Draft Microplate Reader Protocol April 30 2008 S. Tonsor

The Bio-Tek Instruments EL800 Universal Microplate Reader S/N 146704 is located in the Tonsor biochem lab, room 163 CD. The EL800 contains the following filters: Wavelength Position in filter wheel

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The EL800 is connected to a Dell Inspiron 4000 running Windows XP through COM1. The two machines interface through KCJunior software v1.41.8. The manuals for both the plate reader and the KCJunior software are located with the plate reader. The disk for the KCJunior software is in a sleeve taped to the inside of the manual.

- 1) Prepare your samples in a 96-well plate. Record sample i.d.s in a spreadsheet, noting the wells as A1 H12 from upper left to lower right.
- 2) Turn on the EL800 using the switch on the lower right side.
- 3) Turn on the laptop.
  - a. fire up excel
  - b. fire up KCJunior, using the desktop alias
- 4) You will see a number of options in the KCJunior window. The top option is 'READ PLATE'. Select 'READ PLATE'.
- 5) a dialog box will open asking for descriptive information about the plate. Fill in as appropriate and click on 'READ PLATE'.
- 6) A Protocol Definition dialogue box will open. Fill in the following:
  - a. READ METHOD: endpoint
  - b. PRIMARY WAVELENGTH: whatever you need; for RNA 595
  - c. REFERENCE WAVELENGTH: this should show the same ups and downs due to background as the primary and helps the EL 800 account for background noise. Any of the other filters' wavelengths can be used; choosing greater difference between primary and reference may be helpful.
  - d. PLATE GEOMETRY:
    - i. 96-well plate -- 8 x 12
    - ii. 24-well plate -- 4 x 6
  - e. Click o.k. or read plate or whatever it says
- 7) A Read Plate Dialogue box will open.
  - a. enter the plate i.d.
  - b. place the plate on the carrier with A1 in the back left corner.
  - c. click on OK
- 8) The plate will disappear into the EL800 and whirring and bumping noises will ensue. After a minute or so the plate will re-emerge.
- 9) KCJunior will display a matrix of absorbance readings.

- 10) From the File pulldown, select EXPORT DATA.
- 11) The EXPORT DATA dialogue box will open. Check the following:
  - a. EXPORT DESTINATION: Directly to Excel
  - b. output report header
  - c. output data descriptions
  - d. output empty row between plates
  - e. Add Raw Data to the DATA OPTIONS box if it isn't there already.
  - f. Click EXPORT DATA.
- 12) The data will appear in an excel spreadsheet.
  - a. SAVE the spreadsheet to this folder:
  - C:\Documents and Settings\All Users\Documents\KCJUNIOR DATA\ b. close the spreadsheet
- 13) Close KCJunior if you are done and turn off the EL800 and Dell laptop.

Load the plate