Tutorial 2: Comparing multiple groups
Comparing Multiple Groups

• User can choose ANOVA tool to compare multiple groups of data.
• There are three ways to activate ANOVA
  1. from TOOL panel.
  2. from pull-down menu.
  3. from selected dataset (database panel).
     We recommend the third way.
Comparing Multiple Groups – continued.

User can use ANOVA to compare multiple groups. Running ANOVA is similar to running T-test (see tutorial 1 for detail about T-test).

Right-click the selected datasets, choose “Analysis” ⇒ T-Test/ANOVA.

Activate ANOVA from database panel
Comparing multiple groups - continued

Assign data into 3 groups based on compound (B, C, D)

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T-Test fold changes are computed as grp1/grp2, so "up" regulation will mean grp1 > grp2 in any further analysis.
Options for ANOVA

<table>
<thead>
<tr>
<th>Gene identifiers to include</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Genbank Acc</td>
<td>✔</td>
</tr>
<tr>
<td>Gene Mfr ID</td>
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<td>GEN_DESCR_MFR</td>
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<tr>
<td>REFSEQ</td>
<td>✔</td>
</tr>
<tr>
<td>SPOTID</td>
<td>✔</td>
</tr>
</tbody>
</table>

Dataset Naming

Hybridization names are always included.

- [ ] add sample name(s) to hybridization names
- [ ] add dye name(s) to hybridization names

Data options

- [ ] Subtract backgrounds when present (raw datasets only)
- [ ] Apply log (base 2) to expression values
- [ ] Exclude spots flagged as bad
Comparing Multiple Groups - continued

User can further filter the results by setting criteria like P value, fold change, etc.

ANOVA result of 3 groups
Comparing Multiple Groups - continued

Options for Pairwise T-test
Comparing Multiple Groups - continued

Pairwise T-test result

Group 1 vs group 2

Group 1 vs group 3

Group 2 vs group 3

Clicking one of the three group button will bring the detail table of the comparing result. See Next slide.
Comparing Multiple Groups - continued
Two-way ANOVA

- Two-way ANOVA is available in ArrayTrack v3.4 and up.
- The Two-way ANOVA analysis will involve more than one independent variable (factor). The example in slide #4 is about ANOVA using only one factor (compound). The following example will use two factors (dose, compound).
- Two-way ANOVA would not only be able to assess both dose and compound in the same test, but also find out if there is an interaction between the parameters.
Two-way ANOVA

Right-click the selected dataset, choose “Analysis” ⇒ “Two-way ANOVA”.

Before assign the groups, please select the factors and levels. Here we select factor A (2 doses: 0 and 2) and factor B (3 compounds: B, C, D) While radio button A is selected, highlight dose 0 group and assign to group 1. In the same way assign dose 2 group to group 2.
Two-way ANOVA

After finishing factor A assignment, select radio button B. Then highlight hybs with compound B, assign them to Level 1 for factor B.
Two-way ANOVA

Similiarly assign compound C to level 2, compound D to level 3.
Two-way ANOVA

Here is the finished assignment for factors and levels. Click “Next” button.
Two-way ANOVA

Here is the default option for two-way ANOVA. Click “Do Tests” button.
Two-way ANOVA

This is the result of two-way ANOVA test. The P value for factor A and B is listed: P(A), P(B), B(A*B).