
**User's Guide:
Chromeleon[®] Operational Qualification and
Performance Qualification Software Templates
for ICS-3000 Instruments
Revision 7.12**

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Document 065448 Part A
March 2013, Thermo Fisher Scientific Corporation
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PRINTING HISTORY

Revision 7.10 September 2011

Revision 7.11 December 2011

Revision 7.12 March 2013

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1 Introduction

Operational and Performance Qualification are becoming increasingly important to analytical laboratories. Documented evidence must be provided to demonstrate the integrity of data collected and qualify the results obtained on laboratory instrumentation.

The following institutes set and control industry instrumentation standards:

NIST The National Institute of Standards and Technology is an administrative body of the US Department of Commerce. Together with industry, the NIST develops new technologies and sets technological standards countrywide. Thermo Fisher Scientific includes NIST traceable standards as part of the qualification kit.

ASTM The American Society for Testing and Materials sets technical standards and rules for the industry worldwide, specifically; E 1657 – 98: Standard Practice for Testing Variable-Wavelength Photometric Detectors in Liquid Chromatography. E1511 – 93: Standard Practice for Testing Conductivity Detectors used in Liquid Chromatography. E19.09.07: Proposed Standard Practice for Evaluation of Gradient HPLC pumps – Draft.

USP The United States Pharmacopeia (USP) is an official public standards–setting authority for all prescription and over–the–counter medicines and other health care products manufactured or sold in the United States. USP establishes documentary and reference standards to ensure quality analytical instrument qualification process.

These standards are written in broad terms to make them as widely applicable as possible. All stipulate general requirements specifying instruments must be fit for purpose, technically checked in defined periods, calibrated and make sure that test results are documented according to international quality standards. The OQ/PQ procedure used to qualify Thermo Fisher Scientific Ion Chromatography Systems meets the requirements established by NIST, ASTM and USP.

This OQ/PQ procedure provides qualification testing for **ICS-3000** systems composed of any modules listed in section 1.6.

OQ/PQ results can be influenced by unusual laboratory conditions. For example, direct sunlight and close proximity to air conditioning ducts will cause thermal irregularities that will affect baseline drift on conductivity detectors. In these cases, it may happen that the manufacturer's specifications are not met. Ensure that the instrument is operated in suitable environmental conditions or for PQ, the specifications may be adjusted accordingly.

All OQ/PQ activities must be formally recorded, all instruments must be clearly identified, and test reports and supporting documentation must include the following information:

- Module name, type and serial number
- Software and firmware version
- Test data
- Service interval
- Signature of the Thermo Fisher Scientific Service Engineer/Qualification Executor who performed the OQ/PQ
- Signature of the customer who reviewed and accepted the OQ/PQ results

1.1 How to use this manual

It is essential that the operator read and understand this manual completely before implementing any OQ or PQ process.

1. Use the table of contents to determine which tests are required for the system to be qualified and refer to the recommended sections. This will help keep track of what steps to follow during system setup and preparation. It will also aid in selecting the appropriate sequences to run the tests.
2. For the ICS-3000 system, electrochemistry cells in Integrated Amperometry mode are supported for qualification tests. For the Integrated Amperometry detection qualification a pH/Ag/AgCl reference electrode is required for all tests. The ICS-3000 electrochemistry detector DC amperometry and cyclic voltammetry modes are not supported for OQ/PQ testing. When qualifying Integrated Amperometry detection a disposable amino acid gold working electrode is used; other electrodes are not supported.
3. Use the **Table of Contents** and the **List of Tables** as reference guides to locate information quickly.

The electronic version of the OQ/PQ operator's manual contains numerous hypertext links that can take you to other locations within the file. These links include:

- Table of contents entries
- Cross-references (underlined in blue) to sections, figures, tables, etc.

Chapter 1 Introduction

Introduces the conventions used in performing OQ/PQ with the ICS-3000 systems; including the basic requirements, industry standards and manual summary.

Chapter 2 Overview of the Tests

Provides an overview of the tests available to be performed.

Chapter 3 Explanation of Test Procedures

Describes the theory behind each test, the OQ/PQ operating procedures and evaluation of the results..

Chapter 4 Qualification Preparation

Step by step preparation instructions for the OQ/PQ software, hardware installation and the details of the materials in ship kits.

Chapter 5 Performing the Qualification

Detailed operating instructions on how to perform the OQ/PQ including test order, time table, sequence initialization and troubleshooting tips.

Chapter 6 Troubleshooting

Lists problems and presents step-by-step procedures for how to isolate and eliminate the cause of each problem.

Chapter 7 Example Reports

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1.4 Operational Qualification (OQ)

According to “The Development and application of guidance on equipment qualification of analytical instruments” by P. Bedson and M. Sargent [Accred. Qual. Assur. (1996) 1: 265 – 274] the following definition applies:

“The purpose of OQ is to prove and document that an analytical system functions according to its operating specification while the specific environmental conditions are taken into account. In his specification, the supplier has therefore to define exactly the conditions that must be observed.”

OQ should be performed after initial system installation or when an additional new module is added to an existing system. OQ should be done according to the instructions outlined in this manual and all tests should be performed by a qualified Thermo Fisher Scientific Service Engineer. Any hand-written notes made during the performance of the tests must become part of the test results, in accordance with Good Laboratory Practice (GLP) and current Good Manufacturing Practice (cGMP).

1.5 Performance Qualification (PQ)

According to “The Development and application of guidance on equipment qualification of analytical instruments” by P. Bedson and M. Sargent [Accred. Qual. Assur. (1996) 1: 265 – 274] the following definition applies:

“The purpose of PQ is to prove and document that an analytical system functions according to a specification that is suitable for the system’s routine operation. A system is subject to wear when being operated. Thus, it may happen that the supplier’s specification is no longer met. This means: The test procedures used for OQ are also used for PQ, but the tolerances of PQ are less restrictive than those for OQ. Using the same test procedures simplifies the handling.”

PQ should be performed at regular intervals after the initial installation and OQ. Thermo Fisher Scientific recommends performing PQ every six months. PQ should be done according to the instructions outlined in this manual and all tests should be performed by a qualified Thermo Fisher Scientific Service Engineer. Any hand-written notes made during the performance of the tests must become part of the test results, in accordance with Good Laboratory Practice (GLP) and current Good Manufacturing Practice (cGMP).

1.6 Basic Requirements for Successful OQ and PQ

It is a requirement that the most current revision of the OQ/PQ software templates is used in order for a qualification to be considered valid. It is the responsibility of the qualification engineer to determine the most up to date revision of the templates available and ensure that the revision is used before proceeding with this qualification.

It is recommended that the qualification be performed using the most current Chromeleon[®] service pack version (Contact Thermo Fisher Scientific Technical Support for current version).

It is recommended that the qualification be performed using the most current firmware version for each system or module (Contact Thermo Fisher Scientific Technical Support for current version).

Operational Qualification and Performance Qualification are system-specific test procedures. The procedures described below are valid for IC systems comprising of the following equipment (only one type of detector and one pump can be qualified concurrently):

Pump: ICS-3000 SP or ICS-3000 DP

Manual injection or any of the following autosamplers: AS, AS-DV, AS-AP or AS40

AS-AP autosampler 10 mL tray (P/N 074938) with ICS-3000 CD (pull or push mode) and 1.5 mL tray (P/N 074936) with ICS-3000 CD and ED (push mode only).

Absorbance detector: AD25, ICS Series VWD or ICS Series PDA

Conductivity detector: ICS-3000 CD

Electrochemical detector: ICS-3000 ED

Fluorescence detector: RF-2000 with UCI-50, UCI-100 or UI-20 interface

Eluent Generator: ICS-3000 EG

Note: Ensure that the seal wash option is turned off during the qualification. After the OQ/PQ tests are finalized, be sure to re-enable the seal wash option.

In addition, please see the table below for the required Chromeleon® version for each module:

Table 1. ICS-3000 and Chromeleon Compatibility

Module	Minimum Chromeleon Software Version
AS-AP	7.1 SR1c
AS	7.1 SR1
AS-DV	7.1 SR1
AD25	7.1 SR1
PDA-100	7.1 SR1
ICS-3000 SP or DP	7.1 SR1
ICS-3000 EG	7.1 SR1
ICS-3000 DC	7.1 SR1
ICS-3000 CD	7.1 SR1
ICS-3000 ED	7.1 SR1
FLUKE189/289	7.1 SR1
ICS Series VWD	7.1 SR1
ICS Series PDA	7.1 SR1c
ICS-3000 TC	7.1 SR1
RF-2000 with UCI-50, UCI-100 or UI20 interface	7.1 SR1

It is possible to qualify modules in systems that include modules that are not listed in this section; however some modification of the included programs may be required. Such modifications are not within the scope of this document.

It is not possible to qualify multiple detectors on a single system, with the following exceptions: (a) An RF-2000 detector must be qualified in series with a conductivity detector, (b) To qualify the AS-AP or AS autosampler with the injector linearity test (partial loop injections) a 250 µL or larger injection syringe is required.

For the ICS-3000, it is not possible to qualify multiple pumps on a single system. If multiple pumps are configured on a single Chromeleon 7.1 Instrument, the second pump will be ignored by the instrument control programs or instrument methods.

2 Overview of the Tests

The following table provides an overview on the parameters to be tested and the limits recommended for OQ and PQ.

- Template sequences (with programs specific to the Instrument Configuration are generated based on the master sequence of the OQ/PQ templates and the system configuration in the IC Instrument).
- The sequences are copied from each test that is to be performed on the same subject system.
- The Chromeleon software will automatically create a default unique name and directory to contain all template sequences.
- Test sequences must be run in the order they are listed. Do not change the order.

Table 2. OQ/PQ Specification Limits: ICS-3000 CD Baseline Noise/Drift and Detector Linearity

Instrument	Parameter	Description	Limits	
			OQ	PQ
ICS-3000 CD	Baseline Noise	ASTM Type I deionized water with low total organic carbon (TOC)* is pumped through the detector at a flow rate of 1.0 mL/min.	1.0 nS	2.0 nS
	Baseline Drift		10.0 nS/hr	20.0 nS/hr
	Detector Linearity	5 Nitrate solutions from the OQ/PQ standards kit P/N 063540 are injected in different concentrations. Concentration (ppm) and peak height (μS) are represented on a graph. The correlation coefficient for the resulting line and the deviations from it indicate the linearity.	r ≥0.999 RSD ≤5.0 %	r ≥0.999 RSD ≤5.0 %
AD25 and ICS Series VWD	Baseline Noise	ASTM Type I deionized water with low TOC is pumped through the detector at a flow rate of 1.0 mL/min. Wavelength 254 nm and 520 nm.	30 μAU	40 μAU
	Baseline Drift		200 μAU/hr	200 μAU/hr
	Wavelength Accuracy	Holmium oxide maxima are compared with the theoretical maxima.	± 2 nm	± 2 nm
	Detector Linearity	5 Caffeine solutions from the OQ/PQ standards kit P/N 063540 are injected in different concentrations. The absorbance is measured at a wavelength of 272 nm. Concentration (ppm) and peak height (mAU) are represented on a graph. The correlation coefficient for the resulting line indicates the linearity.	r ≥0.999 RSD ≤5.0 %	r ≥0.999 RSD ≤5.0 %

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Table 3. OQ/PQ Specification Limits: ICS-3000 PDA and ED Baseline Noise/Drift and Detector Linearity

Instrument	Parameter	Description	Limits	
			OQ	PQ
PDA-100, ICS Series PDA	Baseline Noise	ASTM Type I deionized water with low total organic carbon (TOC) s pumped through the detector at a flow rate of 1.0 mL/min. Observed wavelengths are set for simultaneous monitoring at 254 nm and 520 nm	0.04 mAU	0.10 mAU
	Baseline Drift		0.5 mAU/hr	1.0 mAU/hr
	Wavelength Accuracy	Holmium oxide maxima are compared with the theoretical maxima.	± 2 nm	± 2 nm
	Detector Linearity	5 Caffeine solutions from the OQ/PQ standards kit P/N 063540 are injected in different concentrations. The absorbance is measured at a wavelength of 272 nm. Concentration (ppm) and peak height (mA) are represented on a graph. The correlation coefficient for the resulting line (and the deviations from it) indicates the linearity.	r ≥0.999 RSD ≤5.0% (up to 1.5 AU)	r ≥0.999 RSD ≤5.0% (up to 1.5 AU)
ICS-3000 ED	Baseline Noise	50 mM NaOH is pumped through the detector cell at a flow rate of 0.25 mL/min. A Au-AAA certified disposable working electrode P/N 060082 is used running the Amino Acids (pH, Ag/AgCl Reference) waveform.	Int. Amp: ≤100 pC	Int. Amp: ≤160 pC
	Baseline Drift		Int. Amp: ≤1000 pC/20 min.	Int. Amp: ≤1250 pC/ 20 min.
	Detector Linearity	5 Threonine solutions from the ED OQ/PQ standards kit P/N 063541 are injected in different concentrations. Concentration (µM) and peak height (nC) are represented by a plot. The correlation coefficient for the resulting line (and the deviations from it) indicates the linearity.	r ≥0.999 RSD ≤5.0 %	r ≥0.999 RSD ≤5.0 %

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Table 4. OQ/PQ Specification Limits: ICS-3000 PDA and ED Baseline Noise/Drift and Detector Linearity

Instrument	Parameter	Description	Limits	
			OQ	PQ
Fluorescence Detectors. RF-2000	Baseline Noise	ASTM Type I deionized water with low TOC is pumped through the detector at a flow rate of 1.0 mL/min. Excitation (EX) WL = 350 nm. Emission (EM) WL is adjusted changed from 450 to 394 nm to give a response between 40 and 80 mV. Noise is measured over thirty, 30-second intervals.	<0.3 mV Minimum Signal 40 mV Maximum Signal 80 mV	<0.3 mV Minimum Signal 40 mV Maximum Signal 80 mV
	Wavelength Accuracy	The EM WL is changed from 380 to 410 nm in steps of 1 nm every 15 seconds. The maximum of the emission spectrum is determined as the maximum of the signal.	≤10 nm	≤10 nm
All injectors and samplers	Injector Precision	10 injections of a defined standard at 25 µL each of the same standard are analyzed. The percent relative standard deviation (%RSD) of the peak areas indicates the reproducibility of the full loop injection volume	RSD ≤0.8 %	RSD ≤1.0 %
	Carryover	Following the injection of a highly concentrated test sample, a blank sample is injected and then a water blank sample is injected. The peak area measured in this chromatogram compared with the peak area of the highly concentrated sample indicates the amount of carryover. If there is any peak area measured in the water blank sample, that area is subtracted from the area of the blank sample before it is compared to the highly concentrated sample.	≤0.1 %	≤0.1 %

	Linearity of Injector Volume	5 injections of the same standard sample are analyzed. The injection volumes are different (10 µL to 50 µL). Injection volume and peak area are represented on a plot of µL vs. nC in ED, µL vs. µS in CD and µL vs. mA in absorbance detectors. The correlation coefficient of the resulting line (and the deviations from it) indicates the linearity. This test does not apply to capillary systems.	r ≥0.999 RSD ≤1.0 %	r ≥0.999 RSD ≤1.0 %
AS only	Sample Preparation Precision (if present – not supported on the AS with ICS-3000 ED detection)	3 samples of identical concentration are prepared from the same starting vial into 3 separate vials and injected. The relative standard deviation of the peak areas indicates the reproducibility of the sample preparation.	RSD ≤1.5 %	RSD ≤2.0 %
AS-AP only	Temperature Accuracy (if present)	The sample temperature is measured using an external thermometer in a standard vial. The vial is filled with water and measured at 15.0 °C.	15.0 ± 2.0 °C	15.0 ± 4.0 °C
AS-AP pH	pH Accuracy	The sample temperature calibration is measuring the pH at 4, 7 and 10. The reading is stable if the reading and the three data points are within 0.2 pH unit of each other.	+/- pH 0.1	+/- pH 0.2
AS-AP Conductivity	Conductivity Accuracy	The conductivity calibration procedure consists of determining the conductivity zero point of the accessory and then measuring the conductivity of a 1000 µS standard.	+/- 30 µS	+/- 50 µS

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Table 5. OQ/PQ Specification Limits: ICS-3000 EG Current, SP/DP Flow Rate Accuracy/Precision and DC Column TC Temperature

Instrument	Parameter	Description	Limits	
			OQ	PQ
ICS-3000 EG	Current Qualification	The eluent generator (EG) is commanded to produce a series of different concentrations (using the current cartridge type configured with a concentration range between 0 mM to 100 mM). At each EG concentration setting the current (mA) produced by the EG is measured and compared with theoretical values.	See Modular Validation Report at: OQ_PQ_Modular_Validation	
ICS-3000 SP, ICS-3000 DP	Gradient Step Accuracy	A step gradient of channels A and B, then C and D is programmed and measured. Theoretical signal heights are compared with recorded signal heights for each step. The deviations between theoretical values and measured values indicate the step accuracy. For channels A and C deionized water is used, for channels B and D 0.3% v/v acetone (absorbance detectors), or 1 mM KCl (conductivity detectors) is used.	≤2.0 %	≤2.0 %
	Gradient Step Noise	For each step of the gradient run described above the noise is determined relative to the signal height.	≤0.2 % for CD, VWD, PDA ≤0.5 % for ED	≤0.2 % for CD, VWD, PDA ≤0.5 % for ED
	Flow Rate Accuracy	Water is pumped at a flow rate of 1.00 mL/min into a weighed beaker for exactly 5 minutes. The mass of the water is measured, the flow rate of the pump calculated and compared with the theoretical value. This experiment is repeated four more times.	1.00 ± 0.05 mL/min	1.00 ± 0.05 mL/min

	Flow Rate Precision	Water is pumped at a flow rate of 1.00 mL/min into a weighed beaker for exactly 5 minutes. This procedure is repeated four more times. The relative standard deviation of the five measurements is calculated.	$\leq 2.0 \%$	$\leq 2.0 \%$
ICS-3000 DC	Column TC and Compartment, Temperature Verification	The Analytical DC column temperature is set to 30 °C and the Analytical DC compartment temperature (if present in a dual temperature zone DC) is set to 20 °C for the duration of the entire qualification. At the end of the qualification the temperature of the column heater is measured with a calibrated external thermometer and compared with the theoretical value.	$\pm 2.0 \text{ }^{\circ}\text{C}$	$\pm 2.5 \text{ }^{\circ}\text{C}$

*ASTM Type 1 DI Water TOC Specification: < 50 ppb.

Note: The instrument's specifications can only be met if the conditions that are described in the accompanying manual are strictly observed.

Note: The AD25, VWD, PDA, and PDA-100 Detector UV and visible lamps must be turned on at least two hours prior to performing the qualification.

3 Explanation of Test Procedures

3.1 Flow Rate Accuracy and Precision Qualification

3.1.1 Theory

The accuracy and precision of a pump is qualified by accurately measuring 5 aliquots of eluent into a tared sample beaker for exactly 5 minutes each. The injection valve is used to measure the 5 minute aliquot and the volume of each aliquot is determined gravimetrically. The backpressure coil is placed before the injection valve and equal lengths of tubing are used for eluent and waste to ensure that there is no backpressure change when switching the valve from waste to sample.

3.1.2 Performing the Test and Evaluating the Results

The Flow Rate Accuracy and Precision Qualification is run using the sequence called XQ_Flow_Rate. The five weight measurements for the aliquots are entered manually into the report and converted to volume when the temperature is entered. Accuracy is measured by comparing the theoretical value (5.00 mL in 5.00 min) to each of the five aliquots and precision is measured by determining the relative standard deviation of the five aliquots.

3.2. Eluent Generator Current Qualification

3.2.1 Theory

The Eluent Generator Current Qualification is determined by performing an EG step gradient using varying eluent concentration set points. The EG applies a specific current to the EG cartridge depending on the concentration of eluent and type of cartridge selected. An IC Qualification test box (P/N 072005) is used in place of the EG cartridge and the applied current for each step is measured using an ammeter or voltmeter that is connected to the test cell. No injection is performed. The Qualification test box must be configured to KOH as the cartridge type.

3.2.2 Performing the Test and Evaluating the Results

The Eluent Generator current qualification test is the next test in the sequence queue and the sequence is called XQ_EG_Current or XQ_EG_Dual_Current. Current readings in mA are displayed on the voltmeter for each concentration step in the gradient. Values for each step are manually recorded (please enter at least three figures after the decimal in each entry) or can be automatically downloaded into the Report Template when using a Fluke 189/289 multimeter or equivalent multimeter (calibrated with up to 300 mV DC scale range and the multimeter needs to display three decimal places) which is configured in the instruments as a device driver. The sequence follows an XQ_Stop sequence that will pause the system to allow connection of the Qualification Test Box and recording of the results.

3.3 Warm up

The warm up sequence is used to ensure that all parts of the system are responding appropriately and to record logs to the audit trail (later used to update the information in the report definition file). It is called XQ_Warm_Up where; X is P for PQ and O for OQ. The warm up sequences from the AS-AP directory will inject samples. The warm up sequences from the AS-AP directory inject low level standards.

3.4 Baseline Noise and Drift (Conductivity and Absorbance Detector)

3.4.1 Theory

A 20-minute detector baseline background trace is collected using deionized water as the eluent at a flow rate of 1.00 mL/min. For ICS Series VWD and PDA, AD25 and PDA-100 detectors, data is collected at 254 nm and 520 nm. No injection is performed. Then a short equilibration is done at 272 nm.

Noise is calculated by partitioning the 20-minute baseline signal into 20 one-minute intervals. The noise is the distance between two parallel lines through the measured minimum and maximum values and the regression line. The measured noise values are then averaged to establish the final value. To calculate drift, the Chromeleon software uses a method of least squares and calculates a regression line from the measured values. The slope of the regression line indicates the drift of the measured signal, and the amount of the slope indicates the amount of drift.

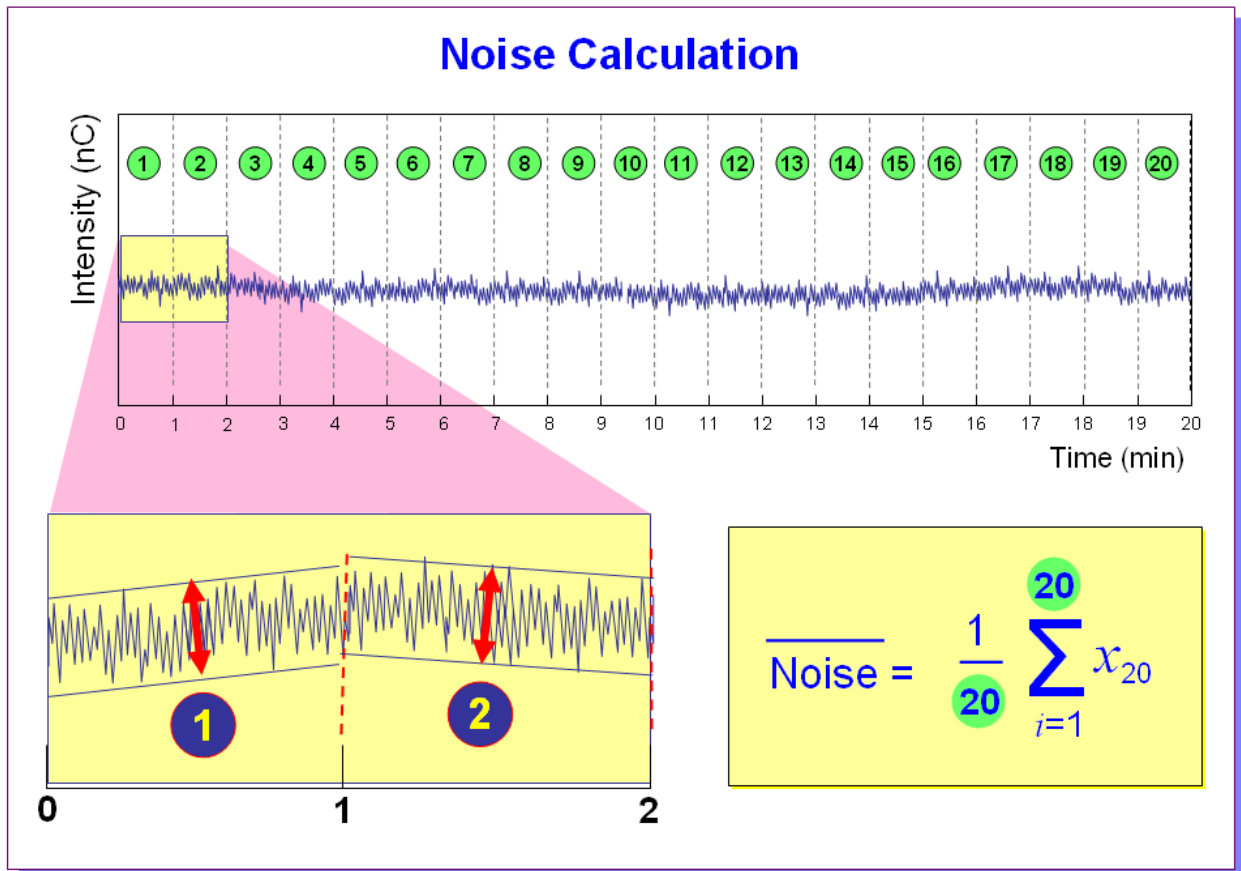


Figure 1. Noise Calculation

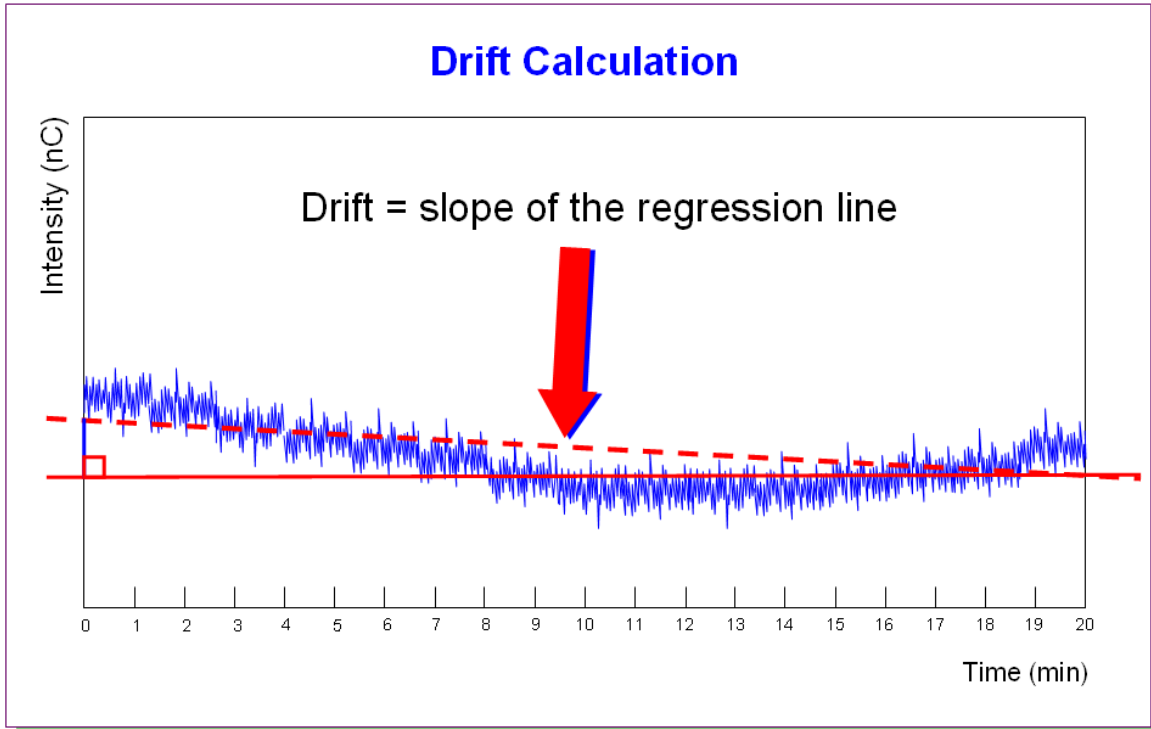


Figure 2. Drift Calculation

The slope of the regression line indicates the drift of the measured signal, and the amount of the slope indicates the amount of drift.

3.4.2 Performing the Test and Evaluating the Results

Noise and drift evaluation for all IC detectors is the next test in the sequence queue and is called XQ_Detector_Noise_Drift where, X is P for PQ and O for OQ and Detector is CD for conductivity detectors, UV_Vis for AD20/25 detectors, VWD and PDA for PDA-100 detectors.

Note: Ensure that the seal wash option is turned off during the qualification. After the OQ/PQ tests are finalized, be sure to re-enable the seal wash option.

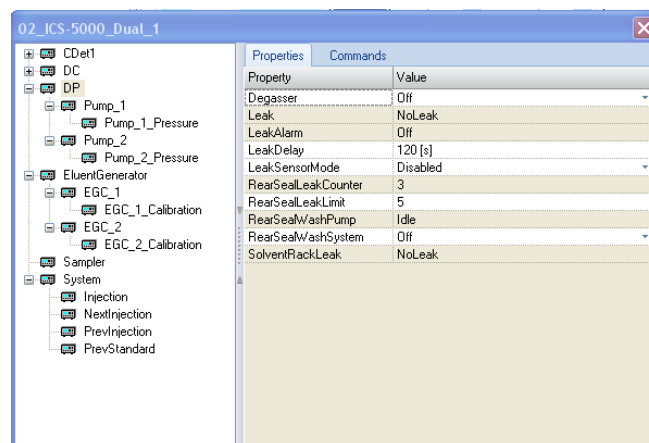


Figure 3. Rear seal wash option

3.5 Integrated Amperometry Noise and Drift

3.5.1 Theory

A 20-minute detector baseline is collected using 50 mM NaOH as the eluent at a flow rate of 0.25 mL/min. An Amino Acid disposable gold working electrode (Thermo Fisher Scientific P/N 060082) is used running the Amino Acid (pH using Ag/AgCl Reference electrode) waveform. No Injection is performed.

3.5.2 Performing the Test and Evaluating the Results

Noise and drift evaluation for the Integrated Amperometry detector is the only test in the sequence queue and is called XQ_ED_Amp_Noise_Drift.

Noise is calculated by partitioning the 20-minute baseline signal into 20 one-minute intervals. The noise is the distance between two parallel lines through the measured minimum and maximum values and the regression line. The measured noise values are then averaged to establish the final value. To calculate drift, the Chromeleon software uses a method of least squares and calculates a regression line from the measured values. The slope of the regression line indicates the drift of the measured signal).

3.6 Injector Precision

3.6.1 Theory

Injector precision is determined by making ten replicate injections of a standard using a fixed sample volume.

PDA/VWD: A 40 ppm caffeine standard is injected when using absorbance detection at a wavelength of 272 nm. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

Conductivity Detector: A 50 ppm nitrate standard is injected when using conductivity detection. Deionized water is used as the eluent at a flow rate of 1.00 ml/min with conductivity.

Electrochemical Detector: A 10 μ M threonine standard is injected when using ICS-3000 ED detector in Integrated Amperometry mode. A 50 mM manually prepared NaOH is used as eluent at a flow rate of 0.25 mL/min with a 2 mm x 50 mm CarboPac PA1 (P/N 057179) guard column is used. Amino Acid disposable gold working electrode (Thermo Fisher Scientific P/N 060082) is used with the Amino Acids (pH, using Ag/AgCl Reference electrode) waveform.

3.6.2 Performing the Test and Evaluating the Results

Injector precision evaluation is the next test in the sequence queue and the samples are called Inj Precision_1 through Inj Precision_10 in the XQ_Detector_Inj_Precision sequence, where X is O for OQ, P for PQ, and Detector is CD for conductivity detectors and ED for amperometry detectors. The concentration of the nitrate or caffeine standard and the sample volume injected is included in the QNT file used for injector linearity.

Precision is determined by calculating the relative standard deviation of the peak areas for the ten injections.

3.7 Sample Carryover

3.7.1 Theory

Sample carryover is determined by injecting a highly concentrated sample, followed by a blank sample and then a second blank injection. The blanks in this case are deionized water.

PDA/ VWD: Caffeine at a concentration of 80 ppm is injected when this test is performed with an absorbance detector.

Conductivity Detector: Nitrate at a concentration of 1000 ppm is injected when this test is performed with a conductivity detector.

Electrochemical Detector: A 10 μ M threonine standard is injected when using ICS-3000 ED detector in Integrated Amperometry mode. A 50 mM manually prepared NaOH is used as eluent at a flow rate of 0.25 mL/min. with a 2 mm x 50 mm CarboPac PA1 (P/N 057179) guard column is used.

Ideally, the chromatogram of the two blank injections should be identical. If, however, a difference is detected, the difference between the peak areas is calculated and the ratio of this difference to the highly concentrated sample is calculated. ASTM Type I water is used as the eluent at a flow rate of 1.00 mL/min for absorbance and conductivity detectors.

3.7.2 Performing the Test and Evaluating the Results

Sample carryover evaluation is included in the same sequence as the Injector Precision test of the sequence queue and the samples are called Water, Carry Over and Water_CO in the XQ_Detector_Inj_Precision_CO sequence.

A peak area ratio is calculated between the blank and high concentration sample to determine sample carryover.

3.8 Detector Linearity

3.8.1 Theory

VWD/PDA: Detector linearity for absorbance detectors is determined at 272 nm using a sequence of five caffeine standard injections at 15, 40, 60, 80 and 100 ppm. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

Conductivity Detector: Detector linearity for conductivity detectors uses a sequence of five nitrate standard injections at 5, 10, 25, 50, and 100 ppm. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

Electrochemical Detector: Linearity for Integrated Amperometry detectors uses a sequence of five Threonine standard injections at 1, 2, 4, 8, and 10 μ M. A 50 mM manually prepared NaOH is used as eluent at a flow rate of 0.25 mL/min with a 2 mm x 50 mm CarboPac PA1 (P/N 057179) guard column is used.

3.8.2 Performing the Test and Evaluating the Results

Detector linearity samples are called Detector linearity_1 through Detector linearity_5 in the XQ_Detector_Linearity sequence. The concentrations for the nitrate, caffeine and the Threonine standards are included in the QNT file used for detector linearity. It is recommended to enter the

actual concentration of the standards from the certification of analysis into the quantification files to achieve the real results.

Depending on which injection module used, the peak height of the sample with the highest concentration may exceed 1500 mAU. This is usually not within the linearity range of UV detectors. Thus, the limits for the regression coefficient and the relative standard deviation may not be met. In this case, reduce the injection volume for all samples used for the linearity check so that the peak height of the sample with the highest concentration is in the linearity range of the tested detector, i.e., usually below 1500 mAU. As an exception, a linearity range of up to 2500 mAU is specified for the supported VWD with an analytical flow cell installed.

If an autosampler is used when the PDA or PDA-100 detectors are tested, only 10 µL of sample will be injected by default. However, when qualifying the Thermo Fisher Scientific detectors using an autosampler, 15 µL of sample will be injected.

3.9 Injection Volume Linearity

3.9.1 Theory

Injection volume linearity is determined by making five injections of a standard using injection volumes of 10, 20, 30, 40 and 50 µL.

PDA/ VWD: A 60 ppm caffeine standard is injected when using absorbance detection at a wavelength of 272 nm. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

Conductivity Detector: A 50 ppm nitrate standard is injected when using conductivity detection. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

Electrochemical Detector: An 4 µM Threonine standard is injected when using integrated amperometry detection. 50 mM NaOH is used as the eluent at a flow rate of 0.25 mL/min. An Amino Acid disposable gold working electrode (Thermo Fisher Scientific P/N 060082) and a 2 mil x 1 mm Teflon gasket (Thermo Fisher Scientific P/N 060141) are used with the Amino Acids (pH, using Ag/AgCl Reference electrode) waveform.

This test is only performed on autosamplers with variable injection volume (e.g. AS or AS-AP) for analytical systems. A 250 µL syringe is required to be configured in the AS-AP to run the injection volume linearity test.

3.9.2 Performing the Test and Evaluating the Results

Injection volume linearity evaluation is the next test in the sequence queue and the sequence is called XQ_Detector_Inj_Linearity. The concentrations for the nitrate, caffeine and threonine standard and the injection volumes are included in the QNT file used for detector linearity.

3.10 Sample Preparation Precision (AS Only)

3.10.1 Theory

Sample Preparation precision (conductivity detection for IC systems; absorbance detection for analytical systems only; electrochemical detection is not supported) is determined by making three separate injections using a fixed injection volume. Each of the three standards is prepared individually using the autosampler's sample preparation option. The autosampler prepares the three samples by first transferring 15 µL of a 1000 ppm caffeine or nitrate standard into three empty sample vials followed by the addition of 1485 µL of water to each of the sample vials. Injections are then made from each vial. Deionized water is used as the eluent at a flow rate of 1.00 mL/min in

analytical systems. If an absorbance detector is used the wavelength is set to 272 nm. This test is only performed on autosamplers with the sample preparation option installed.

3.10.2 Performing the Test and Evaluating the Results

Sample prep precision evaluation is the next test in the sequence queue and the sequence is called XQ_Detector_Samp_Prep.

Required configuration for running a successful sample preparation experiment:

- AS configured with a 10.0 mL tray (P/N 026374).
- System set up with conductivity or an absorbance detector.

Precision is determined by calculating the relative standard deviation (RSD) of the peak areas from the three vials.

3.11 Gradient Accuracy

3.11.1 Theory

Gradient proportioning accuracy is determined by performing a step gradient using varying compositions of deionized water and either 0.3 % Acetone with absorbance detectors or 1 mM KCl for conductivity detectors. When performing the test with an absorbance detector, deionized water is used for eluents A and C and 0.3 % Acetone is used for eluents B and D with a wavelength setting of 254 nm. With conductivity detection eluents B and D are 1 mM KCl.

When performing the test with an electrochemical detector, 50 mM NaOH is used for eluents A and B and 50 mM NaOH with 8.3 μ M glucose is used for eluents B and D.

Gradient composition is measured at 0, 20, 50, 80, and 100 % of eluents B and D at a flow rate of 1.00 mL/min in analytical systems. Step noise (ripple) is measured for each step in the gradient. No injection is performed.

3.11.2 Performing the Test and Evaluating the Results

Gradient Accuracy evaluation is called XQ_GP_Grad_Accuracy.

The measured step height is compared to the theoretical step height and a percent deviation is calculated. Step noise is evaluated for all steps. Each step has a defined one-minute interval where noise is evaluated. Based on a method of least squares, Chromeleon calculates a regression line from the measured values of each interval. The noise is the distance between two parallel lines through the measured minimum and maximum values and the regression line.

3.12 Column/Compartment TC Temperature Qualification

3.12.1 Theory

The temperature of a column/compartment TC is qualified by measuring the temperature of the column/compartment with a calibrated thermometer after allowing the column/compartment and thermometer to equilibrate for a minimum of 15 minutes. The temperature is recorded from the thermometer.

3.12.2 Performing the Test and Evaluating the Results

During the sequence queue the column compartment is set to 30 °C and the Analytical DC compartment temperature is set to 20 °C. The Column Compartment Temperature Qualification is performed after the completion of the sequence queue. The recorded temperature is manually entered into the report.

3.13 Wavelength Accuracy of Absorbance Detectors

3.13.1 Theory

Wavelength accuracy is evaluated using a holmium oxide filter. Three holmium oxide absorbance maxima are compared with the theoretical maxima at 360.9 nm, 418.0 nm and 536.6 nm for ICS Series PDA and VWD detectors.

3.13.2 Performing the Test and Evaluating the Results

For AD25 and PDA-100 detectors the wavelength accuracy test is the first test in the sequence queue and is called XQ_Wavelength_Accuracy where, X is P for PQ and O for OQ. The Wavelength Accuracy Test program executes the verification and the results are logged to the audit trail by the Wavelength Accuracy Report program.

3.14 Sample Temperature Accuracy of Autosamplers (AS-AP only)

3.14.1 Theory

The sample temperature accuracy mainly depends on the cooling and heating accuracy of the AS-AP autosampler, the insulation of the sample compartment, and the thermal transfer from the carousel to the vial.

3.14.2 Performing the Test

The sample temperature accuracy is determined with the help of an external thermometer. The temperature sensor is placed in a standard vial (10 mL or 1.5 mL) filled with water. The carousel cover must be closed during the test. The autosampler temperature is set to a specified temperature. When the set temperature is reached, the sample (water) temperature is recorded over a period of 30 minutes. Within the 30 minutes, the sample temperature reaches a stable value. The program will keep the carousel from moving during the run. The temperature accuracy is the temperature difference between the sample temperature and the nominal autosampler temperature.

3.15 Sample pH Conductivity verification (AS-AP only)

3.15.1 Theory

The sample pH Conductivity qualification is evaluated by checking the pH calibration and pH accuracy. The sample temperature calibration is measuring the pH at 4, 7 and 10. The reading is stable if the reading and the three data points are within 0.2 pH unit of each other. The pH measurement of the accessory must be calibrated before you begin using the accessory and then periodically thereafter. During pH calibration, the pH of a pH 7 buffer solution is measured first and *the offset is determined*. Then, the pH of either a pH 4 or a pH 10 buffer solution is measured

and a slope between pH 7 and pH 4 (or pH 10) is determined. Optionally, the pH of a third buffer solution (pH 4 or pH 10) can be measured and a slope between pH 7 and the third buffer determined. The conductivity calibration procedure consists of determining the conductivity zero point of the accessory and then measuring the conductivity of a 1000 μ S standard. Chromeleon determines the calibration offset and slope and calculates a new cell constant.

4 Qualification Preparation

4.1 Materials in Kit

Before starting the qualification procedure confirm that all the materials needed are present. If any items from these tables are not present, do not continue with the qualification process until these items are available.

Table 6. Materials/Parts Description for OQ/PQ

Part Description	Part Number	Quantity
OQ/PQ User's Manual Part A Version 7.10 (this document)	065448 Part A	1
Tubing, 0.010" (0.25 mm) ID x 60" (150 cm) length black PEEK <ul style="list-style-type: none"> Connect pump to injection valve, backpressure tubing to detector, etc. 	042690	60" (152 cm)
Red PEEK Tubing 0.005" (0.125 mm) ID x 39" (99 cm) length <ul style="list-style-type: none"> Generate system backpressure and delay volume for detector and injector tests 	044211	39" (99 cm)
Yellow PEEK Tubing (0.003", 0.075 mm ID) <ul style="list-style-type: none"> Generate system backpressure and delay volume for detector and injector tests in Analytical Systems only 	049715	39" (99 cm)
25 µL Sample Loop <ul style="list-style-type: none"> AS-DV/Manual injection 	042857	1
100 µL Sample Loop <ul style="list-style-type: none"> AS-AP, AS 	042951	1
<ul style="list-style-type: none"> 3 – Way mixing tees Connect eluent lines and eluent bottles 	024314	2
High-pressure bolt, 10-32	074449	6
High-pressure ferrules, double cone, 10-32	074373	6
Qualification labels	050837	6
Certificate of Functional Performance	031423	6
NIST Traceable Caffeine Standards Kit <ul style="list-style-type: none"> Absorbance detectors 	060253	1
NIST Traceable Nitrate Standards Kit <ul style="list-style-type: none"> Conductivity detectors 	060254	1
Test Binder	035863	1
Restrictor Microbore, Gradient Test	049630	1

Table 7. OQ/PQ Test Kits Description

Part Description	Part Number
IC OQ/PQ Kit for IC systems (PROD,KIT,OQ/PQ,ICS,CD ABS)	063540
IC OQ/PQ Kit for ICS-5000 or ICS-3000 Analytical ED detectors (PROD,KIT,OQ/PQ,ICS,ED)	063541

4.2 Additional Required Materials

The following items will be needed:

Table 8. Additional Materials Needed

Description	Quantity
ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered) <ul style="list-style-type: none"> All eluent, standard, and sample preparation 	2 L
Reagent Grade or better Potassium Chloride for OQ/PQ gradient tests using conductivity detection <ul style="list-style-type: none"> Gradient pump tests using conductivity detectors 	1 g
HPLC grade Acetone for OQ/PQ gradient tests using optical absorbance detection <ul style="list-style-type: none"> Gradient pump tests using optical detectors 	3 mL
Sodium Hydroxide 50/50 w/w (Low Carbonate) (Thermo Fisher Scientific P/N 033465) <ul style="list-style-type: none"> Prepare eluent for Integrated Amperometry qualification 	~ 5.2 mL
NIST Traceable Threonine Standards Kit for OQ/PQ (P/N 063541) <ul style="list-style-type: none"> Integrated Amperometry qualification on an Analytical ICS-5000* 	1
Disposable Amino Acid AAA Working Electrode (Thermo Fisher Scientific P/N 060082) <ul style="list-style-type: none"> Integrated Amperometry qualification on an ICS-5000* 	1
CarboPac PA1 guard 2 mm (Thermo Fisher Scientific P/N 057179) <ul style="list-style-type: none"> Integrated Amperometry qualification on an analytical ICS-5000* 	1
Glucose Sigma-Aldrich P/N G8644 in ED Analytical OQPQ kits only (P/N 080150)	1
2 mil x 1 mm Teflon Gasket (P/N 060141)	1
Ammeter or digital volt multimeter (DVM) with milliamp (300 mA scale) setting for OQ/PQ tests using an EG. Must be capable of displaying 0.00 to at least 220 mA. A Fluke Model 189, 289 or equivalent DVM will allow for automated EG Current testing.	1
Digital thermometer. Temperature accuracy test can be performed with a Dostmann P600 Thermometer or equivalent. The P600 is included in the Column Thermostat PQ Kit (P/N 5705.0050A)	1
Type K temperature sensor for Dostmann P600 Thermometer (P/N 6820.0010)	1
Thermo Fisher Scientific analytical conductivity cell (detector) <ul style="list-style-type: none"> Detector, injector and gradient pump tests using conductivity detectors in conductivity mode 	1
AS or AS-AP in pull mode: 10 mL vials are required for AS-AP pull mode and AS Sample Preparation Qualification	10
AS-AP: 1.5 mL vial tray (P/N 066332)	1
AS-AP: 1.5 mL vials (P/N 061696)	7
Black tubing 48" (P/N 042690)	1
AS-DV 5.0 mL vials with filter caps (P/N 038141) if qualifying an AS-DV autosampler	20

100 mL volumetric flasks (Class A, 100 ± 0.08 mL) <ul style="list-style-type: none"> • <i>Standard preparation for AS-DV and Manual Injection</i> 	5
1000 mL volumetric flasks (Class A, 1000 ± 0.3 mL) <ul style="list-style-type: none"> • <i>Eluent preparation</i> 	At least 1
Transfer Pipettes or Pipettor	0.5 – 10.0 mL
Calibrated Analytical Balance - capable of mass recording to the nearest milligram or better <ul style="list-style-type: none"> • <i>Weigh KCl for eluent preparation</i> • <i>Weigh eluent for the Flow Rate Accuracy and Precision test</i> 	1
Calibration and Check Standard pH 4 (P/N 033238)	1
Calibration and Check Standard pH10 (P/N 033240)	1
1000 uS Standard (P/N 080172)	1
Computer workstation running Chromeleon™ Version 7.10 (with SR1) or later (later versions required for some configurations, see Chapter 1.6)	1

*** These items are needed for ICS-3000 system qualification in ED mode. These items are included in the ICS-3000 ED Qualification kit (Thermo Fisher Scientific P/N 063541)**

Deionized water should be vacuum filtered through a 0.2 or 0.45 µm nylon or other water compatible membrane exhibiting very low extractable.

All volumetric ware, pipettes and standard eluent storage bottles should be made of low extractable polymeric materials such as polypropylene or polystyrene.

A 48" black tubing (P/N 042690) is recommended to be added to the yellow tubing (P/N 049715) from the injection vale to the conductivity detector to reduce pump ripple and cell noise.

4.3 Preparation of Standards

Two standards are provided in the OQ/PQ kit. Use the Caffeine Kit, P/N 060253 to qualify systems with an absorbance detector (AD25, ICS Series VWD or PDA or PDA-100). Use the Nitrate kit, Part Number 060254 to qualify systems with a conductivity detector.

To qualify an ICS-3000 system with an ED detector in Integrated Amperometry mode use the Threonine standard kit for ED qualification (Thermo Fisher Scientific P/N 063542). ED detector qualification does not support AS40 and AS-DV autosamplers.

4.3.1 Caffeine standard preparation for the Injector Precision test (AS-DV and AS40 autosamplers only)

(AS-DV and AS40 Autosamplers only) For the Injector Precision test prepare a 40 ppm caffeine standard using the 1000 ppm caffeine standard vials included in the Caffeine Standards Kit. Pipette 4.0 mL of the 1000 ppm caffeine standard into a 100 mL volumetric flask. Fill the flask to the mark with ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered).

(AS-DV and AS40 Autosamplers and Manual Injection) For the Carryover test use the 1000 ppm caffeine standard included in the Caffeine Standards Kit.

4.3.2 Nitrate standard preparation for the Injector Precision test (AS-DV and AS40 autosamplers only)

For the Injector Precision test prepare a 50 ppm nitrate standard using the 1000 ppm nitrate standard ampoule included in the Nitrate Standards Kit. Pipette 5.0 mL of the 1000 ppm nitrate standard into a 100 mL volumetric flask. Fill the flask to the mark with ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered).

For the Carryover test use the included 1000 ppm nitrate standard found the in the nitrate standards kit without dilution.

4.4 Preparation of Eluents

All eluents must be prepared using ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered). Eluent should be freshly prepared prior to the execution of the OQ/PQ tests.

4.4.1 Preparation of Eluents for use with Conductivity Detection

Prepare the following eluents and connect them to lines A, B, C and D of the pump. All eluents must be degassed. Eluent lines A and C can be connected to one eluent bottle containing 1 L of water by a mixing tee. Eluent lines B and D can be connected to one eluent bottle containing 1 L of 1 mM KCl by a mixing tee.

Note: Eluents B, C and D are needed for the Gradient Pump Accuracy Test only and should be prepared only if the pump configured has gradient capabilities.

Table 9. Eluent Channels for Gradient Performance Test with Conductivity Detector

Eluent	Description
A	Deionized water
B	1.0 mM KCl

C	Deionized water
D	1.0 mM KCl

10 mM KCl Stock Solution Preparation:

Prepare one liter of 10 mM KCl by weighing 0.746 g of KCl. Place the KCl in a 1 L volumetric flask. Fill the flask to the mark with ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered).

1 mM KCl Preparation:

Pipette 100 mL of the 10 mM KCl solution into another 1 L volumetric flask, filling the flask to the mark with ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered). The 1 mM KCl solution should be fresh prepared fresh daily.

Remember to ...

- Degas all eluents and autosampler wash solutions. Degassing is not required, but is recommended for most consistent results.
- Connect eluent lines A and C to one eluent bottle containing 1 L of deionized water equipped with a 3-way mixing tee.
- Connect eluent lines B and D to one eluent bottle containing 1 L of 1 mM KCl equipped with a 3-way mixing tee.
- Eluent bottles for ICS-3000 pumps do not need to be pressurized.
- Ensure that the rear seal wash supply bottle is filled with fresh deionized water or the rear seal wash pump is turned off from the F8 command window.

4.4.2 Preparation of Eluents for use with Absorbance Detection

These eluents are for used for qualification with an ICS Series Variable Wavelength Detector (VWD) or Photodiode Array Detector(PDA), AD25 Absorbance Detector or PDA-100 Photodiode Array Detector.

Prepare the following eluents and connect them to lines A, B, C and D of the pump. All eluents must be degassed. Eluent lines A and C can be inserted into one eluent bottle containing 1 L of water. Eluent lines B and D can be inserted into one bottle containing 1 L of 0.3 % Acetone in Water.

Note: Eluents B, C and D are needed for the Gradient Pump Accuracy Test only and should be prepared only if the pump configured has gradient capabilities. Do not prepare these eluents if the pump configured is an ICS-3000 Isocratic pump since it does not have gradient mixing capabilities.

Table 10. Eluent Channels for Gradient Performance Test with Conductivity Detector

Eluent	Description
A	Deionized water
B	1.0 mM KCl
C	Deionized water
D	1.0 mM KCl

0.3 % Acetone in Water Preparation:

Pipette 3 mL of HPLC grade Acetone into a 1 L volumetric flask. Fill flask to the mark with ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered) . Eluents should be freshly prepared on a daily basis.

Remember to ...

- Degas all eluents.
- Connect eluent lines A and C to one eluent bottle containing 1 L of deionized water using a 3-way mixing tee.
- Connect eluent lines B and D to one eluent bottle containing 1 L of 0.3 % Acetone in water using a 3-way mixing tee.
- Eluent bottles on ICS-3000 pumps do not need to be pressurized.

4.4.3 Preparation of Eluent for use with Integrated Amperometry Detection

There are two different eluents used for qualification with an ICS-3000 Electrochemical Detector in Integrated Amperometry mode.

For Flow Test (initial Flow test):

Eluent	Description
A	Deionized water

For all other ED Tests :

Eluent	Description
A	50 mM NaOH

The preparation of 50 mM NaOH for the ED tests should be performed after the Pump Flow Rate test has been completed to ensure minimal potential contamination of 50 mM NaOH eluent. Do not make the 50 mM NaOH eluent until section 5.3.2. All eluents must be degassed. Eluent lines A should be connected to one eluent bottle containing 1 L of 50 mM NaOH. After the 50 mM NaOH has been connected to line A, the line should be thoroughly primed.

50 mM NaOH Preparation:

Fill eluent bottle with 997.4 mL of ASTM Type I (or better) deionized, degassed water (18.0 Megohm-cm, filtered). Pipette 2.6 mL of 50/50 (w/w) NaOH solution into the bottle. Cap, cover with Helium (or Nitrogen) head pressure and mix. The 50 mM NaOH solution should be prepared daily and care should be taken to minimize exposure to CO₂ gas.

Remember to ...

- Degas all eluents.
- Connect eluent line A to one eluent bottle containing 1 L of 50 mM NaOH.
- Pressurize eluent bottles with Helium (or Nitrogen) gas.

4.4.4 Preparation of Eluent for use in Gradient Performance Test with Integrated Amperometry Detection

Prepare the following eluents and connect them to lines A, B, C and D of the pump. All eluents must be degassed. Eluent lines A and C can be connected to one eluent bottle containing 50 mM NaOH by a mixing tee. Eluent lines B and D can be connected to one eluent bottle containing 50 mM NaOH with 8.3 μ M Glucose by a mixing tee.

Note: Eluents B, C and D are needed for the Gradient Pump Accuracy Test only and should be prepared only if the pump configured has gradient capabilities.

Table 11. Eluent Channels for Gradient Performance Test with Electrochemical Detector

Eluent	Description
A	50 mM NaOH
B	8.3 μ M Glucose in 50 mM NaOH
C	50 mM NaOH
D	8.3 μ M Glucose in 50 mM NaOH

50 mM NaOH Preparation:

Fill eluent bottle with 994.8 mL of ASTM Type I (or better) deionized, degassed water (18.0 Megohm-cm, filtered). Pipette 2.6 mL of 50/50 (w/w) NaOH solution into the bottle. Cap, cover with Helium (or Nitrogen) head pressure and mix. The 50 mM NaOH solution should be prepared daily and care should be taken to minimize exposure to CO₂ gas.

8.3 μ M Glucose in 50 mM NaOH Preparation:

Fill eluent bottle with 1000 mL of previously prepared 50 mM NaOH. Pipette 15 μ L of 555 mM Glucose, cover with Helium (or Nitrogen) head pressure and mix. The 50 mM NaOH solution should be prepared daily and care should be taken to minimize exposure to CO₂ gas.

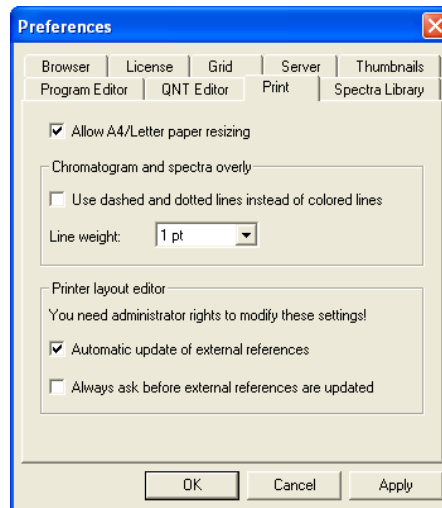
Remember to ...

- Degas all eluents.
- Connect eluent lines A and C to one eluent bottle containing 1 L of 50 mM NaOH using a 3-way mixing tee.
- Connect eluent lines B and D to one eluent bottle containing 1 L of 50 mM with 8.3 μ M glucose using a 3-way mixing tee.
- Pressurize eluent bottles with Helium (or Nitrogen) gas.
- Use the disposable carbohydrate working electrode (P/N 060082).

4.5 Preparing the IC System

Note: The AD25, VWD, PDA, and PDA-100 Detector UV and visible lamps must be turned on at least two hours prior to performing the qualification.

1. Chromeleon settings: Ensure that Chromeleon is set for Automatic update of external references. Click on the “Automatic update of external references” check box in File → Preferences → Print menu.



2. Open the Chromeleon Instrument Configuration application. Only one pump, one injection valve and one detector can be qualified concurrently (except the RF2000 which must be qualified in series with a conductivity detector), so if more than one pump, one injection valve or one detector are installed, the second pump (Pump_2), the second detector (CDet_2, EDet_2) will be ignored if both detectors are the same. The detector names can be configured in the Instrument Configuration. If the detectors are different, uncheck the detector signal that will not be qualified. If the second pump or detector is to be qualified, remove (or uncheck) the first pump or detector in the Instruments. Make note of any changes made to the system for the purposes of this qualification. After qualification these changes should be reversed. It is helpful to save a backup file of the configuration before you make any changes for qualification.
3. ELUENT GENERATOR: Select the **General** page and confirm the Device Name is “EluentGenerator”. On the Pump Link & Cartridge page confirm the eluent generator is linked to the pump to be qualified and the Offset Volume is set to no more than 400.0 µL.
4. AUTOSAMPLER: Double click on the Autosampler to be qualified (if installed) to view the properties page for the autosampler. Select the **General** page and confirm the Device Name is “Sampler”. Select the **Options** tab and confirm that the options match the configuration of your system. The AS-AP injection mode must be selected at this point. Only one mode is qualified. Select the **Devices** tab and confirm the Autosampler diverter valve status matches your system. Select the **Temperature** page and confirm the Autosampler controlled Column Temperature option matches your system (if installed). Exit the properties page. For the AS-AP autosampler in pull mode, the Flush Volume and Flush Volume2 must be set with F8 commands or from the **More Options** page of the AS-AP panel before beginning qualification. Flush Volume = 2 x (Needle Volume + Needle Line Tubing Volume) µL where Needle Volume = 30 µL and Needle Line Tubing Volume (the volume of tubing between the needle and the injection port) = 0.86 µL/cm (2.2 µL/in) of 0.33-mm (0.013-in) ID PEEK tubing (blue). Set the Flush Volume2 parameter to 30 µL.

NOTE: Make sure inject mode in the panel is consistent with the inject mode in the Instrument Configuration before loading the OQPQ templates.
5. ICS-3000 Pump: Double click on the pump to be qualified to view the properties page for the pump. Select the **Devices** tab and confirm the Device Name is “Pump_1” or “Pump_2”.

6. ICS-3000 DC: Double click on the DC to be qualified to view the general properties. Select the Thermal Controls tab and confirm the heating configuration of the Instruments. Confirm that the device name for the Column TC is "Column_TC" and the device name for the Compartment TC is "Compartment_TC" if installed on the Instruments.
7. DETECTOR: Double click on the detector to be qualified to view the properties page for that detector (for CD and ED this will be in the DC under the detector tab). Select the **General** page and confirm the Device Name is "UV" for AD25 and PDA-100, detectors. For the ED and CD detectors: Open the properties of the DC and click on the **Detectors** tab. Highlight the detector name and click on **Configuration** button. Confirm that the Device Name is "CDet1" for CD detectors or "EDet1" for ED detectors. If both detectors are on the same Instruments, uncheck the detector(s) that is not being qualified (only one detector can be qualified at a time). Click on the **Signals** tab. Confirm the signal name is "CD_1" for CD detectors or "ED_1" for ED detectors. Confirm that the signal factor for absorbance, conductivity, and fluorescence and integrated amperometry detectors is 1.000. For integrated amperometry detectors confirm the signal factor is 0.001. If it is not, change the factor to the correct value for the qualification. Confirm the signal unit is μS for conductivity or nC integrated amperometry respectively. Confirm the signal unit is AU for UV detectors. Confirm the signal unit is mV for fluorescence detectors. If it is not, change the unit to the correct value for the qualification. Exit the properties page.
8. If applicable make sure the detector Output is in 'Offset' mode in the Analog Out Set-Up screen.
9. Make note of any changes and save the configuration.
10. All modules, with the exception of the AD25, RF2000, AS-DV, AS40, must have their serial number set in the moduleware. Qualification of modules without serial numbers, with the exception of the above-mentioned modules, is not supported.
11. Connect Eluents A, B, C and D to the pump and prime each eluent line individually for approximately 3 minutes each, longer if the vacuum degas option is installed (10 minutes).
12. Use the backpressure coil to connect the pump outlet directly to port 2 on the injection valve
Connect the pump line to injection valve port 2 (P). Bypass the column, suppressor, CR-TC, and CRD from the system leaving the gradient mixer in-line; If an Eluent Generator Cartridge is in-line with the IC system, bypass all plumbing to the EGC and connect the pump directly to the injection valve.
 - a. Diagram on how to connect the pump outlet to the valve

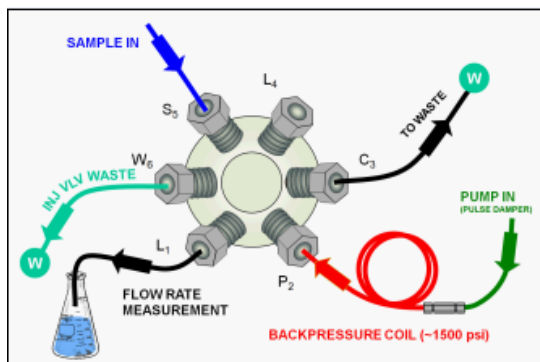


Figure 4. Plumbing diagram, initial equilibration step

13. ICS-3000 DC COLUMN TC: Insert the thermometer probe into the 2nd tubing slot from the bottom of the DC so that the LCD display sits out the side of the column TC. The LCD display of the thermometer should be visible and display the current Column TC temperature.
14. ICS-3000 DC COMPARTMENT TC: Insert the thermometer probe into top tubing slot on the DC so that the LCD display sits out the side of the column TC. The LCD display of the thermometer should be visible and display the current Column TC temperature.
15. ICS-3000 TC: Insert the thermometer probe into the side tubing slot on the TC so that the LCD display sits out the side of the TC. The LCD display of the thermometer should be visible and display the current TC temperature.
16. RF-2000 DETECTOR: Make sure the fluorescence detector is plumbed in series with a conductivity detector. The RF-2000 detector will be qualified during the other tests. The tests will not be performed correctly if the RF-2000 detector is the only detector found on the system.
17. Thoroughly rinse the autosampler and manual injector fluidics with water. This can be accomplished on the AS and AS-AP by flushing the sample syringe six times and on the AS-DV and the AS40 by running two or three rinse 5.0 mL vials
18. AS-AP or AS: For ICS-3000 Analytical systems using an AS-AP or AS Autosampler, install the 100 µL sample loop on the injection valve. Loop volume must be set in the instrument configuration for the AS-AP.

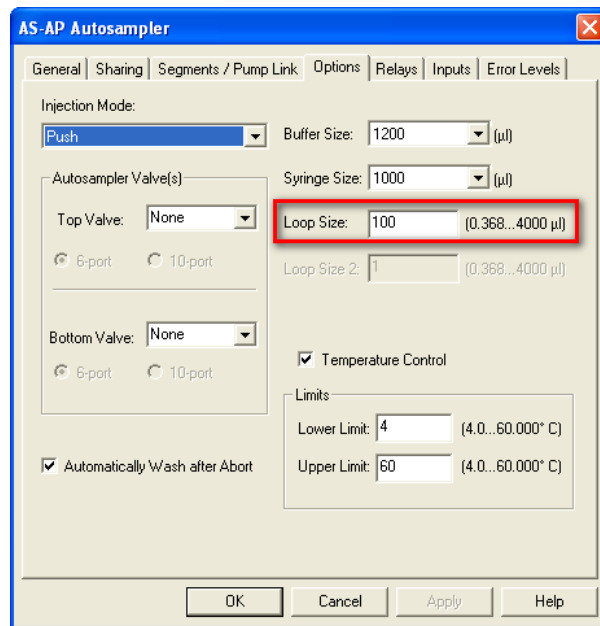


Figure 5. AS-AP Instrument Configuration

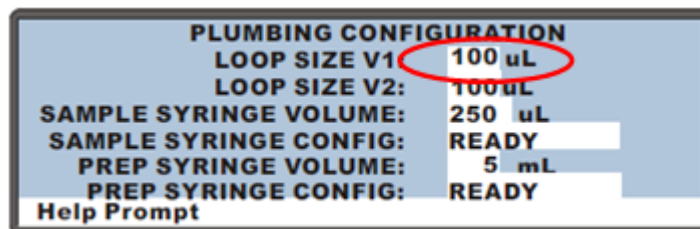


Figure 6. AS Autosampler LCD Control Panel Display

Ensure that the Loop SIZE V1 volume is changed to 25 μ L

19. AS-DV/AS40 or Manual Injector: For systems using an AS-DV or an AS40 Autosampler or manual injector, install the 25 μ L sample loop on the injection valve. Connect the AS40 **Load** connector to **Relay_1** on integrated systems or to **Relay_1** on the pump for modular systems.

Note: All programs using the AS-DV and the AS40 are designed to run with the 5.0 mL vials (*i.e.*, trigger relay at -2.40 minutes). If 0.5 mL vials are to be used programs will have to be manually modified to trigger the relay at -1.20 minutes).

If using 5.0 mL vials, 1 Nitrate standards kit (P/N 063541) is needed to complete the OQ or PQ.

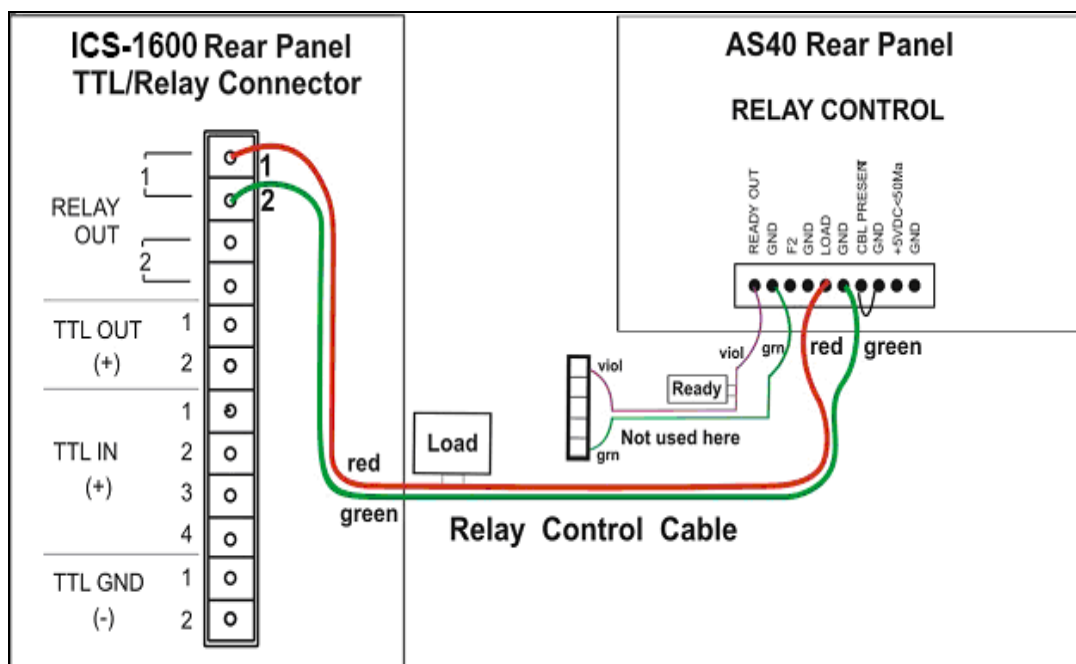


Figure 7. Remote Control Cabling to AS40 Autosampler

20. Select 100% of Eluent A (water) and a flow rate of 1.00 mL/min. Turn the pump flow on and let the system equilibrate for a minimum of 15 minutes.
21. NEW TUBING: When running a qualification on a system with new tubing, such as a new backpressure coil, new sample loop or a complete new system, it is recommended to rinse the tubing with carbonate eluent before proceeding with the qualification. This can be achieved by injecting two or three times with 0.5 M Sodium Carbonate solution while running deionized water at 1.00 mL/min, or by running 9 mM Sodium Carbonate as the eluent for 10 minutes before rinsing with deionized water for 20 minutes. The latter is recommended for new systems being qualified for the first time.

When running a qualification on a system configured with electrochemical detection, it is recommended to rinse the system with 1.0 M hydroxide before proceeding with the

qualification. To prepare 1.0 M hydroxide, prepare from a 50/50 w/w sodium hydroxide solution (never from hydroxide pellets), by pipetting 52 mL of sodium hydroxide in water to make 1 L of solution.

22. For the AS-AP autosampler pipette the standards from the standards kit vials directly into 1.5 mL or 10.0 mL autosampler vials and place them in the position/order according to table 14 below. For the AS-DV autosampler, pipette the standards from the standards kit vials directly into autosampler vials and place them in the position/order according to Table 12 below. For AS, place the vials in the tray, as per Table 11. For the Injector Precision and Carryover standards transfer the Injector Precision standards prepared in Section 4.3 into 10 autosampler vials and the Carryover standard into one autosampler vial. To qualify an IC system without an autosampler, use the AS-DV table for the injection order and inject the samples from the standards kit ampoules manually.
23. To qualify an IC system without an autosampler, use the AS-DV/AS40 table for the injection order and inject the samples from the standards kit ampoules manually.
24. ICS-3000 INTEGRATED AMPEROMETRY: Cut the label off the vials (included in the ICS-3000 ED qualification kit) and place the vial directly into autosampler in the position order according to the table below. Note that even though the main portion of the label has been cut off, the vial is still properly loaded. For the AS-AP (push mode only) or AS autosampler this will require the 1.5 mL AP-AP tray (P/N 074936) or AS autosampler tray (P/N 062481).
25. ICS-3000 ELUENT GENERATOR: Double click on the eluent generator to be qualified to view the properties page for the eluent generator. Select the **General** page and confirm the Device Name is "EluentGenerator". On the "cartridge" tab confirm that the eluent generator is linked to the pump to be qualified. Confirm that there is a serial number for a KOH or a MSA cartridge. If a serial number is not available for the KOH cartridge, contact Thermo Fisher Scientific Technical Support. **Connect the EG test box with the ammeter to the EGC_1 or EGC_2 connector.**
26. For systems using an ICS-3000 EG:
 - a. Disconnect the Eluent Generator Cartridge cable from the Eluent Generator EGC connector.
 - b. If installed, disconnect the CR-TC cable from the CR-TC connector.
 - c. Turn on the ammeter or multimeter and set the display to DC mA. This test requires mA readings from 0.00 to at least 220 mA.
 - d. Connect the positive lead of the ammeter to the red post connector labeled "Amps" on the IC Qualification Test Box (P/N 072005)
 - e. Connect the negative lead of the ammeter to the black post connector labeled "COMMON" on the IC Qualification Test Box.

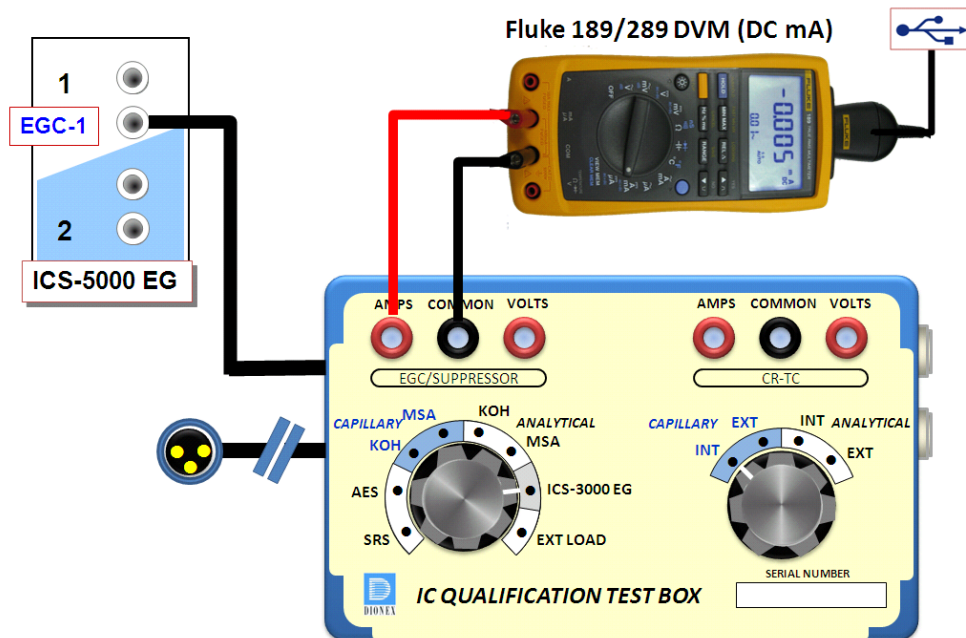


Figure 8. ICS-3000 EG Current Test Setup

- f. From the console in the Thermo Fisher Scientific Chromatography Management System, go to the navigation pane and select the pump_EluGen_detector panel appropriate for the system being qualified. Go to Control and select the instruments configured with the Eluent Generator to be qualified (for Chromeleon 6.7+, click on the Default Panel Tab set).
- g. From the Eluent Generator section of the panel, select the cartridge type to be used. The panel will recognize previously configured KOH cartridges even though the IC Qualification Test Box is plugged into the cartridge connector. Do not switch the test box settings during operation as this can damage the test box.

27. FLUKE 189/289 MULTIMETER:

- a. Install the Fluke 189/289 multimeter or equivalent into the instruments to be qualified.
- b. The Chromeleon device driver for the Fluke 189/289 multimeter is included with Chromeleon 7.1 (Fluke 189, 289).
- c. Connect the Fluke 189/289 multimeter to the IC Qualification Test Box (P/N 072005) as described in the **important notes** of this section
- d. The Fluke 189/289 multimeter is only used for systems that will enable automated downloading of EG current qualification results into the validation reports.
- e. To correctly configure the Fluke 189/289 device driver:
 - i. Connect the Fluke USB/IR (Rev II or greater) cable to the computer USB port; Windows XP will automatically detect and install the USB driver.
 - ii. Verify through the Windows Hardware Device Manager for the actual COM port assignment.

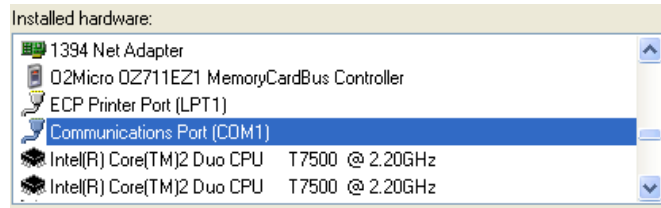


Figure 9. COM Port Assignment

- iii. Configure the Fluke 189 or 289 Chromeleon Device Driver in the instruments, save the instrument configuration file and exit then shut down the Chromeleon Server.
- iv. Restart the Chromeleon Server and open the Fluke 189/289 CM device driver.
- v. Select the COM port from the COM drop down selection to match the COM number assigned in the Windows Hardware Device Manager above then save and exit from the Chromeleon Server.
- f. It is recommended that the Power off time out (PrOff) for the Fluke189/289 be set to a value greater than the time it will take to perform the Operational Qualification to avoid inadvertent shutdown of the meter which will terminate the OQ or PQ sequence.
- g. Ensure that the meter is set to record in 'DC mA' (instead of 'AC mA').
- h. See Fluke 189/289 operator's manual for instructions on these settings.

IMPORTANT NOTES:

- a. It is important that the Fluke 189/289 multimeter is installed AND connected before loading the OQ/PQ templates.
- b. Follow the instructions in the Chromeleon 7.1 help topics for the installation of the Fluke 189/289 multimeter Windows USB driver and for the Chromeleon device driver for the Fluke 189/289 multimeter and the Fluke 189 defaults to DC mA.
- c. Follow the Fluke 189/289 multimeter instructions for adjusting the default 'PrOFF' setting in the meter. It is recommended to adjust this parameter to a longer time (e.g., several hours or more) to avoid having the meter auto power off during your OQ/PQ test. In the event that the meter is powered off during the qualification testing the queue sequences will be aborted.

Table 12. AS Autosampler Sample Vial Order

Conductivity Detector with AS	Absorbance or Photodiode Array Detector with AS	ICS-3000 Integrated Amperometry Detector with AS	Vial Tray Position for AS
Water	Water	Water	1
5 ppm Nitrate Std			2
10 ppm Nitrate Std			3
25 ppm Nitrate Std			4
50 ppm Nitrate Std			5
100 ppm Nitrate Std			6
1000 ppm Nitrate Std			7
Empty Vial*			8
Empty Vial*			9
Empty Vial*			10
	15 ppm Caffeine Std		11
	40 ppm Caffeine Std		12
	60 ppm Caffeine Std		13
	80 ppm Caffeine Std		14
	100 ppm Caffeine Std		15
	1000 ppm Caffeine Std		16
	Empty Vial*		17
	Empty Vial*		18
	Empty Vial*		19
	----		20
	----	1 µM Threonine Std	21
	----	2 µM Threonine Std	22
	----	4 µM Threonine Std	23
	----	8 µM Threonine Std	24
	----	10 µM Threonine Std	25
	----	10 µM Threonine Std	26

- Vials only need to be present if the detector listed at the top of the table is to be qualified.
- Users need to reconnect the detectors when using the absorbance detectors.
- Don't forget to add the water vial in position 1 for the ED qualification test.

Table 13. AS-DV/AS40 Autosampler or Manual Injection Sample Order (Conductivity or Absorbance Only)

Qualification	Conductivity Detector with AS-DV/AS40	Sample Order on AS-DV	Absorbance or Photodiode Array Detector with AS-DV/AS40	Sample Order on AS-DV
Warm Up	Water	Vial 1	Water	None
Injector Precision	50 ppm Nitrate Std*	Vial 2	40 ppm Caffeine Std*	Vial 20
	50 ppm Nitrate Std*	Vial 3	40 ppm Caffeine Std*	Vial 21
	50 ppm Nitrate Std*	Vial 4	40 ppm Caffeine Std*	Vial 22
	50 ppm Nitrate Std*	Vial 5	40 ppm Caffeine Std*	Vial 23
	50 ppm Nitrate Std*	Vial 6	40 ppm Caffeine Std*	Vial 24
	50 ppm Nitrate Std*	Vial 7	40 ppm Caffeine Std*	Vial 25
	50 ppm Nitrate Std*	Vial 8	40 ppm Caffeine Std*	Vial 26
	50 ppm Nitrate Std*	Vial 9	40 ppm Caffeine Std*	Vial 27
	50 ppm Nitrate Std*	Vial 10	40 ppm Caffeine Std*	Vial 28
	50 ppm Nitrate Std*	Vial 11	40 ppm Caffeine Std*	Vial 29
Carry Over	1000 ppm Nitrate Std	Vial 12	80 ppm Caffeine Std*	Vial 30
	Water	Vial 13	Water	Vial 31
	Water	Vial 14	Water	Vial 32
Detector Linearity	5 ppm Nitrate Std	Vial 15	15 ppm Caffeine Std	Vial 33
	10 ppm Nitrate Std	Vial 16	40 ppm Caffeine Std	Vial 34
	25 ppm Nitrate Std	Vial 17	60 ppm Caffeine Std	Vial 35
	50 ppm Nitrate Std	Vial 18	80 ppm Caffeine Std	Vial 36
	100 ppm Nitrate Std	Vial 19	100 ppm Caffeine Std	Vial 37

* AS-DV/AS40 users: Do not use the standard from the standards kit for the Injector Precision and Absorbance carryover samples; instead, use the standards prepared in Section 4.3 of this manual.

* AS-DV/AS40 users: ICS-3000 Integrated Amperometry is not supported.

Table 14. AS-AP Autosampler Sample Order

Conductivity Detector with AS-AP Push/Pull Mode	Absorbance Detector with AS-AP Push/Pull Mode	Electrochemical Detector with AS-AP Push Mode	Vial Tray Position
Water	Water		BA1
5 ppm Nitrate Std	15 ppm Caffeine Std		BA2
10 ppm Nitrate Std	40 ppm Caffeine Std		BA3
25 ppm Nitrate Std	60 ppm Caffeine Std		BA4
50 ppm Nitrate Std	80 ppm Caffeine Std		BA5
100 ppm Nitrate Std	100 ppm Caffeine Std		BB1
1000 ppm Nitrate Std	1000 ppm Caffeine Std		BB2
		Water	RA1
		1 µM Threonine Std	RA2
		2 µM Threonine Std	RA3
		4 µM Threonine Std	RA4
		8 µM Threonine Std	RA5
		10 µM Threonine Std	RA6
		10 µM Threonine Std	RA7

Note: AS-AP pull mode requires the use of 10 mL vials. Qualification with ED detection is not supported with AS-AP pull mode.

Note: To run sequences with 1.5 mL vials, it is important to set the puncture offset to 3 mm in the commands before you start. The puncture offset commands allows the AS-AP to realign the x-axis of the 1.5m/10 mL vial setting.

4.6 OQ/PQ Sequence Installation

For performing OQ and PQ checks in Chromeleon 7.1, it is not required to create and copy the sequence templates from a Chromeleon CD. An Instrument Qualification Wizard automatically performs these steps. The wizard creates the sequences to be run. No instrument-specific sequence templates are created.

If using an AS autosampler configured for sequential injections, manually put the diverter valve in the correct position for the instrument.

Tip: Please note that Chromeleon 7 terminology is different from the terminology used in Chromeleon 6.80. For details, refer to the 'Glossary - Chromeleon 7,' which is available in the Documents folder of your Chromeleon 7 installation.

Go to Tools → Instrument Qualification to open Instrument Qualification Wizard. For this step, Chromeleon 7.1 must display the instrument pane with the system to be tested selected (Figure 9) otherwise the error “Instrument not connected” will occur.

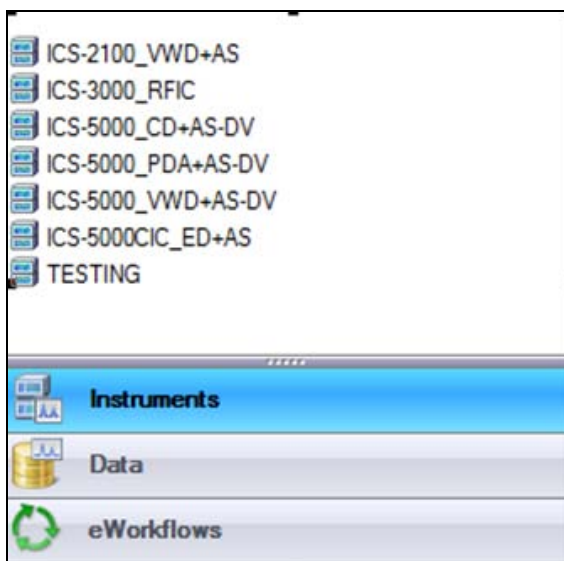


Figure 10. Instrument Pane for Chromeleon 7.1

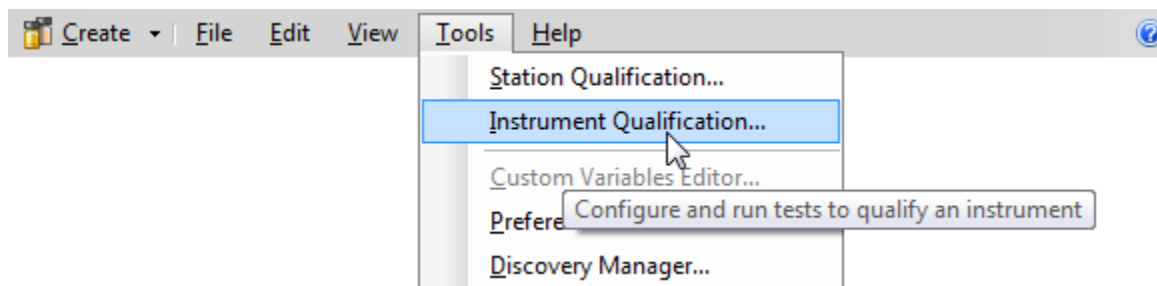


Figure 11. Starting the Instrument Qualification Wizard for Chromeleon 7.1

Select the qualification type: Installation (qualification of the installation), Operational (qualification in the working environment), or Performance Qualification (qualification during routine operation)

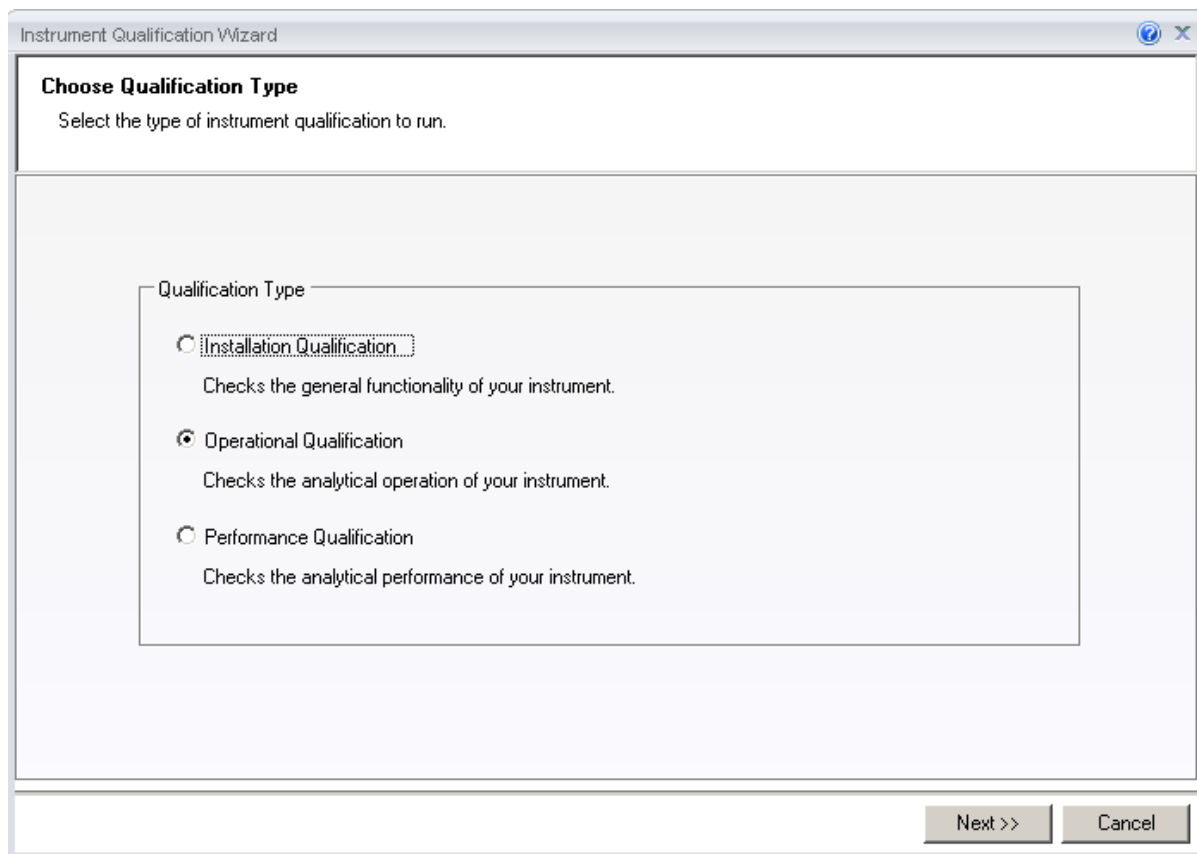


Figure 12. Selecting the qualification type for Chromeleon 7.1

Select the instrument to qualify.

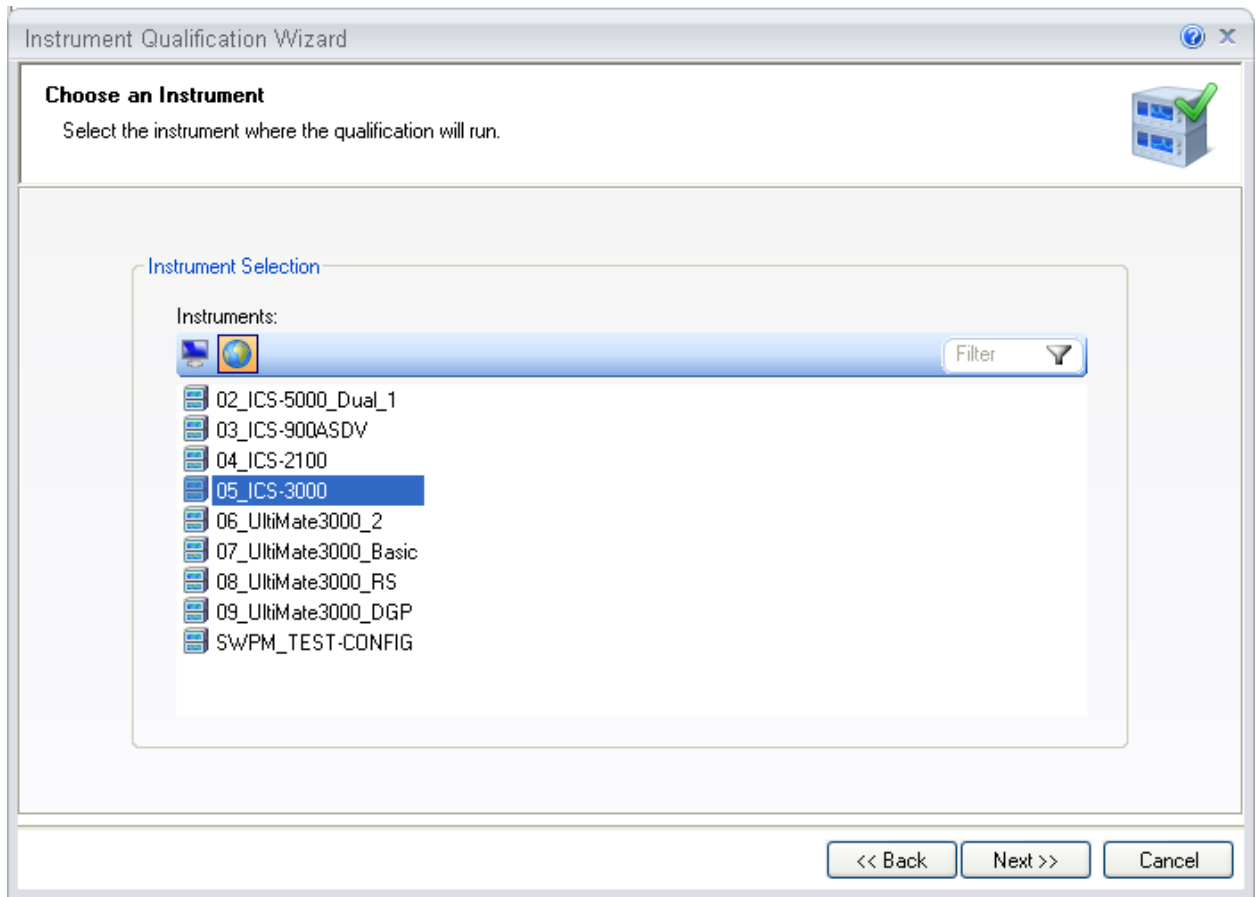
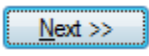


Figure 13. Selecting the instrument

Click  to connect the selected instrument to Chromeleon.

A list of sequences (tests) is displayed. The list is adapted to the instrument configuration of the selected instrument as defined in the Chromeleon Instrument Configuration Manager.

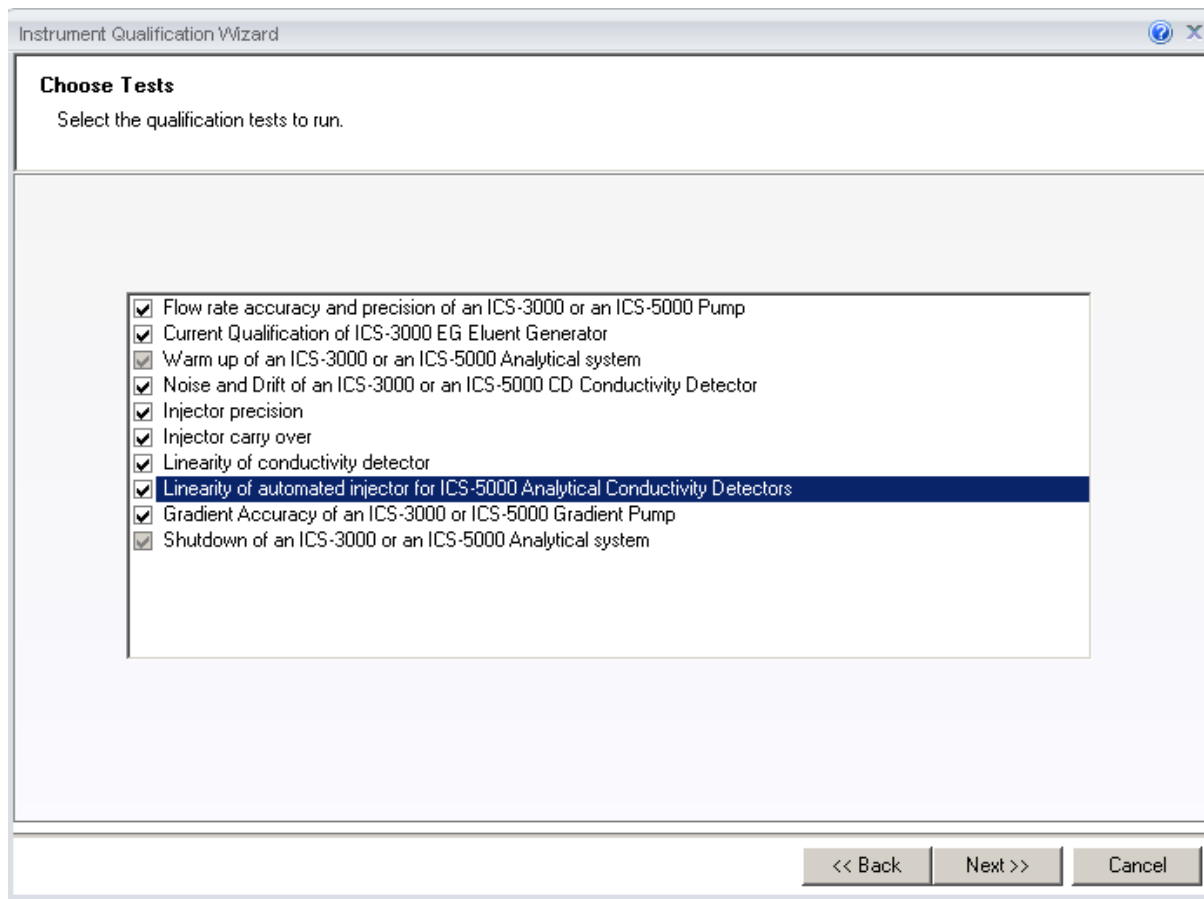


Figure 14. List of sequences for the selected instrument

Select the sequences that needed for the tests to be performed. Mandatory tests, such as "**Warm up**", are shown in the list, but the selection cannot be changed.

On the last wizard page, select a unique name under which the OQ and/or PQ sequence directory for this instrument is saved.

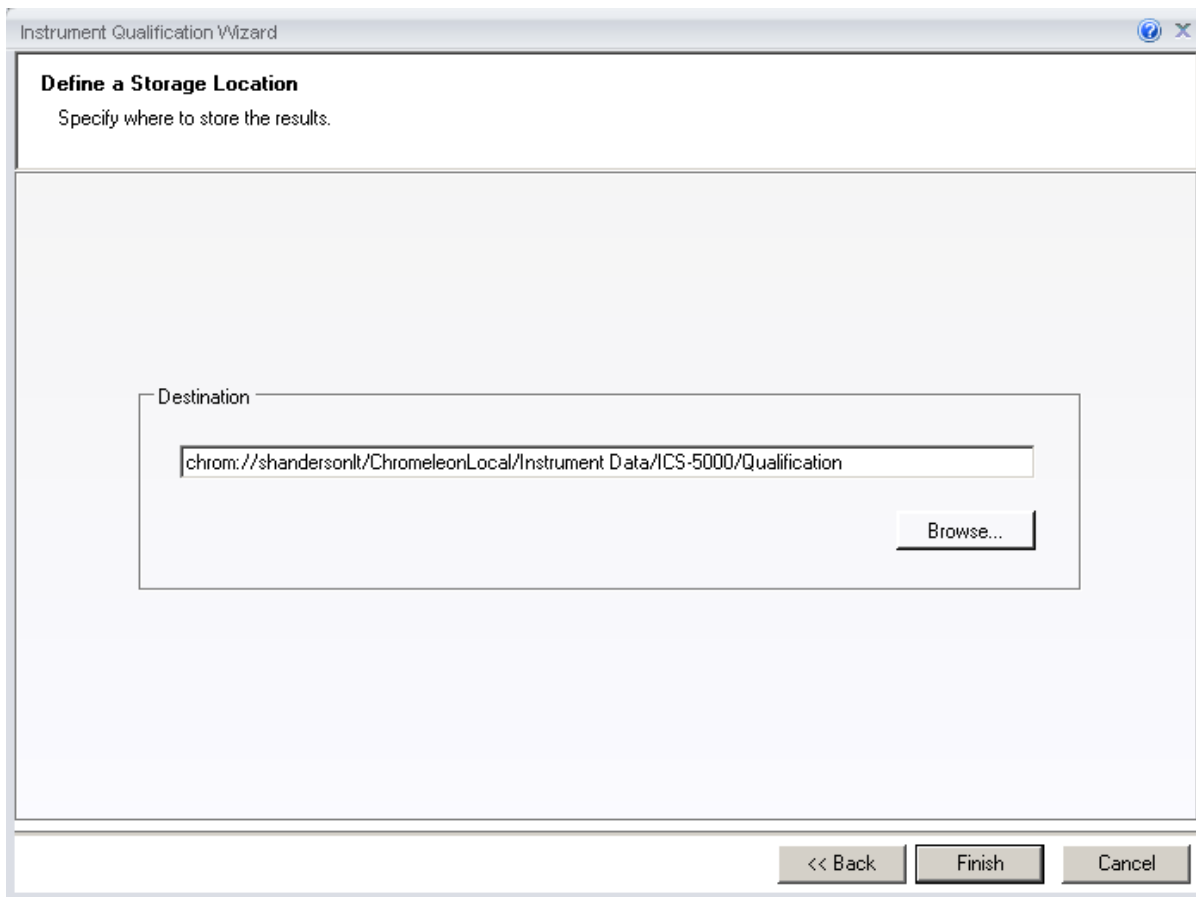


Figure 15. Selecting the storage location

4.7 OQ/PQ Sequence Execution

When the qualification wizard has been completed, the selected sequences are created and are automatically added to the instrument queue. A progress window shows which steps have been performed:

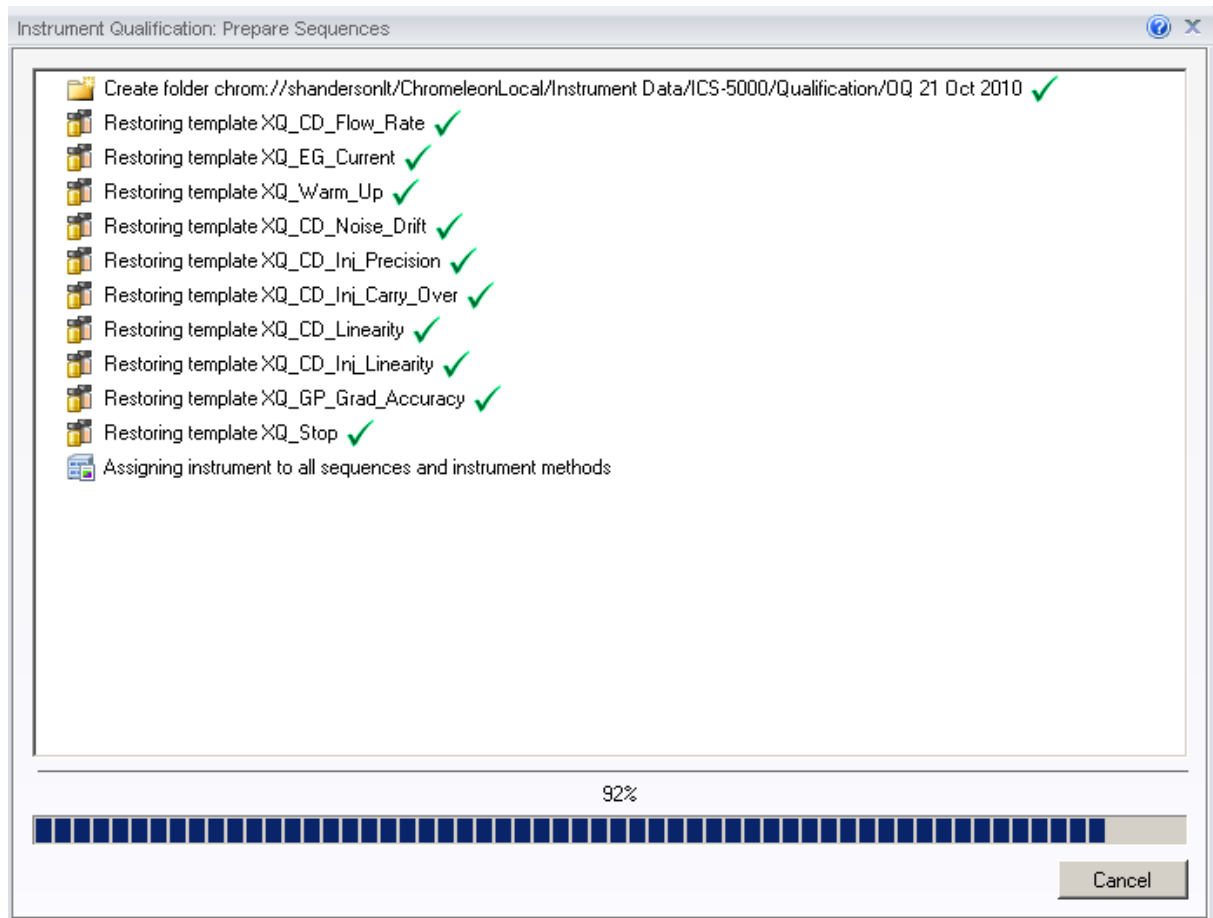


Figure 16. Progress during sequence creation

The Instrument View dialog box will open, displaying the Queue tab. As soon as the queue is started, Chromeleon runs the sequences. The wizard should copy the sequences to the destination selected then auto-populate the instrument queue. If the error “Instrument not connected” appears, make certain that the Chromeleon 7.1 must be displaying the instrument pane with the system to be tested.

The OQ/PQ qualification tests are performed in the following order:

- a. Flow Rate Test
- b. Eluent Generator Current Test (only if an EG is present)
- c. Warm Up

- d. Wavelength Accuracy
- e. Detector Noise and Drift Test
- f. Injector Precision Test
- g. Carryover Test
- h. Detector Linearity Test
- i. Injector Linearity Test (only if an AS or AS-AP is present)
- j. Sample Preparation Precision Test (only if an AS is present with the Sample Prep Option and the 10 mL Tray Type installed with the conductivity or the absorbance detector)
- k. Gradient Accuracy Test (only if an analytical gradient pump is present)
- l. Column TC and/or Compartment TC
- m. Sample Temperature Accuracy of Autosamplers (only for AS-AP Autosamplers)

Note: Test 'a' and 'l' is done manually and does not require a sequence template. The Instrument Method will pause before the Eluent Generator Current Qualification Test and the Flow-Rate Accuracy and Precision Test as these tests require user intervention.

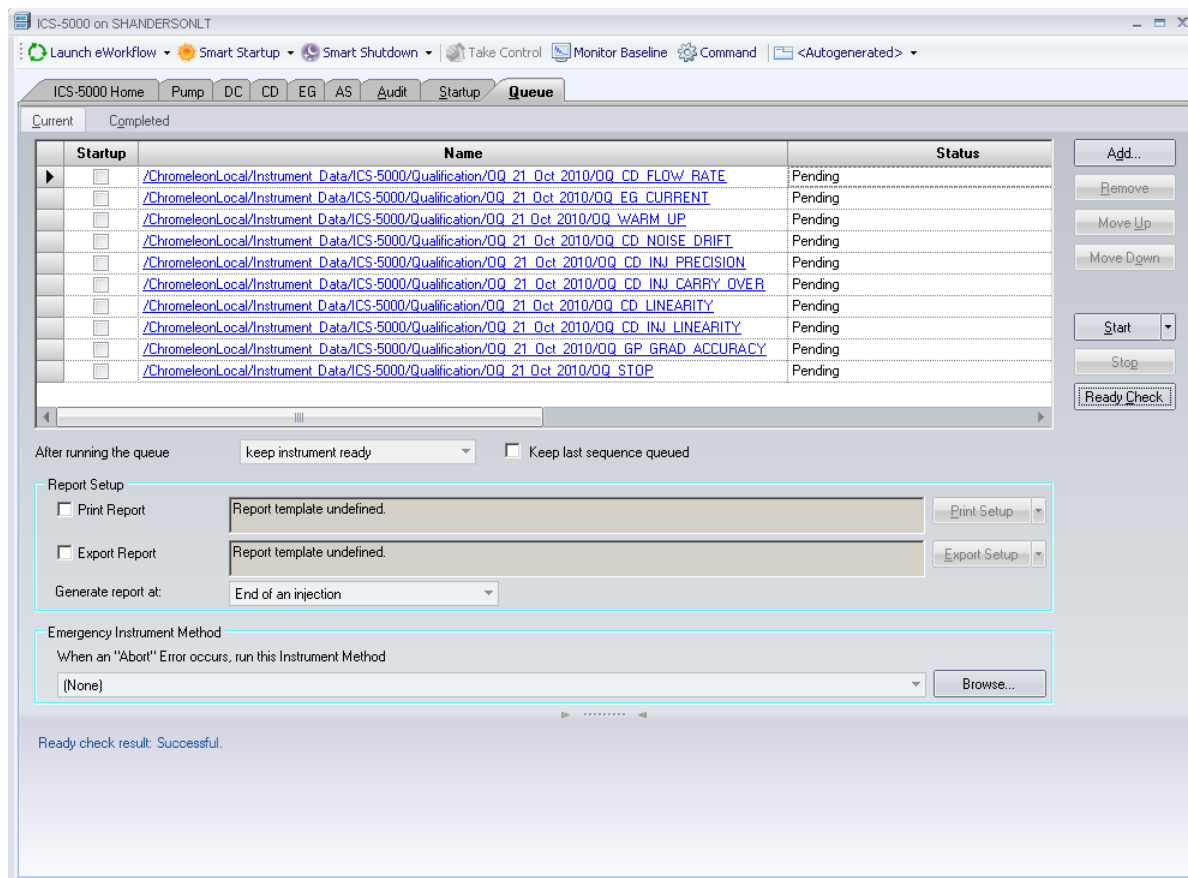


Figure 17. Sequence queue for Chromeleon 7.1

4.8 Evaluating the Test Sequences

The qualification sequences are saved under the path that was selected in the wizard. Each sequence also includes a separate report template.

To edit the report, open the report and remove the protection (if any) from the SPECIFICATION sheet. Enter the following information:

Name of the customer and name of the tester

Sample information such as batch number, expiration date, and actual concentration of the standard.

Name of the item that is used to generate the backpressure [default: capillary (L: 15 m; ID: 0.18 mm)]

Do not change any of the other report sheets. The report contains many references between data sheets. If you insert or delete lines and columns, these references will be lost and the calculations will be wrong.

To make sure that Chromeleon reads and processes the data in the report correctly, always print the report from the Chromeleon Console.

In the Data category, right-click the sequence for which you want to print the report, and then click "Print Report".

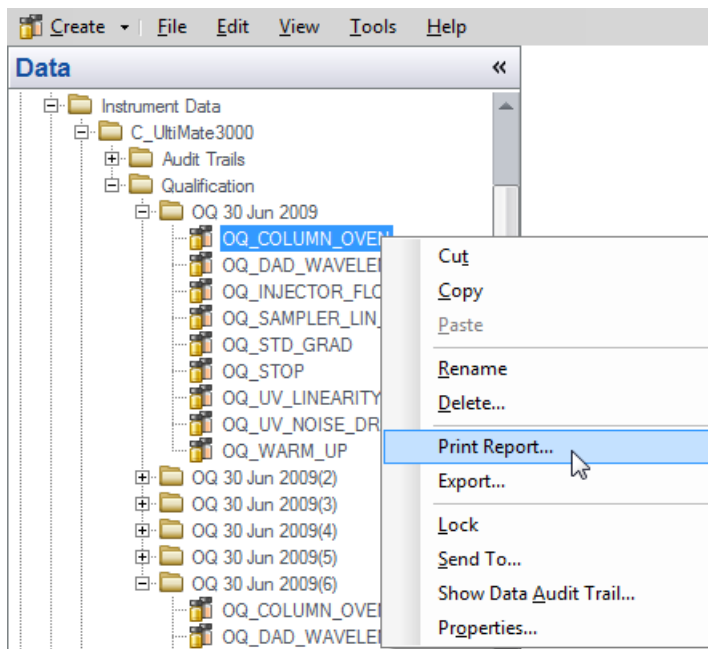


Figure 18. Selecting Print

Note: For ICS-5000 Integrated Amperometry System: Before executing the sequences, first turn on the pump flow and the cell current. Click on the “ePanel from the pump tab, start the pump flowing 50 mM NaOH at 0.25 mL/min. Click on the **EC detector tab**, Select Cell mode of “IntAmp”, then click on the **Waveform** button, use the drop down menu to choose the waveform **Amino Acids (pH/Ag/AgCl Reference)**, then click on the **Exit** button. Turn the cell voltage to **On**.

Note: If an Eluent Generator is configured on the system, the following message may occur at the Ready Check: “Flow rate not specified at time 0.0, using a flow setting 1.00.” This refers to the Eluent Generator flow rate, not the pump flow rate. It is not necessary to take any action since the Eluent Generator is not used for this part of the qualification. Proceed with starting the queue.

Note: For systems with a shared autosampler there might be a number of *informational* messages indicating that “Exclusive Access” has not been acquired or released. This is normal and occurs because the qualification does not release the autosampler until the qualification is complete.

Repeating Individual Tests

It may be necessary to repeat individual tests. If so, review Section 6 where problems are described according to which individual tests might fail. According to Good Laboratory Practice (GLP), all tests following the one that failed must be repeated. The entire OQ/PQ qualification is designed such that almost of the performance tests require that the previous one to have passed successfully.

For example, if the test regarding the linearity of a CD detector fails, the results regarding the linearity of the injector are questionable as the linearity of the detector is a prerequisite for testing the injector linearity.

Table 15. Sequence File Names and description

Sequence Name	Check
XQ_Flow_Rate	Pump flow-rate accuracy and precision tests for ICS-3000 or ICS-5000 pumps
XQ_EG_Current	Current qualification of an EG Eluent Generator
XQ_EG_Dual_Current	Current qualification of an Dual EG configuration (ICS-5000 with both EGC configured on one Instrument)
XQ_CD_Noise_Drift	Noise and drift test for conductivity detectors
XQ_DC_Amp_Noise_Drift	Noise and drift test for DC amperometry detectors
XQ_Int_Amp_Noise_Drift	Noise and drift test for integrated amperometry detectors
XQ_PDA_Noise_Drift	Noise and drift test for photodiode array detectors
XQ_UV_Vis_Noise_Drift	Noise and drift test for absorbance detectors (excluding PDA detectors)
XQ_Wavelength_Accuracy	Wavelength accuracy test for the VWD and PDA absorbance detectors
XQ_Abs_Inj_Precision	Injector precision test for systems with an absorbance detector
XQ_Abs_Inj_Carryover	Injector carryover tests for systems with an absorbance detector
XQ_CD_Inj_Precision	Injector precision test for systems with an conductivity detector
XQ_CD_Inj_Carry_Over	Injector carryover test for systems with an conductivity detector
XQ_ED_Warm_Up	Warm-up sequence for ICS-5000 systems with ED detector
XQ_ED_Noise_Drift	Noise and drift test for ICS-5000 amperometry detectors (ED in Integrated Amperometry mode)
XQ_ED_Inj_Precision	Injector precision for ICS-5000 systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Carryover	Injector carryover for ICS-5000 systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Linearity	Detector Linearity for ICS-5000 systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Inj_Linearity	Injector linearity for ICS-5000 systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Stop	The sequence turns off the pump and cell for ICS-5000 systems with amperometry detector (ED in Integrated Amperometry mode).
XQ_Abs_Linearity	Detector linearity test for absorbance detectors
XQ_CD_Linearity	Detector linearity test for conductivity detectors
XQ_Abs_Inj_Linearity	Injector linearity test for systems with an absorbance detector

XQ_CD_Inj_Linearity	Injector linearity test for systems with an detector
XQ_Abs_Samp_Prep	Injector sample preparation precision test for systems with an absorbance detector
XQ_CD_Samp_Prep	Injector sample preparation precision test for systems with an conductivity detector
XQ_GP_Grad_Accuracy	Gradient accuracy and noise test for systems with a gradient pump
XQ_CD_Noise_Drift	Noise and drift test for conductivity detectors
XQ_DC_Amp_Noise_Drift	Noise and drift test for DC amperometry detectors
XQ_Int_Amp_Noise_Drift	Noise and drift test for integrated amperometry detectors
XQ_PDA_Noise_Drift	Noise and drift test for photodiode array detectors
XQ_UV_Vis_Noise_Drift	Noise and drift test for absorbance detectors (excluding PDA detectors)
XQ_Stop	The sequence slows down the flow rate to 0.25 mL/min to conserve eluent for overnight runs

5 Performing the Qualification

5.1 OQ/PQ Test Order

The OQ/PQ qualification tests are performed in the following order:

1. Flow Rate Accuracy and Precision Qualification
2. Eluent Generator Current Qualification Test
3. Warm Up
4. Wavelength Accuracy Test (only for VWD, and PDA Absorbance Detectors)
5. Detector Noise and Drift Test
6. Injector Precision Test
7. Carryover Test
8. Detector Linearity Test
9. Injector Linearity Test (only for AS and AS-AP autosamplers)
10. Sample Preparation Precision Test (only for AS and AS50 autosamplers with the Sample Prep Option and 10 mL Tray Type installed with conductivity or absorbance detectors)
11. Gradient Accuracy Test (only for gradient pumps)
12. Column TC and/or Compartment TC
13. Sample Temperature Accuracy of Autosamplers (only for AS-AP autosamplers)

5.2 OQ/PQ Test Timetable

Table 16. OQ/PQ Test Timetable

Test	Method	Material Required	Equilibration Time	Approx. Run Time
Flow Rate Accuracy and Precision All pump models	Sequence	Eluent (water) Backpressure loop	15 min	30 min
Wavelength Accuracy AD25, ICS Series VWD	Sequence	Eluent (water)	Lamps – 2 h 10 min w/pump	6 min
Wavelength Accuracy PDA-100, ICS Series PDA	Sequence	Eluent (water)	Lamps – 2 h 10 min w/pump	6 min
Conductivity Noise / Drift	Sequence	Water at 1.0 mL/min	5 min	20 min
Int. Amperometry Noise / Drift	Sequence	Eluent (50 mM OH)	5 min	20 min
AS-DV/AS40, Manual Injector Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity	Sequence	Eluent (water) Standards Backpressure loop	10 min	1 hr, 10 min
AS-AP, AS Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity Injector Linearity	Sequence	- Eluent (water) - Standards - Backpressure loop	10 min	2 hr, 10 min
AS-AP with Temp. Control Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity Injector Linearity Sample Temp. Accuracy	Sequence	Eluent (water) Standards Backpressure loop Type K temperature sensor Calibrated Digital Thermometer	10 min	2 hr, 55 min

AS with Sample Prep Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity Injector Linearity Sample Prep Precision	Sequence	Eluent (water) Standards Backpressure loop	10 min	2 hr, 40 min
Eluent Generator Current Qualification	Sequence	Test cell Multimeter	N/A	16 min
Gradient Accuracy	Sequence	Eluents Backpressure loop	10 min	62 min
Fluorescence Noise and Wavelength Accuracy	Sequence	Eluent (water)	Lamps – 2 h 10 min w/pump	25 min

5.3 Starting the Sequences



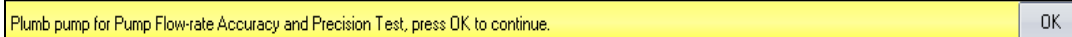
For AS Autosamplers with the Sequential Injection Option: Before starting any sequences, first verify that none of the instruments configured on the computer have “Acquired Exclusive Access” to the autosampler, or column TC, or compartment TC. If this is the case release the “Exclusive Access” command before proceeding. Once the qualification sequences begin, the system being qualified will gain and retain exclusive access to the autosampler, column TC and compartment TC until the “Stop” sequence is run and “Exclusive Access” is released. “Exclusive Access” can be released by going to the command function (F8), choosing the module from the list, and executing the “ReleaseExclusiveAccess” command.

5.3.1 Flow Rate Accuracy and Precision Qualification

The Flow Rate Accuracy and Precision test is designed to qualify the flow rate accuracy and precision of all pumps.

Thermo Fisher Scientific cannot guarantee tests performed outside the scope of the published OQ/PQ protocol.

1. Ensure the pump is disconnected from the Chromeleon software. The system should be in hold mode and a message box displaying “Plumb pump for Pump Flow Rate Accuracy and Precision Test, press OK to continue” should be visible.



2. Stop the pump from the front control panel of the module if it is not already stopped.
3. Locate the backpressure coil. . Use a coil made of 37" of red PEEK tubing (049715).
4. Connect one end of the backpressure coil directly to the outlet of the pump, after gradient mixers if applicable and the other to port 2 (P) of the injector valve.

5. Prepare two lengths of black tubing, 0.010 (0.25 mm) ID, of equal length, approximately 12" (40 cm). Connect one of these lines to port 1 (L) of the injector valve. The tubing connected to port 1 (L) of the injector valve will be the eluent line that goes to the "sample beaker."
6. Connect the other line to port 3 (C) of the injector valve. The tubing connected to port 3 (C) of the injector valve will be the waste line.
7. Run the waste line to a waste beaker and the eluent line to a sample beaker. The plumbing to the valve should look like the figure below.

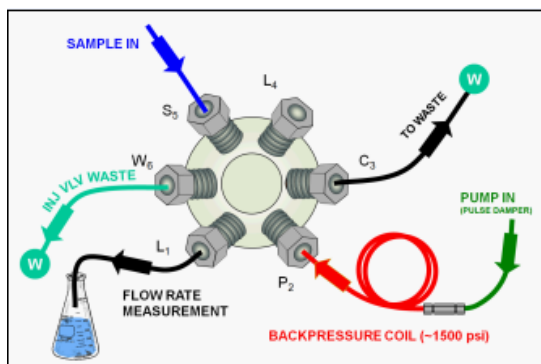


Figure 19. Plumbing schematic for Pump Flow Rate Test

8. Click OK on the Chromeleon software. The system should reconnect to the module and/or resume the program in run mode.
9. The program will equilibrate the pump for 15 minutes before starting the test. During this time eluent flow should pass through the waste line. Ensure that sufficient backpressure is observed during this equilibration time.
10. At time 0, the injector will switch to the eluent line.
11. At time 2.00, the injector will switch back to the waste line and the Chromeleon software will ask you to "Please weigh the eluent container. Enter results under segment 0. Press OK to continue." The program will be in hold mode and will wait until OK is pressed before continuing. Weigh the eluent container, but do not zero the balance used for weighing. Record the weight as segment 0. Click OK to continue.

Please weigh the eluent container. Enter results under segment 0. Press OK to continue.

Note: The template for this report is available on the PQ_OQ CD and can be used for manual data collection and data calculations.

Path: PQ_OQ/IC_Templates/OQ_PQ_Modular_Validation

12. The Chromeleon software will switch the injector back to the eluent line for exactly 5 minutes. After this time the injector will switch to the waste line and a the software will display the message "Please weigh the eluent container. Enter results under segment 1. Press OK to continue". The instrument will be in hold mode and will wait until OK is pressed before continuing. Weigh the eluent container, but do not zero the balance used for weighing. Record the weight as segment 1. Click OK to continue.
13. Repeat 12 for segments 2 through 5.

14. After the sequence is finished open the Water_FR sample from the Console. Select the Flow Rate sheet at the bottom of the page and enter the Segment values and room temperature into the sheet. Save the worksheet by clicking on the Chromeleon Icon on the top left of the User Interface and select sav . A written or balance printout of the weights should be signed and dated to accompany data entered on the sheet

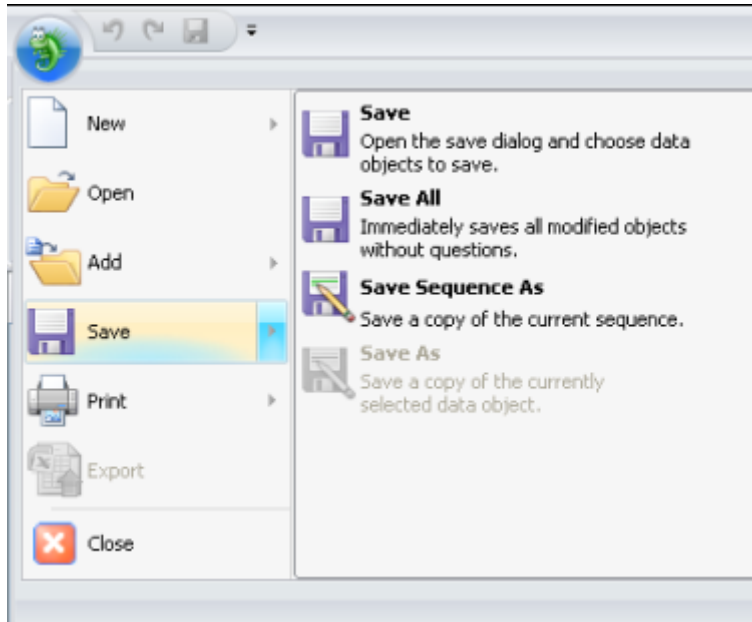


Figure 20. Selecting Save

15. Print the Flow Rate Accuracy and Precision report by clicking on the Chromeleon Icon on the top left of the User Interface and select print. Choose a printer and press OK to begin printing.

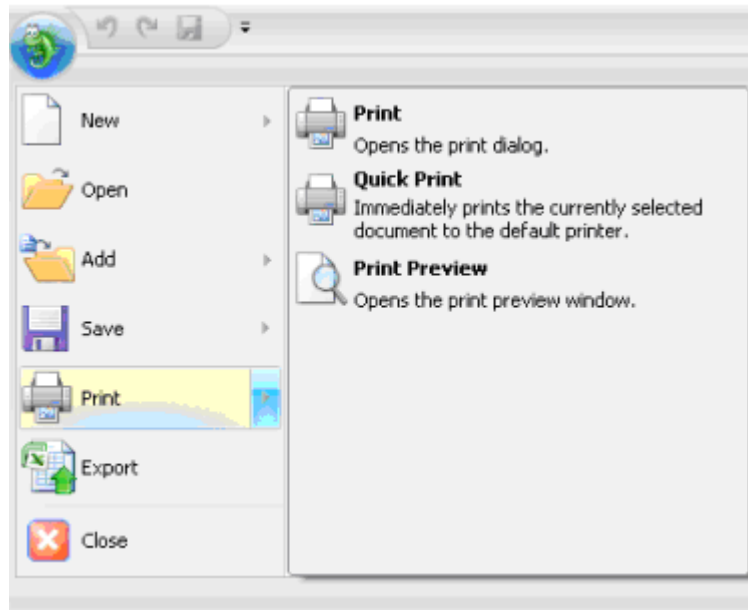


Figure 21. Selecting Print

16. If any part of the test does not pass, make the appropriate repair or calibrations and rerun the test. Refer to Section 5 for troubleshooting assistance.
17. Plumb the system for the remainder of the qualification.

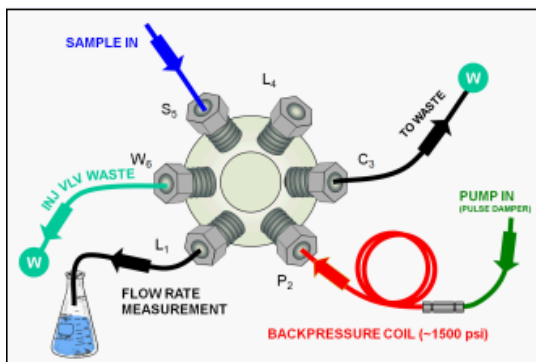


Figure 22. Plumbing diagram, initial equilibration step

18. Ensure the pump is disconnected from the Chromeleon software. The system should be in hold mode and a message box displaying "Plumb the system for the remainder of the qualification tests, press OK to continue" should be visible.
 - a. ICS-3000 Conductivity or Absorbance: Connect the backpressure tubing to the heat exchanger outlet line. Attach the loop end to the inlet of the conductivity or absorbance cell, thus bypassing all columns and suppressors.

It is important to connect the Backpressure Loop assembly upstream from the loop or unusual peak shapes may occur.

Note: A piece of 0.003" tubing may be used to provide approximately 1500 psi (\pm 100 psi) backpressure and sufficient delay.

- b. ICS-3000 Integrated Amperometry: Calibrate the reference electrode (see operators manual), then assemble the ED cell with an AAA certified disposable gold working electrode (P/N 060082) and mount the ED cell onto the ED detector. Connect the two cell leads (yellow and blue) to the appropriate ports on the detector. Connect a 24-inch piece of 0.003" PEEK tubing to port three (C) of the injection valve, connect the other end to the inlet of a CarboPac PA1 guard (2-mm) column. To the outlet of the guard column connect a small length (approx 15-cm) of 0.010" tubing. Attach the loop end to the inlet of the Integrated Amperometry Cell.

Note: When loading the Threonine standards in the AS or AS-AP autosampler, it may be necessary to cut the label off the Threonine vial for proper fit into the AS or AS-AP sample tray. For this reason, the concentration has been printed on the part of the label that will remain on the vial.

5.3.2 Eluent Generator Current Qualification

The Eluent Generator Current Qualification test ICS-3000 EG (single or dual) is configured with either a KOH or MSA cartridge (or both).

1. **Resume** the Chromeleon 6 queue or Chromeleon 7 Queue by clicking OK. This sequence is an Eluent Generator step gradient. Each step is held until the user clicks OK to continue. There may be a delay of up to 0.4 minutes between when the Eluent Generator program steps to the next concentration and when the ammeter or multimeter registers the new milliamp reading. Record the milliamp reading for each step on a note pad. These values will be entered into the **EG_CURRENT**, **EG_DUAL_CURRENT** or **EluGen Current Qualification** sheet after the run is complete. The run time is 11.0 minutes plus pause time.

Note: The template of this report is available on the PQ_OQ CD and can be used for manual data collection and data calculations.

Template name: OQ_PQ_Modular_Validation

2. When the run is complete, open the report file by double clicking on the Water_EG sample of the XQ_EG_Current sequence. Locate the row of sheet names at the bottom of the report and click on the **EG_CURRENT**, **EG_DUAL_CURRENT** or **EluGen Current Qualification** sheet.
3. Enter the milliamp readings that were recorded in step 1 for each concentration value in the **EG_CURRENT**, **EG_DUAL_CURRENT** or **EluGen Current Qualification** sheet. The deviation and pass/fail results will be calculated automatically.
4. Print the EG Current Qualification report by clicking on the Chromeleon Icon on the top left of the User Interface and select print. Choose a printer and press OK to begin printing.

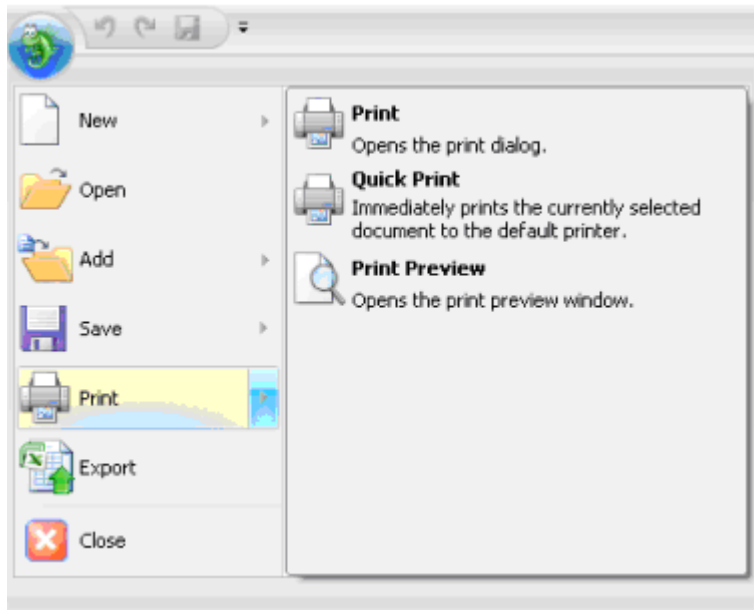


Figure 23. Selecting Print

5. If any part of the test does not pass, make the appropriate repair/adjustments and rerun the test. Refer to Section 5 for troubleshooting assistance.
6. To qualify another cartridge, select the new cartridge type in the control panel, add the XQ_EG_Current, XQ_EG_Dual_Current or XQ_IC_EG_Current sequence to the queue list, set the status of the Water_EG and Stop samples to single and repeat steps 1 - 4.

5.3.3 Performing the Main Qualification

The main part of the OQ/PQ consists of the tests 4 – 12 of the OQ/PQ Test Order, and this part of the qualification requires that an absorbance or conductivity detector be configured on the system being qualified.

If the qualification will be performed using a manual injection valve, use full loop injections and overfill the loop a minimum volume equal to of 3 times the sample loop volume. Inject standards at the beginning of each sample in the sequence. There will be a 2.5-minute delay before the sample is injected. It is advisable to manually edit the programs to replace the 2.5-minute delay with a message command to hold the system until the sample is ready.

Replace the “-2.500 Pump_Relay_1.Closed Duration=130.00” line with “-0.100 Message "Load the sample into the injection loop, press OK to continue"”

1. The sequences must be installed as described in Section 4.6.
2. The sequences must be set up as described in Section 4.7.

If the queue is not started already go to queue and Edit. Start the queue. Run times are as follows:

- Systems with an AS-DV, AS40 or manual injection have a run time of approximately 1h, 10 min.
- Systems with an AS or AS-AP, but no sample prep have a run time of approximately 2h, 10 min.
- Systems with an AS-AP with Injection Temperature Accuracy Test have a run time of approximately 2 h, 40 min.

- Systems with an AS and sample prep have a run time of approximately 2h, 40 min.
- Systems with a gradient pump, add an extra 60 minutes to the above times.

At the end of the main qualification the system will automatically stop.

For systems using an Eluent Generator, the program will pause before the Eluent Generator Current Qualification Test, proceed to section 5.3.2. The program will pause before the Flow Rate Accuracy and Precision Test, proceed to section 3.3.4 before continuing to step 3.

3. When the queue is finished, go to the Chromeleon console right click on each completed sequence present in the qualification, select the **Report** and **Print** the report. The Eluent Generator Current Qualification, Flow Rate Accuracy and Precision Qualification, Column Heater Accuracy qualification have already been printed.
4. Review the report for Pass/Fail results. If any test does not pass, make the appropriate instrument repair and/or adjustments and rerun all tests from the original point of failure on. Refer to Section 5 for troubleshooting assistance.

5.3.4 Column/Compartment TC Temperature Qualification

The column/compartment TC temperature qualification can be used to qualify the temperature accuracy of the column/compartment TC on an ICS-3000. After completion of all other tests record the reading on the digital thermometer. See 5.2.5 to set up the Thermocouple on to the digital thermometer.

Open the Water_WU sample in the Warm Up sequence from the console. Select the Temperature Qualification tab at the bottom of the page Enter the measured temperature of the Column TC in the "Measured Column Temperature" cell and if the Compartment TC is installed, enter the measured temperature of the Compartment TC in the "Measured Compartment Temperature" cell.

Note: The template of this report is stored in the data vault of the respective sequence. Please refer to Chromeleon 7 Quick Start Guide on how to access report templates.

Template name: OQ_PQ_Modular_Validation

If the test does not pass, make the appropriate repair/adjustments and rerun the test. Refer to Section 5 for troubleshooting assistance.

Print the Column TC Temperature Verification report by selecting "Print" from the File menu. Choose a printer and press OK to begin printing.

5.3.5 Turning off watermarks in the reports

Some users prefer not having watermarks in the back of the report. The procedure below describes how to turn off watermarks in the reports. The procedure can only be performed if the user has sufficient privileges granted by the administration.

1. Go to the Instrument Category
2. In the Chromeleon Console, Go to the Tools menu.
3. Select Administrations Console

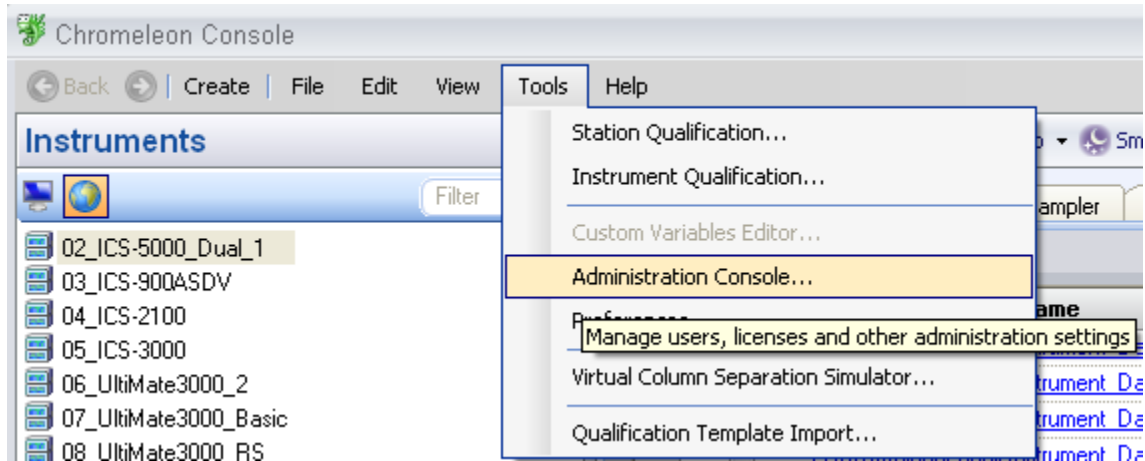


Figure 24. **Selecting Administration Console**

4. Select Global Policies within the Administration Console.

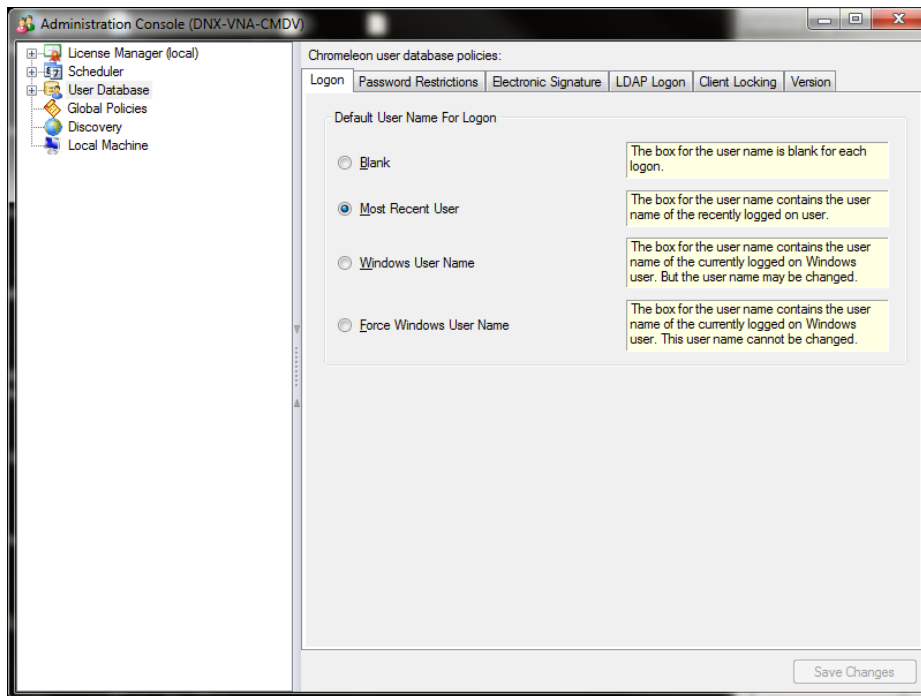


Figure 25 **Selecting Global Policies**

5. Select "Reporting" tab, then uncheck "Print with watermark"

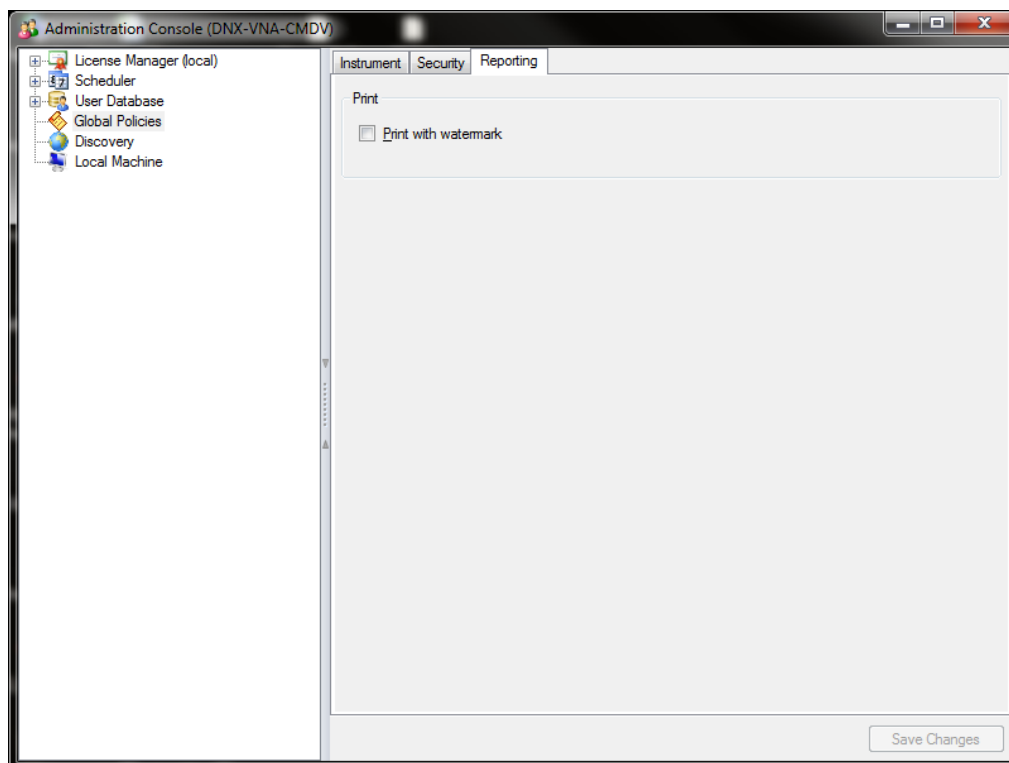


Figure 26. **Selecting reporting**

5.3.6 Sample Temperature Accuracy (AS-AP autosampler only)

This section describes how the sample temperature accuracy is determined for the following Thermo Fisher Scientific autosamplers: AS-AP. For this test, only the autosampler is required. The other modules of the IC system are not required.

Test Procedure

The following table lists the materials required for performing the test.

Part No.	Description	Quantity
6820.0010	Type K temperature sensor for P600 thermometers	1
5705.0050A	Column Thermostat PQ Kit	1

In addition, a standard polypropylene vial (10 mL) is required. Fill the vial with water (do not cap).

Connecting and Configuring the System

System connections

- Connect the Type K temperature sensor to the P600 thermometer and set the sensor type and calibration values as described in the instructions for the thermometer.



Figure 27. P600 Thermometer and the Type K Sensor

- Fill an open standard polypropylene vial (10 mL) with water and place it at sample position BC1 if you have a 10 mL rack or sample position RB1 if you have a small vial rack.
- Insert the temperature sensor into the vial at a right angle until the tip touches the vial bottom.



Figure 28. P600 Thermometer with the Type K sensor inserted into the vial

- Rotate the carousel cover until it closes completely.



Figure 29. Cover closed

- **Configuration**
Configuration is not required.
- **Preparing Chromeleon**
To qualify the sample temperature accuracy, select the following sequence:
SAMPLER_TEMP_ACC

Performing the Check

Set the sample temperature to 15 °C. When the nominal temperature is reached, the external thermometer is used to record the sample temperature over a period of 30 minutes.

Tip: Do not perform any autosampler commands during the test. Moving the needle arm or carousel may damage the thermometer or autosampler.

Duration

The test takes approximately 45 minutes.

5.3.7 AS-AP pH and Conductivity Qualification

This section describes how the sample pH and conductivity accuracy is determined for the following Thermo Fisher Scientific autosampler: AS-AP. For this test, the autosampler and

other modules of the IC system are required. Please refer to the operator manual for pH and conductivity detector set up. The pH qualification should be performed after the standard qualification. The AS-AP pH and conductivity qualification will be set up in push mode only.

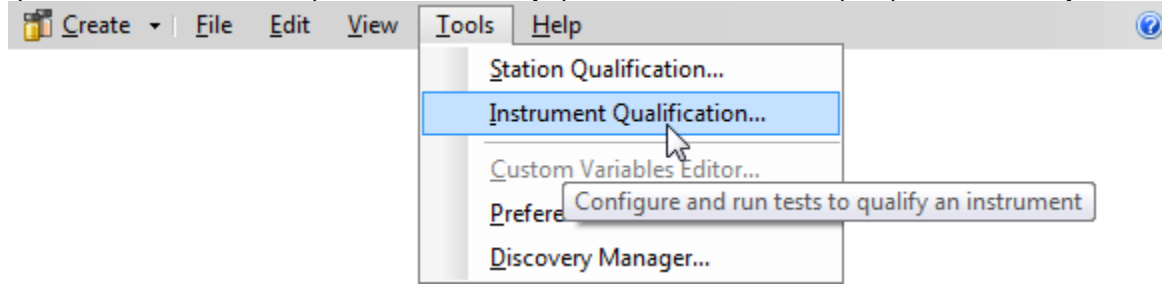


Figure 30. Instrument Pane for Chromeleon 7.1

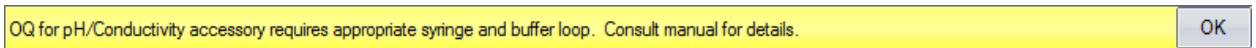


Figure 31. Starting the Instrument Qualification Wizard for Chromeleon 7.1

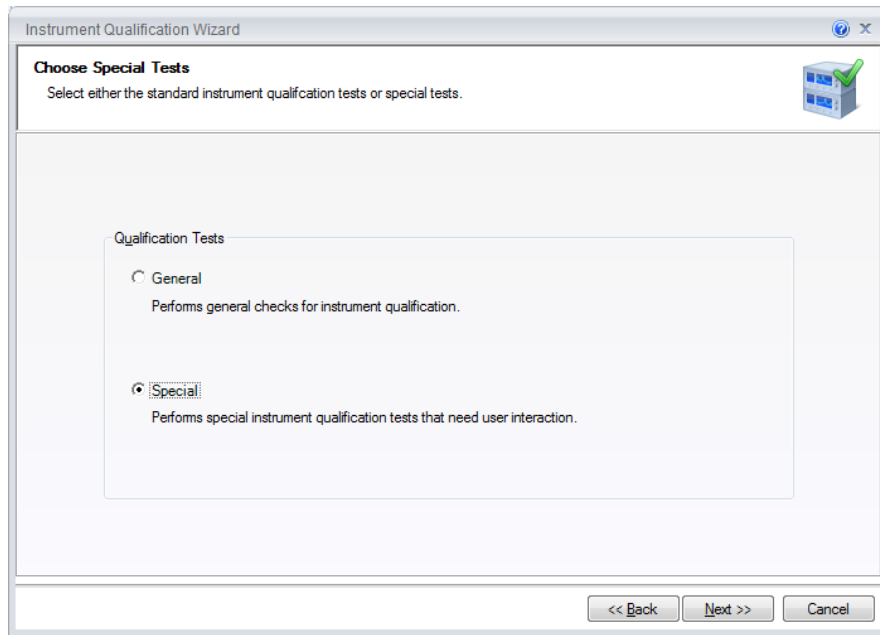


Figure 32. Select special for the Qualification Tests

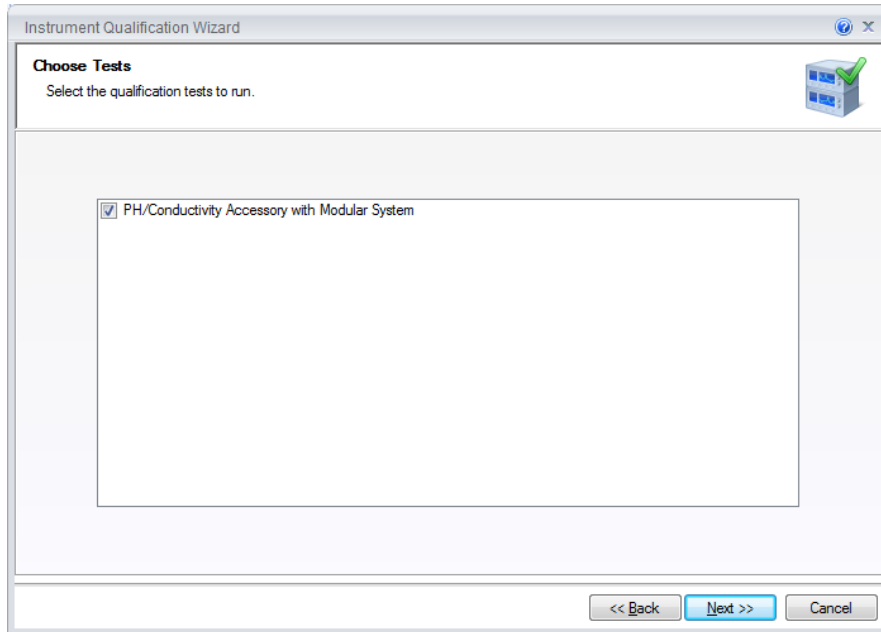


Figure 33. The pH/ Conductivity Tests will be selected if the instrument is configured correctly.

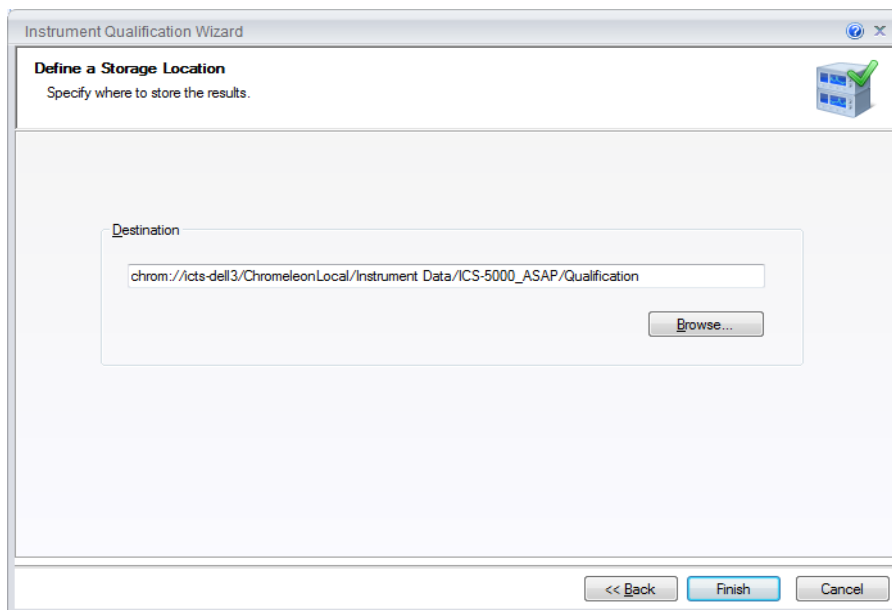


Figure 34. Select finish

#	Chromatogram	Name	Type	Level	Position	Volume [μL]	Instrument Method	Processing Method
1	None	wARMUP	Blank	01	RA1	100.0000	wARMUP	Validation
2	None	pH_OFFSET_CALIBR...	Blank	01	RA1	100.0000	pH7_CALIBRATION	Validation
3	None	pH_SLOPE_CALIBR...	Blank	01	RA2	100.0000	pH4_CALIBRATION	Validation
4	None	pH_SLOPE_CALIBR...	Blank	01	RA3	100.0000	pH10_CALIBRATION	Validation
5	None	pH_VERIFICATION(p...	Blank	01	RA1	100.0000	pH_MEASUREMENT	Validation
6	None	pH_VERIFICATION(p...	Blank	01	RA2	100.0000	pH_MEASUREMENT	Validation
7	None	pH_VERIFICATION(p...	Blank	01	RA3	100.0000	pH_MEASUREMENT	Validation
8	None	CD_CALIBRATION	Blank	01	RA4	100.0000	CD_CALIBRATION	Validation
9	None	CD_MEASUREMENT...	Blank	01	RA5	100.0000	CD_MEASUREMENT	Validation

Figure 35. Sequences

Performing the Check

1. Fill the vials according to the table below.

Table 17. OQ/PQ Test Timetable

Qualification	Standard	Sample Order on AS-P
pH Measurement	pH OFFSET Calibration	RA1
pH Measurement	pH Slope Calibration	RA2
pH Measurement	pH Slope Calibration	RA3
CD Calibration	Blank	RA4
CD Measurement	Conductivity Standard	RA5

2. Start the sequence.
3. A pop out screen will remind the user to configure the AS-AP with the 5mL syringe and the 8500 µL buffer loop before the qualification begins.

OQ for pH/Conductivity accessory requires appropriate syringe and buffer loop. Consult manual for details.

OK

Figure 36. Reminder Screen

Duration

The test takes approximately 1 hour.

5.3.8 Qualification Comments

The comments section of the report is provided as a place to record any comments, test failures, or configuration changes that occur during the qualification.

To use the comment section, open the Water_WU sample in the Warm Up sequence from the console. Select the Comment tab at the bottom of the page, turn on layout mode and enter any comments. This page can also be printed and comments entered manually.

Note: The template of this report is stored in the data vault of the respective sequence. Please refer to Chromeleon 7 Quick Start Guide on how to access report templates.

Template name: OQ_PQ_Modular_Validation

5.4 What to do if a Test Fails

If at any point the instrument results do not pass within Thermo Fisher Scientific specified limits, the instrument must be serviced. See Chapter 6 for troubleshooting assistance. When the repair is complete, rerun all tests from the original point of failure on in the sequence. All actions for failures and repairs should be recorded on the “comments” report. For example, if the instrument passes tests 1, 2, and 3 and fails test 4, make the appropriate repairs, record the action, and then repeat tests 4 – 12 only.

When rerunning the tests, restart the Instruments OQ or Instrument PQ procedure from the Qualification menu and uncheck all passed tests prior to the test that failed. All tests after the test that failed should be left checked even if they have previously passed. The Warm Up sequence is always required and must be rerun.

5.5 OQ/PQ Completion

5.5.1 Customer Review

1. The printed results should be reviewed by the instrument owner/user.
2. When the qualification is accepted, both the Customer and Qualification Executor need to sign the printed reports.
3. Fill out the Qualification Stickers and Certificates of Functional Performance for the qualified modules.
4. Make a copy of the certificates for FSR qualification, multimeter if used, temperature probe if used, and original standard certificate(s).
5. Place the reports and certificates into the binder provided in the OQ/PQ kit and leave it with the customer.

5.5.2 Accuracy of Reports

In accordance with Good Laboratory Practice (GLP), calculations used to determine the results in the printed reports use more significant figures than those shown and are rounded down for the final report. When reproducing the calculations by hand it may be necessary to obtain numbers from the reports after increasing the number of displayed significant figures.

The number of displayed significant figures can be adjusted through the report publisher. Open the sequence containing the samples for the test being examined and double click on a sample. The

sample data should be shown. Under the View menu choose Printer Layout to open the report. Highlight the numbers being used for the hand calculations, click on the Format menu and select number. The number of digits displayed can be increased by changing the number type; for example, entering 0.00000 into the Type field will increase the number of displayed decimal places to 5. **Always use more significant figures for hand calculations than is shown in the printed report.**

5.5.3 Returning the System to the Original Configuration

1. If the Signal factor was modified in the system preparation, or a module was renamed it must be set back to its original value. Open the Chromeleon Instrument Configuration application. Double click on the detector/s that were qualified to view the properties page. Select the **Signals** page to change the factor back to the original value and the **General** page to change the name back to the original value. Exit the properties page and save the configuration.
2. Disconnect the Backpressure Coil and restore the system plumbing to its original configuration with the appropriate columns, suppressors, and Eluent Generator in line.
3. Remove the sample loop and reinstall the original sample loop back onto the injection valve. For systems using an AS Autosampler, go to the **Main Menu** on the front panel of the autosampler. Access the **Module Set Up** screen and then go to **Plumbing Configuration**. Change the sample loop size back to its original value.
4. Reconnect the original eluents to the pump and prime each line individually to purge the eluents used in the OQ/PQ tests.
5. Flush the system with an eluent recommended by the customer and confirm that there are no leaks.

6 Troubleshooting

6.1 General Hints

If the system pressure is above 2000 psi after connecting the PEEK Backpressure coil with a flow rate of 1.00 mL/min, the coil has a blockage. Re-cut the ends of the coil or replace it.

Dips before or after the main nitrate or caffeine peak indicate extra dead volume in the system. This is caused by small voids between the end of a liquid line and the bottom of the port. Individually disconnect each line from the injection valve to the detector cell. Push the ferrule and fitting up and away from the end of the tubing. Make sure the tubing is cut straight. If the end of the tubing is not cut straight, re-cut the end. When reinstalling all liquid connections, first insert the tubing all the way into the port until it touches the bottom. Hold the tubing in this position while inserting and tightening ferrule and fitting down into the port.

6.2 Failure of Individual Tests

6.2.1 Conductivity Detector Tests

Test	Reason	Action
Baseline Noise	Poor water quality	Replace with an alternate water source
	Air bubbles in flow cell	Flush out flow cell
	Cell temperature regulation failure	Replace cell
Drift	System not equilibrated	Allow longer flush/equilibration time
	Lab temperature not stable	Regulate temperature (block vents, close windows, etc.)
	Water not degassed	Degas water or turn degas unit onto "Always On" setting.
Detector Linearity	Concentration of standards is incorrect	Obtain new fresh standards
	Liquid leak	Find and repair source of leak
	Injection volume not consistent (manual injections only)	Overfill sample loop at least 4x when loading
	The peak height of the sample with the highest concentration is not in the linearity range specified for the detector, i.e., usually > 1500 mAU (for Thermo Fisher Scientific and PDA detectors).	Reduce the injection volume for all samples used for the detector linearity check so that the peak height of the sample with the highest concentration is in the linearity range of the detector, i.e., usually < 1500 mAU.

6.2.2 Pump

Test	Reason	Action
Gradient Step Accuracy	Air in system	Prime pump – prime each eluent line individually for 3 – 5 minutes, flush system
	System not equilibrated	Allow longer flush/equilibrium time
	Old eluent in vacuum degas chamber	Prime pump – prime each line individually for 3 – 5 minutes
	Incorrect eluent concentration for eluent B and D	Remake 0.3% Acetone, 1 mM KCl solutions or 8.3 μ M glucose in 50 mM NaOH
	Pump eluents might need to be pressurized	Pressurize eluent bottles with 5 psi helium or nitrogen
	Eluent bottles not vented	Vent eluent bottles
	Pump is not primed	Prime the pump
Step Noise (ripple)	Eluent lines not conditioned with new eluent	Prime all 4 eluent lines for at least 15 minutes
	Air in system	Prime pump, flush system
	Air in flow cell	Flush flow cell with iso-propyl alcohol, ensure adequate backpressure on flow cell
	System not equilibrated	Allow longer flush/equilibrium time
	Check valve or piston seal failure	Replace check valve/piston seal
Flow Rate Accuracy and Precision	Void time from pump to detector too long or short	Adjust tubing length accordingly
	Flow rate out of calibration	Re-calibrate the flow rate of the pump

6.2.3 Autosampler

Test	Reason	Action
Injector Precision	Air injected from vial or from sample syringe when doing manual injections Air bubbles in AS sample syringe Liquid leak in autosampler or injection valve Poor water quality in flush reservoir	Too little sample volume in vial – refill vial or flush bubbles out of injection syringe Flush out syringe Find and repair source of leak Change water and flush sampler fluidics
Injection Volume Linearity	Detector failure Defective sample syringe	See Detector linearity troubleshooting section Replace sample syringe
Carryover	Contaminated injection valve Contamination in autosampler Contamination in sample vial or in syringe used for manual injections	Replace injection valve rotor and stator face Flush/replace autosampler liquid lines Use new sample vial for deionized water blank, rinse vial thoroughly before using, flush syringe or use a new one
Sample Prep Precision	Air injected from vial Air bubbles in AS sample syringe or prep syringe and tubing Defective prep syringe Liquid leak in autosampler or injection valve Poor water quality in Sample Prep reservoir	Too little sample volume in vial, refill vial Flush out syringe Replace prep syringe Find and repair source of leak Change water and flush sampler fluidics
Tray Temperature Accuracy	Thermometer not calibrated	Calibrate Thermometer

6.2.4 Integrated Amperometry Detector Tests

Test	Reason	Action
Baseline Noise	Electrochemical cell not connected properly	Reconnect test cell cable to amperometry cell connection on SP board
	Electrochemical cell defective	Replace DWE, reference electrode or electrochemical cell
Drift	Detector not equilibrated	Allow longer equilibration time
	Lab temperature not stable	Regulate temperature (block vents, close windows, etc.)

6.2.5 Eluent Generator (ICS-3000 EG)

Test	Reason	Action
Current	Qualification Test box not connected properly	Reconnect test cell cable to the Eluent Generator, confirm ammeter or multimeter leads are making contact with test cell wires
	Ammeter or multimeter on wrong setting	Ammeter or multimeter should be set to read current (mA). Range should be set to view 0.00 to 300.00 mA
	Qualification Test box defective	Replace Qualification Test Box

6.2.6 Column Thermostat

Test	Reason	Action
Temperature Qualification	Column Compartment not calibrated	Recalibrate the Column Heater
	Thermometer not calibrated	Calibrate Thermometer
	Thermometer not fitted in column compartment correctly	Fit thermometer in column compartment correctly

7 Example Report

Please refer to the ICS-5000 example reports

**User's Guide:
Chromeleon[®] Operational Qualification and
Performance Qualification Software Templates
for Integrated Ion Chromatography Instruments,
ICS-1000, ICS-1100, ICS-1500, ICS-1600,
ICS-2000 and ICS-2100
Revision 7.12**

2013 Thermo Fisher Scientific

Document No. 065448 Part B
March 2013, Thermo Fisher Scientific Corporation
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PRINTING HISTORY

Revision 7.10 September 2011

Revision 7.11 December 2011

Revision 7.12 March 2013

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1 Introduction

Operational and Performance Qualification are becoming increasingly important to analytical laboratories. Documented evidence must be provided to demonstrate the integrity of data collected and qualify the results obtained on laboratory instrumentation.

The following institutes set and control industry instrumentation standards:

NIST The National Institute of Standards and Technology is an administrative body of the US Department of Commerce. Together with the industry, the NIST develops new technologies and sets technological standards countrywide. Thermo Fisher Scientific includes NIST traceable standards as part of the qualification kit.

ASTM The American Society for Testing and Materials sets technical standards and rules for the industry worldwide, specifically; E 1657 – 98: Standard Practice for Testing Variable-Wavelength Photometric Detectors in Liquid Chromatography. E1511 – 93: Standard Practice for Testing Conductivity Detectors used in Liquid Chromatography. E19.09.07: Proposed Standard Practice for Evaluation of Gradient HPLC pumps – Draft.

USP The United States Pharmacopeia (USP) is an official public standards-setting authority for all prescription and over-the-counter medicines and other health care products manufactured or sold in the United States. USP establishes documentary and reference standards to ensure quality analytical instrument qualification process.

These standards are written in broad terms to make them as widely applicable as possible. All stipulate general requirements specifying instruments must be fit for purpose, technically checked in defined periods, calibrated and make sure that test results are documented according to international quality standards. The OQ/PQ procedure used to qualify Thermo Fisher Scientific Ion Chromatography Systems meets the requirements established by NIST, ASTM and USP.

This OQ/PQ procedure provides qualification testing for **ICS-1000, ICS-1100, ICS-1500, ICS-1600, ICS-2000** and **ICS-2100** systems including additional components from the table in section 1.6.

OQ/PQ results can be influenced by unusual laboratory conditions. For example, direct sunlight and close proximity to air conditioning ducts will cause thermal irregularities that will affect baseline drift on conductivity detectors. In these cases, it may happen that the manufacturer's specifications are not met. Ensure that the instrument is operated in suitable environmental conditions or for PQ, the specifications may be adjusted accordingly.

All OQ/PQ activities must be formally recorded, all instruments must be clearly identified, and test reports and supporting documentation must include the following information:

- Module name, type and serial number
- Software and firmware version
- Test data
- Service interval
- Signature of the Thermo Fisher Scientific Service Engineer/Qualification Executor who performed the OQ/PQ
- Signature of the customer who reviewed and accepted the OQ/PQ results

1.1 How to Use This Manual

It is essential that the operator read and understand this manual completely before implementing any OQ or PQ process.

1. Use the table of contents to determine which tests are required for the system to be qualified and refer to the recommended sections. This will help keep track of what steps to follow during system setup and preparation. It will also aid in selecting the appropriate sequences to run the tests.
2. Use the **Table of Contents** and the **List of Tables** as reference guides to locate information quickly.

The electronic version of the OQ/PQ operator's manual contains numerous hypertext links that can take you to other locations within the file. These links include:

- Table of contents entries
- Cross-references (underlined in blue) to sections, figures, tables, etc.

Chapter 1 Introduction

Introduces the conventions used in performing OQ/PQ with the ICS-1000, ICS-1100, ICS-1500, ICS-1600, ICS-2000 and ICS-2100; including the basic requirements, industry standards and manual summary.

Chapter 2 Overview of the Tests

Provides an overview of the tests available to be performed.

Chapter 3 Explanation of Test Procedures

Describes the theory behind each test, describes OQ/PQ operating procedures and evaluation.

Chapter 4 Qualification Preparation

Provides step by step preparation instructions for the OQ/PQ software, hardware installation and the details of the materials in ship kits.

Chapter 5 Performing the Qualification

Provides detailed operating instructions on how to perform OQ/PQ including test order, time table, sequence initialization and troubleshooting.

Chapter 6 Troubleshooting

Lists problems and presents step-by-step procedures for how to isolate and eliminate the cause of each problem.

Chapter 7 Example Report

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1.4 Operational Qualification (PQ)

According to “The Development and application of guidance on equipment qualification of analytical instruments” by P. Bedson and M. Sargent [Accred. Qual. Assur. (1996) 1: 265 – 274] the following definition applies:

“The purpose of OQ is to prove and document that an analytical system functions according to its operating specification while the specific environmental conditions are taken into account. In this specification, the supplier has therefore to define exactly the conditions that must be observed.”

OQ should be performed after initial system installation or when an additional new module is added to an existing system. OQ should be done according to the instructions outlined in this manual and all tests should be performed by a qualified Thermo Fisher Scientific Service Engineer. Any hand-

written notes made during the performance of the tests must become part of the test results, in accordance with Good Laboratory Practice (GLP) and current Good Manufacturing Practice (cGMP).

1.5 Performance Qualification (PQ)

According to "The Development and application of guidance on equipment qualification of analytical instruments" by P. Bedson and M. Sargent [Accred. Qual. Assur. (1996) 1: 265 – 274] the following definition applies:

"The purpose of PQ is to prove and document that an analytical system functions according to a specification that is suitable for the system's routine operation. A system is subject to wear when being operated. Thus, it may happen that the supplier's specification is no longer met. This means: The test procedures used for OQ are also used for PQ, but the tolerances of PQ are less restrictive than those for OQ. Using the same test procedures simplifies the handling."

PQ should be performed at regular intervals after the initial installation and OQ. Thermo Fisher Scientific recommends performing PQ every six months. PQ should be done according to the instructions outlined in this manual and all tests should be performed by a qualified Thermo Fisher Scientific Service Engineer. Any hand-written notes made during the performance of the tests must become part of the test results, in accordance with Good Laboratory Practice (GLP) and current Good Manufacturing Practice (cGMP).

1.6 Basic Requirements for Successful OQ and PQ

It is recommended that the most current revision of the OQ/PQ software templates is used in order for a qualification to be considered valid. It is the responsibility of the qualification engineer to determine the most up to date revision of the templates available and ensure that revision is used before proceeding with this qualification or if the customers SOP require an earlier OQ/PQ version to be used.

It is recommended that the qualification be performed using the most current Chromeleon[®] service pack version (Contact Thermo Fisher Scientific Technical Support for current version).

It is recommended that the qualification be performed using the most current firmware version for each system or module (Contact Thermo Fisher Scientific Technical Support for current version).

Operational Qualification and Performance Qualification are system-specific test procedures. The procedures described below are valid for IC systems comprised of the following equipment (only one type of detector and one pump can be qualified concurrently):

Pump: ICS-1000, ICS-1100, ICS-1500, ICS-1600, ICS-2000 or ICS-2100.

Manual injection or any of the following autosamplers: AS, AS-DV, AS40 or AS50.

AS-AP (push or pull mode) autosampler 10 mL vial trays (P/N 074938): with ICS-1100, 1600 and 2100.

Conductivity detector: detector integrated into ICS-1000, ICS-1100, ICS-1500, ICS-1600, ICS-2000, or ICS-2100.

Eluent Generator: ICS-2000 or ICS-2100

Absorbance detector: AD25, ICS Series VWD or ICS Series PDA.

Table 1. ICS-1000/1100/1500/1600/2000/2100 and Chromeleon Compatibility

Module	Minimum Chromeleon Software Version
AS-AP	7.1 SR1
AS	7.1 SR1
AS-DV	7.1 SR1
ICS-1000/1500/2000	7.1 SR1
ICS-1100/1600/2100	7.1 SR1
FLUKE189/289	7.1 SR1
ICS Series VWD	7.1 SR1
ICS Series PDA	7.1 SR1
AD25	7.1 SR1

It is possible to qualify modules in systems that include modules that are not listed here; however some modification of the included programs may be required. Such modifications are not within the scope of this document.

It is not possible to qualify multiple detectors on a single system, with the following exceptions: Two ICS-1000, ICS-1100, ICS-1500, ICS-1600, ICS-2000, ICS-2100 systems can be qualified simultaneously when used with an AS50 (USB version), AS-AP autosampler (with a 5000 μ L injection syringe) or AS autosampler (with a 5000 or 10,000 μ L injection syringe). Simultaneous injection does not support partial loop injection by the AS or AS-AP autosampler. To qualify the AS-AP or AS autosampler with the injector linearity test (partial loop injections) a 250 μ L injection syringe is required. The buffer size should always be set at 1200 μ L.

2 Overview of the Tests

The following table provides an overview on the parameters to be tested and the limits recommended for OQ and PQ.

- Template sequences (with programs specific to the Instrument Configuration are generated based on the master sequence of the OQ/PQ templates and the system configuration in the IC Instrument.
- The sequences are copied from each test that is performed on the same system.
- The Chromeleon software will automatically create a default unique name and directory to contain all template sequences.
- Do not alter the test sequence order.
- Note: The instrument's specifications can only be met if the conditions that are described in the accompanying manual are strictly observed.

Table 2. OQ/PQ Specification Limits

Instrument	Parameter	Description	Limits	
			OQ	PQ
ICS-1000/1100 ICS-1500/1600 ICS-2000/2100 Pump	Flow Rate Accuracy	ASTM Type I deionized water with low total organic carbon (TOC)* water is pumped at a flow rate of 1.00 mL/min into a weighed beaker for exactly 5 minutes. The mass of the water is measured, the flow rate of the pump calculated and compared with the theoretical value. This experiment is repeated four more times.	1.00 ± 0.05 mL/min	1.00 ± 0.05 mL/min
	Flow Rate Precision		≤2.0%	≤2.0%
ICS-1000/1100 ICS-1500/1600 ICS-2000/2100 Conductivity Detector	Baseline Noise	ASTM Type I deionized water with low total organic carbon (TOC)* is pumped through the detector at a flow rate of 1.0 mL/min	1.0 nS	2.0 nS
	Baseline Drift		10.0 nS/hr	20.0 nS/hr
	Detector Linearity	5 Nitrate standard solutions OQ/PQ standards kit P/N 063540 are injected in increasing concentrations. Concentration (ppm) and peak height (µS) are represented on a graph. The correlation coefficient of the resulting line and the deviations from it indicate the linearity.	r ≥0.999 RSD ≤5.0 %	r ≥0.999 RSD ≤5.0 %
ICS-1000/1100 (With optional column heater) ICS-1500/1600 ICS-2000/2100	Temperature Verification	The column heater is set to 40 °C for the duration of the entire qualification. At the end of the qualification the temperature of the column heater is measured with a calibrated external thermometer and compared with the theoretical value.	± 2.0 °C	± 2.5 °C

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Instrument	Parameter	Description	Limits	
			OQ	PQ
ICS-2000/ICS-2100 EG	Current Qualification	The eluent generator is commanded to produce a series of different concentrations (using the current (mA) cartridge type configured with a concentration range between 0 mM to 100 mM). At each EG concentration setting the current produced by the eluent generator is measured and compared with theoretical values.	OQ_PQ_Integrated_Validation	

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Instrument	Parameter	Description	Limits	
			OQ	PQ
PDA-100, ICS-Series PDA	Baseline Noise	ASTM Type 1 deionized with low total organic carbon (TOC) * water is pumped through the detector at 1.0 mL/min. Observed wavelengths are set for simultaneous monitoring at 254 nm and 520 nm	0.04 mAU	0.10 mAU
	Baseline Drift		0.5 mAU/hr	1.0 mAU/hr
	Wavelength Accuracy	Holmium oxide maxima are compared with the theoretical maxima.	± 2 nm	± 2 nm
	Detector Linearity	5 Caffeine solutions from the OQ/PQ standards kit P/N 063540 are injected in different concentrations. The absorbance is measured at a wavelength of 272 nm. Concentration (ppm) and peak height (mA) are represented on a graph. The correlation coefficient for the resulting line (and the deviations from it) indicates the linearity	r ≥0.999 RSD ≤5.0% (up to 1.5 AU)	r ≥0.999 RSD ≤5.0 % (up to 1.5AU)
AD25 and ICS Series VWD	Baseline Noise	ASTM Type 1 deionized with low TOC water is pumped through the detector at 1.0 mL/min. Wavelength 254 nm and 520 nm.	30 µAU	40 µAU
	Baseline Drift		200 µAU/hr	200 µAU/hr
AD25 and ICS Series VWD	Wavelength Accuracy	Holmium oxide maxima are compared with the theoretical maxima.	± 2 nm	± 2 nm
	Detector Linearity	5 Caffeine solutions from the OQ/PQ standards kit P/N 063540 are injected in different concentrations. The absorbance is measured at a wavelength of 272 nm. Concentration (ppm) and peak height (mA) are represented on a graph. The correlation coefficient for the resulting line (and the deviations from it) indicates the linearity	r ≥0.999 RSD ≤5.0 % (up to 1.5 AU)	r ≥0.999 RSD ≤5.0 % (up to 1.5 AU)

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Note: The AD25, VWD, PDA, and PDA-100 Detector UV and visible lamps must be turned on at least two hours prior to performing the qualification.

Instrument	Parameter	Description	Limits	
			OQ	PQ
All injectors and samplers	Injector Precision	10 injections of a defined standard at 25 μ L each of the same standard are analyzed. The percent relative standard deviation (%RSD) of the peak areas indicates the reproducibility of the injection volume.	RSD \leq 0.8 %	RSD \leq 1.0 %
	Carryover	Following the injection of a highly concentrated test sample, a blank sample is injected and then a water blank sample is injected. The peak area measured in this chromatogram compared with the peak area of the highly concentrated sample indicates the amount of Carryover. If there is any peak area measured in the water blank sample, that area is subtracted from the area of the blank sample before it is compared to the highly concentrated sample.	\leq 0.1 %	\leq 0.1 %
AS-AP, AS, AS50	Linearity of Injector Volume	5 injections of the same standard sample are analyzed. The injection volumes are different (10 μ L to 50 μ L). Injection volume and peak area are represented on a plot of μ L vs. μ S. The correlation coefficient of the resulting line (and the deviations from it) indicates the linearity.	r \geq 0.999 RSD \leq 1.0 %	r \geq 0.999 RSD \leq 1.0 %
AS, AS50	Sample Preparation Precision (if present)	3 samples of identical concentration are prepared from the same starting vial into 3 separate vials and injected. The relative standard deviation of the peak areas indicates the reproducibility of the sample preparation procedure	RSD \leq 1.5 %	RSD \leq 2.0 %
AS-AP pH	pH Accuracy	The sample temperature calibration is measuring the pH at 4, 7 and 10. The reading is stable if the reading and the three data points are within 0.2 pH unit of each other.	+/- pH 0.1	+/- pH 0.2
AS-AP Conductivity	Conductivity Accuracy	The conductivity calibration procedure consists of determining the conductivity zero point of the accessory and then measuring the conductivity of a 1000 μ S standard.	+/- 30 μ S	+/- 50 μ S
AS-AP	Temperature Accuracy (if present)	The sample temperature is measured using an external thermometer in a standard vial. The standard vial is filled with water (measured at 15.0 $^{\circ}$ C).	15.0 \pm 2.0 $^{\circ}$ C	15.0 \pm 4.0 $^{\circ}$ C

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb

3 Explanation of the Test Procedures

3.1 Flow Rate Accuracy and Precision Qualification

3.1.1 Theory

The accuracy and precision of a pump is qualified by accurately measuring 5 aliquots of eluent into a tared sample beaker for exactly 5 minutes each. The injection valve is used to measure the 5 minute aliquot and the volume of each aliquot is determined gravimetrically. The backpressure coil is placed before the injection valve and equal lengths of tubing are used for eluent and waste to ensure that there is no backpressure change when switching the valve from waste to sample.

3.1.2 Performing the Test and Evaluating the Results

The Flow Rate Accuracy and Precision Qualification are run using the sequence called XQ_Flow_Rate. The five weight measurements for the aliquots are entered manually into the report and converted to volume when the temperature is entered. Accuracy is measured by comparing the theoretical value (5.00 mL in 5.00 min) to each of the five aliquots and precision is measured by determining the relative standard deviation of the five aliquots.

3.2 Eluent Generator Current Qualification

3.2.1 Theory

The Eluent Generator Current Qualification is determined by performing an EG step gradient using varying eluent concentration set points. The Eluent Generator applies a specific current to the EluGen cartridge depending on the concentration of eluent and type of cartridge selected. An IC Qualification test box (P/N 072005) is used in place of the EluGen cartridge and the applied current for each step is measured using an ammeter or voltmeter that is connected to the test cell. No injection is performed.

3.2.2 Performing the Test and Evaluating the Results

The Eluent Generator current qualification test is run using the sequence called XQ_EG_Current or XQ_EG_Dual_Current. The current readings in mA are displayed on the voltmeter for each concentration step in the gradient. Values for each step are manually recorded (please enter at least three figures after the decimal in each entry) or can be automatically downloaded into the Report Template when using a Fluke 189/289 or equivalent multimeter (calibrated with up to 300 mV DC scale range and the multimeter needs to display three decimal places) which is configured in the Instrument as a module using the appropriate device driver. The sequence follows an XQ_Stop sequence that will pause the system to allow connection of the qualification test box and recording of the results.

3.3 Warm up

The warm up sequence is used to ensure that all parts of the system are responding appropriately and to record logs to the audit trail (later used to update the information in the report definition file). It is called XQ_Warm_Up where; X is P for PQ and O for OQ. The warm up sequence is necessary for all qualifications. The warm up sequences from the AS-AP directory inject the low level standards.

3.4 Baseline Noise and Drift

3.4.1 Theory

A 20-minute detector baseline background trace is collected using deionized water as the eluent at a flow rate of 1.00 mL/min. For ICS Series VWD and PDA, AD25 and PDA-100 detectors, data is

collected at 254 nm and 520 nm. No injection is performed. Then a short equilibration is done at 272 nm.

Noise is calculated by partitioning the 20-minute baseline signal into 20 one-minute intervals. The noise is the distance between two parallel lines through the measured minimum and maximum values and the regression line. The measured noise values are then averaged to establish the final value. To calculate drift, the Chromeleon software uses a method of least squares and calculates a regression line from the measured values. The slope of the regression line indicates the drift of the measured signal, and the amount of the slope indicates the amount of drift.

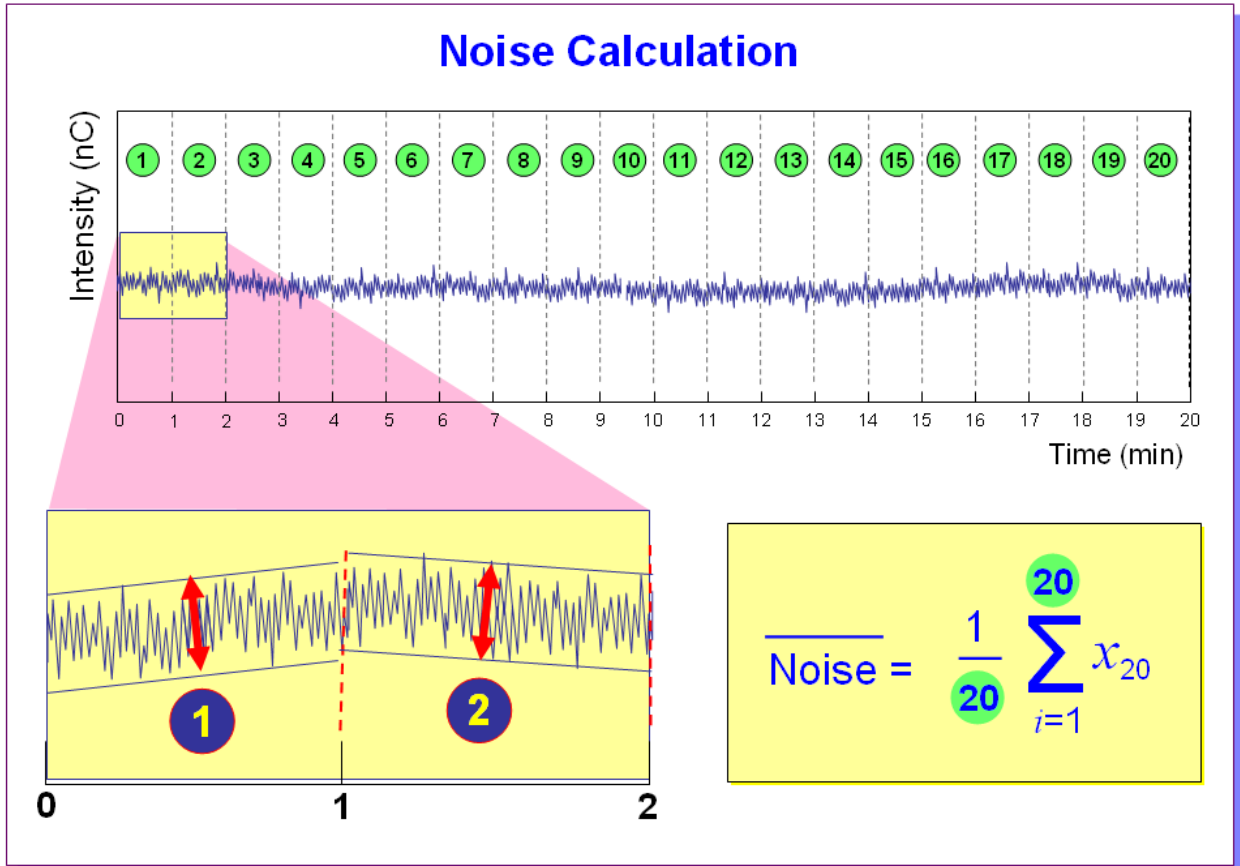


Figure 1. Noise Calculation

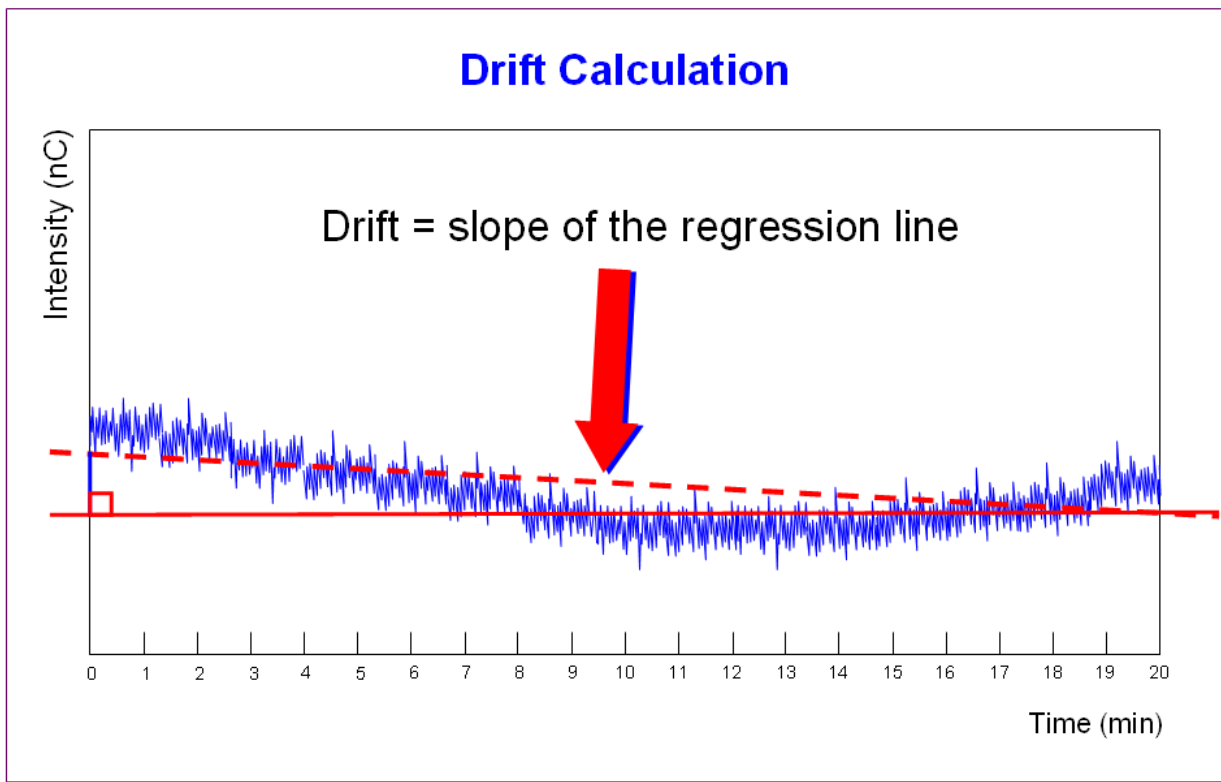


Figure 2. Drift Calculation

The slope of the regression line indicates the drift of the measured signal, and the amount of the slope indicates the amount of drift.

3.4.2 Performing the Test and Evaluating the Results

Noise and drift evaluation for all IC detectors is run using the sequence called XQ_Detector_Noise_Drift where X is P for PQ and O for OQ and Detector is CD for conductivity detectors, VWD Detector, and UV_Vis for PDA

3.5 Injector Precision

3.5.1 Theory

Injector precision is determined by performing ten replicate injections of a standard using a fixed sample volume.

PDA/ VWD: A 40 ppm caffeine standard is injected when using absorbance detection at a wavelength of 272 nm. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

Conductivity Detector: A 50 ppm nitrate standard is injected when using conductivity detection. Deionized water is used as the eluent at a flow rate of 1.00 mL/min with conductivity or absorbance detection.

3.5.2 Performing the Test and Evaluating the Results

Injector precision evaluation is the next test in the sequence queue and the samples are called Inj Precision_1 through Inj Precision_10 in the XQ_Detector_Inj_Precision sequence, where X is O

for OQ, P for PQ, and Detector is CD for conductivity detectors and UV for absorbance detectors. The concentration of the nitrate or caffeine standard and the sample volume injected is included in the QNT file used for injector linearity.

Precision is determined by calculating the relative standard deviation of the peak areas for the ten injections.

3.6 Sample Temperature Accuracy of Autosamplers (AS-AP only)

3.6.1 Theory

The sample temperature accuracy mainly depends on the cooling and heating accuracy of the AS-AP autosampler, the insulation of the sample compartment, and the thermal transfer from the carousel to the vial.

3.6.2 Performing the Check

The sample temperature accuracy is determined with an external thermometer. The temperature sensor is placed in a standard polypropylene vial (10 mL) filled with water. The carousel cover must be closed during the test. The autosampler temperature is set to a nominal temperature (e.g., 10 °C), depending on the autosampler type. When the set temperature is reached, the sample (water) temperature is recorded over a period of 30 minutes. Within the 30 minutes, the sample temperature reaches a stable value. The temperature accuracy is the temperature difference between the sample temperature and the nominal autosampler temperature.

3.7 Sample Carryover

3.7.1 Theory

Sample carryover is determined by injecting a highly concentrated sample, followed by a blank sample and then a second blank injection. The blanks in this case are deionized water.

Conductivity Detector: Nitrate at a concentration of 1000 ppm is injected when this test is performed with a conductivity detector. Caffeine at a concentration of 80 ppm is injected when this test is performed with an absorbance detector.

Ideally, the chromatogram of the two blank injections should be identical. If, however, a difference is detected, the difference between the peak areas is calculated and the ratio of this difference to the highly concentrated sample is calculated. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

3.7.2 Performing the Test and Evaluating the Results

Sample carryover evaluation is included in the same sequence as the Injector Precision test. The samples are called Water, Carryover and Water_CO in the XQ_Detector_Inj_Precision_CO sequence.

A peak area ratio is calculated between the blank and high concentration sample to determine sample carryover.

3.8 Detector Linearity

3.8.1 Theory

VWD/PDA: Detector linearity for absorbance detectors is determined at 272 nm using a sequence of five caffeine standard injections at 15, 40, 60, 80 and 100 ppm. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

Conductivity Detector: Detector linearity for conductivity detectors uses a sequence of five nitrate standard injections at 5, 10, 25, 50, and 100 ppm. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

3.8.2 Performing the Test and Evaluating the Results

Detector linearity samples are called Detector linearity_1 through Detector linearity_5 in the *XQ_Detector_Linearity* sequence. The concentrations of the standards are included in the QNT file used for detector linearity. It is recommended to enter the actual concentration of the standards from the certification of analysis into the quantification files to achieve the real results.

Depending on which injection module used, the peak height of the sample with the highest concentration may exceed 1500 mAU. This is usually not within the linearity range of UV detectors. Thus, the limits for the regression coefficient and the relative standard deviation may not be met. In this case, reduce the injection volume for all samples used for the linearity check so that the peak height of the sample with the highest concentration is in the linearity range of the tested detector, i.e., usually below 1500 mAU. As an exception, a linearity range of up to 2500 mAU is specified for the supported VWD with an analytical flow cell installed.

If an autosampler is used when the PDA or PDA-100 detectors are tested, only 10 µL of sample will be injected by default. However, when qualifying the Thermo Fisher Scientific detectors using an autosampler, 15 µL of sample will be injected.

3.9 Injection Volume Linearity

3.9.1 Theory

Injection volume linearity is determined by making five injections of a standard using injection volumes of 10, 20, 30, 40 and 50 µL. A 60 ppm caffeine standard is injected when using absorbance detection at a wavelength of 272 nm. A 50 ppm nitrate standard is injected when using conductivity detection. Deionized water is used as the eluent at a flow rate of 1.00 mL/min. This test is only performed on autosamplers with variable injection volume capability (AS-AP, AS or AS50).

3.9.2 Performing the Test and Evaluating the Results

Injection volume linearity evaluation is run using the sequence called *XQ_Detector_Inj_Linearity*. The concentration of the nitrate or caffeine standard and the injection volumes are included in the QNT file used for detector linearity.

3.9 Sample Preparation Precision (AS only)

3.9.1 Theory

Sample Preparation precision is determined by making three separate injections using a fixed injection volume. Each of the three standards is prepared individually using the autosampler's sample preparation option. The autosampler prepares the three samples by first transferring 15 µL of a 1000 ppm caffeine or nitrate standard into three empty sample vials and then adding 1485 µL of water to each of the sample vials. Injections are then made from each vial. Deionized water is used as the eluent at a flow rate of 1.00 mL/min. If an absorbance detector is used the wavelength is set to 272 nm. This test is only performed on autosamplers with the sample prep option installed.

3.9.2 Performing the Test and Evaluating the Results

Sample prep precision evaluation is run using the sequence called *XQ_Detector_Samp_Prep*.

Required configuration for running a successful sample preparation experiment: Note: Sample Prep only supports 10 mL vials.

- AS configured with a 10.0 mL tray (P/N 026374).
- System is configured with a conductivity or an absorbance detector

Precision is determined by calculating the relative standard deviation (RSD) of the peak areas for the three injections.

3.10 Column Heater Temperature Qualification

3.10.1 Theory

The temperature of a column heater is qualified by measuring the plate temperature of the column heater after allowing the column heater plate and calibrated thermometer to equilibrate for a minimum of 15 minutes. The temperature is read off the calibrated thermometer, giving the temperature of the column heater plate. The temperature displayed by the front panel and the software is the calculated liquid temperature.

3.10.2 Performing the Test and Evaluating the Results

During the sequence queue the column heater is set to 40 °C. The Column Heater Temperature Qualification is performed after the completion of the sequence queue. The recorded plate temperature is entered into the report and automatically converted to liquid temperature.

3.11 Wavelength Accuracy of Absorbance Detectors

3.11.1 Theory

Wavelength accuracy is evaluated using a holmium oxide filter. Three holmium oxide absorbance maxima are compared with the theoretical maxima at 360.9 nm, 418.0 nm and 536.6 nm for ICS Series PDA and VWD detectors.

3.11.2 Performing the Test and Evaluating the Results

The wavelength accuracy test is run using the sequence called XQ_Wavelength_Accuracy where X is P for PQ and O for OQ. The Wavelength Accuracy Test program executes the verification and the results are logged to the audit trail by the Wavelength Accuracy Report program.

3.12 Sample pH Conductivity verification (AS-AP only)

3.12.1 Theory

The sample pH Conductivity qualification is evaluated by checking the pH calibration and pH accuracy. The sample temperature calibration is measuring the pH at 4, 7 and 10. The reading is stable if the reading and the three data points are within 0.2 pH unit of each other. The pH measurement of the accessory must be calibrated before you begin using the accessory and then periodically thereafter. During pH calibration, the pH of a pH 7 buffer solution is measured first and *the offset is determined*. Then, the pH of either a pH 4 or a pH 10 buffer solution is measured and a slope between pH 7 and pH 4 (or pH 10) is determined. Optionally, the pH of a third buffer solution (pH 4 or pH 10) can be measured and a slope between pH 7 and the third buffer determined. The conductivity calibration procedure consists of determining the conductivity zero point of the accessory and then measuring the conductivity of a 1000 µS standard. Chromeleon determines the calibration offset and slope and calculates a new cell constant.

4 Qualification Preparation

4.1 Qualification Kit, Part Number 063540

Before starting the qualification procedure confirm that all the materials needed are present.

Table 3. Components of OQ/PQ Kit, P/N 063540

Part Description	Part Number	Quantity
OQ/PQ User's Manual Part B Version 7.10 (this document)	065448 Part B	1
Tubing, 0.010" (0.25 mm) ID x 60" (150 cm) length black PEEK <ul style="list-style-type: none"> Connect pump to injection valve, backpressure tubing to detector, etc. 	042690	60" (152 cm)
Red PEEK Tubing 0.005" (0.125 mm) ID x 39" (99 cm) length <ul style="list-style-type: none"> Generate system backpressure and delay volume for detector and injector tests 	044211	39" (99 cm)
Yellow PEEK Tubing (0.003", 0.075 mm ID) <ul style="list-style-type: none"> Generate system backpressure and delay volume for detector and injector tests in Analytical Systems only 	049715	39" (99 cm)
25 µL Sample Loop <ul style="list-style-type: none"> AS-DV/Manual injection 	042857	1
100 µL Sample Loop <ul style="list-style-type: none"> AS-AP, AS 	042951	1
<ul style="list-style-type: none"> 3- Way mixing tees Connect eluent lines and eluent bottles 	024314	2
High-Pressure, fittings bolt, 10-32	074449	6
High-Pressure, ferrules double cone 10-32	074373	6
Qualification labels	050837	6
Certificate of Functional Performance	031423	6
NIST Traceable Caffeine Standards Kit <ul style="list-style-type: none"> Absorbance detectors 	060253	1
NIST Traceable Nitrate Standards Kit <ul style="list-style-type: none"> Conductivity detectors 	060254	1
Test Binder	035863	1
Restrictor Microbore, Gradient Test	049630	1

Table 4. Required Kit for ICS configurations

Part Description	Part Number
IC OQ/PQ Kit for IC systems (PROD,KIT,OQ/PQ,ICS,CD ABS)	063540

4.2 Additional Required Materials

The following items will be needed:

Table 5. Additional Materials Needed

Description	Quantity
ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered) <ul style="list-style-type: none"> All eluent, standard, and sample preparation 	2 L
Reagent Grade or better Potassium Chloride for OQ/PQ gradient tests using conductivity detection <ul style="list-style-type: none"> Gradient pump tests using conductivity detectors 	1 g
HPLC grade Acetone for OQ/PQ gradient tests using optical absorbance detection <ul style="list-style-type: none"> Gradient pump tests using optical detectors 	3 mL
Sodium Hydroxide 50/50 w/w (Low Carbonate) (Thermo Fisher Scientific P/N 033465) <ul style="list-style-type: none"> Prepare eluent for Integrated Amperometry qualification 	~5.2 mL
Ammeter or digital volt multimeter (DVM) with milliamp (300 mA scale) setting for OQ/PQ tests using an EG. Must be capable of displaying 0.00 to at least 220 mA. A Fluke Model 189, 289 or equivalent DVM will allow for automated EG Current testing.	1
Thermo Fisher Scientific analytical conductivity cell (detector) <ul style="list-style-type: none"> Detector, injector and gradient pump tests using conductivity detectors in conductivity mode 	1
AS or AS-AP in pull mode: 10 mL vials are required for AS-AP pull mode and AS Sample Preparation Qualification	10
AS-AP: 1.5 mL vial tray (P/N 066332)	1
AS-AP: 1.5 mL vials (P/N 061696)	7
Column Thermostat PQ Kit (P/N 5705.0050A)	1
Type K temperature sensor for P600 thermometers (P/N 6820.0010)	1
Black tubing 48" (P/N 042690)	1
AS-DV 5.0 mL vials with filter caps (P/N 038141) if qualifying an AS-DV autosampler	20
100 mL volumetric flasks (Class A, 100 ± 0.08 mL) <ul style="list-style-type: none"> Standard preparation for AS-DV and Manual Injection 	1
1000 mL volumetric flasks (Class A, 1000 ± 0.3 mL) <ul style="list-style-type: none"> Eluent preparation 	At least 1
Transfer Pipettes or Pipettor	0.5 – 10.0 mL
Calibrated Analytical Balance - capable of mass recording to the nearest milligram or better <ul style="list-style-type: none"> Weigh KCl for eluent preparation Weigh eluent for the Flow Rate Accuracy and Precision test 	1
Calibration and Check Standard pH 4 (P/N 033238)	1
Calibration and Check Standard pH10 (P/N 033240)	1
1000 uS Standard (P/N 080172)	1
Computer workstation running Chromeleon™ Version 7.10 (with SR1) or later (later versions required for some configurations, see Chapter 1.5)	1

De-ionized water should be vacuum filtered through a 0.2 µm or 0.45 µm nylon or other water compatible membrane exhibiting very low extractable.

All volumetric ware, pipettes and standard eluent storage bottles should be made of low extractable polymeric materials such as polypropylene or polystyrene.

4.3 Preparation of Standards

Two standards kits are provided in the OQ/PQ kit. Use the Caffeine Kit, Part Number 060253 to qualify systems with an absorbance detector (AD25, ICS Series PDA or VWD, PDA-100). Use the Nitrate kit, Part Number 060254 to qualify systems with a conductivity detector.

4.3.1 Caffeine standard preparation for the Injector Precision and Carryover tests

AS-DV and AS40 Autosamplers only:

AS-DV Autosampler only: For the Injector Precision test prepare a 40 ppm caffeine standard using the 1000 ppm caffeine standard ampoule included in the Caffeine Standards Kit. Pipette 4.0 mL of the 1000 ppm caffeine standard into a 100 mL volumetric flask. Fill the flask to the mark with ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered).

AS-DV Autosampler and Manual Injection: For the Carryover test use the 80 ppm caffeine standard included in the Caffeine Standards Kit.

4.3.2 Nitrate standard preparation for the Injector Precision test (AS-DV autosampler and manual injections only)

For the Injector Precision test prepare a 50 ppm nitrate standard using the 1000 ppm nitrate standard ampoule included in the Nitrate Standards Kit. Pipette 5.0 mL of the 1000 ppm nitrate standard into a 100 mL volumetric flask. Fill the flask to the mark with ASTM Type 1 (or better) deionized water (18.0 Megohm-cm, filtered).

For the carryover test use the included 1000 ppm nitrate standard found the in the nitrate standards kit without dilution.

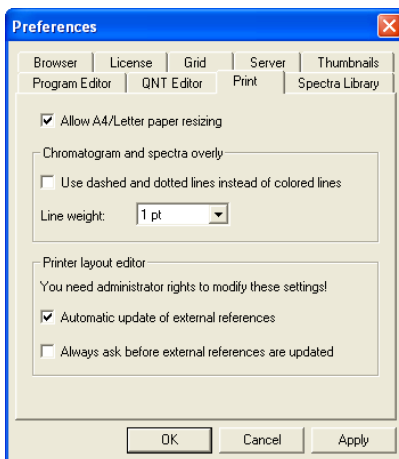
4.4 Preparation of Eluents

All eluents must be prepared using ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered). Eluent should be freshly prepared.

4.5 Preparing the IC System

Note: The AD25, VWD, and PDA UV and visible lamps must be turned on at least two hours prior to performing the qualification.

1. Chromeleon settings: Ensure that Chromeleon is set for Automatic update of external references. Click on the "Automatic update of external references" check box in File → Preferences → Print menu.



2. Open the Chromeleon Instrument Configuration application. Make note of any changes made to the system for the purposes of this qualification. After qualification these changes should be reversed. It is helpful to save a backup file of the configuration before you make any changes for qualification.
3. **AUTOSAMPLER:** Double click on the Autosampler to be qualified (if installed) to view the properties page for the autosampler. Select the **General** tab and confirm the Device Name is "Sampler". Select the **Options** tab and confirm the options match the configuration of your system. The AS-AP injection mode must be selected at this point. Only one mode is qualified. Select the **Devices** tab and confirm the Autosampler diverter valve status matches your system. (An AS40 autosampler is controlled only by relay, and does not show up as a device in the instrument configuration). For the AS-AP autosampler in pull mode, the Flush Volume and Flush Volume2 must be set with F8 commands or from the **More Options** page of the AS-AP panel before beginning qualification. Flush Volume = 2 x (Needle Volume + Needle Line Tubing Volume) μ L where Needle Volume = 30 μ L and Needle Line Tubing Volume (the volume of tubing between the needle and the injection port) = 0.86 μ L/cm (2.2 μ L/in) of 0.33-mm (0.013-in) ID PEEK tubing (blue). Set the Flush Volume2 parameter to 30 μ L.

NOTE: Make sure inject mode in the panel is consistent with the inject mode in the server configuration before loading the OQPQ templates. In addition, the temperature control must be checked in the instrument configuration and the panel in order to run the sample temperature accuracy test in the sequence.

4. **ICS-2000, ICS-2100:** Double click on the ICS system to be qualified to view the properties page for the system. Select the General page and confirm the Device Name is "Pump_ECD". Select the Options page and confirm the Installed Options match the configuration of the system. If a degasser is installed, ensure it is not in Cycle mode for the duration of the qualification. Select the Signals page and confirm the ECD_1 signal is active, the unit is μ S and the factor is set to 1.000.

Select the **EluentGenerator** page and confirm the EGC-1 or EGC_2 cartridge is installed, the Device Name is "EluentGenerator" and a valid eluent generator cartridge serial number is entered. The user must rename EGC-2 to EGC-1 in the instrument configuration if the Qualification test box is connected to EGC-2.

5. If applicable make sure the detector Output is in 'Offset' mode in the Analog Out Set-Up screen on the front panel.
6. Make note of any changes and save the configuration.
7. All modules, with the exception of the AS40, must have their serial number set in the moduleware. Qualification of modules without serial numbers, with the exception of the AS40, is not supported.

8. Connect the eluent bottle to the pump and prime the eluent line for approximately 3 minutes; at least 10 minutes if the vacuum degas option is installed.
9. Connect the pump line to injection valve port 2 (P). Bypass the column, suppressor, CR-TC, and CRD (if any are installed) from the system leaving the gradient mixer in-line; use the backpressure coil to connect the pump outlet directly to port 2 on the injection valve. This is illustrated in Figure 3 below. If an Eluent Generator Cartridge is in-line with the IC system, bypass all plumbing to the EGC and connect the pump directly to the injection valve.
10. ICS-2000 or ICS-2100: Disconnect the Pulse Damper Out line (0.030" tubing exiting from left and below the detector cell) and connect it directly to the Injection Valve port 2 (P), thus bypassing the eluent generator.

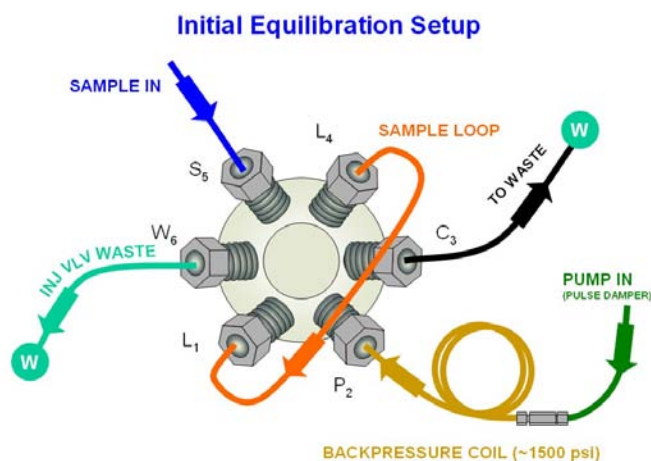


Figure 3. Plumbing diagram, initial equilibration step

11. ICS-1000/1100 (WITH COLUMN HEATER INSTALLED), ICS-1500/1600 and ICS-2000/2100. Open and remove the column heater plastic cover by removing the two thumbscrews. Remove the column heater front plate by removing the six thumbscrews. Unclip and remove the column and insert the thermometer probe in its place so that the LCD display sits out the top of the column heater with the probe lowered into the column clips. Replace the column heater front plate and the plastic cover, leaving the thermometer LCD display out of the top of the cover and the probe in the column heater assembly. The LCD display of the thermometer should be visible and display the current column heater plate temperature. (Column heater plate temperature may vary slightly from the column heater liquid temperature).
12. Thoroughly rinse the autosampler fluidics with water. This can be accomplished on the AS-AP, AS or AS50 by flushing the sample syringe two or three times and on the AS40 or AS-DV by running two or three rinse vials.
13. AS-AP, AS, AS50: For systems using an AS-AP, AS or AS50, install the 100 μ L sample loop on the injection valve. For the AS-AP, Loop volume must be set to 100 μ L in instrument configuration.

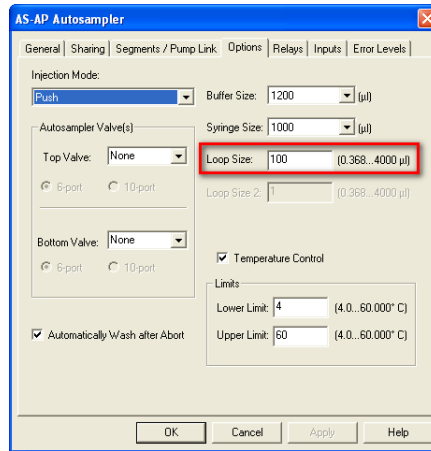


Figure 4. AS-AP Instrument Configuration

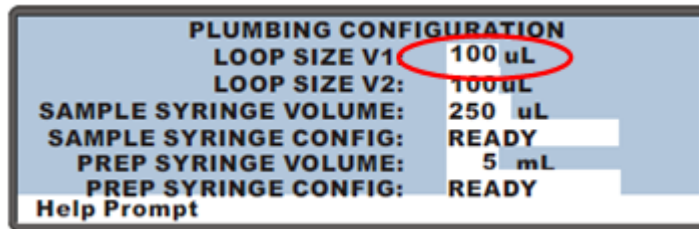


Figure 5. AS Autosampler LCD Control Panel Display

Ensure that the Loop SIZE V1 volume is changed to 100 µL

From the front panel of the AS or AS50, go to the Main Menu, select Module Set-Up and then Plumbing Configuration. If necessary change the sample loop size to 100 µL.

14. AS40, AS-DV or manual injector: For systems using an AS40 Autosampler or manual injector, install the 25 µL sample loop on the injection valve. The AS40 Load connectors of the relay control cable (P/N 047946) should be connected to **Relay_1** on integrated systems. This is illustrated below in Figure 4. The AS-DV connects to the Chromeleon PC via a USB cable.

Note: All programs using the AS40 are designed to run with full vial sampling from the 5.0 mL vials (i.e. trigger relay at -2.40 minutes). If 0.5 mL vials are to be used programs will have to be manually modified to trigger the relay at -1.20 minute).

If using 5.0 mL vials, you will need 1 Nitrate standards kit to complete the OQ or PQ.

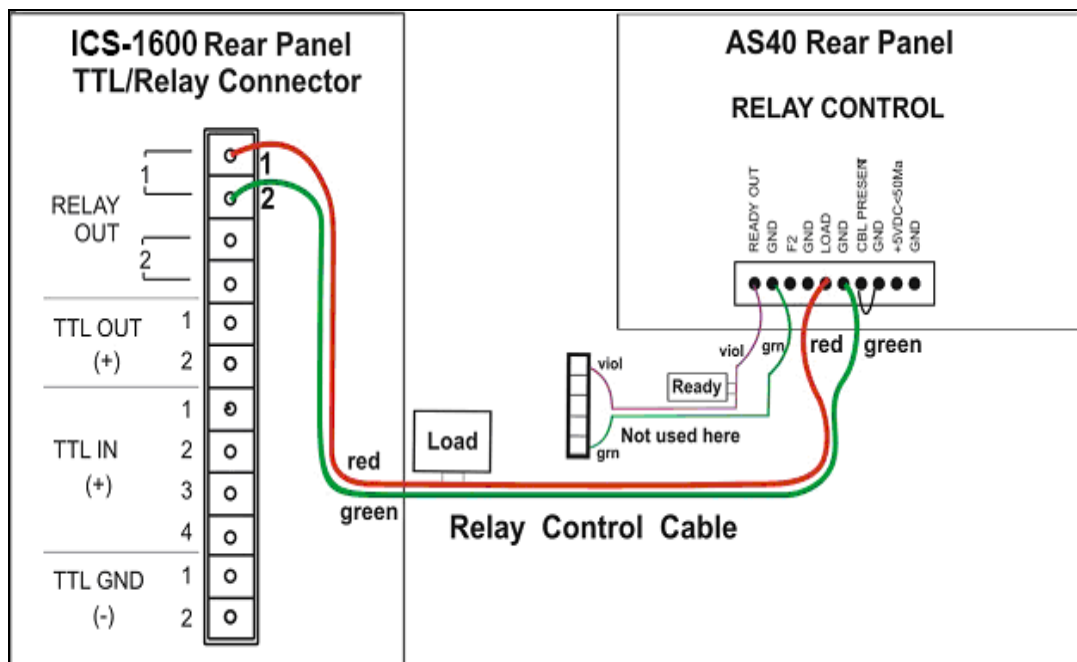


Figure 6. Remote Control Cabling to AS40 Autosampler

15. Set the flow rate to 1.00 mL/min. Turn the pump flow on and let the system equilibrate for a minimum of 15 minutes or longer as necessary until the system has equilibrated by monitoring the detector output for a stable response. If a degasser is present ensure it is not in cycle mode.
16. **NEW TUBING:** When running a qualification on a system with new tubing, such as a new backpressure coil or a new sample loop, it is recommended to rinse the tubing with carbonate eluent before proceeding with the qualification. This can be achieved by injecting a 0.5 M Sodium Carbonate solution two or three times while running deionized water at 1.00 mL/min, or by running 9 mM Sodium Carbonate as the eluent for 10 minutes before rinsing with deionized water for 20 minutes. Since ICS-1100, 1600 and 2100 systems ship with pre-rinsed tubing, it is only necessary to carry out this step when new pieces of tubing have been added.

To prepare 0.5 M sodium carbonate dissolve 0.53 g of sodium carbonate in water to make a total volume of 10 mL of solution. To prepare 9 mM sodium carbonate dissolve 0.955 g of sodium carbonate in water to make 1 L of solution.

17. **CONDUCTIVITY DETECTOR:** Pipette the standards from the standards kit vials directly into the 1.5 mL or 10 mL autosampler vials and place them in the position / order according to Table 5, 6 or 7 below. For the Injector Precision and Carryover standards (AS40 or AS-DV Autosamplers only) transfer the Injector Precision standards prepared in Section 4.3 into 10 autosampler vials. To qualify an IC system without an autosampler, use the AS40/AS-DV table for the injection order and inject the samples directly from the standards kit vials.
18. For systems using an ICS-2000/ICS-2100 EG:
 - a. Disconnect the Eluent Generator Cartridge cable from the Eluent Generator EGC connector.
 - b. If installed, disconnect the CR-TC cable from the CR-TC connector.
 - c. Plug the EG/AES/SRS Supply Input cable (4-pin connector) on the IC Qualification Test Box into the Eluent Generator EGC connector.

Note: This test requires the IC Qualification Test Box. EG Qualification Test Box is no longer supported for OQ/PQ qualifications.

- d. Turn on the ammeter or multimeter and set the display to mA. This test requires mA readings from 0.00 to at least 220 mA.
- e. Connect the positive lead of the ammeter to the red post connector labeled "EG Internal Load" on the IC Qualification Test Box.
- f. Connect the negative lead of the ammeter to the black post connector labeled "COM" on the IC Qualification Test Box.
- g. From the console in the Chromeleon software, go to the Panel directory and select the pump_EluGen_detector panel appropriate for the system being qualified. Go to Control and select the instrument configured with the Eluent Generator to be qualified (or click on the "Default Panel Tab set").
- h. From the Eluent Generator section of the panel, select the cartridge type to be used. The panel will recognize previously configured cartridges even though the IC Qualification Test Box is plugged into the cartridge connector.

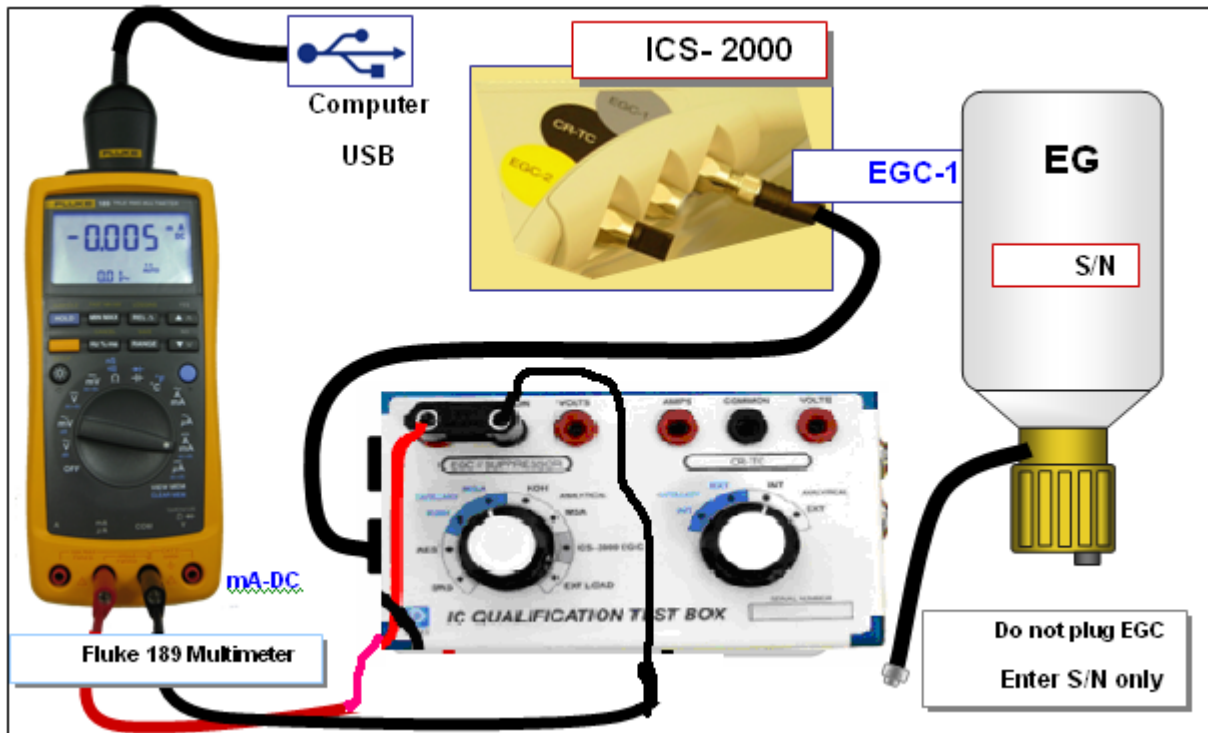


Figure 7. IC Qualification Test Box and Fluke Multimeter Setup

19. **FLUKE 189/289 MULTIMETER:** Install the Fluke 189/289 multimeter or equivalent into the instrument to be qualified. The driver for the Fluke 189/289 multimeter is included with Chromeleon 6.8 or later. Connect the Fluke 189/289 multimeter to the IC Qualification Test Box as described in step 18 of this section. The Fluke 189/289 multimeter is only used for systems that require EG current qualification. It is recommended that the "Power off time out" for the Fluke189/289 be set to a value greater than the time it will take to perform the Operational Qualification. See Fluke189 or 289 operator's manual for instructions on how to set this value.

- a. It is important that the Fluke 189/289 multimeter is installed AND connected before loading the OQ/PQ templates.
- b. Follow the instructions in the Chromeleon Help topics for the installation of the Fluke 189/289 multimeter Windows USB driver and for the Chromeleon device driver for the Fluke 189/289 multimeter.
- c. Follow the Fluke 189/289 multimeter instructions for adjusting the default 'PrOFF' setting in the meter. It is recommended to adjust this parameter to a longer time (e.g., several hours or more) to avoid having the meter auto power off during your OQ/PQ tests.

Table 6. AS/AS50 Autosampler Sample Order

Conductivity Detector with AS	Absorbance or Photodiode Array Detector with AS/AS50	Vial Tray Position for AS
Water	Water	1
5 ppm Nitrate Std		2
10 ppm Nitrate Std		3
25 ppm Nitrate Std		4
50 ppm Nitrate Std		5
100 ppm Nitrate Std		6
1000 ppm Nitrate Std		7
Empty Vial*		8
Empty Vial*		9
Empty Vial*		10
	15 ppm Caffeine Std	11
	40 ppm Caffeine Std	12
	60 ppm Caffeine Std	13
	80 ppm Caffeine Std	14
	100 ppm Caffeine Std	15
	1000 ppm Caffeine Std	16
	Empty Vial*	17
	Empty Vial*	18
	Empty Vial*	19

- Vials only need to be present if the detector listed at the top of the column is to be qualified

*Needed only if qualifying AS/AS50 Sample Preparation Option. **10 mL vials are required for Sample Preparation Qualification.**

Table 7. AS-AP Autosampler Sample Order

Conductivity Detector with AS-AP Push/Pull Mode	Absorbance Detector with AS-AP Push Mode	Vial Tray Position
Water	Water	BA1
5 ppm Nitrate Std	15 ppm Caffeine Std	BA2
10 ppm Nitrate Std	40 ppm Caffeine Std	BA3
25 ppm Nitrate Std	60 ppm Caffeine Std	BA4
50 ppm Nitrate Std	80 ppm Caffeine Std	BA5
100 ppm Nitrate Std	100 ppm Caffeine Std	BB1
1000 ppm Nitrate Std	1000 ppm Caffeine Std	BB2

Note: AS-AP pull mode requires the use of 10 mL vials.

Note: To run sequences with 1.5 mL vials, it is important to set the puncture offset to 3 mm in the commands before you start. The puncture offset commands allows the AS-AP to realign the x-axis of the 1.5m/10 mL vial setting.

Table 8. AS/ AS50 Autosampler Sample Order

Qualification	Conductivity Detector with AS-DV/AS40	Absorbance Detector with AS-DV/AS40	Sample Order
Warm Up	Water	Water	Vial 1
Injector Precision	50 ppm Nitrate Std*	40 ppm Caffeine Std*	Vial 2
	50 ppm Nitrate Std*	40 ppm Caffeine Std*	Vial 2
	50 ppm Nitrate Std*	40 ppm Caffeine Std*	Vial 2
	50 ppm Nitrate Std*	40 ppm Caffeine Std*	Vial 3
	50 ppm Nitrate Std*	40 ppm Caffeine Std*	Vial 3
	50 ppm Nitrate Std*	40 ppm Caffeine Std*	Vial 3
	50 ppm Nitrate Std*	40 ppm Caffeine Std*	Vial 4
	50 ppm Nitrate Std*	40 ppm Caffeine Std*	Vial 4
	50 ppm Nitrate Std*	40 ppm Caffeine Std*	Vial 4
	50 ppm Nitrate Std*	40 ppm Caffeine Std*	Vial 5
Carryover	1000 ppm Nitrate Std	80 ppm Caffeine Std*	Vial 12
	Water	Water	Vial 13
	Water	Water	Vial 14
Detector Linearity	5 ppm Nitrate Std	15 ppm Caffeine Std	Vial 15
	10 ppm Nitrate Std	40 ppm Caffeine Std	Vial 16
	25 ppm Nitrate Std	60 ppm Caffeine Std	Vial 17
	50 ppm Nitrate Std	80 ppm Caffeine Std	Vial 18
	100 ppm Nitrate Std	100 ppm Caffeine Std	Vial 19

*AS40/AS-DV users: For the Injector Precision and Absorbance Carryover samples, use the standards prepared in Section 4.3 of this manual.

4.6 OQ/PQ Sequence Installation

For performing OQ and PQ checks in Chromeleon 7.1, it is not required to create and copy the sequence templates from a Chromeleon CD. An Instrument Qualification Wizard automatically performs these steps. The wizard creates the sequences to be run. No instrument-specific sequence templates are created.

If using an AS autosampler configured for sequential injections, manually put the diverter valve in the correct position for the Instrument.

Tip: Please note that Chromeleon 7 terminology is different from the terminology used in Chromeleon 6.80. For details, refer to the 'Glossary - Chromeleon 7,' which is available in the Documents folder of your Chromeleon 7 installation.

Go to Tools → Instrument Qualification to open Instrument Qualification Wizard. For this step, Chromeleon 7.1 must display the instrument pane with the system to be tested selected (Figure 9) otherwise the error “Instrument not connected” will occur.

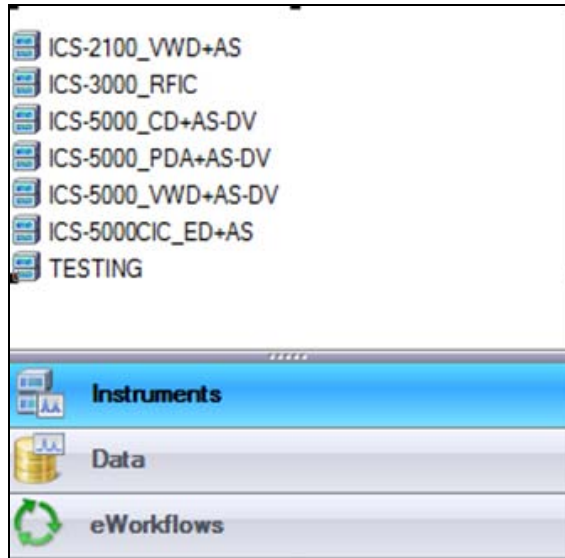


Figure 8. Instrument Pane for Chromeleon 7.1

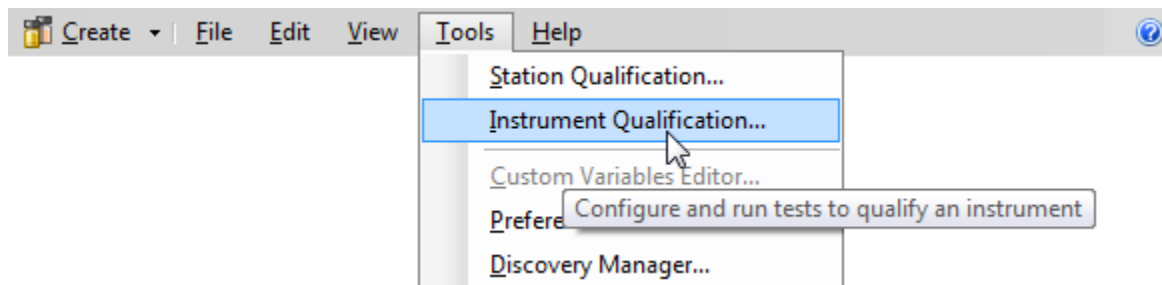


Figure 9. Starting the Instrument Qualification Wizard for Chromeleon 7.1

Select the qualification type: Installation (qualification of the installation), Operational (qualification in the working environment), or Performance Qualification (qualification during routine operation)

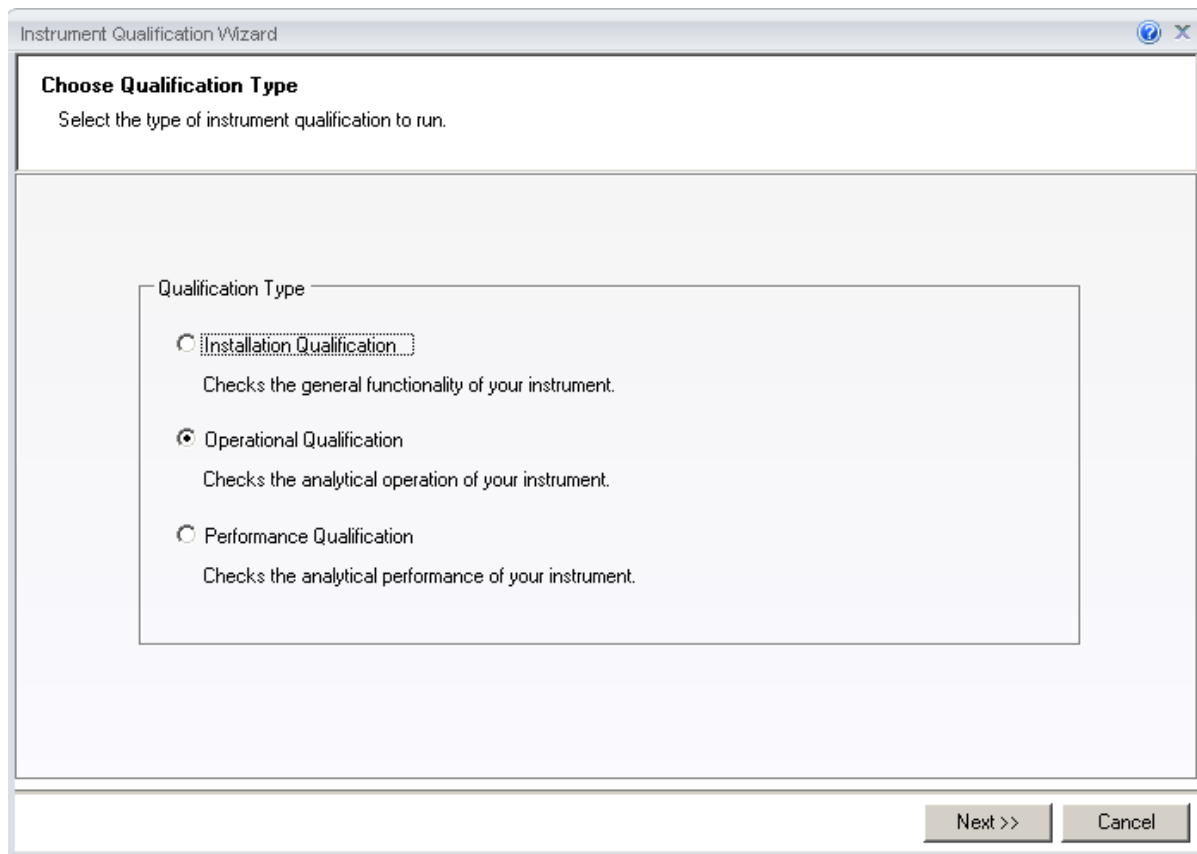


Figure 10. Selecting the qualification type for Chromeleon 7.1

Select the instrument to qualify.

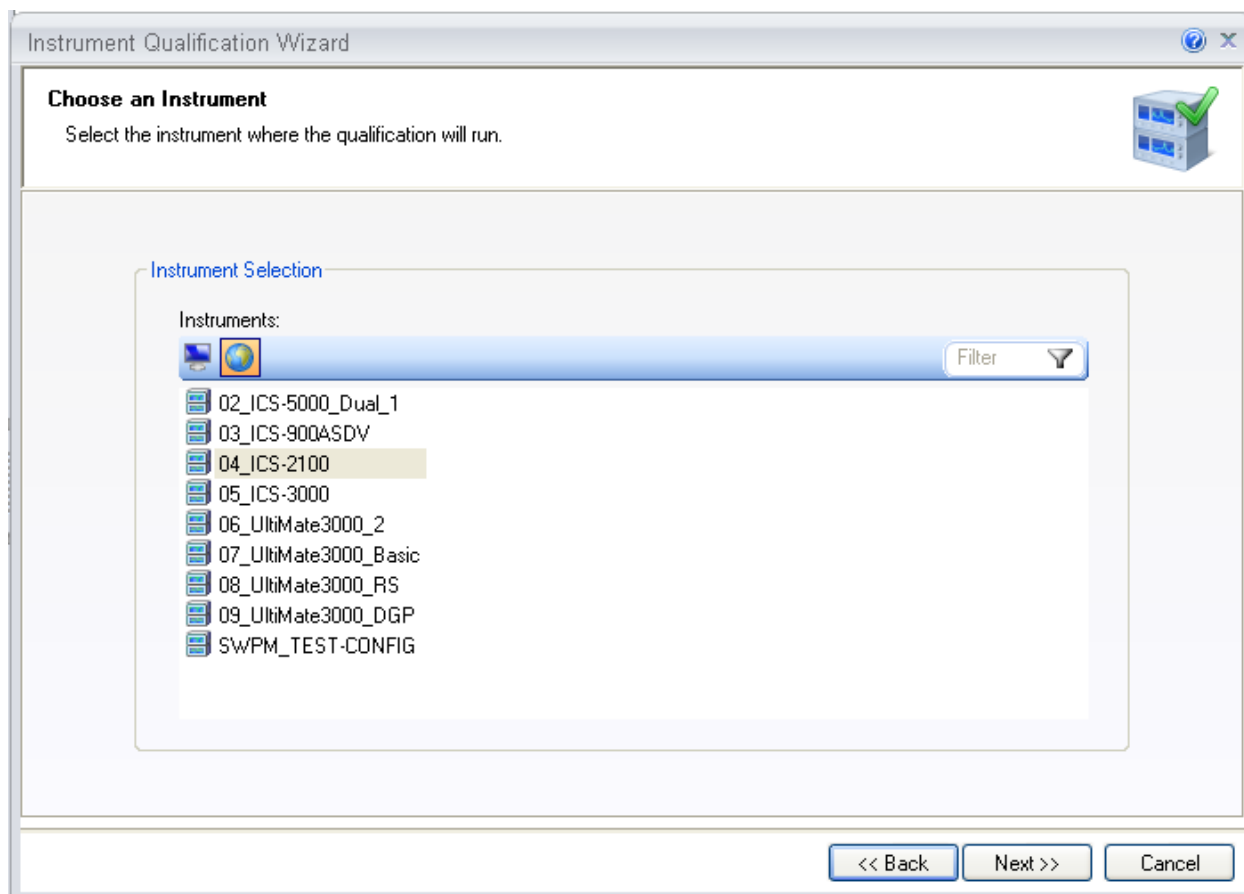
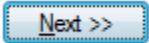


Figure 11. Selecting the instrument

Click  to connect the selected instrument to Chromeleon.

A list of sequences (tests) is displayed. The list is adapted to the instrument configuration of the selected instrument as defined in the Chromeleon Instrument Configuration Manager.

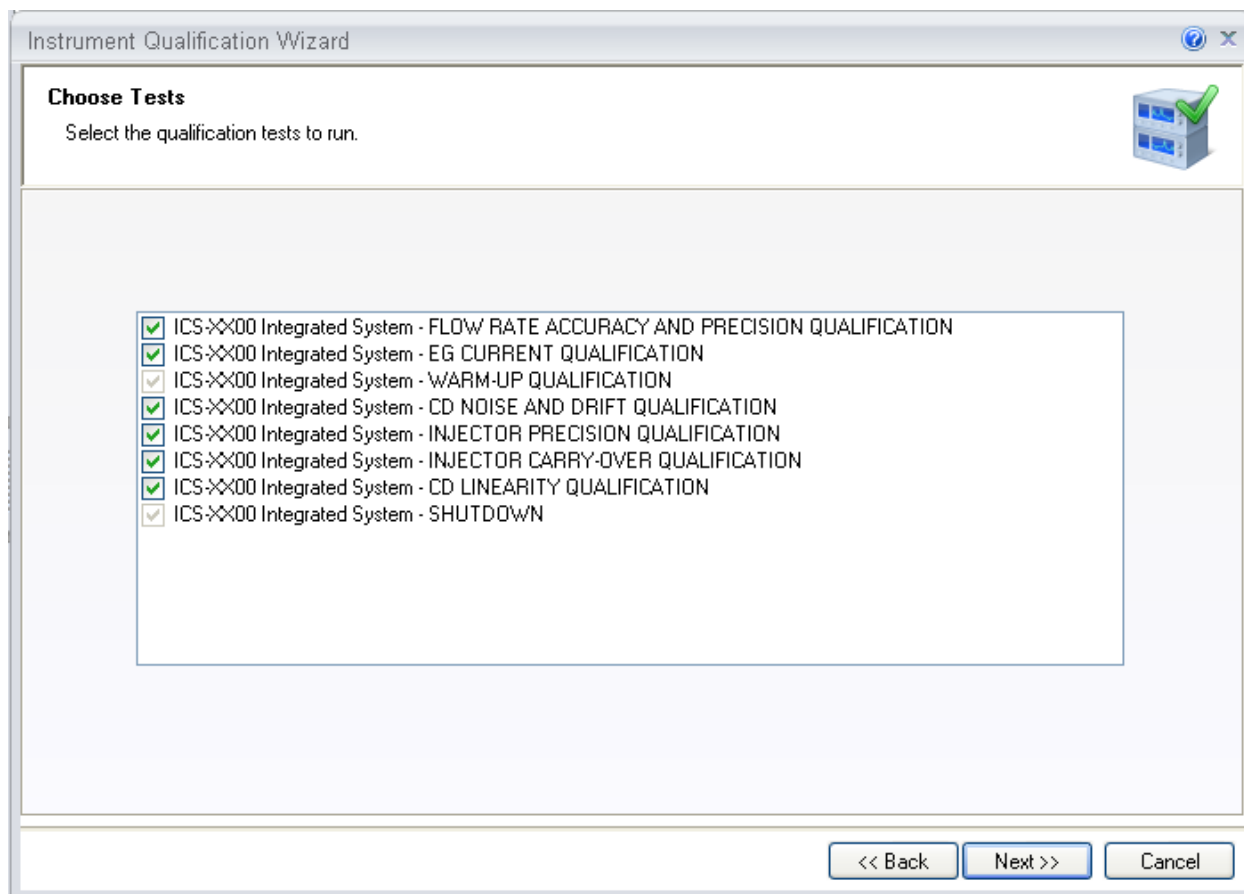


Figure 12. List of sequences for the selected instrument

Select the sequences that needed for the tests to be performed. Mandatory tests, such as "**Warm up**", are shown in the list, but the selection cannot be changed.

On the last wizard page, select a unique name under which the OQ and/or PQ sequence directory for this instrument is saved.

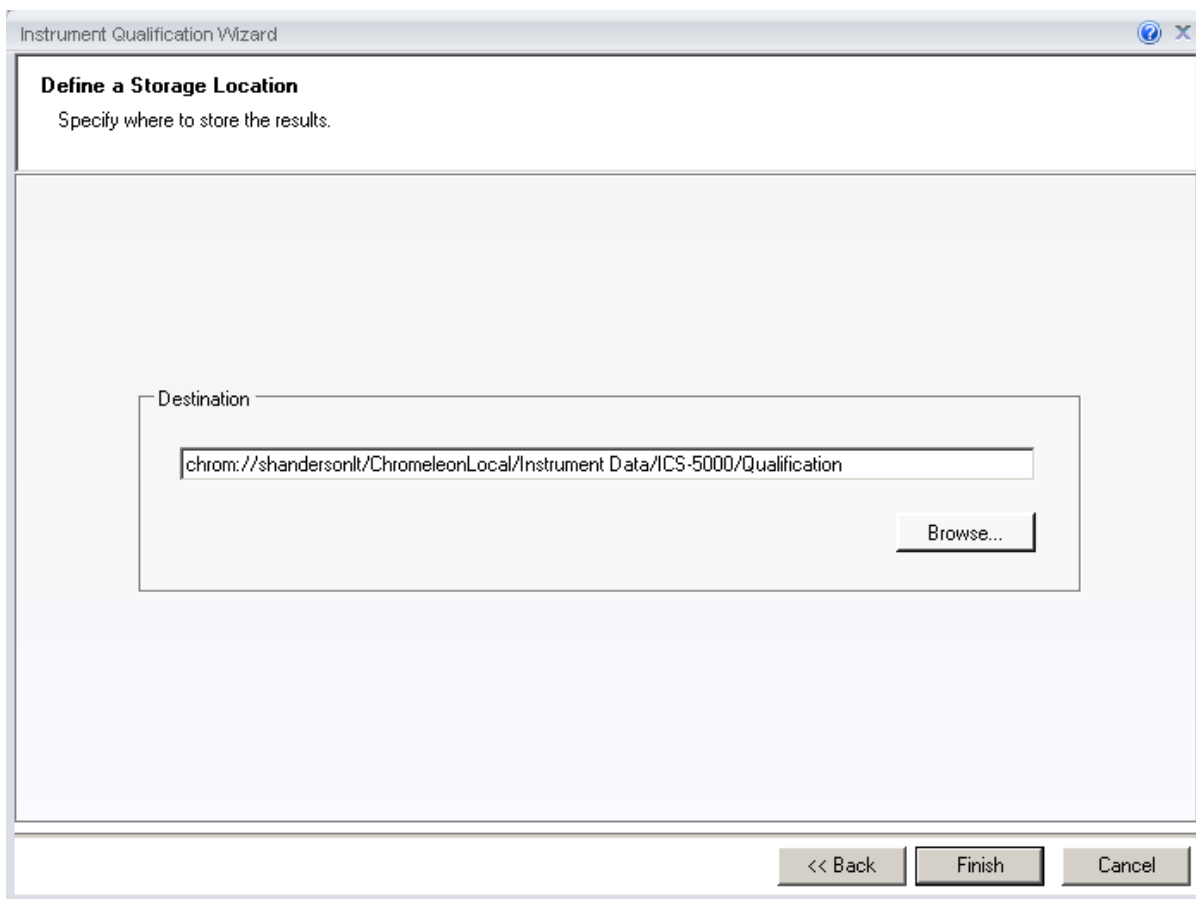


Figure 13. Selecting the storage location

4.7 OQ/PQ Sequence Execution

When the qualification wizard has been completed, the selected sequences are created and are automatically added to the instrument queue. A progress window shows which steps have been performed:

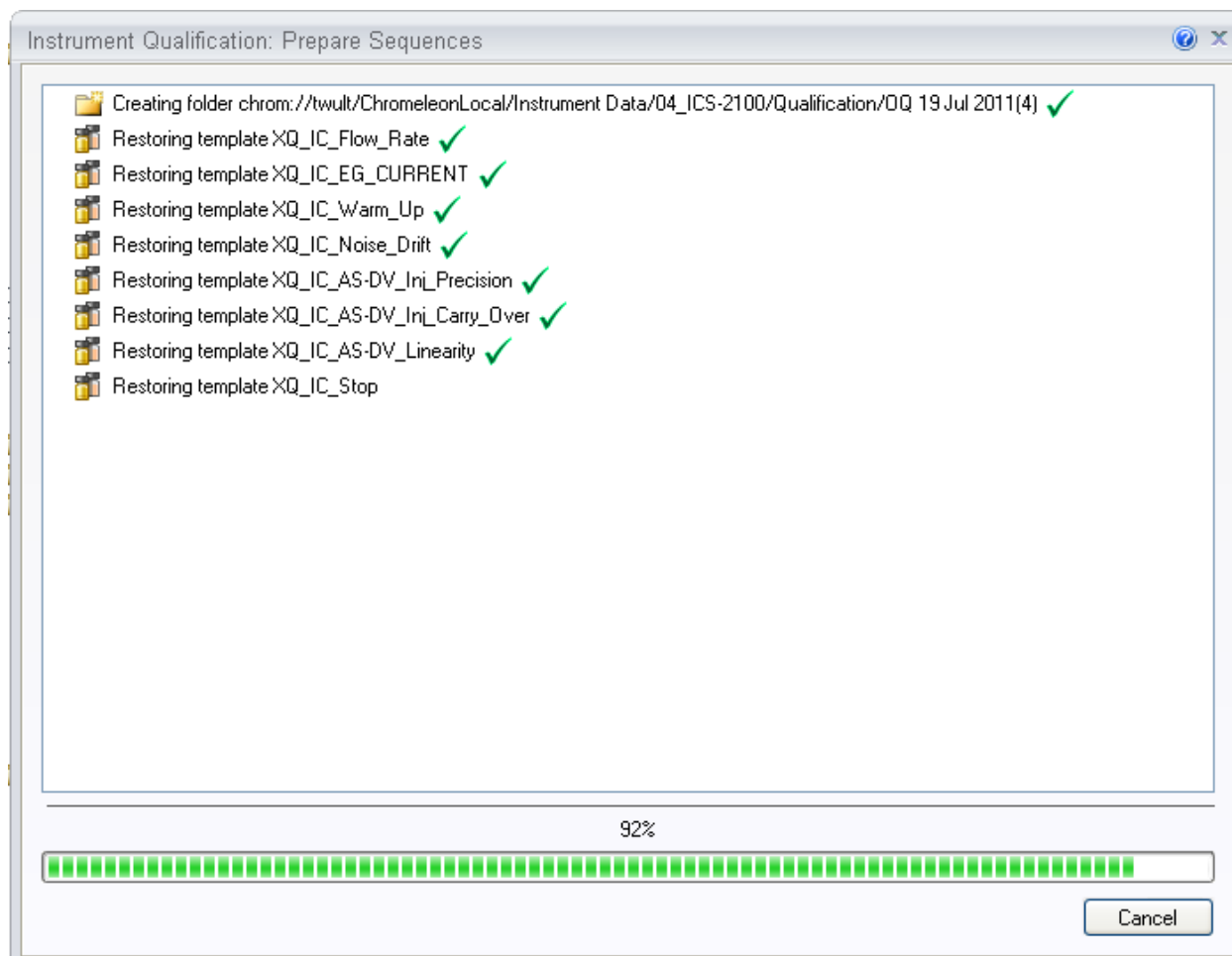


Figure 14. Progress during sequence creation

The Instrument View dialog box will open, displaying the Queue tab. As soon as the queue is started, Chromeleon runs the sequences. The wizard should copy the sequences to the destination selected then auto-populate the instrument queue. If the error “Instrument not connected” appears, make certain that the Chromeleon 7.1 must be displaying the instrument pane with the system to be tested.

The OQ/PQ qualification tests are performed in the following order:

- a. Flow Rate Test
- b. Eluent Generator Current Test (only if an EG is present)
- c. Warm Up
- d. Wavelength Accuracy

- e. Detector Noise and Drift Test
- f. Injector Precision Test
- g. Carryover Test
- h. Detector Linearity Test
- i. Injector Linearity Test (only if an AS or AS-AP is present)
- j. Sample Preparation Precision Test (only if an AS is present with the Sample Prep Option and the 10 mL Tray Type installed with the conductivity or the absorbance detector)
- k. Gradient Accuracy Test (only if an analytical gradient pump is present)
- l. Column TC and/or Compartment TC
- m. Sample Temperature Accuracy of Autosamplers (only for AS-AP Autosamplers)

Note: Test 'a' and 'l' is done manually and does not require a sequence template. The Instrument Method will pause before the Eluent Generator Current Qualification Test and the Flow-Rate Accuracy and Precision Test as these tests require user intervention.

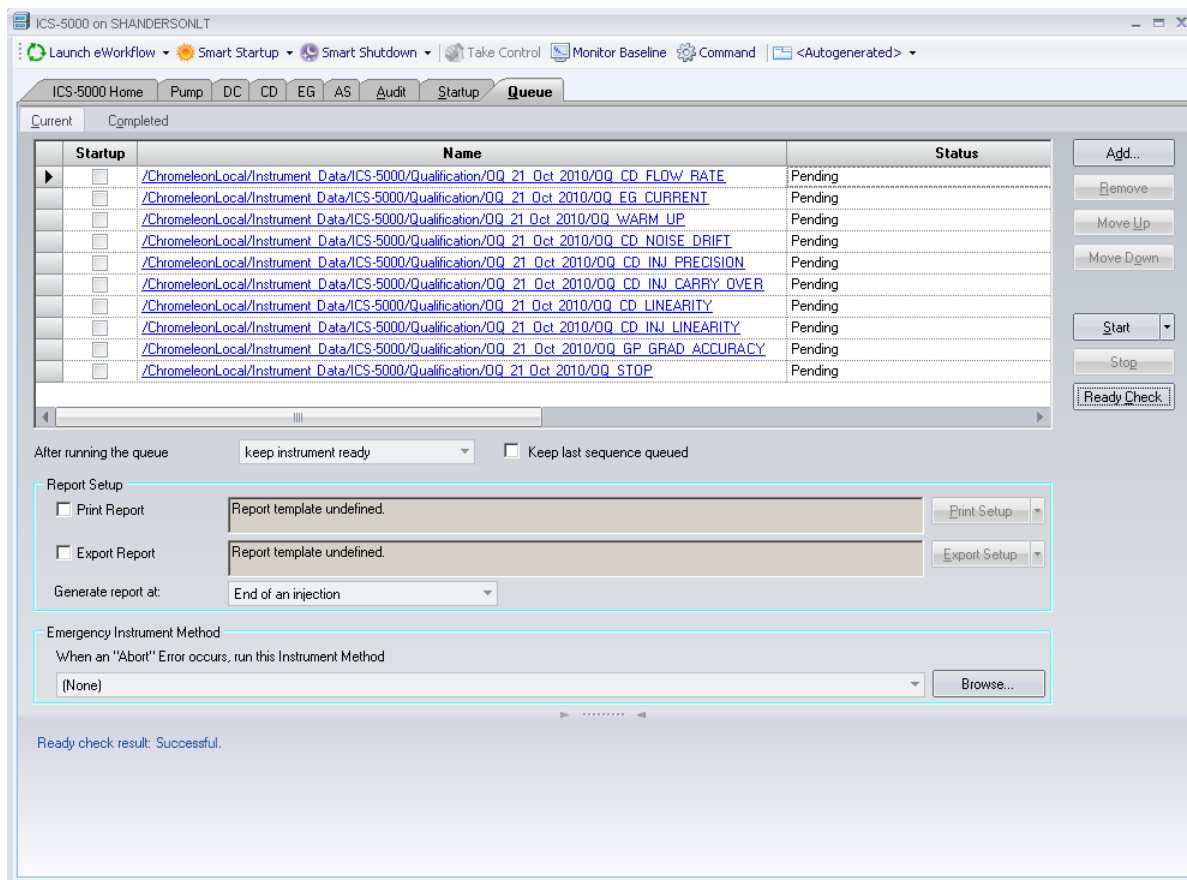


Figure 15. Sequence queue for Chromeleon 7.1

Evaluating the Test Sequences

The qualification sequences are saved under the path that was selected in the wizard. Each sequence also includes a separate report template.

To edit the report, open the report and remove the protection (if any) from the SPECIFICATION sheet. Enter the following information:

Name of the customer and name of the tester

Sample information such as queue number, expiration date, and actual concentration of the standard.

Name of the item that is used to generate the backpressure [default: capillary (L: 15 m; ID: 0.18 mm)]

Do not change any of the other report sheets. The report contains many references between data sheets. If you insert or delete lines and columns, these references will be lost and the calculations will be wrong.

To make sure that Chromeleon reads and processes the data in the report correctly, always print the report from the Chromeleon Console.

In the Data category, right-click the sequence for which you want to print the report, and then click **"Print Report"**.

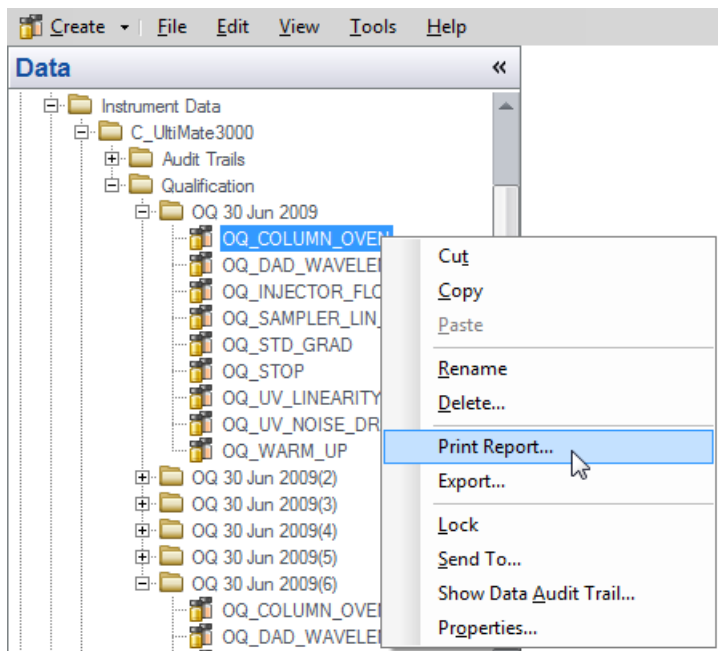


Figure 16. Selecting Print

Note: If an Eluent Generator is configured on the system, the following message may occur at the Ready Check: “Flow rate not specified at time 0.0, using a flow setting 1.00.” This refers to the Eluent Generator flow rate, not the pump flow rate. It is not necessary to take any action since the Eluent Generator is not used for this part of the qualification. Proceed with starting the queue.

Note: For systems with a shared autosampler there might be a number of *informational* messages indicating that “Exclusive Access” has not be acquired or released. This is normal and occurs because the qualification does not release the autosampler until the qualification is complete.

Repeating Individual Tests

It may be necessary to repeat individual tests. If so, review Section 6 where problems are described according to which individual tests might fail. According to Good Laboratory Practice (GLP), all tests following the one that failed must be repeated. The entire OQ/PQ qualification is designed such that almost of the performance tests require that the previous one to have passed successfully.

For example, if the test regarding the linearity of a CD detector fails, the results regarding the linearity of the injector are questionable as the linearity of the detector is a prerequisite for testing the injector linearity.

Table 9. Sequence File Names and description

Sequence Name	Check
XQ_Flow_Rate	Pump flow-rate accuracy and precision tests for ICS-3000 or ICS-5000 pumps
XQ_EG_Current	Current qualification of an EG Eluent Generator
XQ_EG_Dual_Current	Current qualification of an Dual EG configuration (ICS-5000 with both EGC configured on one Instrument)
XQ_CD_Noise_Drift	Noise and drift test for conductivity detectors
XQ_DC_Amp_Noise_Drift	Noise and drift test for DC amperometry detectors
XQ_Int_Amp_Noise_Drift	Noise and drift test for integrated amperometry detectors
XQ_PDA_Noise_Drift	Noise and drift test for photodiode array detectors
XQ_UV_Vis_Noise_Drift	Noise and drift test for absorbance detectors (excluding PDA detectors)
XQ_Wavelength_Accuracy	Wavelength accuracy test for the VWD and PDA absorbance detectors
XQ_Abs_Inj_Precision	Injector precision test for systems with an absorbance detector
XQ_Abs_Inj_Carryover	Injector carryover tests for systems with an absorbance detector
XQ_CD_Inj_Precision	Injector precision test for systems with an conductivity detector
XQ_CD_Inj_Carry_Over	Injector carryover test for systems with an conductivity detector
XQ_ED_Warm_Up	Warm-up sequence for ICS-5000 systems with ED detector
XQ_ED_Noise_Drift	Noise and drift test for ICS-5000 amperometry detectors (ED in Integrated Amperometry mode)
XQ_ED_Inj_Precision	Injector precision for ICS-5000 systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Carryover	Injector carryover for ICS-5000 systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Linearity	Detector Linearity for ICS-5000 systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Inj_Linearity	Injector linearity for ICS-5000 systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Stop	The sequence turns off the pump and cell for ICS-5000 systems with amperometry detector (ED in Integrated Amperometry mode).
XQ_Abs_Linearity	Detector linearity test for absorbance detectors
XQ_CD_Linearity	Detector linearity test for conductivity detectors
XQ_Abs_Inj_Linearity	Injector linearity test for systems with an absorbance detector

XQ_CD_Inj_Linearity	Injector linearity test for systems with an detector
XQ_Abs_Samp_Prep	Injector sample preparation precision test for systems with an absorbance detector
XQ_CD_Samp_Prep	Injector sample preparation precision test for systems with an conductivity detector
XQ_GP_Grad_Accuracy	Gradient accuracy and noise test for systems with a gradient pump
XQ_CD_Noise_Drift	Noise and drift test for conductivity detectors
XQ_DC_Amp_Noise_Drift	Noise and drift test for DC amperometry detectors
XQ_Int_Amp_Noise_Drift	Noise and drift test for integrated amperometry detectors
XQ_PDA_Noise_Drift	Noise and drift test for photodiode array detectors
XQ_UV_Vis_Noise_Drift	Noise and drift test for absorbance detectors (excluding PDA detectors)
XQ_Stop	The sequence slows down the flow rate to 0.25 mL/min to conserve eluent for overnight runs

5 Performing the Qualification

5.1 OQ/PQ Test Order

The OQ/PQ qualification tests are performed in the following order:

- 1 Flow Rate Accuracy and Precision test
- 2 Eluent Generator Current Qualification Test (only if an ICS-2000 or ICS-2100 is present).
- 3 Warm Up
- 4 Detector Noise and Drift Test
- 5 Injector Precision Test
- 6 Carryover Test
- 7 Detector Linearity Test
- 8 Injector Linearity Test (only if an AS-AP, AS or AS50 autosampler is present)
- 9 Sample Preparation Precision Test (only if an AS or AS50 is present with the Sample Prep Option and the 10 mL Tray Type installed)
- 10 Column Heater Temperature Verification (only for ICS-1100 or ICS-1000 with column heater option, ICS-1500, ICS-1600, ICS-2000 and ICS-2100 systems)
- 11 Sample Temperature Accuracy of Autosamplers (only for AS-AP autosamplers)

5.2 OQ/PQ Test Timetable

If the system to be tested includes an autosampler all the tests after the Flow Rate Accuracy and Precision Test are run automatically. In the case where manual injections are to be used the test time needed is shown here:

Table 10. OQ/PQ Test Timetable

Test	Procedure Type	Material Required	Equilibration Time	Approx. Run Time
Flow Rate Accuracy and Precision All pump models	Sequence	Eluent (water) Backpressure loop	15 min	30 min
Wavelength Accuracy AD25, ICS Series VWD	Sequence	Eluent (water)	Lamps – 2 h 10 min w/pump	6 min
Wavelength Accuracy PDA-100, ICS Series PDA	Sequence	Eluent (water)	Lamps – 2 h 10 min w/pump	6 min
AS-DV/AS40/Manual Inj Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity	Sequence	Eluent (water) Standards Backpressure loop	10 min	1 hr and 10 min
AS-AP/AS/AS50 Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity Injector Linearity	Sequence	Eluent (water) Standards Backpressure loop	10 min	2 hr and 10 min
AS-AP with Temp. Control Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity Injector Linearity Sample Temp. Accuracy	Sequence	Eluent (water) Standards Backpressure loop Type K temperature sensor	10 min	2 hr and 55 min
AS/AS50 with Sample Prep Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity Injector Linearity Sample Prep Precision	Sequence	Eluent (water) Standards Backpressure loop	10 min	2 hr and 40 min
Eluent Generator Current Qualification	Sequence	Test cell Multimeter	N/A	16 min
Column Heater Temperature Verification	Manual	Calibrate Thermometer	15 min minimum	0 min

5.3 Starting the Sequences



For AS-AP or AS Autosamplers with the Sequential Injection Option: Before starting any sequences, first verify that none of the instrument configured on the computer have “Acquired Exclusive Access” to the autosampler. If this is the case release the “Exclusive Access” command before proceeding. Once the qualification sequences begin, the system being qualified will gain and retain exclusive access to the autosampler until the “Stop” sequence is run and “Exclusive Access” is released. “Exclusive Access” can be released by going to the command function (F8), choosing the module from the list, and executing the “ReleaseExclusiveAccess” command.

5.3.1 Flow Rate Accuracy and Precision Qualification

The Flow Rate Accuracy and Precision test is designed to qualify the flow rate accuracy and precision of all ICS series pumps.

Thermo Fisher Scientific cannot guarantee tests performed outside the scope of the published OQ/PQ protocol.

1. Ensure the pump is disconnected from the Chromeleon software. The system should be in hold mode and a message box displaying “Plumb pump for Pump Flow rate Accuracy and Precision Test, press OK to continue” should be visible.

Plumb pump for Pump Flow-rate Accuracy and Precision Test, press OK to continue.

OK

2. Stop the pump from the front control panel of the module if it is not already stopped. ICS-1100 users: if the pump is not already stopped, it may be necessary to click OK on the message box, manually stop the queue and turn off the pump from the control panel. Remember to reset the status to single before resuming the test.
3. Locate the backpressure coil (P/N 044211 or 049715)
4. Connect one end of the backpressure coil directly to the outlet of the pump, after gradient mixers if applicable and the other to port 2 (P) of the injector valve.
5. Prepare two lengths of black tubing, 0.010 (0.25 mm) ID, of equal length, approximately 12” (40 cm). Connect one of these lines to port 1 (L) of the injector valve. The tubing connected to port 1 (L) of the injector valve will be the eluent line that goes to the “sample beaker”.
6. Connect the other line to port 3 (C) of the injector valve. The tubing connected to port 3 (C) of the injector valve will be the waste line.
7. Run the waste line to a waste beaker and the eluent line to a sample beaker. The plumbing to the valve should look like the Figure 6 below. Turn the pump on (except ICS-1100).

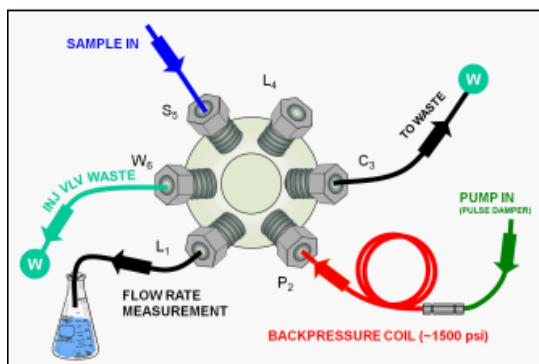


Figure 17. Plumbing schematic for Pump Flow Rate Test

8. Click OK in the Chromeleon software. The system should reconnect to the module and/or resume the program in run mode. ICS-1100 users: the pump should turn on at this point.
9. The program will equilibrate the pump for 15 minutes before starting the test. During this time eluent flow should pass through the waste line. Ensure that sufficient backpressure is observed during this equilibration time.
10. At time 0, the injector will switch to the eluent line.
11. At time 2.00, the injector will switch back to the waste line and the Chromeleon software will ask you to "Please weigh the eluent container. Enter results under segment 0. Press OK to continue." The instrument will be in hold mode and will wait until OK is pressed before continuing. Weigh the eluent container, but **DO NOT ZERO THE BALANCE AFTER WEIGHING**. Record the weight as segment 0. Click OK to continue.

Please weigh the eluent container. Enter results under segment 0. Press OK to continue. OK

12. The Chromeleon software will switch the injector back to the eluent line for exactly 5 minutes. After this time the injector will switch to the waste line and a message "Please weigh the eluent container. Enter results under segment 1. Press OK to continue" will be displayed. The instrument will be in hold mode and will wait until OK is pressed before continuing. Weigh the eluent container, but **DO NOT ZERO THE BALANCE AFTER WEIGHING**. Record the weight as segment 1. Click OK to continue.
13. Repeat 12 for segments 2 through 5.
14. After the sequence is finished open the Water_FR sample from the Console. Select the Flow Rate sheet at the bottom of the page and enter the Segment values and room temperature into the sheet. Save the worksheet by clicking on the Chromeleon Icon on the top left of the User Interface and select save.

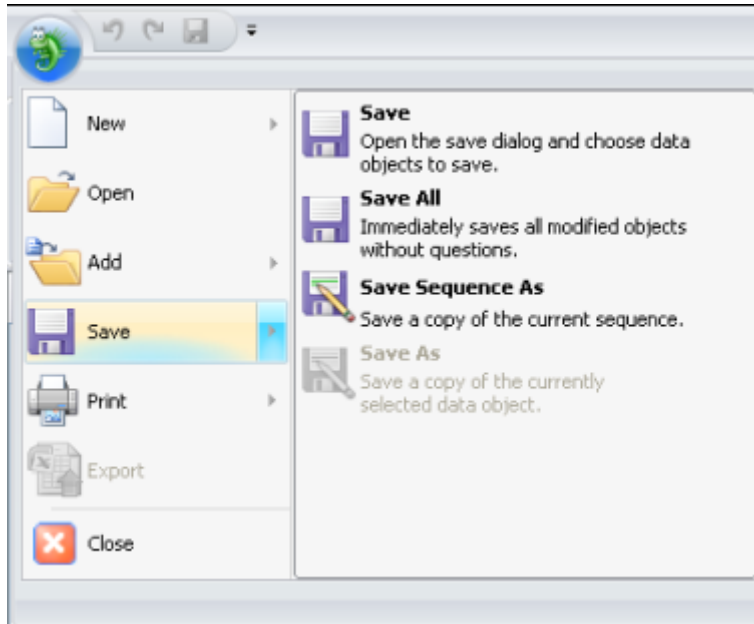


Figure 18. Selecting Save

15. Print the Flow Rate Accuracy and Precision report by clicking on the Chromeleon Icon on the top left of the User Interface and select print. Choose a printer and press OK to begin printing.

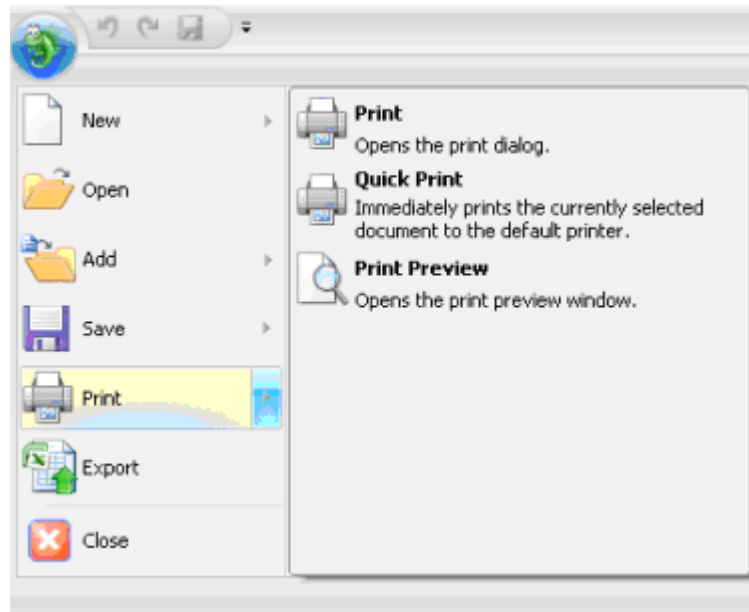


Figure 19. Selecting Print

16. If any part of the test does not pass, make the appropriate repair or calibrations and rerun the test. Refer to Section 5 for troubleshooting assistance.
17. Plumb the system for the remainder of the qualification as in Figure 7 below.

18. Ensure the pump is disconnected from the Chromeleon software. The system should be in hold mode and a message box displaying "Plumb the system for the remainder of the qualification tests, press OK to continue" should be visible.
19. Connect a length of yellow tubing sufficient to produce about 1500 psi backpressure to the inlet of the conductivity or absorbance cell, thus bypassing all columns and suppressors.

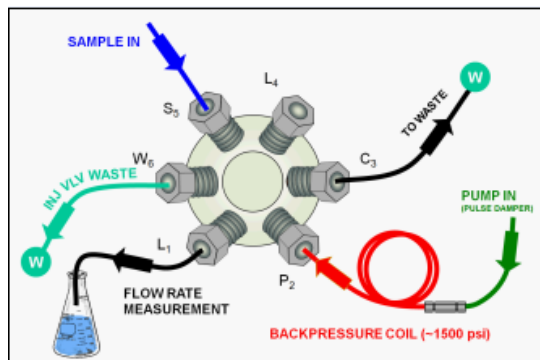


Figure 20. Plumbing schematic for OQ/PQ Performance Tests after Pump Flow Rate Test

5.3.2 Eluent Generator Current Qualification

The Eluent Generator Current Qualification test (j) will be used for ICS-2000 or ICS-2100 configured with either a KOH or MSA cartridge.

1. **Resume** the Queue by clicking OK. This sequence is an Eluent Generator step gradient. Each step is held until the user clicks OK to continue. There may be a delay of up to 0.4 minutes between when the Eluent Generator program steps to the next concentration and when the ammeter or multimeter registers the new milliamp reading. Record the milliamp reading for each step on a note pad. These values will be entered into the **EG_CURRENT**, **EG_DUAL_CURRENT** or **EluGen Current Qualification** sheet after the run is complete. The run time is 11.0 minutes plus pause time.

Note: The template of this report is stored in the data vault of the respective sequence. Please refer to Chromeleon 7 Quick Start Guide on how to access report templates.

Template name: OQ_PQ_Integrated_Validation

2. When the run is complete, open the report file by double clicking on the Water_EG sample of the XQ_EG_Current sequence. Locate the row of sheet names at the bottom of the report and click on the **EG_CURRENT**, **EG_DUAL_CURRENT** or **EluGen Current Qualification** sheet.
3. Enter the milliamp readings that were recorded in step 1 for each concentration value in the **EG_CURRENT**, **EG_DUAL_CURRENT** or **EluGen Current Qualification** sheet. The deviation and pass/fail results will be calculated automatically.
4. Print the EG Current Qualification report by clicking on the Chromeleon Icon on the top left of the User Interface and select print. Choose a printer and press OK to begin printing.

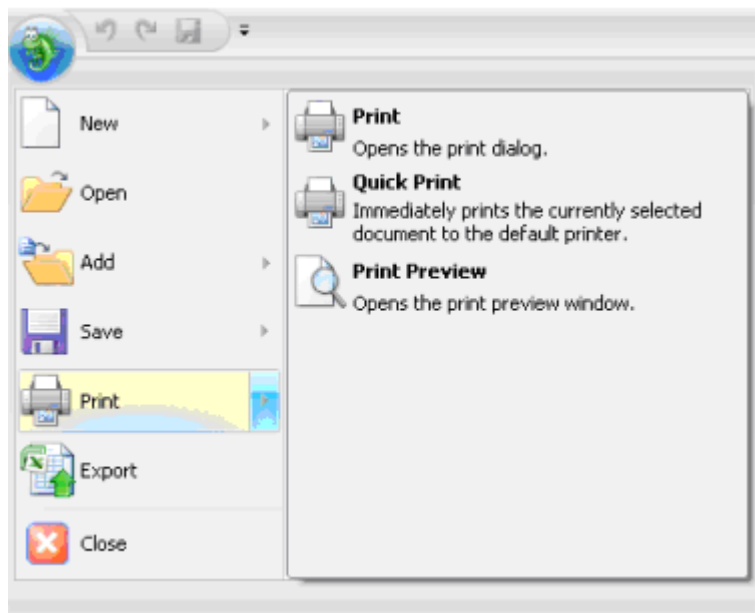


Figure 21. Selecting Print

5. If any part of the test does not pass, make the appropriate repair/adjustments and rerun the test. Refer to Section 5 for troubleshooting assistance.
6. To qualify another cartridge, select the new cartridge type in the control panel, add the XQ_EG_Current, XQ_EG_Dual_Current or XQ_IC_EG_Current sequence to the queue list, set the status of the Water_EG and Stop samples to single and repeat steps 1 - 4.

5.3.3 Performing the Main Qualification

The main part of the OQ/PQ consists of the tests d – k, as shown in Section 5.1. This part of the qualification requires that an absorbance or conductivity detector be configured on the system being qualified.

If the qualification will be performed using a manual injection valve, use full loop injections and overfill the loop a minimum volume equal to 4 times the sample loop volume. Inject standards according to the sequence table. With an AS40 autosampler there will be a 2.4-minute delay before the sample is injected.

If manual injection is used it is essential to manually edit the programs to replace the 2.4-minute delay with a message command to hold the system until the sample is ready. Replace the “– 2.400 Pump_Relay_1.Closed Duration=10.00” line with “–0.100 Message "Load the sample into the injection loop, press OK to continue"”

1. The IC must be set up and running as described in Section 4.5.
2. The sequences must be installed as described in Section 4.6.
3. The sequences must be set up as described in Section 4.7.
4. If the queue is not started already go to Queue and Edit. Start the Queue. Run times are as follows:

Systems with an AS-DV, AS40 or manual injection have a run time of approximately 1 h and 10 min.

Systems with an AS-AP, AS or AS50 but no sample prep have a run time of approximately 2 h and 10 min.

Systems with an AS or AS50 and sample prep have a run time of approximately 2 h and 40 min.

At the end of the main qualification the system will automatically stop.

For systems using an Eluent Generator, the program will pause before the Eluent Generator Current Qualification Test, proceed to section 2. The program will pause before the Flow Rate Accuracy and Precision Test, proceed to section 3.3.4 before continuing to step 5.

5. When the queue is finished, go to the Chromeleon console right click on each completed sequence present in the qualification, select the **Report** and **Print** the report. The Eluent Generator Current Qualification, Flow Rate Accuracy and Precision Qualification, Column Heater Accuracy qualification have already been printed.
6. Review the report for Pass/Fail results. If any test does not pass, make the appropriate instrument repair and/or adjustments and rerun all tests from the original point of failure on. Refer to Section 5 for troubleshooting assistance.

5.3.4 Column Heater Temperature Qualification

The column heater temperature qualification can be used to qualify the temperature accuracy of the column heater fitted to an ICS-2100, ICS-2000, ICS-1600, ICS-1500, ICS-1100 or ICS-1000. After completion of all other tests record the reading on the calibrated thermometer.

1. Open the Water_WU sample in the Warm Up sequence from the console. Select the Column Heater Temp Qualification tab at the bottom of the page. Enter the measured temperature in the Measured Plate Temp. cell.

Note: The template of this report is stored in the data vault of the respective sequence. Please refer to Chromeleon 7 Quick Start Guide on how to access report templates.

Template name: OQ_PQ_Integrated_Validation

2. The measured liquid temperature and its deviation from theory will be calculated automatically. If the test does not pass, make the appropriate repair/adjustments and rerun the test. Refer to Section 5 for troubleshooting assistance.
3. Print the Column Heater Temperature Verification report by selecting "Print" from the File menu. Choose a printer and press OK to begin printing.

5.3.5 Turning off watermarks in the reports

Some users prefer not having watermarks in the back of the report. The procedure below describes how to turn off watermarks in the reports. The procedure can only be performed if the user has sufficient privileges granted by the administration.

1. Go to the Instrument Category
2. In the Chromeleon Console, Go to the Tools menu.
3. Select Administrations Console

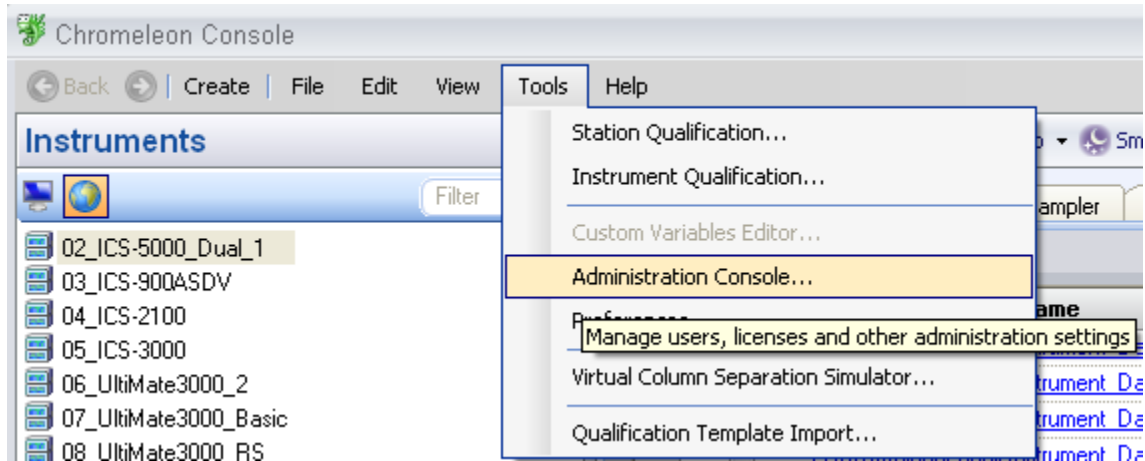


Figure 22. **Selecting Administration Console**

4. Select Global Policies within the Administration Console.

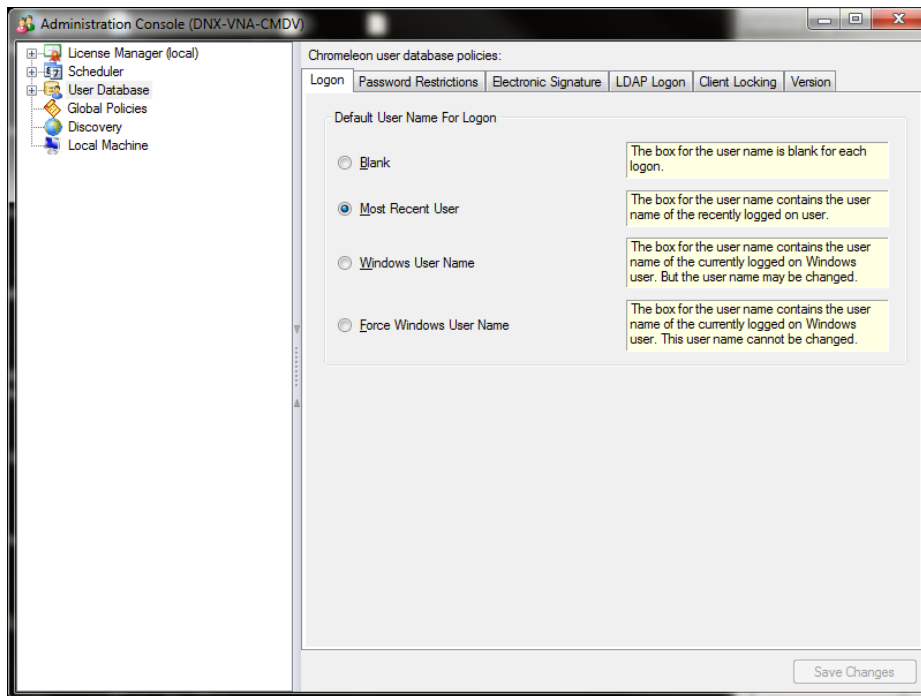


Figure 23. **Selecting Global Policies**

5. Select "Reporting" tab, then uncheck "Print with watermark"

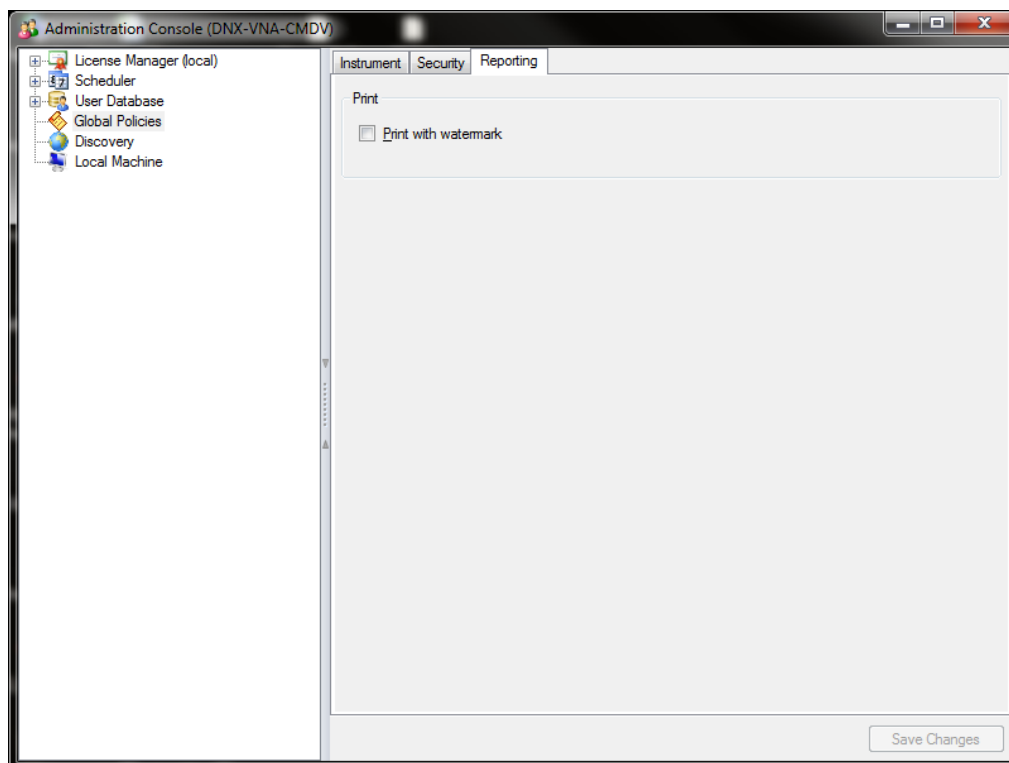


Figure 24. Selecting reporting

5.3.6 Sample Temperature Accuracy (AS-AP autosampler only)

This section describes how the sample temperature accuracy is determined for the following Thermo Fisher Scientific autosamplers: AS-AP. For this test, only the autosampler is required. The other modules of the IC system are not required.

Test Procedure

The following table lists the materials required for performing the test.

Part No.	Description	Quantity
6820.0010	Type K temperature sensor for P600 thermometers	1
5705.0050A	Column Thermostat PQ Kit	1

In addition, a standard polypropylene vial (10 mL) is required. Fill the vial with water (do not cap).

Connecting and Configuring the System

System connections

- Connect the Dostmann P600 Thermometer and the Type K temperature sensor. Make the settings (that is, adapt the sensor type and calibration values) as described in the instructions for the P600 Thermometer.



Figure 25. P600 Thermometer and the Type K sensor



Figure 26. P600 Thermometer and the Type K sensor

- Fill an open standard polypropylene vial (10 mL) with water and place it at sample position BC1.
- Insert the thermocouple into the vial at a right angle until the tip touches the vial bottom.



Figure 27. Temperature sensor inserted into the vial

- Rotate the carousel until the carousel cover closes completely.



Figure 28. Temperature sensor inserted into the vial

Configuration

Configuration is not required.

- **Preparing Chromeleon**
To qualify the sample temperature accuracy, select the following sequence:
SAMPLER_TEMP_ACC

Performing the Test

Set the sample temperature to 15 °C. When the nominal temperature is reached, the external thermometer is used to record the sample temperature over a period of 30 minutes.

Tip: Do not perform any autosampler commands during the test. Moving the needle arm or carousel may damage the thermometer or autosampler. The user can use a calibrated thermometer such as the Dostmann P600 or equivalent.

Duration

The test takes approximately 45 minutes.

5.3.7 AS-AP pH and Conductivity Qualification

This section describes how the sample pH and conductivity accuracy is determined for the following Thermo Fisher Scientific autosampler: AS-AP. For this test, the autosampler and other modules of the IC system are required. Please refer to the operator manual for pH and conductivity detector set up. The pH qualification should be performed after the standard qualification. The AS-AP pH and conductivity qualification will be set up in push mode only.

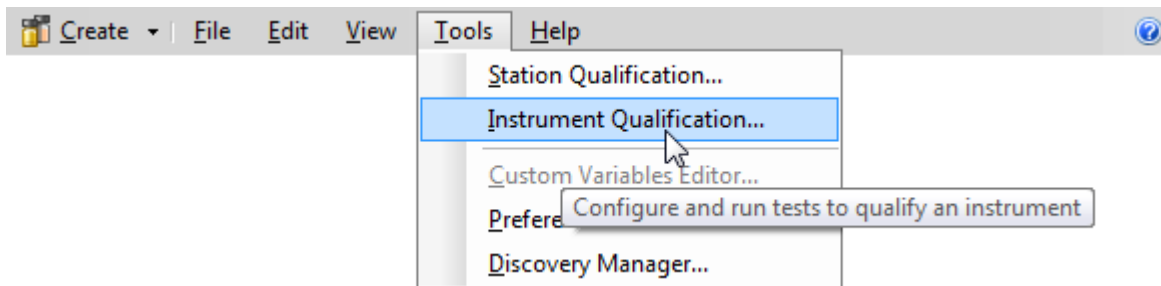


Figure 29. Instrument Pane for Chromeleon 7.1

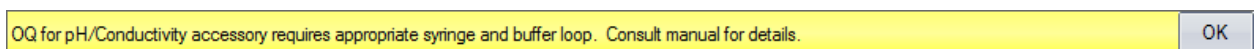


Figure 30. Starting the Instrument Qualification Wizard for Chromeleon 7.1

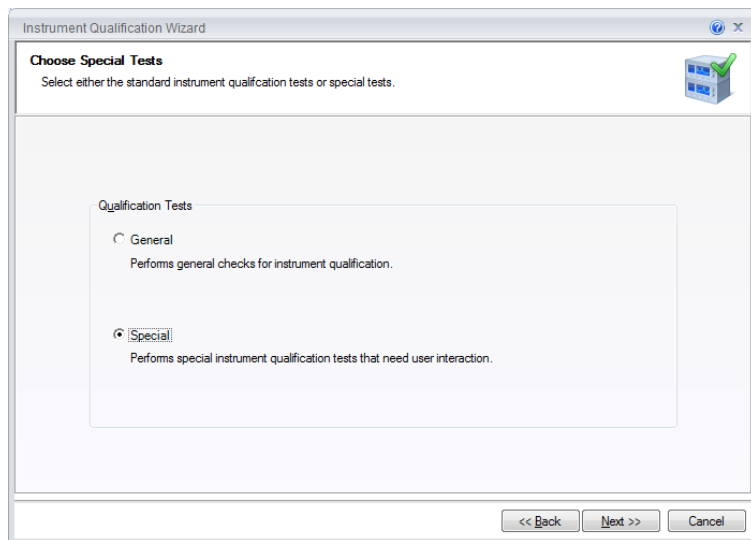


Figure 31. Select special for the Qualification Tests

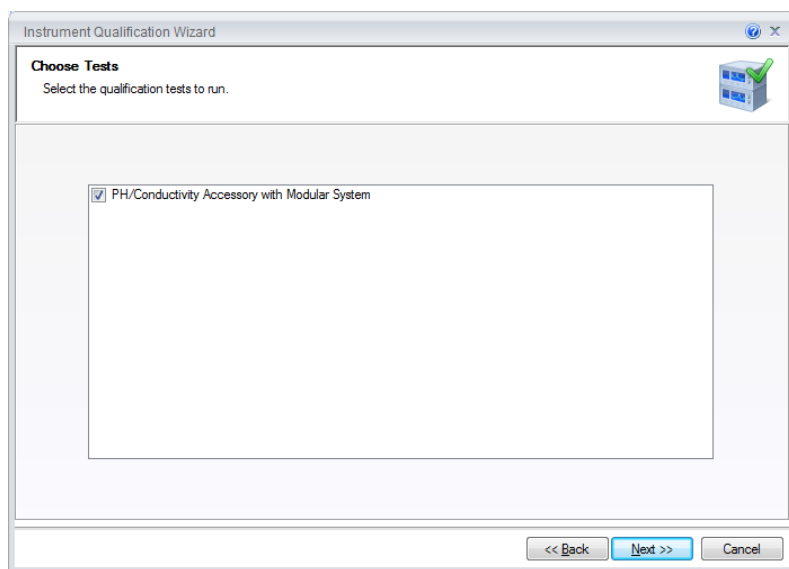


Figure 32. The pH/ Conductivity Tests will be selected if the instrument is configured correctly.

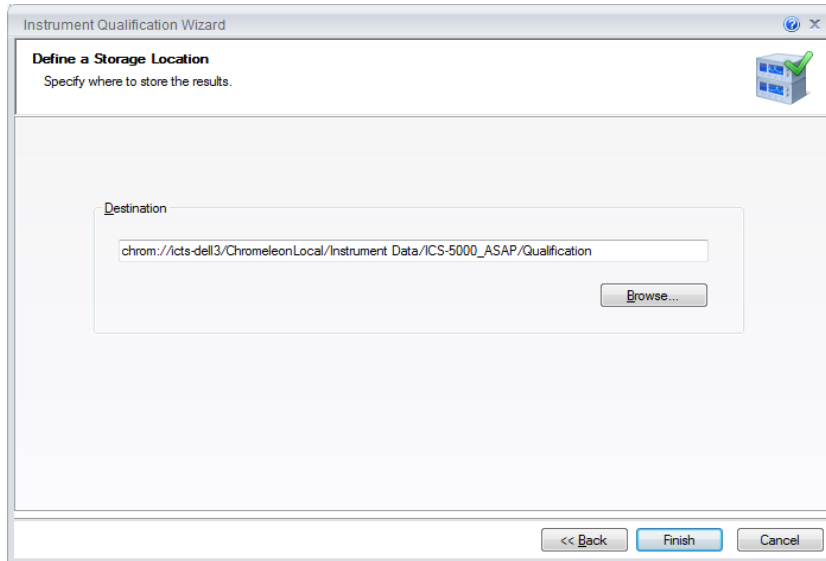


Figure 33. Select finish

#	Chromatogram	Name	Type	Level	Position	Volume [µL]	Instrument Method	Processing Method
1	None	WARMUP	Blank	01	RA1	100.0000	WARMUP	Validation
2	None	pH_OFFSET_CALIBR...	Blank	01	RA1	100.0000	pH7_CALIBRATION	Validation
3	None	pH_SLOPE_CALIBR...	Blank	01	RA2	100.0000	pH4_CALIBRATION	Validation
4	None	pH_SLOPE_CALIBR...	Blank	01	RA3	100.0000	pH10_CALIBRATION	Validation
5	None	pH_VERIFICATION(p...	Blank	01	RA1	100.0000	pH_MEASUREMENT	Validation
6	None	pH_VERIFICATION(p...	Blank	01	RA2	100.0000	pH_MEASUREMENT	Validation
7	None	pH_VERIFICATION(p...	Blank	01	RA3	100.0000	pH_MEASUREMENT	Validation
8	None	CD_CALIBRATION	Blank	01	RA4	100.0000	CD_CALIBRATION	Validation
9	None	CD_MEASUREMENT...	Blank	01	RA5	100.0000	CD_MEASUREMENT	Validation

Figure 34. Sequences

Performing the Check

1. Fill the vials according to the table below.

Table 11. OQ/PQ Test Timetable

Qualification	Standard	Sample Order on AS-P
pH Measurement	pH OFFSET Calibration	RA1
pH Measurement	pH Slope Calibration	RA2
pH Measurement	pH Slope Calibration	RA3
CD Calibration	Blank	RA4
CD Measurement	Conductivity Standard	RA5

2. Start the sequence.
3. A pop out screen will remind the user to configure the AS-AP with the 5mL syringe and the 8500 µL buffer loop before the qualification begins.

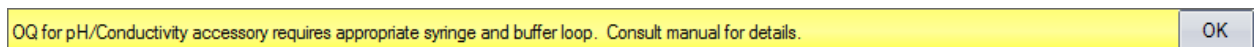


Figure 35. Reminder Screen

Duration

The test takes approximately 1 hour.

5.3.8 Qualification Comments

The comments section of the report is provided as a place to record any comments, test failures, or configuration changes that occur during the qualification.

To use the comment section, open the Water_WU sample in the Warm Up sequence from the console. Select the Comment tab at the bottom of the page. Enter any comments. This page can also be printed and comments entered manually.

To print the Comment report, select "Print" from the File menu. Choose a printer and press OK to begin printing.

Note: The template of this report is stored in the data vault of the respective sequence. Please refer to Chromeleon 7 Quick Start Guide on how to access report templates.

Template name: OQ_PQ_Integrated_Validation

5.4 What to do if a Test Fails

If at any point the instrument results do not pass within Thermo Fisher Scientific specified limits, the instrument must be serviced. See Chapter 6 for troubleshooting assistance. When the repair is complete, rerun all tests from the original point of failure on in the sequence. All actions for failures and repairs should be recorded on the "comments" report. For example, if the instrument passes tests a, b, and c and fails test d, make the appropriate repairs, record the action, and then repeat tests d – o only.

When rerunning the tests, restart the Instruments OQ or Instrument PQ procedure from the Qualification menu and uncheck all passed tests prior to the test that failed. All tests after the test that failed should be left checked even if they have previously passed. The Warm Up sequence is always required and must be rerun.

5.5 OQ/PQ Completion

5.5.1 Customer Review

1. The printed results should be reviewed by the instrument owner/user.
2. When the qualification is accepted, both the Customer and Qualification Executor need to sign the printed reports.
3. Fill out the Qualification Stickers and Certificates of Functional Performance for the qualified modules.

- Place the reports and certificates into the binder provided in the OQ/PQ kit and leave it with the customer.

5.5.2 Accuracy of reports

In accordance with Good Laboratory Practice, calculations used to determine the results in the printed reports use more significant figures than those shown and are rounded down for the final report. When reproducing the calculations by hand it may be necessary to obtain numbers from the reports after increasing the number of displayed significant figures.

The number of displayed significant figures can be adjusted through the report publisher. Open the sequence containing the samples for the test being examined and double click on a sample. The sample data should be shown. Highlight the numbers being used for the hand calculations, click on the Format menu and select number. The number of digits displayed can be increased by changing the number type; for example, entering 0.00000 into the Type field will increase the number of displayed decimal places to 5. **Always use more significant figures for hand calculations than are shown in the printed report.**

5.5.3 Return the System to the Original Configuration

- If the Signal factor was modified in the system preparation, or a module was renamed it must be set back to its original value. Open the Chromeleon Instrument Configuration application. Double click on the detector/s qualified to view the **properties** page. Select the **Signals** page to change the factor back to the original value and the **General** page to change the name back to the original value. Exit the properties page and save the configuration.
- Disconnect the Backpressure Coil and restore the system plumbing to its original configuration with the appropriate columns, suppressors, and Eluent Generator in line.

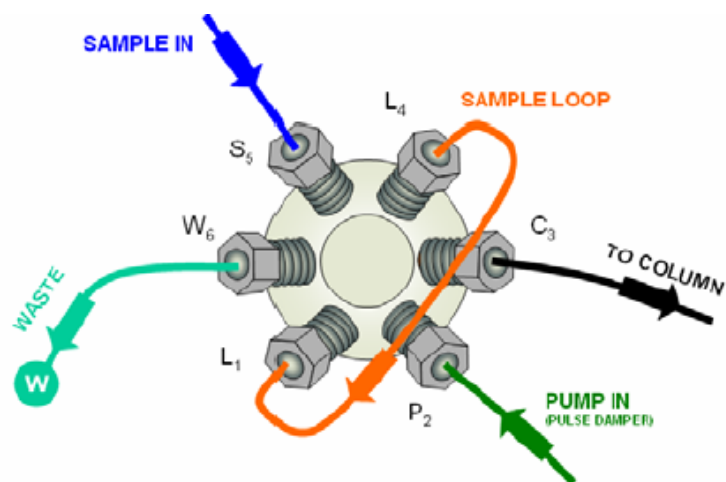


Figure 36. Final valve plumbing diagram

- Remove the sample loop and reinstall the original sample loop back onto the injection valve, as in Figure 8 above. For systems using an AS or AS50 Autosampler, go to the **Main Menu** on the front panel of the autosampler. Access the **Module Set Up** screen and then go to **Plumbing Configuration**. Change the sample loop size back to its original value. For systems using the AS-AP Autosampler, open Instrument Configuration and go to the Options tab for the AS-AP Autosampler and change the sample loop size back to the original value

- 4 Reconnect the original eluents to the pump and prime each line individually to purge the eluents used in the OQ/PQ tests.
- 5 Flush the system with an eluent recommended by the customer and confirm that there are no leaks.

6 Troubleshooting

6.1 General Hints

If the system pressure is above 2000 psi after connecting the PEEK Backpressure coil with a flow rate of 1.00 mL/min, the coil has a blockage. Re-cut the ends of the coil or replace it.

Dips before or after the main nitrate or caffeine peak indicate extra dead volume in the system. This is caused by small voids between the end of a liquid line and the bottom of the port. Individually disconnect each line from the injection valve to the detector cell. Push the ferrule and fitting up and away from the end of the tubing. Make sure the tubing is cut straight; if it is not, re-cut the end. When reinstalling all liquid connections, first insert the tubing all the way into the port until it touches the bottom. Hold the tubing in this position while inserting and tightening ferrule and fitting down into the port.

6.2 Failure of Individual Tests

6.2.1 Conductivity Detector Tests

Table 12. Troubleshooting Conductivity Detector Test Failure

Test	Reason	Action
Baseline Noise	Poor water quality	Replace with an alternate water source
	Air bubbles in flow cell	Flush out flow cell
	Cell temperature regulation failure	Replace cell
Drift	System not equilibrated	Allow longer flush/equilibration time
	Lab temperature not stable	Regulate temperature (block vents, close windows, etc.)
	Water not degassed	Degas water or turn degas unit onto "Always On" setting.
Detector Linearity	Concentration of standards is incorrect	Obtain new fresh standards
	Liquid leak	Find and repair source of leak
	Injection volume not consistent (manual injections only)	Overfill sample loop at least 4x when loading
	The peak height of the sample with the highest concentration is not in the linearity range specified for the detector, i.e., usually > 1500 mAU (for Thermo Fisher Scientific and PDA detectors).	Reduce the injection volume for all samples used for the detector linearity check so that the peak height of the sample with the highest concentration is in the linearity range of the detector, i.e., usually < 1500 mAU.

6.2.2 Pump Tests

Table 13. Troubleshooting Pump Test Failure

Test	Reason	Action
Flow Rate Accuracy and Precision	Flow rate out of calibration	Re-calibrate the flow rate of the pump
	Leaking piston seals	Replace piston seals
	Faulty check valves	Replace check valves

6.2.3 Autosampler Tests

Table 14. Troubleshooting Autosampler Test Failure

Test	Reason	Action
Injector Precision	Air injected from vial or from sample syringe when doing manual injections	Too little sample volume in vial – refill vial or flush bubbles out of injection syringe
	Air bubbles in autosampler sample syringe	Flush out syringe
	Liquid leak in autosampler or injection valve	Find and repair source of leak
	Poor water quality in flush reservoir	Change water and flush sampler fluidics
Injection Volume Linearity	Detector failure	See Detector linearity troubleshooting section
	Defective sample syringe	Replace sample syringe
Carryover	Contaminated injection valve	Replace injection valve rotor and stator face
	Contamination in autosampler	Flush/replace autosampler liquid lines
	Contamination in sample vial or in syringe used for manual injections	Use new sample vial for deionized water blank, rinse vial thoroughly before using, flush syringe or use a new one
Sample Prep Precision	Air injected from vial	Too little sample volume in vial, refill vial
	Air bubbles in AS sample syringe or prep syringe	Flush out syringe
	Defective prep syringe	Replace prep syringe
	Liquid leak in autosampler or injection valve	Find and repair source of leak
	Poor water quality in Sample Prep reservoir	Change water and flush sampler fluidics
Tray Temperature Accuracy	Thermometer not calibrated	Obtain a new calibrated thermometer

6.2.4 Eluent Generator Tests (ICS-2000/2100)

Table 15. Troubleshooting Eluent Generator Test Failure

Test	Reason	Action
Current	Qualification Test box not connected properly	Reconnect test cell cable to the Eluent Generator, confirm ammeter or multimeter leads are making contact with test cell wires
	Ammeter or multimeter on wrong setting	Ammeter or multimeter should be set to read current (mA). Range should be set to view 0.00 to 300.00 mA
	Qualification test box defective	Replace qualification test box

6.2.5 Column Heater Tests

Table 16. Troubleshooting Column Heater Test Failure

Test	Reason	Action
Temperature Qualification	Column Compartment not calibrated	Recalibrate the Column Heater
	Fluke Multimeter not calibrated	Obtain a new calibrated Fluke Multimeter
	Thermometer not fitted in column compartment correctly	Fit thermometer in column compartment correctly

7 Example Report

The example report is for an ICS-2100 configured with an AS-AP Autosampler.

**User's Guide:
Chromleon[®] Operational Qualification and
Performance Qualification Software Templates
for ICS-5000/ICS-5000⁺ Analytical Instruments
Revision 7.12**

2013 Thermo Fisher Scientific

Document 065448 Part E
March 2013, Thermo Fisher Scientific
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PRINTING HISTORY

Revision 7.10, September 2011

Revision 7.11, December 2011

Revision 7.12, March 2013

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1 Introduction

Operational and Performance Qualification are becoming increasingly important to analytical laboratories. Documented evidence must be provided to demonstrate the integrity of data collected and qualify the results obtained on laboratory instrumentation.

The following institutes set and control industry instrumentation standards:

NIST The National Institute of Standards and Technology is an administrative body of the US Department of Commerce. Together with industry, the NIST develops new technologies and sets technological standards countrywide. Thermo Fisher Scientific includes NIST traceable standards as part of the qualification kit.

ASTM The American Society for Testing and Materials sets technical standards and rules for the industry worldwide, specifically; E 1657 – 98: Standard Practice for Testing Variable-Wavelength Photometric Detectors in Liquid Chromatography. E1511 – 93: Standard Practice for Testing Conductivity Detectors used in Liquid Chromatography. E19.09.07: Proposed Standard Practice for Evaluation of Gradient HPLC pumps – Draft.

USP The United States Pharmacopeia (USP) is an official public standards–setting authority for all prescription and over–the–counter medicines and other health care products manufactured or sold in the United States. USP establishes documentary and reference standards to ensure quality analytical instrument qualification process.

These standards are written in broad terms to make them as widely applicable as possible. All stipulate general requirements specifying instruments must be fit for purpose, technically checked in defined periods, calibrated and make sure that test results are documented according to international quality standards. The OQ/PQ procedure used to qualify Thermo Fisher Scientific Ion Chromatography Systems meets the requirements established by NIST, ASTM and USP.

This OQ/PQ procedure provides qualification testing for **ICS-5000/ICS-5000⁺** analytical systems composed of any modules listed in section 1.6.

OQ/PQ results can be influenced by unusual laboratory conditions. For example, direct sunlight and close proximity to air conditioning ducts will cause thermal irregularities that will affect baseline drift on conductivity detectors. In these cases, it may happen that the manufacturer's specifications are not met. Ensure that the instrument is operated in suitable environmental conditions or for PQ, the specifications may be adjusted accordingly.

All OQ/PQ activities must be formally recorded, all instruments must be clearly identified, and test reports and supporting documentation must include the following information:

- Module name, type and serial number
- Software and firmware version
- Test data
- Service interval
- Signature of the Thermo Fisher Scientific Service Engineer/Qualification Executor who performed the OQ/PQ
- Signature of the customer who reviewed and accepted the OQ/PQ results

1.1 How to use this manual

It is essential that the operator read and understand this manual completely before implementing any OQ or PQ process.

1. Use the table of contents to determine which tests are required for the system to be qualified and refer to the recommended sections. This will help keep track of what steps to follow during system setup and preparation. It will also aid in selecting the appropriate sequences to run the tests.
2. For the ICS-5000/ICS-5000+ system, electrochemistry cells in Integrated Amperometry mode are supported for qualification tests. For the Integrated Amperometry detection qualification a pH/Ag/AgCl reference electrode is required for all tests. The ICS-5000/ICS-5000+ electrochemistry detector DC amperometry and cyclic voltammetry modes are not supported for OQ/PQ testing. When qualifying Integrated Amperometry detection a disposable amino acid gold working electrode is used; other electrodes are not supported.
3. Use the **Table of Contents** and the **List of Tables** as reference guides to locate information quickly.

The electronic version of the OQ/PQ operator's manual contains numerous hypertext links that can take you to other locations within the file. These links include:

- Table of contents entries
- Cross-references (underlined in blue) to sections, figures, tables, etc.

Chapter 1 Introduction

Introduces the conventions used in performing OQ/PQ with the ICS-5000/ICS-5000⁺ analytical systems; including the basic requirements, industry standards and manual summary.

Chapter 2 Overview of the Tests

Provides an overview of the tests available to be performed.

Chapter 3 Explanation of Test Procedures

Describes the theory behind each test, the OQ/PQ operating procedures and evaluation of the results.

Chapter 4 Qualification Preparation

Step by step preparation instructions for the OQ/PQ software, hardware installation and the details of the materials in ship kits.

Chapter 5 Performing the Qualification

Detailed operating instructions on how to perform the OQ/PQ including test order, time table, sequence initialization and troubleshooting tips.

Chapter 6 Troubleshooting

Lists problems and presents step-by-step procedures for how to isolate and eliminate the cause of each problem.

Chapter 7 Example Reports

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1.4 Operational Qualification (OQ)

According to “The Development and application of guidance on equipment qualification of analytical instruments” by P. Bedson and M. Sargent [Accred. Qual. Assur. (1996) 1: 265 – 274] the following definition applies:

“The purpose of OQ is to prove and document that an analytical system functions according to its operating specification while the specific environmental conditions are taken into account. In his specification, the supplier has therefore to define exactly the conditions that must be observed.”

OQ should be performed after initial system installation or when an additional new module is added to an existing system. OQ should be done according to the instructions outlined in this manual and all tests should be performed by a qualified Thermo Fisher Scientific Service Engineer. Any hand-written notes made during the performance of the tests must become part of the test results, in accordance with Good Laboratory Practice (GLP) and current Good Manufacturing Practice (cGMP).

1.5 Performance Qualification (PQ)

According to “The Development and application of guidance on equipment qualification of analytical instruments” by P. Bedson and M. Sargent [Accred. Qual. Assur. (1996) 1: 265 – 274] the following definition applies:

“The purpose of PQ is to prove and document that an analytical system functions according to a specification that is suitable for the system’s routine operation. A system is subject to wear when being operated. Thus, it may happen that the supplier’s specification is no longer met. This means: The test procedures used for OQ are also used for PQ, but the tolerances of PQ are less restrictive than those for OQ. Using the same test procedures simplifies the handling.”

PQ should be performed at regular intervals after the initial installation and OQ. Thermo Fisher Scientific recommends performing PQ every six months. PQ should be done according to the instructions outlined in this manual and all tests should be performed by a qualified Thermo Fisher Scientific Service Engineer. Any hand-written notes made during the performance of the tests must become part of the test results, in accordance with Good Laboratory Practice (GLP) and current Good Manufacturing Practice (cGMP).

1.6 Basic Requirements for Successful OQ and PQ

It is recommended that the most current revision of the OQ/PQ software templates is used in order for a qualification to be considered valid. It is the responsibility of the qualification engineer to determine the most up to date revision of the templates available and ensure that the revision is used before proceeding with this qualification or if the customers SOP require an earlier OQ/PQ version to be used.

It is recommended that the qualification be performed using the most current Chromeleon[®] service pack version (Contact Thermo Fisher Scientific Technical Support for current version).

It is recommended that the qualification be performed using the most current firmware version for each system or module (Contact Thermo Fisher Scientific Technical Support for current version).

Operational Qualification and Performance Qualification are system-specific test procedures. The procedures described below are valid for IC systems comprising of the following equipment (only one type of detector and one pump can be qualified concurrently):

- Pump: ICS-5000/ICS-5000⁺ SP or ICS-5000/ICS-5000⁺ DP (Analytical version)
- Manual injection or any of the following autosamplers: AS, AS-DV or AS-AP
- AS-AP autosampler 10 mL tray (P/N 074938) with ICS-5000/ICS-5000⁺ CD (pull or push mode) and 1.5 mL tray (P/N 074936) with ICS-5000/ICS-5000⁺ CD and ED (push and push sequential mode).
- Absorbance detector: ICS Series VWD or ICS Series PDA.
- Conductivity detector: ICS-5000/ICS-5000⁺ CD
- Electrochemical detector: ICS-5000/ICS-5000⁺ ED
- Eluent Generator: ICS-5000/ICS-5000⁺ EG

Note: Ensure that the seal wash option is turned off during the qualification. After the OQ/PQ tests are finalized, be sure to re-enable the seal wash option.

In addition, Chromeleon[®] version 7.1 or higher is required and the following requirements must also be met:

Table 1. ICS-5000/ICS-5000⁺ and Chromeleon Compatibility

Module	Minimum Chromeleon Software Version
AS-AP	7.1 SR1c
AS	7.1 SR1
AS-DV	7.1 SR1
ICS-5000/ICS-5000 ⁺ SP or DP	7.1 SR1
ICS-5000/ICS-5000 ⁺ EG	7.1 SR1
ICS-5000/ICS-5000 ⁺ DC	7.1 SR1

ICS-5000/ICS-5000 ⁺ CD	7.1 SR1
ICS-5000/ICS-5000 ⁺ ED	7.1 SR1
FLUKE189/289	7.1 SR1
ICS Series VWD	7.1 SR1
ICS Series PDA	7.1 SR1
ICS-5000/ICS-5000 ⁺ TC	7.1 SR1

It is possible to qualify modules in systems that are not listed in this section; however some modification of the included programs may be required. Such modifications are not within the scope of this document. For the ICS-5000/ICS-5000⁺, it is not possible to qualify multiple pumps on a single system. If multiple pumps are configured on a single Chromeleon Timebase, the second pump will be ignored by the programs. To qualify the AS-AP or AS autosampler with the injector linearity test (partial loop injections) a 250 µL injection syringe is required.

2 Overview of the tests

The following table provides an overview on the parameters to be tested and the limits recommended for OQ and PQ.

- Template sequences (with programs specific to the Instrument Configuration are generated based on the master sequence of the OQ/PQ templates and the system configuration in the IC Instrument).
- The sequences are copied from each test that is to be performed on the subject system.
- The Chromeleon software will automatically create a default unique name and directory to contain all template sequences.
- Test sequences must be run in the order they are listed. Do not alter the test sequence order.

Table 2. OQ/PQ Specification Limits: ICS-5000/ICS-5000⁺ CD Baseline Noise/Drift and Detector Linearity

Instrument	Parameter	Description	Limits	
			OQ	PQ
ICS-5000/ICS-5000 ⁺ CD	Baseline Noise	ASTM Type I Deionized water with low total organic carbon (TOC)* is pumped through the detector at a flow rate of 1.0 mL/min.	1.0 nS	2.0 nS
	Baseline Drift		10.0 nS/hr 3.33 nS/20 min	20.0 nS/hr 6.67 nS/20 min
	Detector Linearity	5 Nitrate solutions from the OQ/PQ standards kit P/N 063540 are injected in different concentrations. Concentration (ppm) and peak height (µS) are represented on a graph. The correlation coefficient for the resulting line and the deviations from it indicate the linearity.	r ≥0.999 RSD ≤5.0 %	r ≥0.999 RSD ≤5.0 %
ICS Series VWD	Baseline Noise	ASTM Type I Deionized water with low TOC is pumped through the detector at a flow rate of 1.0 mL/min. Wavelength 254 nm and 520 nm.	30 µAU	40 µAU
	Baseline Drift		200 µAU/hr	200 µAU/hr
	Wavelength Accuracy	Holmium oxide maxima are compared with the theoretical maxima.	± 2 nm	± 2 nm
	Detector Linearity	5 Caffeine solutions from the OQ/PQ standards kit P/N 063540 are injected in different concentrations. The absorbance is measured at a wavelength of 272 nm. Concentration (ppm) and peak height (mAU) are represented on a graph. The correlation coefficient for the resulting line indicates the linearity.	r ≥0.999 RSD ≤5.0 %	r ≥0.999 RSD ≤5.0 %

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Table 3. OQ/PQ Specification Limits: ICS-5000/ICS-5000⁺ PDA and ED Baseline Noise/Drift and Detector Linearity

Instrument	Parameter	Description	Limits	
			OQ	PQ
ICS Series PDA	Baseline Noise	ASTM Type I Deionized water with low total organic carbon (TOC) is pumped through the detector at a flow rate of 1.0 mL/min. Observed wavelengths are set for simultaneous monitoring at 254 nm and 520 nm	0.04 mAU	0.10 mAU
	Baseline Drift		0.16 mAU/20 min 0.5 mAU/hr	0.33 mAU/20 min 1.0 mAU/hr
	Wavelength Accuracy	Holmium oxide maxima are compared with the theoretical maxima.	± 2 nm	± 2 nm
	Detector Linearity	5 Caffeine solutions from the OQ/PQ standards kit P/N 063540 are injected in different concentrations. The absorbance is measured at a wavelength of 272 nm. Concentration (ppm) and peak height (mAU) are represented on a graph. The correlation coefficient for the resulting line (and the deviations from it) indicates the linearity.	r ≥0.999 RSD ≤5.0 % ≤ 1500 mAU	r ≥0.999 RSD ≤5.0 % ≤ 1500 mAU
ICS-5000/ICS-5000 ⁺ ED	Baseline Noise	50 mM NaOH is pumped through the detector cell at a flow rate of 0.25 mL/min. A Au-AAA certified disposable working electrode (P/N 060082) is used running the Amino Acids (pH, Ag/AgCl Reference) waveform.	Int. Amp: ≤ 100 pC	Int. Amp: ≤ 160 pC
	Baseline Drift		Int. Amp: ≤3000 pC/hr ≤1000 pC/20 min	Int. Amp: ≤3750 pC/hr ≤1250 pC/20 min
	Detector Linearity	5 Threonine solutions from the ED OQ/PQ standards kit P/N 063541 are injected in different concentrations. Concentration (µM) and peak height (nC) are represented by a plot. The correlation coefficient for the resulting line (and the deviations from it) indicates the linearity.	r ≥0.999 RSD ≤5.0 %	r ≥0.999 RSD ≤5.0 %

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Table 4. OQ/PQ Specification Limits: Injector Precision, Carryover and Sample Preparation

Instrument	Parameter	Description	Limits	
			OQ	PQ
All injectors and samplers	Injector Precision	10 injections of a defined standard at 25 μ L each of the same standard are analyzed. The percent relative standard deviation (%RSD) of the peak areas indicates the reproducibility of the injection volume.	RSD \leq 0.8 %	RSD \leq 1.0 %
	Carryover	Following the injection of a highly concentrated test sample, a blank sample is injected and then a water blank sample is injected. The peak area measured in this chromatogram compared with the peak area of the highly concentrated sample indicates the amount of carryover. If there is any peak area measured in the water blank sample, that area is subtracted from the area of the blank sample before it is compared to the highly concentrated sample.	\leq 0.1 %	\leq 0.1 %
AS-AP, AS	Linearity of Injector Volume	5 injections of the same standard sample are analyzed. The injection volumes are different (10 μ L to 50 μ L). Injection volume and peak area are represented on a plot of μ L vs. nC in ED and μ L vs. μ S in CD. The correlation coefficient of the resulting line (and the deviations from it) indicates the linearity.	r \geq 0.999 RSD \leq 1.0 %	r \geq 0.999 RSD \leq 1.0 %
AS	Sample Preparation Precision (if present – not supported on the AS with ICS-5000/ICS-5000 ⁺ ED detection)	3 samples of identical concentration are prepared from the same starting vial into 3 separate vials and injected. The relative standard deviation of the peak areas indicates the reproducibility of the sample preparation.	RSD \leq 1.5 %	RSD \leq 2.0 %
AS-AP	Temperature Accuracy (if present)	The sample temperature is measured using an external thermometer in a standard vial. The vial is filled with water and measured at 15.0 $^{\circ}$ C.	15.0 \pm 2.0 $^{\circ}$ C	15.0 \pm 4.0 $^{\circ}$ C
AS-AP pH	pH Accuracy	The sample temperature calibration is measuring the pH at 4, 7 and 10. The reading is stable if the reading and the three data points are within 0.2 pH unit of each other.	+/- pH 0.1	+/- pH 0.2

AS-AP Conductivity	Conductivity Accuracy	The conductivity calibration procedure consists of determining the conductivity zero point of the accessory and then measuring the conductivity of a 1000 µS standard.	+/- 30 µS	+/- 50 µS
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*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Table 5. OQ/PQ Specification Limits: ICS-5000/ICS-5000⁺ EG Current, SP/DP Flow Rate Accuracy/Precision and DC Column TC Temperature

Instrument	Parameter	Description	Limits	
			OQ	PQ
ICS-5000/ICS-5000 ⁺ EG	Current Qualification	The eluent generator (EG) is commanded to produce a series of different concentrations (using the current cartridge type configured with a concentration range between 0 mM to 100 mM). At each EG concentration setting the current (mA) produced by the EG is measured and compared with theoretical values.	See Modular Validation Report at: OQ_PQ_Modular_Validation	
ICS-5000/ICS-5000 ⁺ SP, ICS-5000/ICS-5000 ⁺ DP	Gradient Step Accuracy	A step gradient of channels A and B, then C and D is programmed and measured. Theoretical signal heights are compared with recorded signal heights for each step. The deviations between theoretical values and measured values indicate the step accuracy. For channels A and C Deionized water is used, for channels B and D 0.3% v/v acetone (absorbance detectors), 8.3 µM glucose (electrochemical detectors) or 1 mM KCl (conductivity detectors) is used.	≤2.0 %	≤2.0 %
	Gradient Step Noise	For each step of the gradient run described above the noise is determined relative to the signal height.	≤0.2 % for CD, VWD, PDA ≤0.5 % for ED	≤0.2 % for CD, VWD, PDA ≤0.5 % for ED

	Flow Rate Accuracy (Analytical Pump)	Water is pumped at a flow rate of 1.00 mL/min into a weighed beaker for exactly 5 minutes. The weight of the water is measured, the flow rate of the pump calculated and compared with the theoretical value. This experiment is repeated four more times.	1.00 ± 0.05 mL/min	1.00 ± 0.05 mL/min
	Flow Rate Precision	Analytical Systems: Water is pumped at a flow rate of 1.00 mL/min into a weighed beaker for exactly 5 minutes. This procedure is repeated four more times. The relative standard deviation of the five measurements is calculated.	≤2.0 %	≤2.0 %
ICS-5000/ICS-5000 ⁺ DC	Column TC and Compartment, Temperature Verification	The Analytical DC column temperature is set to 30 °C and the Analytical DC compartment temperature is set to 20 °C for the duration of the entire qualification. At the end of the qualification the temperature of the column heater is measured with a calibrated external thermometer and compared with the theoretical value.	± 2.0 °C	± 2.5 °C

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Note: The instrument's specifications can only be met if the conditions that are described in the accompanying manual are strictly observed.

Note: The VWD and PDA Detector UV and visible lamps must be turned on at least two hours prior to performing the qualification.

3 Explanation of Test Procedures

3.1 Flow Rate Accuracy and Precision Qualification

3.1.1 Theory

The accuracy and precision of an analytical pump is qualified by accurately measuring 5 aliquots of eluent into a tared sample beaker for exactly 5 minutes each. The injection valve is used to measure the 5 minute aliquot and the volume of each aliquot is determined gravimetrically. The backpressure coil is placed before the injection valve and equal lengths of tubing are used for eluent and waste to ensure that there is no backpressure change when switching the valve from waste to sample.

3.1.2 Performing the Test and Evaluating the Results

The Flow Rate Accuracy and Precision Qualification are run using the sequence called XQ_Flow_Rate. The five weight measurements for the aliquots are entered manually into the report and converted to volume when the temperature is entered. Accuracy is measured by comparing the theoretical value (5.00 mL in 5.00 min) to each of the five aliquots and precision is measured by determining the relative standard deviation of the five aliquots.

3.2 Eluent Generator Current Qualification

3.2.1 Theory

The Eluent Generator Current Qualification is determined by performing an EG step gradient using varying eluent concentration set points. The EG applies a specific current to the EG cartridge depending on the concentration of eluent and type of cartridge selected. An IC Qualification test box (P/N 072005) is used in place of the EG cartridge and the applied current for each step is measured using an ammeter or voltmeter that is connected to the test cell. No injection is performed.

3.2.2 Performing the Test and Evaluating the Results

The Eluent Generator current qualification test is the next test in the sequence queue and the sequence is called XQ_EG_Current or XQ_EG_Dual_Current. The current readings in mA are displayed on the voltmeter for each concentration step in the gradient. Values for each step are manually recorded (please enter at least three figures after the decimal in each entry) or can be automatically downloaded into the Report Template when using a Fluke 189/289 or equivalent multimeter (calibrated with up to 300 mV DC scale range and the multimeter needs to display three decimal places) which is configured in the instrument as a module using the appropriate device driver. The sequence follows an XQ_Stop sequence that will pause the system to allow connection of the qualification test box and recording of the results.

3.3 Warm up

The warm up sequence is used to ensure that all parts of the system are responding appropriately and to record logs to the audit trail (later used to update the information in the report definition file). It is called XQ_Warm_Up where; X is P for PQ and O for OQ. The warm up sequences from the AS-AP directory inject the low level standards.

3.4 Baseline Noise and Drift (Conductivity and Absorbance Detector)

3.4.1 Theory

A 20-minute detector baseline background trace is collected using Deionized water as the eluent at a flow rate of 1.00 mL/min. For ICS Series VWD and PDA detectors, data is collected at 254 nm and 520 nm. No injection is performed. Then a short equilibration is done at 272 nm.

Noise is calculated by partitioning the 20-minute baseline signal into 20 one-minute intervals. The noise is the distance between two parallel lines through the measured minimum and maximum values and the regression line. The measured noise values are then averaged to establish the final value. To calculate drift, the Chromeleon software uses a method of least squares and calculates a regression line from the measured values. The slope of the regression line indicates the drift of the measured signal, and the amount of the slope indicates the amount of drift.

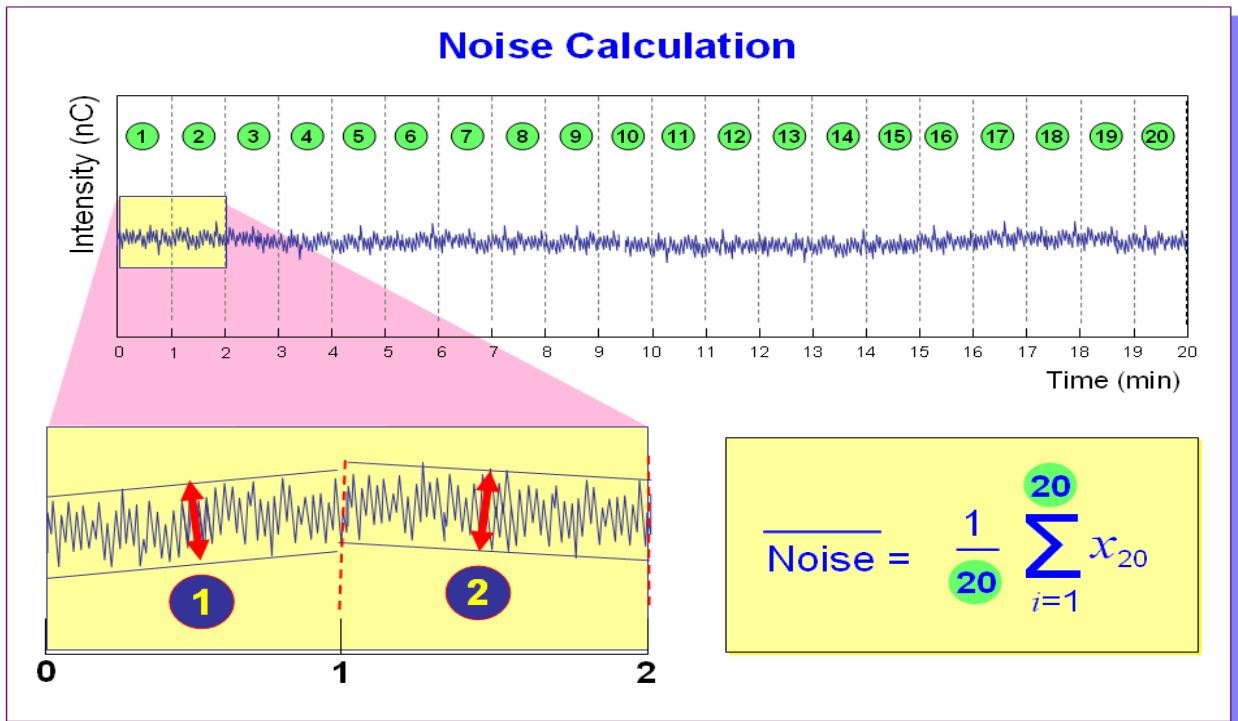


Figure 1. Noise Calculation

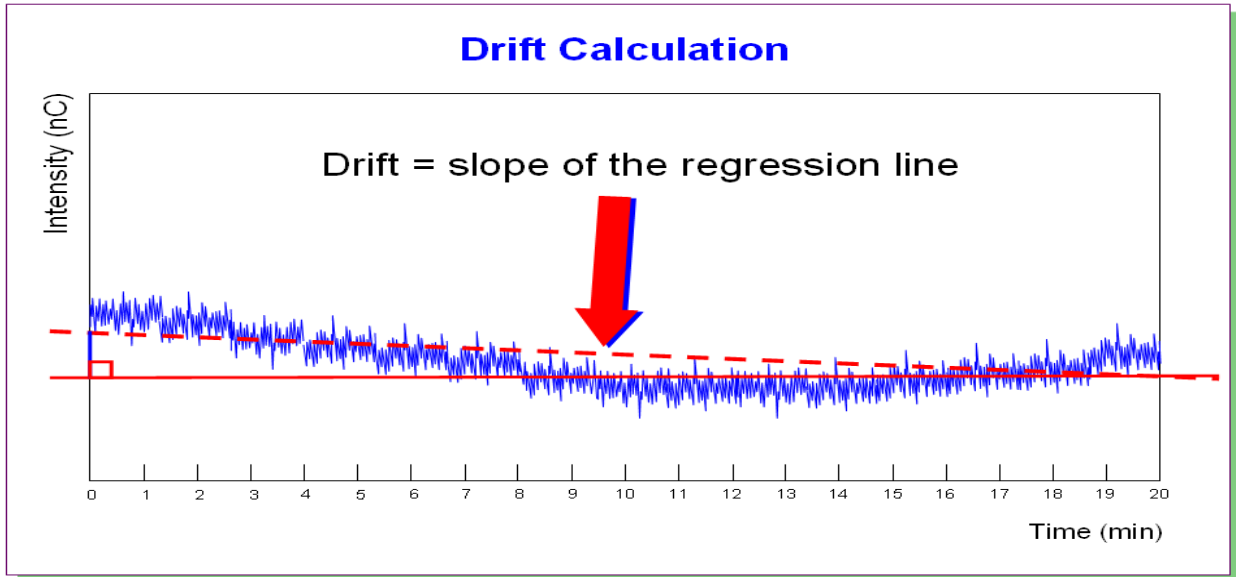


Figure 2. Drift Calculation

The slope of the regression line indicates the drift of the measured signal, and the amount of the slope indicates the amount of drift.

3.4.2 Performing the Test and Evaluating the Results

Noise and drift evaluation for all IC detectors is the next test in the sequence queue and is called XQ_Detector_Noise_Drift where X is P for PQ and O for OQ and Detector is CD for conductivity detectors, UV_Vis for PDA and VWD detectors.

Note: Ensure that the seal wash option is turned off from the Chromeleon F8 commands window during the qualification. After the OQ/PQ tests are finalized, be sure to re-enable the seal wash option.

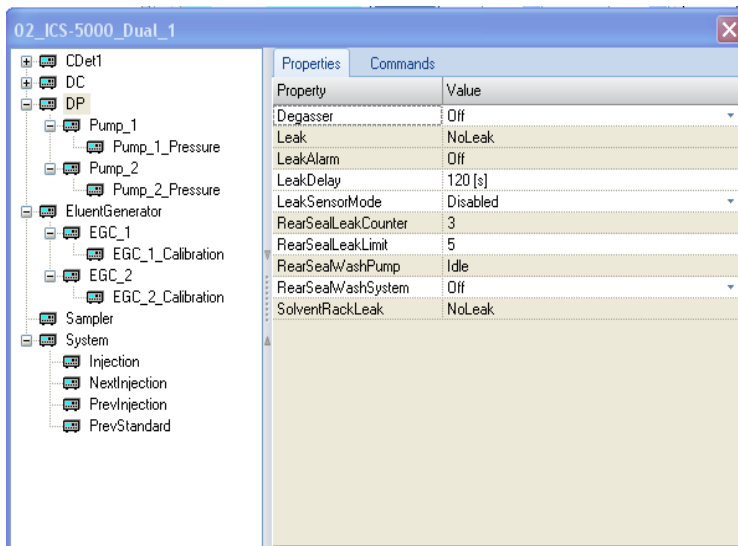


Figure 3. Rear seal wash option in the F8 commands window

3.5 Noise and Drift (Integrated Amperometry)

3.5.1 Theory

A 20-minute detector baseline is collected using 50 mM NaOH as the eluent at a flow rate of 0.25 mL/min. An Amino Acid disposable gold working electrode (Thermo Fisher Scientific P/N 060082) is used along with the Amino Acid (pH/Ag/AgCl Reference) waveform. This test requires a pH/Ag/AgCl reference electrode. No injection is performed.

3.5.2 Performing the Test and Evaluating the Results

Noise and drift evaluation for Integrated Amperometry detector is the only test in the sequence Queue and is called XQ_ED_Amp_Noise_Drift.

Noise is calculated by partitioning the 20-minute baseline signal into 20 one-minute intervals. The noise is the distance between two parallel lines through the measured minimum and maximum values and the regression line. The measured noise values are then averaged to establish the final value. To calculate drift, the Chromeleon software uses a method of least squares and calculates a regression line from the measured values. The slope of the regression line indicates the drift of the measured signal, and the amount of the slope indicates the amount of drift (Figure 1 and 2).

Note: Ensure that the seal wash option is turned off during the qualification. After the OQ/PQ tests are finalized, be sure to re-enable the seal wash option.

3.6 Injector Precision

3.6.1 Theory

Injector precision is determined by performing ten replicate injections of a standard using a fixed sample volume.

PDA/VWD: A 40 ppm caffeine standard is injected when using absorbance detection at a wavelength of 272 nm. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

Conductivity Detector: A 50 ppm nitrate standard is injected when using conductivity detection. Deionized water is used as the eluent at a flow rate of 1.00 mL/min with conductivity.

Electrochemical Detector: A 10 μ M threonine standard is injected when using ICS-5000/ICS-5000⁺ ED detector in Integrated Amperometry mode. A 50 mM manually prepared NaOH is used as eluent at a flow rate of 0.25 mL/min with a 2 mm x 50 mm CarboPac PA1 (P/N 057179) guard column. An Amino Acid disposable gold working electrode (Thermo Fisher Scientific P/N 060082) and a 2 mil x 1 mm Teflon gasket (Thermo Fisher Scientific P/N 060141) are used with the Amino Acids (pH, using Ag/AgCl Reference electrode) waveform.

3.6.2 Performing the Test and Evaluating the Results

Injector precision evaluation is the next test in the sequence queue and the samples are called Inj Precision_1 through Inj Precision_10 in the XQ_Detector_Inj_Precision sequence, where X is O for OQ, P for PQ, and Detector is CD for conductivity detectors, ED for amperometry detectors and UV for absorbance detectors. The concentration of the nitrate or caffeine or threonine standard and the sample volume injected is included in the QNT file used for injector linearity.

Precision is determined by calculating the relative standard deviation of the peak areas for the ten injections.

3.7 Sample Carryover

3.7.1 Theory

Sample carryover is determined by injecting a highly concentrated sample, followed by a blank sample and then a second blank injection. The blanks in this case are Deionized water.

PDA/ VWD: Caffeine at a concentration of 80 ppm is injected when this test is performed with an absorbance detector.

Conductivity Detector: Nitrate at a concentration of 1000 ppm is injected when this test is performed with a conductivity detector.

Electrochemical Detector: A 10 μ M threonine standard is injected when using ICS-5000/ICS-5000⁺ ED detector in Integrated Amperometry mode. A 50 mM manually prepared NaOH is used as eluent at a flow rate of 0.25 mL/min with a 2 mm x 50 mm CarboPac PA1 (P/N 057179) guard column. An Amino Acid disposable gold working electrode (Thermo Fisher Scientific P/N 060082) and a 2 mil x 1 mm Teflon gasket (Thermo Fisher Scientific P/N 060141) are used with the Amino Acids (pH, using Ag/AgCl Reference electrode) waveform.

Ideally, the chromatogram of the two blank injections should be identical. If, however, a difference is detected, the difference between the peak areas is calculated and the ratio of this difference to the highly concentrated sample is calculated. ASTM Type 1 water is used as the eluent at a flow rate of 1.00 mL/min for absorbance and conductivity detectors.

3.7.2 Performing the Test and Evaluating the Results

Sample carryover evaluation is included in the same sequence as the Injector Precision test of the sequence queue and the samples are called Water, Carryover and Water_CO in the *XQ_Detector_Inj_Precision_CO* sequence.

A peak area ratio is calculated between the blank and high concentration sample to determine sample carryover.

3.8 Detector Linearity

3.8.1 Theory

VWD/PDA: Detector linearity for absorbance detectors is determined at 272 nm using a sequence of five caffeine standard injections at 15, 40, 60, 80 and 100 ppm. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

Conductivity Detector: Detector linearity for conductivity detectors uses a sequence of five nitrate standard injections at 5, 10, 25, 50, and 100 ppm. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

Electrochemical Detector: Linearity for Integrated Amperometry detectors uses a sequence of five Threonine standard injections at 1, 2, 4, 8, and 10 μ M. A 50 mM manually prepared NaOH is used as eluent at a flow rate of 0.25 mL/min with a 2 mm x 50 mm CarboPac PA1 (P/N 057179) guard column. An Amino Acid disposable gold working electrode (Thermo Fisher Scientific P/N 060082) and a 2 mil x 1 mm Teflon gasket (Thermo Fisher Scientific P/N 060141) are used with the Amino Acids (pH, using Ag/AgCl Reference electrode) waveform.

3.8.2 Performing the Test and Evaluating the Results

Detector linearity samples are called Detector linearity_1 through Detector linearity_5 in the *XQ_Detector_Linearity* sequence. The concentrations of the nitrate, caffeine and threonine standards are included in the QNT file used for detector linearity. It is recommended to enter the actual concentration of the standards from the certification of analysis into the quantification files to achieve the real results.

Depending on which injection module used, the peak height of the sample with the highest concentration may exceed 1500 mAU. This is usually not within the linearity range of UV detectors. Thus, the limits for the regression coefficient and the relative standard deviation may not be met. In this case, reduce the injection volume for all samples used for the linearity check so that the peak height of the sample with the highest concentration is in the linearity range of the tested detector, i.e., usually below 1500 mAU. As an exception, a linearity range of up to 2500 mAU is specified for the supported VWD with an analytical flow cell installed.

If an AS or AS-AP autosampler is used when the PDA or PDA-100 detectors are tested, only 10 μL of sample will be injected by default. However, when qualifying the Thermo Fisher Scientific VWD detectors using an AS autosampler, 15 μL of sample will be injected.

3.9 Injection Volume Linearity

3.9.1 Theory

Injection volume linearity is determined by making five injections of a standard using injection volumes of 10, 20, 30, 40 and 50 μL .

PDA/ VWD: A 60 ppm caffeine standard is injected when using absorbance detection at a wavelength of 272 nm. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

Conductivity Detector: A 50 ppm nitrate standard is injected when using conductivity detection. Deionized water is used as the eluent at a flow rate of 1.00 mL/min.

Electrochemical Detector: An 4 μM Threonine standard is injected when using integrated amperometry detection. 50 mM NaOH is used as the eluent at a flow rate of 0.25 mL/min. An Amino Acid disposable gold working electrode (Thermo Fisher Scientific P/N 060082) and a 2 mil x 1 mm Teflon gasket (Thermo Fisher Scientific P/N 060141) are used with the Amino Acids (pH, using Ag/AgCl Reference electrode) waveform.

This test is only performed on autosamplers with variable injection volume (e.g. AS or AS-AP) for analytical systems. A 250 μL syringe is required to be configured in the AS-AP to run the injection volume linearity test.

3.9.2 Performing the Test and Evaluating the Results

Injection volume linearity evaluation is run using the sequence called *XQ_Detector_Inj_Linearity*. The concentration of the standards and the injection volumes are included in the QNT file used for detector linearity.

3.10 Sample Preparation Precision (AS only)

3.10.1 Theory

Sample Preparation precision (conductivity detection for IC systems; absorbance detection for analytical systems only; electrochemical detection is not supported) is determined by making three separate injections using a fixed injection volume. Each of the three standards is prepared

individually using the autosampler's sample preparation option. The autosampler prepares the three samples by first transferring 15 μL of a 1000 ppm caffeine or nitrate standard into three empty sample vials followed by the addition of 1485 μL of water to each of the sample vials. Injections are then made from each vial. Deionized water is used as the eluent at a flow rate of 1.00 mL/min in analytical systems. If an absorbance detector is used the wavelength is set to 272 nm. This test is only performed on autosamplers with the sample preparation option installed.

3.10.2 Performing the Test and Evaluating the Results

Sample prep precision evaluation is the next test in the sequence queue and the sequence is called XQ_Detector_Samp_Prep.

Required configuration for running a successful sample preparation experiment:

- AS configured with a 10.0 mL tray (P/N 026374). Note: Sample Prep only supports 10 mL vials.
- System set up with conductivity or an absorbance detector.

Precision is determined by calculating the relative standard deviation (RSD) of the peak areas from the three vials.

3.11 Gradient Proportioning Accuracy

3.11.1 Theory

Gradient proportioning accuracy is determined by performing a step gradient using varying compositions of deionized water and either 0.3 % Acetone with absorbance detectors or 1 mM KCl for conductivity detectors. When performing the test with an absorbance detector, deionized water is used for eluents A and C and 0.3 % Acetone is used for eluents B and D with a wavelength setting of 254 nm. With conductivity detection eluents B and D are 1 mM KCl.

When performing the test with an electrochemical detector, 50 mM NaOH is used for eluents A and B and 50 mM NaOH with 8.3 μM glucose is used for eluents B and D.

Gradient composition is measured at 0, 20, 50, 80, and 100 % of eluents B and D at a flow rate of 1.00 mL/min in analytical systems. Step noise (ripple) is measured for each step in the gradient. No injection is performed.

3.11.2 Performing the Test and Evaluating the Results

Gradient Accuracy evaluation is called XQ_GP_Grad_Accuracy.

The measured step height is compared to the theoretical step height and a percent deviation is calculated. Step noise is evaluated for all steps. Each step has a defined one-minute interval where noise is evaluated. Based on a method of least squares, Chromeleon calculates a regression line from the measured values of each interval. The noise is the distance between two parallel lines through the measured minimum and maximum values and the regression line.

3.12 Column/Compartment TC Temperature Qualification

3.12.1 Theory

The temperature of a column/compartment TC is qualified by measuring the temperature of the column/compartment with a calibrated thermometer after allowing the column/compartment and thermometer to equilibrate for a minimum of 15 minutes. The temperature is recorded from the thermometer.

3.12.2 Performing the Test and Evaluating the Results

During the sequence queue the column compartment is set to 30 °C and the Analytical DC compartment temperature is set to 20 °C. The Column Compartment Temperature Qualification is performed after the completion of the sequence queue. The recorded temperature is manually entered into the report.

3.13 Wavelength Accuracy of Absorbance Detectors

3.13.1 Theory

Wavelength accuracy is evaluated using a holmium oxide filter. Three holmium oxide absorbance maxima are compared with the theoretical maxima at 360.9 nm, 418.0 nm and 536.6 nm for ICS Series PDA and VWD detectors.

3.13.2 Performing the Test and Evaluating the Results

For the VWD or the PDA detectors the wavelength accuracy test is the first test in the sequence queue and is called XQ_Wavelength_Accuracy where, X is P for PQ and O for OQ. The Wavelength Accuracy Test program executes the verification and the results are logged to the audit trail by the Wavelength Accuracy Report program.

3.14 Sample Temperature Accuracy of Autosamplers (AS-AP only)

3.14.1 Theory

The sample temperature accuracy mainly depends on the cooling and heating accuracy of the AS-AP autosampler, the insulation of the sample compartment, and the thermal transfer from the carousel to the vial.

3.14.2 Performing the Test

The sample temperature accuracy is determined with the help of an external thermometer. The temperature sensor is placed in a standard polypropylene vial (10 mL or 1.5 mL) filled with water. The carousel cover must be closed during the test. The autosampler temperature is set to a specified temperature. When the set temperature is reached, the sample (water) temperature is recorded at the end of the 30 minute period. Within the 30 minutes, the sample temperature reaches a stable value. During the run, the program will keep the carousel from moving. The temperature accuracy is the temperature difference between the sample temperature and the nominal autosampler temperature.

3.15 Sample pH Conductivity (AS-AP only)

3.15.1 Theory

The sample pH qualification is evaluated by checking the pH calibration and pH accuracy. The sample temperature calibration is measuring the pH at 4, 7 and 10. The reading is stable if the reading and the three data points are within 0.1 pH unit of each other. The pH measurement of the accessory must be calibrated before you begin using the accessory and then periodically thereafter. During pH calibration, the pH of a pH 7 buffer solution is measured first and *the offset is determined*. Then, the pH of either a pH 4 or a pH 10 buffer solution is measured and a slope between pH 7 and pH 4 (or pH 10) is determined. Optionally, the pH of a third buffer solution (pH 4 or pH 10) can be measured and a slope between pH 7 and the third buffer determined. The conductivity calibration procedure consists of determining the conductivity zero point of the accessory and then measuring the conductivity of a 1000 µS standard. Chromeleon determines the calibration offset and slope and calculates a new cell constant.

4 Qualification Preparation

Materials required for system qualification depend on the system configuration. Table 6 lists the kits available to be used with the ICS-5000/ICS-5000⁺ Analytical configurations. Table 7 describes the base kit for several instrument configurations. Tables 7b and 7c describe consumable items used with each of the two kits.

Changes or additions to a system configuration may require additional materials.

4.1 Materials in Kits

Table 6. Required Kits for ICS-5000/ICS-5000⁺ Analytical Configurations

Part Description	Part Number
IC OQ/PQ Kit for IC systems (PROD,KIT,OQ/PQ,ICS,CD ABS)	063540
IC OQ/PQ Kit for ICS-5000/ICS-5000 ⁺ or ICS-3000 Analytical ED detectors (PROD,KIT,OQ/PQ,ICS,ED)	063541

Table 7. Hardware items in 063540 and 063541 Kits

Part Description	Part Number	Quantity
3 – Way mixing tees <ul style="list-style-type: none"> Connect eluent lines and eluent bottles 	024314	2
Certificate of Functional Performance	031423	6
Test Binder	035863	1
Tubing, 0.010" (0.25 mm) ID x 60" (150 cm) length black PEEK <ul style="list-style-type: none"> Connect pump to injection valve, backpressure tubing to detector, etc. 	042690	60" (152 cm)
25 µL Sample Loop <ul style="list-style-type: none"> AS-DV or Manual injection 	042857	1
100 µL Sample Loop <ul style="list-style-type: none"> AS-AP or AS autosamplers 	042951	1
Red PEEK Tubing 0.005" (0.125 mm) ID x 39" (99 cm) length <ul style="list-style-type: none"> Generate system backpressure and delay volume for detector and injector tests 	044211	39" (99 cm)
Restrictor Microbore, Gradient Test	049630	1
Yellow PEEK Tubing (0.003", 0.075 mm ID) <ul style="list-style-type: none"> Generate system backpressure and delay volume for detector and injector tests in Analytical Systems only 	049715	39" (99 cm)
Qualification labels	050837	6

OQ/PQ User's Manual Part E Version 7.10 (this document)	065448 Part E	1
High-pressure ferrules, double cone, 10-32	074373	6
High-pressure bolt, 10-32	074449	6

Table 7b. Consumables in 063540 Kit for Conductivity or Absorbance Detection

Part Description	Part Number	Quantity
NIST Traceable Caffeine Standards Kit <ul style="list-style-type: none"> • <i>Absorbance detectors</i> 	060253	1
NIST Traceable Nitrate Standards Kit <ul style="list-style-type: none"> • <i>Conductivity detectors</i> 	060254	1

Table 7c. Consumables in 063541 Kit for Amperometric (ED) Detection

Part Description	Part Number	Quantity
CarboPac PA1 Guard 2 mm	057179	1
Disposable Amino Acid AAA Working Electrodes, 3 w/ gasket	060082	1
NIST Traceable Threonine Standards Kit for OQ/PQ	063542	1
STD, GLUCOSE, 10% SOLUTION, 100mL	080150	1

4.2 Additional Required Materials

The following additional items will be needed:

Table 8. Additional Materials Needed

Description	Quantity
ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered) <ul style="list-style-type: none"> All eluent, standard, and sample preparation 	2 L
Reagent Grade or better Potassium Chloride (KCl) for OQ/PQ gradient tests using conductivity detection <ul style="list-style-type: none"> Gradient pump tests using conductivity detectors 	1 g
HPLC grade Acetone for OQ/PQ gradient tests using optical absorbance detection <ul style="list-style-type: none"> Gradient pump tests using optical detectors 	3 mL
Sodium Hydroxide 50/50 w/w% (Low Carbonate) (Thermo Fisher Scientific P/N 033465) <ul style="list-style-type: none"> Prepare eluent for Integrated Amperometry qualification 	~5.2 mL
Ammeter or digital volt multimeter (DVM) with milliamp (300 mA scale) setting for OQ/PQ tests using an EG. Must be capable of displaying 0.00 to at least 220 mA. A Fluke Model 189, 289 or equivalent DVM will allow for automated EG Current testing.	1
Digital thermometer. Temperature accuracy test can be performed with a Dostmann P600 Thermometer or equivalent. The P600 is included in Kit (P/N 5705.0050A)	1
Type K temperature sensor for Dostmann P600 Thermometer (P/N 6820.0010)	1
Thermo Fisher Scientific analytical conductivity cell (detector) <ul style="list-style-type: none"> Detector, injector and gradient pump tests using conductivity detectors in conductivity mode 	1
AS or AS-AP in pull mode: 10 mL vials (P/N 074228 or 055058) are required for AS-AP pull mode and AS Sample Preparation Qualification	10
AS-AP: 1.5 mL vial tray (P/N 066332)	1
AS-AP: 1.5 mL vials (P/N 061696)	7
Black tubing 48" (P/N 042690)	1
AS-DV 5.0 mL vials with filter caps (P/N 038141) if qualifying an AS-DV autosampler	20
100 mL volumetric flasks (Class A, 100 ± 0.08 mL) <ul style="list-style-type: none"> Standard preparation for AS-DV and Manual Injection 	1
1000 mL volumetric flasks (Class A, 1000 ± 0.3 mL) <ul style="list-style-type: none"> Eluent preparation 	At least 1
Transfer Pipettes or Pipettor	0.5 – 10.0 mL
Calibrated Analytical Balance - capable of mass recording to the nearest milligram or better <ul style="list-style-type: none"> Weigh KCl for eluent preparation Weigh eluent for the Flow Rate Accuracy and Precision test 	1
Calibration and Check Standard pH 4 (P/N 033238)	1
Calibration and Check Standard pH10 (P/N 033240)	1
1000 uS Standard (P/N 080172)	1
Computer workstation running Chromeleon™ Version 7.10 (with SR1) or later (later versions required for some configurations, see Chapter 1.6)	1

Deionized water should be vacuum filtered through a 0.2 or 0.45 µm nylon or other water compatible membrane exhibiting very low extractables.

A 48" black tubing (P/N 042690) is recommended to be added to the yellow tubing (P/N 049715) from the injection valve to the conductivity detector to reduce pump ripple and cell noise.

All volumetric ware, pipettes and standard eluent storage bottles should be made of low extractable polymeric materials such as polypropylene or polystyrene

4.3 Preparation of Standards

Two standard kits are provided in the OQ/PQ kit. Use the Caffeine Kit, P/N 060253 to qualify systems with an absorbance detector (ICS Series VWD or PDA). Use the Nitrate kit, P/N 060254 to qualify systems with a conductivity detector.

To qualify an ICS-5000/ICS-5000⁺ system with an ED detector in Integrated Amperometry mode use the Threonine standard kit for ED qualification (Thermo Fisher Scientific P/N 063542). ED detector qualification does not support AS-DV autosamplers.

4.3.1 Caffeine standard preparation for the Injector Precision and Carryover tests (AS-DV and manual injection only)

AS-DV Autosampler only: For the Injector Precision test prepare a 40 ppm caffeine standard using the 1000 ppm caffeine standard ampoule included in the Caffeine Standards Kit. Pipette 4.0 mL of the 1000 ppm caffeine standard into a 100 mL volumetric flask. Fill the flask to the mark with ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered).

AS-DV Autosampler and Manual Injection: For the Carryover test use the 80 ppm caffeine standard included in the Caffeine Standards Kit.

4.3.2 Nitrate standard preparation for the Injector Precision test (AS-DV and manual injection only)

For the Injector Precision test prepare a 50 ppm nitrate standard using the 1000 ppm nitrate standard vials included in the Nitrate Standards Kit. Pipette 5.0 mL of the 1000 ppm nitrate standard into a 100 mL volumetric flask. Fill the flask to the mark with ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered).

For the Carryover test use the included 1000 ppm nitrate standard found in the nitrate standards kit without dilution.

4.4 Preparation of Eluents

All eluents must be prepared using ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered). Eluent should be freshly prepared prior to the execution of the OQ/PQ tests.

4.4.1 Preparation of Eluents for use with Conductivity Detection

Prepare the following eluents and connect them to lines A, B, C and D of the pump. All eluents must be degassed. Eluent lines A and C can be connected to one eluent bottle containing 1 L of water by a mixing tee. Eluent lines B and D can be connected to one eluent bottle containing 1 L of 1 mM KCl by a mixing tee.

Note: Eluents B, C and D are needed for the Gradient Pump Accuracy Test only and should be prepared only if the pump configured has gradient capability.

Table 9. Eluent Channels for Gradient Performance Test with Conductivity Detector

Eluent	Description
A	Deionized water
B	1.0 mM KCl
C	Deionized water
D	1.0 mM KCl

10 mM KCl Stock Solution Preparation:

Prepare one liter of 10 mM KCl by weighing 0.746 g of KCl. Place the KCl in a 1 L volumetric flask. Fill the flask to the mark with ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered).

1 mM KCl Preparation:

Pipette 100 mL of the 10 mM KCl solution into another 1 L volumetric flask, filling the flask to the mark with ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered). The 1 mM KCl solution should be prepared fresh daily.

Remember to ...

- Degas all eluents and autosampler wash solutions. Degassing is not required, but is recommended for most consistent results.
- Connect eluent lines A and C to one eluent bottle containing 1 L of deionized water equipped with a 3-way mixing tee.
- Connect eluent lines B and D to one eluent bottle containing 1 L of 1 mM KCl equipped with a 3-way mixing tee.
- Eluent bottles for ICS-5000/ICS-5000⁺ pumps do not need to be pressurized.
- Ensure that the rear seal wash supply bottle is filled with fresh deionized water or the rear seal wash pump is turned off from the F8 commands window.

4.4.2 Preparation of Eluents for use with Absorbance Detection

These eluents are used for qualifications with an ICS-Series Variable Wavelength (VWD) or ICS-Series Photodiode Array Detector (PDA).

Prepare the following eluents and connect them to lines A, B, C and D of the pump. All eluents must be degassed. Eluent lines A and C can be inserted into one eluent bottle containing 1 L of water. Eluent lines B and D can be inserted into one bottle containing 1 L of 0.3 % acetone in water.

Note: Eluents B, C and D are needed for the Gradient Pump Accuracy Test only and should be prepared only if the pump configured has gradient capabilities. Do not prepare these eluents if the pump configured is an ICS-5000/ICS-5000⁺ isocratic pump since it does not have gradient mixing capabilities.

Table 10. Eluent Channels for Gradient Performance Test with Absorbance Detector

Eluent	Description
A	Deionized water
B	0.3 % Acetone
C	Deionized water
D	0.3 % Acetone

0.3 % Acetone in Water Preparation:

Pipette 3 mL of HPLC grade Acetone into a 1 L volumetric flask. Fill flask to the mark with ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered). Eluents should be freshly prepared on a daily basis.

Remember to ...

- Degas all eluents and autosampler wash solutions. Degassing is not required, but is recommended for most consistent results.
- Connect eluent lines A and C to one eluent bottle containing 1 L of deionized water using a 3-way mixing tee.
- Connect eluent lines B and D to one eluent bottle containing 1 L of 0.3 % Acetone in water using a 3-way mixing tee.
- Eluent bottles on ICS-5000/ICS-5000⁺ pumps do not need to be pressurized.
- Ensure the new seal wash solution is renewed or the new seal wash pump is turned off.

4.4.3 Preparation of Eluent for use with Integrated Amperometry Detection in Analytical systems

There are two different eluents used for qualification with an ICS-5000/ICS-5000⁺ Electrochemical Detector in Integrated Amperometry mode:

For Flow Test (initial Flow test):

Eluent	Description
--------	-------------

A	Deionized water
---	-----------------

For all other ED Tests:

Eluent	Description
A	50 mM NaOH

The preparation of 50 mM NaOH for the ED tests should be performed after the Pump Flow Rate test has been completed to ensure minimal potential contamination of 50 mM NaOH eluent. Do not make the 50 mM NaOH eluent until section 5.3.2. All eluents must be degassed. Eluent lines A should be connected to one eluent bottle containing 1 L of 50 mM NaOH. After the 50 mM NaOH has been connected to line A, the line should be thoroughly primed.

50 mM NaOH Preparation:

Fill eluent bottle with 997.4 mL of ASTM Type I (or better) deionized, degassed water (18.0 Megohm-cm, filtered). Pipette 2.6 mL of 50/50 (w/w) NaOH solution into the bottle. Cap, cover with Helium (or Nitrogen) head pressure and mix. The 50 mM NaOH solution should be prepared daily and care should be taken to minimize exposure to CO₂ gas.

Remember to ...

- Degas all eluents and autosampler wash solutions. Degassing is not required, but is recommended for most consistent results.
- Connect eluent line A to one eluent bottle containing 1 L of 50 mM NaOH.
- Pressurize eluent bottles with Helium (or Nitrogen) gas.
- Use the disposable AAA Gold working electrode (P/N 060082)

4.4.4 Preparation of Eluent for use in Gradient Performance Test with Integrated Amperometry Detection

Prepare the following eluents and connect them to lines A, B, C and D of the pump. All eluents must be degassed. Eluent lines A and C can be connected to one eluent bottle containing 50 mM NaOH by a mixing tee. Eluent lines B and D can be connected to one eluent bottle containing 50 mM NaOH with 8.3 μM Glucose (P/N 080150) by a mixing tee.

Note: Eluents B, C and D are needed for the Gradient Pump Accuracy Test only and should be prepared only if the pump configured has gradient capabilities.

Table 11. Eluent Channels for Gradient Performance Test with Electrochemical Detector

Eluent	Description
A	50 mM NaOH
B	8.3 μM Glucose in 50 mM NaOH
C	50 mM NaOH
D	8.3 μM Glucose in 50 mM NaOH

50 mM NaOH Preparation:

Fill the eluent bottle with 997.4 mL of ASTM Type I (or better) deionized, degassed water (18.0 Megohm-cm, filtered). Pipette 2.6 mL of 50/50 (w/w) NaOH solution into the bottle. Cap, cover with Helium (or Nitrogen) head pressure and mix. The 50 mM NaOH solution should be prepared daily and care should be taken to minimize exposure to CO₂ gas.

8.3 μM Glucose in 50 mM NaOH Preparation:

Fill the eluent bottle with 1000 mL of previously prepared 50 mM NaOH. Pipette 15 μL of 555 mM Glucose, cover with Helium (or Nitrogen) head pressure and mix. The 50 mM NaOH solution should be prepared daily and care should be taken to minimize exposure to CO₂ gas.

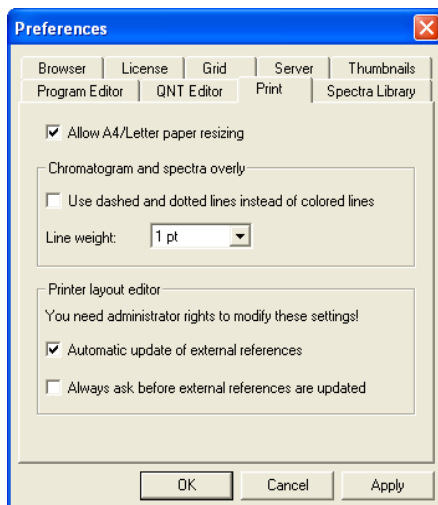
Remember to ...

- Degas all eluents and autosampler wash solutions. Degassing is not required, but is recommended for most consistent results.
- Connect eluent lines A and C to one eluent bottle containing 1 L of 50 mM NaOH using a 3-way mixing tee.
- Connect eluent lines B and D to one eluent bottle containing 1 L of 50 mM with 8.3 μM glucose using a 3-way mixing tee.
- Pressurize eluent bottles with Helium (or Nitrogen) gas.
- Use the disposable carbohydrate working electrode (P/N 060082)

4.5 Preparing the IC System

Note: The ICS Series VWD and PDA Detector UV and visible lamps must be turned on at least two hours prior to performing the qualification to ensure proper equilibration..

1. Chromeleon settings: Ensure that Chromeleon is set for Automatic update of external references. Click on the "Automatic update of external references" check box in File → Preferences → Print menu.



2. Open the Chromeleon Instrument Configuration application. Only one pump, one injection valve and one detector can be qualified concurrently, so if more than one pump, one injection valve or one detector are installed, the second pump (Pump_2), the second detector (CDet_2, EDet_2) will be ignored if both detectors are the same. The detector names can be configured in the Instrument Configuration. If the detectors are different, uncheck the detector signal that will not be qualified. If the second pump or detector is to be qualified, remove (or uncheck) the first pump or detector in the Timebase. Make note of any changes made to the system for the purposes of this qualification. After qualification these changes should be reversed. It is helpful to save a backup file of the configuration before you make any changes for qualification.
3. AUTOSAMPLER: Double click on the Autosampler to be qualified (if installed) to view the properties page for the autosampler. Select the **General** tab and confirm the Device Name is "Sampler". Select the **Options** tab and confirm the options match the configuration of your system. The AS-AP injection mode must be selected at this point. Only one mode is qualified. Select the **Devices** tab and confirm the Autosampler diverter valve status matches your system. For the AS-AP autosampler in pull mode, the Flush Volume and Flush Volume2 must be set with F8 commands or from the **More Options** page of the AS-AP panel before beginning qualification. Flush Volume = 2 x (Needle Volume + Needle Line Tubing Volume) μ L where Needle Volume = 30 μ L and Needle Line Tubing Volume (the volume of tubing between the needle and the injection port) = 0.86 μ L/cm (2.2 μ L/in) of 0.33-mm (0.013-in) ID PEEK tubing (blue). Set the Flush Volume2 parameter to 30 μ L

NOTE: Make sure inject mode in the commands is consistent with the inject mode in the Instrument Configuration before loading the OQPQ templates

4. ICS-5000/ICS-5000⁺ Pump: Double click on the pump to be qualified to view the properties page for the pump. Select the **Devices** tab and confirm the Device Name is "Pump_1" or "Pump_2".
5. ICS-5000/ICS-5000⁺ DC: Double click on the DC to be qualified to view the general properties. Select the Thermal Controls tab and confirm the heating configuration of the Timebase. Confirm that the device name for the Column TC is "Column_TC" and the device name for the Compartment TC is "Compartment_TC" if installed on the Timebase.
6. DETECTOR: Double click on the detector to be qualified to view the properties page for that detector (for CD and ED this will be in the **DC** under the **Detector** tab). Select the **General** page and confirm the Device Name is "UV" for the VWD and the PDA detectors. For the ED and CD detectors: Open the properties of the DC and click on the **Detectors** tab. Highlight the detector name and click on **Configuration** button. Confirm that the Device Name is "CDet1" for CD detectors or "EDet1" for ED detectors. If both detectors are in the same Timebase, uncheck the

detector(s) that is not being qualified (only one detector can be qualified at a time). Click on the **Signals** tab. Confirm the signal name is “CD_1” for CD detectors or “ED_1” for ED detectors. Confirm that the signal factor for absorbance and conductivity detectors is 1.000. For integrated amperometry detectors confirm the signal factor is 0.001. If it is not, change the factor to the correct value for the qualification. Confirm the signal unit is μS for conductivity or nC integrated amperometry respectively. Confirm the signal unit is mAU for absorbance detectors. If it is not, change the unit to the correct value for the qualification. Exit the properties page.

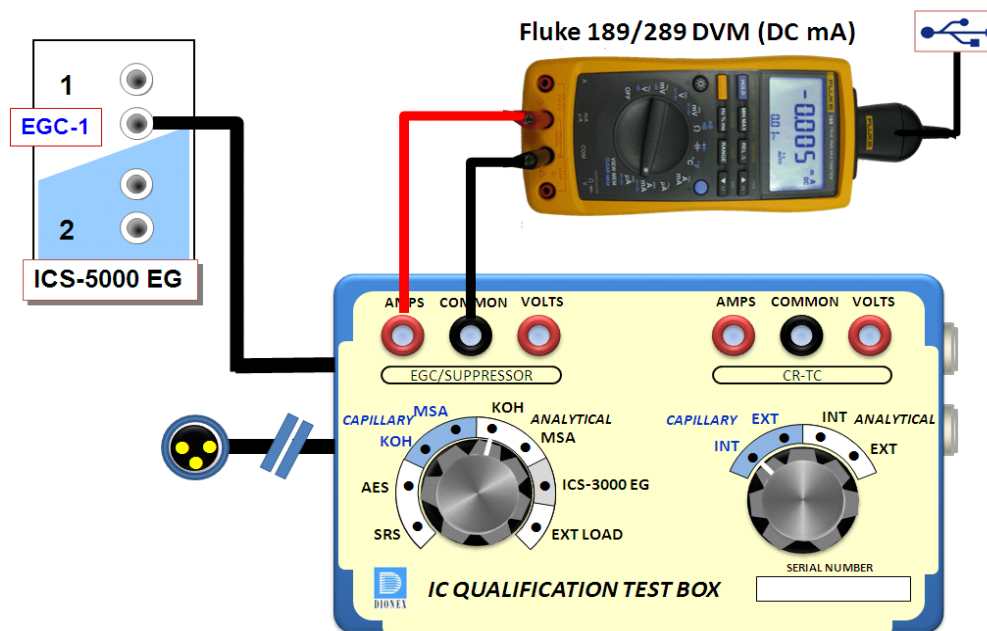


Figure 4. ICS-5000/ICS-5000+ EG Current Test Setup

7. If applicable ensure that the detector Output is in 'Offset' mode in the Analog Out Set-Up screen.
8. Make note of any changes and save the configuration.
9. All modules must have their serial number set in the moduleware. Qualification of modules without serial numbers is not supported.
10. Connect Eluents A, B, C and D to the pump (if present) and prime each eluent line individually for approximately 10 minutes each.
11. Use the backpressure coil to connect the pump outlet directly to port 2(P) on the injection valve. Bypass the column, suppressor, CR-TC, and CRD from the system leaving the gradient mixer in-line; if an Eluent Generator Cartridge is installed, bypass all plumbing to the EGC and connect the pump directly to the injection valve.

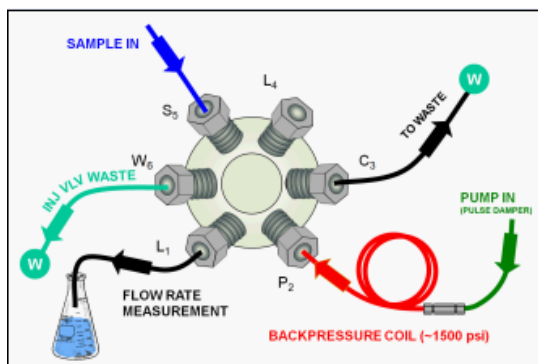


Figure 5. Plumbing diagram, initial equilibration step

12. ICS-5000/ICS-5000⁺ DC COLUMN TC: Insert the thermometer probe into the 2nd tubing slot from the bottom of the DC so that the LCD display sits out the side of the column TC. The LCD display of the thermometer should be visible and display the current Column TC temperature.
13. ICS-5000/ICS-5000⁺ DC COMPARTMENT TC: Insert the thermometer probe into top tubing slot on the DC so that the LCD display sits out the side of the compartment. The LCD display of the thermometer should be visible and display the current Compartment TC temperature.
14. ICS-5000/ICS-5000⁺ TC: Insert the thermometer probe into the side tubing slot on the TC so that the LCD display sits out the side of the TC. The LCD display of the thermometer should be visible and display the current TC temperature.
15. Thoroughly rinse the autosampler and/or manual injector fluidics with water. This can be accomplished on the AS and AS-AP Autosampler by flushing the sample syringe six times the sample syringe volume and on the AS-DV by running two or three 5.0 mL rinse vials
16. AS-AP or AS: For ICS-5000/ICS-5000⁺ systems using an AS-AP or AS Autosampler, install the 100 μ L sample loop on the injection valve. Loop volume must be set in Instrument Configuration for the AS-AP.

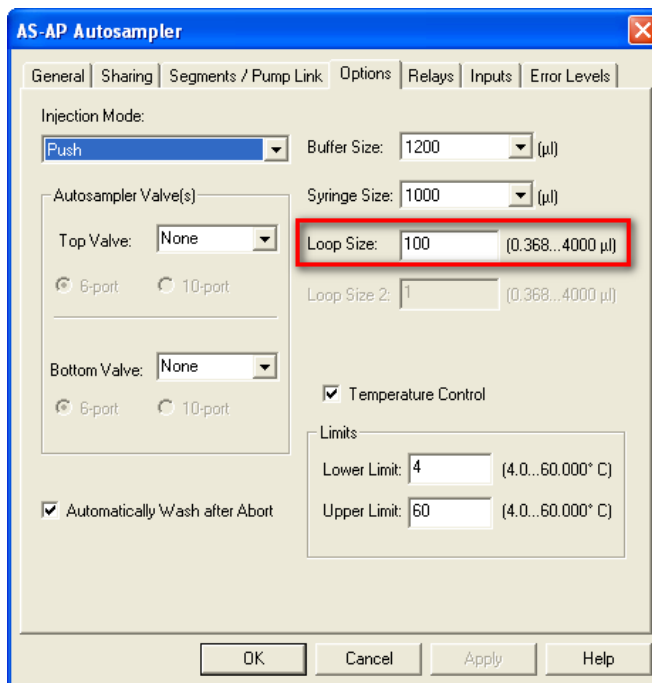


Figure 6. AS-AP Instrument Configuration

From the front panel of the AS, press the **Main Menu** button, select **Module Set-Up** and then **Plumbing Configuration**. Ensure that the sample loop size is set to 100 µL in the AS front control panel.

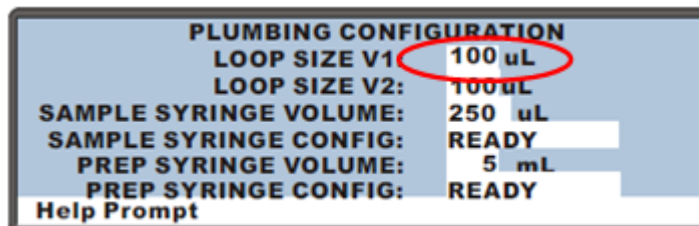


Figure 7. AS Autosampler LCD Control Panel Display

Ensure that the Loop SIZE V1 volume is changed to 100 µL

17. AS-DV or Manual Injector: For systems using an AS-DV Autosampler or manual injector, install the 25 µL sample loop on the injection valve.

Note: All programs using the AS-DV are designed to run with the 5.0 mL vials (*i.e.*, trigger relay at -2.40 minutes). If 0.5 mL vials are to be used, programs will have to be manually modified to trigger the relay at -1.20 minutes).

If using 5.0 mL vials, 1 Nitrate standards kit (P/N 060254) is needed to complete the OQ or PQ.

18. Select 100 % of Eluent A (water) and a flow rate of 1.00 mL/min. Turn the pump flow on and let the system equilibrate for a minimum of 15 minutes.

NEW TUBING: When running a qualification on a system with new tubing, such as a new backpressure coil, new sample loop or a complete new system, it is recommended to rinse the tubing with carbonate eluent before proceeding with the qualification. This can be achieved by doing two or three injections with 0.5 M Sodium Carbonate solution while running Deionized water at 1.00 mL/min or by running 9 mM Sodium Carbonate as the eluent for 10 minutes before rinsing with Deionized water for 20 minutes. The latter is recommended for new systems being qualified for the first time.

To prepare 0.5 M sodium carbonate dissolve 0.53 g of sodium carbonate in water to make a total volume of 10 mL of solution. To prepare 9 mM sodium carbonate dissolve 0.955 g of sodium carbonate in water to make 1 L of solution.

When running a qualification on a system configured with electrochemical detection, it is recommended to rinse the system with 1.0 M hydroxide before proceeding with the qualification. To prepare 1.0 M hydroxide, prepare from a 50/50 w/w sodium hydroxide solution (never from hydroxide pellets), by pipetting 52 mL of sodium hydroxide in water to make 1 L of solution.

19. **CONDUCTIVITY OR ABSORBANCE:** For the AS-AP autosampler pipette the standards from the standards kit vials directly into 1.5 mL or 10.0 mL autosampler vials and place them in the position / order according to Table 13. For the AS-DV autosampler, pipette the standards from the standards kit vials directly into the autosampler vials and place them in the position / order according to Table 14. For AS, place the vials in the tray, as per Table 12. For the AS-DV Injector Precision and Carryover standards transfer the Injector Precision standards prepared in Section 4.2 into 10 autosampler vials and the Carryover standard into one autosampler vial. To qualify an IC system without an autosampler, use the table 14 for the injection order and inject the samples from the standards kit ampoules manually.
20. **ICS-5000/ICS-5000⁺ INTEGRATED AMPEROMETRY:** Cut the label off the 1.5 mL standards vials (included in the ICS-5000/ICS-5000⁺ ED qualification kit) and place the vial directly into autosampler in the position order according to the table below. Note that even though the main portion of the label has been cut off, the vial is still properly loaded. For the AS-AP (push mode only) or AS autosampler this will require the 1.5 mL AP-AP tray (P/N 074936) or AS autosampler tray (P/N 062481).
21. **ICS-5000/ICS-5000⁺ ELUENT GENERATOR:** Double click on the eluent generator to be qualified to view the properties page for the eluent generator. Select the **General** page and confirm the Device Name is "EluentGenerator". On the "cartridge" tab confirm that the eluent generator is linked to the pump to be qualified. Confirm that there is a serial number for a KOH or a MSA cartridge. If a serial number is not available for the KOH or MSA cartridge, contact Thermo Fisher Scientific Technical Support. Connect the EG test box to the EGC_1 or EGC_2 connection.

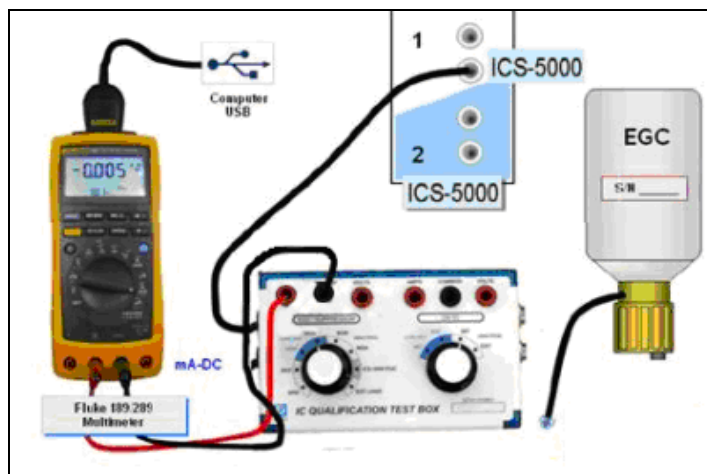


Figure 8. ICS-5000/ICS-5000⁺ EG Current Test Setup

22. For systems using an ICS-5000/ICS-5000⁺ EG:
 - a. Disconnect the Eluent Generator Cartridge cable from the Eluent Generator module at the connector.
 - b. If installed, disconnect the CR-TC cable from the CR-TC connector.
 - c. Turn on the ammeter or multimeter and set the display to DC mA. This test requires mA readings from 0.00 to at least 220 mA.
 - d. Connect the positive lead of the ammeter to the red post connector labeled “Amps” on the IC Qualification Test Box (P/N 072005)
 - e. Connect the negative lead of the ammeter to the black post connector labeled “COMMON” on the IC Qualification Test Box.

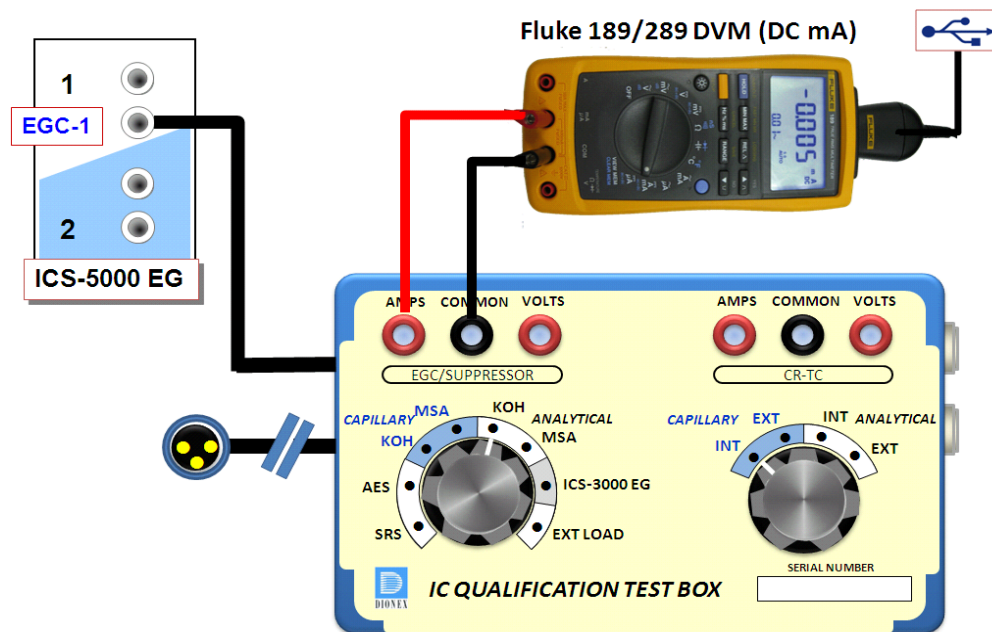


Figure 9. ICS-5000/ICS-5000⁺ EG Current Test Setup

- f. From the Console in the Chromeleon 7.1 software, go to the Navigation Pane and select the pump_EluGen_detector panel appropriate for the system being qualified. Go to Control and select the Instrument configured with the Eluent Generator to be qualified (click on the Default ePanel Tab)
- g. From the Eluent Generator section of the panel, always select KOH as the cartridge type. The panel will show previously configured cartridges even though the IC Qualification Test Box is plugged into the cartridge connector. Do not switch the test box settings during operation as this can damage the test box.

23. FLUKE 189/289 MULTIMETER:

- a. Install the Fluke 189/289 multimeter or equivalent into the Instrument to be qualified.
- b. The Chromeleon device driver for the Fluke 189/289 multimeter is included with Chromeleon 7.1
- c. Connect the Fluke 189/289 multimeter or equivalent to the IC Qualification Test Box (P/N 072005) as described in the **important notes** of this section.
- d. The Fluke 189/289 multimeter is only used for systems that will enable automated downloading of EG current qualification results into the validation reports.
- e. To correctly configure the Fluke 189/289 device driver:
 - i. Connect the Fluke USB/IR (Rev II or greater) cable to the computer USB port; Windows will automatically detect and install the USB driver.
 - ii. Verify through the Windows Hardware Device Manager for the actual COM port assignment.

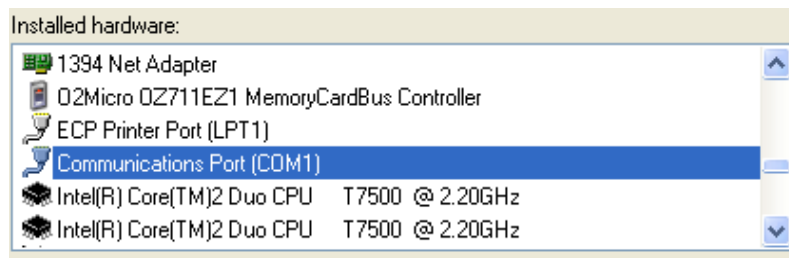


Figure 10. COM Port Assignment in the Windows Device Manager for the Fluke meter

- iii. Configure the Fluke 189 or 289 Chromeleon Device Driver in the Timebase, save the Instrument Configuration file and exit then shut down the Chromeleon Server.
 - iv. Restart the Chromeleon Server and open the Fluke 189/289 CM device driver.
 - v. Select the COM port from the COM drop down selection to match the COM number assigned in the Windows Hardware Device Manager above then save and exit from the Chromeleon Server.
- f. It is recommended that the Power off time out (PrOff) for the Fluke189/289 be set to a value greater than the time it will take to perform the Operational Qualification to avoid inadvertent shutdown of the meter which will terminate the OQ or PQ sequence.
- g. Ensure that the meter is set to record in 'DC mA' (instead of 'AC mA').
- h. See Fluke 189/289 operator's manual for instructions on these settings.

IMPORTANT NOTES:

- a. It is important that the Fluke 189/289 multimeter is installed AND connected before loading the OQ/PQ templates.
- b. Follow the instructions in the [Chromeleon 7.1](#) help topics for the installation of the Fluke 189/289 multimeter Windows USB driver and for the Chromeleon device driver for the Fluke 189/289 multimeter. The Fluke 289 defaults to AC mA and the Fluke 189 defaults to DC mA.
- c. Follow the Fluke 189/289 multimeter instructions for adjusting the default 'PrOFF' setting in the meter. It is recommended to adjust this parameter to a longer time (e.g., several hours or more) to avoid having the meter auto power off during your OQ/PQ. In the event that the meter is powered off during the qualification testing the queue sequences will be aborted.

Table 12. AS Autosampler Sample Vial Order

Conductivity Detector with AS	Absorbance or Photodiode Array Detector with AS	ICS-5000/ICS-5000 ⁺ Integrated Amperometry Detector with AS	Vial Tray Position for AS
Water	Water	Water	1
5 ppm Nitrate Std			2
10 ppm Nitrate Std			3
25 ppm Nitrate Std			4
50 ppm Nitrate Std			5
100 ppm Nitrate Std			6
1000 ppm Nitrate Std			7
Empty Vial*			8
Empty Vial*			9
Empty Vial*			10
	15 ppm Caffeine Std		11
	40 ppm Caffeine Std		12
	60 ppm Caffeine Std		13
	80 ppm Caffeine Std		14
	100 ppm Caffeine Std		15
	1000 ppm Caffeine Std		16
	Empty Vial*		17
	Empty Vial*		18
	Empty Vial*		19
			20
		1 µM Threonine Std	21
		2 µM Threonine Std	22
		4 µM Threonine Std	23
		8 µM Threonine Std	24
		10 µM Threonine Std	25
		10 µM Threonine Std	26

* Vials only need to be present if the detector listed at the top of the table is to be qualified.

* Users need to reconnect the detectors when using the absorbance detectors.

* Don't forget to add the water vial in position 1 for the ED qualification test.

Table 13. AS-AP Autosampler Sample Order

Conductivity Detector with AS-AP Push/Pull Mode	Absorbance Detector with AS-AP Push/Pull Mode	Electrochemical Detector with AS-AP Push Mode	Vial Tray Position
Water	Water		BA1
5 ppm Nitrate Std	15 ppm Caffeine Std		BA2
10 ppm Nitrate Std	40 ppm Caffeine Std		BA3
25 ppm Nitrate Std	60 ppm Caffeine Std		BA4
50 ppm Nitrate Std	80 ppm Caffeine Std		BA5
100 ppm Nitrate Std	100 ppm Caffeine Std		BB1
1000 ppm Nitrate Std	1000 ppm Caffeine Std		BB2
		Water	RA1
		1 µM Threonine Std	RA2
		2 µM Threonine Std	RA3
		4 µM Threonine Std	RA4
		8 µM Threonine Std	RA5
		10 µM Threonine Std	RA6
		10 µM Threonine Std	RA7

Note: AS-AP pull mode requires the use of 10 mL vials. Qualification with ED detection is not supported with AS-AP in pull mode.

Note: To run sequences with 1.5 mL vials, it is important to set the puncture offset to 3 mm in the commands before you start. The puncture offset commands allows the AS-AP to realign the x-axis of the 1.5m/10 mL vial setting.

Table 14. AS-DV Autosampler or Manual Injection Sample Order (Conductivity or Absorbance Only)

Qualification	Conductivity Detector with AS-DV	Sample Order on AS-DV	Absorbance or Photodiode Array Detector with AS-DV	Sample Order on AS-DV
Warm Up	Water	Vial 1	Water	None
Injector Precision	50 ppm Nitrate Std*	Vial 2	40 ppm Caffeine Std*	Vial 20
	50 ppm Nitrate Std*	Vial 2	40 ppm Caffeine Std*	Vial 21
	50 ppm Nitrate Std*	Vial 2	40 ppm Caffeine Std*	Vial 22
	50 ppm Nitrate Std*	Vial 3	40 ppm Caffeine Std*	Vial 23
	50 ppm Nitrate Std*	Vial 3	40 ppm Caffeine Std*	Vial 24
	50 ppm Nitrate Std*	Vial 3	40 ppm Caffeine Std*	Vial 25
	50 ppm Nitrate Std*	Vial 4	40 ppm Caffeine Std*	Vial 26
	50 ppm Nitrate Std*	Vial 4	40 ppm Caffeine Std*	Vial 27
	50 ppm Nitrate Std*	Vial 4	40 ppm Caffeine Std*	Vial 28
	50 ppm Nitrate Std*	Vial 5	40 ppm Caffeine Std*	Vial 29
Carryover	1000 ppm Nitrate Std	Vial 12	80 ppm Caffeine Std*	Vial 30
	Water	Vial 13	Water	Vial 31
	Water	Vial 14	Water	Vial 32
Detector Linearity	5 ppm Nitrate Std	Vial 15	15 ppm Caffeine Std	Vial 33
	10 ppm Nitrate Std	Vial 16	40 ppm Caffeine Std	Vial 34
	25 ppm Nitrate Std	Vial 17	60 ppm Caffeine Std	Vial 35
	50 ppm Nitrate Std	Vial 18	80 ppm Caffeine Std	Vial 36
	100 ppm Nitrate Std	Vial 19	100 ppm Caffeine Std	Vial 37

* AS-DV users: Do not use the standard from the standards kit for the Injector Precision and Absorbance Carryover samples; instead, use the standards prepared in Section 4.3 of this manual.

* AS-DV users: ICS-5000/ICS-5000⁺ Integrated Amperometry is not supported.

4.6 OQ/PQ Sequence Installation

For performing OQ and PQ checks in Chromeleon 7.1, it is not required to create and copy the sequence templates from a Chromeleon CD. An Instrument Qualification Wizard automatically performs these steps. The wizard creates the sequences to be run. No instrument-specific sequence templates are created.

If using an AS autosampler configured for sequential injections, manually put the diverter valve in the correct position for the Instrument.

Tip: Please note that Chromeleon 7 terminology is different from the terminology used in Chromeleon 6.80. For details, refer to the 'Glossary - Chromeleon 7,' which is available in the Documents folder of your Chromeleon 7 installation. Make sure the user takes control of the panel before starting the instrument qualification.

Go to Tools → Instrument Qualification to open Instrument Qualification Wizard. For this step, Chromeleon 7.1 must display the instrument pane with the system to be tested selected (Figure 11) otherwise the error “Instrument not connected” will occur.

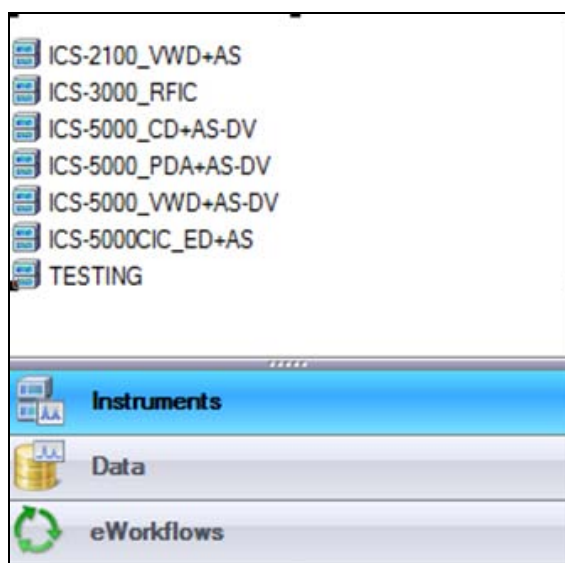


Figure 11. Instrument Pane for Chromeleon 7.1

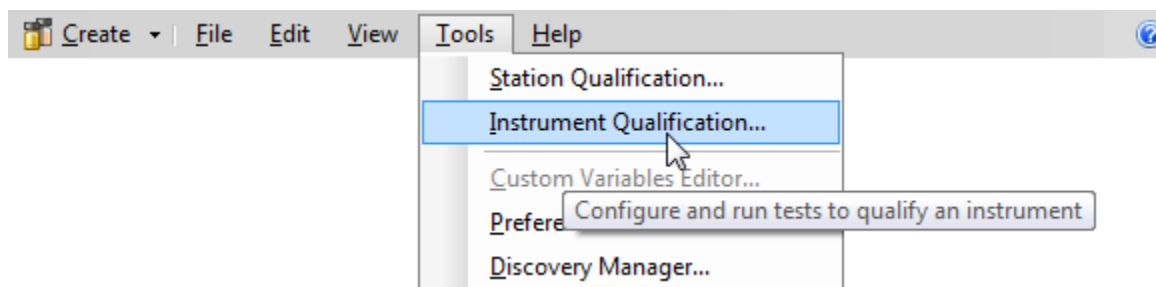


Figure 12. Starting the Instrument Qualification Wizard for Chromeleon 7.1

Select the qualification type: Installation (qualification of the installation), Operational (qualification in the working environment), or Performance Qualification (qualification during routine operation)

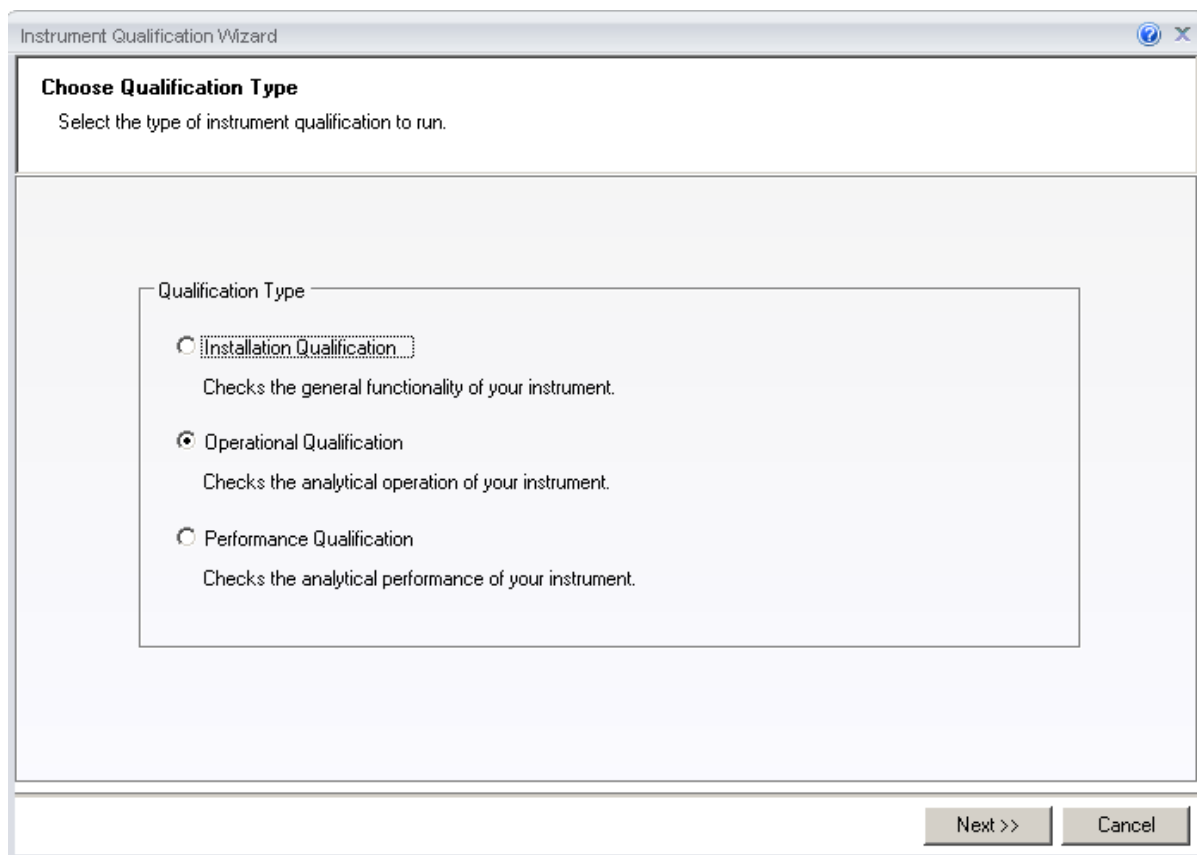


Figure 13. Selecting the qualification type for Chromeleon 7.1

Select the instrument to qualify.

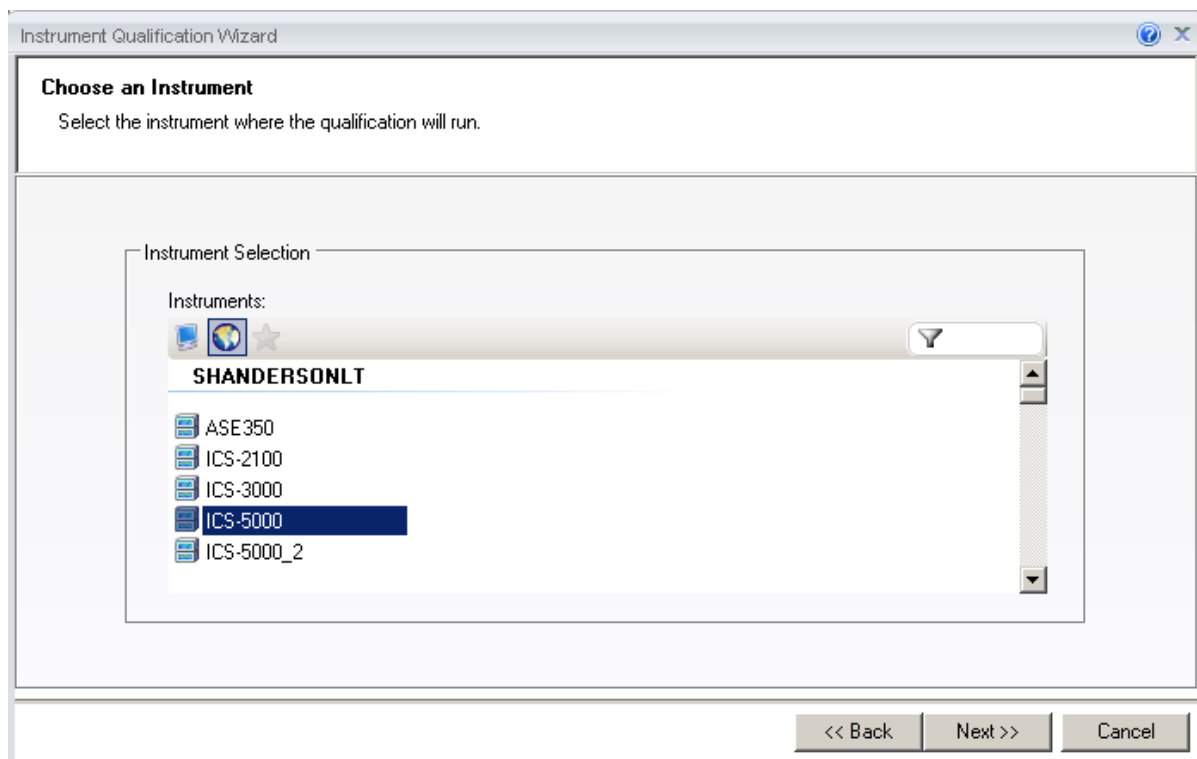
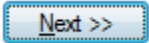


Figure 14. Selecting the instrument

Click  to connect the selected instrument to Chromeleon.

A list of sequences (tests) is displayed. The list is adapted to the instrument configuration of the selected instrument as defined in the Chromeleon Instrument Configuration Manager.

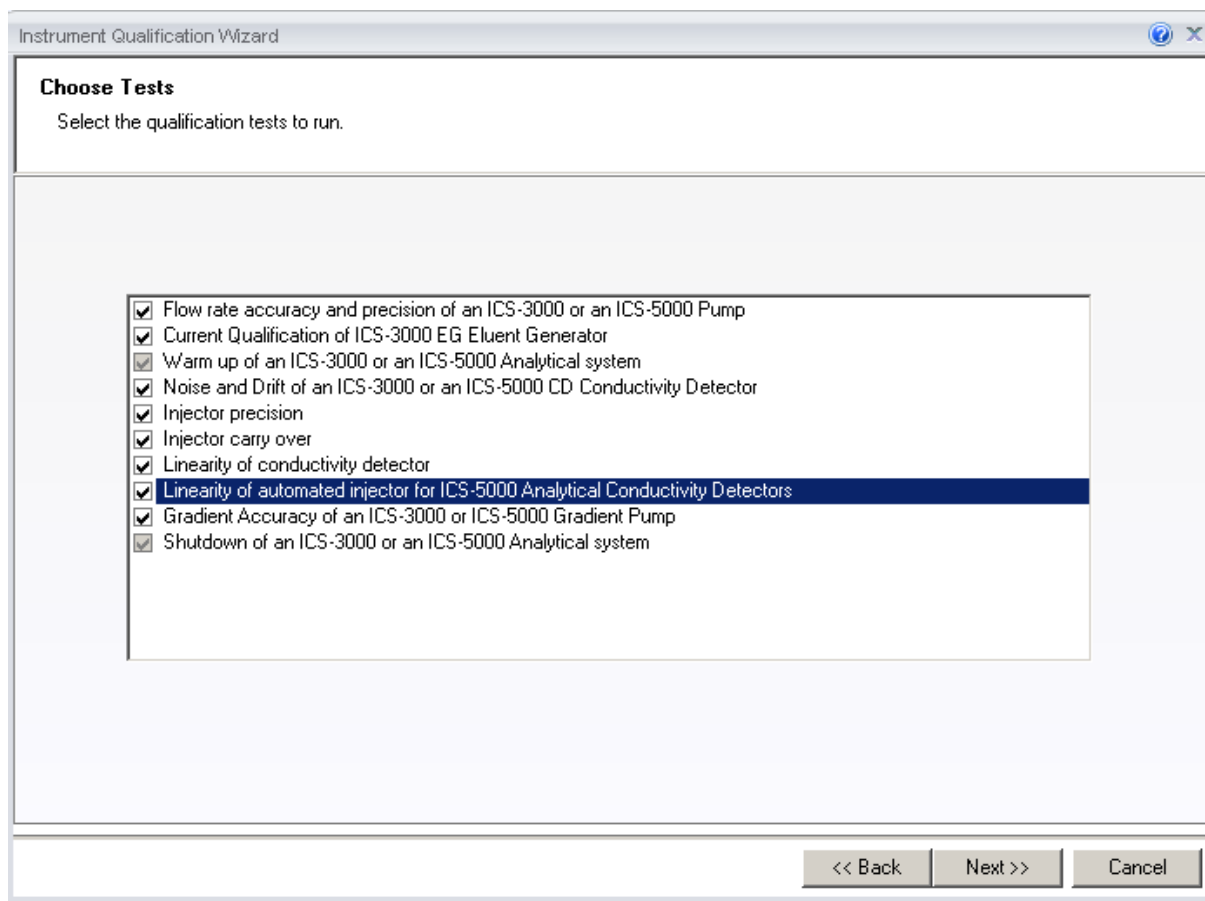


Figure 15. List of sequences for the selected instrument

Select the sequences that needed for the tests to be performed. Mandatory tests, such as "**Warm up**", are shown in the list, but the selection cannot be changed.

On the last wizard page, select a unique name under which the OQ and/or PQ sequence directory for this instrument is saved.

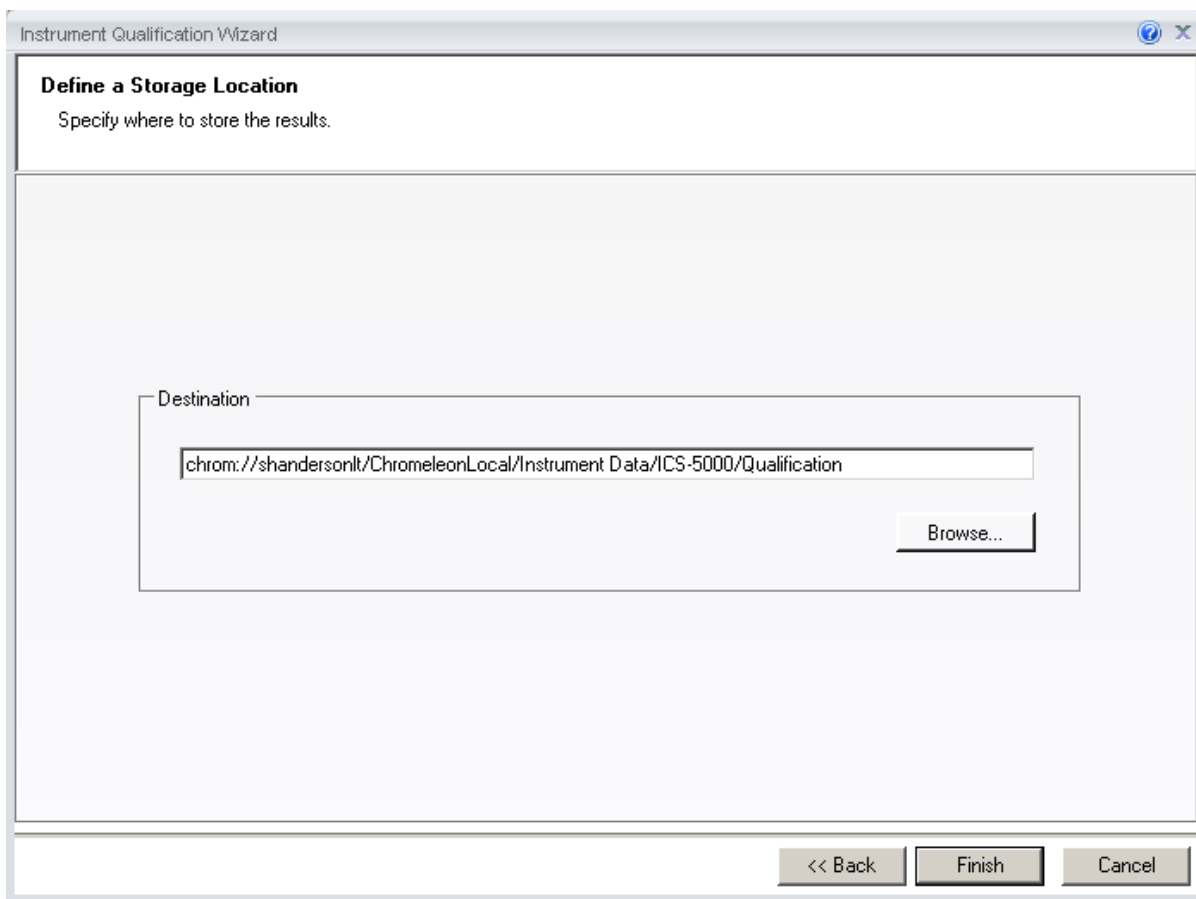


Figure 16. Selecting the storage location

4.7 OQ/PQ Sequence Execution

When the qualification wizard has been completed, the selected sequences are created and are automatically added to the instrument queue. A progress window shows which steps have been performed:

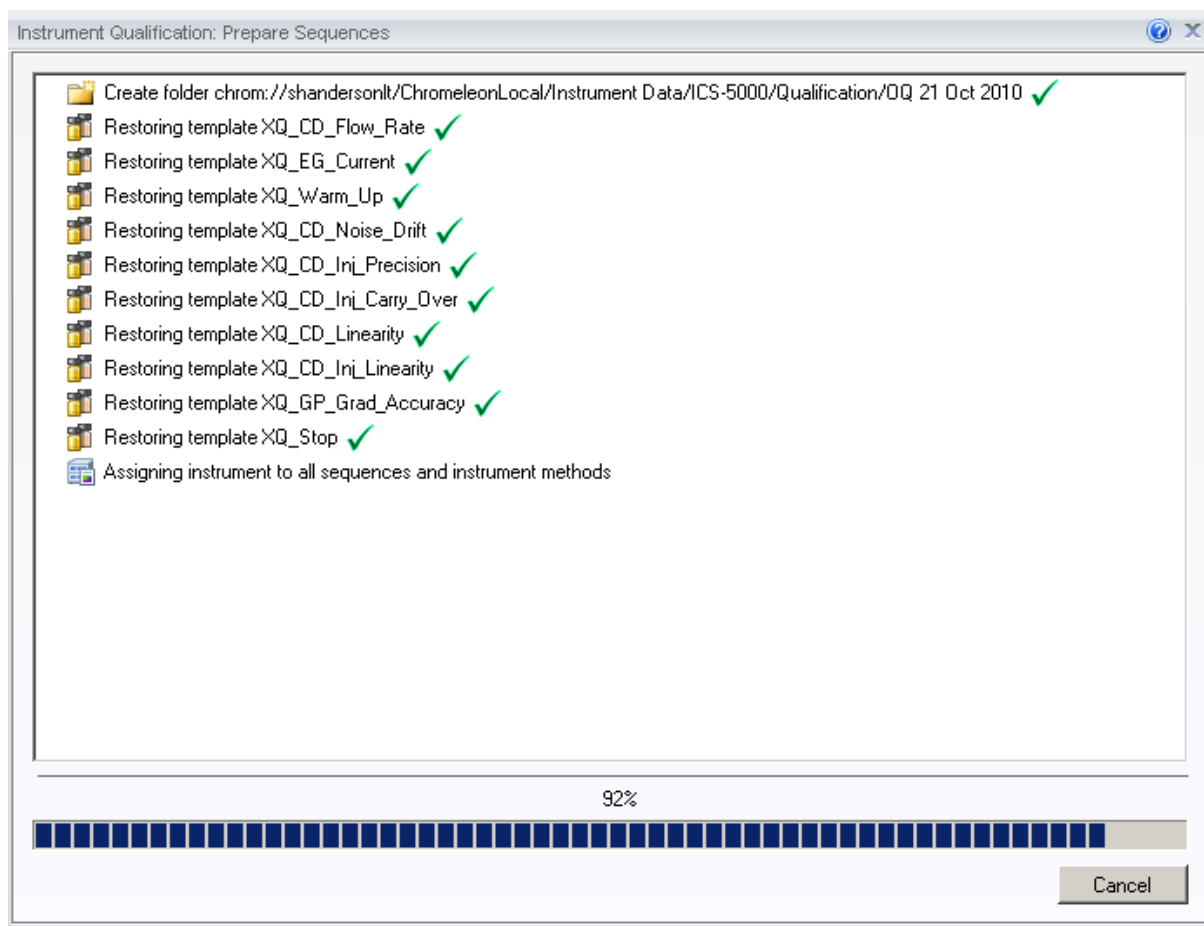


Figure 17. Progress during sequence creation

The Instrument View dialog box will open, displaying the Queue tab. As soon as the queue is started, Chromeleon runs the sequences. The wizard should copy the sequences to the destination selected then auto-populate the instrument queue. If the error “Instrument not connected” appears, make certain that the Chromeleon 7.1 must be displaying the instrument pane with the system to be tested.

The OQ/PQ qualification tests are performed in the following order:

- Flow Rate Test
- Eluent Generator Current Test (only if an EG is present)
- Warm Up
- Wavelength Accuracy
- Detector Noise and Drift Test

- f. Injector Precision Test
- g. Carryover Test
- h. Detector Linearity Test
- i. Injector Linearity Test (only if an AS or AS-AP is present)
- j. Sample Preparation Precision Test (only if an AS is present with the Sample Prep Option and the 10 mL Tray Type installed with the conductivity or the absorbance detector)
- k. Gradient Accuracy Test (only if an analytical gradient pump is present)
- l. Column TC and/or Compartment TC
- m. Sample Temperature Accuracy of Autosamplers (only for AS-AP Autosamplers)

Note: Test 'a' and 'l' is done manually and does not require a sequence template. The Instrument Method will pause before the Eluent Generator Current Qualification Test and the Flow-Rate Accuracy and Precision Test as these tests require user intervention.

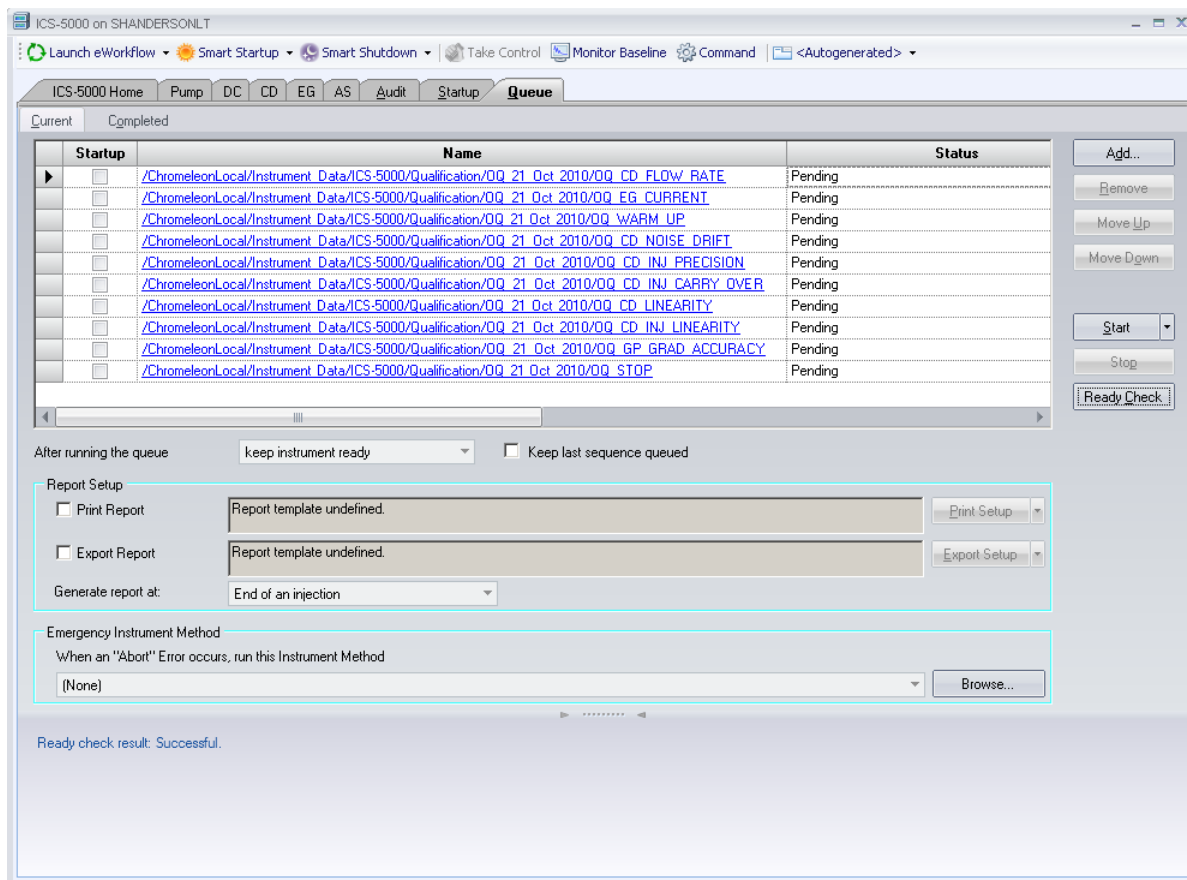


Figure 18. Sequence queue for Chromeleon 7.1

4.8 Evaluating the Test Sequences

The qualification sequences are saved under the path that was selected in the wizard. Each sequence also includes a separate report template.

To edit the report, open the report and remove the protection (if any) from the SPECIFICATION sheet. Enter the following information:

Name of the customer and name of the tester

Sample information such as queue number, expiration date, and actual concentration of the standard.

Name of the item that is used to generate the backpressure [default: capillary (L: 15 m; ID: 0.18 mm)]

Do not change any of the other report sheets. The report contains many references between data sheets. If you insert or delete lines and columns, these references will be lost and the calculations will be wrong.

To make sure that Chromeleon reads and processes the data in the report correctly, always print the report from the Chromeleon Console.

In the Data category, right-click the sequence for which you want to print the report, and then click **"Print Report"**.

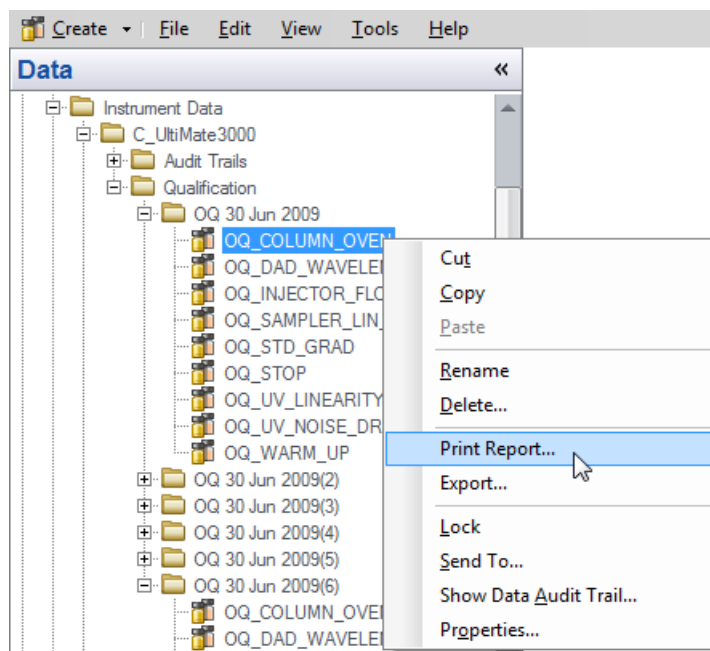


Figure 19. Selecting Print

Note: For ICS-5000/ICS-5000[±] Integrated Amperometry System: Before executing the sequences, first turn on the pump flow and the cell current. Click on the “**Default Panel Tabset**”. From the pump tab, start the pump flowing 50 mM NaOH at 0.25 mL/min. Click on the **EC detector tab**, Select Cell mode of “IntAmp”, then click on the **Waveform** button, use the drop down menu to choose the waveform **Amino Acids (pH/Ag/AgCl Reference)**, then click on the **Exit** button. Turn the cell voltage to **On**. PdH reference electrode is not supported in OQ/PQ configurations.

Note: If an Eluent Generator is configured on the system, the following message may occur at the Ready Check: “Flow rate not specified at time 0.0, using a flow setting 1.00.” This refers to the Eluent Generator flow rate, not the pump flow rate. It is not necessary to take any action since the Eluent Generator is not used for this part of the qualification. Proceed with starting the queue.

Note: For systems with a shared autosampler there might be a number of *informational* messages indicating that “Exclusive Access” has not been acquired or released. This is normal and occurs because the qualification does not release the autosampler until the qualification is complete.

Repeating Individual Tests

It may be necessary to repeat individual tests. If so, review Section 6 where problems are described according to which individual tests might fail. According to Good Laboratory Practice (GLP), all tests following the one that failed must be repeated. The entire OQ/PQ qualification is designed such that almost of the performance tests require that the previous one to have passed successfully.

For example, if the test regarding the linearity of a CD detector fails, the results regarding the linearity of the injector are questionable as the linearity of the detector is a prerequisite for testing the injector linearity.

Table 15. Sequence File Names and description

Sequence Name	Check
XQ_Flow_Rate	Pump flow-rate accuracy and precision tests for ICS-3000 or ICS-5000/ICS-5000 ⁺ pumps
XQ_EG_Current	Current qualification of an EG Eluent Generator
XQ_EG_Dual_Current	Current qualification of an Dual EG configuration (ICS-5000/ICS-5000 ⁺ with both EGC configured on one Instrument)
XQ_CD_Noise_Drift	Noise and drift test for conductivity detectors
XQ_DC_Amp_Noise_Drift	Noise and drift test for DC amperometry detectors
XQ_Int_Amp_Noise_Drift	Noise and drift test for integrated amperometry detectors
XQ_PDA_Noise_Drift	Noise and drift test for photodiode array detectors
XQ_UV_Vis_Noise_Drift	Noise and drift test for absorbance detectors (excluding PDA detectors)
XQ_Wavelength_Accuracy	Wavelength accuracy test for the VWD and PDA absorbance detectors
XQ_Abs_Inj_Precision	Injector precision test for systems with an absorbance detector
XQ_Abs_Inj_Carryover	Injector carryover tests for systems with an absorbance detector
XQ_CD_Inj_Precision	Injector precision test for systems with an conductivity detector
XQ_CD_Inj_Carry_Over	Injector carryover test for systems with an conductivity detector
XQ_ED_Warm_Up	Warm-up sequence for ICS-5000/ICS-5000 ⁺ systems with ED detector
XQ_ED_Noise_Drift	Noise and drift test for ICS-5000/ICS-5000 ⁺ amperometry detectors (ED in Integrated Amperometry mode)
XQ_ED_Inj_Precision	Injector precision for ICS-5000/ICS-5000 ⁺ systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Carryover	Injector carryover for ICS-5000/ICS-5000 ⁺ systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Linearity	Detector Linearity for ICS-5000/ICS-5000 ⁺ systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Inj_Linearity	Injector linearity for ICS-5000/ICS-5000 ⁺ systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Stop	The sequence turns off the pump and cell for ICS-5000/ICS-5000 ⁺ systems with amperometry detector (ED in Integrated Amperometry mode).

XQ_Abs_Linearity	Detector linearity test for absorbance detectors
XQ_CD_Linearity	Detector linearity test for conductivity detectors
XQ_Abs_Inj_Linearity	Injector linearity test for systems with an absorbance detector
XQ_CD_Inj_Linearity	Injector linearity test for systems with an detector
XQ_Abs_Samp_Prep	Injector sample preparation precision test for systems with an absorbance detector
XQ_CD_Samp_Prep	Injector sample preparation precision test for systems with an conductivity detector
XQ_GP_Grad_Accuracy	Gradient accuracy and noise test for systems with a gradient pump
XQ_CD_Noise_Drift	Noise and drift test for conductivity detectors
XQ_DC_Amp_Noise_Drift	Noise and drift test for DC amperometry detectors
XQ_Int_Amp_Noise_Drift	Noise and drift test for integrated amperometry detectors
XQ_PDA_Noise_Drift	Noise and drift test for photodiode array detectors
XQ_UV_Vis_Noise_Drift	Noise and drift test for absorbance detectors (excluding PDA detectors)
XQ_Stop	The sequence slows down the flow rate to 0.25 mL/min to conserve eluent for overnight runs

5 Performing the Qualification

5.1 OQ/PQ Test Order

The OQ/PQ qualification tests are performed in the following order:

1. Flow Rate Accuracy and Precision Qualification
2. Eluent Generator Current Qualification Test
3. Warm Up
4. Wavelength Accuracy Test (only for VWD, and PDA Absorbance Detectors)
5. Detector Noise and Drift Test
6. Injector Precision Test
7. Carryover Test
8. Detector Linearity Test
9. Injector Linearity Test (only for AS-AP and AS Autosamplers)
10. Sample Preparation Precision Test (only for AS Autosamplers with the Sample Prep Option and 10 mL Tray Type installed with conductivity or absorbance detectors)
11. Gradient Accuracy Test (only for gradient pumps)
12. Column TC and/or Compartment TC
13. Sample Temperature Accuracy of Autosamplers (only for AS-AP Autosamplers)

Table 16. OQ/PQ Test Timetable

Test	Method	Material Required	Equilibration Time	Approx. Run Time
Flow Rate Accuracy and Precision All pump models	Sequence	Eluent (water) Backpressure loop	15 min	52 min
Wavelength Accuracy ICS Series VWD and PDA	Sequence	Eluent (water)	Lamps – 2 h 10 min w/pump	6 min
Conductivity Noise / Drift	Sequence	Water at 1.0 mL/min	5 min	20 min
Int. Amperometry Noise / Drift	Sequence	Eluent (50 mM OH)	5 min	20 min
AS-DV, Manual Injector Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity	Sequence	Eluent (water) Standards Backpressure loop	10 min	1 hr, 10 min
AS-AP, AS Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity Injector Linearity	Sequence	Eluent (water) Standards Backpressure loop	10 min	2 hr, 10 min
AS-AP with Temp. Control Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity Injector Linearity Sample Temp. Accuracy	Sequence	Eluent (water) Standards Backpressure loop Type K temperature sensor Calibrated Digital Thermometer	10 min	2 hr 55 min
AS with Sample Prep Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity Injector Linearity Sample Prep Precision	Sequence	Eluent (water) Standards Backpressure loop	10 min	2 hr, 40 min
Eluent Generator Current Qualification	Sequence	Test cell Multimeter	N/A	16 min
AS-AP pH Conductivity Accuracy Test	Sequence	Calibration pH Standards	N/A	1 hr
EG Gradient Accuracy w/ CD or ED	Sequence	Eluents Backpressure loop	10 min	1 hr

5.2 Starting the Sequences



For AS and AS-AP Autosamplers with the Sequential Injection Option: Before starting any sequences, first verify that none of the instruments configured on the computer have “Acquired Exclusive Access” to the autosampler, or column TC, or compartment TC. If this is the case release the “Exclusive Access” command before proceeding. Once the qualification sequences begin, the system being qualified will gain and retain exclusive access to the autosampler, column TC and compartment TC until the “Stop” sequence is run and “Exclusive Access” is released. “Exclusive Access” can be released by going to the command function (F8), choosing the module from the list, and executing the “ReleaseExclusiveAccess” command.

5.2.1 Flow Rate Accuracy and Precision Qualification

The Flow Rate Accuracy and Precision test is designed to qualify the flow rate accuracy and precision of all pumps. A 6-port valve is required to run the IC flow rate test.

1. Ensure the pump is disconnected from the Chromeleon software. The system should be in hold mode and a message box displaying “Plumb pump for Pump Flow Rate Accuracy and Precision Test, press **OK** to continue” should be visible.



Plumb pump for Pump Flow-rate Accuracy and Precision Test, press OK to continue.

2. Stop the pump from the front control panel of the module if it is not already stopped.
3. Locate the backpressure coil. Use a coil made of 37" of red PEEK tubing (049715) for the analytical system.
4. Connect one end of the backpressure coil directly to the outlet of the pump, after gradient mixers if applicable and the other to port 2 (P) of the injector valve.
5. Prepare two lengths of black tubing, 0.010" (0.25 mm) ID, of equal length, approximately 12" (40 cm). Connect one of these lines to port 1 (L) of the injector valve. The tubing connected to port 1 (L) of the injector valve will be the eluent line that goes to the “sample beaker.”
6. Connect the other line to port 3 (C) of the injector valve. The tubing connected to port 3 (C) of the injector valve will be the waste line.
7. Run the waste line to a waste beaker and the eluent line to a sample beaker. The plumbing to the valve should look like the figure below.

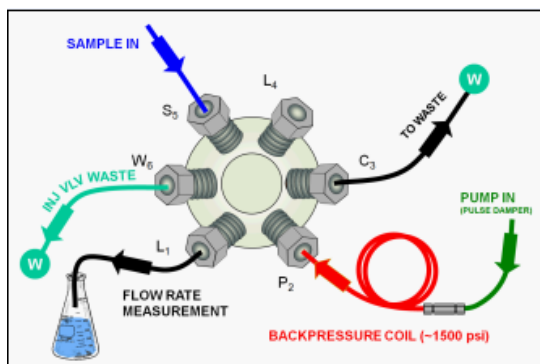


Figure 20. Plumbing schematic for Pump Flow Rate Test

8. Click **OK** on the Chromeleon software. The system should reconnect to the module and/or resume the program in run mode.
9. The program will equilibrate the pump for 15 minutes before starting the test. During this time eluent flow should pass through the waste line. Ensure that sufficient backpressure is observed during this equilibration time.
10. At time 0, the injector will switch to the eluent line.
11. At time 2.00, the injector will switch back to the waste line and the Chromeleon software will ask you to "Please weigh the eluent container. Enter results under segment 0. Press OK to continue." The program will be in hold mode and will wait until OK is pressed before continuing. Weigh the eluent container, but do not zero the balance used for weighing. Record the weight as segment 0. Click **OK** to continue.

Please weigh the eluent container. Enter results under segment 0. Press OK to continue.

OK

Note: The report is available in the flow rate sheet of the Report Designer and can be used for manual data collection and data calculations.

12. The Chromeleon software will switch the injector back to the eluent line for exactly 5 minutes. After this time the injector will switch to the waste line and a message "Please weigh the eluent container. Enter results under segment 1. Press OK to continue" will be displayed. The program will be in hold mode and will wait until OK is pressed before continuing. Weigh the eluent container, but do not zero the balance used for weighing. Record the weight as segment 1. Click **OK** to continue.
13. Repeat 12 for segments 2 through 5.

After the sequence is finished open the Water_FR sample from the Console. Select the Flow Rate sheet at the bottom of the page and enter the Segment values and room temperature into the sheet. Save the worksheet by clicking on the Chromeleon Icon on the top left of the User Interface and select save. A written or balance printout of the weights should be signed and dated to accompany data entered on the sheet

14.

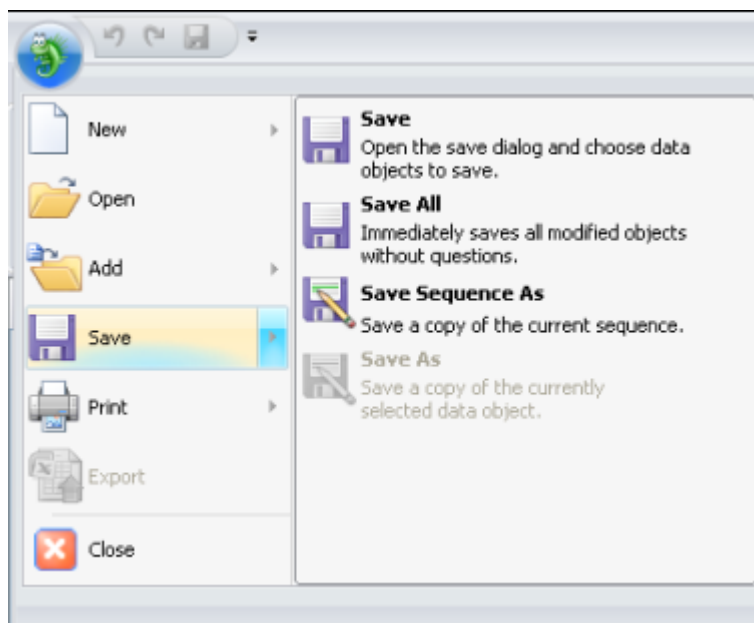


Figure 21. Selecting Save

15. Print the Flow Rate Accuracy and Precision report by clicking on the Chromeleon Icon on the top left of the User Interface and select print. Choose a printer and press OK to begin printing.

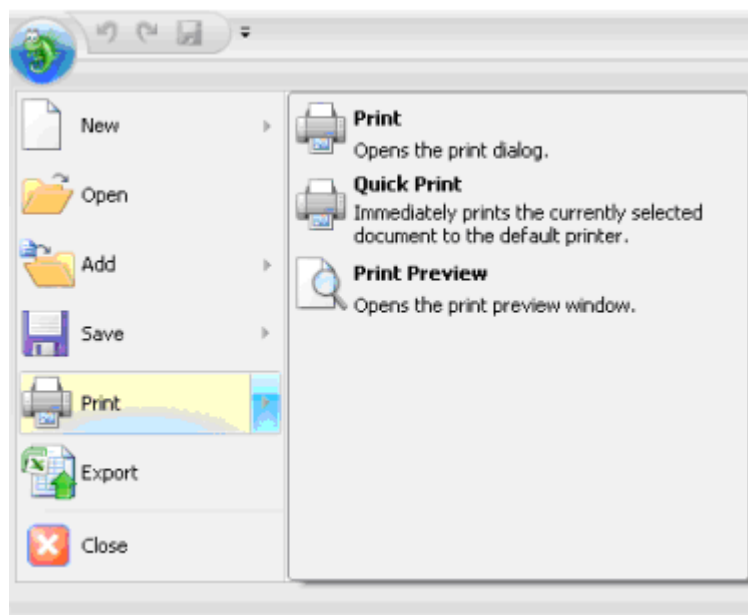


Figure 22. Selecting Print

16. If any part of the test does not pass, make the appropriate repair or calibrations and rerun the test. Refer to Section 5 for troubleshooting assistance.

17. Plumb the system for the remainder of the qualification.

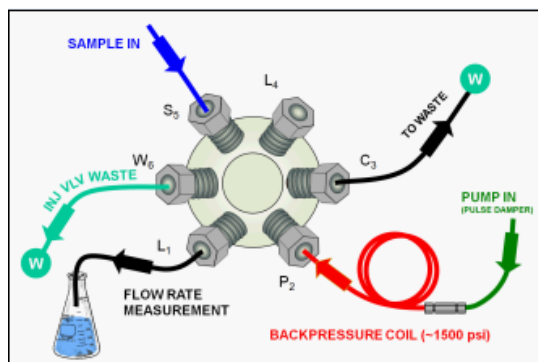


Figure 23. Plumbing schematic for OQ/PQ performance test after pump flow rate test

18. Ensure the pump is disconnected from the Chromeleon software. The system should be in hold mode and a message box displaying “Plumb the system for the remainder of the qualification tests, press **OK** to continue” should be visible.
 - a. ICS-5000/ICS-5000[±] Conductivity or Absorbance: Connect the backpressure tubing to the heat exchanger outlet line. Attach the loop end to the inlet of the conductivity or absorbance cell, thus bypassing all columns and suppressors.

It is important to connect the Backpressure Loop assembly upstream from the loop or unusual peak shapes may occur.

Note: A piece of 0.003” tubing may be used to provide approximately 1500 psi (± 100 psi) backpressure and sufficient delay.

- b. ICS-5000/ICS-5000[±] Integrated Amperometry: Calibrate the reference electrode (see operators manual), then assemble the ED cell with an AAA certified disposable gold working electrode (P/N 060082) and mount the ED cell onto the ED detector. Connect the two cell leads (yellow and blue) to the appropriate ports on the detector. Connect a 24-inch piece of 0.003” PEEK tubing to port three (C) of the injection valve, connect the other end to the inlet of a CarboPac PA1 guard (2-mm) column. To the outlet of the guard column connect a small length (approx 15-cm) of 0.010” tubing. Attach the loop end to the inlet of the Integrated Amperometry Cell.

Note: When loading the Threonine standards in the AS-AP or AS autosampler, it may be necessary to cut the label off the Threonine vial for proper fit into the AS or AS-AP sample tray. For this reason, the concentration has been printed on the part of the label that will remain on the vial.

5.2.2 Eluent Generator Current Qualification

The Eluent Generator Current Qualification test ICS-5000/ICS-5000⁺ EG (single or dual) needs to be configured with a KOH cartridge.

When using a FLUKE 189/289 multimeter, the measured current is automatically entered into the **EG_CURRENT, EG_DUAL_CURRENT** sheet.

1. **Resume** the Chromeleon 7 Queue by clicking **OK**. This sequence is an Eluent Generator step gradient. Each step is held until the user clicks OK to continue. There may be a delay of up to

0.4 minutes between when the Eluent Generator program steps to the next concentration and when the ammeter or multimeter registers the new milliamp reading. Record the milliamp reading for each step on a note pad. These values will be entered into the **EG_CURRENT**, **EG_DUAL_CURRENT** or **EluGen Current Qualification** sheet after the run is complete. The run time is 11.0 minutes plus pause time.

Note: The template of this report is stored in the data vault of the respective sequence. Please refer to Chromeleon 7 Quick Start Guide on how to access report templates.

Template name: OQ_PQ_Modular_Validation

2. When the run is complete, open the report file by double clicking on the Water_EG sample of the XQ_EG_Current sequence. Locate the row of sheet names at the bottom of the report and click on the **EG_CURRENT**, **EG_DUAL_CURRENT** or **EluGen Current Qualification** sheet.
3. Enter the milliamp readings that were recorded in step 1 for each concentration value in the **EG_CURRENT**, **EG_DUAL_CURRENT** or **EluGen Current Qualification** sheet. The deviation and pass/fail results will be calculated automatically.
4. Print the EG Current Qualification report by clicking on the Chromeleon Icon on the top left of the User Interface and select print. Choose a printer and press OK to begin printing.

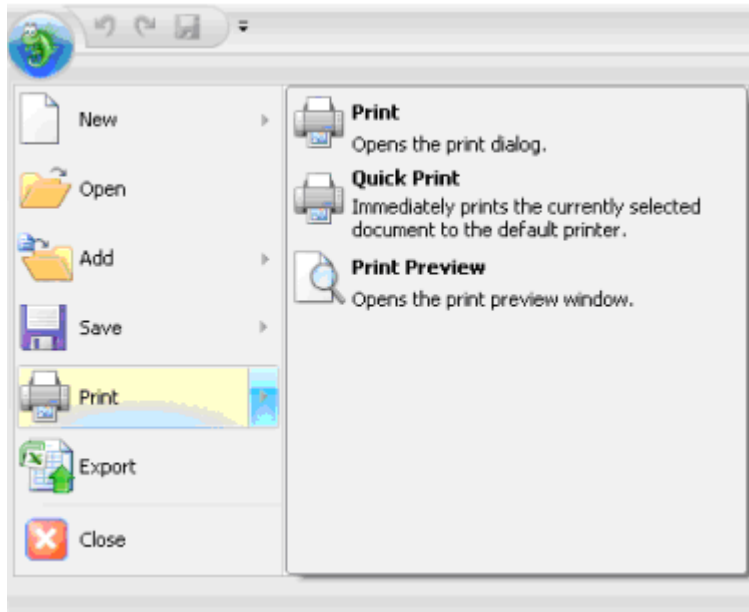


Figure 24. Selecting Print

5. If any part of the test does not pass, make the appropriate repair/adjustments and rerun the test. Refer to Section 5 for troubleshooting assistance.
6. To qualify another cartridge, select the new cartridge type in the control panel, add the XQ_EG_Current, XQ_EG_Dual_Current or XQ_IC_EG_Current sequence to the queue list, set the status of the Water_EG and Stop samples to single and repeat steps 1 - 4.

5.2.3 Performing the Main Qualification

The main part of the OQ/PQ consists of the tests 5 – 12 of the OQ/PQ Test Order, and this part of the qualification requires that an absorbance or conductivity detector be configured on the system being qualified.

If the qualification will be performed using a manual injection valve, use full loop injections and overfill the loop a minimum volume equal to 5 times the sample loop volume. Inject standards at the beginning of each sample in the sequence. There will be a 2.4 minute delay before the sample is injected. It is advisable to manually edit the programs to replace the 2.4 minute delay with a message command to hold the system until the sample is ready.

Replace the “-2.400 Pump_Relay_1.Closed Duration=130.00” line with “-0.100 Message "Load the sample into the injection loop, press OK to continue"”

1. The sequences must be installed as described in Section 4.6.
2. The sequences must be set up as described in Section 4.7.

If the queue is not started already go to queue and Edit. Start the queue. Run times are as follows:

- Systems with an AS-DV or manual injection have a run time of approximately 1 h, 10 min.
- Systems with an AS-AP or AS but no sample prep have a run time of approximately 2 h, 10 min.
- Systems with an AS-AP with Injection Temperature Accuracy Test have a run time of approximately 2 h, 40 min.
- Systems with an AS and sample prep have a run time of approximately 2 h, 40 min.
- Systems with a gradient pump, add an extra 60 min to the above times.

At the end of the main qualification the system will turn off and the automatically stop.

For systems using an Eluent Generator, the program will pause before the Eluent Generator Current Qualification Test, proceed to section 5.2.1 for re-plumbing information. The program will pause before the Flow Rate Accuracy and Precision Test, proceed to section 5.2.2 before continuing to step 3.

3. When the queue is finished, go to the Chromeleon console right click on each completed sequence present in the qualification, select the **Report** and **Print** the report. The Eluent Generator Current Qualification, Flow Rate Accuracy and Precision Qualification, Column Heater Accuracy qualification have already been printed.
4. Review the report for Pass/Fail results. If any test does not pass, make the appropriate instrument repair and/or adjustments and rerun all tests from the original point of failure on. Refer to Section 5 for troubleshooting assistance.

5.2.4 Turning off watermarks in the reports

Some users prefer not having watermarks in the back of the report. The procedure below describes how to turn off watermarks in the reports. The procedure can only be performed if the user has sufficient privileges granted by the administration.

1. Go to the Instrument Category
2. In the Chromeleon Console, Go to the Tools menu.
3. Select Administrations Console

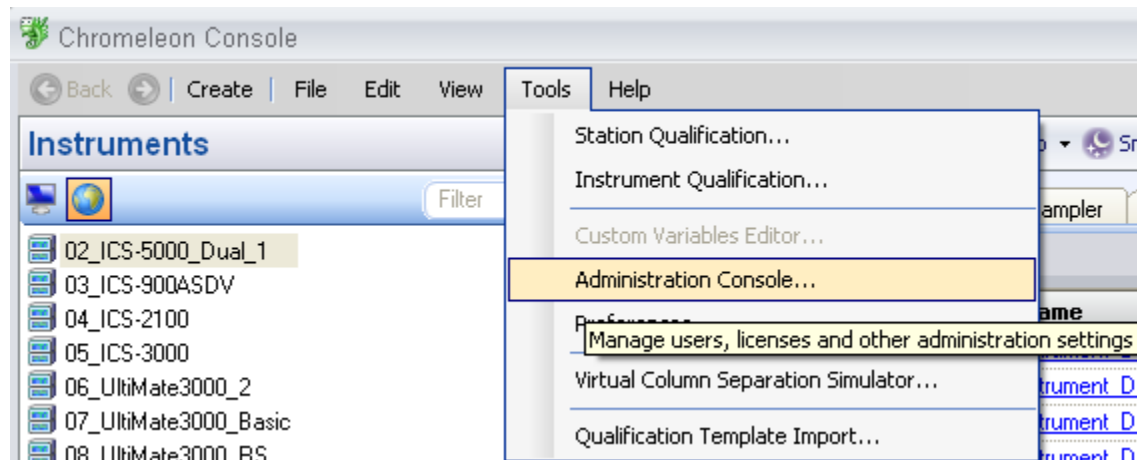


Figure 25. **Selecting Administration Console**

4. Select Global Policies within the Administration Console.

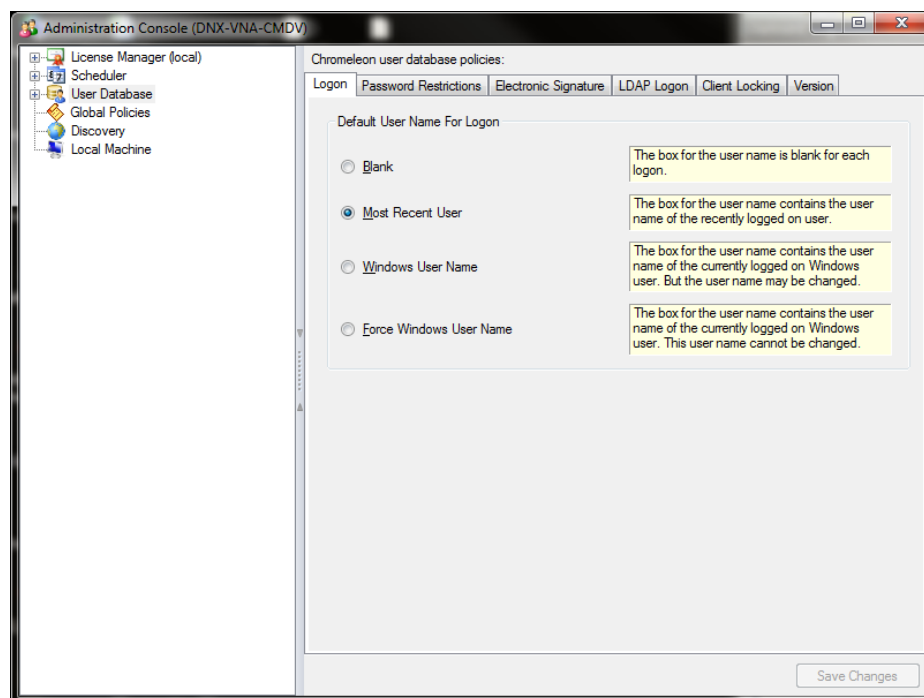


Figure 26. **Selecting Global Policies**

5. Select "Reporting" tab, then uncheck "Print with watermark"

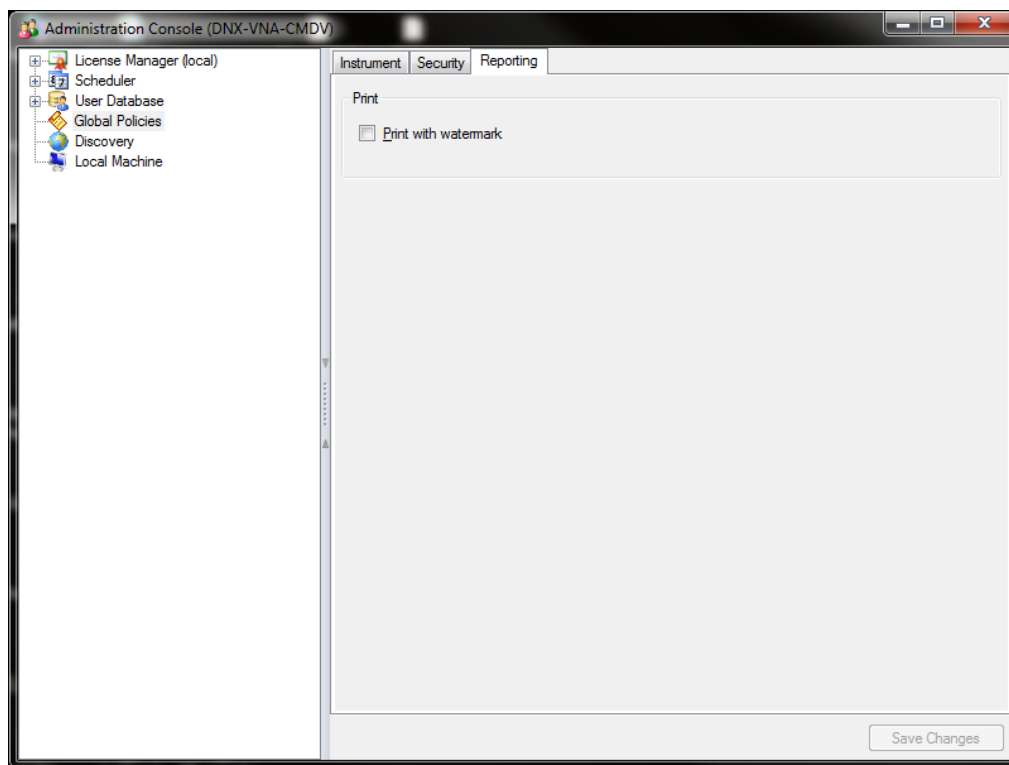


Figure 27. Selecting reporting

5.2.5 Column/Compartment TC Temperature Qualification

The column/compartment TC temperature qualification can be used to qualify the temperature accuracy of the column/compartment TC on an ICS-5000/ICS-5000⁺. After completion of all other tests record the reading on the digital thermometer. See 5.2.5 to set up the Thermocouple on to the digital thermometer.

Open the Water_WU sample in the Warm Up sequence from the console. Select the **Temperature Qualification** tab at the bottom of the page. Enter the measured temperature of the Column TC in the "Measured Column Temperature" cell and if the Compartment TC is installed, enter the measured temperature of the Compartment TC in the "Measured Compartment Temperature" cell.

Note: The template of this report is available on the PQ_OQ CD and can be used for manual data collection and data calculations

Template name: OQ_PQ_Modular_Validation

If the test does not pass, make the appropriate repair/adjustments and rerun the test. Refer to Section 6 for troubleshooting assistance.

Print the Column TC Temperature Verification report by selecting **Print** from the File menu. Choose a printer and press **OK** to begin printing.

5.2.6 Sample Temperature Accuracy (AS-AP autosampler only)

This section describes how the sample temperature accuracy is determined for the following Thermo Fisher Scientific autosampler: AS-AP. For this test, only the autosampler is required. The other modules of the IC system are not required.

Test Procedure

The following table lists the materials required for performing the test.

Part No.	Description	Quantity
6820.0010	Type K temperature sensor for P600 thermometers	1
5705.0050A	Column Thermostat PQ Kit	1

Note: The temperature accuracy test can be performed with a P600 thermometer or equivalent.

In addition, a standard polypropylene vial (10.0 mL) is required. Fill the vial with water (do not cap).

Connecting and Configuring the System

System connections

- Connect the Type K temperature sensor to the P600 thermometer and set the sensor type and calibration values as described in the instructions for the thermometer.



Figure 28. P600 Thermometer and Type K Sensor

- Fill an open standard polypropylene vial (10 mL) with water and place it at sample position BC1 if you have a 10 mL rack or sample position RB1 if you have a small vial rack.
- Insert the temperature sensor into the vial at a right angle until the tip touches the vial bottom.



Figure 29. P600 Thermometer with the Type K sensor inserted into the vial

- Rotate the carousel cover until it closes completely.



Figure 30. Carousel cover closed

- **Configuration**
Configuration is not required.
- **Preparing Chromeleon**
To qualify the sample temperature accuracy, select the following sequence:
SAMPLER_TEMP_ACC

Performing the Check

Set the sample temperature to 15 °C. When the nominal temperature is reached, the external thermometer is used to record the sample temperature over a period of 30 minutes.

Tip: Do not perform any autosampler commands during the test. Moving the needle arm or carousel may damage the thermometer or autosampler.

Duration

The test takes approximately 1 hr.

5.2.7 AS-AP pH and Conductivity Qualification

This section describes how the sample pH and conductivity accuracy is determined for the following Thermo Fisher Scientific autosampler: AS-AP. For this test, the autosampler and other modules of the IC system are required. Please refer to the operator manual for pH and conductivity detector set up. The pH qualification should be performed after the standard qualification. The AS-AP pH and conductivity qualification will be set up in push mode only.

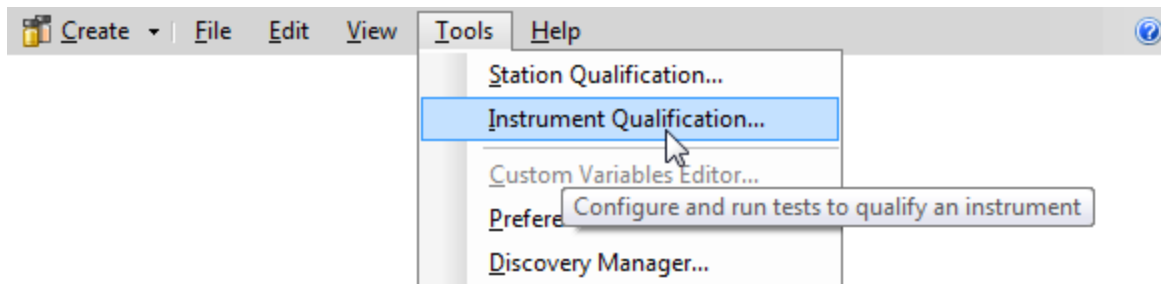


Figure 31. Instrument Pane for Chromeleon 7.1

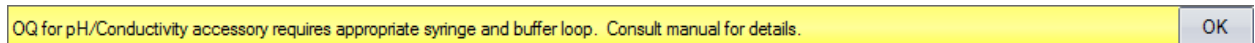


Figure 32. Starting the Instrument Qualification Wizard for Chromeleon 7.1

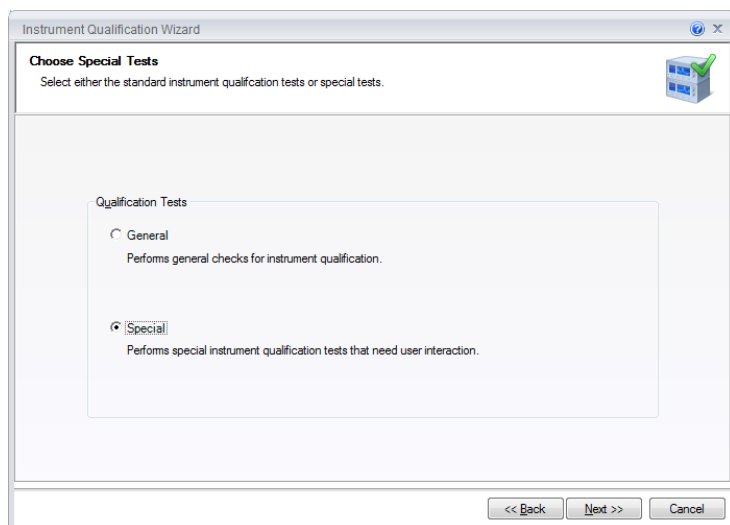


Figure 33. Select special for the Qualification Tests

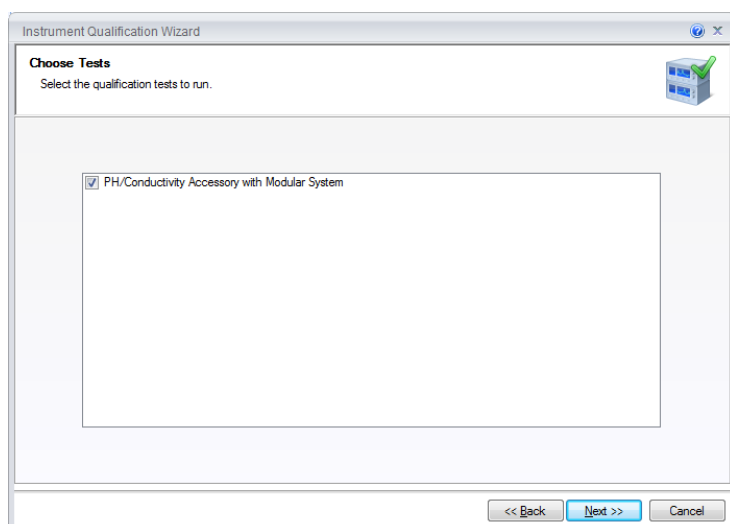


Figure 34. The pH/ Conductivity Tests will be selected if the instrument is configured correctly.

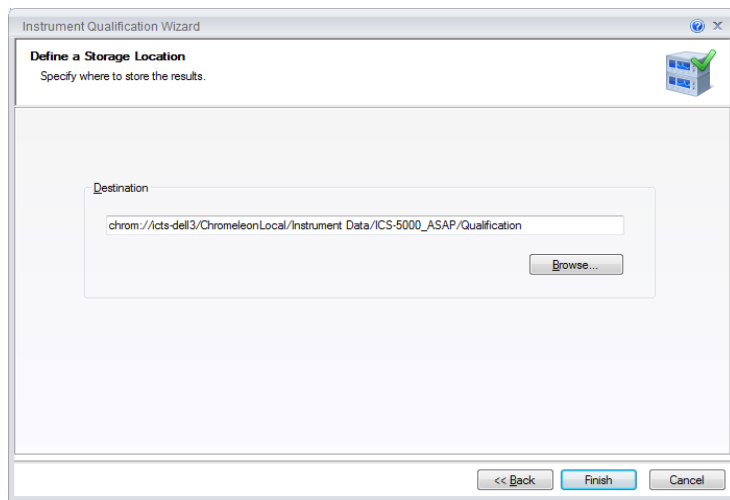


Figure 35. Select finish

#	Chromatogram ▶	Name	Type	Level	Position	Volume [µL]	Instrument Method	Processing Method
1	None	WARMUP	Blank	01	RA1	100.0000	WARMUP	Validation
2	None	pH_OFFSET_CALIBR...	Blank	01	RA1	100.0000	pH7_CALIBRATION	Validation
3	None	pH_SLOPE_CALIBR...	Blank	01	RA2	100.0000	pH4_CALIBRATION	Validation
4	None	pH_SLOPE_CALIBR...	Blank	01	RA3	100.0000	pH10_CALIBRATION	Validation
5	None	pH_VERIFICATION(p...	Blank	01	RA1	100.0000	pH_MEASUREMENT	Validation
6	None	pH_VERIFICATION(p...	Blank	01	RA2	100.0000	pH_MEASUREMENT	Validation
7	None	pH_VERIFICATION(p...	Blank	01	RA3	100.0000	pH_MEASUREMENT	Validation
8	None	CD_CALIBRATION	Blank	01	RA4	100.0000	CD_CALIBRATION	Validation
9	None	CD_MEASUREMENT	Blank	01	RA5	100.0000	CD_MEASUREMENT	Validation

Figure 36. Sequences

Performing the Check

1. Fill the vials according to the table below.

Table 17. OQ/PQ Test Timetable

Qualification	Standard	Sample Order on AS-P
pH Measurement	pH OFFSET Calibration	RA1
pH Measurement	pH Slope Calibration	RA2
pH Measurement	pH Slope Calibration	RA3
CD Calibration	Blank	RA4
CD Measurement	Conductivity Standard	RA5

2. Start the sequence.
3. A pop out screen will remind the user to configure the AS-AP with the 5mL syringe and the 8500 µL buffer loop before the qualification begins.

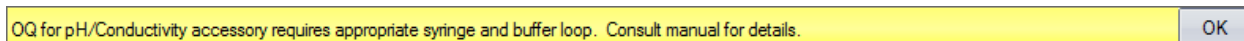


Figure 37. Reminder Screen

Duration

The test takes approximately 1 hour.

5.2.8 Qualification Comments

The comments section of the report is provided as a place to record any comments, test failures, or configuration changes that occur during the qualification.

To use the comment section, open the Water_WU sample in the Warm Up sequence from the Console. Select the Comment tab at the bottom of the page. Enter any comments. This page can also be printed and comments entered manually.

To print the Comment report, select **Print** from the **File** menu. Choose a printer and press **OK** to begin printing.

Note: The template of this report is stored in the data vault of the respective sequence. Please refer to Chromeleon 7 Quick Start Guide on how to access report templates.

Template name: OQ_PQ_Modular_Validation

5.3 What to do if a Test Fails

If at any point the instrument results do not pass within Thermo Fisher Scientific specified limits, the instrument must be serviced. See Chapter 6 for troubleshooting assistance. When the repair is complete, rerun all tests from the original point of failure on in the sequence. All actions for failures and repairs should be recorded on the "comments" report. For example, if the instrument passes tests 1, 2, and 3 and fails test 4, make the appropriate repairs, record the action, and then repeat tests 4 – 12 only.

When rerunning the tests, restart the Instruments OQ or Instrument PQ procedure from the Qualification menu and uncheck all passed tests prior to the test that failed. All tests after the test that failed should be left checked even if they have previously passed. The Warm Up sequence is always required and must be rerun.

5.4 OQ/PQ Completion

5.4.1 Customer Review

1. The printed results should be reviewed by the instrument owner/user.
2. When the qualification is accepted, both the Customer and Qualification Executor need to sign the printed reports.
3. Fill out the Qualification Stickers and Certificates of Functional Performance for the qualified modules.
4. Make a copy of the certificates for FSR qualification, multimeter if used, temperature probe if used, and original standard certificate(s).
5. Place the reports and certificates into the binder provided in the OQ/PQ kit and leave it with the customer.

5.4.2 Accuracy of Reports

In accordance with Good Laboratory Practice (GLP), calculations used to determine the results in the printed reports use more significant figures than those shown and are rounded down for the final report. When reproducing the calculations by hand it may be necessary to obtain numbers from the reports after increasing the number of displayed significant figures.

The number of displayed significant figures can be adjusted through the report publisher. Open the sequence containing the samples for the test being examined and double click on a sample. The sample data should be shown. Highlight the numbers being used for the hand calculations, click on the Format menu and select number. The number of digits displayed can be increased by changing the number type; for example, entering 0.00000 into the Type field will increase the number of displayed decimal places to 5. **Always use more significant figures for hand calculations than is shown in the printed report.**

5.4.3 Returning the System to the Original Configuration

1. If the Signal factor was modified in the system preparation, or a module was renamed it must be set back to its original value. Open the Chromeleon Instrument Configuration application. Double click on the detector qualified to view the **properties** page. Select the **Signals** page to change the factor back to the original value and the **General** page to change the name back to the original value. Exit the properties page and save the configuration.
2. Disconnect the Backpressure Coil and restore the system plumbing to its original configuration with the appropriate columns, suppressors, and Eluent Generator in line.
3. Remove the sample loop and reinstall the original sample loop back onto the injection valve. For systems using an AS Autosampler, go to the **Main Menu** on the front panel of the autosampler. Access the **Module Set Up** screen and then go to **Plumbing Configuration**. Change the sample loop size back to its original value. For systems using an AS-AP Autosampler, open Instrument Configuration and go to the **Options** tab for the AS-AP Autosampler and change the sample loop size back to the original value.
4. Reconnect the original eluents to the pump and prime each line individually to purge the eluents used in the OQ/PQ tests.
5. Flush the system with an eluent recommended by the customer and confirm that there are no leaks.

6 Troubleshooting

6.1 General Hints

If the system pressure is above 2000 psi after connecting the PEEK Backpressure coil with a flow rate of 1.00 mL/min, the coil has a blockage. Re-cut the ends of the coil or replace it.

Dips before or after the main nitrate or caffeine peak indicates extra dead volume in the system, this may be caused by small voids between the end of a liquid line and the bottom of the port. Individually disconnect each line from the injection valve to the detector cell. Push the ferrule and fitting up and away from the end of the tubing. For analytical systems only: Make sure the tubing is cut straight. If it is not, re-cut the end.

6.2 Failure of Individual Tests

6.2.1 Conductivity Detector Tests

Test	Reason	Action
Baseline Noise	Poor water quality	Replace with an alternate water source
	Air bubbles in flow cell	Flush out flow cell
	Cell temperature regulation failure	Replace cell
Drift	System not equilibrated	Allow longer flush/equilibration time
	Lab temperature not stable	Regulate temperature (block vents, close windows, etc.)
	Water not degassed	Degas water or turn degas unit onto "Always On" setting.
Detector Linearity	Concentration of standards is incorrect	Obtain new fresh standards
	Liquid leak	Find and repair source of leak
	Injection volume not consistent (manual injections only)	Overfill sample loop at least 4x when loading
	The peak height of the sample with the highest concentration is not in the linearity range specified for the detector, i.e., usually > 1500 mAU (for Thermo Fisher Scientific and PDA detectors).	Reduce the injection volume for all samples used for the detector linearity check so that the peak height of the sample with the highest concentration is in the linearity range of the detector, i.e., usually < 1500 mAU.

6.2.2 Pump

Test	Reason	Action
Gradient Step Accuracy	Air in system	Prime pump – prime each eluent line individually for 3 – 5 minutes, flush system
	System not equilibrated	Allow longer flush/equilibrium time
	Old eluent in vacuum degas chamber	Prime pump – prime each line individually for 3 – 5 minutes
	Incorrect eluent concentration for eluent B and D	Remake 0.3 % Acetone, 1 mM KCl solutions or 8.3 μ M glucose in 50 mM NaOH
	Pump eluents might need to be pressurized	Pressurize eluent bottles with 5 psi helium or nitrogen
	Eluent bottles not vented	Vent eluent bottles
	Pump is not primed	Prime the pump
Step Noise (ripple)	Eluent lines not conditioned with new eluent	Prime all 4 eluent lines for at least 15 minutes
	Air in system	Prime pump, flush system
	Air in flow cell	Flush flow cell with iso-propyl alcohol, ensure adequate backpressure on flow cell
	System not equilibrated	Allow longer flush/equilibrium time
	Check valve or piston seal failure	Replace check valve/piston seal
Flow Rate Accuracy and Precision	Void time from pump to detector too long or short	Adjust tubing length accordingly
	Flow rate out of calibration	Re-calibrate the flow rate of the pump

6.2.3 Autosampler

Test	Reason	Action
Injector Precision	Air injected from vial or from sample syringe when doing manual injections	Too little sample volume in vial – refill vial or flush bubbles out of injection syringe
	Air bubbles in AS or AS-AP sample syringe	Flush out syringe
	Liquid leak in autosampler or injection valve	Find and repair source of leak
	Poor water quality in flush reservoir	Change water and flush sampler fluidics
Injection Volume Linearity	Detector failure	See Detector linearity troubleshooting section
	Defective sample syringe	Replace sample syringe
Carryover	Contaminated injection valve	Replace injection valve rotor and stator face
	Contamination in autosampler	Flush/replace autosampler liquid lines
	Contamination in sample vial or in syringe used for manual injections	Use new sample vial for deionized water blank, rinse vial thoroughly before using, flush syringe or use a new one
Sample Prep Precision	Air injected from vial	Too little sample volume in vial, refill vial
	Air bubbles in AS sample syringe or prep syringe	Flush out syringe
	Defective prep syringe	Replace prep syringe
	Liquid leak in autosampler or injection valve	Find and repair source of leak
Tray Temperature Accuracy	Poor water quality in Sample Prep reservoir	Change water and flush sampler fluidics
	Thermometer not calibrated	Calibrate thermometer

6.2.4 Integrated Amperometry Detector Tests

Test	Reason	Action
Baseline Noise	Electrochemical cell not connected properly	Reconnect test cell cable to amperometry cell connection on SP board
	Electrochemical cell defective	Replace DWE, reference electrode or electrochemical cell
Drift	Detector not equilibrated	Allow longer equilibration time
	Lab temperature not stable	Regulate temperature (block vents, close windows, etc.)

6.2.5 Eluent Generator

Test	Reason	Action
Current	Qualification Test box not connected properly	Reconnect test cell cable to the Eluent Generator, confirm ammeter or multimeter leads are making contact with test cell wires
	Ammeter or multimeter on wrong setting	Ammeter or multimeter should be set to read current (mA). Range should be set to view 0.00 to 300.00 mA
	Qualification cell defective	Replace Qualification test cell

6.2.6 Column Thermostat

Test	Reason	Action
Temperature Qualification	Column Compartment not calibrated	Recalibrate the Column Heater
	Thermometer not calibrated	Calibrate thermometer
	Thermometer not fitted in column compartment correctly	Fit thermometer in column compartment correctly

7 Example Reports

The example reports are for:

An ICS-5000/ICS-5000⁺ Analytical system configured with a CD Detector, DP Gradient Pump, and an AS-AP Autosampler.

An ICS-5000/ICS-5000⁺ Analytical system configured with a ED Detector, DP Gradient Pump, and an AS-AP Autosampler.

**User's Guide:
Chromleon[®] Operational Qualification and
Performance Qualification Software Templates
for ICS-5000/ICS-5000⁺ Capillary Instruments
Revision 7.12**

2013 Thermo Fisher Scientific

Document 065448 Part F
March 2013, Thermo Fisher Scientific Corporation
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PRINTING HISTORY

Revision 7.10, September 2011

Revision 7.11, December 2011

Revision 7.12, March 2013

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1 Introduction

Operational and Performance Qualification are becoming increasingly important to analytical laboratories. Documented evidence must be provided to demonstrate the integrity of data collected and qualify the results obtained on laboratory instrumentation.

The following institutes set and control industry instrumentation standards:

NIST The National Institute of Standards and Technology is an administrative body of the US Department of Commerce. Together with industry, the NIST develops new technologies and sets technological standards countrywide. Thermo Fisher Scientific includes NIST traceable standards as part of the qualification kit.

ASTM The American Society for Testing and Materials sets technical standards and rules for the industry worldwide, specifically; E 1657 – 98: Standard Practice for Testing Variable-Wavelength Photometric Detectors in Liquid Chromatography. E1511 – 93: Standard Practice for Testing Conductivity Detectors used in Liquid Chromatography. E19.09.07: Proposed Standard Practice for Evaluation of Gradient HPLC pumps – Draft.

USP The United States Pharmacopeia (USP) is an official public standards–setting authority for all prescription and over–the–counter medicines and other health care products manufactured or sold in the United States. USP establishes documentary and reference standards to ensure quality analytical instrument qualification process.

These standards are written in broad terms to make them as widely applicable as possible. All stipulate general requirements specifying instruments must be fit for purpose, technically checked in defined periods, calibrated and make sure that test results are documented according to international quality standards. The OQ/PQ procedure used to qualify Thermo Fisher Scientific Ion Chromatography Systems meets the requirements established by NIST, ASTM and USP.

This OQ/PQ procedure provides qualification testing for **ICS-5000/ICS-5000⁺** capillary systems composed of any modules listed in section 1.6.

The OQ/PQ results can be influenced by unusual laboratory conditions. For example, direct sunlight and close proximity to air conditioning ducts will cause thermal irregularities that will affect baseline drift on conductivity detectors. In these cases, it may happen that the manufacturer's specifications are not met. Ensure that the instrument is operated in suitable environmental conditions or for PQ, the specifications may be adjusted accordingly.

All OQ/PQ activities must be formally recorded, all instruments must be clearly identified, and test reports and supporting documentation must include the following information:

1. Module name, type and serial number
2. Software and firmware version
3. Test data
4. Service interval
5. Signature of the Thermo Fisher Scientific Service Engineer/Qualification Executor who performed the OQ/PQ
6. Signature of the customer who reviewed and accepted the OQ/PQ results

1.1 How to use this manual

It is essential that the operator read and understand this manual completely before implementing any OQ or PQ process.

1. Use the table of contents to determine which tests are required for the system to be qualified and refer to the recommended sections. This will help keep track of what steps to follow during system setup and preparation. It will also aid in selecting the appropriate sequences to run the tests.
2. For the ICS-5000/ICS-5000⁺ capillary system, electrochemistry cells in Integrated Amperometry mode are supported for qualification tests. For the Integrated Amperometry detection qualification, a pH/Ag/AgCl reference electrode is required for all tests in capillary format. The ICS-5000/ICS-5000⁺ electrochemistry detector modes for DC amperometry and cyclic voltammetry are not supported for OQ/PQ testing. When qualifying Integrated Amperometry detection a disposable gold working electrode is used; other electrodes are not supported.
3. Use the **Table of Contents** and the **List of Tables** as reference guides to locate information quickly.

The electronic version of the OQ/PQ operator's manual contains hypertext links that can take you to other locations within the file. These links include:

- Table of contents entries
- Cross-references (underlined in blue) to sections, figures, tables, etc.

Chapter 1 Introduction

Introduces the conventions used in performing OQ/PQ with the ICS-5000/ICS-5000⁺ capillary systems; including the basic requirements, industry standards and manual summary.

Chapter 2 Overview of the Tests

Provides an overview of the tests available to be performed.

Chapter 3 Explanation of Test Procedures

Describes the theory behind each test, the OQ/PQ operating procedures and evaluation of the results.

Chapter 4 Qualification Preparation

Step by step preparation instructions for the OQ/PQ software, hardware installation and the details of the materials in ship kits.

Chapter 5 Performing the Qualification

Detailed operating instructions on how to perform the OQ/PQ including test order, time table, sequence initialization and troubleshooting tips.

Chapter 6 Troubleshooting

Lists problems and presents step-by-step procedures for how to isolate and eliminate the cause of each problem.

Chapter 7 Example Reports

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1.4 Operational Qualification (OQ)

According to “The Development and application of guidance on equipment qualification of analytical instruments” by P. Bedson and M. Sargent [Accred. Qual. Assur. (1996) 1: 265 – 274] the following definition applies:

“The purpose of OQ is to prove and document that an analytical system functions according to its operating specification while the specific environmental conditions are taken into account. In his specification, the supplier has therefore to define exactly the conditions that must be observed.”

OQ should be performed after initial system installation or when an additional new module is added to an existing system. OQ should be done according to the instructions outlined in this manual and all tests should be performed by a qualified Thermo Fisher Scientific Service Engineer. Any hand-written notes made during the performance of the tests must become part of the test results, in accordance with Good Laboratory Practice (GLP) and current Good Manufacturing Practice (cGMP).

1.5 Performance Qualification (PQ)

According to “The Development and application of guidance on equipment qualification of analytical instruments” by P. Bedson and M. Sargent [Accred. Qual. Assur. (1996) 1: 265 – 274] the following definition applies:

“The purpose of PQ is to prove and document that an analytical system functions according to a specification that is suitable for the system’s routine operation. A system is subject to wear when being operated. Thus, it may happen that the supplier’s specification is no longer met. This means: The test procedures used for OQ are also used for PQ, but the tolerances of PQ are less restrictive than those for OQ. Using the same test procedures simplifies the handling.”

PQ should be performed at regular intervals after the initial installation and OQ. Thermo Fisher Scientific recommends performing PQ every six months. PQ should be done according to the instructions outlined in this manual and all tests should be performed by a qualified Thermo Fisher Scientific Service Engineer. Any hand-written notes made during the performance of the tests must become part of the test results, in accordance with Good Laboratory Practice (GLP) and current Good Manufacturing Practice (cGMP).

1.6 Basic Requirements for Successful OQ and PQ

It is recommended that the most current revision of the OQ/PQ software templates is used in order for a qualification to be considered valid. It is the responsibility of the qualification engineer to determine the most up to date revision of the templates available and ensure that the revision is used before proceeding with this qualification or if the customers SOP require an earlier OQ/PQ version to be used.

It is recommended that the qualification be performed using the most current Chromeleon[®] service pack version (Contact Thermo Fisher Scientific Technical Support for current version).

It is recommended that the qualification be performed using the most current firmware version for each system or module (Contact Thermo Fisher Scientific Technical Support for current version).

Operational Qualification and Performance Qualification are system-specific test procedures. The procedures described below are valid for IC systems comprising of the following equipment (only one type of detector and one pump can be qualified concurrently):

- Pump: ICS-5000/ICS-5000⁺ SP or ICS-5000/ICS-5000⁺ DP (Capillary version)
- Manual injection or any of the following autosamplers: AS, AS-DV or AS-AP
- AS-AP autosampler 10 mL tray (P/N 074938) with ICS-5000/ICS-5000⁺ CD (push mode) and 1.5 mL tray (P/N 074936) with ICS-5000/ICS-5000⁺ CD or ED (push mode only). To qualify the AS-AP, a 250 µL injection syringe is required. The buffer size should always be set at 1200 µL.
- Conductivity detector: ICS-5000/ICS-5000⁺ CD (Capillary version)

- Electrochemical detector: ICS-5000/ICS-5000⁺ ED (Capillary version)
- Eluent Generator: ICS-5000/ICS-5000⁺ EG

Note: Ensure that the wash option is turned off during the qualification.

In addition, Chromeleon[®] version **7.10** or higher is required and the following requirements must also be met:

Table 1. ICS-5000/ICS-5000⁺ and Chromeleon Compatibility

Module	Minimum Chromeleon Software Version
AS-AP	7.1 SR1
AS	7.1 SR1
AS-DV	7.1 SR1
ICS-5000/ICS-5000 ⁺ SP or DP	7.1 SR1
ICS-5000/ICS-5000 ⁺ EG	7.1 SR1
ICS-5000/ICS-5000 ⁺ DC	7.1 SR1
ICS-5000/ICS-5000 ⁺ CD	7.1 SR1
ICS-5000/ICS-5000 ⁺ ED	7.1 SR1
ICS Series VWD	7.1 SR1
FLUKE189/289	7.1 SR1

It is possible to qualify modules in systems that are not listed in this section; however some modification of the included programs may be required. Such modifications are not within the scope of this document.

For the ICS-5000/ICS-5000⁺, it is not possible to qualify multiple pumps on a single system. If multiple pumps are configured on a single Chromeleon Timebase, the second pump will be ignored by the programs.

2 Overview of the tests

The following table provides an overview on the parameters to be tested and the limits recommended for OQ and PQ.

- Template sequences (with programs specific to the Instrument Configuration) are generated based on the master sequence of the OQ/PQ CD (P/N 057601-rev19) and system configuration in the IC Timebase.
- The sequences are copied from each test that is to be performed on the subject system.
- Capillary OQ/PQ runs at 0.010 mL/min.
- Capillary OQ/PQ sequences typically begin with XQ_CIC_...
- The Chromeleon software will automatically create a default unique name and directory to contain all template sequences.
- Do not alter the test sequence order.

Table 2. OQ/PQ Specification Limits: ICS-5000/ICS-5000⁺ CD Baseline Noise/Drift and Detector Linearity

Instrument	Parameter	Description	Limits	
			OQ	PQ
ICS-5000/ICS-5000 ⁺ CD	Baseline Noise	ASTM Type I deionized water with low total organic carbon (TOC)* is pumped through the detector at a flow rate of 0.010 mL/min.	1.0 nS	2.0 nS
	Baseline Drift		10.0 nS/hr 3.33 nS/ 20 min	20.0 nS/hr 6.67 nS/ 20 min
	Detector Linearity	5 Nitrate solutions from the OQ/PQ standards kit P/N 063540 are injected in different concentrations. Concentration (ppm) and peak height (μS) are represented on a graph. The correlation coefficient for the resulting line and the deviations from it indicate the linearity.	r ≥0.999 RSD ≤5.0 %	r ≥0.999 RSD ≤5.0 %
ICS Series VWD	Baseline Noise	ASTM Type I Deionized water with low TOC is pumped through the detector at a flow rate of 1.0 mL/min. Wavelength 254 nm and 520 nm.	50 μAU	50 μAU
	Baseline Drift		4000 μAU/hr	4000 μAU/hr
	Wavelength Accuracy	Holmium oxide maxima are compared with the theoretical maxima.	± 2 nm	± 2 nm
	Detector Linearity	5 Caffeine solutions from the OQ/PQ standards kit P/N 063540 are injected in different concentrations. The absorbance is measured at a wavelength of 272 nm. Concentration (ppm) and peak height (mAU) are represented on a graph. The correlation coefficient for the resulting line indicates the linearity.	r ≥0.999 RSD ≤5.0 %	r ≥0.999 RSD ≤5.0 %

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Table 3. OQ/PQ Specification Limits: ICS-5000/ICS-5000⁺ ED Baseline Noise/Drift

Instrument	Parameter	Description	Limits	
			OQ	PQ
ICS-5000/ICS-5000 ⁺ ED Capillary	Baseline Noise	15 mM of electrolytically generated KOH is pumped through the detector cell at a flow rate of 0.010 mL/min. An Amino Acid disposable gold working electrode (Thermo Fisher Scientific P/N 060082), with a 1 mil x 1 mm PTFE cell gasket (Thermo Fisher Scientific P/N 072161) and the Ag/AgCl pH reference electrode with a cell gasket (Thermo Fisher Scientific P/N 072162) are set up.	Int. Amp: ≤100 pC	Int. Amp: ≤160 pC
	Baseline Drift		Int. Amp: ≤3000 pC/hr ≤1000 pC/20 min	Int. Amp: ≤3750 pC/hr ≤1250 pC/20 min
	Detector Linearity	5 Threonine solutions from the Capillary ED OQ/PQ standards kit P/N 074730 are injected in different concentrations. Concentration (µM) and peak height (nC) are represented by a plot. The correlation coefficient for the resulting line (and the deviations from it) indicates the linearity.	r ≥0.999 RSD ≤5.0 %	r ≥0.999 RSD ≤5.0 %

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Table 4. OQ/PQ Specification Limits: Injector Precision, Carryover and Sample Prep Precision

Instrument	Parameter	Description	Limits	
			OQ	PQ
All injectors and samplers	Injector Precision	10 injections of a defined standard with full loop injection at 25 µL each of the same standard are analyzed. The percent relative standard deviation (%RSD) of the peak areas indicates the reproducibility of the injection volume.	CD Capillary: RSD ≤0.8 % ED Capillary: RSD ≤3.0 %	CD Capillary: RSD ≤1.0 % ED Capillary: RSD ≤3.0 %

	Carryover	Following the injection of a highly concentrated test sample, a blank sample is injected and then a water blank sample is injected. The peak area measured in this chromatogram compared with the peak area of the highly concentrated sample indicates the amount of carryover. If there is any peak area measured in the water blank sample, that area is subtracted from the area of the blank sample before it is compared to the highly concentrated sample.	≤0.1 %	≤0.1 %
AS	Sample Preparation Precision (if present – not supported on the AS with ICS-5000/ICS-5000 ⁺ ED detection)	3 samples of identical concentration are prepared from the same starting vial into 3 separate vials and injected. The relative standard deviation of the peak areas indicates the reproducibility of the sample preparation.	RSD ≤1.5 %	RSD ≤2.0 %
AS-AP	Temperature Accuracy (if present)	The sample temperature is measured using an external thermometer in a standard vial. The vial is filled with water and measured at 15.0 °C.	15.0 ± 2.0 °C	15.0 ± 4.0 °C
AS-AP pH	pH Accuracy	The sample temperature calibration is measuring the pH at 4, 7 and 10. The reading is stable if the reading and the three data points are within 0.2 pH unit of each other.	+/- pH 0.1	+/- pH 0.2
AS-AP Conductivity	Conductivity Accuracy	The conductivity calibration procedure consists of determining the conductivity zero point of the accessory and then measuring the conductivity of a 1000 µS standard.	+/- 30 µS	+/- 50 µS
AS-AP	Temperature Accuracy (if present)	The sample temperature is measured using an external thermometer in a standard vial. The standard vial is filled with water (measured at 15.0 °C).	15.0 ± 2.0 °C	15.0 ± 4.0 °C

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Table 5. OQ/PQ Specification Limits: ICS-5000/ICS-5000⁺ EG Current, SP/DP Flow Rate Accuracy/Precision and DC Column TC Temperature

Instrument	Parameter	Description	Limits	
			OQ	PQ
ICS-5000/ICS-5000 ⁺⁺ EG	Current Qualification	The eluent generator (EG) is commanded to produce a series of different concentrations (using the current cartridge type configured with a concentration range between 0 mM to 200 mM). At each EG concentration setting the current (mA) produced by the EG is measured and compared with theoretical values.	See report at CIC_Modular_Validation	
ICS-5000/ICS-5000 ⁺ SP, ICS-5000/ICS-5000 ⁺ DP	Flow Rate Accuracy (Capillary Pump)	Water is pumped at a flow rate of 0.010 mL/min into a weighed beaker for exactly 10 minutes. The weight of the water is measured, the flow rate of the pump calculated and compared with the theoretical value. This experiment is repeated four more times.	<u>0.010</u> ±0.0005 mL/min	<u>0.010</u> ±0.0005 mL/min
	Flow Rate Precision (Capillary Pump)	Capillary Systems: Water is pumped at a flow rate of 0.010 mL/min into a weighed beaker for exactly 10 minutes. This procedure is repeated four more times. The relative standard deviation of the five measurements is calculated.	≤5.0 %	≤5.0 %
ICS-5000/ICS-5000 ⁺ DC	Column TC and Compartment, Temperature Verification	The Capillary DC column temperature and the IC Cube are set to 30 °C. The Capillary DC compartment temperature is set to 15 °C for the duration of the entire qualification. At the end of the qualification the temperature of the column heater is measured with a calibrated external thermometer and compared with the theoretical value.	± 2.0 °C	± 2.5 °C

*ASTM Type 1 Deionized water Total Organic Carbon Specification: <50 ppb.

Note: The instrument's specifications can only be met if the conditions that are described in the accompanying manual are strictly observed.

3 Explanation of Test Procedures

3.1 Flow Rate Accuracy and Precision Qualification

3.1.1 Theory

The accuracy and precision of a capillary pump is qualified by accurately measuring 5 aliquots of eluent into a tared ~10 mL sample vial (P/N 074731) including tubing (P/N 074483), ferrule (P/N 074373) and nut (P/N 074449) for exactly 10 minutes each. A 6-port 2 position injection valve is used to measure the 10 minute aliquot and the volume of each aliquot is determined gravimetrically. The backpressure coil is placed before the injection valve and equal lengths of tubing (blue color PEEK P/N 074484 0.0063 mm ID 22 cm) are used for eluent and waste to ensure that there is no backpressure change when switching the valve from waste to sample.

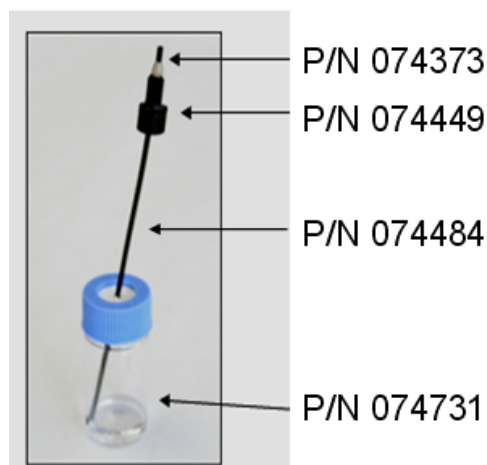


Figure 1. 10 mL Sample Vial with proper set up for flow rate accuracy test.

3.1.2 Performing the Test and Evaluating the Results

The Flow Rate Accuracy and Precision Qualification are run using the sequence called CIC_XQ_Flow_Rate. The five weight measurements for the aliquots are entered manually into the report and converted to volume when the temperature is entered. Accuracy is measured by comparing the theoretical value (0.10 mL in 10.00 min) to each of the five aliquots and precision is measured by determining the relative standard deviation of the five aliquots.

3.2 Eluent Generator Current Qualification

3.2.1 Theory

The Eluent Generator Current Qualification is determined by performing an EG step gradient using varying eluent concentration set points. The EG applies a specific current to the EG cartridge depending on the concentration of eluent and type of cartridge selected. An IC Qualification Test Box (P/N 072005) is used in place of the EG cartridge and the applied current for each step is measured using an ammeter or voltmeter that is connected to the test cell. No injection is performed.

In order to perform the test, the system is first configured with the qualification test box before the batch is started. Once the program is completed, the program prompts the user to disconnect the qualification test box and connect the EGC and CR-TC.

3.2.2 Performing the Test and Evaluating the Results

The Eluent Generator current qualification test is the next test in the sequence queue and the sequence is called XQ_EG_Current or XQ_EG_Dual_Current. The Current readings in mA are displayed on the voltmeter for each concentration step in the gradient. Values for each step are manually recorded (please enter at least three figures after the decimal in each entry) or can be automatically downloaded into the Report Template when using a Fluke 189/289 or equivalent multimeter (calibrated with up to 300 mV DC scale range and the multimeter needs to display three decimal places) which is configured in the Instrument as a module using the appropriate device driver. The sequence follows an XQ_Stop sequence that will pause the system to allow connection of the qualification cell and recording of the results.

3.3 Warm up

The warm up sequence is used to ensure that all parts of the system are responding appropriately and to record logs to the audit trail (later used to update the information in the report definition file). It is called XQ_Warm_Up where; X is P for PQ and O for OQ. The warm up sequence is necessary for all qualifications. The amperometry detector qualifications have a 1 hour warm up period. The warm up sequences from the AS-AP directory inject the low level standards.

3.4 Baseline Noise and Drift (Conductivity Detector and Absorbance Detector)

3.4.1 Theory

A 20-minute detector baseline background trace is collected using deionized water as the eluent at a flow rate of 0.010 mL/min. No injection is performed. For ICS Series VWD, data is collected at 254 nm and 520 nm. No injection is performed. Then a short equilibration is done at 272 nm.

Noise is calculated by partitioning the 20-minute baseline signal into 20 one-minute intervals. The noise is the distance between two parallel lines through the measured minimum and maximum values and the regression line. The measured noise values are then averaged to establish the final value. To calculate drift, the Chromeleon software uses a method of least squares and calculates a regression line from the measured values. The slope of the regression line indicates the drift of the measured signal, and the amount of the slope indicates the amount of drift.

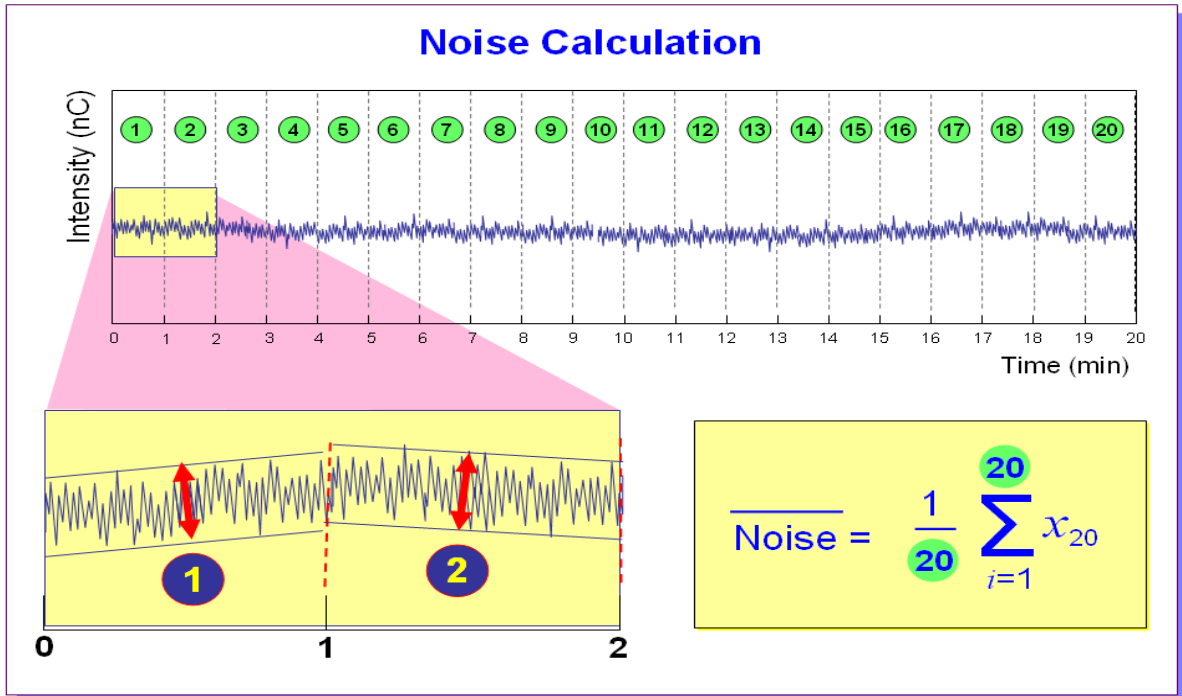


Figure 2. Noise Calculation

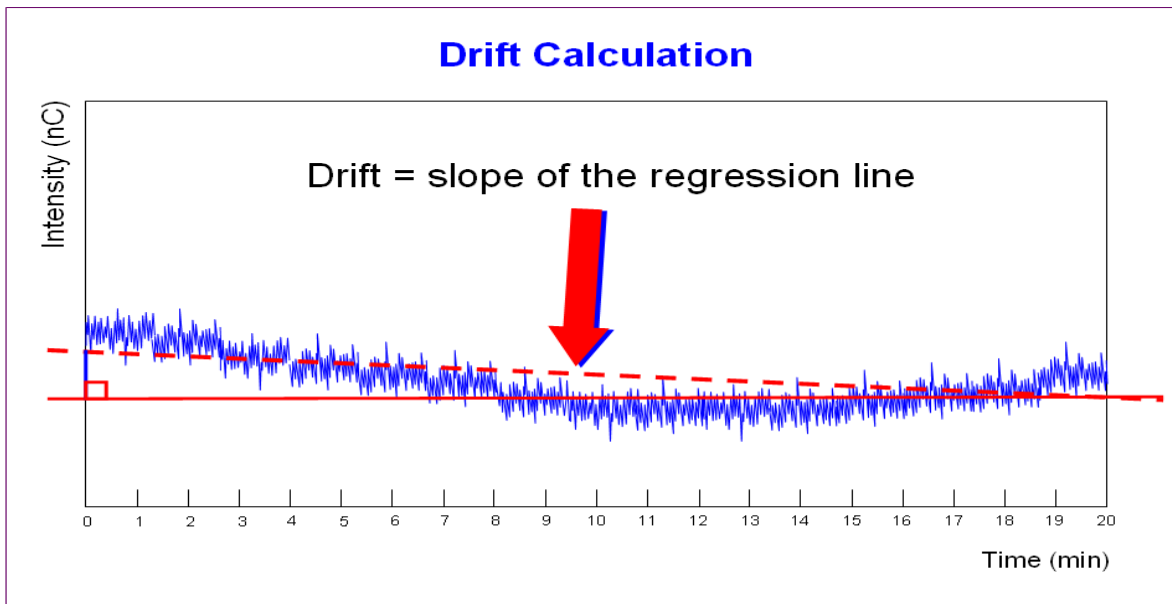


Figure 3. Drift Calculation

The slope of the regression line indicates the drift of the measured signal, and the amount of the slope indicates the amount of drift.

3.4.2 Performing the Test and Evaluating the Results

Noise and drift evaluation for all IC detectors is the next test in the sequence queue and is called *XQ_CIC_Detector_Noise_Drift* where *X* is P for PQ and O for OQ and Detector is CD for conductivity detectors.

Note: Ensure that the wash option is turned off during the qualification.

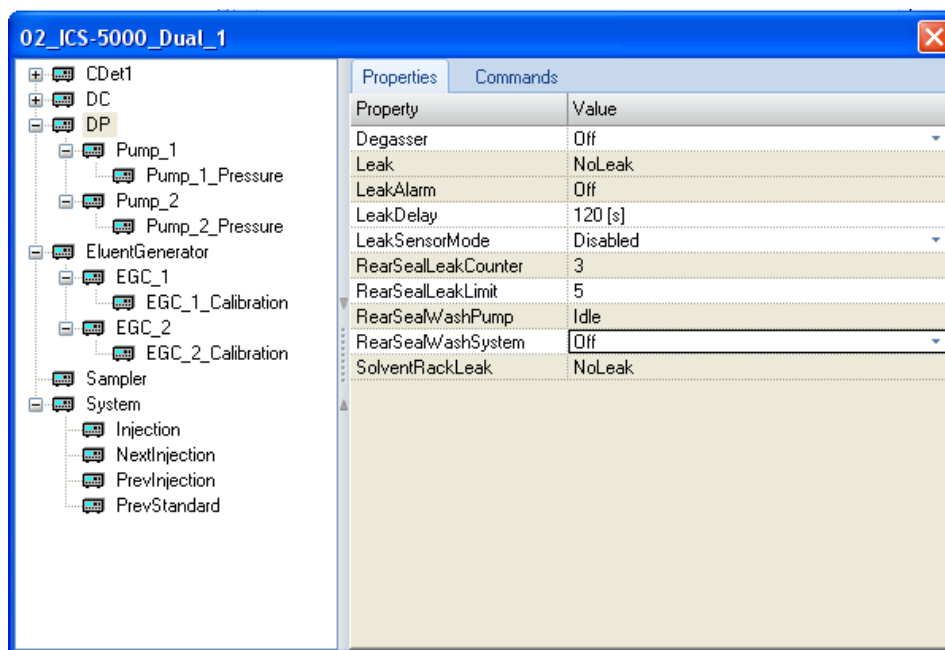


Figure 4. Rear seal wash option

3.5 Wavelength Accuracy of Absorbance Detectors

3.5.1 Theory

Wavelength accuracy is evaluated using a holmium oxide filter. Three holmium oxide absorbance maxima are compared with the theoretical maxima at 360.9 nm, 418.7 nm and 536.2 nm for ICS Series VWD detectors.

3.5.2 Performing the Test and Evaluating the Results

For the VWD detectors the wavelength accuracy test is the first test in the sequence queue and is called *XQ_Wavelength_Accuracy* where, *X* is P for PQ and O for OQ. The Wavelength Accuracy Test program executes the verification and the results are logged to the audit trail by the Wavelength Accuracy Report program.

3.6 Noise and Drift (Integrated Amperometry)

3.6.1 Theory

A 20-minute detector baseline is collected using 15 mM KOH as the eluent at a flow rate of 0.010 mL/min for capillary systems. An Amino Acid disposable gold working electrode (Thermo

Fisher Scientific P/N 060082) is used along with the Amino Acids (pH using Ag/AgCl Reference electrode) waveform. This test requires a Ag/AgCl pH reference electrode. No injection is performed.

3.6.2 Performing the Test and Evaluating the Results

Noise and drift evaluation for Integrated Amperometry a detector is the only test in the sequence queue and is called XQ_CIC_ED_Amp_Noise_Drift.

Noise is calculated by partitioning the 20-minute baseline signal into 20 one-minute intervals. The noise is the distance between two parallel lines through the measured minimum and maximum values and the regression line. The measured noise values are then averaged to establish the final value. To calculate drift, the Chromeleon software uses a method of least squares and calculates a regression line from the measured values. The slope of the regression line indicates the drift of the measured signal, and the amount of the slope indicates the amount of drift (Figure 2 and 3).

3.7 Injector Precision

3.7.1 Theory

Injector precision is determined by performing ten replicate injections of a standard using a fixed sample volume.

Conductivity Detectors: A 50 ppm nitrate standard is injected when using conductivity detection. Deionized water is used as the eluent at a flow rate of 0.010 mL/min and with conductivity detection.

Electrochemical Detectors: A 10 μ M Threonine standard is injected when using ICS-5000/ICS-5000⁺ ED detector in Integrated Amperometry mode. 15 mM of electrolytically generated KOH is used as eluent at a flow rate of 0.010 mL/min. A 0.4 mm x 50 mm CarboPac PA20 (P/N 072073) guard column, an Amino Acid disposable gold working electrode (Thermo Fisher Scientific P/N 060082) and a 1 mm PTFE cell gasket (Thermo Fisher Scientific P/N 072161) are used with the Amino Acid (pH using Ag/AgCl Reference electrode) waveform. Please note that the ED Capillary precision specification is 3.0 %.

3.7.2 Performing the Test and Evaluating the Results

The Injector precision evaluation is the next test in the sequence queue and the samples are called Inj Precision_1 through Inj Precision_10 in the XQ_CIC_Detector_Inj_Precision sequence, where X is O for OQ, P for PQ, and Detector is CD for conductivity detectors and ED for amperometry detectors. The concentration of the nitrate standard and the sample volume injected is included in the QNT file used for injector precision.

Precision is determined by calculating the relative standard deviation (RSD) of the peak areas for the ten injections.

3.8 Sample Carryover

3.8.1 Theory

Sample carryover is determined by injecting a highly concentrated sample, followed by a blank sample and then a second blank injection. The blanks in this case are deionized water. Nitrate at a concentration of 1000 ppm is injected when this test is performed with a conductivity detector.

Conductivity Detectors: Nitrate at a concentration of 1000 ppm is injected when this test is performed with a conductivity detector.

Electrochemical Detectors: A 10 μ M Threonine standard is injected when using ICS-5000/ICS-5000⁺ ED detector in Integrated Amperometry mode. 15 mM of electrolytically generated KOH is used as eluent at a flow rate of 0.010 mL/min. A 0.4 mm x 50 mm CarboPac PA20 (P/N 072073) guard column, an Amino Acid disposable gold working electrode (Thermo Fisher Scientific P/N 060082) and a 1 mm PTFE cell gasket (Thermo Fisher Scientific P/N 072161) are used with the Amino Acid (pH using Ag/AgCl Reference electrode) waveform.

Ideally, the chromatogram of the two blank injections should be identical. If, however, a difference is detected, the difference between the peak areas is calculated and the ratio of this difference to the highly concentrated sample is calculated. Deionized water is used as the eluent at a flow rate of 0.010 mL/min for conductivity detectors.

3.8.2 Performing the Test and Evaluating the Results

Sample carryover evaluation is included in the same sequence as the Injector Precision test of the sequence queue and the samples are called Water, Carryover and Water_CO in the *XQ_CIC_CD_inj_precision_CO* sequence.

A peak area ratio is calculated between the blank and high concentration sample to determine sample carryover.

3.9 Detector Linearity

3.9.1 Theory

Conductivity Detector: Detector linearity for conductivity detectors uses a sequence of five nitrate standard injections at 5, 10, 25, 50, and 100 ppm. Deionized water is used as the eluent at a flow rate of 0.010 mL/min. Note that only capillary VWD absorbance detector is supported for capillary ion chromatography.

Electrochemical Detector: Linearity for Integrated Amperometry detectors uses a sequence of five Threonine standard injections at 1, 2, 4, 8, and 10 μ M. 15 mM of electrolytically generated KOH is used as eluent at a flow rate of 0.010 mL/min. A 0.4 mm x 50 mm CarboPac PA20 (P/N 072073) guard column, an Amino Acid disposable gold working electrode (Thermo Fisher Scientific P/N 060082) and a 1 mm PTFE cell gasket (Thermo Fisher Scientific P/N 072161) are used with the Amino Acid (pH using Ag/AgCl Reference electrode) waveform.

3.9.2 Performing the Test and Evaluating the Results

Detector linearity samples are called Detector linearity_1 through Detector linearity_5 in the *XQ_CIC_Detector_Linearity* sequence. The concentrations of the standards are included in the QNT file used for detector linearity. It is recommended to enter the actual concentration of the standards from the certification of analysis into the quantification files to achieve the real results.

3.10 Sample Temperature Accuracy of Autosamplers (AS-AP only)

3.10.1 Theory

The sample temperature accuracy mainly depends on the cooling and heating accuracy of the AS-AP autosampler, the insulation of the sample compartment, and the thermal transfer from the carousel to the vial.

3.10.2 Performing the Test

The sample temperature accuracy is determined with the help of an external thermometer. The temperature sensor is placed in a standard polypropylene vial (10 mL or 1.5 mL) filled with water. The carousel cover must be closed during the test. The autosampler temperature is set to a specified temperature. When the set temperature is reached, the sample (water) temperature is recorded at the end of the 30 minute period. Within the 30 minutes, the sample temperature reaches a stable value. The program will keep the carousel from moving during the run. The temperature accuracy is the temperature difference between the sample temperature and the nominal autosampler temperature.

3.11 Sample Preparation Precision (AS only)

3.11.1 Theory

Sample Preparation precision (electrochemical and absorbance detection are not supported) is determined by making three separate injections using a fixed injection volume. Each of the three standards is prepared individually using the autosampler's sample preparation option. The autosampler prepares the three samples by first transferring 15 μ L of a 1000 ppm nitrate standard into three empty sample vials followed by the addition of 1485 μ L of water to each. Injections are then made from each of the sample vials. Deionized water is used as the eluent at a flow rate of 0.010 mL/min. This test is only performed on autosamplers with the sample prep option installed. This test is not supported with capillary ED.

3.11.2 Performing the Test and Evaluating the Results

Sample prep precision evaluation is the next test in the sequence queue and the sequence is called *XQ_CIC_Detector_Samp_Prep*.

Required configuration for running a successful sample preparation experiment:

- AS configured with a 10.0 mL tray (P/N 026374). Note: Sample Prep only supports 10 mL vials.
- System set up with a conductivity detector.

Precision is determined by calculating the relative standard deviation (RSD) of the peak areas from the three vials.

3.12 Column/Compartment TC Temperature Qualification

3.12.1 Theory

The temperature of a column/compartment TC is qualified by measuring the temperature of the column/compartment TC with a calibrated thermometer after allowing the column/compartment and thermometer to equilibrate for a minimum of 15 minutes. The temperature is recorded from the

thermometer display, giving the temperature of the column/compartments TC. The temperature displayed by the front panel and the software is the calculated liquid temperature.

3.12.2 Performing the Test and Evaluating the Results

During the sequence queue the column heater is set to 30 °C and the DC compartment temperature is set to 15 °C. The Column Heater Temperature Qualification is performed after the completion of the sequence queue. The recorded temperature is manually entered into the temperature sheet of the report.

3.13 IC Cube Temperature Qualification

3.13.1 Theory

The temperature of the IC Cube is qualified by measuring the temperature of the column cartridge of the IC Cube after allowing the IC Cube to equilibrate for a minimum of 20 minutes. The temperature is recorded from the thermometer display, giving the temperature of the IC Cube temperature. The temperature displayed by the front panel and the software is the measured liquid temperature.

3.13.2 Performing the Test and Evaluating the Results

The IC Cube temperature is set to 30 °C during the warm up sequence. The Column thermostat Temperature Qualification is performed after the completion of the sequence queue. The recorded temperature is manually entered into the temperature sheet of the report.

3.14 Sample pH Conductivity verification (AS-AP only)

3.14.1 Theory

The sample pH Conductivity qualification is evaluated by checking the pH calibration and pH accuracy. The sample temperature calibration is measuring the pH at 4, 7 and 10. The reading is stable if the reading and the three data points are within 0.2 pH unit of each other. The pH measurement of the accessory must be calibrated before you begin using the accessory and then periodically thereafter. During pH calibration, the pH of a pH 7 buffer solution is measured first and *the offset is determined*. Then, the pH of either a pH 4 or a pH 10 buffer solution is measured and a slope between pH 7 and pH 4 (or pH 10) is determined. Optionally, the pH of a third buffer solution (pH 4 or pH 10) can be measured and a slope between pH 7 and the third buffer determined. The conductivity calibration procedure consists of determining the conductivity zero point of the accessory and then measuring the conductivity of a 1000 µS standard. Chromeleon determines the calibration offset and slope and calculates a new cell constant.

4 Qualification Preparation

Materials required for system qualification depend on the system configuration. Table 6 describes the base kit for several instrument configurations Table 7 lists the kits available to be used with the ICS-5000/ICS-5000⁺ Capillary configurations. Table 8 describes the additional materials required for the OQ/PQ tests.

Changes or additions to a system configuration may require additional materials.

4.1 Required Kits for qualification of Capillary ICS-5000/ICS-5000⁺ units

Before starting the qualification procedure, confirm that all the materials needed are present. If any items from these tables are not present, do not continue with the qualification process until these items are available.

Table 6. Materials/Parts Description for OQ/PQ

Part Description	Part Number	Quantity
OQ/PQ User's Manual Part F Version 7.10 (this document)	065448 Part F	1
Tubing, 0.010" (0.25 mm) ID x 60" (150 cm) length black PEEK <ul style="list-style-type: none"> Connect pump to injection valve, backpressure tubing to detector, etc. 	042690	60" (152 cm)
Red PEEK Tubing 0.005" (0.125 mm) ID x 39" (99 cm) length <ul style="list-style-type: none"> Generate system backpressure and delay volume for detector and injector tests 	044211	39" (99 cm)
Yellow PEEK Tubing (0.003", 0.075 mm ID) <ul style="list-style-type: none"> Generate system backpressure and delay volume for detector and injector tests in Analytical Systems only 	049715	39" (99 cm)
TUBING,.001X.062 X 42INCH,PK,NAT	074572	1
VLV POD ASSY,6 PORT,RHEO,PK	061947	1
25 µL Sample Loop <ul style="list-style-type: none"> AS-DV/Manual injection 	042857	1
100 µL Sample Loop <ul style="list-style-type: none"> AS 	042951	1
<ul style="list-style-type: none"> 3-Way mixing tees Connect eluent lines and eluent bottles 	024314	2

High-Pressure, fittings bolt, 10-32	074449	6
High-Pressure, ferrules double cone 10-32	074373	6
Qualification Labels	050837	6
Certificate of Functional Performance	031423	6
NIST Traceable Caffeine Standards Kit <ul style="list-style-type: none"> • Absorbance detectors 	060253	1
NIST Traceable Nitrate Standards Kit <ul style="list-style-type: none"> • Conductivity detectors 	060254	1
TUBE,PCUT,,.0025X.062X8.9,PK,BLUE	074484	2
ASSY,KIT,VIAL,AS,10ML,CAPS,SEPTUM,X2	074731	1
PROD,DE,Au,AAA,PK 3,1GSKT,TSTD	060082	1
GSKT,REF ELEC,AG/AGCL,CAP	072162	1
Test Binder	035863	1
Restrictor Microbore, Gradient Test	049630	1

Table 7. Required kits for specified ICS-5000/ICS-5000⁺ capillary systems

Part Description	Part Number
ICS-5000/ICS-5000 ⁺ OQ/PQ Kit Capillary CD (and/or Optical Detectors)	074735
ICS-5000/ICS-5000 ⁺ OQ/PQ Kit, Hybrid CD (Analytical and Capillary CD)	074802
ICS-5000/ICS-5000 ⁺ OQ/PQ Kit (Capillary ED)	074736
ICS-5000/ICS-5000 ⁺ OQ/PQ Kit for Hybrid ED (Analytical and Capillary ED)	074803

4.2 Additional Required Materials

Table 8. Additional Materials Needed for Qualifications

Description	Quantity
ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered) <ul style="list-style-type: none"> All eluent, standard, and sample preparation 	2 L
EGC KOH (Capillary) for electrochemical detection in Capillary systems (P/N 072076)	1
Ammeter or digital multimeter (DVM) with milliamp (300 mA scale) setting for OQ/PQ tests using an EG. Must be capable of displaying 0.00 to at least 220 mA. A Fluke 189 or 289 or equivalent DVM will allow for automated EG Current testing. <ul style="list-style-type: none"> Eluent Generator current test 	1
Column Thermostat PQ Kit (P/N 5705.0050A)	1
Type K temperature sensor for P600 thermometers (P/N 6820.0010)	1
Thermo Fisher Scientific capillary conductivity cell (detector) <ul style="list-style-type: none"> Detector, injector and gradient pump tests using conductivity detectors in conductivity mode 	1
AS autosampler vials- 10 mL vials are required for AS Sample Preparation Qualification and for capillary IC Flow Accuracy Test (P/N 074731)	2
AS-DV autosampler vials (P/N 038141) if qualifying as AS-DV autosampler	20
Blue PEEK Tubing (0.0025 in, 0.0063 mm ID 8.9" 22 cm length P/N 074484) Required for Capillary IC Flow Accuracy Test	2
100 mL volumetric flasks (Class A, 100 ±0.08 mL) <ul style="list-style-type: none"> Standard preparation for AS-DV and Manual Injection 	5
1000 mL volumetric flasks (Class A, 1000 ±0.3 mL) <ul style="list-style-type: none"> Eluent preparation 	At least 1
Transfer Pipettes or Pipettor	0.5 – 10.0 mL
Calibrated Analytical Balance - capable of mass recording to the nearest milligram or better <ul style="list-style-type: none"> Weigh KCl for eluent preparation Weigh eluent for the Flow Rate Accuracy and Precision test 	1
Calibration and Check Standard pH 4 (P/N 033238)	1
Calibration and Check Standard pH10 (P/N 033240)	1
1000 uS Standard (P/N 080172)	1
Computer workstation running Chromeleon™ Version 7.10 (with SR1) or later (later versions required for some configurations, see Chapter 1.6)	1

Deionized water should be vacuum filtered through a 0.2 or 0.45 µm porous nylon filter or other water compatible membrane exhibiting very low extractables.

Please note that if the AS or AS-AP transfer line is not long enough to reach the second IC Cube, a six to eight inch length of black PEEK tubing can be added to the transfer line. It is necessary to then recalibrate the volume of the transfer line.

All volumetric ware, pipettes and standard eluent storage bottles should be made of low extractable polymeric materials such as polypropylene or polystyrene.

4.2 Preparation of Standards

Two standards are provided in the OQ/PQ kit. Use the Nitrate kit, P/N 060254 to qualify systems with a conductivity detector.

To qualify an ICS-5000/ICS-5000⁺ capillary system with an ED detector in Integrated Amperometry mode use the Threonine standards (P/N 063542) for ED qualification (Thermo Fisher Scientific P/N 074737). ED detectors qualification does not support AS-DV autosamplers.

4.2.1 Nitrate standard preparation for the Injector Precision test (AS-DV and manual injections only)

For the Injector Precision test prepare a 50 ppm nitrate standard using the 1000 ppm nitrate standard vials included in the Nitrate Standards Kit.

Directions: Pipette 5.0 mL of the 1000 ppm nitrate standard into a 100 mL volumetric flask. Fill the flask to the mark with ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered).

For the Carryover test use the included 1000 ppm nitrate standard found in the nitrate standards kit without dilution.

4.3 Preparation of Eluents

Only ASTM Type I (or better) deionized water (18.0 Megohm-cm, filtered) is used as eluent and should be freshly prepared.

4.3.1 Preparation of Eluent for use with Integrated Amperometry Detection in Capillary systems

EGC KOH (Capillary) (P/N 072076) and CR-ATC (P/N 072078) are used to qualify the ED Capillary system.

For Flow Test (initial Flow test) and all other Tests.

Eluent	Description
A	Deionized water

Remember to ...

- Degas the deionized water.
- Set EGC KOH (Capillary) concentration at 15 mM and turn the CR-ATC on.
- Use the disposable AAA Gold working electrode (P/N 060082).

4.4 Preparing the IC System

1. Open the Chromeleon Instrument Configuration application. Only one pump, one injection valve and one detector can be qualified concurrently, so if more than one pump, one injection valve or one detector are installed, the second pump (Pump_2), the second detector (CDet_2, EDet_2) will be ignored if both detectors are the same. The detector names can be configured in the Instrument Configuration.
2. Make note of any changes made to the system for the purposes of this qualification. After qualification these changes should be reversed. It is helpful to save a backup file of the configuration before you make any changes for qualification.
3. **AUTOSAMPLER:** Double click on the Autosampler to be qualified (if installed) to view the properties page for the autosampler. Select the **General** tab and confirm the Device Name is "Sampler". Select the **Options** tab and confirm that the options match the configuration of your system. The AS-AP injection mode must be selected at this point. Only one mode is qualified. Select the **Devices** tab and confirm the Autosampler diverter valve status matches your system. Exit the properties page. For the AS-AP autosampler in pull mode, the Flush Volume and Flush Volume2 must be set with F8 commands or from the **More Options** page of the AS-AP panel before beginning qualification. Flush Volume = 2 x (Needle Volume + Needle Line Tubing Volume) μ L where Needle Volume = 30 μ L and Needle Line Tubing Volume (the volume of tubing between the needle and the injection port) = 0.86 μ L/cm (2.2 μ L/in) of 0.33-mm (0.013-in) ID PEEK tubing (blue). Set the Flush Volume2 parameter to 30 μ L.

NOTE: Make sure inject mode in the panel is consistent with the inject mode in the Instrument Configuration before loading the OQPQ templates. In addition, the temperature control must be checked in the Instrument Configuration and the panel in order to run the sample temperature accuracy test in the sequence.

NOTE: It is recommended to degas the AS-AP wash when qualifying with a capillary system. The user can refer the degas procedure to the AS-AP user manual.

4. **ICS-5000 DC:** Double click on the DC to be qualified to view the general properties. Select the Thermal Controls tab and confirm the heating configuration of the Timebase. Confirm that the device name for the Column TC is "Column_TC" and the device name for the Compartment TC is "Compartment_TC" if installed on the Timebase.
5. **DETECTOR:** Double click on the detector to be qualified to view the properties page for that detector (for CD and ED this will be in the **DC** under the **Detector** tab). Open the properties of the DC and click on the **Detectors** tab. Highlight the detector name and click on **Configuration** button. Confirm that the Device Name is "CDet1" for CD detectors or "EDet1" for ED detectors. If both detectors are in the same Timebase, uncheck the detector(s) that is not being qualified (only one detector can be qualified at a time). Click on the **Signals** tab. Confirm the signal name is "CD_1" for CD detectors or "ED_1" for ED detectors. Confirm that the signal factor for conductivity detectors is 1.000. For integrated amperometry detectors confirm the signal factor is 0.001. If it is not, change the factor to the correct value for the qualification. Confirm the signal unit is μ S for conductivity or nC integrated amperometry respectively. If it is not, change the unit to the correct value for the qualification. Exit the properties page.
6. If applicable, ensure that the detector Output is in 'Offset' mode in the Analog Out Set-Up screen.
7. Make note of any changes (to be documented in the qualification report) and save the configuration.
8. All modules must have their serial number set in the moduleware. Qualification of modules without serial numbers is not supported.

9. Plumb the Eluents A to the pump and if additional eluent lines are present, then prime each eluent line individually for approximately 10 minutes each.
10. Use the 115 mm blue peek tubing (P/N 072189) from the IC Cube Tubing kit (P/N 072186) to connect from the 4-port valve to the detector.
11. Use the backpressure coil (P/N 074572) to connect the pump outlet directly to port 2 on the IC Cube injection valve. Bypass the column, suppressor, CR-TC, and CRD from the system; if an Eluent Generator Cartridge is in-line with the IC system, bypass all plumbing to the EGC and connect the pump directly to the 4-port valve.

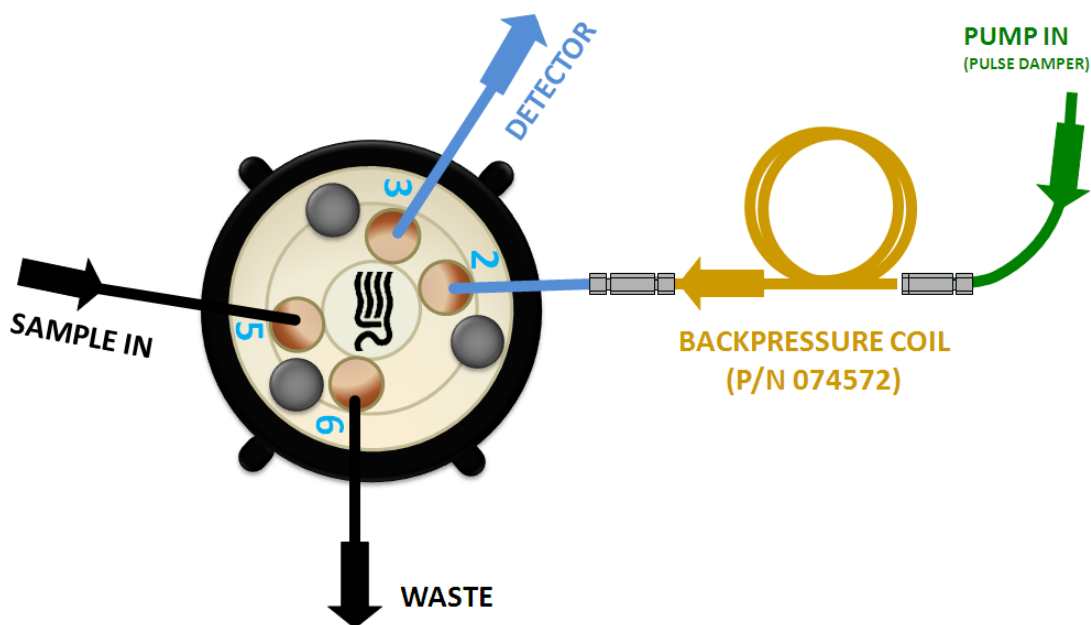


Figure 5. Plumbing diagram, initial equilibration step in a capillary system

12. ICS-5000/ICS-5000[±] DC COLUMN TC: Insert the thermometer probe horizontally into the 2nd tubing slot from the bottom of the TC so that the LCD display sits out the side of the column TC. The LCD display of the thermometer should be visible and display the current Column TC temperature.
13. ICS-5000/ICS-5000[±] IC Cube COLUMN TC: Insert the thermometer probe horizontally into the tubing slot of the IC Cube column cartridge so that the LCD display sits out the side of the IC Cube. The LCD display of the thermometer should be visible and display the current IC Cube Column TC temperature.
14. ICS-5000/ICS-5000[±] DC COMPARTMENT TC: Insert the thermometer probe horizontally into top tubing slot on the DC so that the LCD display of the calibrated thermometer sits out the side of the compartment. The LCD display of the thermometer should be visible and display the current Compartment TC temperature.
15. Thoroughly rinse the autosampler and/or manual injector fluidics with water. This can be accomplished on the AS and AS-AP Autosampler by flushing the sample syringe six times the sample syringe volume and on the AS-DV by running two or three 5.0 mL rinse vials.

16. AS: For ICS-5000/ICS-5000⁺ Capillary systems using an AS or autosampler, a 4-port injection valve in the DC with an internal loop of 0.4 μL is used. From the front panel of the AS, press to the **Main Menu** button, select **Module Set-Up** and then **Plumbing Configuration**. Ensure that the sample loop size is adjusted to 25 μL in the AS front control panel.

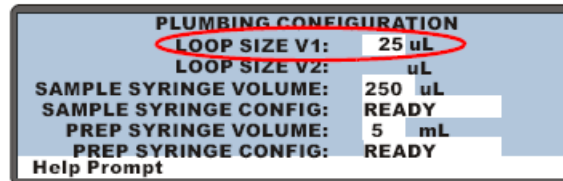


Figure 6. AS Autosampler LCD Control Panel Display.

Ensure that the 'LOOP SIZE V1' volume is changed to 25 μL

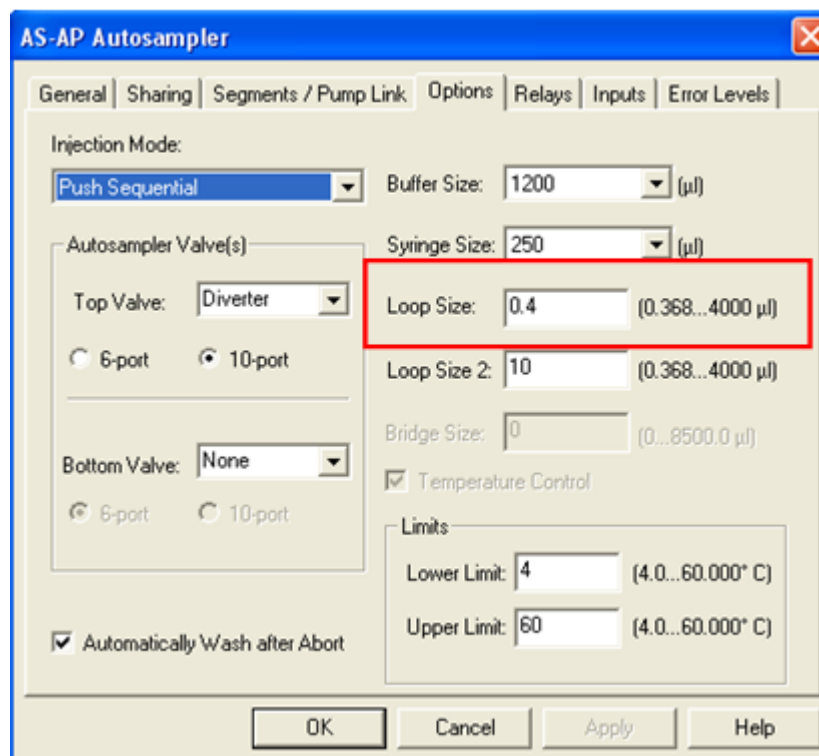


Figure 7. AS-AP Instrument Configuration

17. AS-AP: For ICS-5000/ICS-5000⁺ Capillary systems using an AS-AP autosampler, a 4-port injection valve in the DC with an internal loop of 0.4 μL is used. From the Instrument Configuration of the AS-AP, select the options tab and change the loop size to 0.4 μL . Ensure that the sample loop size is adjusted to 0.4 μL in the AS-AP Instrument Configuration. .

18. For ICS-5000/ICS-5000⁺ Analytical systems using an AS-AP or AS Autosampler, install the 100 μ L sample loop on the injection valve. Loop volume must be set in Instrument Configuration for the AS-AP.
19. AS-DV or Manual Injector: For systems using an AS-DV Autosampler or manual injector connect the transfer line to the 4-port injection valve of the IC Cube.

Note: All programs using the AS-DV are designed to run with the 5.0 mL vials.

1 Nitrate standards kit (P/N 060254) is needed to complete the OQ or PQ.

20. Select 0.010 mL/min and turn the pump flow on and let the system equilibrate for a minimum of 15 minutes.

NEW TUBING: When running a qualification on a system with new tubing, such as a new backpressure coil, new sample loop or a complete new system, it is recommended to rinse the tubing with carbonate eluent before proceeding with the qualification. This can be achieved by doing two or three injections with 0.5 M Sodium Carbonate solution while running deionized water at 0.010 mL/min, or by running 9 mM Sodium Carbonate as the eluent for 10 minutes before rinsing with deionized water for 20 minutes. The latter is recommended for new systems being qualified for the first time.

To prepare 0.5 M sodium carbonate dissolve 0.53 g of sodium carbonate in water to make a total volume of 10 mL of solution. To prepare 9 mM sodium carbonate dissolve 0.955 g of sodium carbonate in water to make 1 L of solution.

When running a qualification on a system configured with electrochemical detection, it is recommended to rinse the system with 1.0 M hydroxide before proceeding with the qualification. To prepare 1.0 M hydroxide, prepare from a 50/50 w/w sodium hydroxide solution (never from hydroxide pellets), by pipetting 52 mL of sodium hydroxide in water to make 1 L of solution.

21. CONDUCTIVITY DETECTOR: Pipette the standards from the standards kit vials directly into autosampler vials and place them in the position / order according to Table 9 or 10 below. For the Injector Precision and Carryover standards (AS-DV Autosampler only) transfer the Injector Precision standards prepared in Section 4.2 into 10 autosampler vials and the Carryover standard into one autosampler vial. To qualify an IC system without an autosampler, use the AS-DV table for the injection order and inject the samples from the standards kit ampoules manually.
22. ICS-5000 INTEGRATED AMPEROMETRY: Cut the label off the 2 mL standards vials (included in the ICS-5000/ICS-5000⁺ ED qualification kit) and place the vial directly into autosampler in the position order according to the table below. Note that even though the main portion of the label has been cut off, the vial is still properly labeled. For the AS-AP autosampler, this will require the 1.5 mL As-AP autosampler tray (P/N 074936) For the AS autosampler this will require the 2 mL AS autosampler tray (P/N 062481).
23. ICS-5000 ELUENT GENERATOR: Double click on the eluent generator to be qualified to view the properties page for the eluent generator. Select the **General** page and confirm the Device Name is "EluentGenerator". On the "cartridge" tab confirm that the eluent generator is linked to the pump to be qualified. Confirm that there is a serial number for a KOH or a MSA cartridge. If a serial number is not available for the KOH or MSA cartridge, contact Thermo Fisher Scientific Technical Support. **Connect the EG test box with the ammeter to the EGC_1 or EGC-2 connector.** The user must rename EGC-2 to EGC-1 in the Instrument Configuration if the Qualification test box is connected to EGC-2.

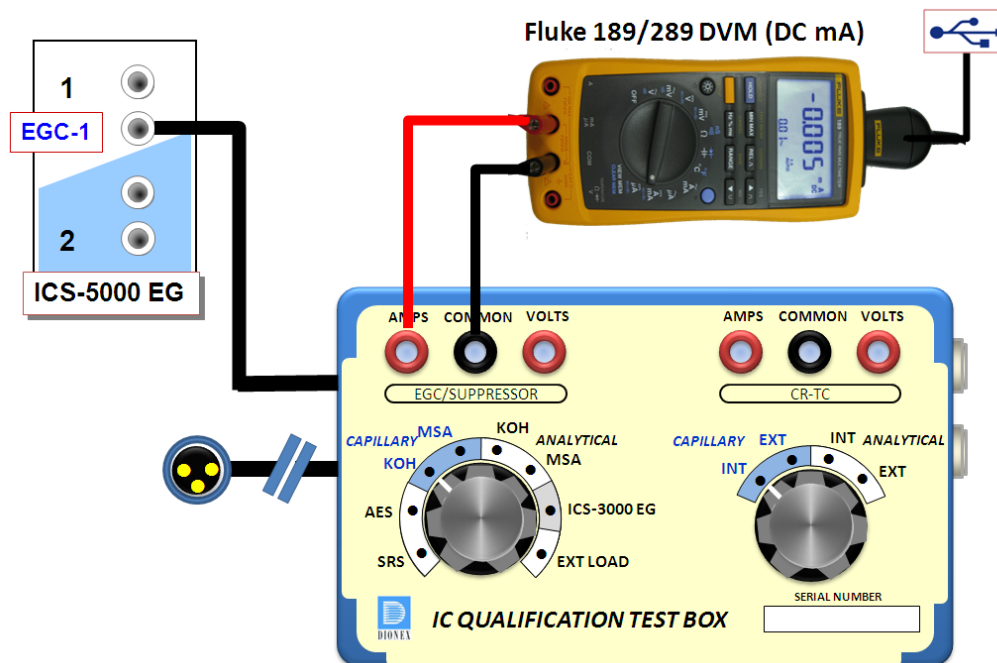


Figure 8. ICS-5000/ICS-5000⁺ EG Current Test Setup

24. For systems using an ICS-5000/ICS-5000⁺ EG:
 - a. Disconnect the Eluent Generator Cartridge cable from the Eluent Generator EGC module at the connector.
 - b. If installed, disconnect the CR-TC cable from the CR-TC connector.
 - c. Turn on the ammeter or multimeter and set the display to DC mA. This test requires mA readings from 0.00 to at least 220 mA.
 - d. Connect the positive lead of the ammeter to the red post connector labeled “Amps” on the IC Qualification Test Box
 - e. Connect the negative lead of the ammeter to the black post connector labeled “COMMON” on the IC Qualification Test Box (P/N 072005).

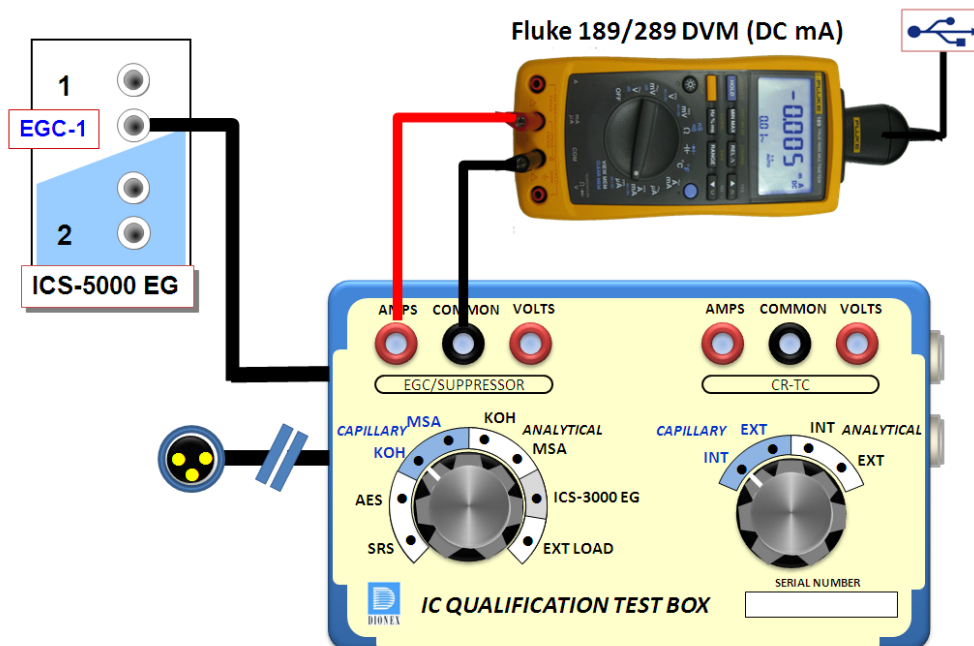


Figure 9. ICS-5000/ICS-5000⁺ EG Current Test Setup

- f. From the Console in the Chromeleon 6.8 software, go to the Navigation Pane and select the pump_EG_detector panel appropriate for the system being qualified. Go to Control and select the Instrument configured with the Eluent Generator to be qualified (click on the Default ePanel).
 - g. From the Eluent Generator section of the panel, select the cartridge type to be used. The panel will show previously configured cartridges even though the IC Qualification Test Box is plugged into the cartridge connector.
25. FLUKE 189/289 MULTIMETER:
- a. Install the Fluke 189/289 multimeter or equivalent into the Instrument to be qualified.
 - b. The Chromeleon device driver for the Fluke 189/289 multimeter is included with Chromeleon 7.1 (Fluke 189, 289).
 - c. Connect the Fluke 189/289 multimeter or equivalent to the IC Qualification Test Box (P/N 072005) as described in the **important notes** of this section.
 - d. The Fluke 189/289 multimeter is only used for systems that will enable automated downloading of EG current qualification results into the validation reports.
 - e. To correctly configure the Fluke 189/289 device driver:
 - i. Connect the Fluke USB/IR (Rev II or greater) cable to the computer USB port; Windows will automatically detect and install the USB driver.
 - ii. Verify through the Windows Hardware Device Manager for the actual COM port assignment.

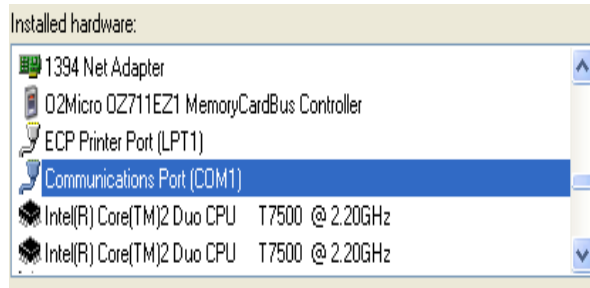


Figure 10. COM Port Assignment

- iii. Configure the Fluke 189 or 289 Chromeleon Device Driver in the Timebase, save the Instrument Configuration file and exit then shut down the Chromeleon Server.
 - iv. Restart the Chromeleon Server and open the Fluke 189/289 CM device driver.
 - v. Select the COM port from the COM drop down selection to match the COM number assigned in the Windows Hardware Device Manager above then save and exit from the Chromeleon Server.
- f. It is recommended that the Power off time out (PrOff) for the Fluke189/289 be set to a value greater than the time it will take to perform the Operational Qualification.
 - g. Ensure that the meter is set to record in 'DC mA' (instead of 'AC mA' the Fluke 289 defaults to AC mA).
 - h. See Fluke 189/289 operator's manual for instructions on these settings.

IMPORTANT NOTES:

- a. It is important that the Fluke 189/289 multimeter is installed AND connected before loading the OQ/PQ templates.
- b. Follow the instructions in the Chromeleon 6.8⁺ help topics for the installation of the Fluke 189/289 multimeter Windows USB driver and for the Chromeleon device driver for the Fluke 189/289 multimeter.
- c. Follow the Fluke 189/289 multimeter instructions for adjusting the default 'PrOFF' setting in the meter. It is recommended to adjust this parameter to a longer time (e.g., several hours or more) to avoid having the meter auto power off during the qualification. In the event that the meter is powered off during the qualification testing the batch sequences will be aborted.

Table 9. AS Autosampler Sample Vial Order

Conductivity Detector with AS	ICS-5000/ICS-5000⁺ Integrated Amperometry Detector with AS	Vial Tray Position for AS
Water	Water	1
5 ppm Nitrate Std		2
10 ppm Nitrate Std		3
25 ppm Nitrate Std		4
50 ppm Nitrate Std		5
100 ppm Nitrate Std		6
1000 ppm Nitrate Std		7
Empty Vial*		8
Empty Vial*		9
Empty Vial*		10
	1 µM Threonine Std	21
	2 µM Threonine Std	22
	4 µM Threonine Std	23
	8 µM Threonine Std	24
	10 µM Threonine Std	25
	10 µM Threonine Std	26

* Vials only need to be present if the detector listed at the top of the table is to be qualified.

* Users need to reconnect the detectors when using the absorbance detectors.

* Don't forget to add the water vial in position 1 for the ED qualification test.

Table 10. AS-DV Autosampler or Manual Injection Sample Order (Conductivity Only)

Qualification	Conductivity Detector with AS-DV	Sample Order on AS-DV
Warm Up	Water	Vial 1
Injector Precision	50 ppm Nitrate Std*	Vial 2
	50 ppm Nitrate Std*	Vial 2
	50 ppm Nitrate Std*	Vial 2
	50 ppm Nitrate Std*	Vial 3
	50 ppm Nitrate Std*	Vial 3
	50 ppm Nitrate Std*	Vial 3
	50 ppm Nitrate Std*	Vial 4
	50 ppm Nitrate Std*	Vial 4
	50 ppm Nitrate Std*	Vial 4
	50 ppm Nitrate Std*	Vial 5
Carryover	1000 ppm Nitrate Std	Vial 12
	Water	Vial 13
	Water	Vial 14
Detector Linearity	5 ppm Nitrate Std	Vial 15
	10 ppm Nitrate Std	Vial 16
	25 ppm Nitrate Std	Vial 17
	50 ppm Nitrate Std	Vial 18
	100 ppm Nitrate Std	Vial 19

* AS-DV users: Do not use the standard from the standards kit for the Injector Precision and; instead, use the standards prepared in Section 4.3 of this manual.

* AS-DV users: ICS-5000/ICS-5000⁺ Integrated Amperometry is not supported.

Table 11. AS-AP Autosampler Sample Order

Conductivity Detector with AS-AP Push/Pull Mode	Absorbance Detector with AS-AP Push/Pull Mode	Electrochemical Detector with AS-AP Push Mode	Vial Tray Position
Water	Water		BA1
5 ppm Nitrate Std	15 ppm Caffeine Std		BA2
10 ppm Nitrate Std	40 ppm Caffeine Std		BA3
25 ppm Nitrate Std	60 ppm Caffeine Std		BA4
50 ppm Nitrate Std	80 ppm Caffeine Std		BA5
100 ppm Nitrate Std	100 ppm Caffeine Std		BB1
1000 ppm Nitrate Std	1000 ppm Caffeine Std		BB2
		Water	RA1
		1 µM Threonine Std	RA2
		2 µM Threonine Std	RA3
		4 µM Threonine Std	RA4
		8 µM Threonine Std	RA5
		10 µM Threonine Std	RA6
		10 µM Threonine Std	RA7

Note: AS-AP pull mode requires the use of 10 mL vials. Qualification with ED detection is not supported with AS-AP pull mode.

Note: To run sequences with 1.5 mL vials, it is important to set the puncture offset to 3 mm in the commands before you start. The puncture offset commands allows the AS-AP to realign the x-axis of the 1.5m/10 mL vial setting.

4.5 OQ/PQ Sequence Installation

For performing OQ and PQ checks in Chromeleon 7.1, it is not required to create and copy the sequence templates from a Chromeleon CD. An Instrument Qualification Wizard automatically performs these steps. The wizard creates the sequences to be run. No instrument-specific sequence templates are created.

If using an AS autosampler configured for sequential injections, manually put the diverter valve in the correct position for the Instrument.

Tip: Please note that Chromeleon 7 terminology is different from the terminology used in Chromeleon 6.80. For details, refer to the 'Glossary - Chromeleon 7,' which is available in the Documents folder of your Chromeleon 7 installation.

Go to Tools → Instrument Qualification to open Instrument Qualification Wizard. For this step, Chromeleon 7.1 must display the instrument pane with the system to be tested selected (Figure 9) otherwise the error "Instrument not connected" will occur.

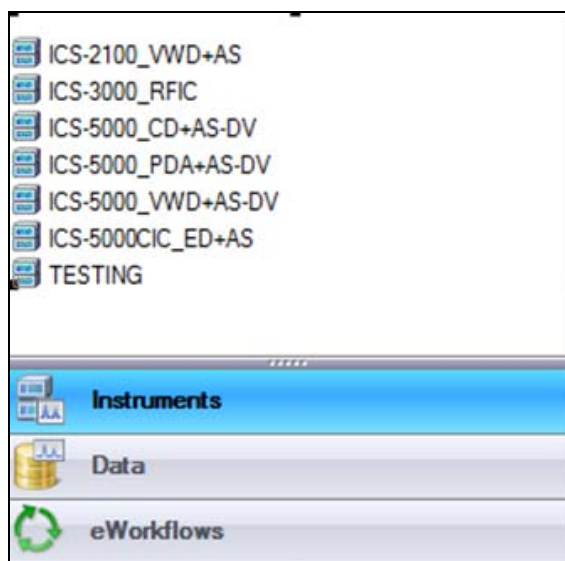


Figure 11. Instrument Pane for Chromeleon 7.1

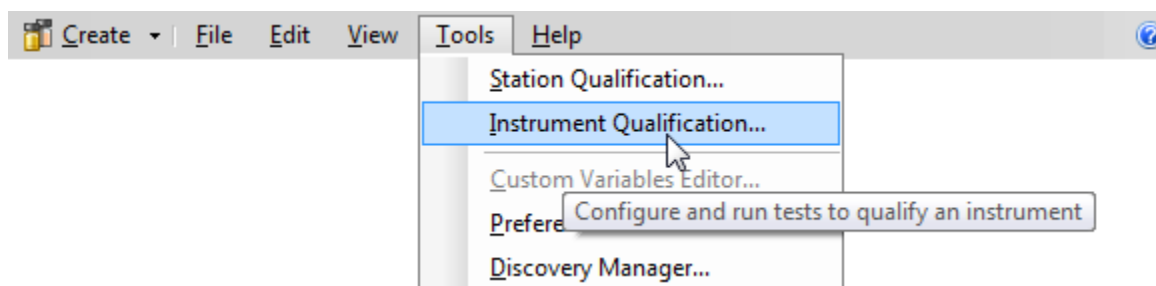


Figure 12. Starting the Instrument Qualification Wizard for Chromeleon 7.1

Select the qualification type: Installation (qualification of the installation), Operational (qualification in the working environment), or Performance Qualification (qualification during routine operation)

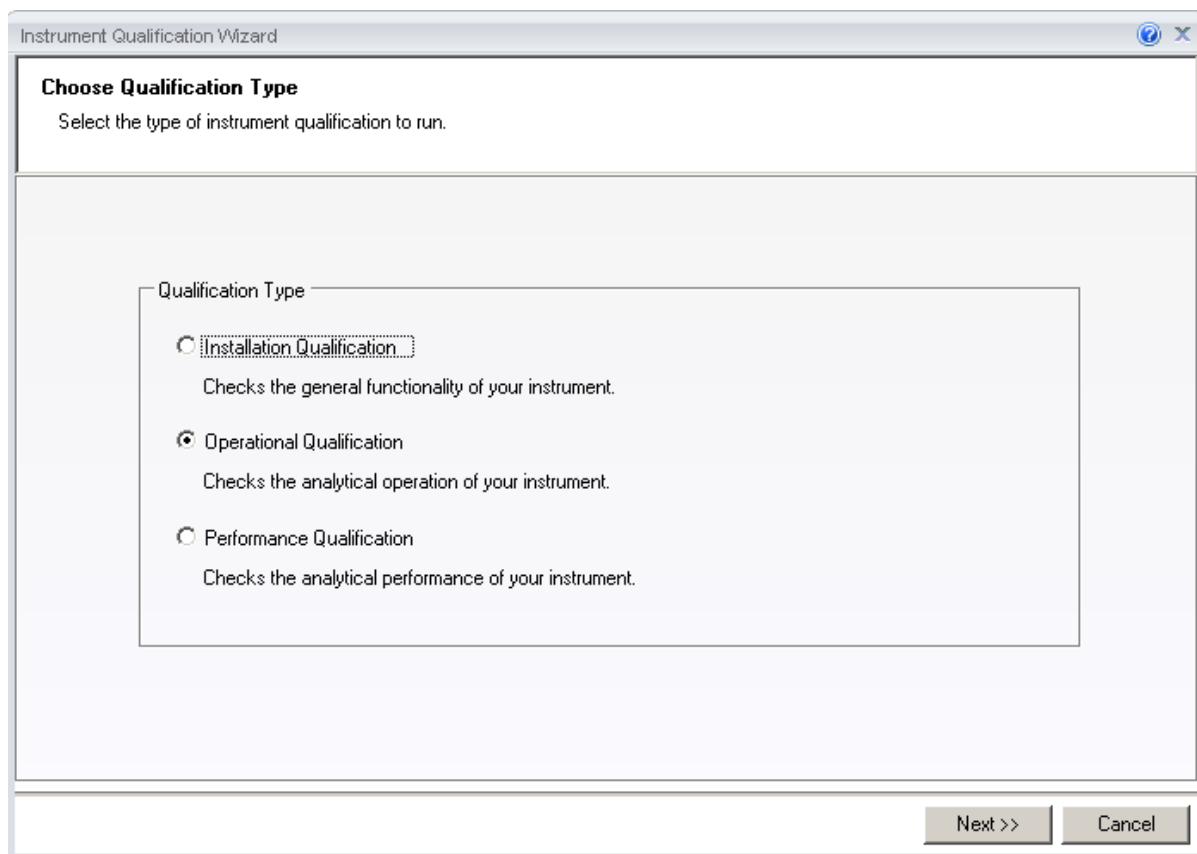


Figure 13. Selecting the qualification type for Chromelon 7.10

Select the instrument to qualify.

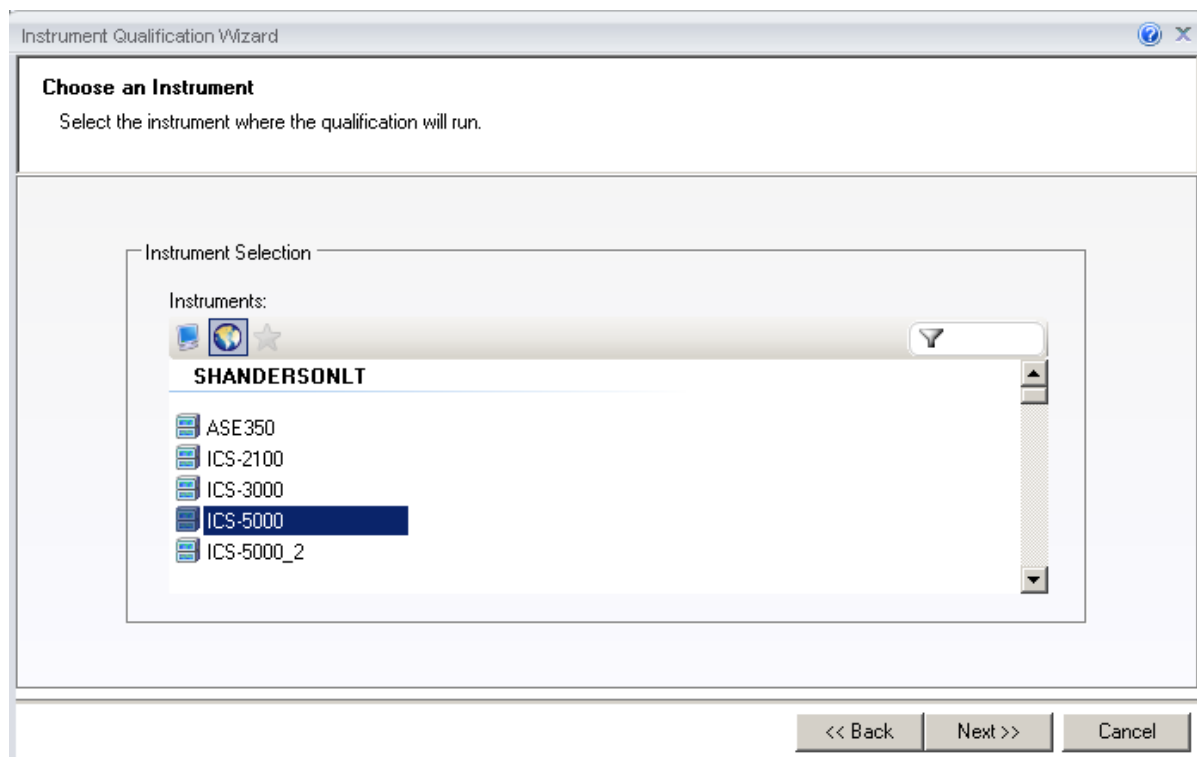
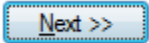


Figure 14. Selecting the instrument

Click  to connect the selected instrument to Chromeleon.

A list of sequences (tests) is displayed. The list is adapted to the instrument configuration of the selected instrument as defined in the Chromeleon Instrument Configuration Manager.

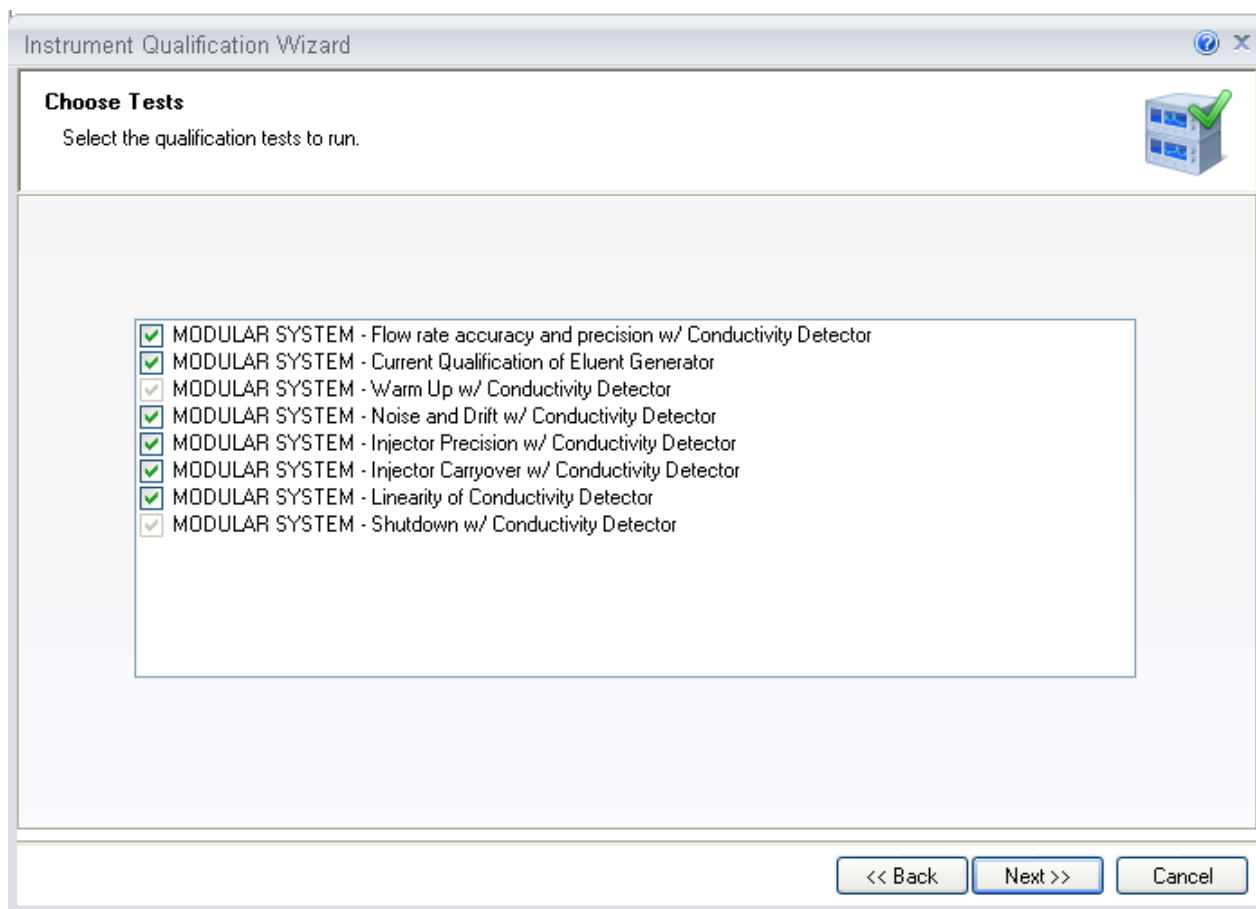


Figure 15. List of sequences for the selected instrument

Select the sequences that needed for the tests to be performed. Mandatory tests, such as **"Warm up"**, are shown in the list, but the selection cannot be changed.

On the last wizard page, select a unique name under which the OQ and/or PQ sequence directory for this instrument is saved.

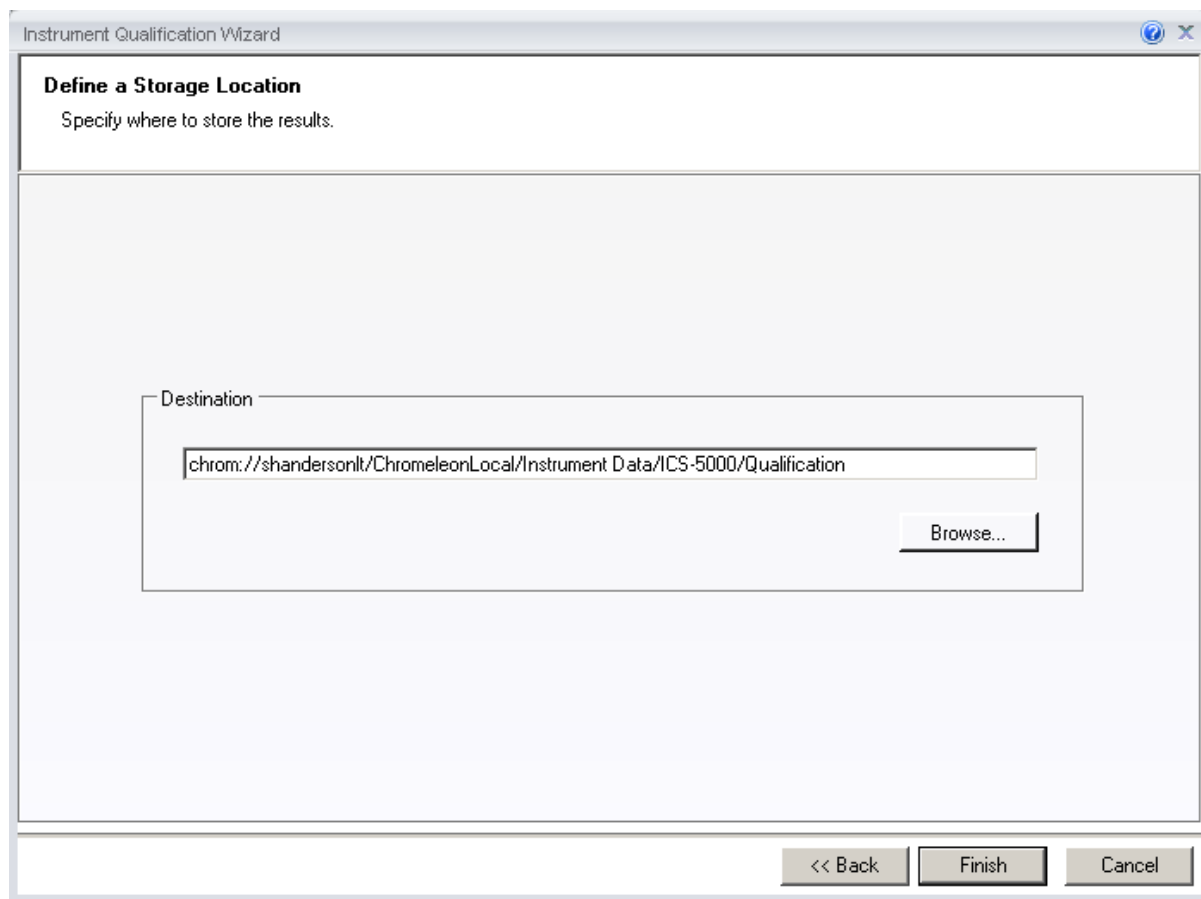


Figure 16. Selecting the storage location

4.6 OQ/PQ Sequence Execution

When the qualification wizard has been completed, the selected sequences are created and are automatically added to the instrument queue. A progress window shows which steps have been performed:

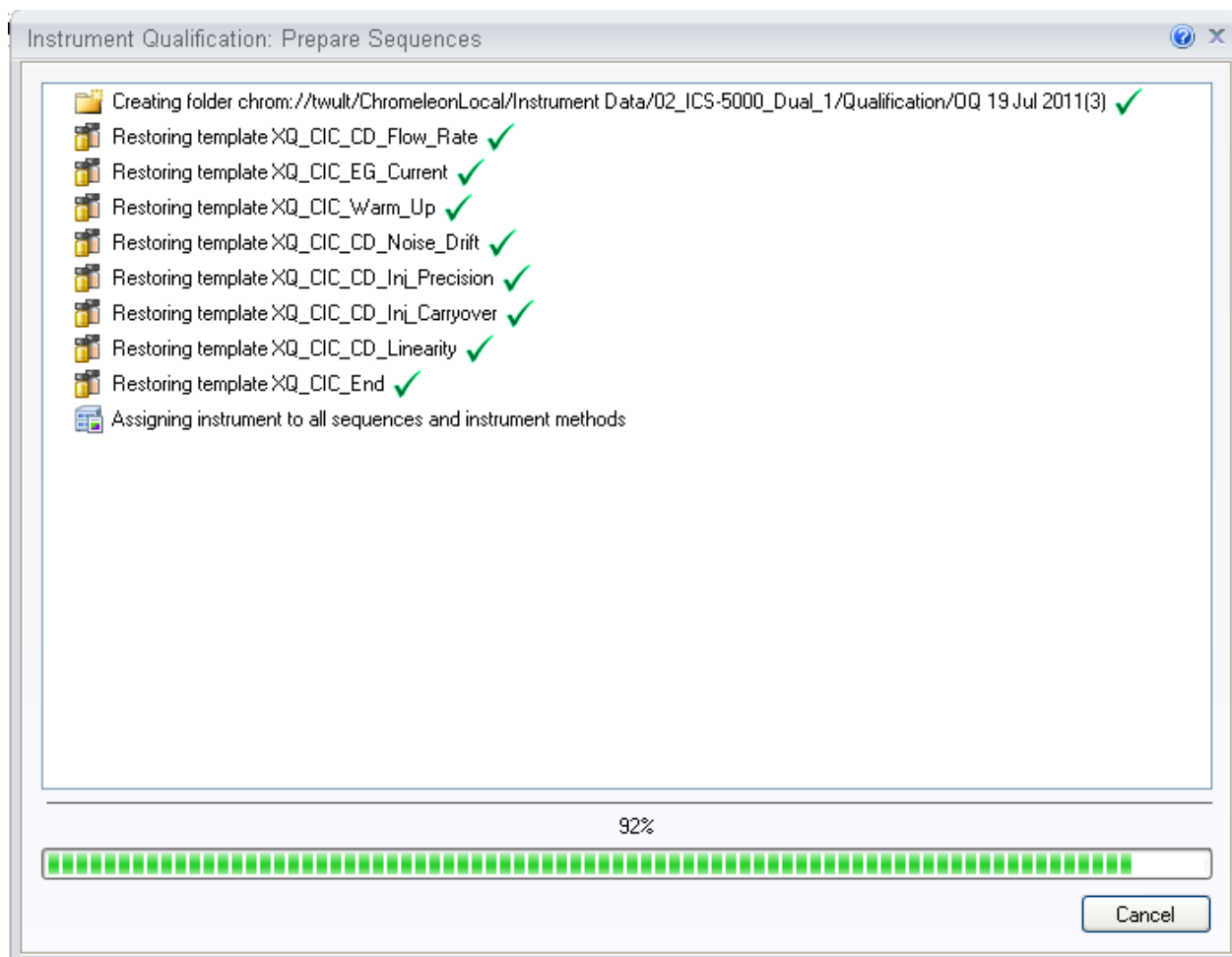


Figure 17. Progress during sequence creation

The Instrument View dialog box will open, displaying the Queue tab. As soon as the queue is started, Chromeleon runs the sequences. The wizard should copy the sequences to the destination selected then auto-populate the instrument queue. If the error “Instrument not connected” appears, make certain that the Chromeleon 7.1 must be displaying the instrument pane with the system to be tested.

The OQ/PQ qualification tests are performed in the following order:

- Flow Rate Test
- Eluent Generator Current Test (only if an EG is present)
- Warm Up
- Wavelength Accuracy

- e. Detector Noise and Drift Test
- f. Injector Precision Test
- g. Carryover Test
- h. Detector Linearity Test
- i. Injector Linearity Test (only if an AS or AS-AP is present)
- j. Sample Preparation Precision Test (only if an AS is present with the Sample Prep Option and the 10 mL Tray Type installed with the conductivity or the absorbance detector)
- k. Gradient Accuracy Test (only if an analytical gradient pump is present)
- l. Column TC and/or Compartment TC
- m. Sample Temperature Accuracy of Autosamplers (only for AS-AP Autosamplers)

Note: Test 'a' and 'l' is done manually and does not require a sequence template. The Instrument Method will pause before the Eluent Generator Current Qualification Test and the Flow-Rate Accuracy and Precision Test as these tests require user intervention.

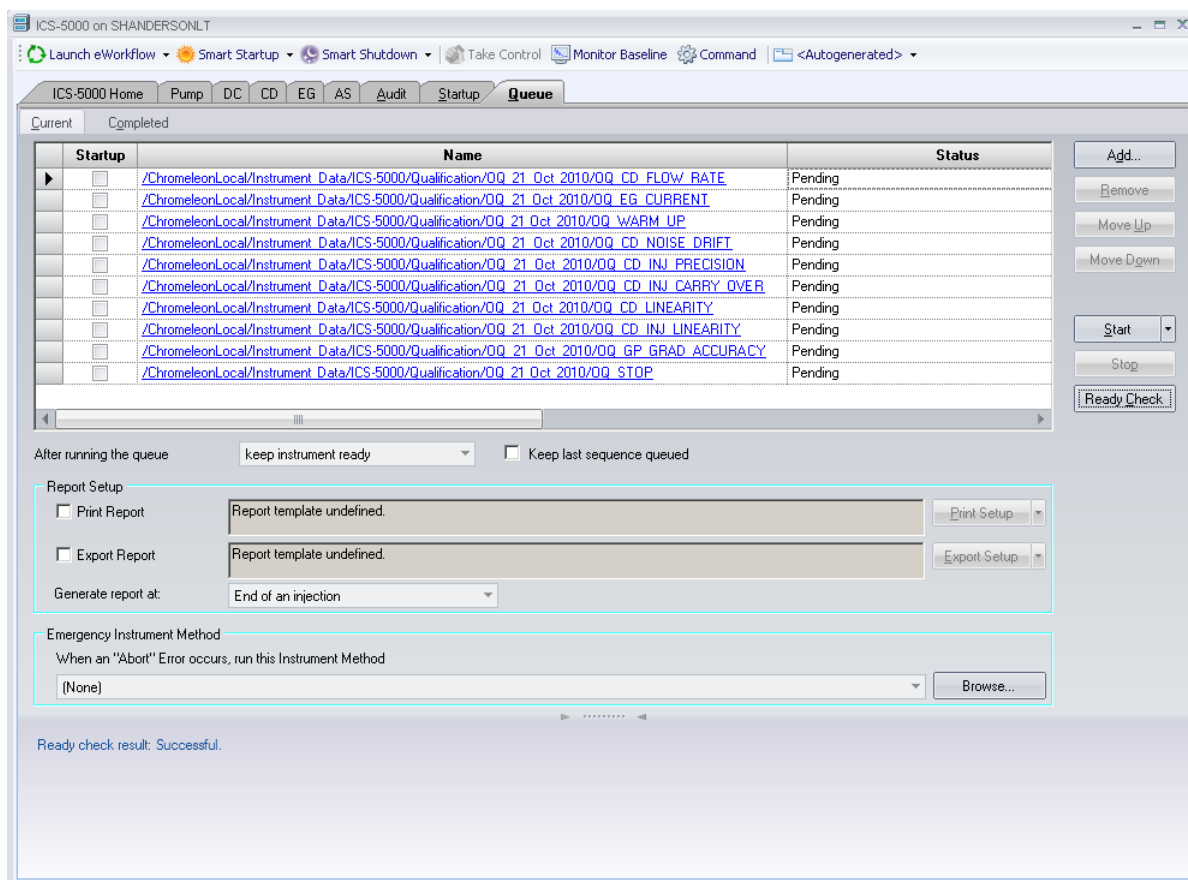


Figure 18. Sequence queue for Chromeleon 7.1

4.7 Evaluating the Test Sequences

The qualification sequences are saved under the path that was selected in the wizard. Each sequence also includes a separate report template.

To edit the report, open the report and remove the protection (if any) from the SPECIFICATION sheet. Enter the following information:

Name of the customer and name of the tester

Sample information such as batch number, expiration date, and actual concentration of the standard.

Name of the item that is used to generate the backpressure [default: capillary (L: 15 m; ID: 0.18 mm)]

Do not change any of the other report sheets. The report contains many references between data sheets. If you insert or delete lines and columns, these references will be lost and the calculations will be wrong.

To make sure that Chromeleon reads and processes the data in the report correctly, always print the report from the Chromeleon Console.

In the Data category, right-click the sequence for which you want to print the report, and then click "Print Report".

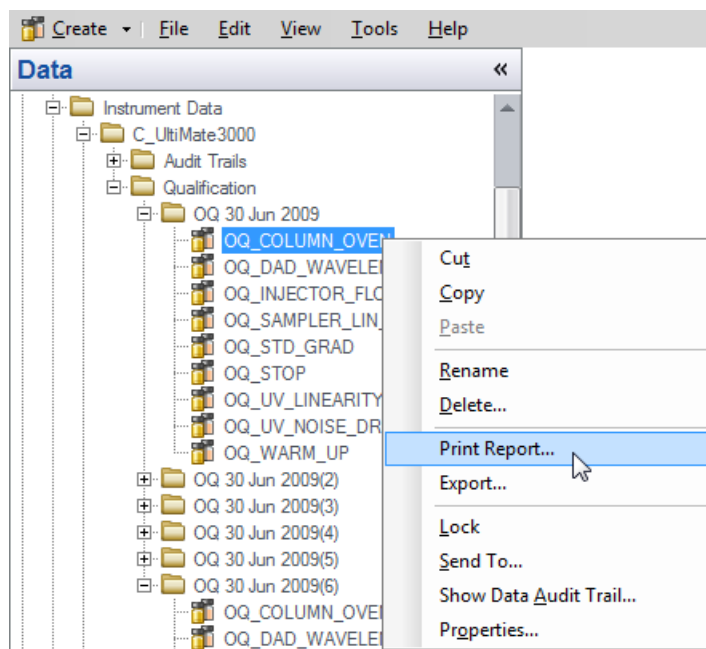


Figure 19. Selecting Print

Note: For ICS-5000/ICS-5000[±] Integrated Amperometry System: Before executing the sequences, first turn on the pump flow and the cell current. Click on the “Default Panel Tabset”. From the pump tab, start the pump flowing 50 mM NaOH at 0.25 mL/min. Click on the **EC detector tab**, Select Cell mode of “IntAmp”, then click on the **Waveform** button, use the drop down menu to choose the waveform **Amino Acids (pH/Ag/AgCl Reference)**, then click on the **Exit** button. Turn the cell voltage to **On**.

Note: If an Eluent Generator is configured on the system, the following message may occur at the Ready Check: “Flow rate not specified at time 0.0, using a flow setting 1.00.” This refers to the Eluent Generator flow rate, not the pump flow rate. It is not necessary to take any action since the Eluent Generator is not used for this part of the qualification. Proceed with starting the batch.

Note: For systems with a shared autosampler there might be a number of *informational* messages indicating that “Exclusive Access” has not been acquired or released. This is normal and occurs because the qualification does not release the autosampler until the qualification is complete.

Repeating Individual Tests

It may be necessary to repeat individual tests. If so, review Section 6 where problems are described according to which individual tests might fail. According to Good Laboratory Practice (GLP), all tests following the one that failed must be repeated. The entire OQ/PQ qualification is designed such that almost of the performance tests require that the previous one to have passed successfully.

For example, if the test regarding the linearity of a CD detector fails, the results regarding the linearity of the injector are questionable as the linearity of the detector is a prerequisite for testing the injector linearity.

Table 12. Sequence File Names and description

Sequence Name	Check
XQ_Flow_Rate	Pump flow-rate accuracy and precision tests for ICS-3000 or ICS-5000/ICS-5000 ⁺ pumps
XQ_EG_Current	Current qualification of an EG Eluent Generator
XQ_EG_Dual_Current	Current qualification of an Dual EG configuration (ICS-5000/ICS-5000 ⁺ with both EGC configured on one Instrument)
XQ_CD_Noise_Drift	Noise and drift test for conductivity detectors
XQ_DC_Amp_Noise_Drift	Noise and drift test for DC amperometry detectors
XQ_Int_Amp_Noise_Drift	Noise and drift test for integrated amperometry detectors
XQ_PDA_Noise_Drift	Noise and drift test for photodiode array detectors
XQ_UV_Vis_Noise_Drift	Noise and drift test for absorbance detectors (excluding PDA detectors)
XQ_Wavelength_Accuracy	Wavelength accuracy test for the VWD and PDA absorbance detectors
XQ_Abs_Inj_Precision	Injector precision test for systems with an absorbance detector
XQ_Abs_Inj_Carryover	Injector carryover tests for systems with an absorbance detector
XQ_CD_Inj_Precision	Injector precision test for systems with an conductivity detector
XQ_CD_Inj_Carry_Over	Injector carryover test for systems with an conductivity detector
XQ_ED_Warm_Up	Warm-up sequence for ICS-5000/ICS-5000 ⁺ systems with ED detector
XQ_ED_Noise_Drift	Noise and drift test for ICS-5000/ICS-5000 ⁺ amperometry detectors (ED in Integrated Amperometry mode)
XQ_ED_Inj_Precision	Injector precision for ICS-5000/ICS-5000 ⁺ systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Carryover	Injector carryover for ICS-5000/ICS-5000 ⁺ systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Linearity	Detector Linearity for ICS-5000/ICS-5000 ⁺ systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Inj_Linearity	Injector linearity for ICS-5000/ICS-5000 ⁺ systems with amperometry detector (ED in Integrated Amperometry mode)
XQ_ED_Stop	The sequence turns off the pump and cell for ICS-5000/ICS-5000 ⁺ systems with amperometry detector (ED in Integrated Amperometry mode).

XQ_Abs_Linearity	Detector linearity test for absorbance detectors
XQ_CD_Linearity	Detector linearity test for conductivity detectors
XQ_Abs_Inj_Linearity	Injector linearity test for systems with an absorbance detector
XQ_CD_Inj_Linearity	Injector linearity test for systems with an detector
XQ_Abs_Samp_Prep	Injector sample preparation precision test for systems with an absorbance detector
XQ_CD_Samp_Prep	Injector sample preparation precision test for systems with an conductivity detector
XQ_GP_Grad_Accuracy	Gradient accuracy and noise test for systems with a gradient pump
XQ_CD_Noise_Drift	Noise and drift test for conductivity detectors
XQ_DC_Amp_Noise_Drift	Noise and drift test for DC amperometry detectors
XQ_Int_Amp_Noise_Drift	Noise and drift test for integrated amperometry detectors
XQ_PDA_Noise_Drift	Noise and drift test for photodiode array detectors
XQ_UV_Vis_Noise_Drift	Noise and drift test for absorbance detectors (excluding PDA detectors)
XQ_Stop	The sequence slows down the flow rate to 0.25 mL/min to conserve eluent for overnight runs

5 Performing the Qualification

5.1 OQ/PQ Test Order

The OQ/PQ qualification tests are performed in the following order:

1. Flow Rate Accuracy and Precision Qualification
2. Eluent Generator Current Qualification Test
3. Warm Up
4. Wavelength Accuracy Test (only for VWD absorbance detectors)
5. Detector Noise and Drift Test
6. Injector Precision Test
7. Carryover Test
8. Detector Linearity Test
9. Sample Preparation Precision Test (only for AS Autosamplers with the Sample Prep Option and 10 mL Tray Type installed with conductivity detectors)
10. Column TC and/or Compartment TC
11. IC Cube Temperature Qualification
12. Injection Temperature Accuracy
13. Sample Temperature Accuracy of Autosamplers (only for AS-AP autosamplers)

Table 13. OQ/PQ Test Timetable

Test	Method	Material Required	Equilibration Time	Approx. Run Time
Flow Rate Accuracy and Precision All pump models	Sequence	Eluent (water) Backpressure loop	15 min	52 min
Conductivity Noise / Drift	Sequence	Water flow rate 1.0 mL/min	5 min	20 min (after 20 minute warm up)
Int. Amperometry Noise / Drift	Sequence	Eluent (15 mM OH)	5 min	20 min (after 60 minute warm up)
AS-DV, Manual Injector Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity	Sequence	Eluent (water) Standards Backpressure loop	10 min	1 hr, 10 min
AS, AS-AP Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity	Sequence	Eluent (water) Standards Backpressure loop	10 min	CD: 2 hr, 10 min ED: 2 hr, 50min
AS-AP with temperature control Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity Injection Temperature Accuracy Test	Sequence	Eluent (water) Standards Backpressure loop Calibrated Digital Thermometer	10 min	CD: 2 hr, 40 min ED: 3 hr, 20min
AS with Sample Prep Warm Up Detector Noise / Drift Injector Precision Carryover Detector Linearity Sample Prep Precision	Sequence	Eluent (water) Standards Backpressure loop	10 min	2 hr, 40 min

Eluent Generator Current Qualification	Sequence	Test cell Multimeter	N/A	16 min
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5.2 Starting the Sequences

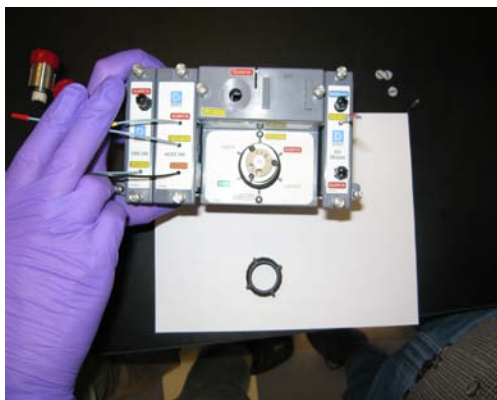


For AS Autosamplers with the Sequential Injection Option: Before starting any sequences, first verify that none of the instruments configured on the computer have “Acquired Exclusive Access” to the autosampler, or column TC, or compartment TC. If this is the case release the “Exclusive Access” command before proceeding. Once the qualification sequences begin, the system being qualified will gain and retain exclusive access to the autosampler, column TC and compartment TC until the “Stop” sequence is run and “Exclusive Access” is released. “Exclusive Access” can be released by going to the command function (F8), choosing the module from the list, and executing the “ReleaseExclusiveAccess” command.

5.2.1 Flow Rate Accuracy and Precision Qualification

The Flow Rate Accuracy and Precision test is designed to qualify the flow rate accuracy and precision of all pumps. A 6-port valve is required to run the IC flow rate test. For capillary IC systems, the 4-port valve needs to be replaced by a 6-port valve. See the procedure on how to replace a 4-port valve by a 6-port valve.

1. Unscrew the locking ring on the outside of the valve and remove the ring.



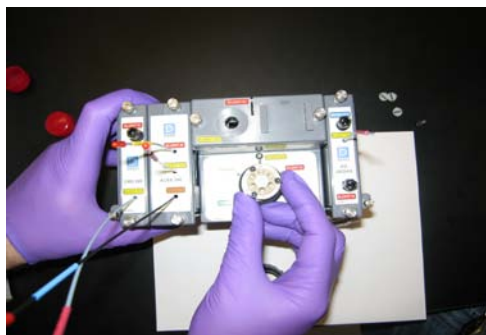
2. Grasp the front of the valve pod and pull out firmly to remove it from the IC cube.



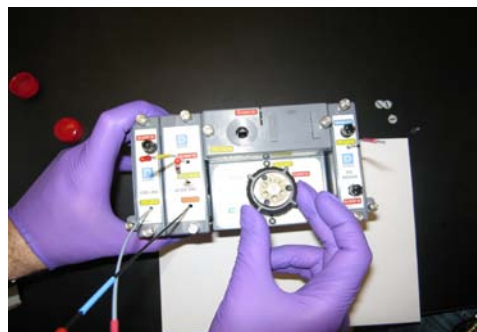
3. Align the slots in the new pod with the runner in the valve holder on the IC Cube.



4. Push the Pod into the holder until it clicks into place.



5. Replace the black locking ring.



1. Ensure the pump is disconnected from the Chromeleon software. The system should be in hold mode and a message box displaying "Plumb pump for Pump Flow rate Accuracy and Precision Test, press **OK** to continue" should be visible.

Plumb pump for Pump Flow-rate Accuracy and Precision Test, press OK to continue. OK

2. Stop the pump from the front control panel of the module if it is not already stopped.
3. Locate the backpressure coil. Use the tan coil (P/N 074572) for the capillary system.
4. Connect one end of the backpressure coil directly to the outlet of the pump and the other to port 2 (P) of the injector valve.
5. Prepare two lengths of blue tubing, 0.0025 (0.0063 in) ID, of equal lengths, approximately 12" (40 cm). Connect one of these lines to port 1 (L) of the injector valve. The tubing connected to port 1 (L) of the injector valve will be the eluent line that goes to the "sample vial."
6. Connect the other line to port 3 (C) of the injector valve. The tubing connected to port 3 (C) of the injector valve will be the waste line.
7. Run the waste line to a waste beaker and the eluent line to a sample beaker. The plumbing to the valve should look like the figure below.

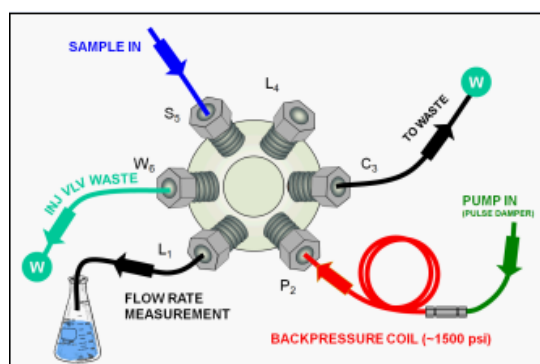


Figure 20. Plumbing schematic for Pump Flow Rate Test

8. Click **OK** on the Chromeleon software. The system should reconnect to the module and/or resume the program in run mode.
9. The program will equilibrate the pump for 15 minutes before starting the test. During this time eluent flow should pass through the waste line. Ensure that sufficient backpressure is observed during this equilibration time.
10. At time 0, the injector will switch to the eluent line.
11. At time 2.00, the injector will switch back to the waste line and the Chromeleon software will ask you to "Please weigh the eluent container. Enter results under segment 0. Press **OK** to continue." The program will be in hold mode and will wait until **OK** is pressed before continuing. Weigh the eluent container, but do not zero the balance used for weighing. Record the weight as segment 0. Click **OK** to continue.

Please weigh the eluent container. Enter results under segment 0. Press OK to continue. OK

Note: The template of this report is stored in the data vault of the respective sequence. Please refer to Chromeleon 7 Quick Start Guide on how to access report templates.

Template name: OQ_PQ_CIC_Modular_Validation

12. The Chromeleon software will switch the injector back to the eluent line for exactly 10 minutes. After this time the injector will switch to the waste line and a message "Please weigh the eluent container. Enter results under segment 1. Press **OK** to continue" will be displayed. The program will be in hold mode and will wait until **OK** is pressed before continuing. Weigh the eluent container including the tubing, nut and ferrule, but do not zero the balance used for weighing. Record the weight as segment 1. Click **OK** to continue.
13. Repeat 12 for segments 2 through 5.
14. After the sequence is finished open the Water_FR sample from the Console. Select the Flow Rate sheet at the bottom of the page and enter the Segment values and room temperature into the sheet. Save the worksheet by clicking on the Chromeleon Icon on the top left of the User Interface and select save. A written or balance printout of the weights should be signed and dated to accompany data entered on the sheet.

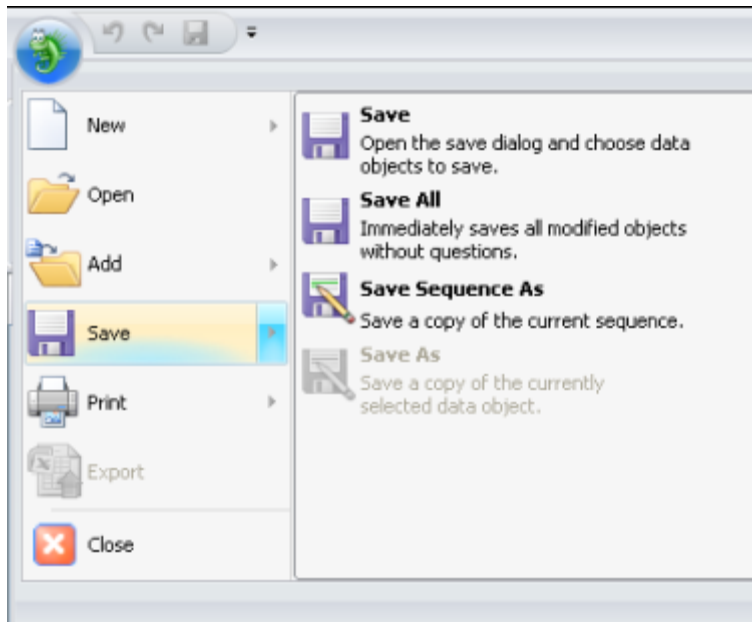


Figure 21. Selecting Save

15. Print the Flow Rate Accuracy and Precision report by clicking on the Chromeleon Icon on the top left of the User Interface and select print. Choose a printer and press OK to begin printing.

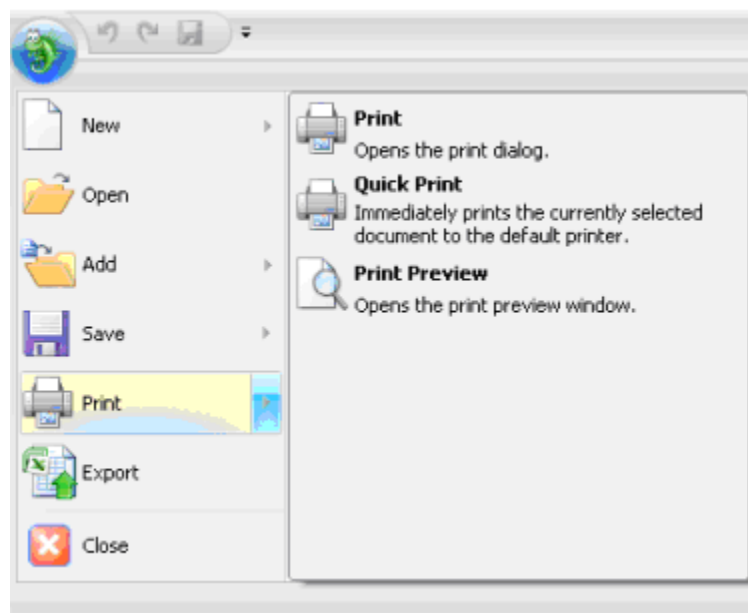


Figure 22. **Selecting Print**

16. Print the Flow Rate Accuracy and Precision report by selecting **Print** from the **File** menu. Choose a printer and press **OK** to begin printing.
17. If any part of the test does not pass, make the appropriate repair or calibrations and rerun the test. Refer to Section 5 for troubleshooting assistance.
18. Plumb the system for the remainder of the qualification.

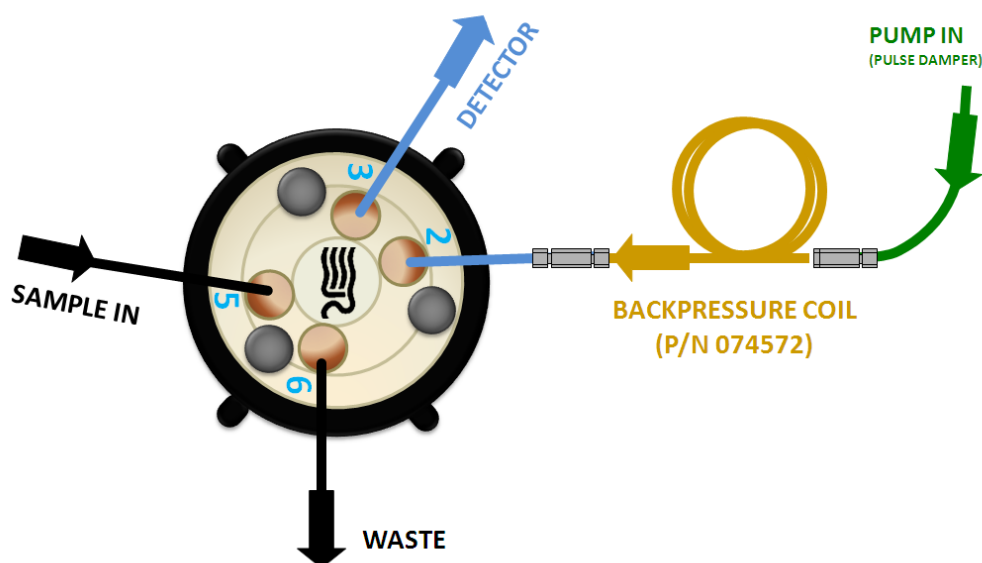


Figure 23. Plumbing diagram, initial equilibration step for a capillary system

19. Ensure the pump is disconnected from the Chromeleon software. The system should be in hold mode and a message box displaying "Plumb the system for the remainder of the qualification tests, press **OK** to continue" should be visible.
 - a. ICS-5000/ICS-5000[±] Conductivity: Connect the backpressure tubing (P/N 074572) to eluent in port of the 4-port valve. Attach the loop end to the inlet of the conductivity cell, thus bypassing all columns and suppressors. Use the 115 mm blue peek tubing (P/N 072189) from the IC Cube Tubing kit (P/N 072186) to connect from the 4-port valve to the detector

It is important to connect the Backpressure Loop assembly upstream from the loop or unusual peak shapes may occur.

- b. ICS-5000/ICS-5000[±] Integrated Amperometry:
 - 1) Locate the ED Cell Inlet Tubing Kit (P/N 074221) shipped with the cell. The kit contains a 17 cm (6.75 in) piece of blue precut capillary tubing (P/N 071870) and the fittings needed for connecting it between the PA20G and the cell inlet.
 - 2) Use a high-pressure (blue) fitting to connect the tubing from the **Eluent Out** port of the capillary EG degas cartridge to the **Eluent In** port (2) of the injection valve. Turn on the pump flow and wait about 60 seconds before tightening the connection.
 - 3) Wait until liquid is flowing out of the **Eluent Out** port (3) of the injection valve, then turn off pump flow.
 - 4) Use a high-pressure (blue) fitting to connect the tubing from the **Eluent In** port of the column cartridge to the **Eluent Out** port (3) of the injection valve. Turn on plump flow and wait until liquid is flowing out of the **Eluent Out** port of the column cartridge.
 - 5) 5) Use a 10-32 fitting bolt (P/N 043275) and 10-32 double-cone ferrule (P/N 043276) to connect the cell inlet tubing (P/N 071870) to the **Eluent Out** port of the column cartridge.
 - 6) 6) Wait until liquid is flowing out of this tubing and then use the long 10-32 fitting bolt (P/N 057227) and split-cone ferrule (P/N 062978) to connect the tubing to the **Cell In** port of the electrochemical detector. Turn the pump flow off.

Refer to Page 99 of the ICS-5000/ICS-5000⁺ Ion Chromatography System Installation Instructions (Document No. 065343) for complete instructions installing the Low-Pressure Fluidic Connections for Electrochemical Detection

5.2.2 Eluent Generator Current Qualification

The Eluent Generator Current Qualification test ICS-5000/ICS-5000⁺ EG (single or dual) configured with either a KOH or MSA cartridge (or both).

When using a FLUKE 189/289 multimeter, the measured current is automatically entered into the **EG_CURRENT**, **EG_DUAL_CURRENT** sheet.

1. **Resume** the Batch by clicking **OK**. This sequence is an Eluent Generator step gradient. Each step is held until the user clicks **OK** to continue. There may be a delay of up to 0.4 minutes between when the Eluent Generator program steps to the next concentration and when the ammeter or multimeter registers the new milliamp reading. Record the milliamp reading for each step on a note pad. These values will be entered into the **EG_CURRENT**, **EG_DUAL_CURRENT** or **EG Current Qualification** sheet after the run is complete. The run time is 11.0 minutes plus pause time.

Note: The template of this report is stored in the data vault of the respective sequence. Please refer to Chromeleon 7 Quick Start Guide on how to access report templates.

Template name: OQ_PQ_CIC_Modular_Validation

2. When the run is complete, open the report file by double clicking on the Water_EG sample of the XQ_EG_Current sequence and select Printer Layout from the View menu. Locate the row of sheet names at the bottom of the report and click on the **EG_CURRENT, EG_DUAL_CURRENT** or **EG Current Qualification** sheet.
3. Go to **Edit** and turn **Layout Mode** on. Enter the milliamp readings that were recorded in step 1 for each concentration value in the **EG_CURRENT, EG_DUAL_CURRENT** or **EG Current Qualification** sheet. The deviation and pass/fail results will be calculated automatically.
4. Print the EG Current Qualification report by selecting **Print** from the **File** menu. Choose a printer and press **OK** to begin printing.
5. If any part of the test does not pass, make the appropriate repair/adjustments and rerun the test. Refer to Section 5 for troubleshooting assistance.
6. To qualify another cartridge, select the new cartridge type in the control panel, add the XQ_EG_Current, XQ_EG_Dual_Current or XQ_IC_EG_Current sequence to the batch list, set the status of the Water_EG and Stop samples to single and repeat steps 1 - 4.

5.2.3 Performing the Main Qualification

The main part of the OQ/PQ consists of the tests 3 – 9 of the OQ/PQ test order, and this part of the qualification requires that an absorbance or conductivity detector be configured on the system being qualified.

If the qualification will be performed using a manual injection valve, use full loop injections and overfill the loop a minimum volume equal to 3 times the sample loop volume. Inject standards at the beginning of each sample in the sequence. There will be a 2.4-minute delay before the sample is injected. It is advisable to manually edit the programs to replace the 2.4-minute delay with a message command to hold the system until the sample is ready.

```
"-2.400 Pump_Relay_1.Closed Duration=130.00"
```

Line with

```
"-0.100 Message "Load the sample into the injection loop, press OK to continue"
```

1. The sequences must be installed as described in Section 4.5.
2. The sequences must be set up as described in Section 4.6.

If the batch is not started already go to Batch and Edit. Start the Batch. Run times are as follows:

- Systems with an AS-DV or manual injection have a run time of approximately 1 h, 10 min.
- Systems with an AS and AS-AP but no sample prep have a run time of approximately 2 h, 10 min.
- Systems with an AS and AS-AP with cooling have a run time of approximately 2 h, 40 min.

- Systems with an AS and sample prep have a run time of approximately 2 h, 40 min.

At the end of the main qualification the capillary system the pump will remain on and continue to run at a flow rate of 0.010 mL/min.

For systems using an Eluent Generator, the program will pause before the Eluent Generator Current Qualification Test, proceed to section 5.2.2. The program will pause before the Flow Rate Accuracy and Precision Test, proceed to section 5.2.4 before continuing to step 3.

3. When the queue is finished, go to the Chromeleon console right click on each completed sequence present in the qualification, select the **Report** and **Print** the report. The Eluent Generator Current Qualification, Flow Rate Accuracy and Precision Qualification, Column Heater Accuracy qualification have already been printed.
4. Review the report for Pass/Fail results. If any test does not pass, make the appropriate instrument repair and/or adjustments and rerun all tests from the original point of failure on. Refer to Section 6 for troubleshooting assistance.

5.2.4 Column/Compartment TC Temperature Qualification

The column/compartment TC temperature qualification can be used to qualify the temperature accuracy of the column/compartment TC on an ICS-5000/ICS-5000⁺. After completion of all other tests record the reading on the digital thermometer. See 5.2.5 to set up the Thermocouple on to the digital thermometer.

The IC Cube temperature qualification is used to qualify the temperature accuracy of the IC Cube temperature on an ICS-5000/ICS-5000⁺. Make sure the temperature probe is inserted into the column cartridge of the IC Cube and the adaptor is inserted into the digital thermometer as shown in Figure 25. Close the DC Door during the temperature qualification. After completion of all other tests record the reading on the fluke meter Make sure the temperature probe is inserted into the column cartridge of the IC Cube.



Figure 24. Thermocouple Adaptor and Thermocouple



Figure 25. Putting the Thermocouple into the column cartridge

1. Open the Water_WU sample in the Warm Up sequence from the Console. Click on **Printer Layout** from the View menu. Select the **Temperature Qualification** tab at the bottom of the page, turn on layout mode and enter the measured temperature of the Column TC in the "Measured Column Temperature" cell and if the Compartment TC is installed, enter the measured temperature of the Compartment TC in the "Measured Compartment Temperature" cell.

Note: The template of this report is stored in the data vault of the respective sequence. Please refer to Chromeleon 7 Quick Start Guide on how to access report templates.

Template name: OQ_PQ_CIC_Modular_Validation

2. If the test does not pass, make the appropriate repair/adjustments and rerun the test. Refer to Section 6 for troubleshooting assistance.
3. Print the Column TC Temperature Verification report by selecting "**Print**" from the File menu. Choose a printer and press **OK** to begin printing.

5.2.5 Turning off watermarks in the reports

Some users prefer not having watermarks in the back of the report. The procedure below describes how to turn off watermarks in the reports. The procedure can only be performed if the user has sufficient privileges granted by the administration.

1. Go to the Instrument Category
2. In the Chromeleon Console, Go to the Tools menu.
3. Select Administrations Console

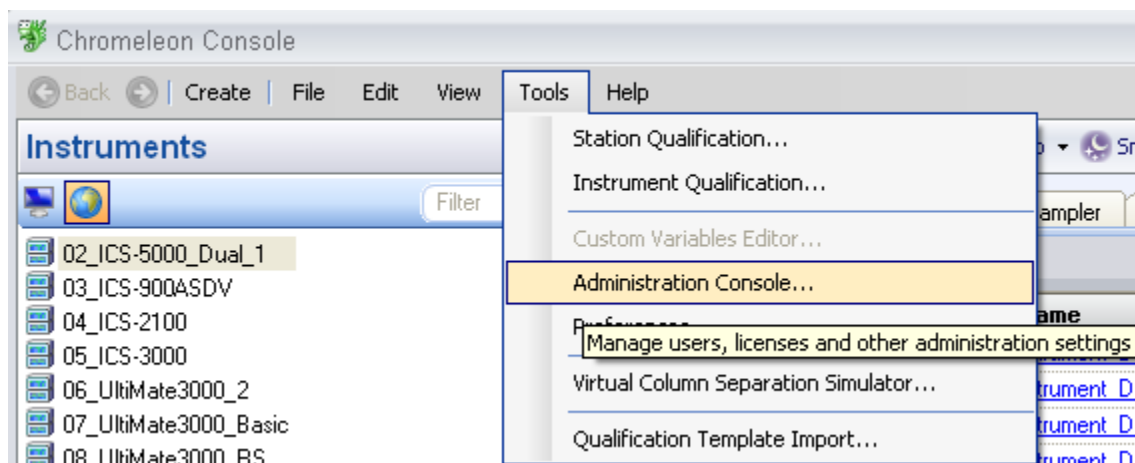


Figure 26. **Selecting Administration Console**

4. Select Global Policies within the Administration Console.

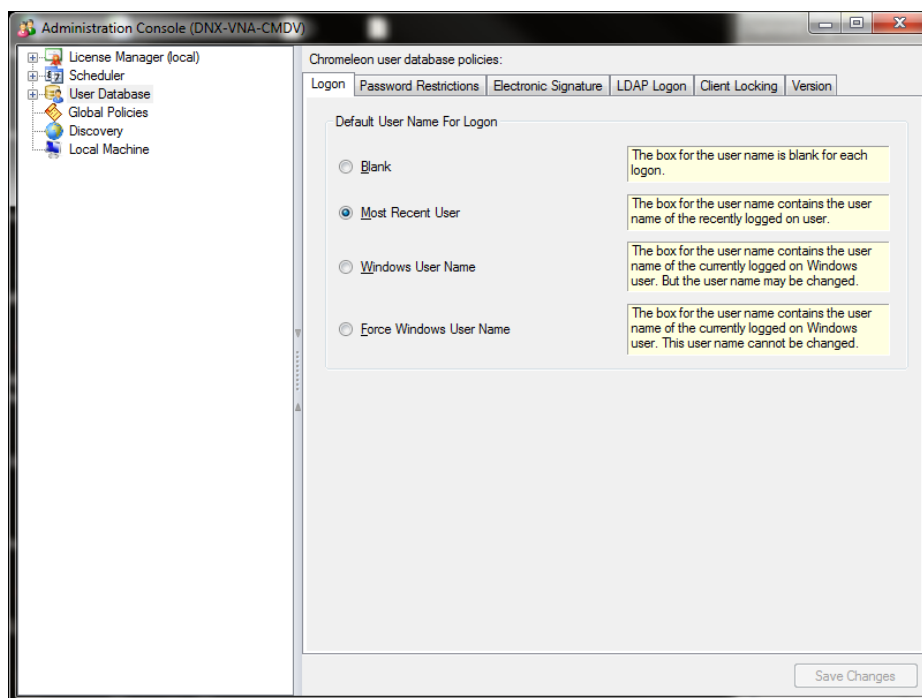


Figure 27. **Selecting Global Policies**

5. Select “Reporting” tab, then uncheck “Print with watermark”

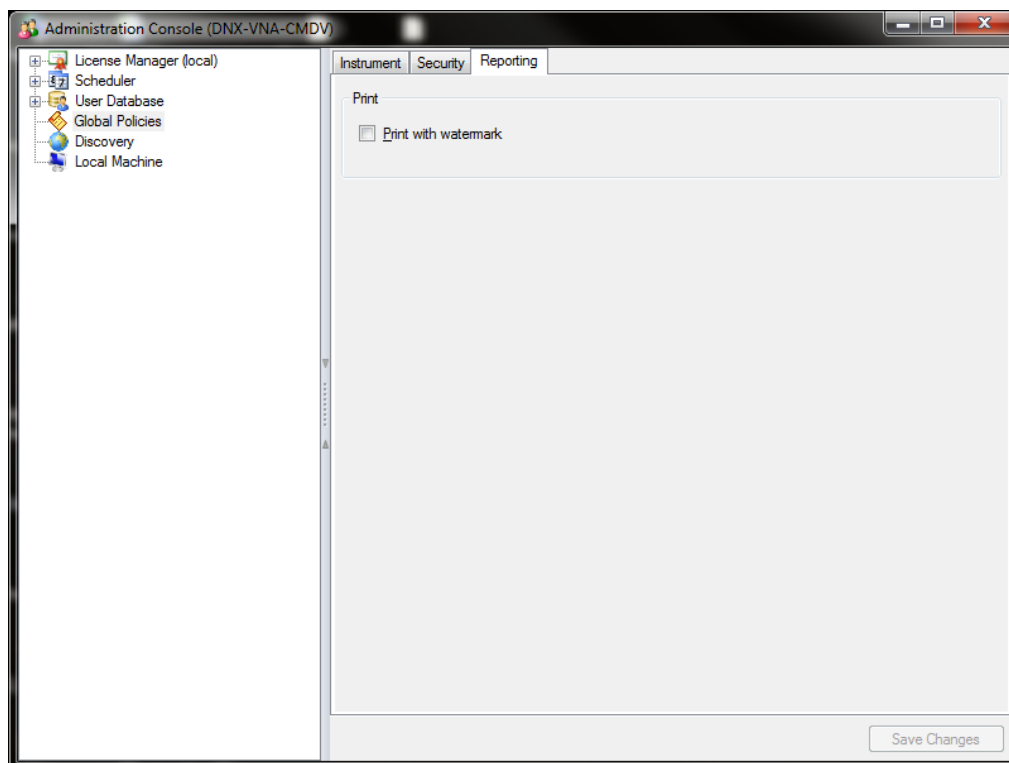


Figure 28. **Selecting reporting**

5.2.6 Sample Temperature Accuracy (AS-AP autosampler only)

This section describes how the sample temperature accuracy is determined for the following Thermo Fisher Scientific autosamplers: AS-AP. For this test, only the autosampler is required. The other modules of the IC system are not required.

Test Procedure

The following table lists the materials required for performing the test.

Part No.	Description	Quantity
6820.0010	Type K temperature sensor for P600 thermometers	1
5705.0050A	Column Thermostat PQ Kit	1

In addition, a standard polypropylene vial (10.0 mL) is required. Fill the vial with water (do not cap).

Connecting and Configuring the System

System connections

- . Connect the Type K temperature sensor to the Dostmann P600 thermometer. and make the settings (that is, adapt the sensor type and calibration values) as described in the instructions for the thermometer.



Figure 29. P600 Thermometer and Type K sensor



Figure 30. Type K sensor

- Fill an open standard polypropylene vial (10 mL) with water and place it at sample position BC1 with CD systems and RB1 with ED systems.
- Insert the temperature sensor into the vial at a right angle until the tip touches the vial bottom.



Figure 31. Temperature sensor inserted into the vial

- Rotate the carousel until the carousel cover closes completely.



Figure 32. Cover closed

- **Configuration**
Configuration is not required.
- **Preparing Chromeleon**
To qualify the sample temperature accuracy, select the following sequence:
SAMPLER_TEMP_ACC

Performing the Check

Set the sample temperature to 15 °C. When the nominal temperature is reached, the external thermometer is used to record the sample temperature over a period of 30 minutes.

Tip: Do not perform any autosampler commands during the test. Moving the needle arm or carousel may damage the thermometer or autosampler. The user can use a calibrated thermometer such as the Dostmann P600 or equivalent.

Duration

The test takes approximately 45 minutes.

5.2.7 AS-AP pH and Conductivity Qualification

This section describes how the sample pH and conductivity accuracy is determined for the following Thermo Fisher Scientific autosampler: AS-AP. For this test, the autosampler and other modules of the IC system are required. Please refer to the operator manual for pH and conductivity detector set up. The pH qualification should be performed after the standard qualification. The AS-AP pH and conductivity qualification will be set up in push mode only.

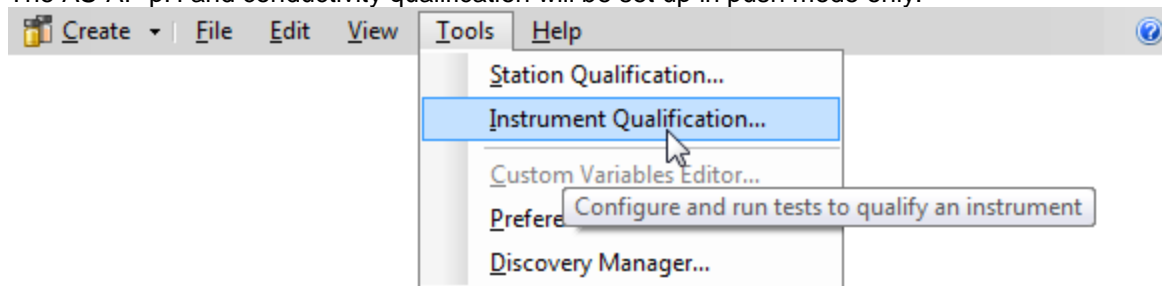


Figure 33. Instrument Pane for Chromeleon 7.1

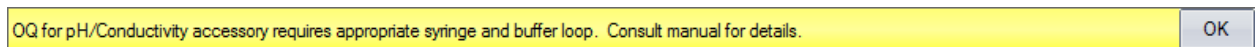


Figure 34. Starting the Instrument Qualification Wizard for Chromeleon 7.1

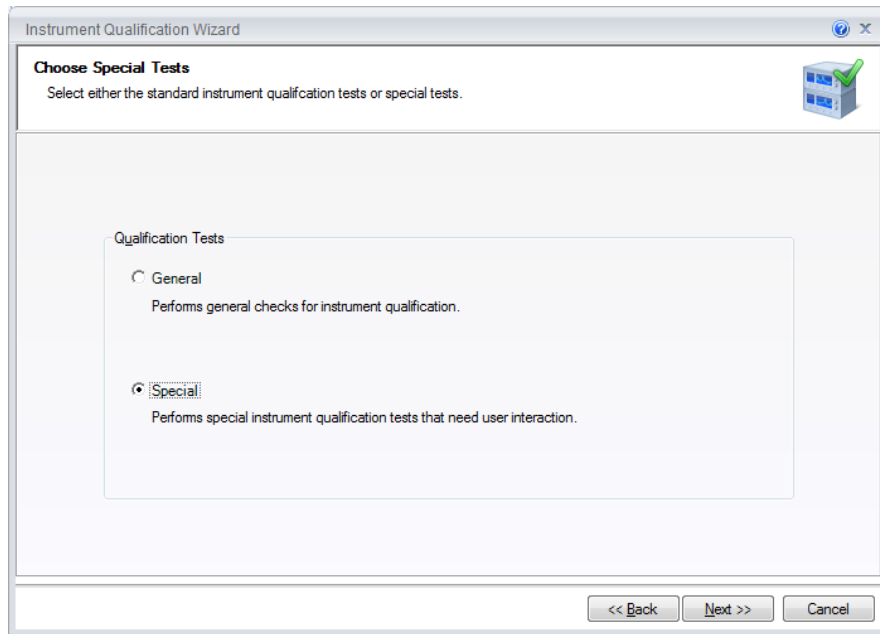


Figure 35. Select special for the Qualification Tests

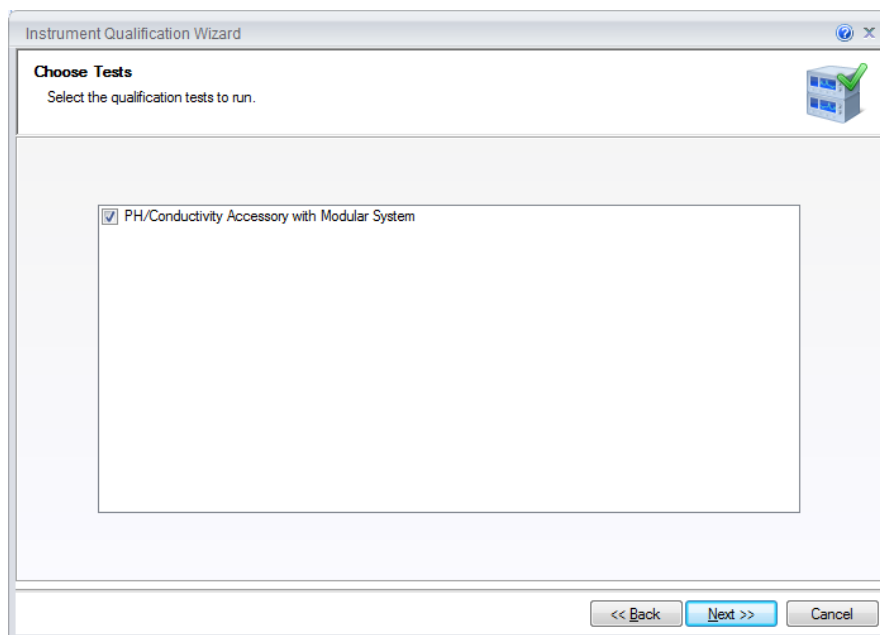


Figure 36. The pH/ Conductivity Tests will be selected if the instrument is configured correctly.

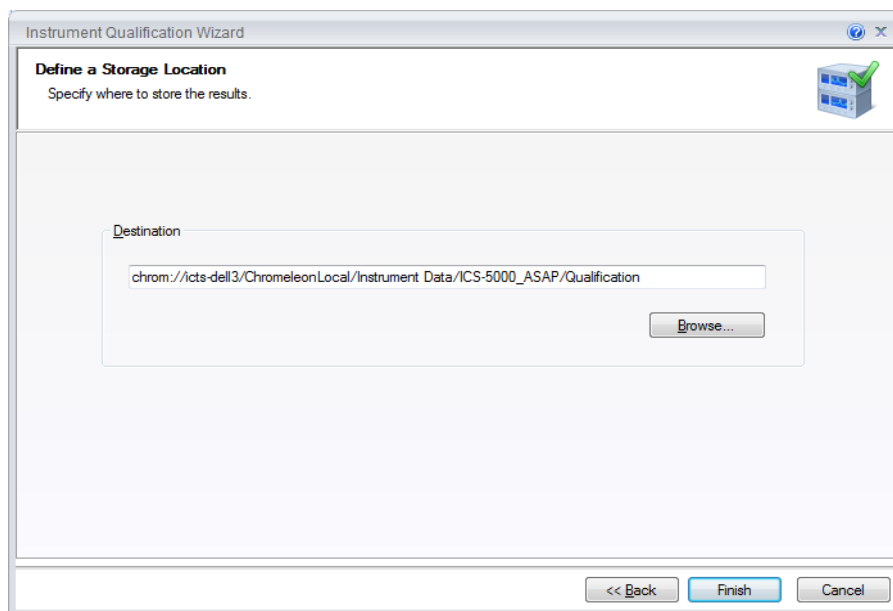


Figure 37. Select finish

#	Chromatogram	Name	Type	Level	Position	Volume [µL]	Instrument Method	Processing Method
1	None	WARMUP	Blank	01	RA1	100.0000	WARMUP	Validation
2	None	pH_OFFSET_CALIBR...	Blank	01	RA1	100.0000	pH7_CALIBRATION	Validation
3	None	pH_SLOPE_CALIBR...	Blank	01	RA2	100.0000	pH4_CALIBRATION	Validation
4	None	pH_SLOPE_CALIBR...	Blank	01	RA3	100.0000	pH10_CALIBRATION	Validation
5	None	pH_VERIFICATION(p...	Blank	01	RA1	100.0000	pH_MEASUREMENT	Validation
6	None	pH_VERIFICATION(p...	Blank	01	RA2	100.0000	pH_MEASUREMENT	Validation
7	None	pH_VERIFICATION(p...	Blank	01	RA3	100.0000	pH_MEASUREMENT	Validation
8	None	CD_CALIBRATION	Blank	01	RA4	100.0000	CD_CALIBRATION	Validation
9	None	CD_MEASUREMENT...	Blank	01	RA5	100.0000	CD_MEASUREMENT	Validation

Figure 38. Sequences

Performing the Check

1. Fill the vials according to the table below.

Table 14. OQ/PQ Test Timetable

Qualification	Standard	Sample Order on AS-P
pH Measurement	pH OFFSET Calibration	RA1
pH Measurement	pH Slope Calibration	RA2
pH Measurement	pH Slope Calibration	RA3
CD Calibration	Blank	RA4
CD Measurement	Conductivity Standard	RA5

2. Start the sequence.
3. A pop out screen will remind the user to configure the AS-AP with the 5mL syringe and the 8500 µL buffer loop before the qualification begins.

OQ for pH/Conductivity accessory requires appropriate syringe and buffer loop. Consult manual for details.

OK

Figure 39. Reminder Screen

Duration

The test takes approximately 1 hour.

5.2.8 Qualification Comments

The comments section of the report is provided as a place to record any comments, test failures, or configuration changes that occur during the qualification.

To use the comment section, open the Water_WU sample in the Warm Up sequence from the Console. Select **Print** from the **File** menu. Choose a printer and press **OK** to begin printing. This page can also be printed and comments entered manually.

To print the Comment report, select **Print** from the **File** menu. Choose a printer and press **OK** to begin printing.

Note: The template of this report is stored in the data vault of the respective sequence. Please refer to Chromeleon 7 Quick Start Guide on how to access report templates.

Template name: OQ_PQ_CIC_Modular_Validation

5.3 What to do if a Test Fails

If at any point the instrument results do not pass within Thermo Fisher Scientific specified limits, the instrument must be serviced. See Chapter 6 for troubleshooting assistance. When the repair is complete, rerun all tests from the original point of failure on in the sequence. All actions for failures and repairs should be recorded on the "comments" report. For example, if the instrument passes tests 1, 2, and 3 and fails test 4, make the appropriate repairs, record the action, and then repeat tests 4 – 9 only.

When rerunning the tests, restart the Instruments OQ or Instrument PQ procedure from the Qualification menu and uncheck all passed tests prior to the test that failed. All tests after the test that failed should be left checked even if they have previously passed. The Warm Up sequence is always required and must be rerun.

5.4 OQ/PQ Completion

5.4.1 Customer Review

1. The printed results should be reviewed by the instrument owner/user.
2. When the qualification is accepted, both the Customer and Qualification Executor need to sign the printed reports.
3. Fill out the Qualification Stickers and Certificates of Functional Performance for the qualified modules.

4. Place the reports and certificates into the binder provided in the OQ/PQ kit and leave it with the customer.

5.4.2 Accuracy of Reports

In accordance with Good Laboratory Practice (GLP), calculations used to determine the results in the printed reports use more significant figures than those shown and are rounded down for the final report. When reproducing the calculations by hand it may be necessary to obtain numbers from the reports after increasing the number of displayed significant figures.

The number of displayed significant figures can be adjusted through the report publisher. Open the sequence containing the samples for the test being examined and double click on a sample. The sample data should be shown. Under the View menu choose Printer Layout to open the report. Highlight the numbers being used for the hand calculations, click on the Format menu and select number. The number of digits displayed can be increased by changing the number type; for example, entering 0.00000 into the Type field will increase the number of displayed decimal places to 5.

Always use more significant figures for hand calculations than is shown in the printed report.

5.4.3 Returning the System to the Original Configuration

1. If the Signal factor was modified in the system preparation, or a module was renamed it must be set back to its original value. Open the Chromeleon Instrument Configuration application. Double click on the detectors qualified to view the **properties** page. Select the **Signals** page to change the factor back to the original value and the **General** page to change the name back to the original value. Exit the properties page and save the configuration.
2. Disconnect the Backpressure Coil and restore the system plumbing to its original configuration with the appropriate columns, suppressors, and eluent generator in line.
3. Remove the sample loop and reinstall the original sample loop back onto the injection valve. For systems using an AS Autosampler, go to the **Main Menu** on the front panel of the autosampler. Access the **Module Set Up** screen and then go to **Plumbing Configuration**. Change the sample loop size back to its original value.
4. Reconnect the original eluents to the pump and prime each line individually to purge the eluents used in the OQ/PQ tests.
5. Flush the system with an eluent recommended by the customer and confirm that there are no leaks.

6 Troubleshooting

6.1 General Hints

If the system pressure is above 2000 psi after connecting the PEEK Backpressure coil with a flow rate of 0.010 mL/min in capillary systems, the coil has a blockage, replace it.

Dips before or after the main nitrate peak indicate extra dead volume in the system. This is caused by small voids between the end of a liquid line and the bottom of the port. Individually disconnect each line from the injection valve to the detector cell. Push the ferrule and fitting up and away from the end of the tubing. Do not cut the capillary tubing. When reinstalling all liquid connections, first insert the tubing all the way into the port until it touches the bottom. Hold the tubing in this position while inserting and tightening ferrule and fitting down into the port.

6.2 Failure of Individual Tests

6.2.1 Conductivity Detector Tests

Test	Reason	Action
Baseline Noise	Poor water quality	Replace with an alternate water source
	Air bubbles in flow cell	Flush out flow cell
	Cell temperature regulation failure	Replace cell
Drift	System not equilibrated	Allow longer flush/equilibration time
	Lab temperature not stable	Regulate temperature (block vents, close windows, etc.)
	Water not degassed	Degas water or turn degas unit onto "Always On" setting.
Detector Linearity	Concentration of standards is incorrect	Obtain new fresh standards
	Liquid leak	Find and repair source of leak
	Injection volume not consistent (manual injections only)	Overfill sample loop at least 4x when loading

6.2.2 Pump

Test	Reason	Action
Step Noise (ripple)	Air in system	Prime pump, flush system
	Air in flow cell	Flush flow cell with iso-propyl alcohol, ensure adequate backpressure on flow cell
	System not equilibrated	Allow longer flush/equilibrium time
	Check valve or piston seal failure	Replace check valve/piston seal
Flow Rate Accuracy and Precision	Void time from pump to detector too long or short	Adjust tubing length accordingly
	Flow rate out of calibration	Re-calibrate the flow rate of the pump

6.2.3 Autosampler

Test	Reason	Action
Injector Precision	Air injected from vial or from sample syringe when doing manual injections	Too little sample volume in vial – refill vial or flush bubbles out of injection syringe
	Air bubbles in AS or AS-AP sample syringe	Flush out syringe
	Liquid leak in autosampler or injection valve	Find and repair source of leak
	Poor water quality in flush reservoir	Change water and flush sampler fluidics
Carryover	Contaminated injection valve	Replace injection valve rotor and stator face
	Contamination in autosampler	Flush/replace autosampler liquid lines
	Contamination in sample vial or in syringe used for manual injections	Use new sample vial for deionized water blank, rinse vial thoroughly before using, flush syringe or use a new one
Sample Prep Precision (AS only)	Air injected from vial	Too little sample volume in vial, refill vial
	Air bubbles in AS sample syringe or prep syringe	Flush out syringe
	Defective prep syringe	Replace prep syringe
	Liquid leak in autosampler or injection valve	Find and repair source of leak
	Poor water quality in Sample Prep reservoir	Change water and flush sampler fluidics

6.2.4 Integrated Amperometry Detector Tests

Test	Reason	Action
Baseline Noise	Electrochemical detector cell not connected properly	Reconnect cable to amperometry cell connection on detector
	Electrochemical detector cell defective	Replace DWE, reference electrode or ED cell.
	CR-ATC defective	Replace CR-ATC
Drift	Detector not equilibrated	Allow longer equilibration time
	Lab temperature not stable	Regulate temperature (block vents, close windows, etc.)
Injector Precision	KOH concentration wrong, retention of Threonine too high or too low	Replace KOH EGC

6.2.5 Eluent Generator

Test	Reason	Action
Current	Qualification Test box not connected properly	Reconnect test cell cable to the Eluent Generator, confirm ammeter or multimeter leads are making contact with test cell wires
	Ammeter or multimeter on wrong setting	Ammeter or multimeter should be set to read current (mA). Range should be set to view 0.00 to 300.00 mA
	Qualification cell defective	Replace qualification test cell

6.2.6 Column Heater

Test	Reason	Action
Temperature Qualification	Column Heater not calibrated	Recalibrate the Column Heater
	Thermometer not calibrated	Calibrate thermometer
	Thermometer not fitted in column heater (compartment) correctly	Fit thermometer in column heater (compartment) correctly

7 Example Report

The example report is for an ICS-5000/ICS-5000⁺ Capillary system configured with a CD Detector, DP Gradient Pump, and an AS-AP Autosampler.