

IGV practical

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1 Introduction

This tutorial will give you a short introduction into using IGV (Integrative Genomics Viewer) to visualize next-generation sequencing data and annotations. We will use the same ChIP-seq dataset we have aligned and peak called in the previous practicals.

2 Loading IGV and basic navigation on the genome browser

1. Open IGV and select the genome build we were using for the alignment in the top left corner (“Human (hg38)”).
2. Select a single chromosome (eg. chr3).
3. Navigate to chr3:39,000,000-43,000,000.
4. Double click on one of the genes to recenter the window on that gene.
5. Navigate to your favorite gene (or ZMAT3).
6. Zoom out to see the surrounding genes.
7. Right click the left side panel next to your genome track and select “Expanded”.

3 Loading your data into IGV

1. Now click File > Load from File... and load the ChIP-seq data files that you generated:
 - the .bam and .tdf files for tp53 and the input (from the ~/Course_Materials/ChIPseq/Alignment_BWA/ folder)
 - the .narrowPeak and bedGraph (ending with _treat_pileup.bdg) files for tp53 (from the ~/Course_Materials/ChIPseq/Peaks/ folder).
2. Visualize all chromosomes - you will see that in our data we only have reads and peaks on chr3.
3. Navigate to ZMAT3.
4. Select the two .tdf files, right click them and select “Autoscale”.
5. Right click them again and select “Set Data Range...” and click OK. (This will set the two tracks to a common scale making it easier to compare signal in the tp53 sample and input files.)

6. Now set the same scale for the two .bam files as well.
7. Set different colours for the tp53 sample tracks and input tracks (Change Track Color (Positive values)...).
8. Zoom out to see the signal of the surrounding genes.
9. Compare the signal in the two files you have loaded.
10. Bookmark this region:
 - Go to Regions > Region Navigator. Click Add, and give your region a name (eg. MyFirstRegion) in the "Description" field. Click "View".
 - This way if you navigate somewhere else on the genome you can always easily access this region from Regions > Region Navigator.

4 Loading data from server

1. Go to File > Load From Server... and select "GC %" from Annotations.

5 Running IGVtools (generating .tdf file)

1. Click Tools > Run igvtools... .
2. Leave the command on "Count" and select the tp53 bam file as input file.
3. Change the output file name so that you don't overwrite the already existing .tdf file that you generated using the command line version of igvtools on Monday.
4. Click Run. This will generate a .tdf file, that you can load later.