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Short manual for the Excel workbook *Response_Surface_1.0.xlsm*

You must have a basic understanding of Design of Experiment, DoE, to use this workbook.

The workbook can be used to make and analyse the following designs:

Sheet: *2 factors*	Central composite design for 2 continuous factors, 8 runs + 2 to 6 centre points = 10 to 14 runs
Sheet: *3 factors*	Central composite design for 3 continuous factors, 14 runs + 2 to 6 centre points = 16 to 20 runs
Sheet: *Box-Behnken*	Box-Behnken designs for 3 continuous factors, 12 runs + 2 to 6 centre points = 14 to 18 runs.

Central composite designs use 3 or 5 levels for each factor.
Box-Behnken designs avoid extreme values (all factors high or low) and use 3 levels for each factor.

Workflow

Detailed instructions for data and parameters are given in all sheets in the workbook (to the right of tables).

There are 5 steps:

1) Set up the design
    a. Decide what design to use (depends on the number of factors, levels and possible runs)
    b. Enter the names and levels of the factors in the sheet *2 factors, 3 factors* or *Box-Behnken*
        i. For the laboratory course: Fill in all part of Table 3 (Headings, assignment of experiments to groups, comments, and comments to each run, especially levels for factors´, that are not a design factor, i.e. constant factors).
        ii. For blocking of Central Composite Designs, assign groups to the runs according to the instructions in the sheet.
        iii. Remember to sort the table according to *Groups.*
        iv. Press the button *Copy Table 3 to Runs* to create a sheet with data for the experimental runs
2) Run the experiments
    a. Experiments should be run in random order if it is possible
3) Enter the results in sheet *Input* (or in the appropriate sheet if you only have one or a few dataset)
4) Analyse the results:
    a. Go to the appropriate sheet and press the *Start* button – data for the first analyte is loaded
    b. Examine the results
    c. Save the results to sheet *Output*:
        i. Click the button *Save results* – results are saved without further actions

ii. Click the button *Save results and load next analyte* results are saved and the next data for the next analyte is loaded

5) Save the workbook under a new name, *Save As….*

Do not use the workbook for more than one DoE design at a time (*2 factors, 3 factors* or *Box-Behnken)*

Examine the results

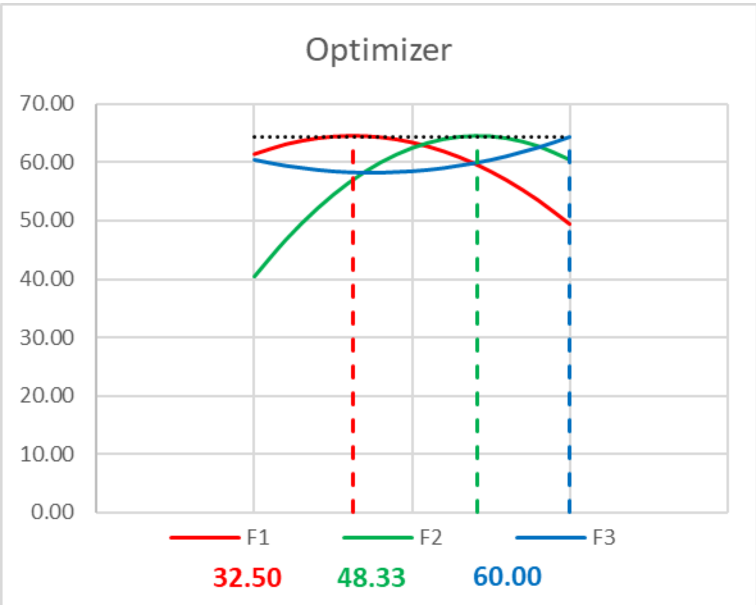
When data are loaded to the analyse sheet, the workbook will automatically find the settings for the maximum response (example from sheet *Box-Behnken* used I the following), see Table 6:

Table 6: Optimization and estimation			
Factor	**Name**	**Value**	**Units**
1	Temp	32.50	Degree
2	Vol	48.33	ml
3	Time	60.00	min
Response	Max	108.79	

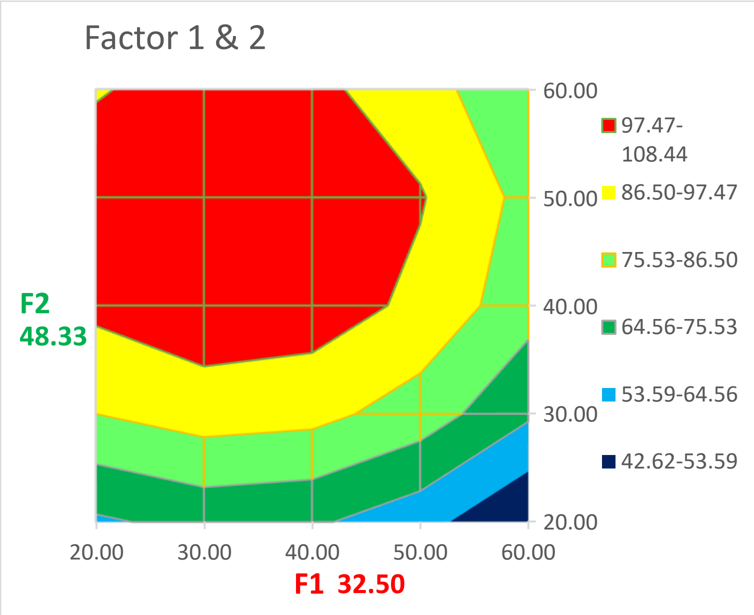
The levels for the factors are given in this table. The text "Max" before the response indicates that is the levels for maximum response. If you press to the button *Minimum* results for the minimum response will be given in the table.

The values in the white cells may be changed, if you want to estimate the response for another level of the factors (but the text "Max" or "Min" for the response will remain unchanged. You may change this text).

There are 4 graphs (2 in sheet *2 factors*) showing the response as function of the factor levels:



This graph shows the response as function of each factor. The optimum level (max or min) of the factors is shown by numbers and by the vertical lines. The line for factor 1, ***F1***, is calculated with the two other factors at the optimum value (48.33 for F2 and 60.00 for F3), and *vice versa* for ***F2*** and *F3.*



This graph shows the response as function of factor1&2 when the 3rd factor is at the value shown in Table 6 (here F3 = 60.0). There are similar plots for factor 1&3 and 2&3.

Two measures of the uncertainty in the results are given in the sheets, example here from sheet *2 factors:*

Table 4: Responses		**Mean**	**Residuals**
72.80553		72.80553	2.74
82.03804		82.03804	2.16
71.10177		71.10177	1.25
92.08177		92.08177	0.66
81.56802		81.56802	-3.99
98.43412		98.43412	-2.82
79.68978		79.68978	-4.90
88.33373		88.33373	-1.92
108.659		108.659	5.63
102.7528		102.7528	-0.27
103.0488		103.0488	0.02
104.453		104.453	1.43
0		0	
0		0	

Table 5: Coefficients		**SE**	**P**
Const	103.02	1.86E+00	0.000
1	0.39	8.32E-02	0.002
2	0.14	8.32E-02	0.127
1^2	-0.02	6.24E-03	0.005
2^2	-0.04	6.24E-03	0.000
12	0.01	5.09E-03	0.187

SE is the standard deviation of the coefficient and P is the probability that the coefficient is different from zero, i.e. if $P < 0.05$ it can be concluded that the coefficient is statistically different from zero.

Residuals is simply the difference between the observed value and the value predicted from the model. A large deviation can indicate that a run is an outlier. The best fix for a missing value or a real outlier is to replace it with the average of the remaining runs.

For blocked designs

1) In Table 1 set the number of blocks = 2

Table 1: Design parameters	
	1.000
No of centre points	6
Blocks:	2

2) Look at P for Block in Table 5:

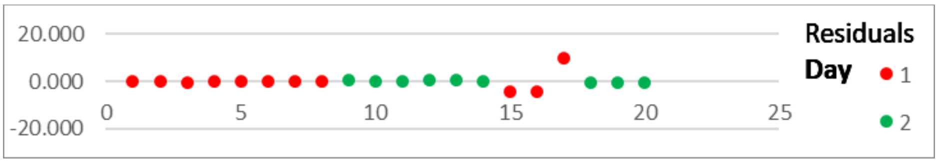
- If P < 0.05 use Block = 2
- If P > 0.05 change to Block = 1

Table 5: Coefficients		SE	P
Const	103.69	1.068329	0.000
1	7.85	1.100226	0.000
2	2.83	1.100226	0.037
1^2	-10.41	1.631899	0.000
2^2	-16.40	1.631899	0.000
12	2.94	1.347496	0.066
Block	-1.99	0.766783	0.036

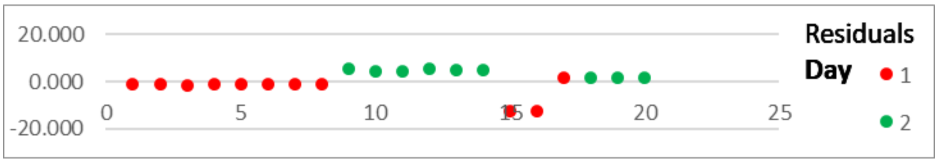
3) The residuals can also be seen as function of the Day in the graph

- (Block = Day)

Example with Block = 2, (block effects removed – random distribution of residuals):



Same data with Block = 1 (results for Day 2 > Day 1):



In this case the data should be analysed with Block = 2 (P = 0.001 i.e. << 0.05 for Block)