# Using the TraVis pies interface

## Select input method and files

The user can choose to either create a new standardized TraVis pies input file from the 3 input files described in the previous section, or upload a previously created standardized TraVis pies input file to move on faster.

Creating a new file will allow the user to upload the three input files and specify which columns contain sample names, cohorts and normalization, and which compounds should be included in the analysis. The standardized TraVis pies input file created this way can be saved for uploading next time the app is used for this data, and will be used as input for this session.

Uploading a previously created file will perform some checks to make sure the format if the input is correct to avoid errors down the line, if this file was not manually modified no problems should be encountered. This input will then be used in the visualization module.

## Input cleaner module

Upload the input .csv files with the data of the experiment. See the example data provided with the article or with TraVis Pies.

* Metadata: Contains at least a column with sample names, and optionally a cohort column and a normalization column. **Note: do not include 12C samples, especially not in the same cohort as respective 13C samples, as their fractional contribution is by default different.**
* Abundance data: Contains the column with sample names that should have the same name as the copy in the metadata. All other columns are abundances of the compounds measured.
* Fractional contribution data: Contains the column with sample names that should have the same name as the copy in the metadata. All other columns are the fractional contributions of the compounds measured, although compounds that are 100% unlabeled can be left out here (automatically happens in some software) and will be processed correctly. The fractional contributions can be formatted as fractions or as percentages.

Depending on whether the input data is consistent between the files, the user will get appropriate errors or warnings, or will be able to further finetune the input by selecting the sample column, and a cohort and normalisation column if present.

The user will then be able to select the compounds that should be taken along, by default all compounds in the input.

If all settings are as desired, click the merge inputdata button to create a standardized TraVis pies input table, which can optionally be saved as a .csv file for quicker input next time it is used. The user can then select a folder and name for the .csv file using the Save to any directory button, or (quicker) copy-pasting a folder path to output the .csv to the textbox, then click the “Save to specified directory” button. Click the “Continue with this data” button to proceed.

## Visualisation

This module shows the pie chart visualization for a compound of choice from the last dataset uploaded or created in the input module, combined with the current visualization settings. It allows users to adapt these settings, see the resulting pie chart plot for a compound of choice before the more time-consuming step of generating and downloading similar pie chart plots for all desired compounds. Below is a rundown of the settings that can be changed.

Once the settings are as desired, click the “Save plots with these settings” button in order to proceed to the output module where images can be generated for all desired compounds using these settings.

### Note on using multiple selections for cohort and others

For compounds, cohorts … multiple selection boxes are used, and these can be confusing at the start. Whether an item in the box is selected (blue) or not (gray) does not affect the output, only whether it is present in the box or not. You can delete an item, or add an item that is available but not currently present in the box by clicking in the box and selecting it from the dropdown list showing all available options currently not present in the box.

### Compound, cohort variable, cohort order and normalization

* Compound: select compound from the dataset should be shown as an example in the visualization app
* Cohort variable: select variable that should be used to group the samples in different cohorts
* Cohorts: Select the cohorts to be shown and their order. The first cohort will be used as a reference for statistical tests
* Use normalized abundances: if available and checked, uses normalized abundances instead of raw abundances for the pie radius and statistical tests.

### Chart layout options

* Maximum charts per row: The number of columns in the facet plot, equaling the maximal amount of pie charts that will be shown on one horizontal line in each figure. If the amount of cohorts exceeds this value, the next line will be used until all cohorts are plotted.
* Position FC label: Position of the label displaying the fractional contribution. If “center”, the FC is shown in the center of the graph. If “slice”, both the contribution of the labelled and unlabeled fractions are shown in their specific slices.
* Slice label distance from center: this is the minimal distance from pie center at which a label can be plotted if Position FC label is “slice”. 1 is the maximal radius a pie can have. Normally a label appears at half the radius of the pie, but if this is smaller than minLabDist it will be plotted at minLabDist distance instead.
* FC label decimals: the amount of decimals shown in the fractional contribution labels
* Add ‘%’ to FC label: if checked adds % to the fractional contribution label (eg. 13% instead of just 13)
* show\_P: if checked hows text relating to P values of statistical significant tests on pie charts
* Add legend: if checked include the labelling color legend for slice colors on the detailed figure. This is never plotted on the concise figure for pathways.
* Add compound name as title: if checked include the compound name on the detailed figure. This is never plotted on the concise figure for pathways.
* Pick colour (un)labeled fraction: select a colour to be used for the (un)labeled fraction slice of the pie
* Pick circle line colour: select a colour to be used for the concentric circles
* Set opacity: change the opacity of the pie slices to see the concentric circle lines more or less distinctly
* Circle linetypes: change the line type (none, solid, dotted…) of the concentric circles

### Chart font options

* Select font: specify the font to be used on the result figures. To get the option to use any but the default font in a local version of this application, a separate script must be run to allow R to use the fonts stored on the machine. Ask Sam if desired.
* Fontsize: change fontsize of specified text on the figure

## Output module

Accessed once the user clicked the button “Save plots with these settings”, this module has a few settings regarding which figures will be generated and the plot type

* The user can select whether they want to output one or both of the following plot types
  + Stand-alone: A detailed figure, depending on chosen settings can contain legend and title .
  + Pathway-compatible: A figure in more concise format never with compound name and legend regardless of settings, more fit for overlaying on pathways.
* The user can select whether they want to output as .png or .tiff. They have similar quality but are suited for different purposes
  + .png is more suited for screen and web display as the files are smaller
  + .TIFF uses a colour scheme that is more compatible with conventional printing, however the files are very big
* The user can make any selection from the compounds in the input to output figures for, by default all

The user can then obtain the images for the desired compounds by selecting or copy-pasting a folder path to output the figures to, then clicking generate figures. A progress bar will pop up to show how many compounds still need to be processed, as this can take some time when 10’s of compounds are supplied