How to Use NeuroTri2-VISDOT

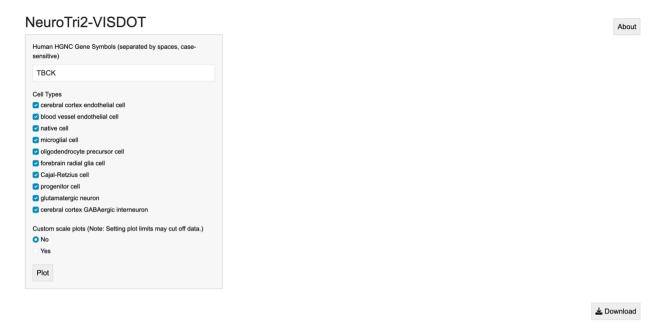
NeuroTri2-VISDOT is an interactive online tool to visualize data from the second trimester human brain development dataset published by Bhaduri et al.

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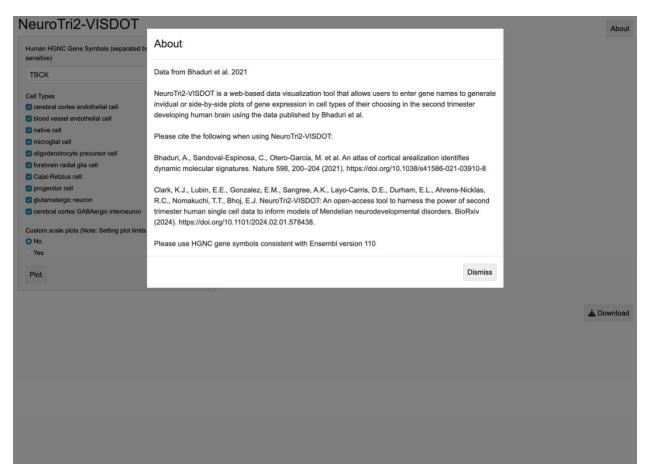
Access

NeuroTri2-VISDOT can be openly accessed at bhojlab.shinyapps.io/NeuroTri2-VISDOT. Code and associated documentation can be found at github.com/kellyclark1/VISDOT. When you visit the web page for NeuroTri2-VISDOT, this will be the first screen you see:



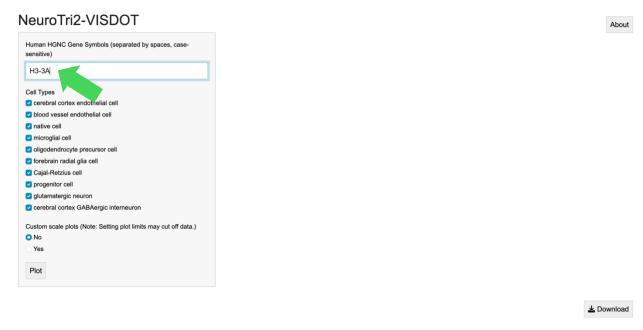
About

When you click the "About" button in the upper right, you will see a pop-up box that contains gene name instructions and current citation information. Please check this tab for information about how to cite the tool and the dataset.

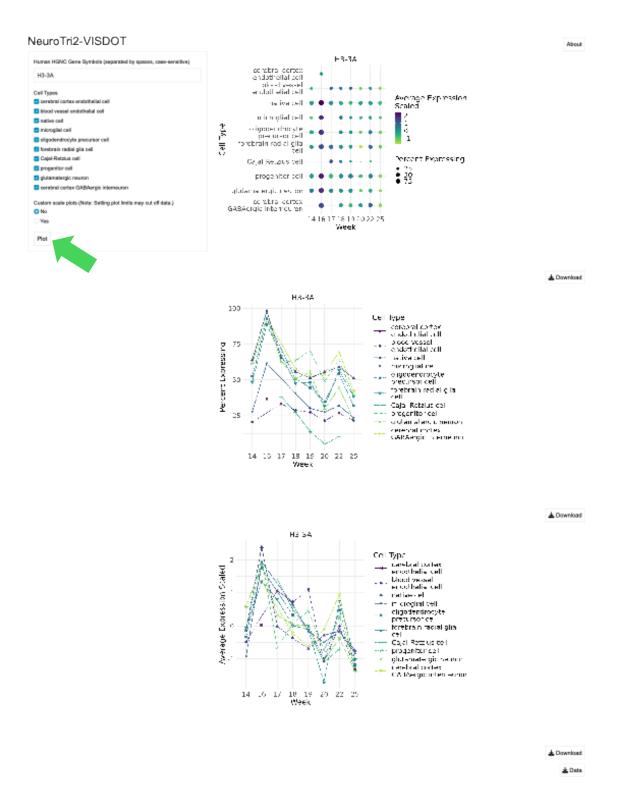


Plot a Gene of Interest

To plot a gene of interest, type in the gene name as the HGNC symbol according to Ensembl version 110. Here, we will use the histone H3.3 gene H3-3A as an example (formerly H3F3A).

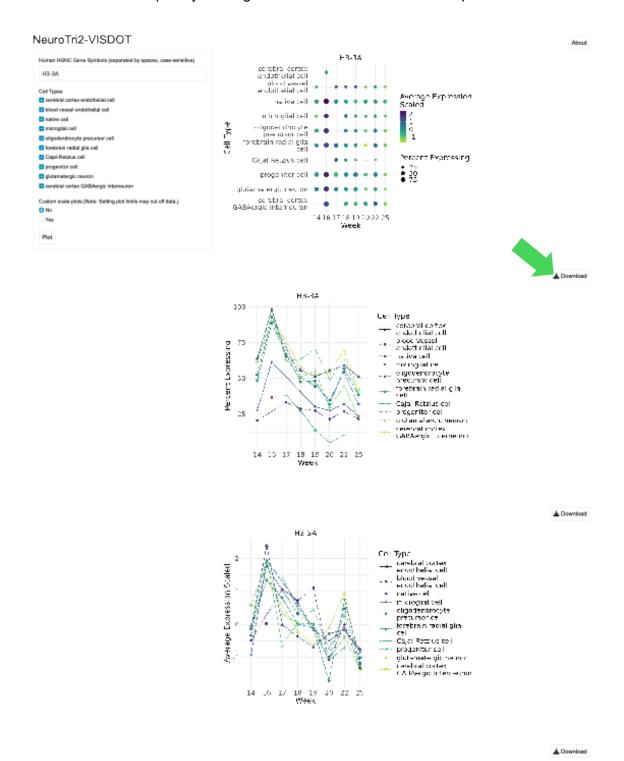


When we click plot (you must click- the enter key will not generate plots, we see each of the three plots like this:



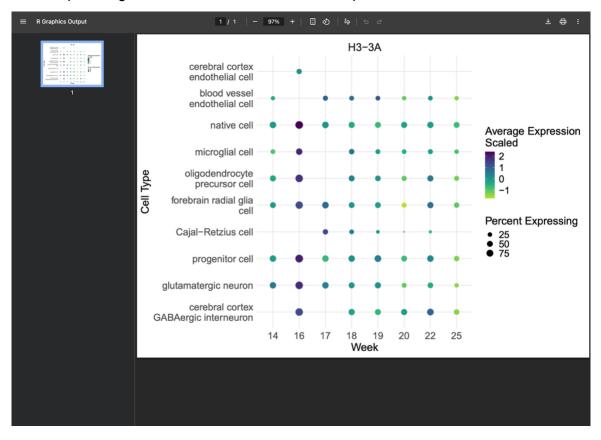
Download Plots

We can download each plot by clicking the download button under that plot:



& Date

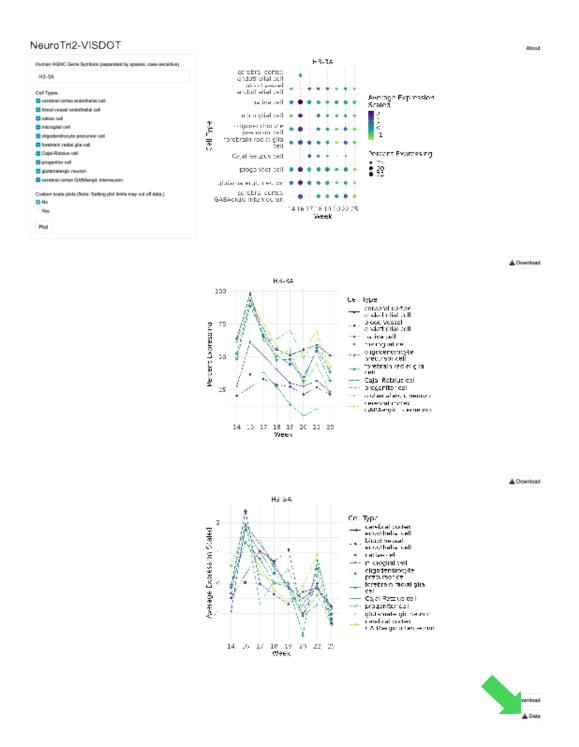
For example, we get this PDF if we download the dot plot:



Plots will be automatically scaled according to the number of genes included.

Download Data

If we want to download the data for our gene of interest, we can use the "Data" button at the bottom of the page to download a comma-separated, spreadsheet-compatible CSV file:

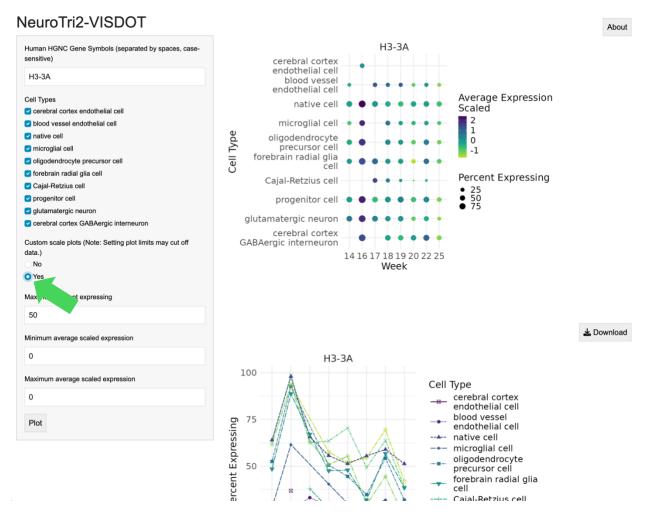


We get a CSV that looks like this, with the gene, cell type, week of prenatal development, percent of that cell type expressing the gene, and the scaled average expression:

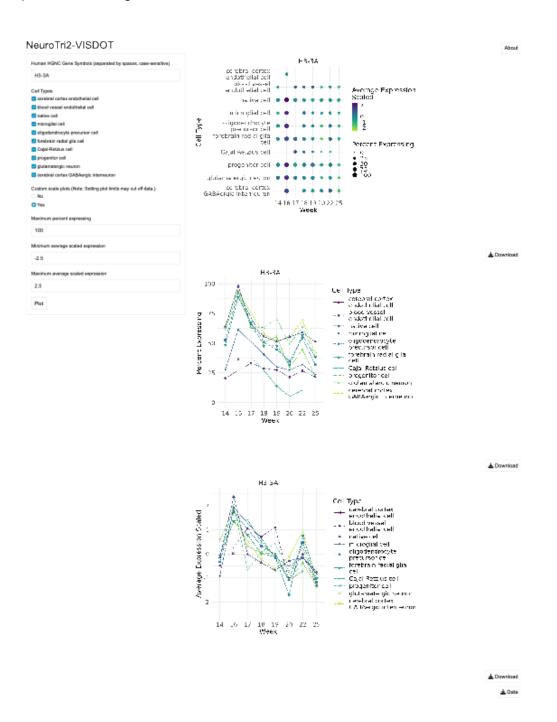
	Α	В	С	D	Е
1	gene	cell.type	week	pct.exp	avg.exp.scaled
2	H3-3A	progenitor ce	19	70.4519481	0.44029863
3	H3-3A	progenitor ce	14	63.1679389	-0.1374569
4	H3-3A	progenitor ce	17	62.5	-0.6634694
5	H3-3A	progenitor ce	20	49.256218	-0.7373942
6	H3-3A	progenitor ce	25	39.1126495	-1.270602
7	H3-3A	progenitor ce	22	63.7869822	0.26860174
8	H3-3A	progenitor ce	18	63.5143319	0.07234076
9	H3-3A	progenitor ce	16	98.7490465	2.0276813
10	H3-3A	cerebral cort	19	52.1129608	-0.6136513
11	H3-3A	cerebral cort	20	54.5121056	-0.1146702
12	H3-3A	cerebral cort	25	41.9718043	-1.3491034
13	H3-3A	cerebral cort	22	69.5599022	0.95002486
14	H3-3A	cerebral cort	18	57.6297616	-0.227546
15	H3-3A	cerebral cort	16	93.6466165	1.35494595
16	H3-3A	native cell	19	51.3421712	-0.6866966
17	H3-3A	native cell	14	64.010989	-0.1327101
18	H3-3A	native cell	17	65.7142857	-0.0119856
19	H3-3A	native cell	20	55.4888422	-0.2861422
20	H3-3A	native cell	25	51.2951106	-0.7485524
21	H3-3A	native cell	22	58.9813406	-0.1543225
22	H3-3A	native cell	18	55.6727246	-0.3688605
23	H3-3A	native cell	16	97.991857	2.3892699
24	H3-3A	oligodendroc	19	44.5747801	-0.0030504
25	H3-3A	oligodendroc	14	52.5423729	-0.3320625
26	H3-3A	oligodendroc	20	34.8471616	-1.0428462
27	H3-3A	oligodendroc	25	31.9155844	-1.1772372
28	H3-3A	oligodendroc	22	54.2930591	0.47522283
29	H3-3A	oligodendroc	18	50.6053269	0.32328306
30	H3-3A	oligodendroc	16	92.7272727	1.75669034
31	H3-3A	forebrain rad	19	47.9633401	-0.1182937
32	H3-3A	forebrain rad	14	48.5344828	-0.0597037

Scaling

We can also custom scale the plots by selecting "Yes" for "Custom scale plots." We see this when we select "Yes":

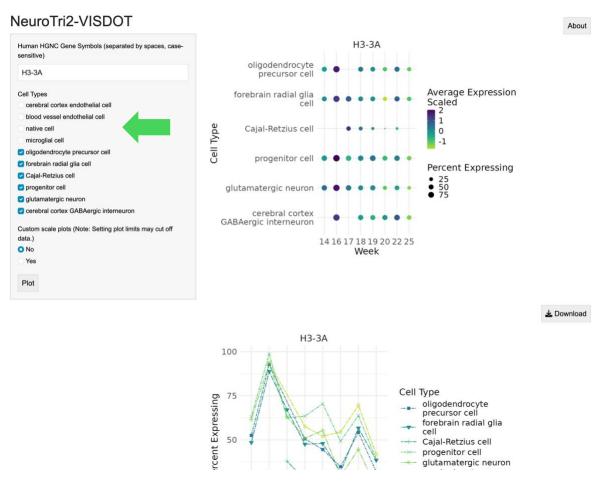


Let's say we want to center 0 on the average expression line plot. We can assign 100 to "Maximum percent expressing," -2.5 to "Minimum average scaled expression," and 2.5 to "Maximum average scaled expression," and click "Plot" again. Now we can see that the limits on the plots have changed:



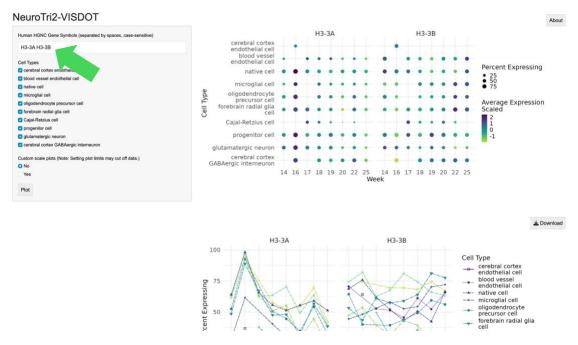
Cell Type Selection

We can also choose which cell types to include on the plots. Maybe we are not interested in looking at endothelial or immune cells. We can uncheck these in the "Cell Types" list, and plot again, this time without those cell types:

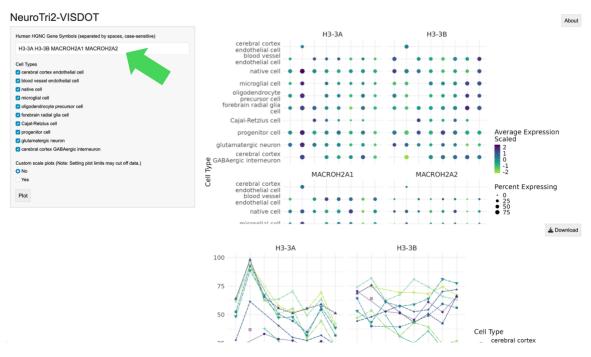


Plotting Multiple Genes

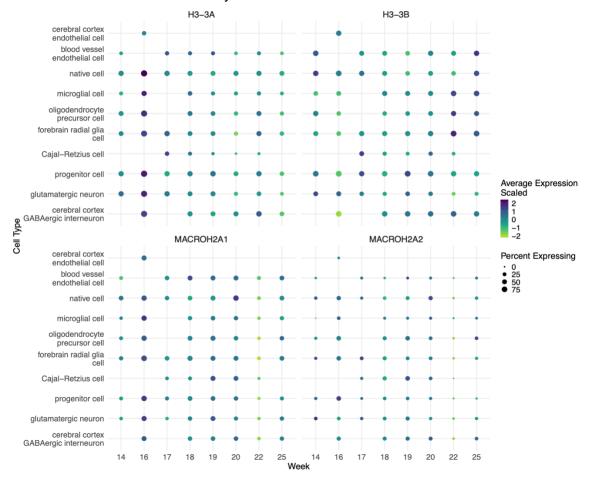
We can plot multiple genes on the same scale by typing in gene names separated by spaces. With two genes, you will get side-by-side plots:



With three or more, the plots will be automatically arranged into equal (or near-equal) numbers of rows and columns. For example, if we plot H3-3A, H3-3A, MACROH2A1, and MACROH2A2, we get two plots by two plots:



You may need to scroll within plotting spaces on the web page, but the plots can also be downloaded and will automatically resize:



The data can be downloaded the same as single-gene plot data. Multiple-gene plots can also be produced with cell type and scaling options in the same way as single-gene plots.

Troubleshooting

Gene X not found.

If you get an error "Gene [gene name] not found," this means that gene name is not present in the data. This may mean that it is not an HGNC gene symbol included in Ensembl version 110, or it may not be present in the dataset. Try checking Ensembl to make sure you have the correct version, and make sure the gene name is in the proper case (input is case-sensitive).

Please enter at least one gene.

If you get this error, there is nothing in the input text box.

Please select at least one cell type.

If you get this error, there are no cell types checked.

Missing data on plots

If you have "floating points" not connected to lines on the line plots and have custom scaled, or you are missing points on the dot plot compared to auto scaled plots, your plot limits are likely narrower than the data range. You can check this by selected "No" for custom scaling, or by downloading the data.