epigenetic data from breast cancer cell lines, surgically-removed breast tumors, patient-derived xenografts, normal breast tissue specimens. Additional tracks add further information such as annotated promoter regions, chromatin accessibility, TFs footprints, and sequence conservation.

Navigate the genome browser

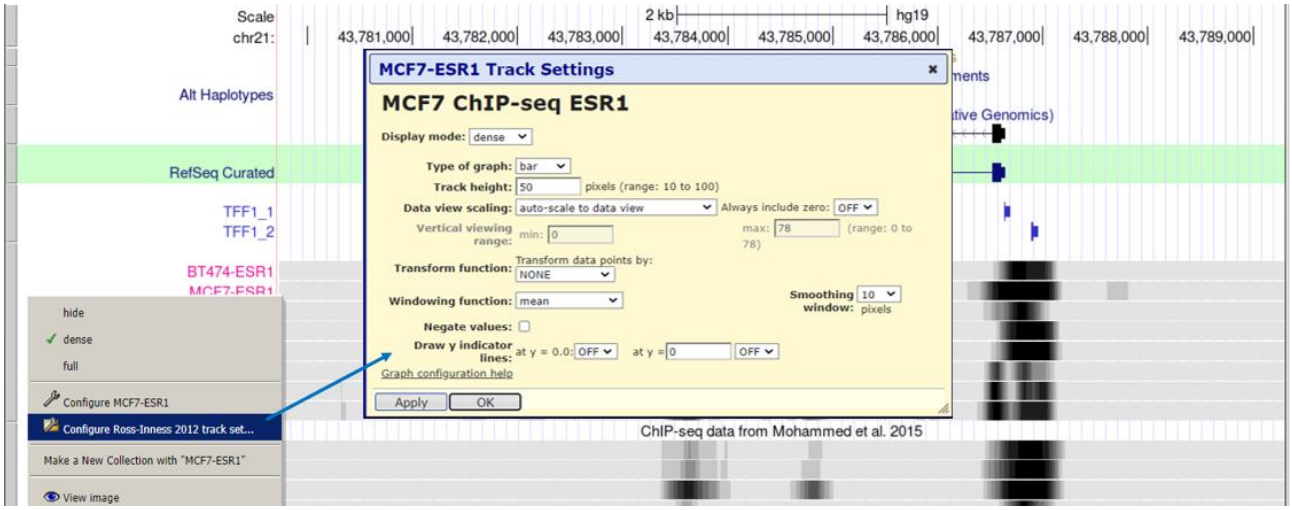
The BC Hub is integrated in the UCSC genome browser and can be accessed from here -> https://ccg.epfl.ch/bchub/.

On the top right on the main browser page the search bar can be used to type in specific genes or indicate the genomic coordinates of a desired region. For example, typing the following text "chr1:1,000,000-1,000,500" will make the browser display to a 500 bp wide region starting at the position 1,000,000 of chromosome 1. Regions of interest can be moved by click-and-drag in the browser window, or zoomed in and out with the zoom buttons. For further general instructions on how to use the UCSC genome browser, please refer to the UCSC genome browser user's guide found at https://genome.ucsc.edu/goldenPath/help/hgTracksHelp.html.



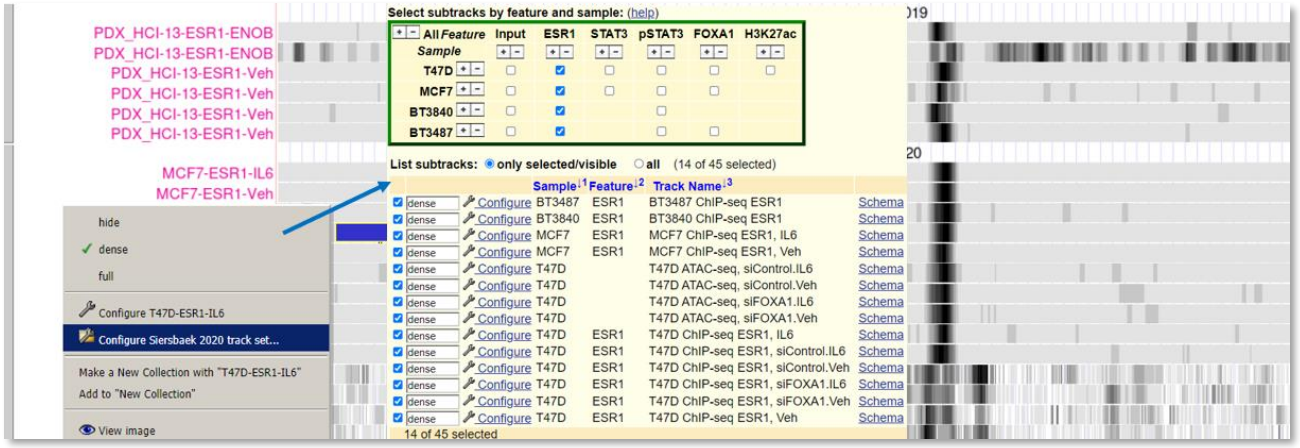
Customize the track order and view

Tracks can be moved up or down to a desired position by dragging the left side of each track. Display settings for each data track can be changed individually by right-clicking on the track itself. By choosing to configure a single track, a pop-up window will appear (as shown in the snapshot here below) to allow users to select the desired display parameters. Individual tracks can be viewed in full or dense mode, or can be hidden from the genome browser (they can be retrieved at any point from the full list of available tracks). Further display settings such as track height, scale, and smoothing window can be changed in the track configuration panel. More information about track settings can be found at https://genome-euro.ucsc.edu/goldenPath/help/hgWiggleTrackHelp.html



Configure the track list

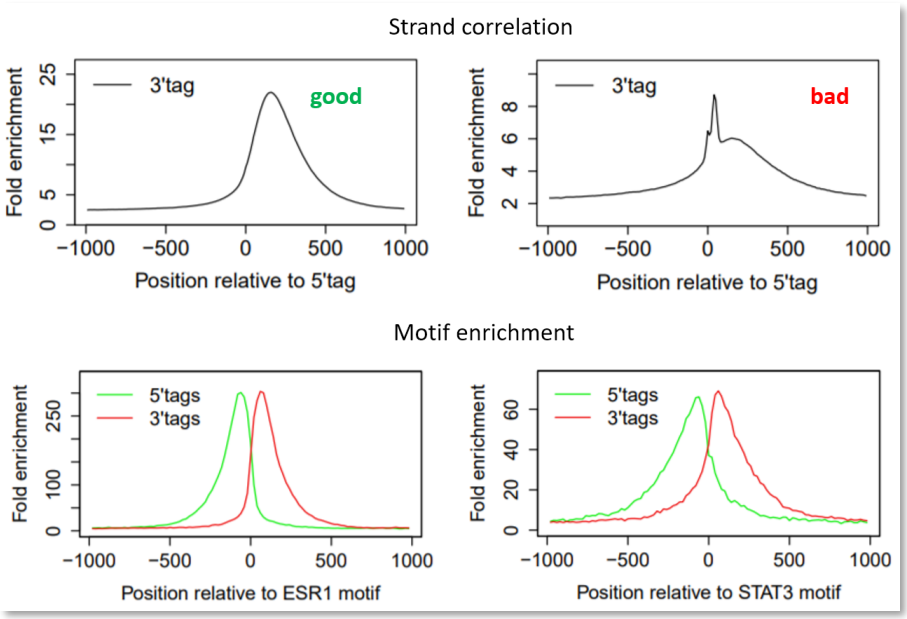
To configure an entire track set, users can select the "configure track set" option in the pop-up window. This will open a new page showing the full track setting options as well as a description of the data track. Subtracks from a given dataset can be selected in the track set configuration settings. Tracks can be selected/unselected individually in the bottom panel, or selected based on properties such as sample type and/or feature in the upper panel.



Retrieve sample information and quality control

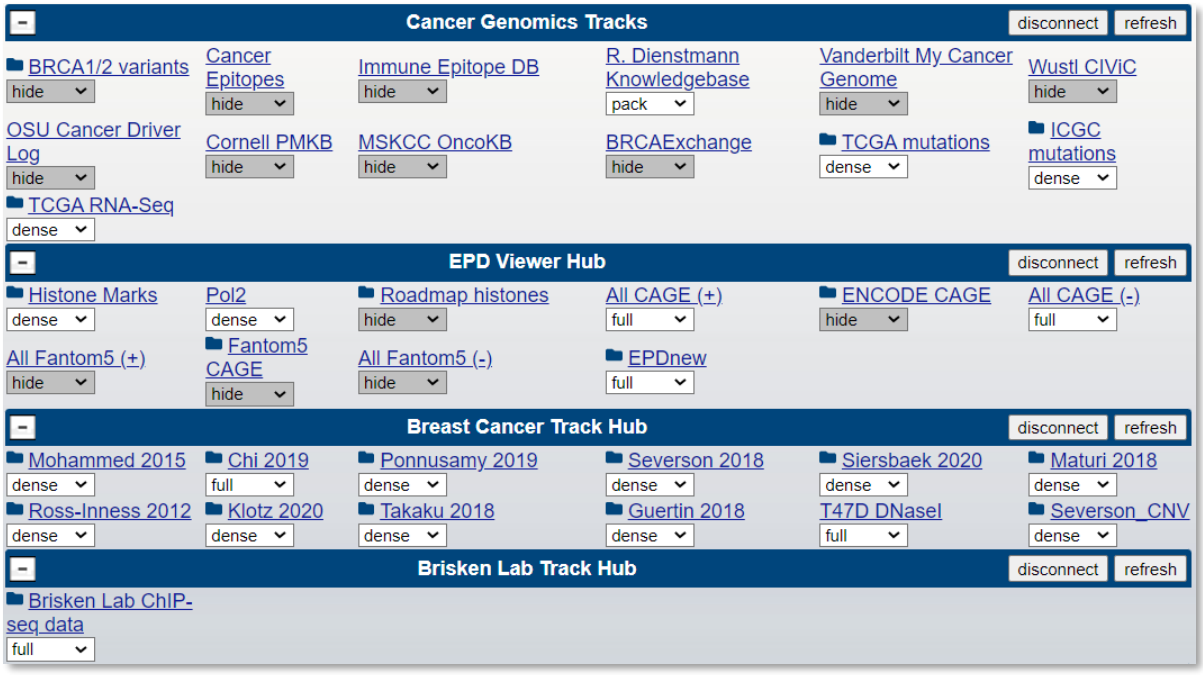
The description of the experiments, the pipeline used for data processing, and references to the original study can be found by scrolling down the track list configuration panel. Furthermore, quality controls reports (according to ENCODE guidelines. PMID: 22955991 https://pubmed.ncbi.nlm.nih.gov/22955991/) can be viewed and downloaded for all the samples processed via the BC Hub standard pipeline. The strand correlation plots represent the distribution of the plus and minus strand reads respectively up-stream and

down-stream of the protein-bound regions. The cross-correlation plot typically produces two peaks: a peak of enrichment corresponding to the predominant fragment length (highest correlation value) and a peak corresponding to the read length ("phantom" peak). Ideal distributions are narrow and sharp, while the presence of shoulders or less-defined shape of the distribution might reflect poor sample quality or technical issues (see examples below). The second quality control report, often named motif enrichment analysis, displays the distribution of positions of the plus and minus strand fragments relative to the sequence motif of the immunoprecipitated transcription factor (for ChIP-seq data). Failed experiment and inputs tend to have flat or noisy distributions.

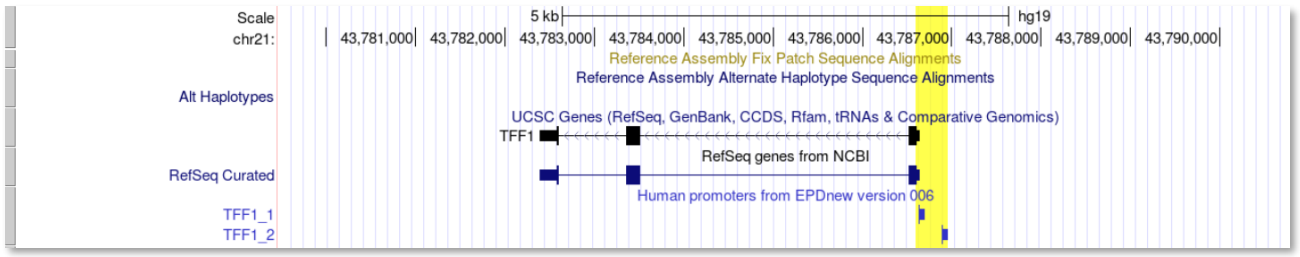


UCSC integrated tracks

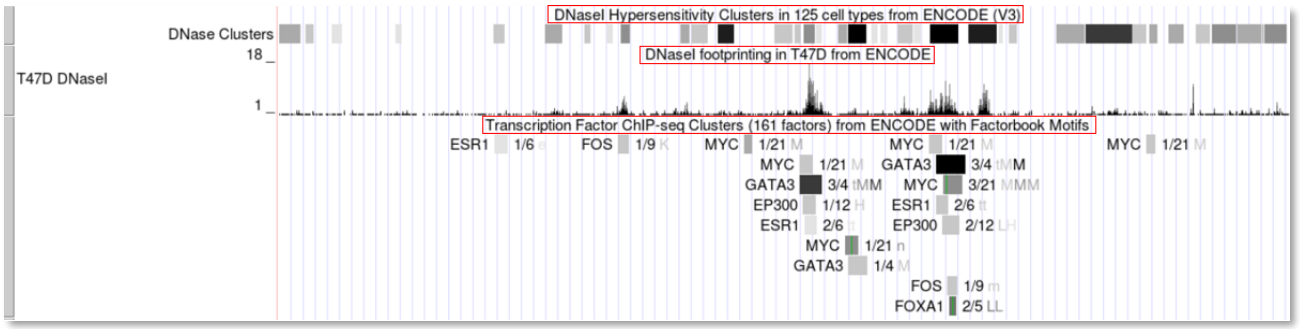
Additional tracks from various sources are included in the BC Hub and can be toggled via the UCSC track manager menu (shown here below), which is located in the main browser page just below the displayed track sets.



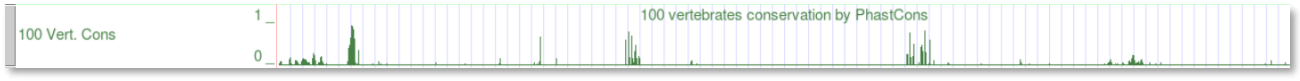
- Eukaryotic Promoter Database (EPD) track (https://epd.epfl.ch//index.php https://academic.oup.com/nar/article/41/D1/D157/1070274) includes a collection 29598 human promoters that have been experimentally validated.



- ENCODE tracks (https://genome.ucsc.edu/ENCODE/): The Encyclopedia of DNA Elements (ENCODE), loaded by default in the UCSC genome browser, is an integrated database of functional regulatory elements in the human genome. BC Hub sessions includes tracks from the ENCODE project. The "Transcription factors ChIP-seq cluster" tracks display transcription factor (TF) bindings derived from ChIP-seq experiments. Binding sites are represented by rectangular boxes, color-coded in grey scale according to the binding strength. A green vertical line appears in the TF binding box when the predicted sequence motif of the TF is recognized in the bound region, implying that the DNA binding is likely to be direct, rather than tethered by other proteins. Right-clicking on the TF binding box redirects the browser to the page containing metadata such as sample information, experimental condition, and sequence motif, if present. TFs displayed in the track can be filtered in the track configuration window. Other ENCODE tracks, such as "DNase Clusters"and "T47D DNase I" tracks, provide information on chromatin accessibility based on hypersensitivity to DNase I cuts. Peaks represent more accessible genomic areas.



- Conservation track (PhastCons): the sequence conservation track displays the degree of sequence conservation among 100 vertebrates species. Highly conserved sequences in non-coding regions are more likely to carry essential functions in genes regulation, thus are areas of interest in epigenetic studies.



What can you do on the Breast Cancer Hub?

Here few of the many scientific questions that can be addressed by exploring the BC Hub:

- *Is the TF binding consistent across different breast cancer cell lines?*
- *Is the TF binding dependent on hormone treatment?*
- *Is the TF binding event shared between normal breast tissue and tumor, or between male and female breast cancer patients?*
- *Does the region correspond to an experimentally-validated promoter?*
- *Are there other TF associated with the genomic region? Is their sequence motif present in the region?*
- *Is the chromatin region in an open, accessible state?*
- *Is the sequence of the region conserved across vertebrates?*