

BIONUMERICS Tutorial:

Importing sugar metabolization data from text files

1 Aim

Sequential measurements that express an evolution of one parameter in function of another (e.g. enzymatic activity, growth curves, etc.) are called *trend type data* and can be imported and analyzed in our BIONUMERICS software package.

In this tutorial you will learn how to import trend data in your BIONUMERICS database, how to fit a curve through the measurement points, and how to deduce parameters from the curve function to compare the samples.

2 Example data

The example text files from which we will import data in this tutorial contain trend data for 6 strains and can be downloaded from the Applied Maths website: go to https://www.applied-maths.com/download/sample-data and click on 'Trend data sample files'.

1. Open one of the files (e.g. BSU1072.txt) to examine the data that will be imported.

<u> </u>	BSU10	72.txt - No	otepad						_		×	
<u>F</u> ile	<u>E</u> dit	F <u>o</u> rmat	<u>V</u> iew	<u>H</u> elp								
Time	(h)		Mannito	1	Inositol	S	orbitol		Rhamnose		Su	~
1		0.20541	83	0.12660	39	0.248595	9	0.77913	91	0.6484	611	
2		0.45484	33	0.16537	58	0.536914	9	0.91040	64	0.8849	223	
3		1.13096	80	0.19482	97	0.584119	7	1.22470	57	1.3633	131	
4		2.09906	84	0.32436	77	0.633242	0	1.20529	99	2.2078	335	
5		3.06171	58	0.53172	84	1.185183	0	1.68168	82	2.7616	652	
6		3.78965	05	1.05980	55	2.346847	3	3.07808	62	3.5830	966	
7		3.98135	29	1.30116	42	3.724370	8	3.91287	13	4.4838	851	
8		4.20038	55	1.83576	35	5.025069	7	5.88500	25	5.5505	135	
9		4.18635	65	1.97819	81	5.636780	2	6.99057	79	5.9768	733	
10		4.31195	95	2.27270	18	6.659298	3	8.63561	79	6.4355	347	
11		4.21921	71	2.31969	73	6.945574	3	9.40257	26	6.7371	534	
12		4.20097	52	2.44953	29	7.196052	8	9.69480	90	6.8251	140	
13		4.28165	67	2.40742	38	7.141610	3	9.81288	60	6.8499	686	
14		4.22048	58	2.47114	.99	7.430006	1	9.76134	89	6,9615	376	¥
/												Ξ.

Figure 1: Text file: an example.

The text file contains a header, followed by the actual data in tabular format. In this table, the first column contains the X-values (= time points, expressed in hours) and the other columns contain the Y-values (= sugar concentration, expressed in mM). The first row describes the curve names (= sugar names).

3 Creating a trend data type experiment

- 1. Create a new database (see tutorial "Creating a new database") or open an existing database.
- 2. In the *Main* window, click on + in the toolbar of the *Experiment types* panel and select **Trend** *data type* from the list (see Figure 2).

Create a new experiment type	?	×
Select what kind of experiment type you	want to cr	eate:
Fingerprint type		^
Character type		
Sequence type		
Matrix type		
🛃 Trend data type		
📥 Spectrum type		
≵ Sequence read set type		
🗠 Composito data pot	_	*
ОК	Car	ncel

Figure 2: New Trend data type.

3. Press <*OK*>, enter a name, for example **Carbon sources** and press <*Finish*> to complete the creation of the new trend data type.

The *Experiment types* panel now lists the trend data type **Carbon sources**.

4. Open the *Trend type* window by double-clicking on **Carbon sources** in the *Experiment types* panel.

The *Trend type* window is initially empty. Trend curves can be added either manually (*Trend-Curves* > *Add new trend curve...*) or during import (see 4).

4 Importing trend data

- 1. Select *File* > *Import...* (, Ctrl+I) to open the *Import* dialog box.
- Choose the option *Import trend data* under the *Trend data type data* item in the tree and press <*Import*> (see Figure 3).
- 3. Press < *Browse* > and browse for the downloaded text files in the Carbon sources folder.

The *Input* wizard page is updated (see Figure 4).

4. Press <*Next*>.

As this is the first time we import trend data into the database, we need to create a new import template by specifying *Import rules*.

All columns detected in the selected text files are listed in the grid.

- Select first row in the list and click < *Edit destination*> or simply double-click on the row. Select "X-axis" as the BIONUMERICS destination field under *Trend curve value* in the *Edit data destination* dialog box and press < *OK*> (see Figure 5).
- 6. Click on "Mannitol" and whilst holding the **Shift**-key, click on "Lactose" to make a multiple selection of the 6 trend data curves.

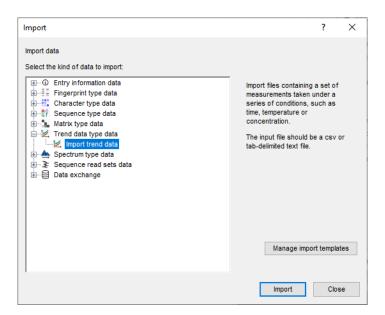


Figure 3: Import tree.

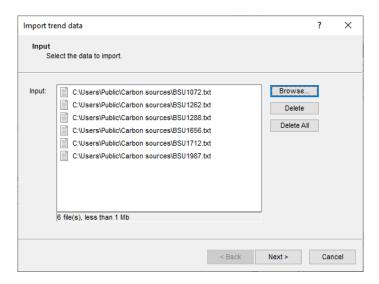


Figure 4: The Input wizard page.

Edit data destination	?	×
 <none></none> Key Entry info field Trend curve value Carbon sources <-> Create new> <-> Xexis Trend data info field 		
ОК	Car	ncel

Figure 5: Edit data destination.

 Press < *Edit destination*>, select "Carbon Sources" under *Trend curve value* as destination and click < *OK*> (see Figure 6).

Edit data destination	?	×
 <none></none> Entry info field Trend curve value Carbon sources Trend data info field 		
ок	Car	icel

Figure 6: Edit data destination.

BIONUMERICS tries to map the column names to existing trend data curves. When there is no trend data curve present with the same name, you will be prompted to create the new trend data curves.

- 8. Press < OK > and then < Yes > to confirm the creation of the curves.
- Select the last row in the grid, i.e. the row containing the *File name*, click <*Edit destination*> and link the field to the "Key" field. Press <*OK*>.

The grid is updated and should now look like in Figure 7.

Source type	Source	Destination type	Destination	
File field	Time (h)	Trend curve value : Carbon sources	X-axis	
File field	Mannitol	Trend curve value : Carbon sources	Mannitol	
File field	Inositol	Trend curve value : Carbon sources	Inositol	
File field	Sorbitol	Trend curve value : Carbon sources	Sorbitol	
File field	Rhamnose	Trend curve value : Carbon sources	Rhamnose	
File field	Sucrose	Trend curve value : Carbon sources	Sucrose	
File field	Lactose	Trend curve value : Carbon sources	Lactose	
File	Name	Entry information	Key	
Edit destination				
Edit destination				

Figure 7: Import rules.

10. Press the *<Preview*> button to verify that all information will correctly be imported in the database (see Figure 8).

Nr.	Key	Х	Mannitol	Inositol	Sorbitol	Rhamnose	Sucrose	Lactose	^
1	BSU1072	1	0.2054183	0.1266039	0.2485959	0.7791391	0.6484611	0.3793394	
2	BSU1072	2	0.4548433	0.1653758	0.5369149	0.9104064	0.8849223	0.3866269	
3	BSU1072	3	1.1309680	0.1948297	0.5841197	1.2247057	1.3633131	0.4657357	
4	BSU1072	4	2.0990684	0.3243677	0.6332420	1.2052999	2.2078335	0.8166276	
5	BSU1072	5	3.0617158	0.5317284	1.1851830	1.6816882	2.7616652	0.8558457	
6	BSU1072	6	3.7896505	1.0598055	2.3468473	3.0780862	3.5830066	1.3821836	
7	BSU1072	7	3.9813529	1.3011642	3.7243708	3.9128713	4.4838851	1.7858596	
8	BSU1072	8	4.2003855	1.8357635	5.0250697	5.8850025	5.5505135	3.6667633	
9	BSU1072	9	4.1863565	1.9781981	5.6367802	6.9905779	5.9768733	5.5060795	
10	BSU1072	10	4.3119595	2.2727018	6.6592983	8.6356179	6.4355347	6.5774936	
11	BSU1072	11	4.2192171	2.3196973	6.9455743	9.4025726	6.7371534	7.1940099	
12	BSU1072	12	4.2009752	2.4495329	7.1960528	9.6948090	6.8251140	7.4915348	
13	BSU1072	13	4.2816567	2.4074238	7.1416103	9.8128860	6.8499686	7.7249221	
14	BSU1072	14	4.2204858	2.4711499	7.4300061	9.7613489	6.9615370	7.6915789	
15	BSU1072	15	4.2253411	2.4187549	7.3197057	10.0599528	6.7954706	7.8750075	
16	BSU1072	16	4.3495493	2.4988415	7.3527352	9.9301089	7.0163252	7.9815472	
17	BSU1072	17	4.3370329	2.4902711	7.3034246	9.8500371	6.9370452	7.7350499	
18	BSU1072	18	4.2500607	2.4750449	7.3284913	10.1294913	6.9384898	7.8073869	~
<								>	

Figure 8: Preview of the parsed data.

- 11. Press < *Next* > to proceed to the *Import links* dialog box.
- 12. Check *Key* under *Import links* and press <*Finish*>.

The import template needs to be saved to be able to use it again later on.

- Enter a Name for the import template (e.g. "Carbon sources template") and optionally a Description. Press < OK >.
- 14. Highlight the newly created template and click < Next>.

This next dialog will indicate that 6 new entries will be created during import (see Figure 9).

Import trend data	?	×
Database links Link the imported records to database entries. Double click on a cell to get an overview.		
Overview In 'All levels' Image: create 6 entries and image: update 0 entries Image: select modified entries Image: select modified entries Image: select modified entries		
< Back Finish	Car	icel

Figure 9: Create 6 new entries.

15. Press < *Finish*> to start the actual import. The progress of the import is shown while database information is added to the BIONUMERICS database.

The entries are displayed in the *Database entries* panel and all entries are automatically selected (see Figure 10).

Sugar metabolization - BioNumerics		- 🗆 ×
File Edit Database Analysis Scripts Window Help		
Experiment types	Database entries	Comparisons
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	 ✓ BSU1262 ✓ BSU1268 ● 	< >>
×	BSU1656 •	Identification projects Decision networks
Entry fields Database design	BSU1712 •	2 + 1 ² ⊗ €. En ∇ <all identific<="" td=""></all>
+ 1 ⊗ 8 1 √ ↑ ↓ «All Entry fields»	BSU1987 •	Name Modified date
Name Field type		
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Database: Sugar metabolization (_DefaultUser_) Entries: Loaded=6, View=6, Selected=6	1 experiments C:\Users\Public\Documents\BioNumerics\Data 80\Sugar metabolization This is a time limit	ed package valid until 2020-12-30

Figure 10: The *Main* window after import of the data.

The imported trend data is stored in the trend data type Carbon sources.

16. Double-click on the experiment **Carbon sources** in the *Experiment types* panel.

The 6 trend data curves are displayed in the *Curves* panel of the *Trend type* window (see Figure 11).

🖆 Trend data type '	Carbon sources'					-		Х
File Settings Tren	dCurves Parameters	Window	Help					
£ 1	≦] ∔ † †©⊡							
Curves				Parameters				
Name	Description			Parameter	Class	Descrip	otion	-
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✓ Inositol								
 Sorbitol 								~
✓ Rhamnose					<		3	>
 Sucrose 				Comparison set	tings			
 Lactose 				Companison set	ungo			_
								~
				Carbon source	ces settings			
				– Lab	els			
				X axis n				
				X axis u				
				Y axis u				
								\sim
Curves Crosslinks	Attachments			– Cur	ves			
Carbon sources								

Figure 11: The Trend type window.

5 Defining parameters

For visualization and comparison purposes, a default curve fit model needs to be specified.

1. In the *Trend type* window select *Settings* > *Default trend curve model...* to call the *Trend curve fit model* dialog box (see Figure 12).

Bacterial growth and activity is usually analyzed using a Logistic growth fit.

- 2. Choose Logistic growth and check both Use offset and Use generalized formula.
- 3. Press < OK > to set the curve model.

Trend curve fit model		?	×
Linear interpolation Cubic splines Lowess Polynomial Linear function Logarithmic function Exponential function Power function Hyperbolic function Gaussian function Logistic growth Gompertz Michaelis-Menten Cumulative normal distribution	Use offset	OK	

Figure 12: Set trend curve model.

Before any analysis can be done, parameters have to be defined. Parameters can be deduced from the model function (*Parameters* > *Model parameters...*) or from the original data points (*Parameters* > *Statistics parameters...*).

- 4. In the *Trend type* window select *Parameters* > *Model parameters...* to call the *Trend curve parameters* dialog box.
- 5. Select Logistic growth and check Use model.

For taxonomy or typing purposes one might be interested in combining data from multiple parameters into one clustering or identification.

6. Enable the parameters *Final value (Max)* (i.e. the maximum value derived from the curve) and *Maximum slope (Smax)* (i.e. the maximum growth rate) (see Figure 13) and press <*Exit*>.

Trend curve parameters		?	\times
Linear function Logarithmic function Exponential function Power function Hyperbolic function Gaussian function Logistic growth Gompertz Michaelis-Menten Cumulative normal distribution Model choices Use offset Use generalized formula	Use model Active parameters Initial value (Min) Final value (Max) Initial exponential growth rate (r) Initial doubling time (Tdoubl) Maximum slope (Smax) Time at maximum slope (TSmax) Time at 5% growth (T05) Time at 95% growth (T95)	Exit	

Figure 13: Trend curve parameters.

The *Parameters* panel now contains the selected curve model parameters to be used for comparison (see Figure 15). Our imported samples can now be compared based on the selected parameters rather than based on the original measurement points.

Select *Parameters > Parameter properties...* to call the *Parameter properties* dialog box (see Figure 14).

Parameter properties	? ×
Color range:	Minimum value: 0.0
R	Maximum value: 10.000
G	Coefficient: Pearson correlation $ \smallsetminus $
Add point Delete point	ОК
Add point Delete point	Cancel

Figure 14: Parameter properties.

- 8. Change the color scale of the selected parameter using the red, green and blue sliders and close the *Parameter properties* dialog box.
- 9. Change the color scale for the other parameter and close the dialog.

The color scales specified will be used when displaying the parameter values in the *Comparison* window (see 6).

🖆 Trend data type 'Carbon sourc	ces'								-		×				
File Settings TrendCurves Pa	arameters Window Help														
6 2 🖾 H+	1 ^c														
Curves			Parameters												
Name Desci	ription	-	Parameter	Class	Description	Minimum	Maximum	Color range			•				
Mannitol		^	Logist:Max	Logistic growth	Final value	0.0	10.000				^				
✓ Inositol			Logist:Smax	Logistic growth	Maximum slope	0.0	10.000								
 Sorbitol 															
 Rhamnose 											~				
 Sucrose 			Logist Smax Logistic growth Maximum slope 0.0 10.000												
✓ Lactose			comparison setun	ys											
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Curves Crosslinks Attachments	Parameters Description Parameter Class Description Maximum Color range Image: Color range Logist Max Logist provid Final value 0.0 10.000 Image: Color range Image: Color range: Color range Image: Color range: C														
Curves Crossinks Attachments	5		L												
Carbon sources											.:				

Figure 15: The *Trend type* window with two parameters defined.

10. Select *Settings* > *General settings...* (11) to call the *Curve settings* dialog box (see Figure 16).

Trend data type	e settings		?	×					
Properties									
X axis name:	Time								
X axis unit:	Hours								
Y axis unit:	mM								
Trend data visualisation									
Include zero in X axis									
Include zero in Y axis									
∠ Label curv	es with description								
	OK Ca	ancel	Appl	y					

Figure 16: General settings.

- 11. Enter "Time" as *X axis name*, "Hours" as *X axis unit*, and "mM" as *Y axis unit* (see Figure 16). Press <*OK*>.
- 12. Close the *Trend type* window with *File* > *Exit*.

6 Displaying trend data

1. Click on a colored dot in the *Experiment presence* panel of the *Main* window representing a trend data type for a particular entry.

The curves for the selected entry are displayed in the experiment card (see Figure 17). Using the pull-down list in the upper left corner of the card you can choose which curves to display.

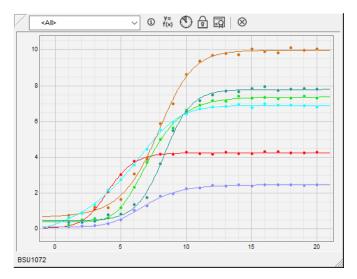


Figure 17: Experiment presence card.

2. Close the experiment card by clicking on the triangular button in the upper left corner.

Trend curves can also be displayed for multiple entries at a time in the same window. This is achieved as follows:

3. Select a number of entries in the database for which trend curves are present using the Ctrland Shift- keys. To select all entries at once select Ctrl+A.

Selected entries are marked by a checked ballot box (

- 4. Double-click on the experiment **Carbon sources** in the *Experiment types* panel to call the *Trend type* window.
- 5. Select *File* > *Create trend data window* () to open the *Trend data* window.

The *Curves panel* displays all curves for all selected entries in a single plot (see Figure 18).

- 6. Close the *Trend data* window with *File* > *Exit*.
- 7. Make sure a few entries are selected in the Main window.
- 8. Highlight the *Comparisons* panel in the *Main* window and select *Edit* > *Create new object...* (+) to create a new comparison for the selected entries.
- 9. Click on the next to the experiment name **Carbon sources** in the *Experiments* panel to display the selected curve model parameters in the *Experiment data* panel (see Figure 19).
- 10. Select *TrendData* > *Show parameter values colors* to display the values of the parameters together with the color as defined in the *Trend type* window (see 5).
- 11. Select a parameter in the *Experiment data* panel, and select *TrendData* > *Sort entries by parameter value* (\downarrow).

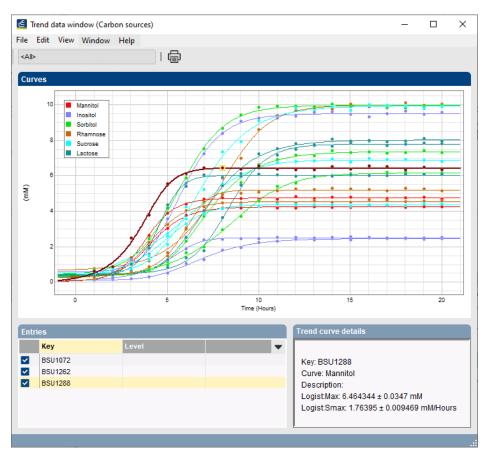


Figure 18: Trend curves displayed for multiple entries at a time.

Comparison																	-		×
File Edit Layout Groups Clustering	Statistics Fingerprints Characters Sequ	uence Trend	Data Rea	dSets Spect	ra Com	posite	Window	w He	elp										
86 € ≭*10:	Carbon sources		≦ 1	1910 1910	ħ.,														
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Carbon sources	Carbon sources																		
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🔁 😣		Mannitol Logist:Max Mannitol	nositol ogist:Ma	Logist:Sm Sorbitol Logist:Max	Sorbitol Logist:Sma	Rhamnose Logist:Max	Rhamnose Logist:Smax	Sucrose Logist:Max	Sucrose Logist:Sm	Lactose Logist:Max	Lactose Logist:Smax		Group	_	Кеу	Ŧ			
Name		4.26 1.0		0.42 7.36		10.12		6.98	0.93	7.87	1.55				BSU1072				-
Û		4.77 1.3		0.60 10.00	1.86	4.57		9.93	1.67	8.02	1.42				BSU1262				
< >		6.46 1.3	6 9.53	1.96 6.26	0.83	5.25	1.39	4.41	0.80	6.06	2.02	Image: A start and a start			BSU1288				
		8.85 2.3	22 5.17	0.87 4.75	0.87	9.22	2.42	4.00	1.32	0.07	0.02	~			BSU1656				
Groups		8.81 1.4		0.21 10.17	2.23	4.80		10.46	2.75	2.61	0.55	~			BSU1712				
		2.71 0.	34 7.20	1.96 8.58	0.88	0.88	0.31	9.20	2.36	1.38	0.21	×			BSU1987				
Size Name 🗸 🗸																			
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~													<			>	<		> <
6 entries in comparison 6 entries select	ted in database																		

Figure 19: The Comparison window.

The entries are sorted according to increasing value of the selected parameter.

- 12. A tab-delimited text file of the entries and trend data values contained in the comparison can be exported with *TrendData* > *Export character table*.
- Selecting *TrendData* > *Create trend data window* () calls the *Trend data* window again (see Figure 18).
- 14. A cluster analysis (*Clustering* > *Calculate* > *Cluster analysis (similarity matrix)...*) can be performed either by comparing the original data points of the curves, or by comparing the parameter values.

15. To calculate correlation and regression on trend data, open the *Trend analysis* window with *TrendData* > *Perform trend analysis* ($\boxed{\mathbb{M}}$).