Using Venn diagrams

https://www.evinet.org

The idea of Venn diagram tool is to allow users working with a pre-computed file of differential expression (DE) values. Often, there are more than one contrast (comparison), and preparing DE lists off-line (e.g. in Excel) and then uploading them to EviNet is not very convenient. And again, there are no Venn diagrams in Excel…

First, you have to prepare a file for the upload: it has to have columns that quantify DE and be understood by the header parser.

At the moment, we support only one DE file format, the one currently used in the example in the project "stemcell" and also downloadable from the downloads directory. Note that the file should contain the keyword ".VENN" (or ".venn").

Given a simplest case of 3 different RNA-seq or microarray samples A, B, and C, your file might have contrasts "A vs. C" and "B vs. C". And you might have compared "A vs. B" as well, if it fits the idea of your experiment. If you caclulated fold change values and accompanied them with p-values and adjusted p-values (FDR), then the header should have at least the following column headers:

```
A_vs_C-FC
A_vs_C-p
A_vs_C-FDR
B_vs_C-FC
B_vs_C-p
B_vs_C-FDR
```

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The most important elements here are the delimiters:

1) condition names should be separated with "vs" and then
2) the value type (one of FC, P, FDR) is separated with "-".

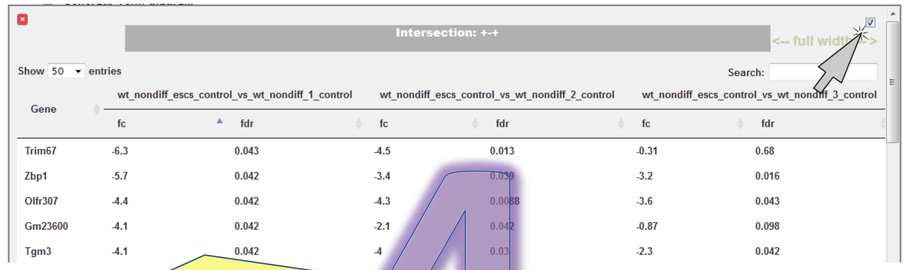
```
m/^(.+)vs(.+)-(FC|FDR|P)/i
```

My_promising_A_vs_tough_control_C-FDR words, e.g.

The parser is case-insensitive.

```
WT_Prog_5(sorted)_Control_vs_WT_Nondiff_ESCs_Control-FC
WT_Prog_5(sorted)_Control_vs_WT_Nondiff_ESCs_Control-p
WT_Prog_5(sorted)_Control_vs_WT_Nondiff_ESCs_Control-FDR
```

The file is submitted in "Altered gene sets" -> "File" -> "Upload local file" by pressing buttons "Display file content" and then "Use Venn diagram". Be patient, uploading can take time and even get crashed. But as soon as it is done, you can always find it in the same section and quickly read its content with "Use Venn diagram".



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You can pop up an arbitrary number of gene lists from one to as many as there are non-zero intersections in the diagram. Each of these lists can be automatically sent to NEA (as soon as you have chosen a network and FGS set).

Note: only visible gene lists with box in the upper right corner checked will be treated as AGS.

A tip: you can also filter/sort genes and copy some/all gene symbols with "Ctrl+mouse down" and then use just these by pasting this new list in "Plain list" section above. Do not forget then to uncheck the box near the button "Generate…". Good luck!

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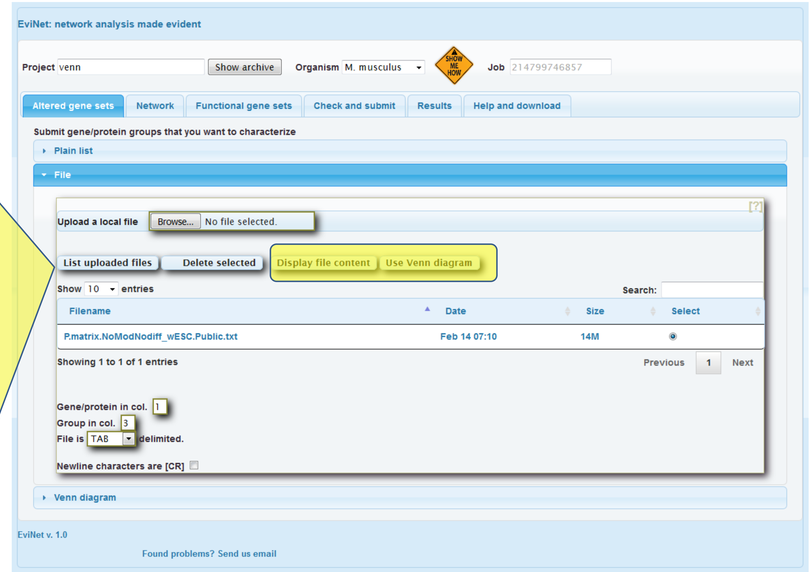
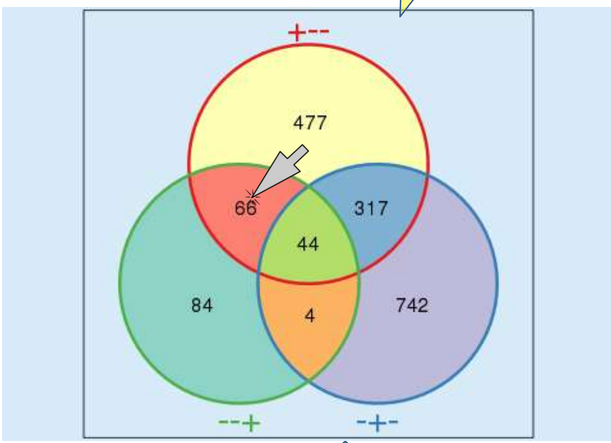
The labeling of diagram areas might look confusing. And it is indeed not trivial to label them without sacrificing readability. We thus propose this convention: labels consist of pluses and minuses which indicate that genes satisfy conditions of respective contrasts (lines of sliders). The order of +/- characters is the same as in the lines. For example, "+--" means that the genes fell within the criteria of contrast 1, "WT_Nondiff_ESCs_Control_vs_WT_Nondiff_1_Control", and did not within those of contrasts 2 and 3. By clicking at the intersection areas, you can get respective gene tables and clarify this notation even further (watch header labels).





After pressing "Altered gene sets", you are going to see a set of controls that reflect the content of your DE file.

The ranges of cut-off sliders are defined by min/max values of each column.

Note that the left and right fold change sliders correspond to down/up-regulation, respectively. Their ranges end/begin at 0. In other words, this would only work with log fold change values.

A tip: if you want to only look at down- or only up-regulated genes, then you should set the other (right or left, respectively) slider in its utmost position, as it is done in the 1st and 2nd lines at the screenshot. This trick should filter out any genes with expression change in that direction. The filtering criteria are applied line-wise, i.e. there will be one DE list from contrast "WT_Nondiff_ESCs_Control_vs_WT_Nondiff_1_Control", where expression pattern of each gene satisfies both the FC and FDR criteria, then another list for "WT_Nondiff_ESCs_Control_vs_WT_Nondiff_2_Control" and yet another for "WT_Nondiff_ESCs_Control_vs_WT_Nondiff_3_Control".

Overlaps of these three will be reflected at the Venn diagram when you press the button "Generate…"

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