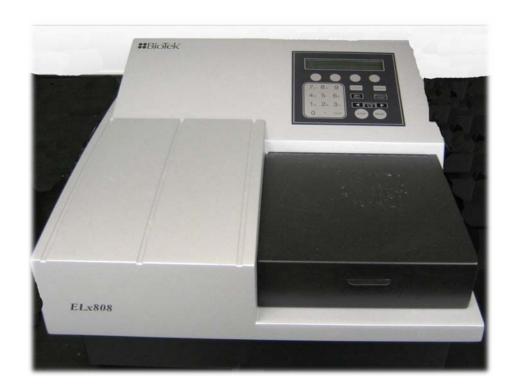
Biotechnology Lab



# BioTek ELx808 Absorbance Microplate Reader

## Bergen County Technical Schools Biotechnology Lab



#### Biotechnology Lab

#### BioTek ELx808 Absorbance Microplate Reader Information

The BioTek ELx808 Absorbance Microplate Reader is a multi-channel absorbance microplate reader that can be used for research and development and diagnostic use. The instrument essentially functions as a spectrophotometer, which measures intensity as a function of the color, or more specifically, the wavelength of light. The instrument has applications that would provide useful for multiple disciplines, from Biology, to Chemistry and Nanotechnology.

Unlike other microplate readers, the ELx808 does not support a wide range of labware. The ELx808 has been designed to only read 96 well plates.

The KC junior software that is installed with the instrument controls all of the measurements, actions, and protocols performed by the ELx808. The software supports detection of only absorbance, with applications that have been designed for use in areas such as Agriculture, Biotech Research, Cell Biology, Cell Counting, Cell Proliferation, ELISA, the Environment, and Neuroscience.

The software allows the user to establish unique protocols to analyze different sample plates. Samples are analyzed in the instrument, and the results stored on the computer's hard drive. The data from the experiments are able to be viewed and analyzed in the KC junior program, as well as in Excel by exporting the data.

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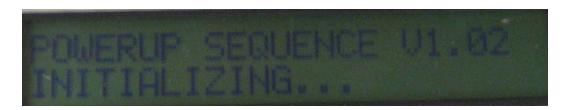


#### BioTek ELx808 Absorbance Microplate Reader Quick Start Guide

 Turn on the BioTek ELx808 Absorbance Microplate Reader by flipping the **Power Switch**, located on lower right side of the instrument.



Allow the instrument to perform its start-up procedure before proceeding.
 The screen on the instrument will show "Powerup Sequence V1.02
 Initializing" while the instrument is starting up.



b. When the instrument stops making noise, it is ready to use. The screen on the instrument will show "Ready" when it is ready to use.



2. On the desktop, click the **KC junior Icon** to start the software.

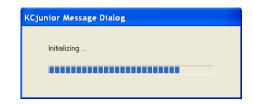


Note: The software will not work correctly if started before the instrument's screen displays "Ready".

Note: The user must be logged in under *donleo* on the computer in order for the software to work correctly with the instrument. Conditions have been optimized in this user setting. If the computer is not logged in as *donleo*, ask the instructor for assistance.

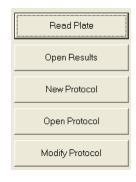
a. Before being able to run anything in the program, a screen will pop-up where the user needs to click **OK**. This will allow the instrument to initialize and communicate with the software.



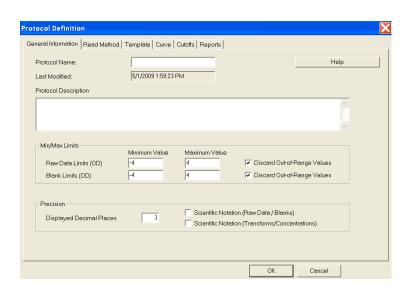


#### Setting up a Protocol

- Once the program starts, the user will have options such as Read Plate or Open Results, and New, Open, or Modify a Protocol on the *Main Menu Screen*.
- 2. Click **New Protocol** to create a new protocol.
- On the Protocol Definition screen that appears, select the General Information Tab.



General Information



a. Enter the **Protocol Name** in the area labeled.



- i. The protocol name should include the user's name, the sampleor experiment, and the data. (Ex StudentX\_ELISA\_1/1/09).
- b. Enter a **Protocol Description**.



- Information about the type of samples being run, wavelength's being used, and other general information can be included in this area.
- c. The remaining portions of this tab do not need to be changed.

Note: Do not click OK at the bottom of the screen until all tabs have been filled in. Only at the end of making the protocol should the OK button be pressed.

4. Select the **Read Methods Tab**.



General Information Read Meth	od Template Curve Cutoffs Reports		
Read Method Type	Endpoint ▼		Help
Primary Wavelength (nm)	<u> </u>		Get Reader Wavelengths
Reference Wavelength (nm)	•		Validate Read Method
Number of Plates	1		
Plate Geometry	8x12 Read A1 H12		
Delay Before Read (sec)	0		
Monitor Well	Monitor Well Params	Shake Method	None
Incubation	Г	Intensity  Duration	1
	37	Continuous Shake	
Read Mode	Normal	эр	

a. First select the **Read Method Type** that will be used for the plate.





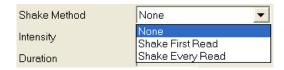
- Endpoint is the type most often used. This method scans the wells at the wavelength selected.
- ii. **Kinetic** is used for plates that need to be read multiple times over a set period of time.
- iii. **MultiWavelength** is used for plates that need to be scanned at more than one wavelength.
- b. Select the **Primary Wavelength** from the *Dropdown Menu*.



- For this instrument, the plates can be read at 405nm, 450nm, 490nm, or 630nm.
- c. A **Reference Wavelength** does not need to be set for the plate.
- d. The plate can be delayed before the read takes place by entering a time in the box next to **Delay Before Read (sec)**.



e. The instrument can be used to shake the plate prior to reading by selecting a **Shake Method** from the *Dropdown Menu*.



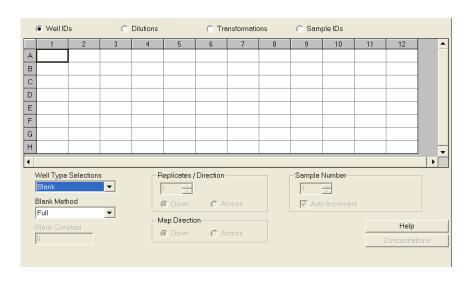
- i. The **Intensity** of the shake can be changed in the *Dropdown Menu*.
- ii. The amount of time in seconds to shake the plate can be entered in the box **Duration**.
- f. The temperature cannot be set for this instrument, so the area **Incubation** and **Temperature** cannot be selected or edited.

g. Once all of the read conditions for the plate are entered, click the **Validate**Read Method Button to make sure
that the protocol can be run correctly.



- If the read method is ok, a pop-up will tell the user that the read method is valid.
- ii. If the read method is not ok, the pop-up will instruct the user on what to change in the protocol.
- 5. Select the **Template Tab**.

Template



- a. In the template tab, the contents of each well in the plate can be assigned.
- b. Make sure that the **Well ID's Button** is selected.
- c. First select **Standards** from the *Dropdown Menu* under **Well Type Selections**.



 d. If there are replicates of the standards in the plate, select the number of replicates, and if they go down or across in Replicates / Direction.



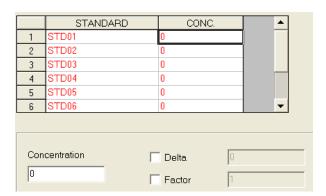
e. The **Sample Number** can be changed if the user does not want the number to start at one.



- f. On the template, click on a box to apply a standard or replicates of a standard to that box.
- g. Repeat steps c f for Samples, Blank, Controls, and Empty wells in the plate. Repeat until the entire plate template is established.
- h. Click the **Concentration Button** to set the concentration of the standards.



Delta



- Click on a standard and enter its concentration. Repeat for each standard.
- ii. Selecting the **Delta Button** will increase the next standard by the value entered.
- iii. Selecting the **Factor Button** will increase the next standard by a factor of the value entered.
- iv. Click OK.

 Click the **Dilutions Button** at the top of the screen if there are dilutions of a sample in the plate. This can only be applied to wells with a sample or control in it.

<ul><li>Dilutions</li></ul>	
Dilution	
1	

- i. Select the sample and enter the dilution factor.
- j. Click the Transformation Button at the top of the screen if formulas need to be applied to the results of samples in certain wells.



- i. Select the sample and enter the formula.
- k. Click the **Sample IDs Button** at the top of the screen to give a name to samples selected in the plate.



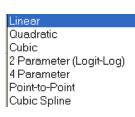
I. Make sure a template has been established for the entire plate.

	1	2	3	4	5	6	7	8	9	10	11	12
Α	STD01	STD01	CTL1	BLK	SMP01	SMP01						
В	STD02	STD02	CTL1	BLK	SMP02	SMP02						
С	STD03	STD03	CTL2	BLK	SMP03	SMP03						
D	STD04	STD04	CTL2	BLK	SMP04	SMP04						
Е	STD05	STD05	CTL3	BLK	SMP05	SMP05						
F	STD06	STD06	CTL3	BLK	SMP06	SMP06						
G	STD07	STD07	CTL4	BLK	SMP07	SMP07						
Н	STD08	STD08	CTL4	BLK	SMP08	SMP08						

6. Click the **Curve Tab** to set up the parameters of the standard curve that can be developed from the run.

Curve

- a. Select a **Curve Method** from the *Dropdown Menu*.
  - Linear is the type of graph used for most runs. Other graph options include Quadratic, Cubic, 2 Parameter, 4 Parameter, Point-to-Point, or Cubic Spine.



b. Enter a **Graph Report Name**.

Graph Report Name

i. The name should say "Standard Curve of ..." whatever the standard used in the plate is.

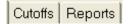
c. For the X and the Y axis, select if the scale should be **Linear** or **Logarithmic**.



d. Enter a **Label** for each axis if the user wants a unique name for each.

Label Absorbance

7. The sections **Cutoffs** and **Reports** do not need to be changed.



8. Click **OK** once the entire protocol is completed.

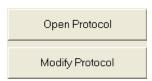


- 9. Before running the protocol, it must be saved.
  - a. Click the **Save Protocol Icon** on the right side of the *Toolbar* (the save icon on the left side is for experiments).



- b. Name the protocol in the provided directory with the user name, type of sample or experiment, and the date.
  - i. Ex. StudentX\_ELISA\_1/1/09 Protocol

Once the protocol is saved, it can be modified or opened at a later time using the options on the first menu page. If the protocol will not be run until a later data or time, it must be opened prior to starting a new run.



#### Running a Protocol on a Sample Plate

- 1. Make sure that an established protocol has been opened in the program.
- 2. Click the **Read Plate Button** on the *Main Menu Screen*.



3. A screen will pop-up. Enter a Results ID in the box.



- a. The results ID should include the student's name, sample, and date.
- 4. A **Plate Description** does not need to be entered, but can be with information about the plate being read.



5. Click **Read Plate** once the information has been entered.



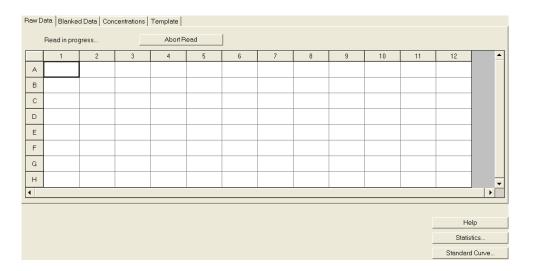
- 6. Place the microplate in the carrier.
- 7. On the pop-up screen, re-enter the result ID that was already given in the area labeled **Plate ID**.



8. Make sure the plate is on the carrier, and click **OK**. The instrument will read the absorbance of the samples in the plate.

#### **Analyzing the Results of the Plate**

1. A screen will pop-up after the run is completed. This contains all of the data from the run.



- a. Click the **Raw Data Tab**. This will provide the user with each well's actual reading from the instrument.
- Raw Data
- b. Click the **Blanked Data Tab**. This subtracts the average value from the wells labeled blank in the template from all other wells in the plate.
- Blanked Data
- c. Click the **Concentrations Tab**. This shows the concentration of each of the samples in the plate based on the absorbance reading and the values of the standards on the plate.
- d. Click the **Template Tab** to see what was in each of the wells in the plate.

Template

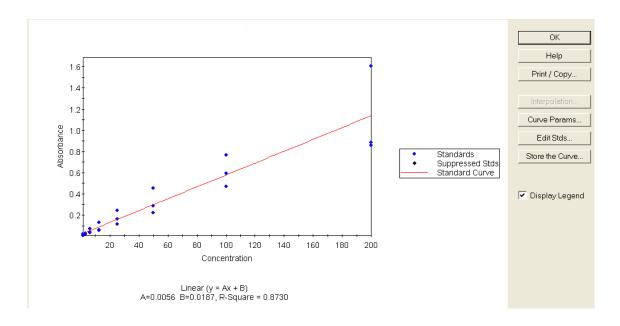
Click the **Statistics Button**. This lists all of the data in list form. It also groups replicates of samples and gives the average value of the replicates.

Statistics...

3. Click the Standard Curve Button.

Standard Curve...

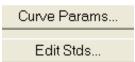
a. This button plots all of the standards from the plate.



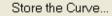
b. An **Equation for the Line** is given on the graph. Sample

concentrations can be determined by entering the absorbance readings into the equation.

c. The parameters set in the protocol for the curve can be adjusted by clicking the Curve Params...
Button, and the standard concentrations can be adjusted by clicking the Edit Stds... Button.



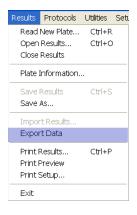
d. Click **Store the Curve** to save the curve. Make sure to name it with the Student's name, sample or experiment, and the date.

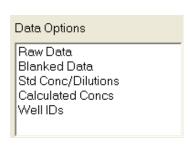


e. The graph and information can be printed from this screen by clicking the **Print / Copy Button**.

Print / Copy...

 To export the information from the run to Excel, click Results → Export Data on the Toolbar.





 Select the different types of data to export to Excel by selecting each and clicking the Add Button.



b. Click the **Export Button** to send the data to Excel.



c. In Excel, the data will show up in a format that matches the wells of the microplate, and will be labeled with what type of information is contained in each set.

12	Blanked Da	ata							
13	0.013833	0.007833	0.001833	0.002833	0.004833	0.001833	0.011833	0.001833	0.032833
14	0.001833	0.004833	-0.00217	0.003833	0.000833	0.003833	0.002833	0.000833	0.007833
15	0.008833	0.047833	0.002833	0.002833	0.003833	0.004833	0.003833	0.001833	0.031833
16	0.013833	0.002833	0.001833	0.001833	-0.00017	0.002833	0.004833	-0.00017	0.002833
17	0.009833	0.007833	0.004833	0.004833	0.005833	0.000833	0.002833	0.004833	0.081833
18	1.607833	0.765833	0.455833	0.242833	0.128833	0.068833	0.028833	0.020833	0.006833
19	0.857833	0.592833	0.287833	0.160833	0.061833	0.036833	0.016833	0.005833	-0.00217
20	0.882833	0.467833	0.222833	0.115833	0.053833	0.030833	0.014833	0.008833	-0.00317
21									
22	Raw Data								
23	0.062	0.056	0.05	0.051	0.053	0.05	0.06	0.05	0.081
24	0.05	0.053	0.046	0.052	0.049	0.052	0.051	0.049	0.056
25	0.057	0.096	0.051	0.051	0.052	0.053	0.052	0.05	0.08
26	0.062	0.051	0.05	0.05	0.048	0.051	0.053	0.048	0.051
27	0.058	0.056	0.053	0.053	0.054	0.049	0.051	0.053	0.13

Note: Results from runs can be accessed anytime after they are saved by clicking the **Open Results Button** on the *Main Menu Screen*.

Open Results

#### Saving Data and Shutting Down the Instrument

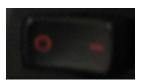
When finished with the experiment and data, click the Save
 Experiment Icon on the left side of the Toolbar to save the results.



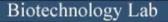
- a. Enter a name for the experiment in the directory that appears. Include the student's name, sample or experiment type, and the date.
- 2. Shut down the KC junior program by clicking the **(X) Button** in the top right corner of the screen.



 Make sure that the microplate was removed from the instrument, and turn the instrument off by pressing the Power Switch on the lower right side of the instrument.



4. Make sure the station is neat before leaving the instrument!



#### BioTek ELx808 Absorbance Microplate Reader Safety Sheet

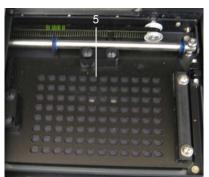
- 1. Samples should be handled according to good laboratory procedures and methods in order to prevent accidents.
- 2. Dispose of all waste solutions according to waste disposal procedures.
- 3. Do not remove any panels or cords from the instrument to avoid electrical shock.
- 4. The instrument has moving parts:
  - a. Do not attempt to exchange microplates while the instrument is operating.
  - b. Keep the work area around the instrument clear to avoid obstruction of the moving parts.
  - c. When the microplate carriage is open, only place a microplate in the carriage to avoid breaking the instrument.
  - d. Be aware of clothing and body parts when instrument doors are opening and closing.
- 5. Cleaning the workstation around the instrument is necessary. Never attempt to clean any internal spaces of the instrument.
- 6. Measurement values may be affected by extraneous particles (such as dust) in the microplate wells. A clean work area is necessary for an accurate reading.
- 7. Food and drinks should not be placed on or near the instrument.
- 8. If any liquid should fall near the instrument, do not operate the instrument. Fluid seepage into internal components creates a potential shock hazard, and can cause the instrument to not work properly.
- Running unauthorized programs or changing preferences on the instrument's computer is not allowed. Laptops are provided for personal use and internet access. Do not plug memory sticks in the instrument's computer unless instructed to do so by the instructor.

#### Biotechnology Lab

#### **BioTek ELx808 Absorbance Microplate Reader Instrument Information**

#### **ELx808 Absorbance Microplate Reader Overview:**





- 1. Instrument Information Screen
- 2. Instrument Information Entry Keys
- 3. Power Switch
- 4. Microplate Carrier Door
- 5. Microplate Carrier