

# Normalize to baseline

If your experimental design includes a sample or a group of samples serving as a baseline control, you can normalize the experimental samples by subtracting or dividing by the baseline sample(s) using the Normalize to baseline task in Partek Flow. For example, in PCR experiments, the delta Ct values of control samples are subtracted from the delta Ct values of experimental samples to obtain delta-delta Ct values for the experimental samples.

The Normalize to baseline option is available in the *Normalization and Scaling* section of the context-sensitive menu (Figure 1) upon selection of any count matrix data node.

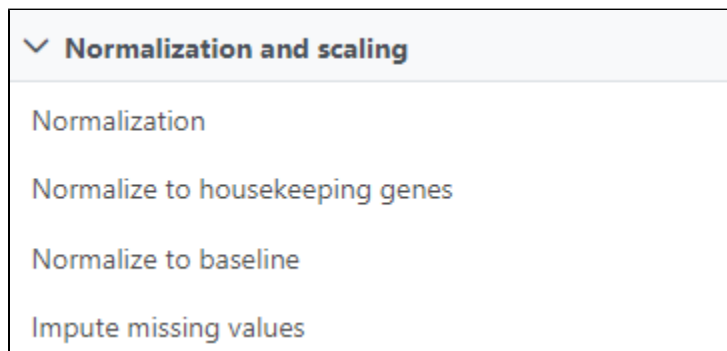


Figure 1. When a data node containing a count matrix is selected, *Normalize to baseline* is available in the toolbox

There are three options to choose the baseline samples:

- use all samples
- use a group
- use matched pairs

## Use all samples to create baseline

To normalize data to all the samples, choose to calculate the baseline using the **mean** or **median** of all samples for each feature, and choose to **subtract baseline** or **ratio to baseline** for the normalization method (Figure 2), and click **Finish**.

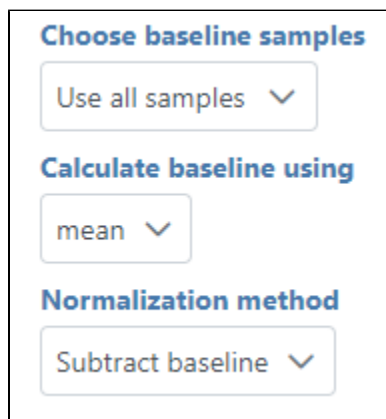


Figure 2. Use the mean or median of all samples as the baseline to normalize the data

## Use a group to create baseline

When there is a subset of samples that serve as the baseline in the experiment, select **use group** for Choose baseline samples. The specific group should be specified using sample attributes (Figure 3).

Choose baseline samples Use group ▼

Attribute with baseline group Treatment ▼

Baseline group Control ▼

Calculate baseline using mean ▼

Normalization method Subtract baseline ▼

Remove baseline samples after normalization ☒

Back Finish

Figure 3. Use a subgroup of samples to create baseline to normalize the data

Choose **use group**, select the attribute containing the baseline group information, e.g. **Treatment** in this example, with the samples with the group **Control** for the **Treatment** attribute used as the baseline. The control samples can be filtered out after normalization by selecting the **Remove baseline samples after normalization** check box.

## Use matched pairs

When using matched pairs, one sample from each pair serves as the control. An attribute specifying the pairs must be selected in addition to an attribute designating which sample in each pair is the baseline sample (Figure 4).

Choose baseline samples Use matched pairs ▼

Pair samples by Pair ▼

Attribute with baseline group Treatment ▼

Baseline group Control ▼

Normalization method Subtract baseline ▼

Remove baseline samples after normalization ☒

Back Finish

Figure 4. Designated pairs and the baseline sample in each pair to normalize by matched pairs

After normalization, all values for the control sample will be either 0 or 1 depending on the normalization method chosen, so we recommend removing baseline samples when using matched pairs.

The output of Normalize to baseline is a Normalized counts data node.

## Additional Assistance

If you need additional assistance, please visit [our support page](#) to submit a help ticket or find phone numbers for regional support.



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