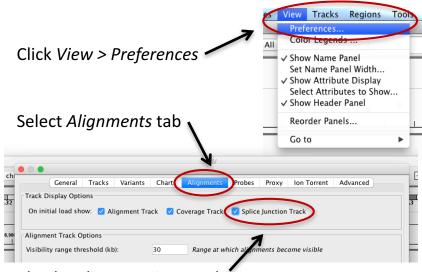
Compute coverage track

1. Set preferences for viewing RNA-seq data



Check Splice Junction Track

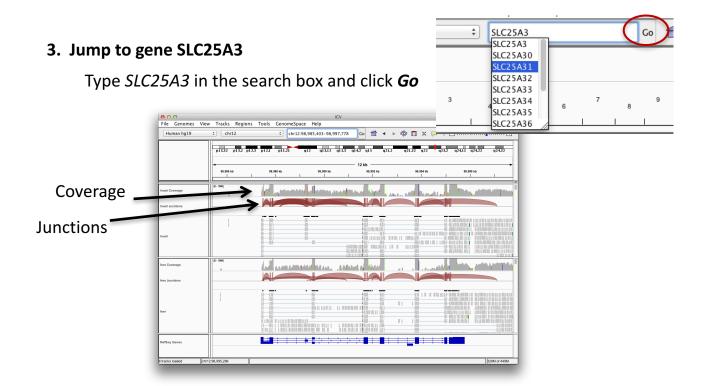
2. Load data

Select *Human hg19* from the genome dropdown menu



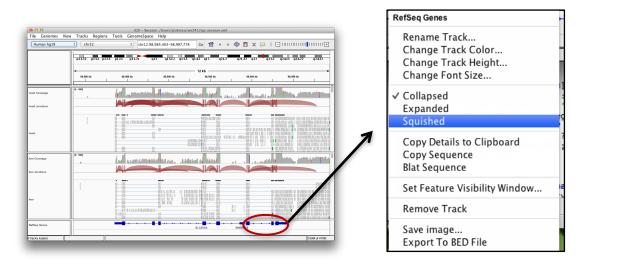
Click File > Load from Server

Open the *Tutorials* menu (Use on Mac, and on Windows) and click on RNA-Seg (Body Map) and then click on OK **Available Datasets Available Datasets** Annotations The Cancer Genome Atlas (i) ENCODE Body Map 2.0 (Illumina HiSeq) Make sure you only open ☐ 1000 Genomes (i) the Tutorials menu. Do **Tutorials** not check the box next to UI Basics (Encode) Tutorials. That will select RNA-Seq (Body Map) everything under Heart Tutorials, but we only Liver want RNA-Seg for this Variants (1000 Genomes) exercise. Cancel OK



4. Expand gene track to see isoforms

Right-click over the RefSeq Genes track, and select Squished



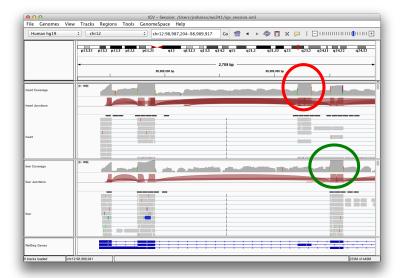
5. Zoom in on first 3 exons

Click and drag in ruler region over area shown



6. Note evidence of alternative splicing.

Observe which isoforms in the RefSeq track are expressed in each tissue.



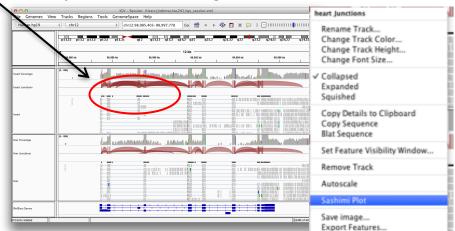
7. Zoom back out to view whole gene

Click the back button in the command bar to zoom out to previous view

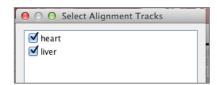


8. Open Sashimi plot

Right-click over junction track or alignments and select "Sashimi Plot"



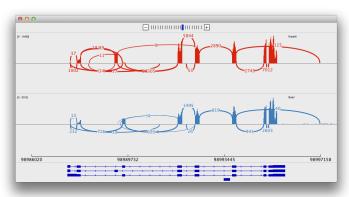
Verify both heart and liver are checked, and click OK



9. Examine Sashimi plot

Note:

- Arcs represent reads spanning exon junctions
- Peaks represent exon coverage



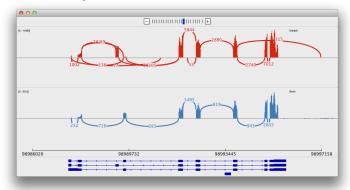
10. Filter out low-count splicing events

Right click over red (heart) track and select **Set Junction Coverage Min**. Enter **50** and click **OK**.

Repeat for blue (liver) track.

Junction Coverage Display Set Exon Coverage Max Set Junction Coverage Min Set Junction Coverage Max Set Color ✓ Show Exon Coverage Data ✓ Text Circle None ✓ Combine Strands Forward Strand Reverse Strand Save Image...

11. Compare with non-filtered view



12. Zoom in on 5' end

Click "+" button 2 times

Click-and-drag tracks to the right to bring the first 3 exons in view.

13. Observe the alternative splicing of the 3rd exon

of the 3rd exon

Compute
coverage -

page 4

