

Standard Operating Procedure for:

Dionex ICS-1000 Ion Chromatography System
with IonPac AS14A Exchange Column
(IC R03.doc)

Missouri State University

and

Ozarks Environmental and Water
Resources Institute (OEWRi)

Prepared by: _____ Date: _____
OEWRi Quality Assurance Coordinator

Approved by: _____ Date: _____
OEWRi Director



Table of Contents

1	Identification of the method.....	3
2	Applicable matrix or matrices.....	3
3	Detection Limit	3
4	Scope of the method	3
5	Summary of method.....	3
6	Definitions.....	4
7	Interferences.....	6
8	Health and safety.....	6
9	Personnel qualifications	6
10	Equipment and supplies	7
11	Reagents and standards	7
13	Quality control	8
14	Calibration and standardization	9
15	Procedure	15
16	Data acquisition, calculations, and reporting.....	24
17	Computer hardware and software.....	26
18	Method performance	26
19	Pollution prevention.....	27
20	Data assessment and acceptable criteria for quality control measures.....	27
21	Corrective actions for out-of-control or unacceptable data	27
22	Waste management.....	28
23	References	28
24	Tables, diagrams and flowcharts.....	22

1 Identification of the method

Use of the Dionex ICS-1000 Ion Chromatography System with IonPac AS14A exchange column with Standard Methods for the Examination of Water and Waste Water Method 4110B.

2 Applicable matrix or matrices

This instrument can be used for natural water, drinking water, industrial water pharmaceutical, and polymer samples.

The US Geological Survey collected anion data in 2001 from samples collected at the gage on Pearson Creek near Springfield, Missouri (site 07050690), (USGS, 2002). This data is typical of what is found in Ozark natural waters. The average concentrations found from those samples are illustrated in Table 1 below. Bromide data was not determined.

Anion	Concentration (mg/l)
Fluoride (n=2)	<0.2
Chloride (n=2)	19.0
Sulfate (n=2)	9.8
Nitrite + Nitrate (n=4)	2.18
Orthophosphate (n=4)	0.021

3 Detection Limit

The method detection limit for fluoride is 2.0µL, chloride is 4.0µL, nitrite is 3.7µL, bromide is 14.0µL, nitrate is 2.7µL, orthophosphate is 14.0µL, and sulfate is 18µL. Analysis requires 17 minutes per sample. The sample volume is 5.0ml. The sample loop is 25µL. The maximum operating pressure is 5000psi (35MPa).

4 Scope of the method

This standard operating procedure provides Missouri State University (MSU) laboratory personnel with guidance on the procedure for operation of the Dionex ICS-1000 Ion Chromatography System with IonPac AS14A exchange column.

5 Summary of method

The sample is automatically loaded into the eluent stream and the pump pushes the eluent and sample through the guard and separator columns. Ion exchange is the mode of separation and is based on the premise that different sample ions migrate through the separator column at different rates due to interactions with ion exchange sites located throughout the column. Then the eluent and sample ions enter a suppressor that selectively enhances ion detection and suppresses the conductivity of the eluent. A conductivity cell measures the electrical conductance of the sample ions as they emerge from the suppressor and produces a signal based on a chemical or physical property of the analyte.

The conductivity cell transmits the signal to a data collection system. The data collection system identifies the ions based on retention time and quantifies each analyte by integrating the peak area or peak height. The data is quantified by comparing the sample peaks in a chromatogram to those produced from a standard solution. The results are displayed as a chromatogram and the concentrations of ionic analytes is automatically determined and tabulated.

6 Definitions

- 6.1 Analytical batch: The set of samples processed at the same time.
- 6.2 Calibration standard: A solution prepared from the primary dilution standard solution or stock standard solutions. The calibration standards are used to calibrate the instrument response with respect to analyte concentration.
- 6.3 Chain of Custody (COC): Used to describe the written record of the collection, possession and handling of samples. Chain of custody forms should be completed as described in the Chain of Custody SOP # 1030R01. Chain of custody (COC) forms are located on a board in Temple Hall 125.
- 6.4 Chromeleon Browser: The portion of the software that allows data management, storage, and retrieval. The left window section illustrates the tree of file and folders. The right window section illustrates detailed information about the currently selected item. See Figure 1.

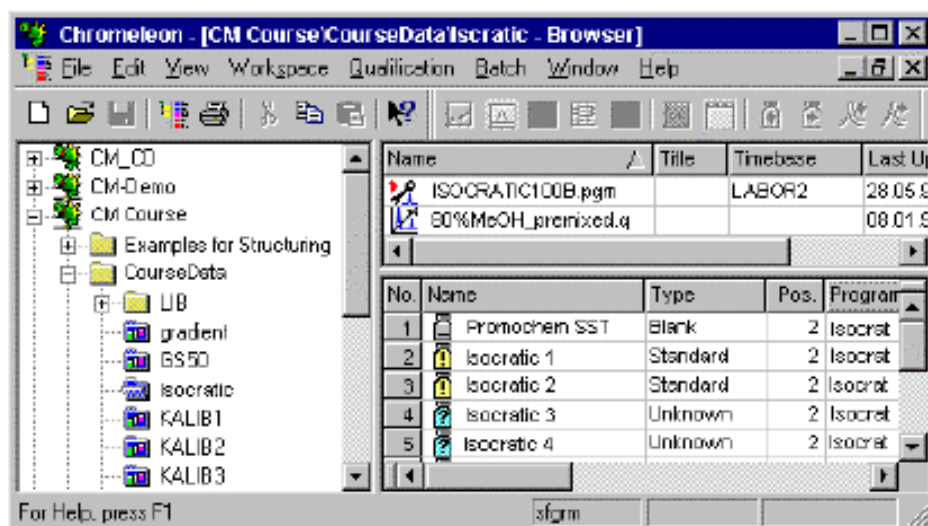


Figure 1. Chromeleon Browser

- 6.5 Conductivity: A measure of the ease with which electrical current flows through a liquid contained between two oppositely charged electrodes.
- 6.6 Control Panel: The portion of the software that controls the ICS-1000.
- 6.7 Eluent: A liquid that transports the sample through the ion chromatography system and a solvent that assists with the separation of ions in the sample.
- 6.8 Field blank (FB) : An aliquot of deionized water treated as a sample in all aspects, including exposure to a sample bottle holding time, preservatives, and all pre-analysis treatments. The purpose is to determine if the field or sample transporting procedures and environments have contaminated the sample.

- 6.9 Field duplicate (FD): Two samples taken at the same time and placed under identical circumstances which are treated identically throughout field and laboratory procedures. Analysis of field duplicates indicates the precision associated with sample collection, preservation, and storage, as well as with laboratory procedures.
- 6.10 Guard Column: A small column that prevents poisoning of the separator column by sorbing organic contaminants and removing particulates.
- 6.11 Instrument calibration: Reference standards with known values are selected based on the range of concentrations expected. The reference standards are measured with the instrument and a functional relationship is established between values of the standards and the corresponding measurements. A commercial standard is diluted and used to perform a 3-point instrument calibration with each batch. The conductivity cell is calibrated every 6 months using a 1.0 mM KCl solution.
- 6.12 Instrument drift: Gradual change in some operating characteristic of a circuit, tube, or other electronic device in a laboratory instrument. This can be measured during a brief period as an effect of warming up or during a long period as an effect of continued use.
- 6.13 Laboratory duplicate (LD): Two aliquots of the same environmental sample treated identically throughout a laboratory analytical procedure. Analysis of laboratory duplicates indicates precision associated with laboratory procedures but not with sample collection, preservation, or storage procedures.
- 6.14 Laboratory reagent blank (LRB): An aliquot of Type 1 water that is used to calibrate the instrument at the beginning of the analyses and is used within the batch analysis sequence for QA/QC. The LRB's purpose is to determine if analytes or interferences are present in the laboratory environment or the apparatus.
- 6.15 Method detection limit (MDL): The lowest level at which an analyte can be detected with 99 percent confidence that the analyte concentration is greater than zero.
- To calculate the MDL:
 - Prepare triplicates of two water samples with low concentrations of non-purgeable organic carbon. The laboratory director or supervisor will choose appropriate samples to use to determine MDL.
 - Analyze all samples.
 - Include all sample processing steps in the determination.
 - Calculate the standard deviation (s).
 - From a table of the one-sided t distribution select the value of t for $7 - 1 = 6$ degrees of freedom at the 99% level. This value is 3.14
 - The product 3.14 times s is the desired MDL.

- 6.13 Minimum Quantification Interval: The lowest level that can be quantitated accurately and is generally defined as four times the method detection limit = 4(MDL).
- 6.14 Relative Percent Difference (RPD): calculated as the difference between a sample and duplicate results, divided by the average of the sample and duplicate results, multiplied by 100%.
- 6.55 Separator Column: The column used to perform a chromatographic separation.
- 6.55 Suppressor: A device used to minimize eluent conductivity and convert sample species to a common form which increases detection sensitivity.

7 Interferences

- 7.1 Substances with coinciding retention times and that produce a detector response of any of the anions of interest could interfere.
- 7.2 Bromate and chlorite may interfere with the determination of chloride and fluoride.
- 7.3 A high concentration of any one ion can interfere with the resolution of other ions.
- 7.4 The instrument is calibrated by the manufacturer and certified by the installer.

8 Health and safety

- 8.1 These analyses involve handling freshwater samples that may contain live microorganisms and therefore pose some threat of infection. Laboratory personnel who are routinely exposed to such water samples are encouraged to protect themselves from water borne illnesses by wearing clean disposable gloves and washing their hands frequently.
- 8.2 Use protective gloves to handle all of the chemical substances necessary for operation of this instrument. Review the MSDS for additional information and safety concerns regarding the chemical substances used throughout these procedures.
- 8.3 All maintenance will be performed by authorized personnel only.
- 8.4 Keep all loose clothing away from mechanical moving parts associated with the carousel and sample tubes.

9 Personnel qualifications

Samples will be analyzed by Missouri State University (MSU) laboratory personnel who have received appropriate training from experienced personnel, prior coursework, and laboratory experience regarding the analyses, and who are familiar with all of MSU's sample handling and labeling procedures and appropriate SOPs.

10 Equipment and supplies

- 10.1 Autosampler Cassettes: loads samples into the instrument, 5.0ml cassettes, Dionex Reorder P/N: 046032.
- 10.2 Chain of Custody Forms: used to describe the written record of the collection, possession and handling of samples. Chain of custody (COC) forms are located on a board in Temple Hall 125. Chain of custody forms should be completed as described in the Chain of Custody SOP # 1030R01.
- 10.3 Filter Syringes: Whatman Autovial Syringeless Filters, pore size 0.45µm, Fisher cat. no. 09-921-20.
- 10.4 Protective Gloves: Nitrile gloves for protection against chemicals.
- 10.5 Vials: 5.0ml plastic, Dionex Reorder P/N: 038008.
- 10.6 Vial Caps: plain caps for 5.0ml vials, Dionex Reorder P/N: 039528.
- 10.7 Tubing for conductivity cell calibration, 0.076mm (ID) YELLOW PEEK tubing, Dionex Reorder P/N: 049715.

11 Reagents and standards

- 11.1 Eluent Solution: 10 ml of AS22 Eluent Concentrate (Dionex Reorder P/N: 063965) is diluted to 1000 ml with Type 1-grade water. Resultant solution contains 4.5 mM sodium carbonate and 1.4 mM sodium bicarbonate. This solution can be used for only 3 weeks.
- 11.2 Conductivity Solution: Dissolve 0.07456 g of reagent-grade KCl in 1 liter of deionized water. Resultant solution concentration: 1.0 mM KCl.
- 11.3 Check Standard Solutions: Use a premade commercial standard (Inorganic Ventures MSU ICAL-2, 500ml) for all check standard solutions. The pre-made commercial standard contains 30 mg/l SO_4 , 0.5 mg/l HPO_4 , 1 mg/l Br^- , 75 mg/l Cl^- , 5 mg/l $\text{NO}_3\text{-N}$, and 1 mg/l F^- . Standards will be stored in plastic bottles and are stable for approximately one month.
 - a. Dilute 1ml of the premade commercial standard to 100ml with Type 1-grade water within a volumetric flask to make Standard 1.
 - b. Dilute 4ml of the premade commercial standard to 100ml with Type 1-grade water within a volumetric flask to make Standard 2.
 - c. Use 10ml of the premade commercial standard for Standard 3.
- 11.4 Type 1-grade water: Fisher cat. no. 9150-5.

12 Sample collection, preservation, shipment and storage

- 12.1 Samples are collected in 500 mL plastic bottles.
- 12.2 Sample bottles are stored on ice for transport to the laboratory.

12.3 Samples are stored in the refrigerator in Temple Hall room 125 prior to analysis.

12.4 Samples will be analyzed within 48 hours of collection.

13 **Quality control**

13.1 Quality control program: The minimum requirements of the quality control program for this analysis consist of an initial demonstration of laboratory capability and the periodic analysis of laboratory reagent blanks and other laboratory solutions as a continuing check on performance. The laboratory must maintain performance records that define the quality of the data that are generated.

- a. Analyses of laboratory blanks are required to demonstrate freedom from contamination. One LRB is analyzed for every 15 samples analyzed.
- b. The laboratory will, on an ongoing basis, demonstrate through calibration verification and analysis of the ongoing precision and recovery sample that the analysis system is in control.
- c. The laboratory will maintain records to define the quality of data that is generated.

13.2 Initial demonstration of performance. The following must be satisfied before the analytical procedure may be used for samples and before a new analyst may analyze samples.

- a. Method Detection Limit (MDL) – To establish the ability to detect the analyte, the analyst shall determine the MDL by carrying through 7 or more separately prepared blanks through the analytical procedures. The average value, \bar{X} , and the standard deviation of the values, s , shall be calculated. The MDL is equal to $3s$ ($3 \times$ standard deviation). The MDL should $\leq 1.0\text{mg/l}$ for all forms of carbon.
- b. Initial Precision and Accuracy– Initial instrument calibration, monthly instrument calibration checks, and individual batch calibration establishes the ability of operators to generate acceptably precise and accurate results. The manufacturer performed an initial 3-point calibration of the instrument. Additional 3-point calibrations are performed with each batch. The measured values must be within 10% of the true value of the standards.

13.3 The LRB is measured along with the standards at the start of the analytical cycle. The LRB must be less than or equal to the required method detection limit to pass the quality control program. A set including the LRB, the laboratory duplicate (LD), and a check standard (Std Check) will be analyzed after every 15 samples. The criteria for these checks are noted in Table 2.

Table 2. Quality control samples and acceptance criteria.

Check	Acceptance Criteria
LRB	\leq MDL
LD	$\pm 20\%$ of the true value
Std Check	$\pm 20\%$ of the true value

13.4 Ten aliquots of sample were filtered through a syringeless filter and then an aliquot of HPLC water was filtered through the syringeless filter. The HPLC filtrate was analyzed. HPLC filtrate was collected after each set of ten sample filtrations up to and including n=50. Data was reviewed and the syringeless filter was determined to be uncompromised through 25 sample filtrations. Blanks filtered after the filtration of 25 samples produced trace amounts of anions therefore the syringeless filters can be used to filter a maximum of 25 samples.

14 Calibration and standardization

14.1 Calibration: Calibration employs a standard curve by plotting detector response in peak height or area of standards (including the reagent blank) versus analyte concentration. The Chromeleon software automatically computes the calibration function and illustrates the calibration plot as well as the calibration coefficients.

$$\text{Amount}_p = f_p(y_p) \times \text{Resp. Fact.}_p \times \text{ISTD Fact} \times \frac{\text{Dil. Factor}_n}{\text{Weight}_n}$$

where: p = peak number
 f_p = calibration function for p calculated during calibration
 y_p = area of peak
 Resp. Fact._p = scaling factor
 $\text{ISTD Fact} = \frac{\text{Amount}_{\text{ISTD(peak table)}}}{\text{Amount}_{\text{ISTD(sample)}}}$ = correction factor for the internal/external standard
 Dil. Factor = dilution factor parameter from the sample list

14.2 Based on this equation, the detector signal of each standard should predict the concentration of the standard to within $\pm 20\%$ accuracy. The correlation coefficient, R^2 value of the equation, should be ≥ 0.995 in order for the equation to be used to predict concentration.

14.3 The instrument will be calibrated with each batch.

14.4 The conductivity cell employs a cell constant to determine conductivity. The cell constant (k) is a factor determined experimentally by measuring the conductance (G) of a standard solution of known equivalent conductivity (\acute{K}).

$$k = \acute{K} / G$$

The value of k depends on the surface area of and distance between the electrode faces in the conductivity detector cell.

$$k = l / A$$


where: l = length

A = area of one electrode

14.5 The conductivity cell must be recalibrated after every 6 months of use. Use the procedures below to calibrate the conductivity cell.

- a. Prepare the conductivity solution as described in the Reagents and Standards Section 11.2.
- b. Remove the 0.076mm (ID) YELLOW PEEK tubing (P/N 049715) listed in the Equipment and Supplies Section 10.7.
- c. Open the Wellness Panel



1. Start up the Chromeleon Software by clicking on . This will open the browser.
2. Click **(+) GGP-3846648_local** within the browser's toolbar.
3. Click **(+) Dionex Templates**.
4. Click **(+) Panels**.
5. Click **(+) Wellness**.
6. Double click **ICS-1000_1500_2000_Wellness_User.pan**.
7. A Wellness Panel similar to Figure 2 should be visible.

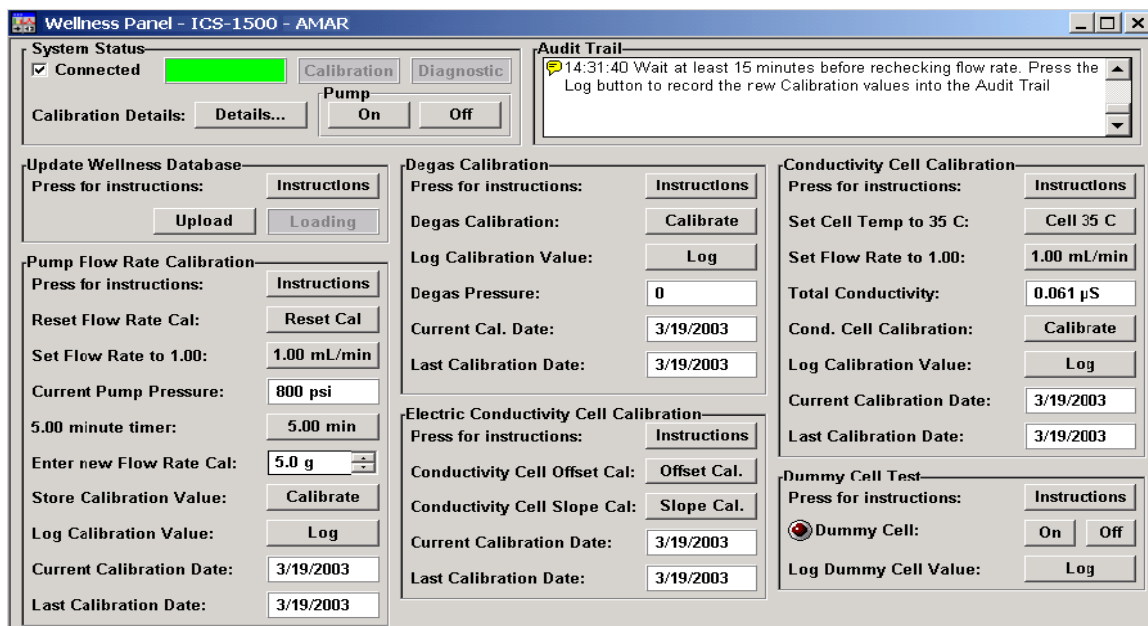
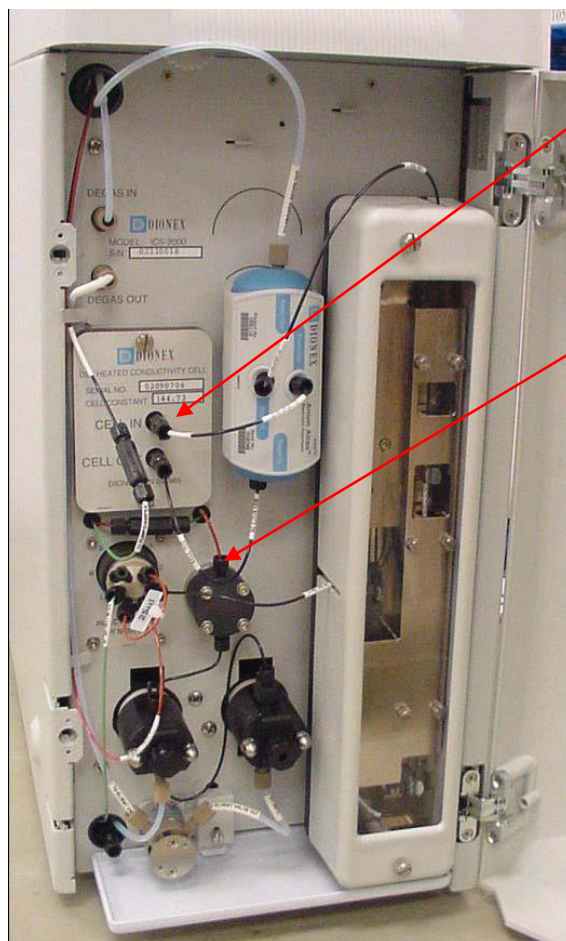


Figure 2. Wellness Panel.

8. Click **Control** on the menu bar.
9. Click **Connect to Timebase**.
- d. Click the **Offset Cal** button within the Electric Conductivity Cell Calibration section of the Wellness Panel.
- e. The Audit Trail box will indicate when the offset calibration is complete, once complete, click the **Slope Cal** button within the Electric Conductivity Cell Calibration section of the Wellness Panel.
- f. When slope calibration is complete, Chromeleon will retrieve the new calibration value from the ICS-1000 and will store it as the current calibration value. Chromeleon will automatically update the Current Calibration Date and Last Calibration Date fields.
- g. Disconnect
 1. Disconnect the BLACK tube from the conductivity CELL IN port (labeled ELUENT OUT --- CELL IN) by unscrewing the black bolt. The bolt is to remain attached to the black tube. See Figure 3 ICS-1000 Component Panel.
 2. Disconnect the RED tube on top of the pressure transducer by unscrewing the black bolt. The tube is approximately 6 cm long, take care not to pull on the tube and keep the bolt attached to the tube.



Black tube from CELL
IN port.

Red tube from
pressure transducer

Figure 3. ICS-1000 Component Panel.

3. The YELLOW PEEK tubing should have ferrules and bolts attached to both ends of the tube. Remove it from its storage container and connect the YELLOW PEEK tubing to the pressure transducer and to the CELL IN by placing the ferrule into the port and sliding the bolt down. Hold the tubing and ferrule in place while screwing the bolts to tighten the tube into each port. Do not strip the bolts by tightening them too much; finger tighten and then use a wrench to tighten $\frac{1}{2}$ to $\frac{3}{4}$ turn.
4. Click the **On** button within the Pump portion of the System Status section of the Wellness Panel. Observe the YELLOW PEEK tubing for any leaks while the pump is on and tighten bolts if necessary with wrench and eighth of a complete turn at a time.
5. Verify that there is a minimum of 1000 psi (7 MPa) of backpressure within the Current Pump Pressure box within the Pump Flow Rate Calibration section of the Wellness Panel. If the

correct pressure is not achieved, disconnect the green pump output tubing from the injection valve labeled TO INL VALVE-P and located at site “P” in Figure 4 below.

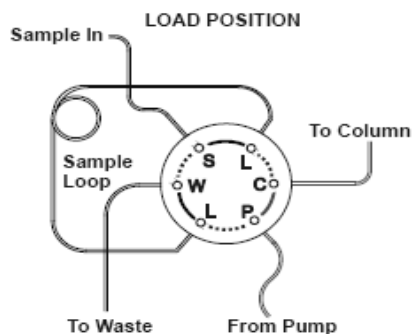


Figure 4. Injection Valve Diagram.

- h. Click the **Cell 35 C** button after the Set Cell Temp to 35 C line within the Conductivity Cell Calibration section of the Wellness Panel.
- i. Monitor the cell temperature by viewing the standard ICS-1000 control panel. When the temperature reaches 35 ° C wait 5 minutes and then continue.
- j. Click the **Off** button within the Pump portion of the System Status section of the Wellness Panel to turn the pump off.
- k. Disconnect the tan union to the eluent bottle and place the tubing connected to the instrument into a flask containing the conductivity solution (KCl) made during step14.5a. See Figure 5 below.



Tan union.

Figure 5. Eluent Bottle.


- l. Click the **On** button within the Pump portion of the System Status section of the Wellness Panel to turn the pump ON.
- m. Click the **1.00 ml/min** button after the Set Flow Rate to 1.00 line within the Conductivity Cell Calibration section of the Wellness Panel.
- n. Wait approximately 20 minutes until the Total Conductivity line within the Conductivity Cell Calibration section of the Wellness Panel stabilizes. Total Conductivity has stabilized if the conductivity fluctuates by only ± 0.5 μS .
- o. Once stable click the **Calibrate** button within the Cond. Cell Calibration line within the Conductivity Cell Calibration section of the Wellness Panel. The Audit Trail box will indicate when the calibration procedure is complete. After calibration, the Chromeleon software will retrieve the new calibration value from the ICS-1000 and store it as the current calibration value. The conductivity reading should be **147.00 \pm 2 $\mu\text{S/cm}$** .
- p. The Audit Trail box will also indicate the cell calibration constant which should be between **130** and **190**. If the cell calibration constant is not between 130 and 190 refer to the troubleshooting section of the operator's manual.
- q. Click the **Log** button within the Log Calibration Value line within the Conductivity Cell Calibration section of the Wellness Panel. Chromeleon will automatically update the Current Calibration Date and the Last Calibration Date fields.
- r. Click the **Off** button within the Pump portion of the System Status section of the Wellness Panel to turn the pump off.
- s. Flush the system
 1. Transfer the eluent tube from the beaker of conductivity calibration (KCl) solution to a beaker of Type 1 water and click the **On** button within the Pump portion of the System Status section of the Wellness Panel.
 2. Wait approximately 25 minutes to allow the conductivity to drop to approximately 1 μS .
 3. Once the conductivity drops to approximately 1 μS , click the **Off** button within the Pump portion of the System Status section of the Wellness Panel.


- t. Reconnect
 - 1. Disconnect the YELLOW PEEK tube by unscrewing the bolts from both ports
 - 2. Reconnect the BLACK tube from the Suppressor ELUENT OUT port to the CELL IN port.
 - 3. Reconnect the GREEN tube (if disconnected) to the injection valve location "P".
 - 4. Reconnect the RED tube to the pressure transducer. *Do not over tighten this tubing.*
 - 5. When all of the tubes are reconnected, click the **On** button within the Pump portion of the System Status section of the Wellness Panel.
 - 6. If the Current Pump Pressure line within the Pump Flow Rate Calibration section of the Wellness Panel exceeds 3000 psi, loosen the RED tubing bolt until pressure drops but no fluid is leaking from the connection.
- u. Check for additional leaks and adjust bolts accordingly.
- v. Click the **Off** button within the Pump portion of the System Status section of the Wellness Panel and close the Wellness Panel. Click **YES** when prompted to save changes.

14.6 Calibration standards and check standards are analyzed with each batch to monitor instrument shift.

15 Procedure

15.1 Pre-analysis Procedures

- a. If the ICS-1000 is not powered on, flip the main switch located on the rear panel of the instrument near the blower vent to power on the instrument.
- b. If the autosampler is not powered on, flip the main switch located on the left side of the back panel.
- c. Turn on the PC and wait for the booting procedure to end.
- d. Open the Chromeleon software by double clicking . This will open the browser.

- e. Within the browser's toolbar click  to open the Default Panel Tabset. A small window displaying 'Connect to Chromelon Server' will appear.
 - f. Click on **(+) My Computer** to expand the menu, highlight **Chromelon Server**, and select **OK**. The Panel Tabset1 window will be displayed. Maximize the window.
 - g. Check the eluent reservoir located on top of the ICS-1000. Approximately 1 L of eluent is required per analysis. Eluent is stable for approximately 2 weeks. If 1L of eluent is not in the reservoir follow the steps below to refill the eluent reservoir.
 1. Unscrew the cap on the top of the eluent bottle.
 2. Fill the reservoir with the solution made in the Reagents and Standards Section 11.1 and screw the cap back onto the eluent bottle.
 3. Click the Pump Settings button within the Pump section of the Panel Tabset1 window to set the eluent level. Enter the correct volume of eluent that is in the reservoir and click **Close**. Note the unit of measure is in liters.
 4. Check the tubing connections from the reservoir to the ICS-1000.
 - h. Ensure that the "Connected" field within the ICS-1000 System section within the Panel Tabset1 window, has a check mark in it and click **Startup**. The pump and the suppressor will make a noticeable sound while engaging. The pressure as indicated in the Pump section within the Panel Tabset1 window will start to increase and the conductivity as indicated in the Detector section within the Panel Tabset1 window will decrease.
- 15.2 If eluent has been changed or if lines are dry or have not been used within 24 hours, prime the eluent lines with a syringe by following the directions below.
- a. Verify that the eluent reservoir is full, the reservoir cap is hand tightened, and the liquid line from the ICS-1000 to the reservoir cap is connected.
 - b. Verify that the waste lines are directed to a waste container.
 - c. Verify that the pump is turned off by viewing the ICS-1000 Pump section within the Panel Tabset1 window. Pump-Off should be displayed, if not click on the **Pump Settings...** button and then click **Off**.
 - d. Remove the front panel from the instrument. Connect a 10 mL syringe to the port in the priming valve on the primary pump head. See Figure 6 below.

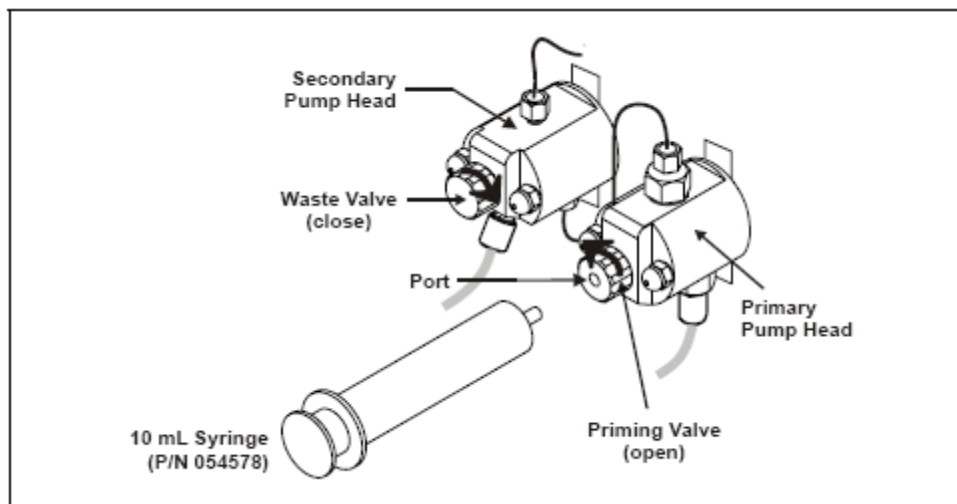



Figure 6. Priming the Eluent Lines

- e. Open the priming valve by turning it one-quarter to one-half turn counterclockwise. Refer to Figure 6.
 - f. Click **Pump Settings** within the ICS-1000 Pump section within the Panel Tabset1 window. Then click the **Open** button within the Eluent Flow Valve section of the Pump Settings window.
 - g. Draw the syringe back to begin pulling eluent through the flow path. It may take several syringe draws to remove all air or previous eluent from the tubing. Transfer eluent drawn into the syringe to the flask labeled "IC waste".
 - h. Use the syringe approximately 4 times (~ 40ml of eluent into the syringe) to thoroughly prime the lines, then close the priming valve. *Do not overtighten the priming valve.*
 - i. Close the eluent valve. Click **Pump Settings** within the ICS-1000 Pump section within the Panel Tabset1 window. Then click the **Closed** button within the Eluent Flow Valve section of the Pump Settings window.
- 15.3 Prime pump following the directions below.
- a. Click on **Pump Settings**.
 - b. Click on **Prime**. A prompt will appear requesting that the waste valve be opened. Refer to Figure 2 and turn the waste valve to the left to open.
 - c. After the waste valve is open, click **OK** within the prompt window.

- d. Allow the instrument to prime until no more bubbles are seen exiting the tubing from the eluent valve lines which could take a maximum of 5 minutes.
- e. Click **Pump Off**.
- f. Close the waste valve.
- g. After priming the pump, click the box next to the pressure reading box within the ICS-1000 Pump section to turn the pump back on. Click the box next to the suppressor reading box within the ICS-1000 Detector section to turn the suppressor back on.

15.4 Set System Operating Conditions

- a. Click  (Acquisition On/Off...) within the main window toolbar to start the data acquisition. A window titled "Data Acquisition – IC" will appear.
- b. Make sure that the fields labeled "Channel_Pressure", "ECD_1", and "ECD_Total" have check marks in them and click **OK**.
- c. This initiates a trace graph. Wait for approximately 15 minutes for the trace graph to flat line before proceeding. Change the y-axis by clicking on the axis and resetting the values to zoom in on the graph and ensure that a true flat line exists. If the graph does not flat line contact the laboratory supervisor.
- d. Click on the **Detector Settings** button within the Detector section within the Panel Tabset1 window and ensure that the total conductivity is 15-23µS, the pressure is 200-3000 psi, the cell temperature is 35 C, and the suppressor current is 37mA.
- e. Click **Autozero**, ensure that the instrument has truly autozeroed, and then click **Close**.
- f. Check the waste bottle for free board volume and dispose of waste properly if the bottle is already ¾ full.

15.5 Setting up a new run

- a. Minimize the Panel Tabset1 window. Navigate to the "OEWR Template" and highlight "OEWR Template". See Figure 7.

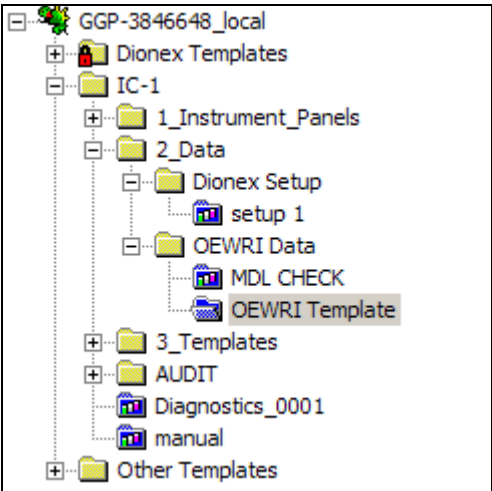


Figure 7. View of the File Explorer within the Chromeleon browser.

- b. Click **File** then select **Save As**. The Save As window will be displayed. Type the project name and sample collection date for the samples as the file name and click **Save**.
- c. Verify that the newly created file is listed under OEWRI Data in the browser. The template provides the correct sample sequence for the standards and laboratory reagent blanks (LRB) for each batch.
- d. Right click within the screen and highlight “Append Sample”. Edit each row to include each sample within the batch. Refer to Figure 8 below.

No.	Name	Type	Pos	Inj. Vol.	Program
1	LRB	Blank	1	20.0	OEWRI pr
2	standard 1	Stand:			
3	standard 2	Stand:			
4	standard 3	Stand:			
5	LRB	Blank			
6	LRB	Unkno			
7	Sample ID	Unkno			

How to...
What's this?
Query
Sync
Copy
Paste
Clear
Insert Sample
Delete Samples
Append Sample
Edit Field

Figure 8. Editing sample sequence

- 15.6 Prepare calibration standard solutions as defined in the Reagents and Standards Section 11.3. Calibration standard solutions will be analyzed with each batch of samples.

15.7 Sample Preparation

- a. Organize the project sample bottles to minimize confusion when preparing samples and entering sample information into the template. A LRB and Std. Check are analyzed after every 15 samples or less as the batch dictates. Two LDs are analyzed for every 15 samples as well. The Std. Check should be standard 1 which is the most dilute of the standards. Shake sample container prior to filtration.
- b. Place the 5 ml plastic vials into the autosampler cassettes.
- c. Use filter syringes to filter samples directly into each vial. The filter syringes contain a 0.45 μm filter and can be used to filter 25 samples. Remember that the first LRB should be in the vial closest to the black dot on the cassette.
 1. Transfer approximately 5 ml from the sample container to the syringe.
 2. Use the plunger to push the sample through the filter and into the vial.
 3. Rinse the plunger with doubly deionized (DDI) water and push approximately 3ml of DDI water through the filter to flush the filter prior to the next sample.
 4. Continue filtering samples into vials making sure not to exceed the maximum amount of samples (25) that can be filtered per vial.
- d. Fill each vial to the first tic mark on the cassette. This mark is found about an inch from the top of the cassette.
- e. Place the caps on the vials. Use the Cap tool to press down the caps. Use the figure below for reference. Figure 9 illustrates the correct position of the cap within the vial. Notice that the larger end of the cap is on the bottom side while the smaller end of the cap is facing the top. The top of the cap should be even with the top of the vial. Use the flat end of the Cap tool to press the cap down to the correct position.

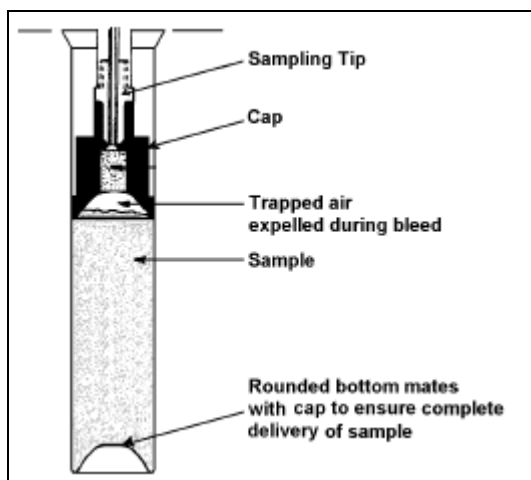



Figure 9: Sample vial with cap ready for analysis

- f. Press the Hold/Run switch located on the front panel of the autosampler to set the sampler to Hold. A green light will display to the left side of the word "hold".
- g. Slide the spring-loaded cassette pusher back and hold it. Place the filled cassettes into the autosampler tray. Make sure that the black dots located on the top corner of the cassette are on the right hand side when loading the cassettes into the autosampler so the cassettes and vials move appropriately through the instrument.
- h. After all the cassettes are in place, allow the pusher to slide forward into place against the last cassette.
- i. Press the Hold-Run switch to Run to initiate the autosampler. The green light will illuminate next to the run side of the switch.

15.8 Sample Analysis

- a. Click  to stop data acquisition. The analysis will not begin if data is being acquired.
- b. Click **Batch** from the Chromeleon Browser toolbar and select Start. Click **Ready Check**. The instrument will perform a self-check prior to the analysis.
- c. After the instrument self-check, the Chromeleon-Check Results window will be displayed. This window indicates the volume of eluent required for the batch analysis. Ensure that the volume in the eluent reservoir is more than the volume indicated.
- d. Click **OK** then click **Start**.

- e. Analysis progression can be checked by noting that the sample being analyzed is highlighted green.
- f. After the three standards have been analyzed, double click on **standard 1**. A new window containing the peak data will be displayed similar to Figure 10 below.

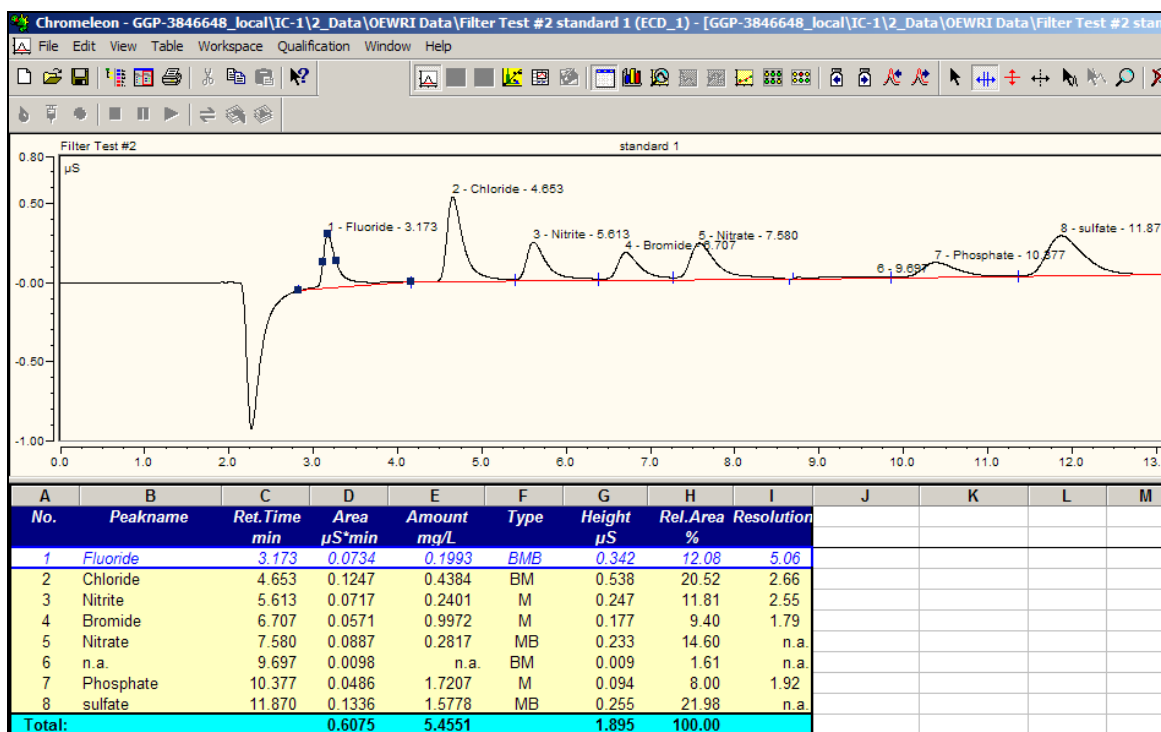



Figure 10. Peak data window.

- g. If the window does not look like Figure 10, stop the analysis and contact the laboratory supervisor.
- h. Open the QNT-Editor by clicking on . At the bottom of the window, click on the Peak Table tab as shown below. Edit the retention times (Ret. Time) to the correct value by looking at the graph shown above the Peak Table. Refer to Figure 11 below.

No.	Peak Name	Ret.Time	Window	Standard	Int.Type	Cal.Type	Peak Type	Group	Cor
1	Fluoride	3.178 min	3.000 RG	External	Area	LOff	Auto		
2	Chloride	4.661 min	3.000 RG	External	Area	LOff	Auto		
3	Nitrite	5.618 min	3.000 RG	External	Area	LOff	Auto		
4	Bromide	6.711 min	5.000 RG	External	Area	LOff	Auto		
5	Nitrate	7.578 min	5.000 RG	External	Area	LOff	Auto		
6	Phosphate	10.384 min	10.000 R	External	Area	LOff	Auto		
7	sulfate	11.881 min	10.000 R	External	Area	LOff	Auto		

Figure 11. QNT-Editor window.

- j. Once the peak table has the correct retention times each peak will be given the correct title (fluoride, chloride, etc.) and the software program will recognize all other peaks from the analysis of each sample within the batch.
- k. After all of the peak retention times have been updated, close the QNT Editor by clicking **File**, then click **Close**. A prompt will be displayed, "Save changes to Method File Dionex setup?", click **Yes** to save the changes.

15.9 Post Sample Analysis Shut Down

- a. After all samples within the batch have been analyzed, ensure that the pump has stopped pumping.
- b. There may be an alarm light on the column, if so, click the display square left of the current display to turn the suppressor on and then click it again to turn it off. The display square will be light grey when the suppressor is off. The display square will be dark grey when the suppressor is on.
- c. When the analysis is complete the autosampler's Hold/Run switch should automatically switch to the **Hold** setting.
- d. Remove all sample vials from the cassettes and retain the used vials and caps. Place the empty cassettes behind the spring loaded cassette pusher.
- e. Return all analysis materials to appropriate locations of the shelf and remove personal items from the station.
- f. Check the waste bottle for free board volume and dispose of waste properly if the bottle is already $\frac{3}{4}$ full. Retighten the cap on the waste bottle.

- g. Do not reuse any vials or caps.

15.10 Export and Save Data

- a. Export the raw data by highlighting the desired file to be exported within the OEWRI Data file. Select File, then Batch Report within the Chromeleon browser window. The Batch Report window will be displayed.
 - 1. Use the down arrow in the window to select Reports\peak analysis OEWRI and ECD_1.
 - 2. Deselect the check mark by Printout.
 - 3. In the **Export options** section, select the box next to Export.
 - 4. Select Excel file format (*.xls) then select Next>.
 - 5. Ensure that the **Peak analysis** and **Calibration** boxes are checked.
 - 6. Click **Finish** to close the Export options box.
 - 7. Select OK on the Batch Report window. A new Batch Report window will be displayed that includes a progress bar.
 - 8. When the bar reaches 100%, select OK.
 - 9. Once data has been exported, the Chromeleon software can be closed.
- b. Data can be viewed by clicking **Start** and then **Documents**. Select My Documents and double click on the **Chromeleon Folder** in the window displayed. Once in the Chromeleon folder, select IC_1, then select IC_1 again, then select 2_Data, and finally select OEWRI Data.

16 Data acquisition, calculations, and reporting

- 16.1 The data from each batch will be saved to the IC computer and the same data will be exported as an Excel or pdf file for project management use.
- 16.2 The ICS-1000 does not perform absolute measurements but determines the value by calculations. Standards of known element content are used to make a series of standards that vary in concentration and are used for instrument calibration. Calibration employs the equation below to represent the relationship between the peak area and concentration. The ICS-1000 software automatically computes the calibration function and illustrates the calibration plot as well as the calibration coefficients.

$$\text{Amount}_p = f_p(y_p) \times \text{Resp. Fact.}_p \times \text{ISTD Fact} \times \frac{\text{Dil. Factor}_n}{\text{Weight}_n}$$

where: p = peak number
 f_p = calibration function for p calculated during calibration
 y_p = area of peak
Resp. Fact. $_p$ = scaling factor
ISTD Fact = $\frac{\text{Amount}_{\text{ISTD(peak table)}}}{\text{Amount}_{\text{ISTD(sample)}}}$ = correction factor
for the internal/external standard
Dil.Factor = dilution factor parameter from the sample list

The calibration coefficients are included in the data file and are printed with a standard report.

- 16.3 Resolution (R) is a measure of the separation between 2 sample components and expressed as the ratio of the distance between the 2 peak maxima to the mean value of the peak width at the baseline.

$$R = 2(t_2 - t_1) / (W_2 + W_1)$$

where: t_2 and t_1 = retention times of components 2 and 1, respectively
 $W_2 + W_1$ = baseline width of peaks 2 and 1 respectively (measured in the same units as the retention time)

- 16.4 Column Efficiency (N) is a measure of the narrowness of analyte bands as they elute from the column. High efficiency is desirable because resolution between closely spaced bands improves with greater efficiency. Column efficiency is proportional to column length. For a symmetrical (Gaussian) peak, column efficiency can be determined by:

$$N = 5.54(t_1 / W_{1/2})^2$$

where: t_1 = peak retention time (sec.)
 $W_{1/2}$ = peak width at $\frac{1}{2}$ height (sec.)

- 16.5 Column Selectivity (a) describes the relative separation of the band maxima between two adjacent peaks and can be determined by:

$$a = (t_2 - t_0) / (t_1 - t_0)$$

where: t_1 and t_2 = retention time of components 1 and 2, respectively

t_0 = retention time of un-retained components

- 16.6 There are no water sample dilutions associated with this method.
- 16.7 Results should be reported to 0.01mg/l.
- 16.8 The evaluation of MDL and precision require calculation of standard deviation. Standard deviations should be calculated as indicated below, where n = number of samples, x = concentration in each sample. Note: This is the sample standard deviation calculated by the STDEV function in Microsoft Excel.

$$s = \left(\frac{\sum x^2 - \frac{(\sum x)^2}{n}}{n - 1} \right)^{1/2}$$

- 16.9 The Method Detection Limit (MDL) is the lowest level at which an analyte can be detected with 99 percent confidence that the analyte concentration is greater than zero and is $3.14(\text{standard deviation}) = 3.14(s)$.
- 16.10 Minimum Quantification Interval is the lowest level that can be quantitated accurately and is generally defined as four times the method detection limit = $4(\text{MDL})$.
- 16.11 All OEWRI project data will be transferred into an OEWRI Project Results template and additional QA/QC data will be added from the original data set.

17 Computer hardware and software

- 17.1 Microsoft Word: this document is prepared using Word.
- 17.2 The Word document file name for this SOP is: IC R01.doc
- 17.3 Chromeleon software controls all aspects of the instrument, data acquisition, and data processing.

18 Method performance

- 18.1 The desired performance criteria for this measurement are:
- Detection limit: carbon = 1mg/l for all forms of carbon.
 - Precision: $\pm 20\%$
 - Accuracy: $\pm 20\%$
 - Minimum Quantification Interval: 0.01mg/l
- 18.2 The operating range for this instrument is 0 to 100,000 mg/l. The sample volume range is 0.05 ml to 4 ml.

18.3 The instrument will be calibrated with each batch.

19 Pollution prevention

All wastes from these procedures shall be collected and disposed of according to existing waste policies within the MSU College of Natural and Applied Sciences. Volumes of reagents made should mirror the number of samples being analyzed. These adjustments should be made to reduce waste.

20 Data assessment and acceptable criteria for quality control measures

20.1 The analyst should review all data for correctness (e.g., calculations).

20.2 Precision values are calculated for pairs of duplicate analyses and are recorded as a percent. The desired precision is $\pm 20\%$.

20.3 The desired detection limit for fluoride is $2.0\mu\text{L}$, chloride is $4.0\mu\text{L}$, nitrite is $3.7\mu\text{L}$, bromide is $14.0\mu\text{L}$, nitrate is $2.7\mu\text{L}$, orthophosphate is $14.0\mu\text{L}$, and sulfate is $18\mu\text{L}$.

20.4 All data is reviewed by the analyst's supervisor or the OEWRI QA coordinator.

21 Corrective actions for out-of-control or unacceptable data

21.1 The results for precision and blank data are compared to the acceptable values for this analysis; $\pm 20\%$ and 0 for all forms, respectively.

21.2 If a precision value exceeds $\pm 20\%$ then the analyst should note that the data are associated with an out-of-control duplicate analysis in the data report given to the QA/QC manager. The upper control limit (UCL) = 20% .

21.3 If a LRB value exceeds the MDL then the analyst should note that the data are associated with a LRB value that exceeds the detection limits in the data report given to the QA/QC manager.

21.4 If data are unacceptable for any reason, the analyst should review their analytical technique prior to conducting this analysis again.

21.5 If there is sufficient volume of sample remaining for all samples, the batch may be re-analyzed. Quality control data from both batches is recorded.

21.6 The instrument may require trouble shooting techniques if the data are unacceptable. Refer to the Troubleshooting section within the Operator's Manual for techniques or inform the laboratory manager of potential issues. The Wellness Panel is used to calibrate the pump flow rate, the vacuum degas assembly, and the conductivity cell as well as to test the conductivity cell electronics and upload calibration values from the instrument. To open the Wellness Panel:

- a. Open the Chromeleon Browser window and expand the Dionex Templates folder.
- b. Expand the Panels folder and then the Wellness folder.

- c. Double-click ICS_1000_1500_2000_Wellness_user.pan in the right pane of the Browser window.
- d. Open the Controls menu and click the name of the ICS-1000 timebase at the bottom of the menu to connect the Wellness Panel to the instrument.

22 Waste management

- 22.1 The wastes generated in this method are not hazardous. Solutions can be discarded in the laboratory drain followed by an equal volume of water.
- 22.2 Outdated chemicals are discarded following the procedures of the MSU Environmental Management Department.

23 References

- 23.1 ICS-1000 Ion Chromatorgraphy System Operator's Manual. Revision 2. September 2005. Dionex Corporation, California, USA.
- 23.2 Standard Methods for the Examination of Water and Waste Water, Method 4110 B. Ion Chromatography with Chemical Suppression of Eluent Conductivity, APHA, 21st Edition, 2005.
- 23.3 U.S. Geological Survey – Water Resources Data Missouri Water Year 2001, Water Data Report MO-01-1, 2002.

24 Tables, diagrams and flowcharts

All tables, diagrams, or flowcharts for this method are in the body of this SOP.