

FAST 2014

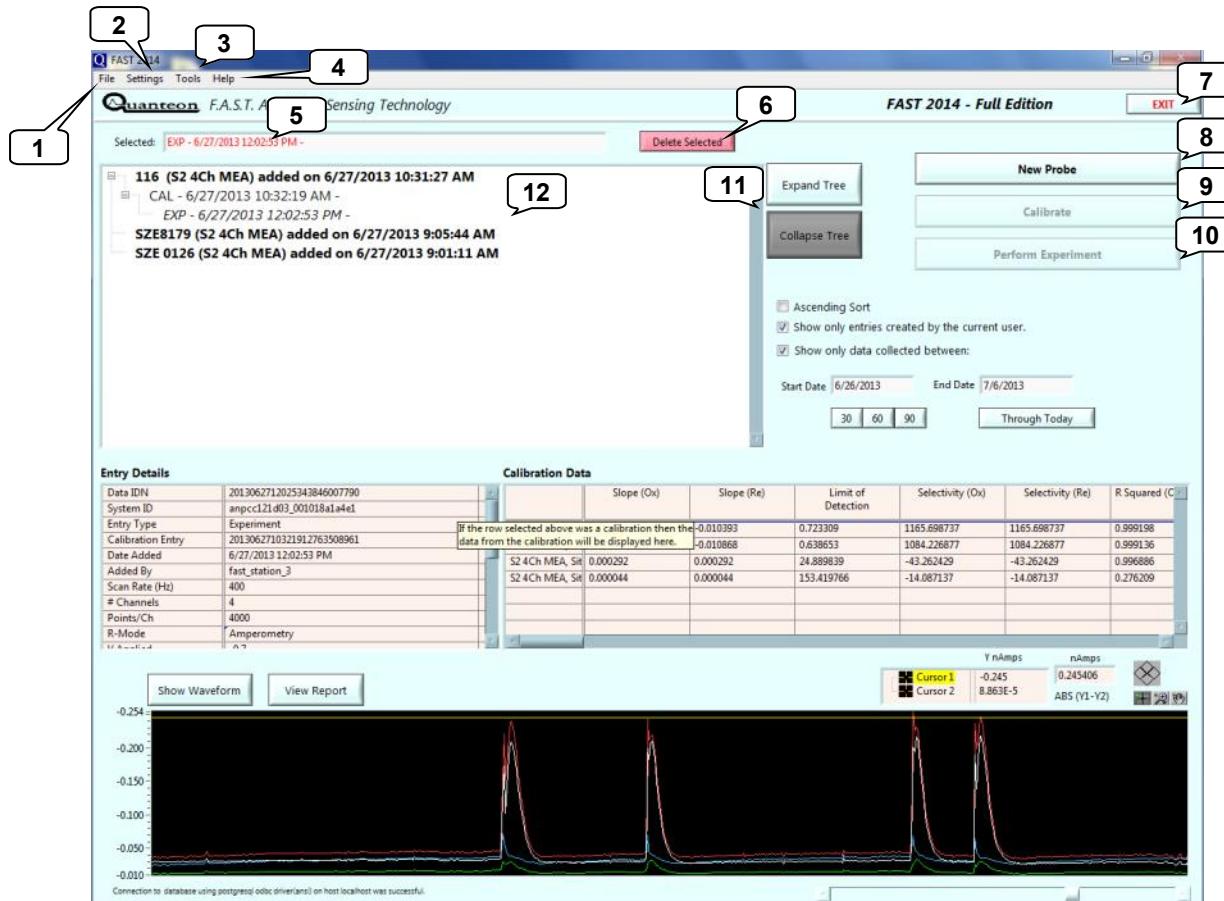
Software Manual



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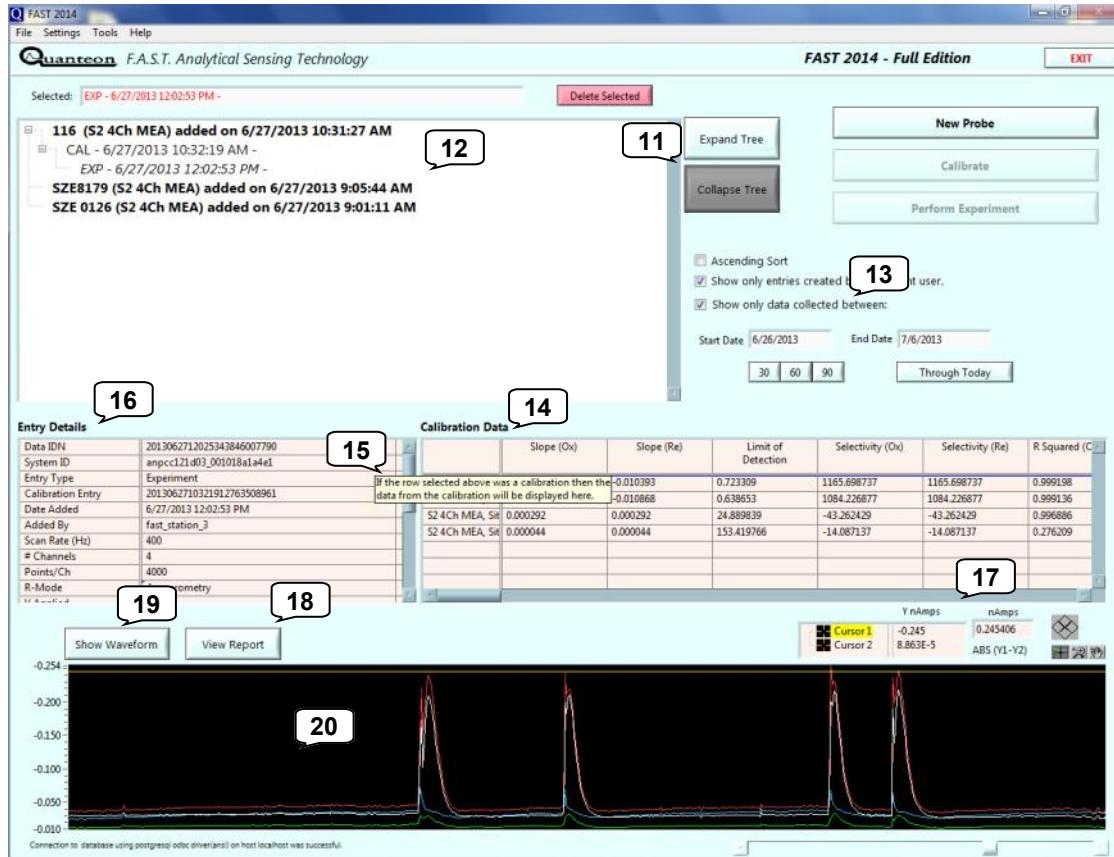
1.0 FAST System Software - Main window



Double click the  on the desktop to open the FAST System Software. The FAST Main window appears. The items on the window are:

1. [File](#) - used to export .html reports and data files for analysis
2. [SETTINGS](#) - Adjust system settings.
3. [TOOLS](#) - Access a concentration calculator.
4. [HELP](#) - Calibrate and perform experiments.
5. Identifies entry for further action by double-clicking on database list.
6. **Delete Selected** (#5) database item (CAUTION: Deletion cannot be undone.)
7. EXIT the FAST program.
8. **STARTING A NEW RECORDING** - Name a New Probe.
9. [Calibrate](#) a new probe. Select after double-clicking a probe in (#12).
10. [Perform Experiment](#). Select after double-clicking a completed calibration (#12).

1.0 FAST System Software - Main window

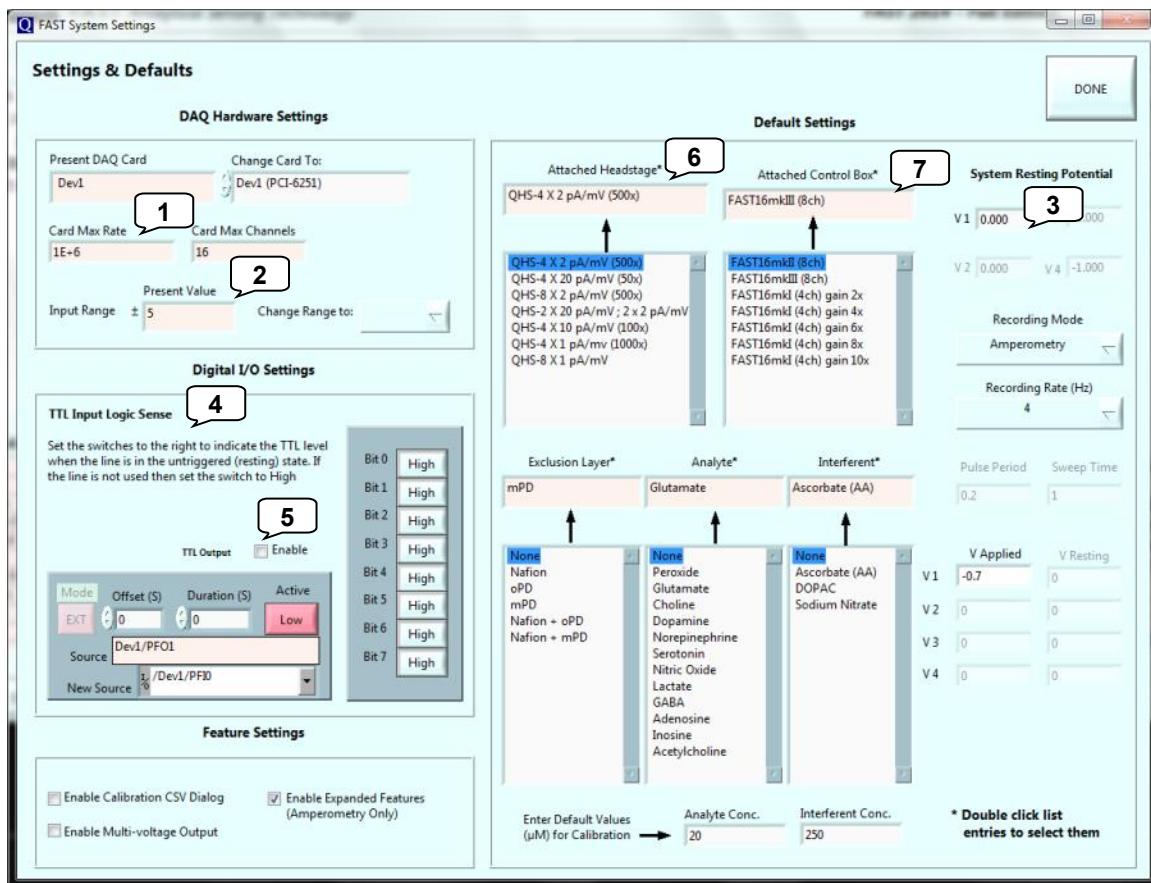


11. Expand Tree or Collapse Tree to expand or contract database list hierarchy.
12. Database lists MEAs/carbon fibers and respective (CAL)ibrations and (EXP)eriments as selected in (#13).
13. Adjust options/selections to modify database range in (#12).
14. Displays calibration results from item selected in (#12).
15. Information pop-ups appear throughout the program. Hover the mouse pointer over entry fields and buttons to reveal the information pop-ups.
16. **Entry Details** - lists recording settings and MEA/carbon fiber information.
17. Cursor values from Show Waveform window.
18. [**View Report**](#) - click button to view HTML report for selected (in #5) item.
19. **Show Waveform** - click button to display traces for selected (in #5) item.
20. Quick view of recording traces. Current (nA) is displayed on the y-axis.

**Amperometry Recordings:
Neurochemistry with MEAs**

1.1 Amperometry Settings & Defaults

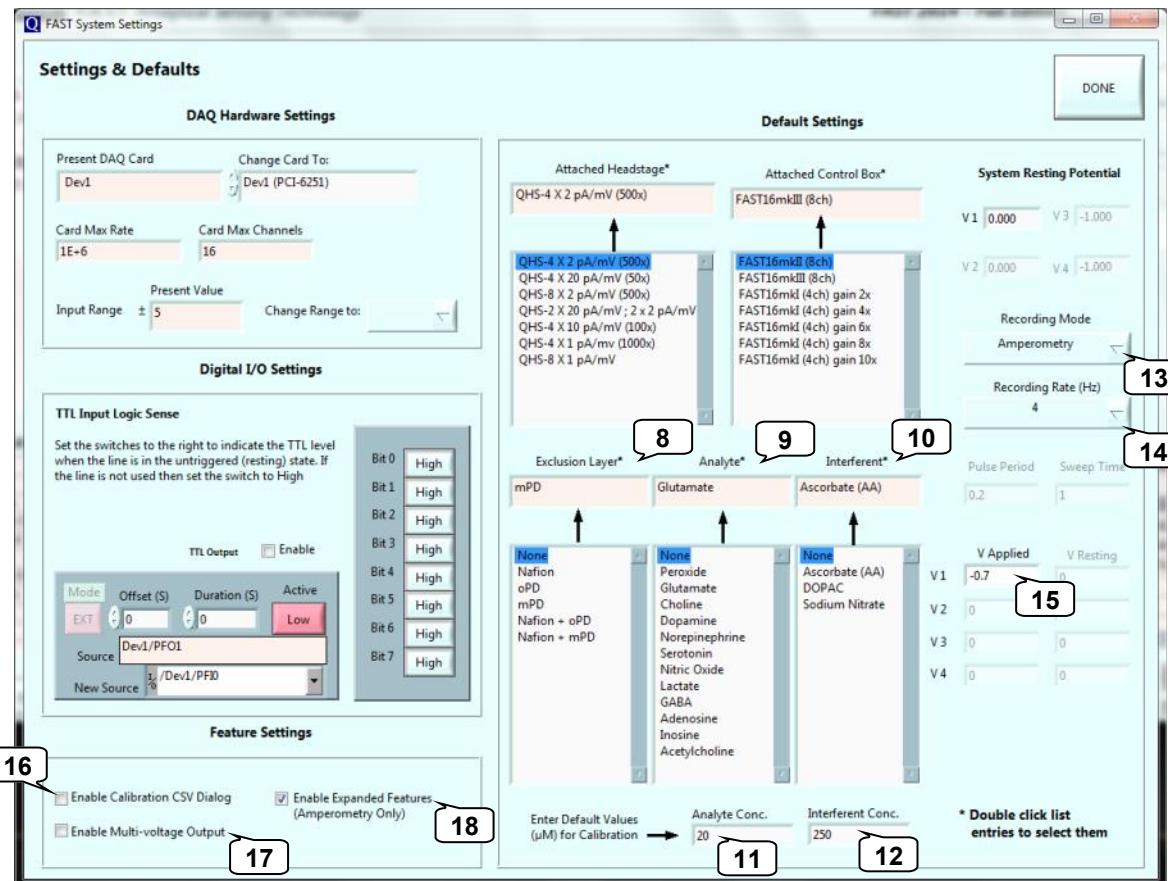
Enzyme-based MEA



- Card Max Rate** - Describes how often the [analog to digital conversion](#) occurs per channel.
- Input Range** - 0.1 to 10V. From drop-down menu, most often select 1 or 2 for best signal detection. Describes [analog to digital conversion range](#).
- System Resting Potential** - Maintains applied potential between recording episodes.
- TTL Input Logic Sense** - Sets the logic level on the digital inputs that indicate the untriggered state of attached equipment.
- TTL Output** - A tick in the box enables the output of a trigger signal that begins on the rising edge of the square wave pulse used in chronoamperometry. Used to synchronize external equipment such as data collection systems to the FAST system.
- Attached Headstage** - Select the headstage that is attached to the FAST control box.
- Attached Control Box** - Select appropriate control box. If selecting "FAST16mkI," match the gain to the one selected on the front panel of the FAST control box.

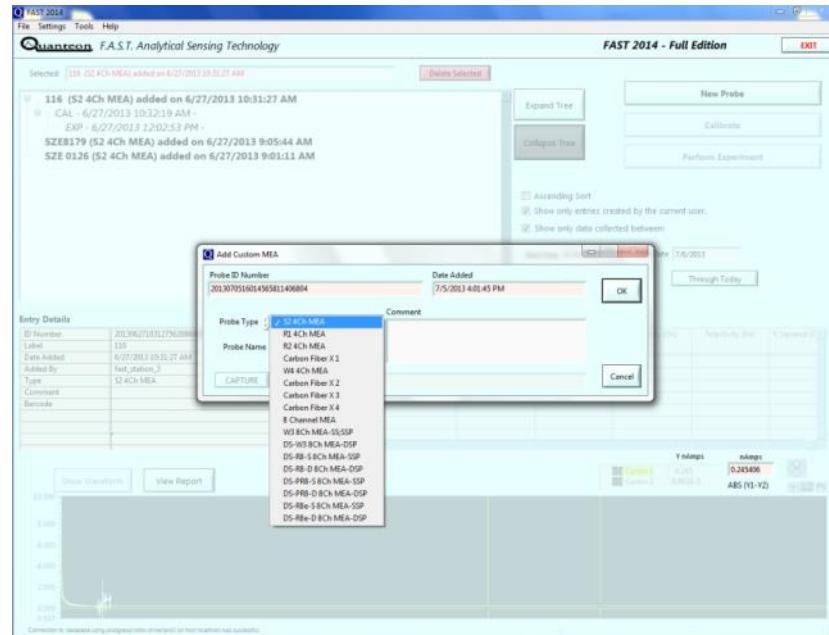
1.1 Amperometry Settings & Defaults

Enzyme-based MEA

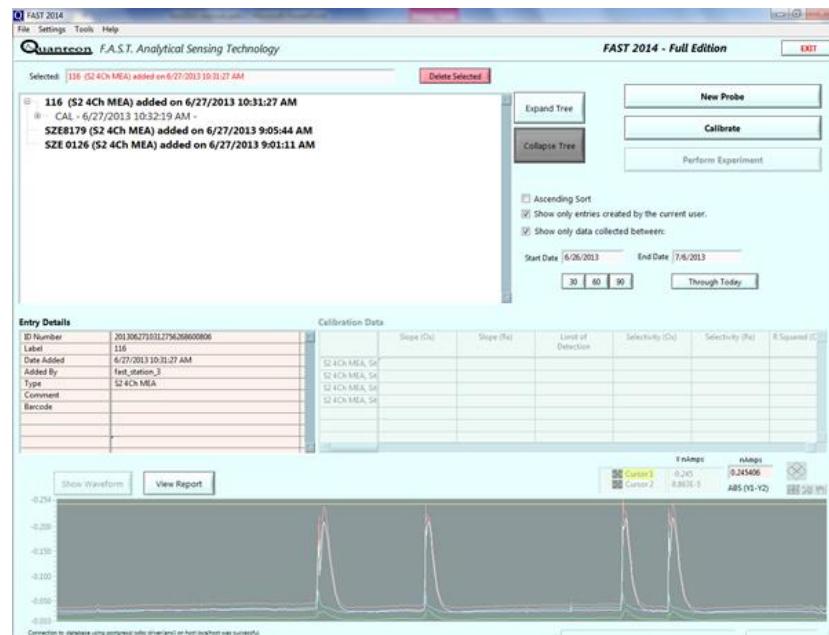


8. **Default Exclusion Layer** - Default exclusion layer, if one, for calibrations. (**Defaults**: double clicking a selection from the list sets/changes the default. The current default is shown in the text box above the list).
9. **Default Analyte** - Default analyte, if one, for calibrations.
10. **Default Interferent** - Default interferent, if one, for calibrations
11. **Analyte Concentration** - Default analyte concentration for calibrations.
12. **Interferent Concentration** - Default interferent concentration for calibrations.
13. **Recording Mode** - MEAs are calibrated in **Amperometry** mode.
14. **Recording Rate (Hz)** - Use the pull down menu to select the rate data is saved to disk.
15. **V applied** - Enter the potential to be applied vs. reference electrode.
16. **Enable Calibration CSV dialog** - Add calibration results to a new or existing spreadsheet.
17. **Enable Multi-voltage Output** - Permits use of multiple applied potentials (advance only).
18. **Enable Expanded Features** - Displays Custom MEA Calibration Setup window.

1.2 Recording with an MEA

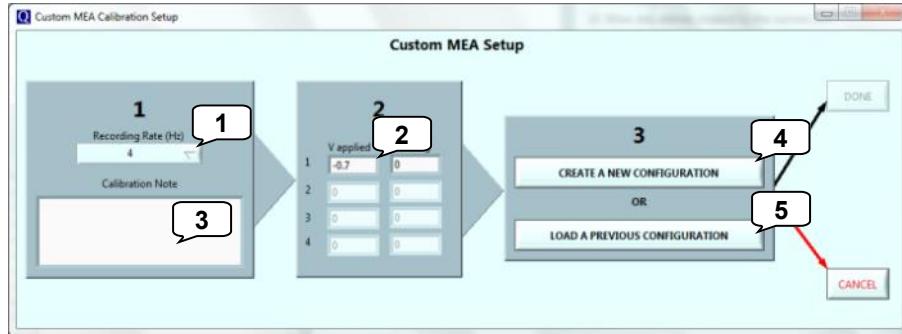


After verifying **Settings**, select **New Probe**. Choose **Probe Type** from the drop down menu, provide a **Probe Name**, and include additional **Comments**, as needed. The information will be displayed in the database list.

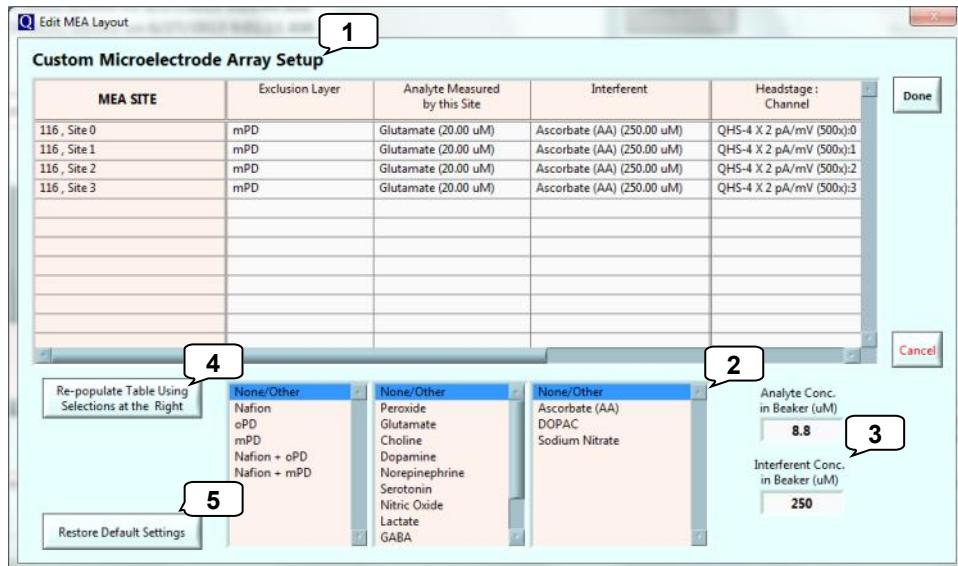


After creating an MEA entry, select the sensor of interest in the database and the **Calibrate** button will become active. Select to begin an [MEA calibration](#).

1.3 Amperometry Calibrations - with expanded features enabled in Defaults and Settings



1. **Recording Rate (Hz)** - Use the pull down menu to select the rate data is saved to disk.
2. **V applied** - Enter the potential to be applied vs. reference electrode.
3. **Calibration Description** - Enter calibration description to aid with database identification.
4. **Create a New Configuration** - Configure new calibration parameters.
5. **Load a Previous Configuration** - Re-use previously defined calibration parameters.



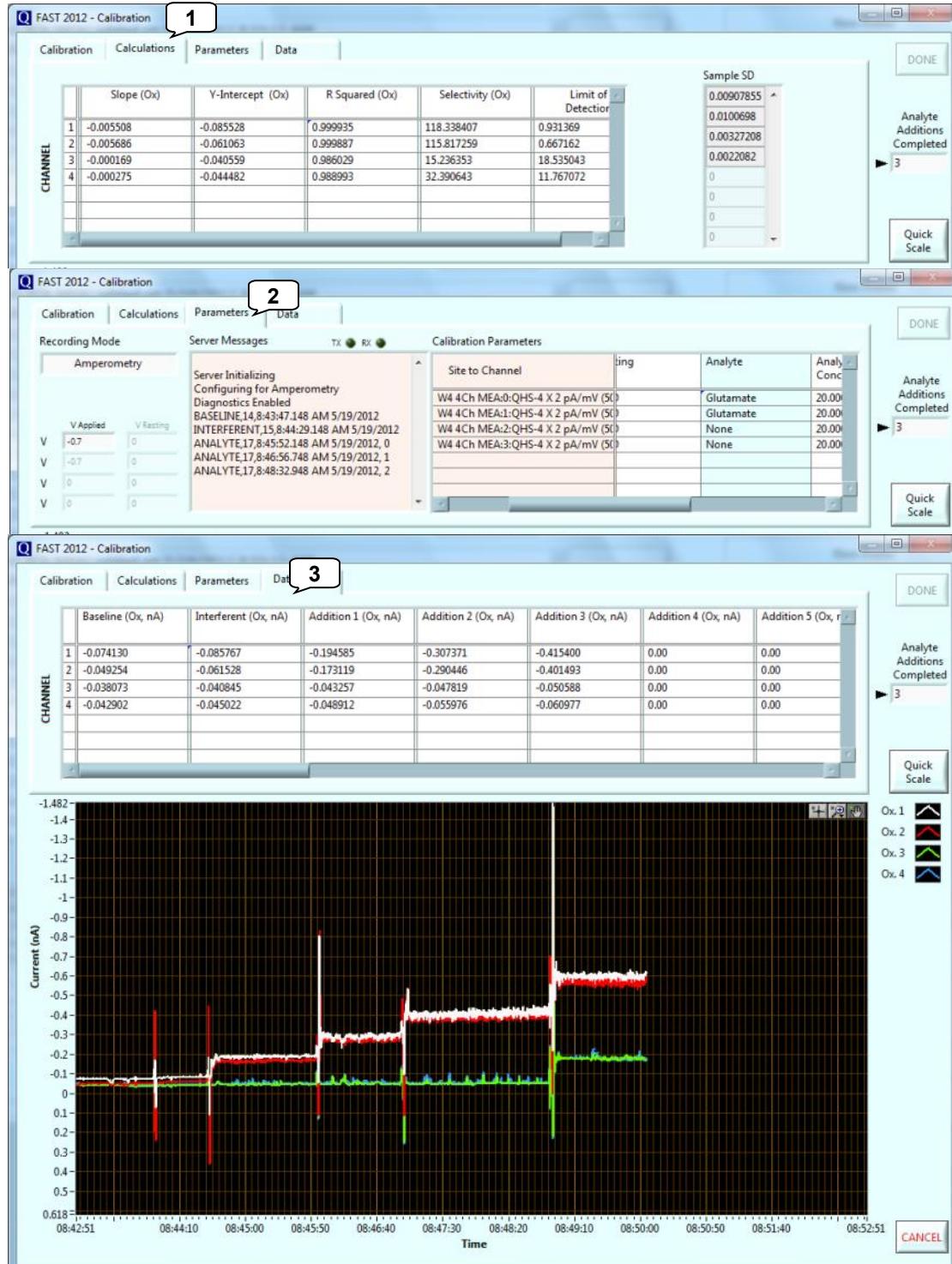
1. **Setup** - Displays MEA settings. Initially populated with default settings.
2. **Calibration Adjustments** - If selecting different values than default, double-click on items to change individual MEA sites in (1) or select (4) to change for all MEA sites.
3. **Analyte & Interferent Concentrations** - If selecting different concentration values than default, enter beaker concentration values.
4. **Re-populate Table** - Select to apply changes from (2) or (3) to MEA setup.
5. **Restore Default Settings** - Select to restore default settings.

1.3 Amperometry Calibrations



1. Current (nA) value for each MEA channel.
2. Graphical display of the current values of individual MEA channels (nA vs. time of day).
3. Magnification and scrolling tools allow for customization of the graphical display.
4. Counter showing total number of additions for calibrations.
5. Re-scale to zoom on the active area of the graph.
6. Active MEA channel colors—modifiable.
7. Click to toggle between displaying/turning off the oscilloscope.
8. Oscilloscope displays wave form or current level generated at microelectrode (in V).
9. Activate **BASELINE** button to begin calibration calculations. Buttons change to **Interferent** then **Analyte** and **Test Substance** based on calibration protocol.
10. Select **Finished** to end calibration calculation.
11. Select **DONE** to close calibration window, otherwise, to abort calibration select **CANCEL**.

1.3 Amperometry Calibrations



- Calculations** - MEA calibration results (including LOD, selectivity, and slope).
- Parameters** - Displays system parameters used for calibration.
- Data** - Displays data used for calibration **Calculations**.

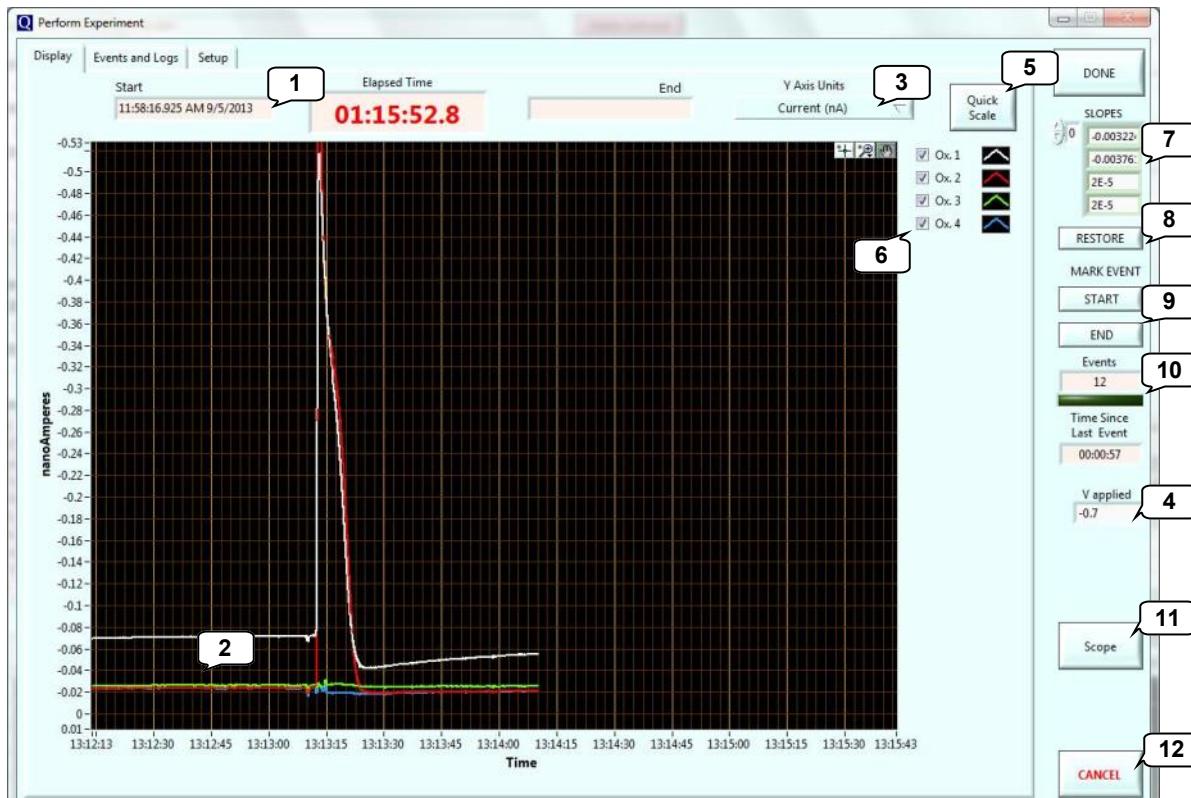
1.4 Amperometry Experiments



After creating a calibration,

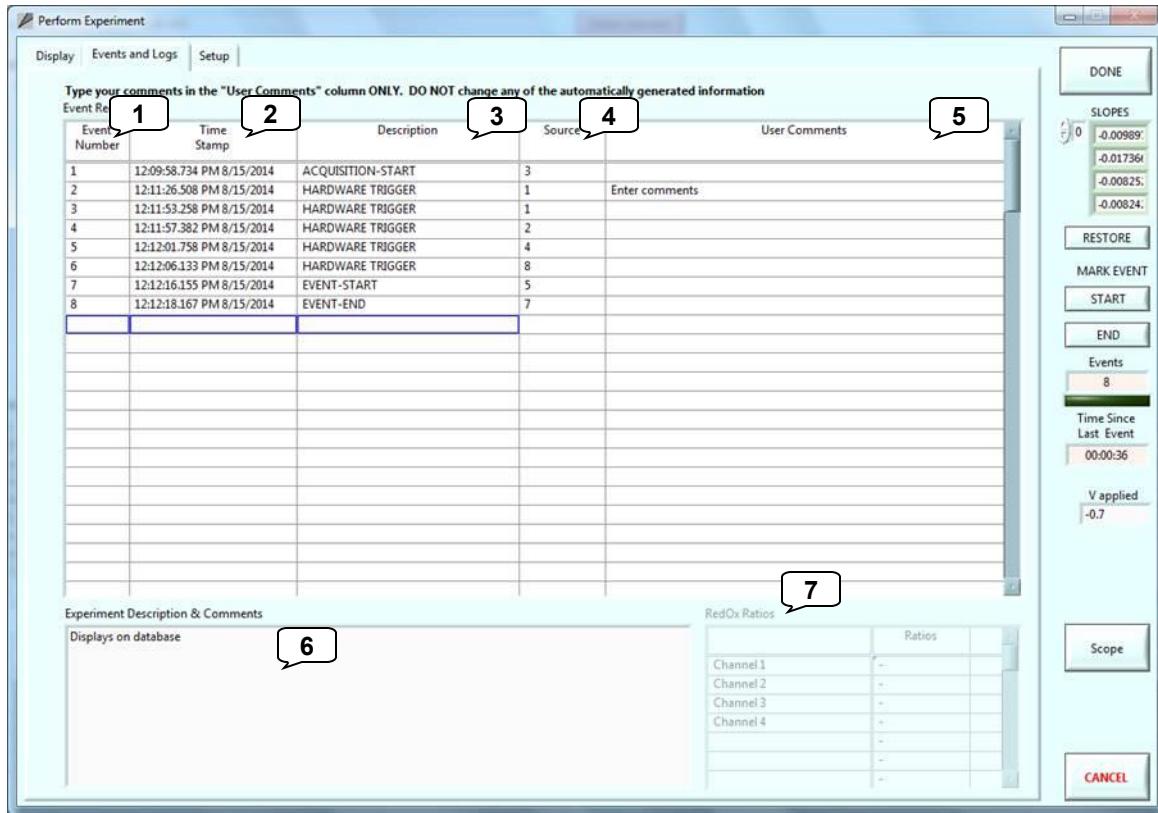
1. Select a calibration to use for an experiment by double-clicking on that calibration.
2. Parameters associated with the MEA and calibration and calibration calculations (LOD, selectivity, Slope) are displayed.
3. Select **PERFORM EXPERIMENT** to open acquisition window and begin an experiment.

1.4 Amperometry Experiments



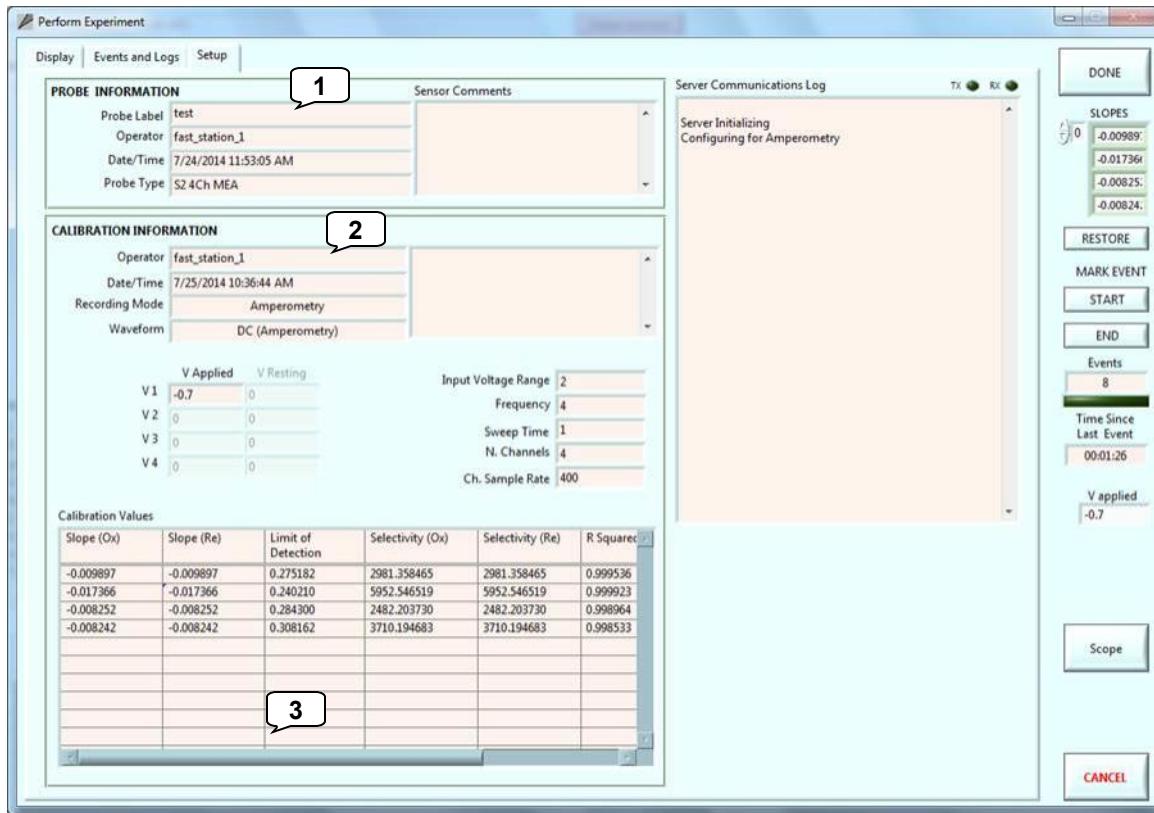
1. **Start** time stamp for acquisition data file, **total elapsed recording time**, and **End** time stamp for acquisition data file.
2. Graphical display of the current (nA or pA) or concentration (μM) values of individual MEA channels (vs. time of day).
3. Toggle between **Current** (nA or pA) and **Concentration** (μM) display for Y-Axis Units.
4. **V applied** - The applied potential may be changed on-the-fly during experiment.
5. Re-scale to zoom on the active area of the graph.
6. Active MEA channel colors—modifiable. Display/hide individual channels.
7. Microelectrode **slopes** from calibration. These are automatically obtained from selected calibration file but can be manually entered as needed.
8. **RESTORE SLOPES** to original calibration file values for selected MEA.
9. Click to signal **START** and **END** of select triggered **EVENT**. Useful for long recording signals, drug, or behavior studies. Adds TTL event mark to data file for analysis.
10. **Events** - Counts number of triggered events and **Time Since Last TTL Event**.
11. **Scope** - Display/Hide oscilloscope.
12. Select **DONE** to close Experiment window, otherwise, to abort experiment select **CANCEL** (data not saved).

1.4 Amperometry Experiments



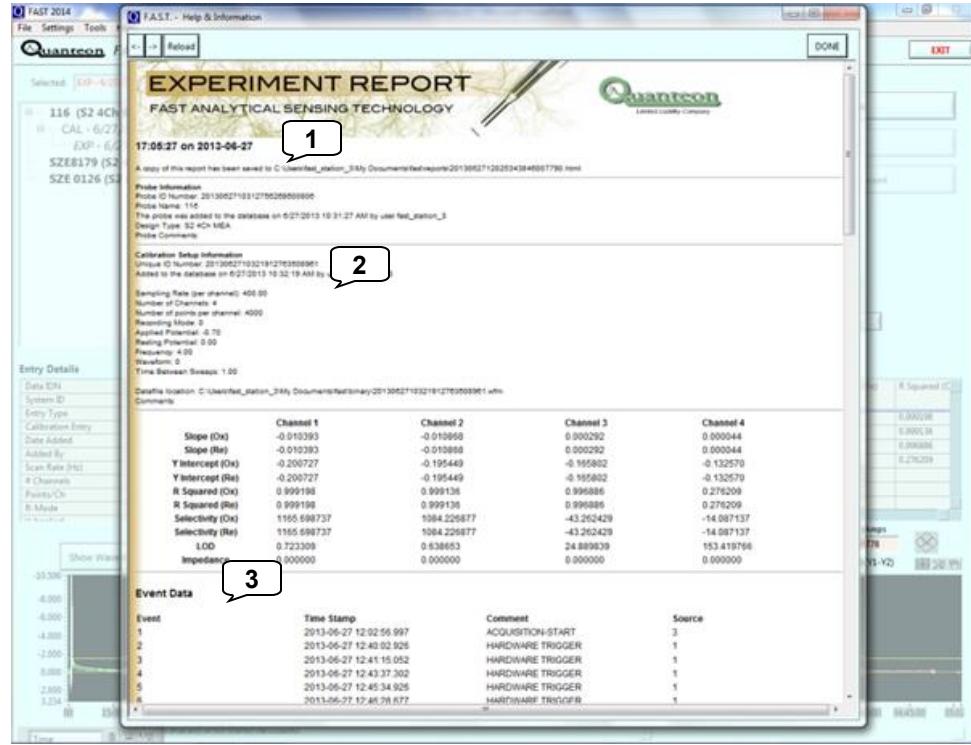
1. **Event Number** - indicates order of TTL events.
2. **Time Stamp** – Time TTL was triggered (Time determined by computer clock).
3. **Description** – Displays the source of the TTL input.
4. **Source** – Displays which TTL input was triggered.
5. **User Comments** - Text field for describing TTL events.
6. **Experiment Description & Comments** - Text field for describing experiment. Viewable in **Browse Database**.
7. **RedOx Ratio** – In chronoamperometry, ratio between reduction current and oxidation current.

1.4 Amperometry Experiments



1. **SENSOR INFORMATION** - Displays MEA parameters.
 2. **CALIBRATION INFORMATION** - Calibration/experiment parameters for selected recording MEA.
 3. **CALIBRATION VALUES** - Displays MEA calibration calculations

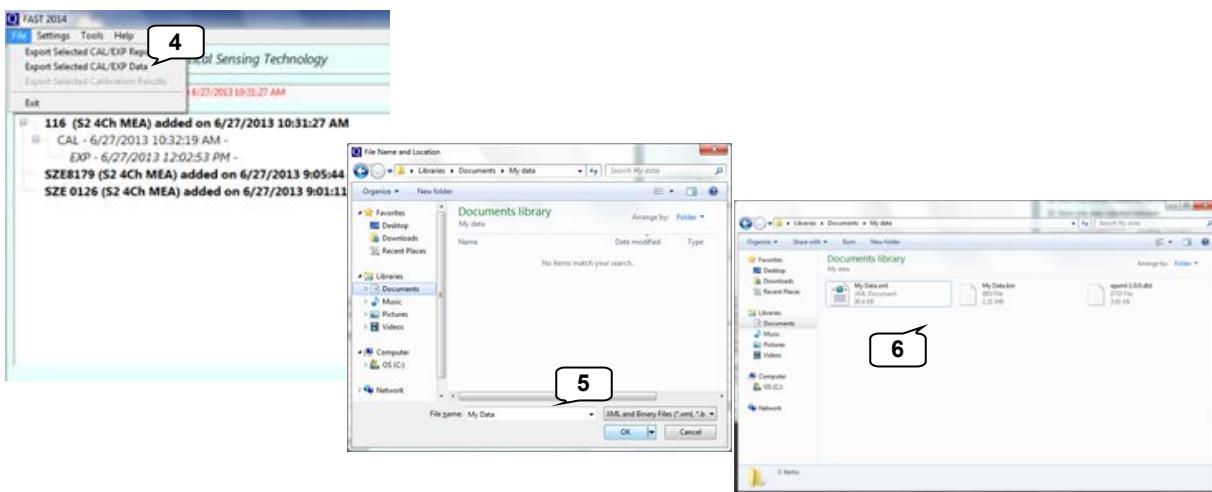
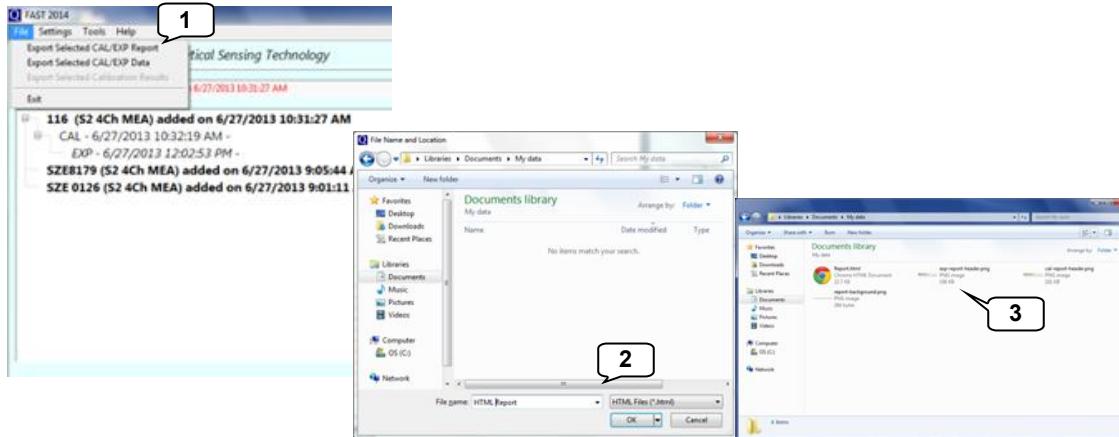
1.4 Amperometry Experiments - Viewing reports



View Report from the Main Screen

1. **Sensor Information** - [Information about MEA](#).
2. **Calibration Setup Information** - Calibration parameters and calculations.
3. **Event Data** - Lists **Event #**, **Time Stamp**, **Comments**, **Source** for the event.

1.4 Amperometry Experiments - Exporting files

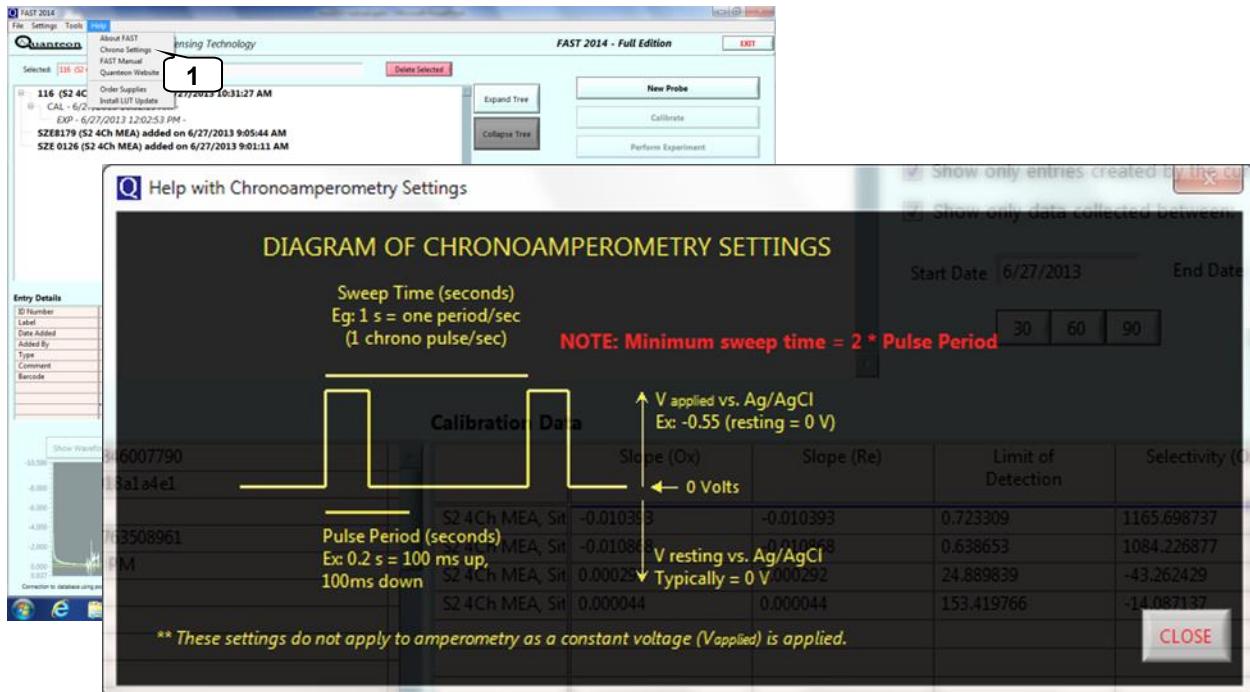


1. **Export Selected CAL/EXP Report** - Used to export a report. [Select entry on the home screen \(#5\)](#)
2. Provide a file name. Exported as .html
3. Exporting the report generates an .html file and header images.
4. **Export Selected CAL/EXP Data** - Export data files used in the [FAST Analysis](#) program a report. [Select entry on the home screen \(#5\)](#)
5. Provide a file name.
6. Exporting data files generates .xml, .bit, .dtd files that are all needed for analysis with [FAST Analysis](#).

**Chronoamperometry Recordings:
Neurochemistry with Carbon Fibers**

2.1 Chronoamperometry Settings & Defaults

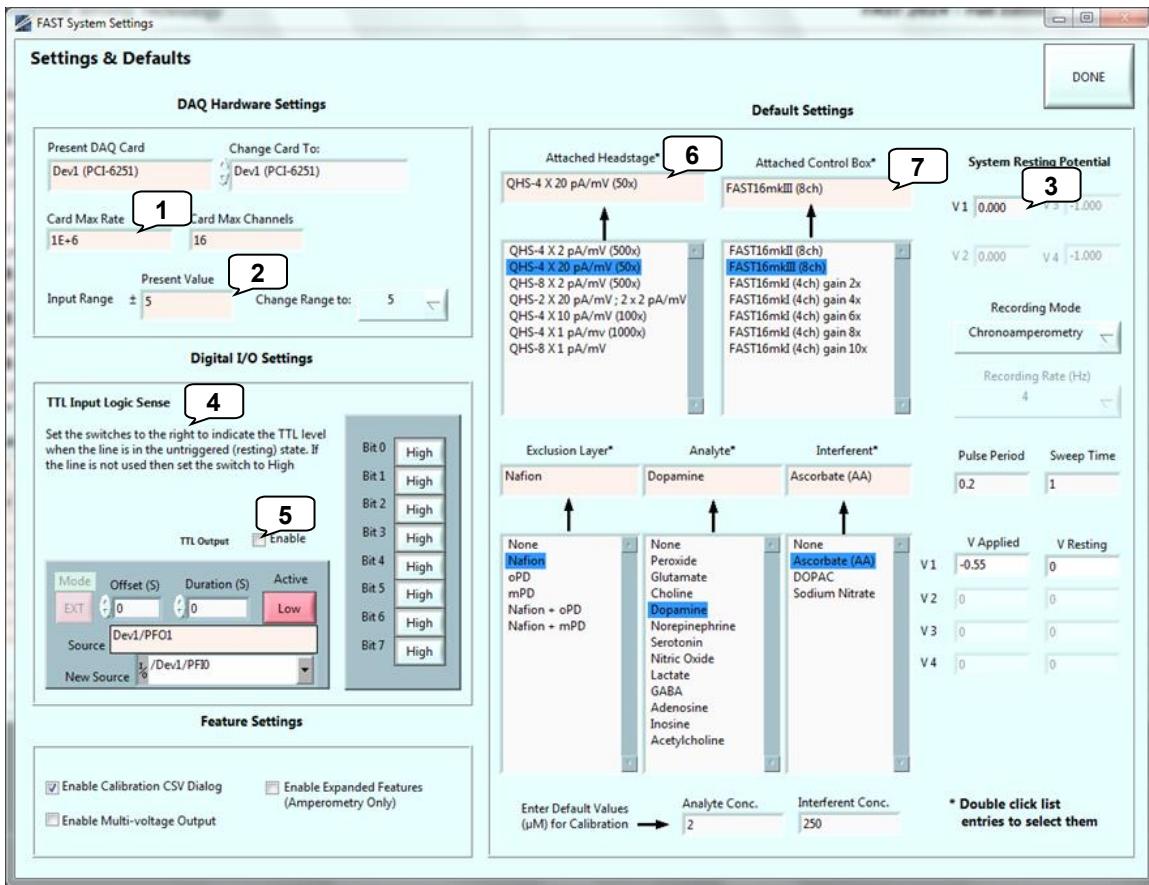
Carbon fibers



- In the main menu, under **Help**, choose **Chrono Settings** to display a diagram of the parameters used in chronoamperometry experiments.
- Pulse Period** - Total duration (in seconds) of the voltage pulse.
- Sweep Time** - Interval (in seconds) between the start of voltage pulses.

2.1 Chronoamperometry Settings & Defaults

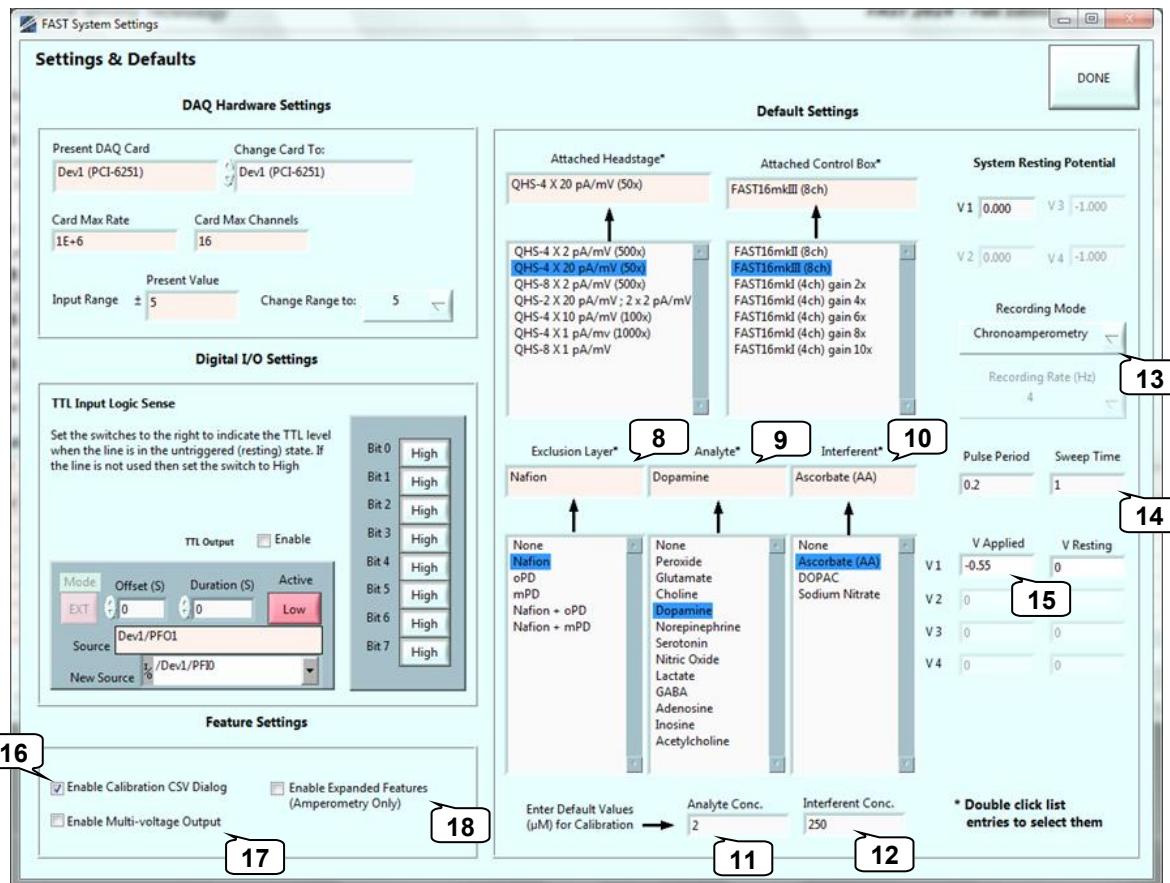
Carbon Fibers



- Card Max Rate** - Describes how often the [analog to digital conversion](#) occurs per channel.
- Input Range** - 0.1 to 10V. From drop-down menu, most often select 5 for best signal detection. Describes [analog to digital conversion range](#).
- System Resting Potential** - Maintains applied potential between recording episodes.
- TTL Input Logic Sense** - Sets the logic level on the digital inputs that indicate the untriggered state of attached equipment.
- TTL Output** - A tick in the box enables the output of a trigger signal that begins on the rising edge of the square wave pulse used in chronoamperometry. Used to synchronize external equipment such as data collection systems to the FAST system.
- Attached Headstage** - Select the headstage that is attached to the FAST control box.
- Attached Control Box** - Select appropriate control box. If selecting "FAST16mkI," match the gain to the one selected on the front panel of the FAST control box.

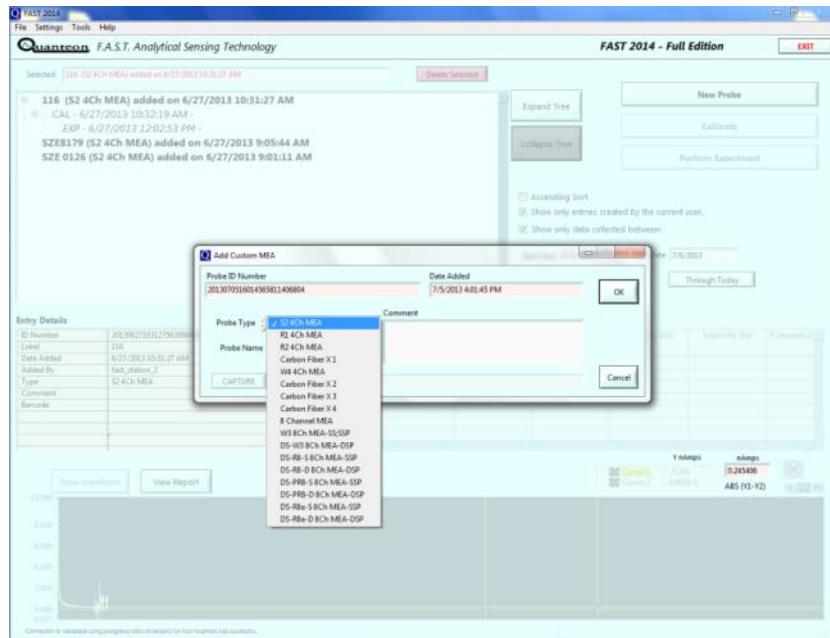
2.1 Chronoamperometry Settings & Defaults

Carbon Fibers

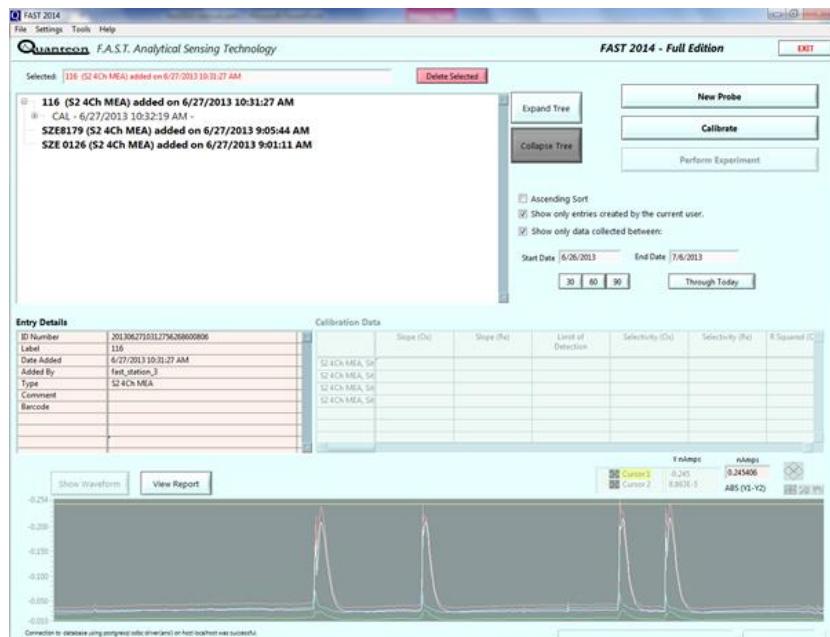


8. **Default Exclusion Layer** - Default exclusion layer, if one, for calibrations. (**Defaults:** double clicking a selection from the list sets/changes the default. The current default is shown in the text box above the list).
9. **Default Analyte** - Default analyte, if one, for calibrations.
10. **Default Interferent** - Default interferent, if one, for calibrations
11. **Analyte Concentration** - Default analyte concentration for calibrations.
12. **Interferent Concentration** - Default interferent concentration for calibrations.
13. **Recording Mode** - Carbon fibers are calibrated in **Chronoamperometry** mode.
14. **Pulse Period, Sweep Time** - Described in [Diagram of Chronoamperometry Settings](#).
15. **V applied** - Enter the potential to be applied vs. reference electrode.
16. **Enable Calibration CSV dialog** - Add calibration results to a new or existing spreadsheet.
17. **Enable Multi-voltage Output** - Permits use of multiple applied potentials (advance only).
18. **Enable Expanded Features** - Displays Custom MEA Calibration Setup window.

2.2 Recording with Chronoamperometry

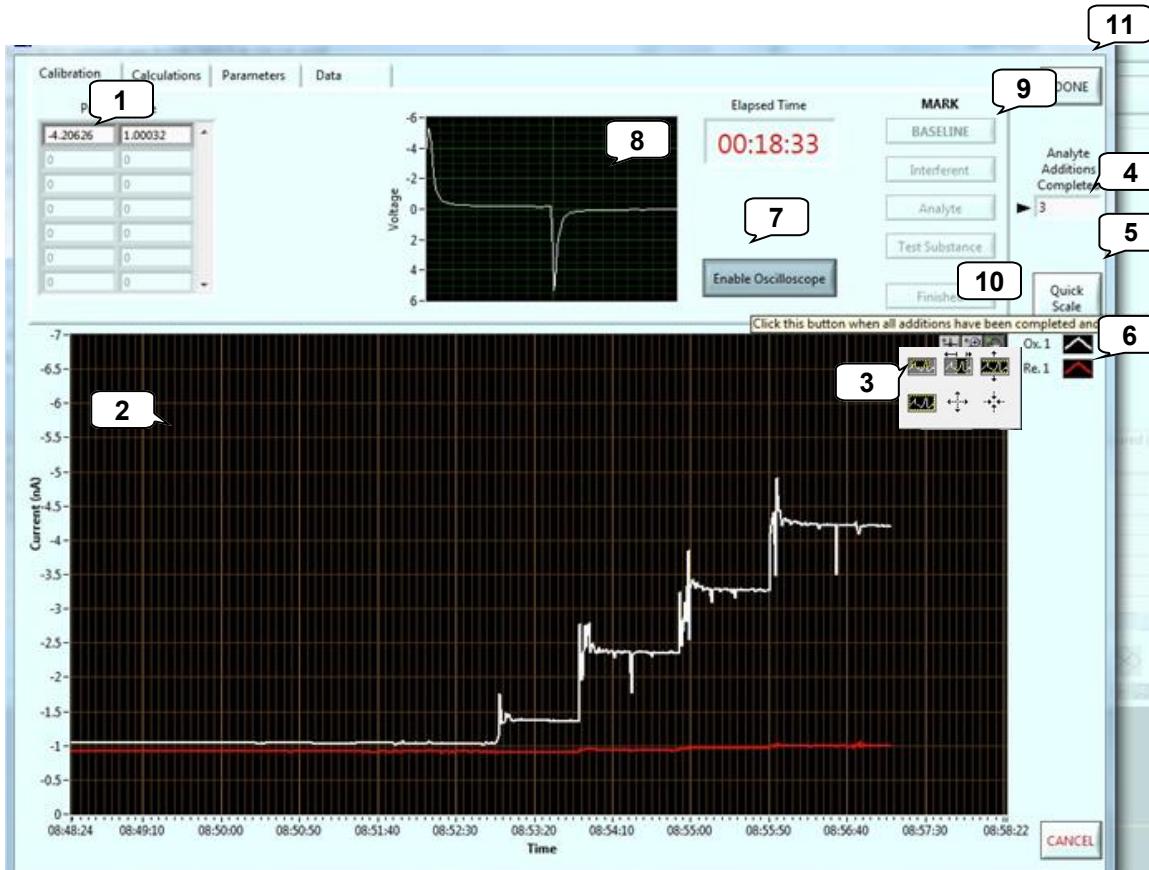


After verifying **Settings**, select **New Probe**. Choose **Probe Type** from the drop down menu (**Carbon Fiber X** *n* for *n* number of active channels), provide a **Probe Name**, and include additional **Comments**, as needed. The information will be displayed in the database list.



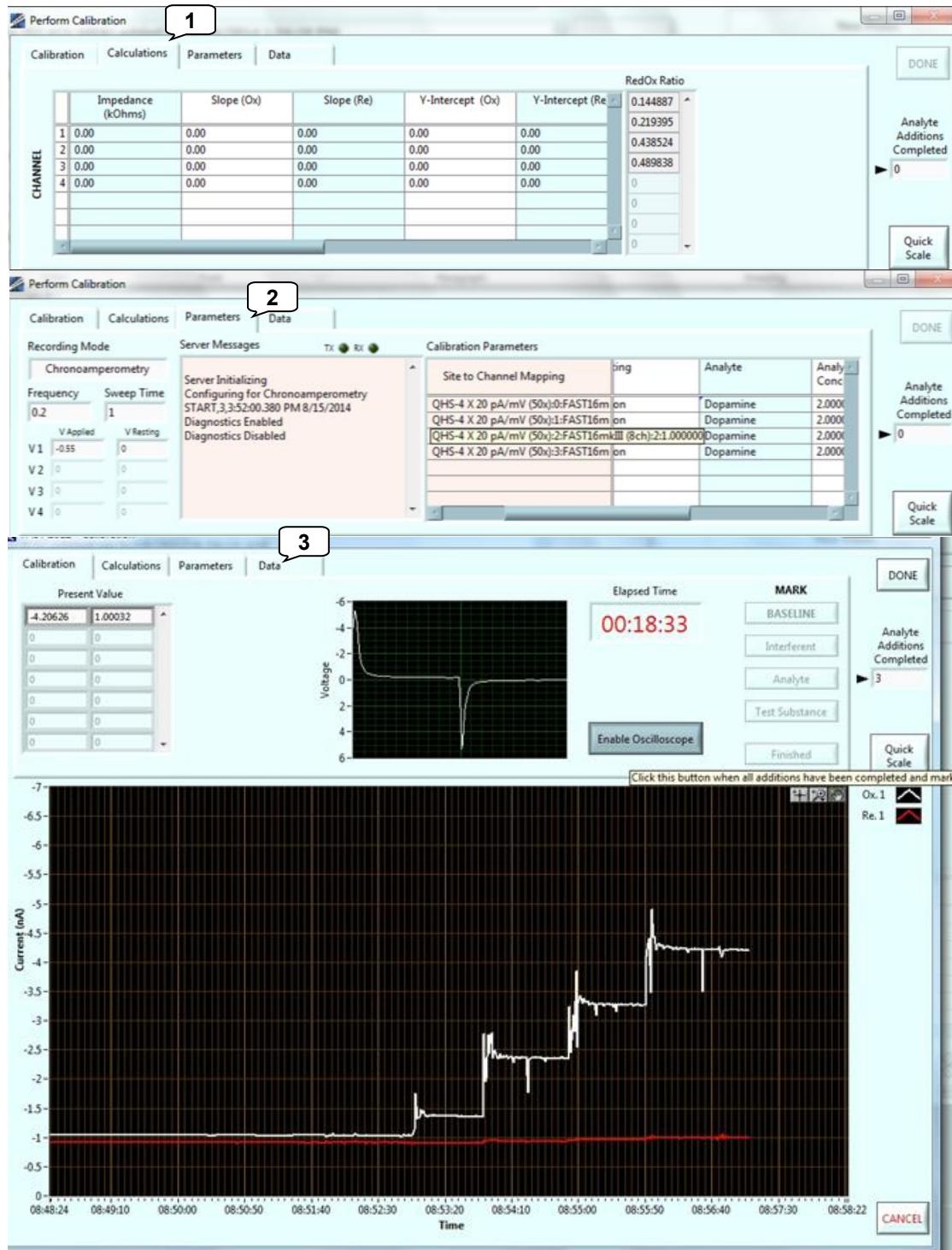
After creating an MEA entry, select the sensor of interest in the database and the **Calibrate** button will become active. Select to begin an [MEA calibration](#).

2.3 Chronoamperometry Calibrations



1. Current (nA) value of oxidation (left) and reduction (right) for each channel.
2. Graphical display of the current values of individual channels (nA vs. time of day).
3. Magnification and scrolling tools allow for customization of the graphical display.
4. Counter showing total number of additions for calibrations.
5. Re-scale to zoom on the active area of the graph.
6. Active microelectrode channel colors for oxidation and reduction —modifiable.
7. Click to toggle between displaying/turning off the oscilloscope.
8. Oscilloscope displays wave form or current level generated at microelectrode (in V).
9. Activate **BASELINE** button to begin calibration calculations. Buttons change to **Interferent** then **Analyte** and **Test Substance** based on calibration protocol.
10. Select **Finished** to end calibration calculation.
11. Select **DONE** to close calibration window, otherwise, to abort calibration select **CANCEL**.

2.3 Chronoamperometry Calibrations



1. **Calculations** - Calibration results (including LOD, selectivity, and slope).
2. **Parameters** - Displays system parameters used for calibration.
3. **Data** - Displays data used for calibration **Calculations**.

2.4 Chronoamperometry Experiments



After creating a calibration,

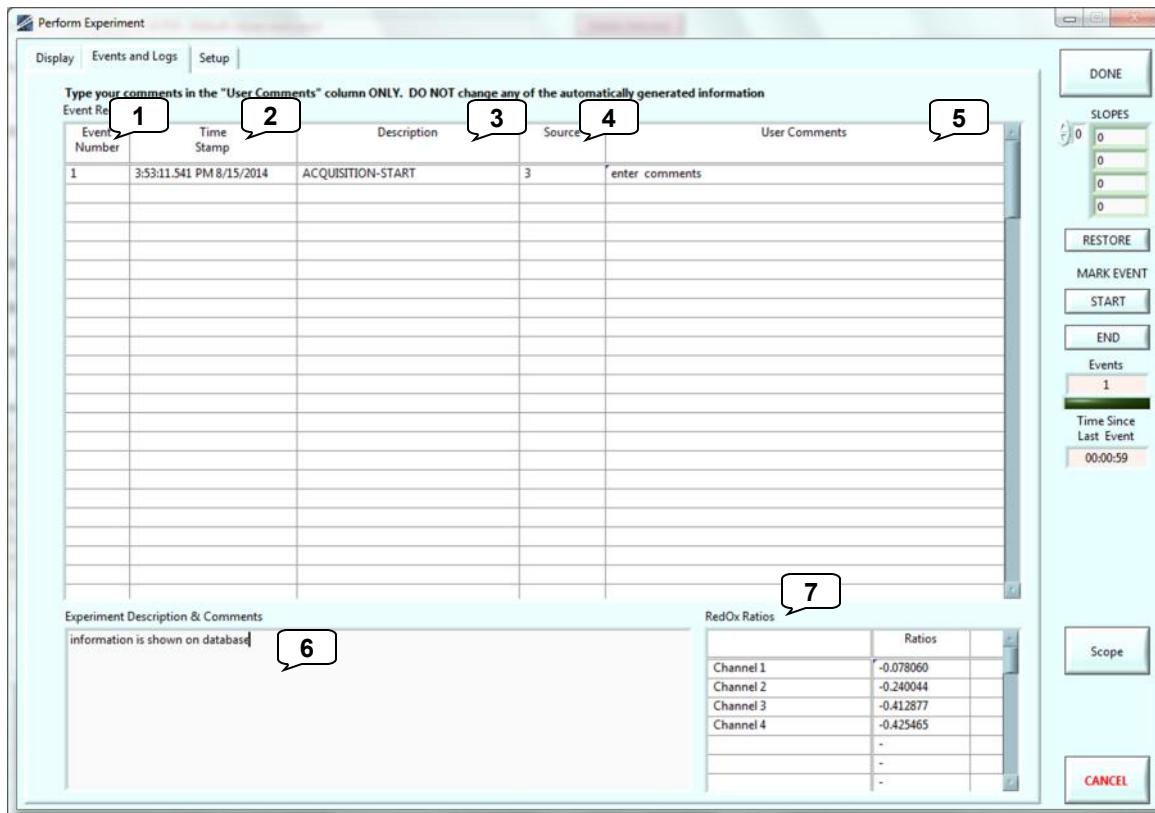
1. Select a calibration to use for an experiment by double-clicking on that calibration.
2. Parameters associated with the MEA and calibration and calibration calculations (LOD, selectivity, Slope) are displayed.
3. Select **PERFORM EXPERIMENT** to open acquisition window and begin an experiment.

2.4 Chronoamperometry Experiments



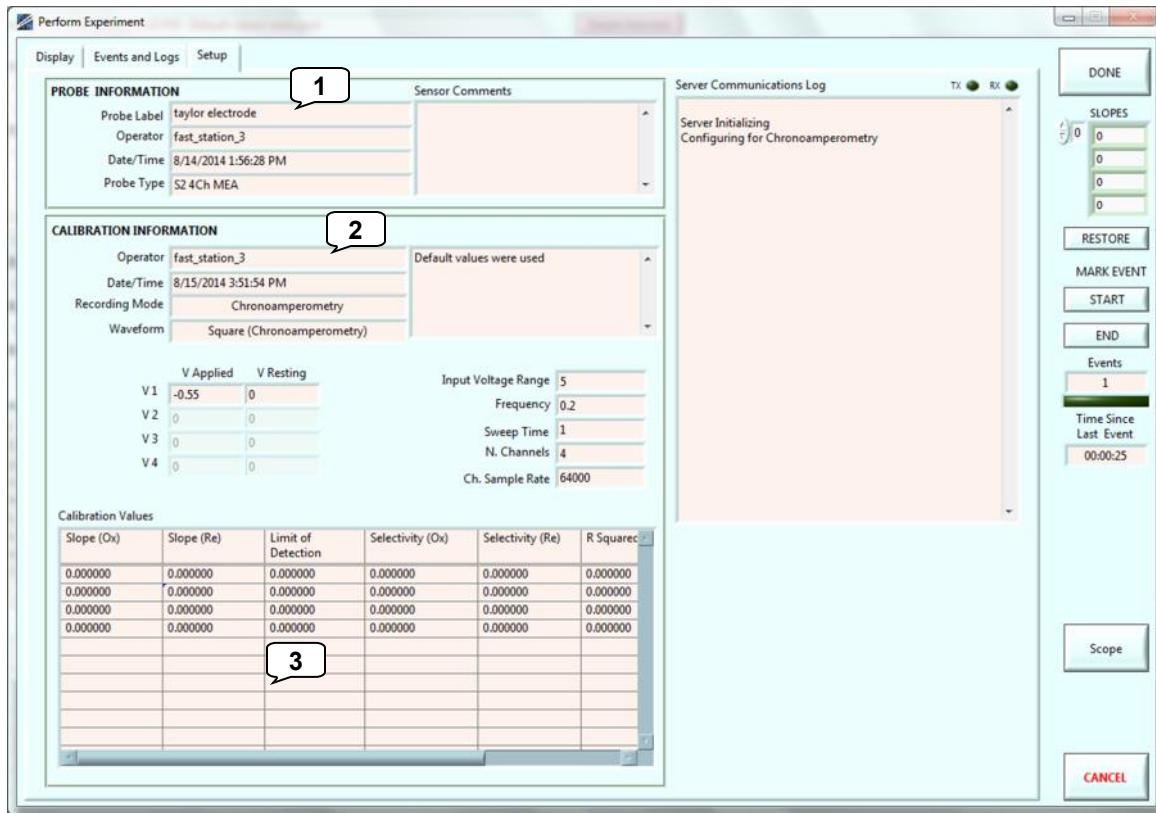
1. Start time stamp for acquisition data file, **total elapsed recording time**, and End time stamp for acquisition data file.
2. Graphical display of the current (nA or pA) or concentration (μM) values of individual MEA channels (vs. time of day).
3. Toggle between **Current** (nA or pA) and **Concentration** (μM) display for Y-Axis Units.
4. **V applied** - [Voltage applied](#).
5. Re-scale to zoom on the active area of the graph.
6. Active microelectrode channel colors for oxidation and reduction —modifiable.
7. Microelectrode **slopes** from calibration. These are automatically obtained from selected calibration file but can be manually entered as needed.
8. **RESTORE SLOPES** to original calibration file values for selected MEA.
9. Click to signal **START** and **END** of select triggered **EVENT**. Useful for long recording signals, drug, or behavior studies. Adds TTL event mark to data file for analysis.
10. **Events** - Counts number of triggered events and **Time Since Last TTL Event**.
11. **V applied** - Voltage applied, adjustable.
12. **Scope** - Display/Hide [oscilloscope](#).
13. Select **DONE** to close Experiment window, otherwise, to abort experiment select **CANCEL** (data not saved).

2.4 Chronoamperometry Experiments



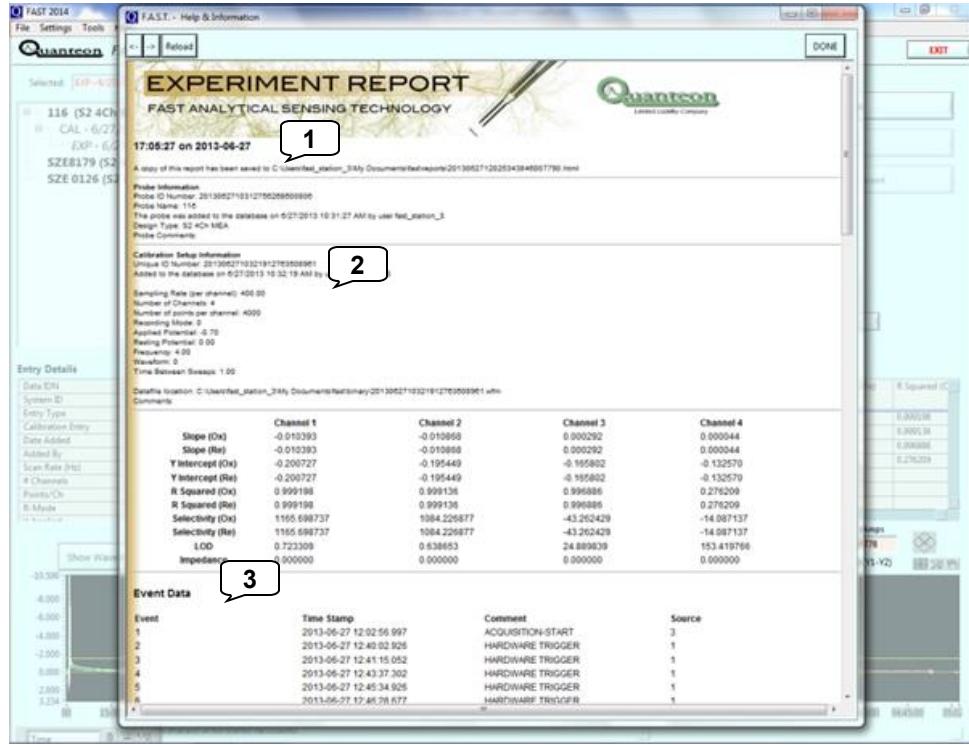
1. **Event Number** - indicates order of TTL events.
 2. **Time Stamp** – Time TTL was triggered (Time determined by computer clock).
 3. **Description** – Displays the source of the TTL input.
 4. **Source** – Displays which TTL input was triggered.
 5. **User Comments** - Text field for describing TTL events.
 6. **Experiment Description & Comments** - Text field for describing experiment. Viewable in **Browse Database**.
 7. **RedOx Ratio** – In chronoamperometry, ratio between reduction current and oxidation current. Used to identify molecules of interest. (~ 0.6 to 0.9: dopamine; ~0.1 to 0.3: serotonin)

2.4 Chronoamperometry Experiments



- 1. SENSOR INFORMATION** - Displays Microelectrode parameters.
- 2. CALIBRATION INFORMATION** - Calibration/experiment parameters for selected recording MEA.
- 3. CALIBRATION VALUES** - Displays MEA calibration calculations

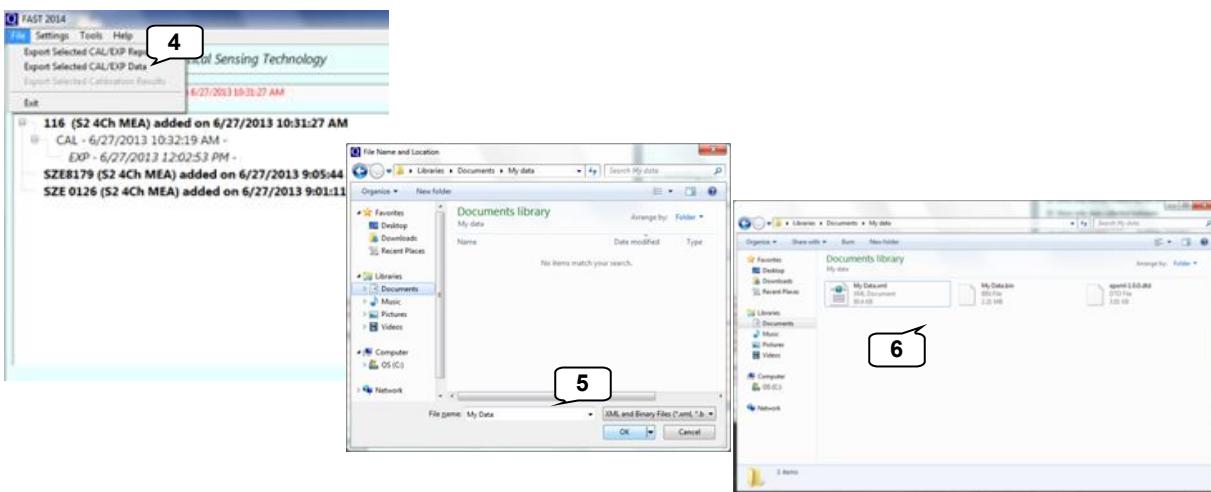
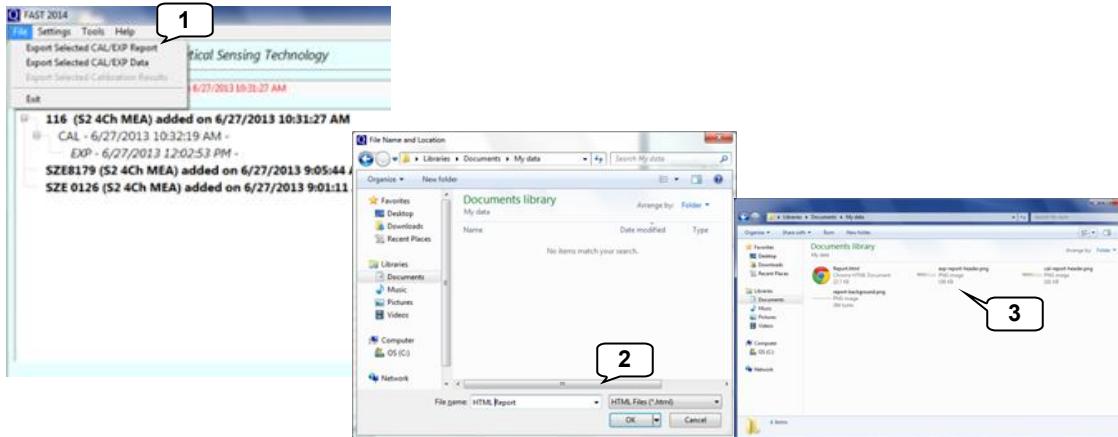
2.4 Chronoamperometry Experiments - Viewing reports



View Report from the Main Screen

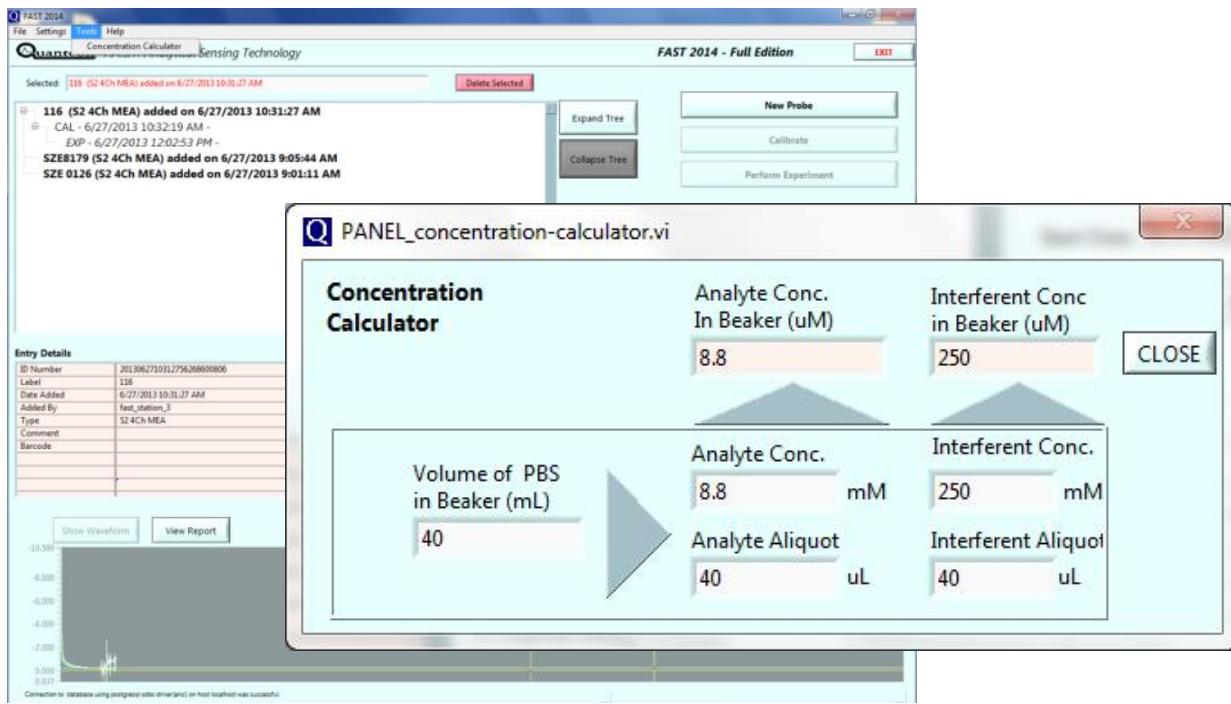
1. **Sensor Information** - [Information about microelectrode](#)
2. **Calibration Setup Information** - Calibration parameters and calculations.
3. **Event Data** - Lists **Event #**, **Time Stamp**, **Comments**, **Source** for the event.

2.4 Chronoamperometry Experiments - Exporting files



1. **Export Selected CAL/EXP Report** - Used to export a report. [Select entry on the home screen \(#5\)](#)
2. Provide a file name. Exported as .html
3. Exporting the report generates an .html file and header images.
4. **Export Selected CAL/EXP Data** - Export data files used in the [FAST Analysis](#) program a report. [Select entry on the home screen \(#5\)](#)
5. Provide a file name.
6. Exporting data files generates .xml, .bit, .dtd files that are all needed for analysis with [FAST Analysis](#).

3.1 Tools - Concentration calculator



1. **Concentration Calculator** - Enter volume used for calibration. Analyte and/or Interferent concentration. Analyte or Interferent aliquot volume and obtain final concentrations in the beaker.

3.2 Preparation of mPD Microelectrode Arrays

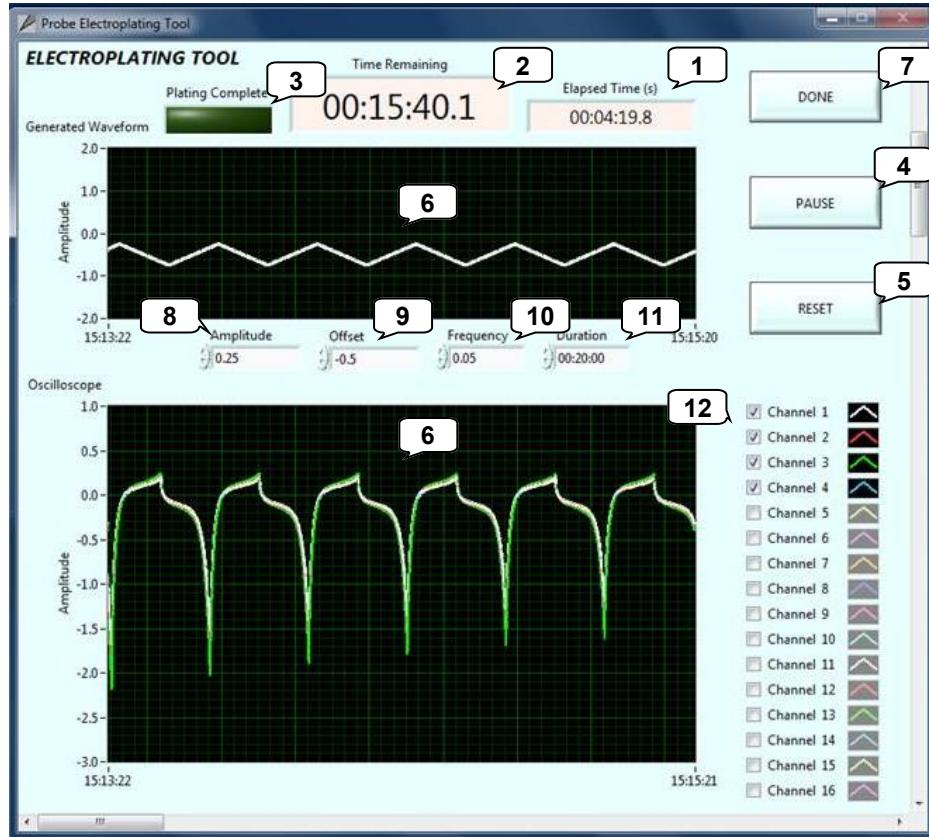
Quick guide to m-PD-plating

Acros Organics: m-Phenylenediamine dihydrochloride, 99%. CAS number: 541-69-5
Catalog number: 216650250 (for 25 g)

1. Bubble nitrogen gas through 125 ml of PBS for 20 minutes
 - This procedure is done to minimize oxidation of mPD by oxygen in the solution.
2. Weigh 0.0905 g of mPhenylenediamine (mPD)
 - a. Use mask and gloves
 - this substance is carcinogenic and easily oxidizes. The substance turns brown on oxidation and leaves ugly stains on fingers and other surfaces.
3. Mix solution on a stirplate in room temperature until it is completely dissolved.
 - a. Do not shake the container because that will re-introduce oxygen into the solution.
4. Place MEA in the headstage (Use either the 500X headstage or the 50X).
5. Pour ~40 ml of mPD solution in a beaker and place a reference electrode in the beaker. The reference used for mPD-plating should be used for this purpose only. Do not use your everyday, BSA reference electrode for this.
6. Verify connections by running calibration and observing exponential-shaped decay.
7. Software: Launch Electroplater  and run program

Calibrate mPD coated MEA per established protocol

3.3 Tools - Electroplating Tool



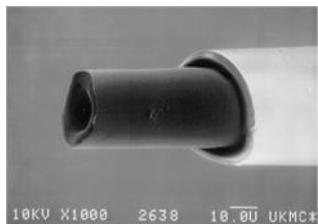
To begin electroplating, launch the electroplating tool program  on the desktop.

1. **Elapsed Time** - Elapsed time (in sec) of running electroplating tool.
2. **Plating Complete** - When electroplating is finished, **green light** turns on.
3. **Time Remaining** - Time left to finish electroplating.
4. **PAUSE** - Unselect to begin electroplating. Select to pause electroplating.
5. **RESET** - Return values to default.
6. **Graphical Display** - Displays electroplating output wave pulse and oscilloscope view.
7. **Done** - Exit Electroplating Tool program.
8. **Peak Amplitude** - Amplitude of the applied voltage pulse.
9. **Offset** - Deviation from zero of the applied voltage pulse.
10. **Frequency** - Number of cycles per second.
11. **Duration** - Length of time the electroplating tool will be applied.
12. Click to display/hide the traces

[Standard protocol](#) for electroplating meta-Phenylenediamine on MEAs is **Duration** : 20 minutes, **Frequency** : 0.05, **Offset** : -0.5, **Peak Amplitude** : 0.25.

4.1 Calibrating Microelectrodes

Monoamines



- | | |
|-----------------------|------------------------|
| Dopamine calibrations | Serotonin calibrations |
| 1. Dopamine | 1. Serotonin |
| 2. PBS | 2. PBS |
| 3. DOPAC | 3. AA |

Accessories

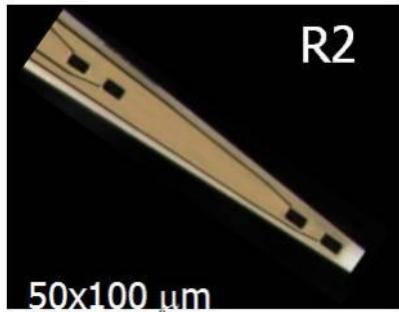
Norepinephrine calibrations

1. Norepinephrine
2. PBS
3. AA

1. Carbon Fiber (Nafion® coated)*

2. One reference electrode (the tip is stored in 3M NaCl at least 24 hours before first use)
3. Battery powered stirrer
4. 50 mL Beaker
5. Magnetic stir bar

Enzyme-based MEAs



Accessories

1. Enzyme-coated MEA (Nafion® coated or mPD plated)*
2. One reference electrode (the tip is stored in 3M NaCl at least 24 hours before first use)
3. Battery powered stirrer
4. 50 mL Beaker
5. Magnetic stir bar

H₂O₂ calibrations

1. Peroxide
2. PBS

Choline calibrations

1. Choline
2. PBS
3. AA
4. Peroxide

Glutamate calibrations

1. Glutamate
2. PBS
3. AA
4. Peroxide

Adenosine calibrations

1. Adenosine
2. Inosine
3. Xanthine
4. PBS
5. AA
6. Peroxide

ACh calibrations

1. ACh
2. PBS
3. AA
4. Choline
5. Peroxide

* Carbon fibers and Microelectrode arrays may be purchased through the [Center for Microelectrode Technology](#)

4.2 Calibration Protocol

Soak microelectrodes for 30 min – 45 min in RT [PBS lite](#) or warmed (35-37°C) [PBS lite](#) for enzyme-based MEAs

Prepare [calibration solutions](#)

Connect sensor to hardware and insert tip into 40 ml of stirred PBS lite

Connect the reference electrode

Begin [amperometry](#) or chronoamperometry calibrations

Allow 5-10 min equilibration; begin once the baseline has stabilized

Mark **BASELINE**

Add 500 ul interferent (stock solutions of: 20 mM AA or 20 mM DOPAC)

Allow ~ 30 sec -1 min between additions/until traces reach new steady-state.

Mark **INTERFERENT**

Add 40 ul of analyte (stock solutions of: 20 mM glutamate or 2 mM DA or other analyte of interest)

Mark 1st addition **ANALYTE**

Add 40 ul analyte

Mark 2nd addition **ANALYTE**

Add 40 ul analyte

Mark 3rd addition **ANALYTE** (Note that 3 additions are standard – but one point calibration could be used)

May select **FINISHED** or may continue with test substances, as necessary.

For enzyme-based MEA, add 40 ul of stock solution of 2mM DA

DO NOT MARK ANALYTE – click on **TEST SUBSTANCE**

Add 40 ul of stock solution of 8.8mM H₂O₂

DO NOT MARK ANALYTE – click on **TEST SUBSTANCE**

Click the **FINISHED** button

After calibration, store microelectrode in PBS lite until use.

4.3 Calibration Solutions

“PBS Lite”

0.05 M Phosphate Buffered Saline

2 Liter recipe

1. Transfer ~1800 mL of deionized, filtered H₂O to a 2L glass container
2. Add, while mixing with a stir bar:

CHEMICAL	F.W.	AMOUNT (G)
SODIUM PHOSPHATE (NaH ₂ PO ₄), <u>MONOBASIC, MONOHYDRATE</u>	137.9	2.8
SODIUM PHOSPHATE (Na ₂ HPO ₄), <u>DIBASIC, ANHYDROUS</u>	141.9	11.34
SODIUM CHLORIDE (NaCl)	58.44	11.68

3. Stir until all chemicals are dissolved. Transfer to a 2L volumetric flask and add dH₂O to bring to volume.
4. Filter through 0.2 µm filter.
5. Test pH, pH ~ 7.2—7.4.

4.3 Common Calibration Solutions

Ascorbic Acid 20mM: (500 µl in 40 ml = 250 µM)

20 mM: 0.18 g of AA in 50 mL of DI H₂O

Make fresh daily.

Acetyl choline 20 mM ACh (40 µl in 40 ml = 20 µM)

20 mM: 0.18 g of ACh in 50 mL of DI H₂O

Make fresh daily.

Adenosine 1 mM: (40 µl in 40 ml = 1µM)

1 mM: 0.13 mg of adenosine in 1 mL of DI H₂O

Make fresh every week

Choline 20mM: (40 µl in 40 ml = 20µM)

20 mM: 0.14 g of choline in 50 mL of DI H₂O

Make fresh every week

Dopamine 2mM: (40 µl in 40 ml = 2 µM)

2 mM: 0.038 g of DA + 10 mL of 0.1M perchloric acid.

Bring volume to 100 mL with DI H₂O.

Make fresh once per month.

DOPAC 20mM: (500 µl in 40 ml = 250 µM)

20 mM: 0.34 g of DOPAC + 1 mL of 0.1M perchloric acid.

Bring volume to 100 mL with DI H₂O.

Make fresh once per month.

L-Glutamate 20 mM: (40 µl in 40 ml = 20 µM)

20 mM: 0.15 g of glutamate in 50 mL of DI H₂O

Make fresh weekly.

H₂O₂ 8.8 mM: (40 µl in 40 ml = 8.8 µM)

8.8 mM 500 µL of H₂O₂ (3%) in 50 mL of DI H₂O

Make fresh every week.

Cont.

4.3 Common Calibration Solutions

Inosine 1 mM: (40 µl in 40 ml = 1µM)

1 mM: 0.13 mg of inosine in 1 mL of DI H₂O

Make fresh every week

Norepinephrine 2 mM: (40 µl in 40 ml = 2 µM)

2 mM: 0.064 g of NE + 10 mL of 0.1M perchloric acid.

Bring volume to 100 mL with DI H₂O.

Make fresh once per month.

Serotonin 2 mM: (40 µl in 40 ml = 2 µM)

2 mM: 0.043 g of 5-HT + 10 mL of 0.1M perchloric acid.

Bring volume to 100 mL with DI H₂O.

Make fresh once per month.

Xanthine 1 mM: (40 µl in 40 ml = 1 µM)

1 mM: 0.15 mg of xanthine in 1 mL of 0.1N NaOH

Make fresh every week

4.4 Analyte ordering information

Description	Distributor	Catalog #	Units
Acetylcholine chloride	Sigma-Aldrich http://www.sigmaaldrich.com	A6625-25 G	25 g
Adenosine	Sigma-Aldrich http://www.sigmaaldrich.com	A9251-5G	5 g
Ascorbic Acid	Sigma-Aldrich http://www.sigmaaldrich.com	A5960-100G	100 g
Choline Chloride	Sigma-Aldrich http://www.sigmaaldrich.com	C1879	500 g
DOPAC	Sigma-Aldrich http://www.sigmaaldrich.com	850217	1 g
Dopamine HCl	Sigma-Aldrich http://www.sigmaaldrich.com	H8502	10 g
Hydrogen Peroxide	Available from local drug store	3 % H ₂ O ₂ solution.	1
Inosine	Sigma-Aldrich http://www.sigmaaldrich.com	I4125-10G	10 g
L-Glutamic acid salt (MSG)	Sigma-Aldrich http://www.sigmaaldrich.com	G-1626	100 g
Norepinephrine bitartrate	Sigma-Aldrich http://www.sigmaaldrich.com	A0937	1g
Serotonin (5-HT)	Sigma-Aldrich http://www.sigmaaldrich.com	H9523-100MG	100 mg
Xanthine	Sigma-Aldrich http://www.sigmaaldrich.com	X7375-10G	10 g

^a Updated March 2010. Information not warranted.

Description	Distributor	Catalog #
Heating & circulating water pump ^a	e.g. Cincinnati Sub-Zero http://www.cszmedical.com	Micro-Temp LT
Magnetic Stirrer ^b (battery operated)	Cole-Parmer http://www.coleparmer.com	EW-04804-01
Reference electrodes	BAS http://www.basinc.com/	MF-2052/MF-2079
Stir Bar	Fisher Scientific http://www.fishersci.com	14-511-92

Common optional/additional accessories

Dip socket	Newark Electronics http://www.newark.com	04M1727 (Mill-Max #210-43-308-41-001000)
Quick Connect for calibration bath	Colder.com http://www.colder.com/	e.g. APCD17006SH ; APCD22006
Glass pipette (o.d. 1 mm, i.d. 0.58 mm, 6")	A-M Systems http://www.a-msystems.com	601500
Gold pin	Allied Electronics Allied Electronics	Mfr. Part #: 220-P02-100
Kopf Model 900 Stereotax ^d	http://www.kopfinstruments.com/Stereotaxic/900.htm	
Meiji Techno EMZ Stereomicroscope ^c	http://www.meijitechno.com/emz.htm	EMZ-5 with SWF10x eyepiece
Reticle		10mm divided into 100 units
Microdrive for Kopf stereotax	Narishige http://products.narishige-group.com/english.html	MO-10 Single-Axis Oil Hydraulic Micromanipulator
Picospritzer III ^e	http://www.parker.com	Picospritzer III
Tubing for micropipette	www.vwr.com	63010-232
Pipette puller	David Kopf instruments http://www.kopfinstruments.com/	730
Silver wire (200µm)	A-M Systems http://www.a-msystems.com	7826500
Sticky Wax	Kerrlab http://www.kerrlab.com/index/kerrlab	00625

^a Used primarily for enzyme-based MEA calibrations.

^b Battery powered stir plates eliminate potential 60 Hz noise that could interfere with calibrations.

^c For craniotomy surgeries and probe placement and monitoring of micropipette pressure ejection of fluids. (10x eyepieces, 0.5x objective), 0.7 – 3.0+ zoom range is very important. Select one with the telescoping arm or boom that has the needed magnification.

^d Holder: Model 1772 Universal Holder places syringes, large diameter electrodes or drill handpieces stereotactically. Holds devices from 2.8 mm to 11 mm diameter.

^e For pressure ejections of solutions *in vivo*. One channel system is fine for most applications. 2 channel is useful if when ejecting multiple agents simultaneously using double-barreled pipettes.

5.1 Glossary

Analog to Digital Conversion (ADC) - In data acquisition, the conversion of analog measurements to digital numbers that the computer can store and use in calculations.

Analog to Digital Conversion Range - Refers to the minimum and maximum values that the analog to digital conversion can span. A fixed number of points span this range and the smaller the range, the greater the resolution of the signal.

Gain - Amplification of a signal.

LUT - Look Up Table file contains FAST system information updates regarding new analytes, coatings, supported DAQ cards, etc. Distributed as a lut-data.ini file. To update, replace the old lut-data.ini file in the Quanteon folder located in the Program Files of the main hard drive (usually the C drive) with this file. Next, go into Manage Database in the FAST software and select, Update Lookup Tables.

Oscilloscope displays wave form or current level generated at microelectrode (V, current converted to V for display).

5.2 Troubleshooting

Problem: I cannot get any waveforms during calibration or electrode verification.

Possible Solutions: Power to the system interface is off. Electrodes are not connected to the headstage. Signal amplification is not high enough for an electrode. The Analog-to-Digital cable has been jarred loose or the board is not connected. Tip of working electrode is not in solution. Potential ground loop in the system. Perform resistor test. Verify the applied voltage between **REF** and **GND/ISO** on the headstage.

Problem: The signal is noisy.

Possible Solution: A/C interference from heating pads or lights. Poor grounding of computer. Bad reference electrode. Damaged or bad working electrode.

Problem: The baseline in Acquisition or the waveforms in Calibration continue to fall.

Possible Solution: Bad reference electrode. Working electrode is overcoated with Nafion® or mPD. The working electrode may need a few more minutes to stabilize.

5.3 Frequently Asked Questions

How is LOD calculated?

LOD is calculated as $3 \times \text{SD}$ of 10 points of the baseline (the baseline SD in the html report)/slope. The units are in μM .

How is selectivity calculated?

Selectivity is ratio of analyte response divided by the interferent response for equal concentrations. So for an interferent addition of $250 \mu\text{M}$ the equation would be $(\text{slope} * 250) / \text{response to interferent in nA}$.

How is slope calculated?

The slope is the sensitivity of the MEA to a given analyte. The calculation is the slope (m) for a first order linear regression. The units are in $\text{nA}/\mu\text{M}$.

What is the y intercept?

The Y-intercept is a parameter that can be calculate, but we have not found utility in it.

What concentration of analyte should I be able to reliably detect if the MEA has a LOD of ~0.47-0.48?

The LOD is in μM and so $>0.5 \mu\text{M}$ concentration of analyte should be reliably detectable. However, in the brain, we usually measure a quieter signal in the absence of fluid motion like in a beaker.

What is the downside of sampling every 0.025 seconds?

Large data files and a noisier recording because the signal is not averaged over 1-2 Hz. The process of converting the analyte to current takes $\sim 800\text{ms}$, so signals faster than that may not be real. However, it appears that one can also measure field potential-like signals at the faster sampling rates. Zhang H, Lin S-C and Nicollelis MAL. J. Neurosci Methods 179: 191-200, 2009.

For further reading:

Gerhardt GA and Burmeister, J. *In vivo Voltammetry for Chemical Analysis of the Nervous System*. In: Encyclopedia of Analytical Chemistry: Instrumentation and Applications, R.A. Meyers, (Ed.), John Wiley & Sons Ltd, Chichester, 2000, pp. 710-731

[Hascup KN, Rutherford EC, Quintero JE, Day BK, Nickell JR, Pomerleau F, Huettl P, Burmeister JJ and Gerhardt GA.](#) Second-by-Second Measures of L-Glutamate and Other Neurotransmitters Using Enzyme-Based Microelectrode Arrays, In: [Electrochemical Methods for Neuroscience](#). Adrian Michael, (Ed), Taylor & Francis Group, LLC, 2007.

For further technical assistance, please contact: support@quanteon.cc

6.0 Appendix

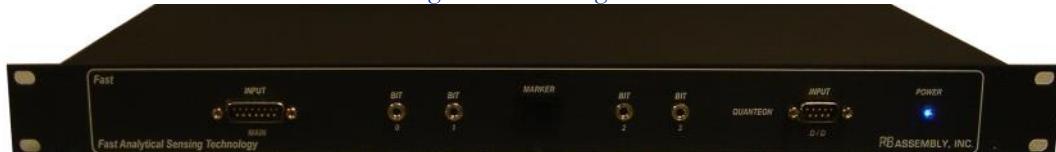
FAST control box

FAST16mkIII rear panel

Bit 0-3

Connection jack for TTL and external device control.

High to Low voltage device.



Main
Pre-amplifier (Headstage)
connection.

Marker
Button for TTL and external
activation of device such as
pressure ejection unit.

Input D/D
Input for 9 additional
TTL and external
device control.
High to Low voltage
devices.

FAST16mkIII rear panel



Ground
Connection jack for
external ground
source. Use only if
needed.

A/D Computer
Connection port for
68 pin cable to A/D
card in computer.

DC Input
Connection jack for 5V DC, 2.0 Amp
external power supply.

I/O

On/Off switch. It is recommended to leave FAST16 unit on at all times to assure optimum operating performance of power supply and amplifiers. Upon initial power up, FAST16 system should reach optimum stability within 1 hour but a slight change in system performance may occur for several hours. This is normal for power supplies and amplifier circuits. In most cases these fluctuations will not affect recording function.

6.0 Appendix

Quick FAST System Setup



1. Setup the all-in-one computer with mouse and keyboard
2. Identify the A/D card and cables



3. Connect one end of the USB to the A/D card. The other end to the computer



OR



4. Connect one end of the 68pin cable to the A/D card. The other end to the back of the FAST system.



5. Power the FAST system with either the AC power or the rechargeable battery

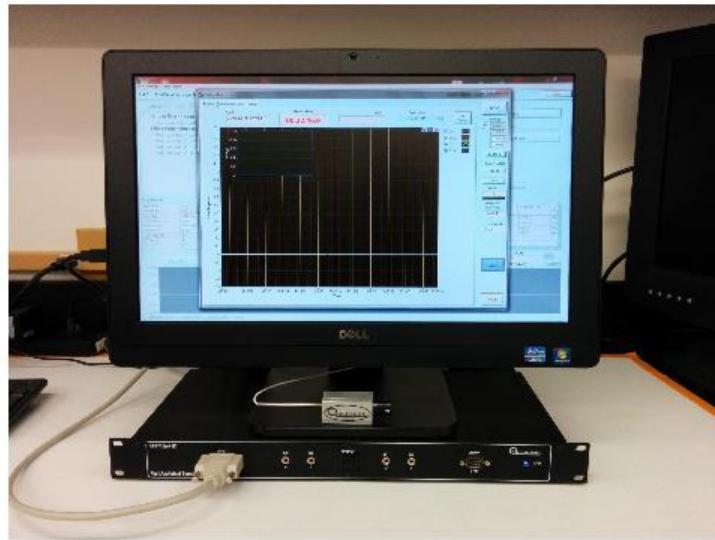
6. Use the 5V AC power supply to power the FAST system. NOT THE 12V for the rechargeable battery

6.0 Appendix

Quick FAST system setup



7. Finish by connecting the cable to the FAST control box, connecting the headstage to the cable, attaching an alligator clip to REF and a MEA connector to the headstage (if applicable)



The rechargeable battery can be recharged by disconnecting from the FAST system, plugging the battery to the 12 V power supply, and connecting to a wall outlet.