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TBS-380
Mini-Fluorometer

Operating Manual

Dated: 08/14/2007
Rev. E

TURNER BIOSYSTEMS

645 N. Mary Avenue • Sunnyvale, CA 94085
Phone: (408) 636-2400 • Toll Free: (888) 636.2401 • Fax: (408) 737-7919
www.turnerbiosystems.com

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User Notes and Information

Date	Notes/Information

1 Introduction

1.1 Description

The TBS-380 Mini-Fluorometer is a dual-channel fluorometer designed for quick, easy, and accurate fluorescence measurements. When properly calibrated with a standard of known concentration, the TBS-380 displays the actual concentration of the measured compound. The new Minicell adaptor design enables the user to measure compounds at a low concentration with a low assay volume (e.g. dsDNA at 1ng/mL using PicoGreen, and RNA at 1 ng/mL using RiboGreen).

1.2 Unpacking and Inspection

Upon receiving your instrument, inspect everything carefully and make sure all accessories are present (refer to the packing list shipped with the instrument). All shipments include:

- TBS-380 Mini-Fluorometer
- TBS-380 User's Manual
- TBS-380 Quick Reference Guide
- Power supply
- RS-232 cable
- Spreadsheet Interface software (one CD) and Instructions

1.2.1 Setup

To get started (see Section 2. Quick View Diagram):

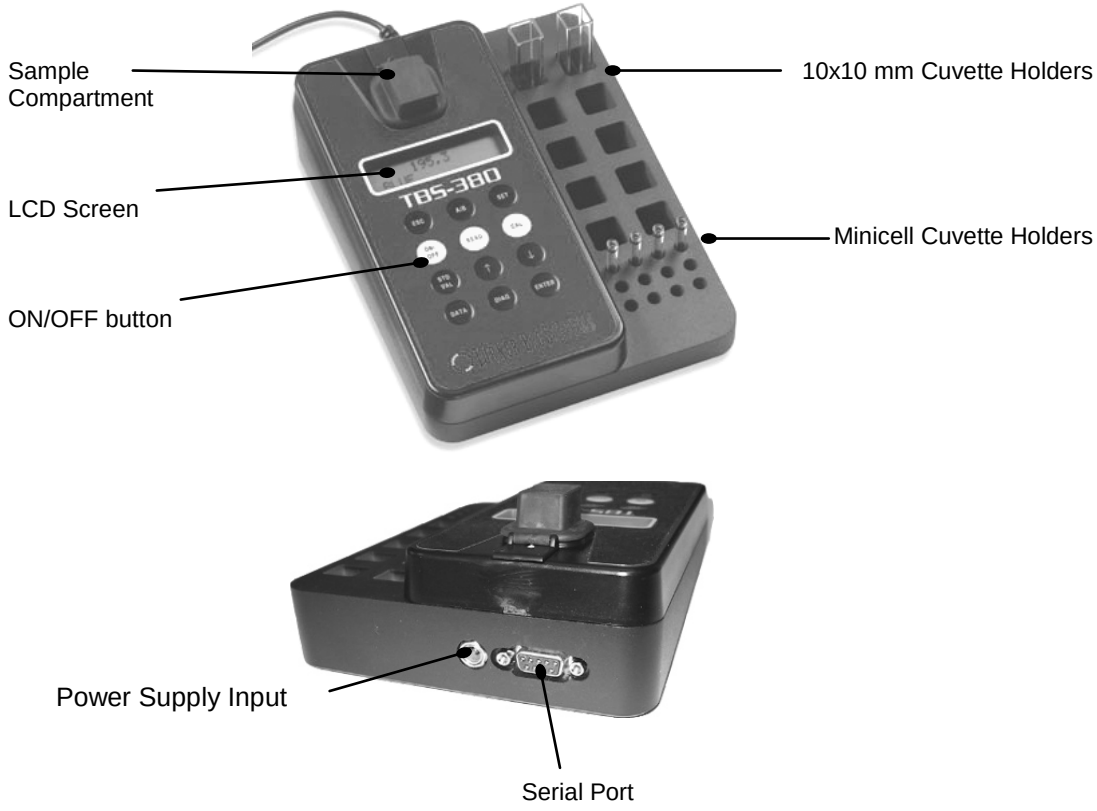
1. Position the instrument on a flat surface.
2. Assemble the power supply: choose the proper AC clip for your outlet and slide it on to the AC adaptor.
3. Plug the power supply into the TBS-380 (see Section 2. Quick View Diagram). **Be sure to use only the power supply provided with your instrument by Turner BioSystems.** Powering the instrument with an unapproved power supply may damage the unit.
4. To turn on the TBS-380, press the **ON/OFF** button. The instrument takes 5 seconds to warm up. After the warm-up, the TBS-380 is ready for operation.
5. To adjust the brightness of the LCD, press the **UP** or **DOWN ARROW** buttons.

1.3 General Information and Precautions

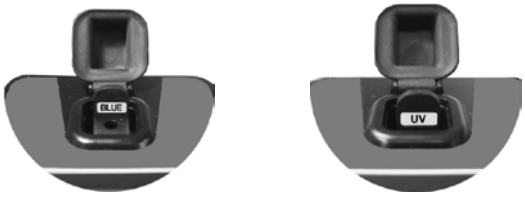
The sample compartment cannot accept glass or quartz cuvettes, except when using Minicell adaptor.

1. A minimum volume of 2 mL is required when using the 10x10 mm cuvette.
2. A minimum volume of 50 μL in a Minicell is required.
3. Avoid having any air bubbles in your sample. They can significantly affect the fluorescence reading.
4. When the Minicell is used, make sure to place the Minicell adaptor in the correct orientation. If BLUE channel is selected, insert the Minicell adaptor in the sample compartment with the tab away from you and the "BLUE" label facing you. If UV channel is selected, insert the Minicell adaptor in the sample compartment with the tab close to you and the "UV" label facing you (see Section 2. Quick View Diagram).

2 Quick View Diagram

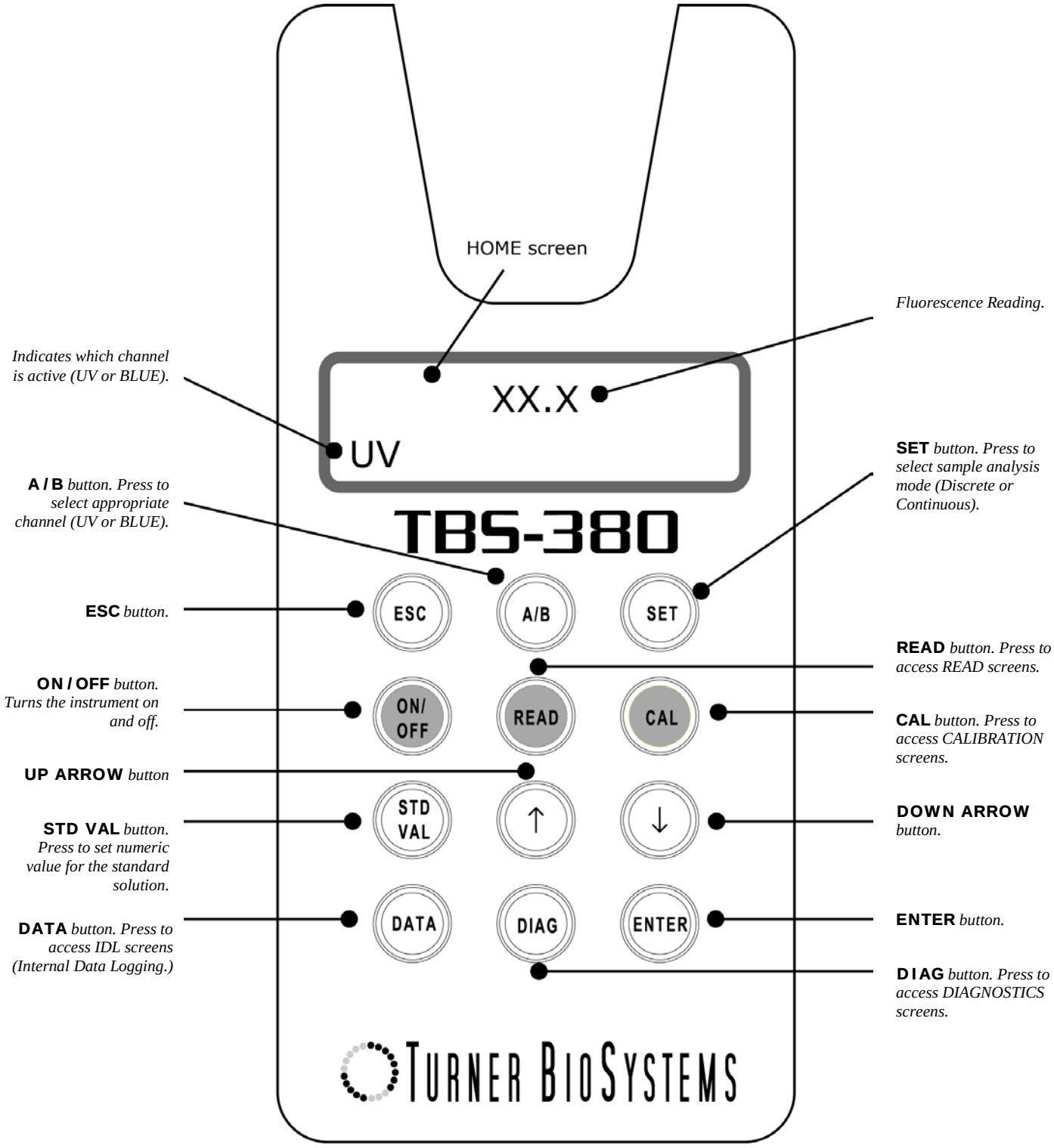


Positioning the Minicell Adaptor



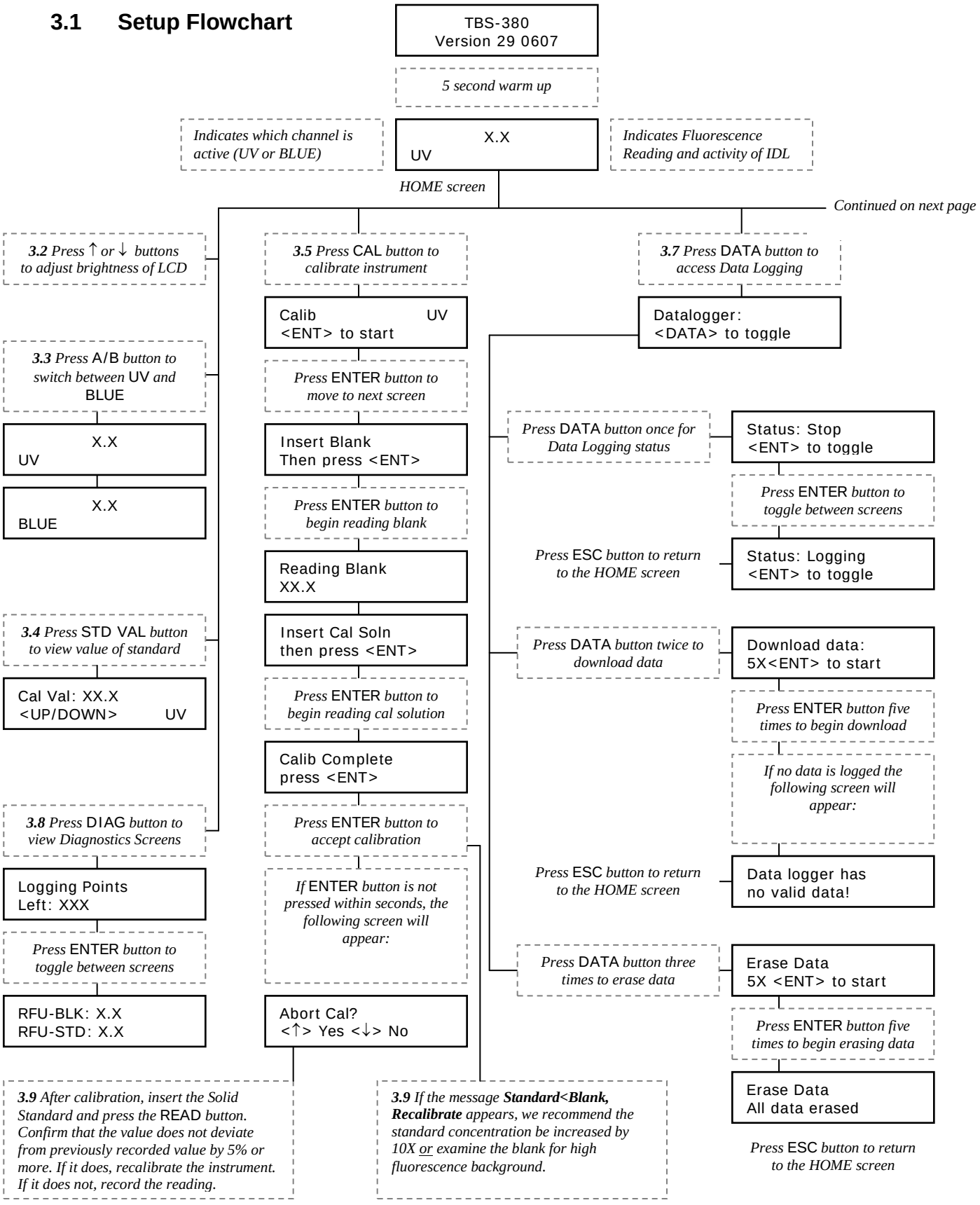
BLUE Position UV Position

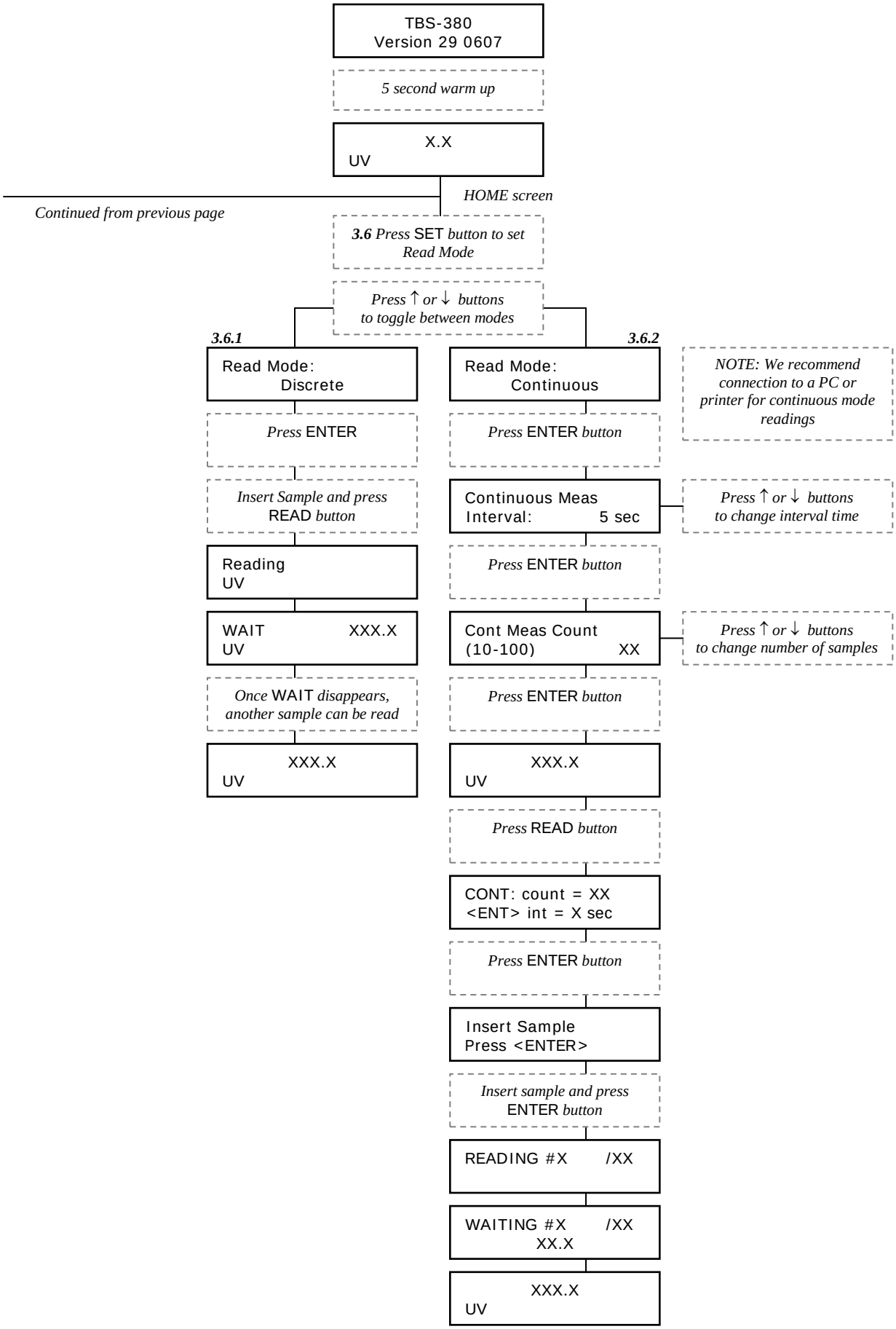
2 Quick View Diagram - Continued



3 Instrument Parameters

3.1 Setup Flowchart




TBS-380
Version 29 0607
5 second warm up
X.X
UV
HOME screen
Continued from previous page
3.6 Press SET button to set Read Mode
Press ↑ or ↓ buttons to toggle between modes
3.6.1
Read Mode:
Discrete
Press ENTER
Insert Sample and press READ button
Reading
UV
WAIT XXX.X
UV
Once WAIT disappears, another sample can be read
XXX.X
UV
3.6.2
Read Mode:
Continuous
NOTE: We recommend connection to a PC or printer for continuous mode readings
Press ENTER button
Continuous Meas
Interval: 5 sec
Press ↑ or ↓ buttons to change interval time
Press ENTER button
Cont Meas Count
(10-100) XX
Press ↑ or ↓ buttons to change number of samples
Press ENTER button
XXX.X
UV
Press READ button
CONT: count = XX
<ENT> int = X sec
Press ENTER button
Insert Sample
Press <ENTER>
Insert sample and press ENTER button
READING #X /XX
WAITING #X /XX
XX.X
XXX.X
UV

3.2 Instrument Power Up

To turn on the TBS-380, press the **ON/OFF** button. The instrument takes 5 seconds to warm up. After the warm-up, the TBS-380 is ready for operation.

To adjust the brightness of the LCD, press the **UP** or **DOWN ARROW** buttons.

3.3 Fluorescence Channels

Choose the appropriate channel for your analysis. To do this, press the **A/B** button to toggle between the 2 channels. The activated channel name will be displayed in the lower left corner of the HOME screen.

UV = Ultra Violet excitation
Blue = Blue excitation

See Appendix A for Optical (Wavelength) Specifications.

3.4 Calibration Standard Value

Before performing a calibration, assign a numeric value between 1 and 999 for your standard solution. (e.g. enter 100 if your calibration standard is 100 ng/mL).

1. Press the **STD VAL** button. Note: If no action is taken within 5 seconds of pressing the **STD VAL** button the instrument will default back to the HOME screen.
2. Use the **UP** or **DOWN ARROW** buttons to adjust the standard value. Holding the button down will activate faster scrolling.
3. When finished, press the **ENTER** button to accept the value and return to the HOME screen.

3.5 Calibration

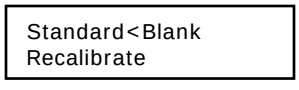
1. Press the **CAL** button.
2. Press the **ENTER** button to start the calibration.
3. Insert your blank and press the **ENTER** button. The TBS-380 will average the blank fluorescence for 5 seconds.

   NOTE*: Be sure to insert the Minicell adaptor in the correct orientation when the Minicell is used (see 1.3.5).*

4. Insert the calibration standard and press the **ENTER** button. The TBS-380 will average the standard fluorescence for 5 seconds.
5. To accept the calibration, press the **ENTER** button when the calibration is complete to accept the calibration. If **ENTER** is not pressed within 10 seconds, a dialog will appear asking the user if aborting the calibration is desired. Press the **UP** or **DOWN ARROW** button to abort or accept the calibration, respectively.

If desired, press the **ESC** button at any time during steps 1-4 to stop the calibration. The TBS-380 will return to the HOME screen and default back to the previous calibration.

NOTE: In the event that the Blank concentration is greater than the Standard concentration, the following error message will appear. We recommend increasing the concentration of the standard by 10X or examine the blank for high fluorescence background.



3.6 Sample Analysis

You can read samples in two reading modes: Discrete or Continuous.

3.6.1 Discrete Mode

In Discrete Mode, the instrument measures and averages the fluorescence signal over a 5-second interval.

1. Press the **SET** button.
2. Use the **UP** or **DOWN ARROW** button to select the “Discrete” mode.
3. Press the **ENTER** button.
4. Insert your sample.
5. Press the **READ** button. The result will be displayed at the top and center of the HOME screen.
6. The top left corner will display “WAIT” for 5 seconds. Once “WAIT” disappears, another sample reading can be performed.

3.6.2 Continuous Mode

In Continuous Mode, the instrument measures and averages the fluorescence signal over a 5-second interval for each sample measurement. The instrument measures the sample repeatedly for a user defined sample number.

1. Press the **SET** button.
2. Use the **UP** or **DOWN ARROW** button to select the “Continuous” mode.
3. Press the **ENTER** button.
4. Use the **UP** or **DOWN ARROW** button to select a measurement interval.
5. Press the **ENTER** button.
6. Use the **UP** or **DOWN ARROW** button to select the number of samples you want to measure.
7. Press the **ENTER** button.
8. Press the **READ** button.
9. The screen will display the setting selected.
10. Press the **ENTER** button.
11. Insert your sample and Press the **ENTER** button again.
12. Once all counts are measured, the result will be displayed at the top center of the HOME screen.
13. To abort the continuous sample measuring, press the **ESC** button and then the **ENTER** button.

3.7 Internal Data Logging (IDL)

The TBS-380 can log up to 1000 data points to the internal memory. The DATA screen controls logging, downloading and erasing of data.

3.7.1 Activating Data Logging

1. Press the **DATA** button.
2. Press the **DATA** button.
3. Press the **ENTER** button to toggle between logging and stop.
4. Press the **ESC** button when finished to return to the HOME screen.

3.7.2 Downloading Data

1. Connect the TBS-380 to the serial port of a PC using the provided or standard interface cable.
2. Launch the Spreadsheet Interface software. See Appendix B for computer requirements and installation instructions.
3. Press the **DATA** button.
4. Press the **DATA** button 2 times.
5. Press the **ENTER** button 5 times to start the data download.
6. Press the **ESC** button when finished to return to the HOME screen.

3.7.3 Erasing Data

1. Press the **DATA** button.
2. Press the **DATA** button 3 times.
3. Press the **ENTER** button 5 times to erase all logged data.
4. Press the **ESC** button when finished to return to the HOME screen.

3.8 Diagnostic Information

1. Press the **DIAG** button to access the DIAGNOSTIC screen.
2. The first screen shows the number of data points available for internal data logging.
3. Press the **ENTER** button to toggle to the RFU (Relative Fluorescence Units) values from the calibration blank and standard.
4. Press the **ESC** button when finished to return to the HOME screen.

3.9 Solid Standard

The Solid Standard is designed to verify the stability of the instrument by checking drift of the calibration. It will also verify if any instrument settings have been changed.

1. After calibration, insert the Solid Standard into the sample compartment of the TBS-380 Mini-Fluorometer. (**NOTE**: always insert it with the tab away from you)
2. Press the **READ** button.
3. Confirm that the current measured value does not deviate from previously recorded value by 5% or more. If the value of the Solid Standard deviates by more than 5%, it is time to recalibrate the instrument (see 3.5).
4. Record the reading.
5. Check the calibration and/or the stability of the instrument by periodically reading the Solid Standard.

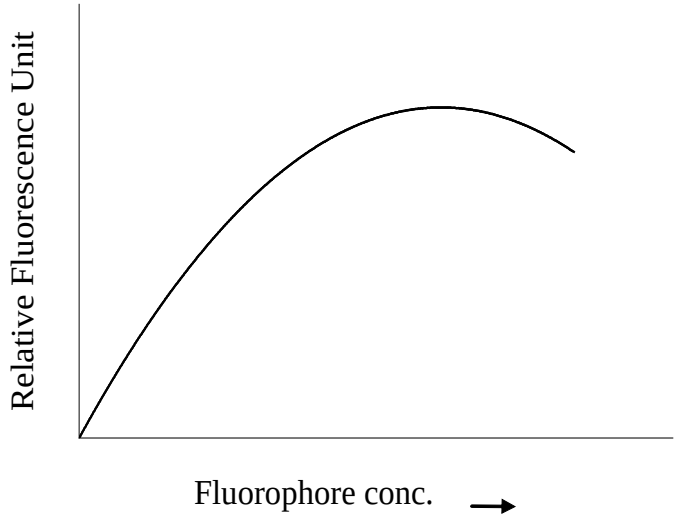
4 General Considerations for Analysis

4.1 Handling Samples

1. Be cautious not to spill samples into the sample chamber. Promptly wipe any spills.
2. The TBS-380 is very sensitive and even small amounts of material left behind from a previous sample may result in errors. Use a clean cuvette or disposable cuvette for each reading.
3. Fill the cuvette to the minimum required volume, significant error will result if it is not filled enough. A minimum volume of 2 mL in a 10x10 mm cuvette or 50 μL in the Minicell is required for best results.
4. When taking readings, the cuvette MUST BE CLEAN AND DRY. Moisture and condensation on the outside can result in erroneous readings.
5. Minute bubbles in samples will also cause errors in readings. Be sure not to introduce bubbles into samples. Particular care must be taken with the Minicell. Tapping the outside cuvette wall slightly will often help dissipate bubbles.

4.2 Linear Range and Quenching

The linear range is the concentration range in which the readout of the TBS-380 is directly proportional to the concentration of the fluorophore. The linear range begins with the smallest detectable concentration and extends to an upper limit (concentration) that is dependent upon the properties of the fluorescent material, the filters used, and the path length. See the diagram below.



A nonlinear relationship is seen at higher concentrations where the fluorescence signal increases slower than the change in concentration. At even higher concentrations, readings will begin to decrease even though the sample concentrations are increasing. This effect is known as “signal quenching.”

Linearity may be checked by diluting a sample 1:1 or some other convenient ratio (be sure to use an appropriate blank for the dilutions). If the sample is within the linear range, the reading will decrease in direct proportion to the dilution. If the reading does not decrease in direct proportion to the dilution or if the reading increases, the sample is beyond the linear range of the fluorophore.

4.3 Temperature Considerations

Fluorescence is temperature sensitive. As the temperature of the sample increases, fluorescence decreases and vice versa. For greatest accuracy, read the blank, standard, and samples at the same temperature.

4.4 Positioning Samples

For low concentration samples, cuvettes will often give slightly different measurements depending upon their orientation in the sample compartment. This is due to defects in the shape of the cuvette that are not visible to the human eye. We recommend that the cuvette be marked at the top and positioned in the sample compartment the same way each time to minimize error.

4.5 Data Quality

The TBS-380 is only as accurate as the standards that are used to calibrate it. It is important to follow correct laboratory practices when preparing all solutions and reagents including standards, samples, and blank.

5 Troubleshooting

The following list defines potential problems that a user may encounter.

5.1 Low or No Reading

- Check with the Solid Standard to determine if the instrument is functioning properly.
- Are all the reagents and standards made to specification? Are any of the reagents expired? Is the dilution done correctly?
- Has the sample been exposed to light for an extended period? Some fluorescent dyes are susceptible to photodegradation. It is recommended that the dye solution be protected from the light.
- Is the right optical channel selected for the application? Has the instrument been calibrated prior to reading?
- Is the sample in a detectable, linear range for the instrument? Sample concentration might be too low or too high to be detected (see 4.2).
- Is the right type of cuvette being used? Methacrylate cuvettes should be used for UV applications.
- Check temperature (see 4.3).
- Does the sample volume meet the minimum requirement? The minimum volume is 2 mL for 10x10 mm cuvette, or 50 μL for Minicell.
- Is the Minicell adaptor inserted in the correct orientation? If the BLUE channel is selected, insert the Minicell adaptor in the sample compartment with the tab away from you and the BLUE label facing you. If UV channel is selected, insert the Minicell adaptor in the sample compartment with the tab close to you and the "UV" label facing you.
- Is the orientation of the Minicell cuvettes consistent (see 4.4)?
- What is the RFU (Relative Fluorescence Unit) of the blank and calibration standard? Press the **DIAG** button and **ENTER** to display the RFU values (see 3.8). If the instrument was calibrated with the highest concentration standard and the RFU of the standard is too close to that of the blank, the sample readings may be out of the instrument's detection limit.

  Ideally:

$$\frac{\text{RFU-STD}}{\text{RFU-Blk}} \geq 3$$

5.2 High or Over Reading

- Dilute samples and standards.

5.3 Fluctuating Results

- Check the Solid Standard to determine if the instrument is functioning properly.
- Check for spills in the sample compartment.
- Check the volume of the sample.
- The minimum volume is 2 mL for 10x10 mm cuvette, 50 μL for the Minicell.
- Check that the reagents and cuvette material are compatible.
- Check temperature (see 4.3).
- Confirm that the RFU-STD is greater than RFU-Blk by factor of ≥ 3.

6 Warranty

6.1 Terms

Turner BioSystems warrants the TBS-380 Mini Fluorometer and any accessories to be free from defects in materials and workmanship under normal use for a period of one year from the time of initial purchase under the following restrictions:

- The instrument and accessories must be installed, powered, and operated in compliance with the directions in this *Operating Manual* and the directions accompanying the accessories.
- Damage incurred during shipping is not covered by warranty.
- Damage resulting from measuring incompatible samples is not covered by warranty.
- Damage resulting from reagent spills is not covered by warranty.
- Damage resulting from contact with corrosive materials or atmosphere is not covered by warranty.
- Damage from salts, proteins and other moderately corrosive materials that are not promptly removed from the instrument are not covered by warranty.
- Damage caused by user modification of the instrument is not covered by warranty.

6.2 Warranty Service

To obtain service during the warranty period, the owner shall take the following steps:

1. Write or call the Turner BioSystems Customer Service and describe as precisely as possible the nature of the problem.

2. Carry out minor adjustments or tests as suggested by the Technical Support Department.

3. If proper the instrument is still not functioning properly, you must obtain an RMA number from the Turner BioSystems Service Department before shipping the instrument. This number is necessary for tracking and repair under warranty.

4. Reference the RMA number on the outside of the package. Pack the instrument well as damage incurred during shipping due to improper packing is not covered by warranty. Purchase shipping insurance for the package and ship it to the Turner BioSystems Service Department. All shipments must be prepaid.

The instrument will be repaired and returned free of charge for any customer in the United States. Turner BioSystems will pay for return shipment and include a reimbursement check for the cost of surface shipment to the Service Department.

Turner BioSystems cannot pay shipping, duties, or documentation costs outside the continental United States. Contact Turner BioSystems for instructions if you are an international customer who made a purchase directly from Turner BioSystems (not from a third party distributor). Customers outside of the United States who have purchased our equipment from an authorized distributor should contact their distributor directly for further instructions. The instrument will be repaired at no charge if it is under warranty.

IMPORTANT: Under no circumstances should the instrument or accessories be returned without prior authorization from Turner BioSystems or our authorized distributor.

Prior correspondence is needed to:

- Eliminate minor problems easily handled on-site in your laboratory in order to save money and minimize equipment downtime.
- Determine the exact nature of the problem so that repairs can be tailored to the defect you have noted.

6.3 Out-of-Warranty Service

Proceed exactly as for Warranty Service, above. If our service department can assist you by phone or correspondence, we will be glad to at no charge.

Repair service will be billed on a basis of time and materials. A complete statement of time spent and materials used will be supplied. Shipment to Turner BioSystems should be prepaid. Your bill will include return shipment freight charges.

Address for Shipment:

Turner BioSystems
645 N. Mary Ave.
Sunnyvale, CA 94085, USA

Phone: 1(408) 636-2400
Toll-Free: 1(888) 636-2401 (US/Canada)
Fax: 1(408) 737-7919

Appendix A: Instrument Specifications

A1. General Specifications

Specification	Description
Size	2.7” x 5.5” x 7.25” (6.9cm x 14cm x 18.42cm)
Weight	1.8 lbs (0.67 kg)
Dynamic range	4 orders of magnitude
ADC Resolution	12 bits
LCD Display	2 x 16 characters
Operating Temperature	59-98 F; 15-40 ^{o}C
Detectors	Photodiodes: measurement capability from 300-1000nm
Calibration Type	Single-point and blank
Alarms	High blank
Cuvette Type	10x10 mm plastic, 10x10 mm Minicell Adaptor
Warm Up Time	5 seconds

A2. Optical Specifications

Light Source	UV LED	Blue LED
Excitation Optics	365–395 nm	465–485 nm
Emission Optics	440–470 nm	515–575 nm

Appendix B: Internal Data Logging

B1. Shipping Checklist

1. RS-232 Cable
2. Spreadsheet Interface software (1 CD) and instructions

Both of these items are necessary for retrieving the stored data in the TBS-380.

B2. Hardware Requirements

1. PC with Windows® 95 or later
2. MS Excel 5.0 or later
3. At least 1 available serial port

B3. Software Installation

1. Exit all Windows programs.
2. Insert the CD and run the setup program.
3. The setup wizard will install the necessary files.
4. When the setup is complete, an icon named “Spreadsheet Interface Software” will appear in the “Programs” menu.
5. Restart your computer.

B4. Connecting

1. Using the provided or standard RS-232 cable, connect the 9 pin connector of the cable into the available serial port of your computer.
2. Plug the opposite end of the cable into the base of the TBS-380.
3. Open the Spreadsheet Interface software.
4. Click on the box to the right of the COM port icon to select the appropriate COM port. This is usually COM port 2.
5. Click on “Start”. The program will find and open MS Excel and should be ready for data transfer. The boxes left of the COM port and MS Excel should both be green.
6. Follow the directions from Section 3.7 for collecting and downloading data from the TBS-380. Data will automatically appear in the MS Excel spreadsheet. Be sure to save this data before closing the software.

B5. Troubleshooting

Difficulties can arise when parameters are set incorrectly or connections with the cable are not tight enough. Here are some common problems.

1. Box to the left of the COM port is red. This means that the COM port is not available. Causes:
   a. Another instrument or program (such as palm pilot/ hot sync) could be occupying the port, making it unavailable. Make sure to close all programs of this type before downloading data
   b. The port selected is incorrect. Follow step 4 of connecting to choose another COM port.
2. All lights are green, but no data has been transferred, even though the instrument shows “All data downloaded”.
   a. The connection between the instrument and the computer is bad. Check and tighten the cable connections. Make sure both ends of the cable are plugged in tightly.

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