

GRaPe 2.0 User Interface Crash Course

Insert Model Name here, special character (e.g. \$%^&) cannot be used.

The screenshot shows the GRaPe 2.0 User Interface. At the top, there is a 'Model Name' input field. Below it are three main sections: 'Reacting Species', 'Enzymes', and 'Reactions'. Each section has a table with columns for No., Name, Concentration, and other relevant fields. Below each table are 'Add' and 'Delete' buttons. At the bottom, there are buttons for 'Generate Header File', 'Insert Fitting Data', 'Run Parameter Estimation', and 'Save'. Annotations with red boxes and numbers 1, 2, and 3 point to the 'Reacting Species', 'Enzymes', and 'Reactions' tables respectively.

Model Name:

Reacting Species

No.	Species	Concentration	Boundary
1			<input type="checkbox"/>

Enzymes

No.	Enzyme	Concentration
1		

Reactions

No.	Enzyme	Activator	Inhibitor	Reaction	PK
1	N/A	N/A	N/A		<input type="checkbox"/>

Number of Species: 0 Number of Enzymes: 0

Number of Reactions: 0

Model Fitting

Box for inserting enzyme information (name & concentration).

Box for inserting reaction information (enzyme that catalyses it*, activator/inhibitor, product and substrates**)

*if enzyme involved are unknown, pseudo name and concentration of 1 can be used.
**to add product and substrates users need to double click on the reaction cell for a different interface to appear.

Box for inserting metabolites' information (name, initial concentration and boundary condition*) within the model

*boundary condition TRUE would make the concentration fixed and vice versa

***NUMBERS indicate ideal information entering order.
****ADD and DELETE button lets users add/delete information count for each boxes.

Example use of GRaPe 2.0

Step 1: Add all known metabolites and its initial concentration (if they are unknown, concentration of 0.1 is suggested)

*concentration unit used in this software is up to the user's choosing (not explicitly stated in software), they should however be standardised across metabolite, proteins and flux.

The screenshot displays the GRaPe 2.0 software interface. At the top, the title bar reads "GRaPe 2.0". Below it is a "File" menu bar. The main workspace is divided into several sections:

- Model Name:** A text input field with the placeholder "Insert Model Name".
- Reacting Species:** A table with columns: No., Species, Concentration, and Boundary. It contains 6 rows of data.
- Enzymes:** A table with columns: No., Enzyme, and Concentration. It contains 1 row of data.
- Reactions:** A table with columns: No., Enzyme, Activator, Inhibitor, Reaction, and PK. It contains 1 row of data.

Below the tables, there are counters and buttons for each section:

- Number of Species:** 6. Buttons: Add, Delete.
- Number of Enzymes:** 0. Buttons: Add, Delete.
- Number of Reactions:** 0. Buttons: Add, Delete.

At the bottom, there is a "Model Fitting" section with a text input field and four buttons: "Generate Header File", "Insert Fitting Data", "Run Parameter Estimation", and "Save".

Logos for SBML and a tree icon are visible in the top right corner.

No.	Species	Concentration	Boundary
1	IN	10	<input checked="" type="checkbox"/>
2	OUT	10	<input checked="" type="checkbox"/>
3	A	18	<input type="checkbox"/>
4	B1	41	<input type="checkbox"/>
5	B2	9	<input type="checkbox"/>
6	C	52	<input type="checkbox"/>

No.	Enzyme	Concentration
1		

No.	Enzyme	Activator	Inhibitor	Reaction	PK
1	N/A	N/A	N/A		<input type="checkbox"/>



Step 2: Add all known enzymes and its concentration (if they are unknown, concentration of 1 is suggested)

*if enzyme for an reaction is unknown OR if it is a pseudo reaction, a pseudo name can be used.

GRaPe 2.0

File

Model Name:



Reacting Species

No.	Species	Concentration	Boundary
1	IN	10	<input checked="" type="checkbox"/>
2	OUT	10	<input checked="" type="checkbox"/>
3	A	18	<input type="checkbox"/>
4	B1	41	<input type="checkbox"/>
5	B2	9	<input type="checkbox"/>
6	C	52	<input type="checkbox"/>

Enzymes

No.	Enzyme	Concentration
1	IMPORT	50
2	Diverge	11
3	Converge	22
4	EXPORT	35

Number of Species: 6

AddDelete

Number of Enzymes: 4

AddDelete

Reactions

No.	Enzyme	Activator	Inhibitor	Reaction	PK
1	N/A	N/A	N/A		<input type="checkbox"/>

Number of Reactions: 0

AddDelete

Model Fitting

Generate Header FileInsert Fitting DataRun Parameter EstimationSave

Step 3: Add reaction information. Drop down menu for enzyme/activator/inhibitor*.

*Enzyme information is compulsory, while activator and inhibitor are optional

**Double click on the reaction cell to access interface to select substrate and production, as well as their stoichiometry

GRaPe 2.0

Model Name:

Reacting Species

No.	Species	Concentration	Boundary
1	IN	10	<input checked="" type="checkbox"/>
2	OUT	10	<input checked="" type="checkbox"/>
3	A	18	<input checked="" type="checkbox"/>
4	B1	41	<input checked="" type="checkbox"/>
5	B2	9	<input checked="" type="checkbox"/>
6	C	52	<input checked="" type="checkbox"/>

Number of Species: 6

Enzymes

No.	Enzyme	Concentration
1	IMPORT	50
2	Diverge	11
3	Converge	22
4	EXPORT	35

Number of Enzymes: 4

Reactions

No.	Enzyme	Activator	Inhibitor	Reaction	PK
1	IMPORT	N/A	N/A		<input type="checkbox"/>
2	IMPORT	N/A	N/A		<input type="checkbox"/>
3	Diverge	IN	IN		<input type="checkbox"/>
4	Converge	OUT	OUT		<input type="checkbox"/>
5	EXPORT	A	A		<input type="checkbox"/>
6	EXPORT	B1	B1		<input type="checkbox"/>
7	EXPORT	B2	B2		<input type="checkbox"/>
8	EXPORT	C	C		<input type="checkbox"/>

Number of Reactions: 0

Model Fitting

Please build your reaction

Substrate(s): N/A

Stoichiometry: 1

Product(s): N/A

Stoichiometry: 1

OK Cancel

Number of Species: 6

Number of Enzymes: 4

Reactions

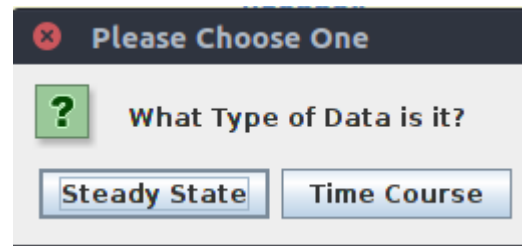
No.	Enzyme	Activator	Inhibitor	Reaction	PK
1	IMPORT	N/A	N/A	R1: (1) IN <-> (1) A	<input type="checkbox"/>
2	Diverge	N/A	N/A	R2: (1) A <-> (1) B1 + (1) B2	<input type="checkbox"/>
3	Converge	N/A	N/A	R3: (1) B1 + (1) B2 <-> (1) C	<input type="checkbox"/>
4	EXPORT	N/A	N/A	R4: (1) C <-> (1) OUT	<input type="checkbox"/>

Number of Reactions: 4

Model Fitting

Generate Header File Insert Fitting Data Run Parameter Estimation Save

4a:



Step 4: Make the fitting data file. (example used here are for steady state data, for time course data, a column would be added to indicate time point)

4a: click on generate header file for fitting data.

4b: move the names and IDs to a spreadsheet to edit the information much more easily.

4c: keep only IDs and expected values for metabolites/flux.

NOTE: move the data back into a text file and make sure there isn't any trailing new lines

What header file looks like in a text file.

```
Copy to Excel.
Remove the Names.
Keep only the IDs.
Paste data back into txt file to be inserted into GRaPe, make sure to avoid empty lines.

IMPORT E1
Diverge E2
Converge E3
EXPORT E4
IN S1
OUT S2
A S3
B1 S4
B2 S5
C S6
IMPORT R1
Diverge R2
Converge R3
EXPORT R4
```

4b:

	A	B	C
1	IMPORT	E1	
2	Diverge	E2	
3	Converge	E3	
4	EXPORT	E4	
5	IMPORT	R1	0.544777
6	Diverge	R2	0.544777
7	Converge	R3	0.544777
8	EXPORT	R4	0.544777
9	IN	S1	10
10	OUT	S2	10
11	A	S3	6.40291
12	B1	S4	32.2015
13	B2	S5	0.201489
14	C	S6	0.00152386

4c:

	A	B
1	R1	0.544777
2	R2	0.544777
3	R3	0.544777
4	R4	0.544777
5	S1	10
6	S2	10
7	S3	6.40291
8	S4	32.2015
9	S5	0.201489
10	S6	0.00152386

Step 4d: OPTIONAL

If a value is given to the steady state fitting data, GRaPe 2.0 would use the value to change the model's selected enzyme values during parameter estimation process.

This is useful for providing fitting data that has multiple conditions, which normally affect protein concentration within the system to an extent, and result in different metabolite and flux output.

As seen in the example, some information can be left out for the fitting process.

	A	B	C	D	E	F
1	E1	50	60	30	58	58
2	E2	11	15	22	15	15
3	E3	22	15	11	16	16
4	E4	35	42	29	32	32
5	R1	0.544777	0.676	0.569	N/A	0.79
6	R2	0.544777	0.676	0.569	N/A	0.79
7	R3	0.544777	0.676	0.569	N/A	0.79
8	R4	0.544777	0.676	0.569	N/A	0.79
9	S1	10	10	10	10	15
10	S2	10	10	10	10	8
11	S3	6.40291	6.27	N/A	6.27	8
12	S4	32.2015	0.2734	N/A	0.31	0.39
13	S5	0.201489	32.27	N/A	32.3	32.4
14	S6	0.00152386	0.0016	N/A	0.002	0.0023

Step 5: After inserting fitting data, user can run the parameter estimation process.

5a: Select the hyperparameters for the genetic algorithm.

Population: Number of models to be solved (high values would lead to longer parameter estimation time)

Max Generations: Maximum number of generations to go for before stopping even if optimal parameters aren't found. (high values would lead to longer parameter estimation time)

Plateau limit: Number of generations where there is no further increase in fitness score. After reaching the given number, the process would stop.

Plague: Process of removing unfit individuals and maintaining only the initial population size at given number of generation.

*After the process is done as seen in the example where it says 'Estimation Complete!', user can click the 'Save' button to save the model with estimated parameters.

GRAPe 2.0

File

Model Name:

Reacting Species

No.	Species	Concentration	Boundary
1	IN	10	<input checked="" type="checkbox"/>
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3	A	18	<input type="checkbox"/>
4	B1	41	<input type="checkbox"/>
5	B2	9	<input type="checkbox"/>
6	C	52	<input type="checkbox"/>

Enzymes

No.	Enzyme	Concentration
1	IMPORT	50
2	Diverge	11
3	Converge	22
4	EXPORT	35

Number of Species: 6

Number of Enzymes: 4

Reactions

No.	Enzyme	Activator	Inhibitor	Reaction	PK
1	IMPORT	N/A	N/A	R1: (1) IN <=> (1) A	<input type="checkbox"/>
2	Diverge	N/A	N/A	R2: (1) A <=> (1) B1 + (1) B2	<input type="checkbox"/>
3	Converge	N/A	N/A	R3: (1) B1 + (1) B2 <=> (1) C	<input type="checkbox"/>
4	EXPORT	N/A	N/A	R4: (1) C <=> (1) OUT	<input type="checkbox"/>

Number of Reactions: 4

Model Fitting

5a:

Parameters for Genetic Algorithm

Starting Population:

Max Generations:

Plateau Limit:

Plague at every N Generation:

Parameter Estimation Progress

starting size: 55

INITIAL EVALUATION DONE

BEST IS: 1.3415371701633495E-4

Generation: 1	Best:	1.3415371701633495E-4
Generation: 2	Best:	2.283770521055304E-4
Generation: 3	Best:	3.322727960773794E-4
Generation: 4	Best:	3.322727960773794E-4
Generation: 5	Best:	3.624346973882424E-4
Generation: 6	Best:	3.624346973882424E-4
Generation: 7	Best:	4.753958946637581E-4
Generation: 8	Best:	5.767447824938422E-4
Generation: 9	Best:	5.970684255129281E-4
Generation: 10	Best:	6.597651891794998E-4
Generation: 11	Best:	6.727344805993176E-4

Estimation Complete!