

Application News

Ion Chromatograph

Seamless Ion Chromatography Workflow Using Nexera™ IC and IC Solution — Application to Anion Suppressor IC —

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User Benefits

- ◆ IC Solution, dedicated software for ion chromatography, streamlines the entire workflow from analysis to data processing.
- ◆ The eluent switching valve and dedicated mixer enable automatic eluent preparation.
- ◆ The dilution function of autosampler enables automatic preparation of calibration standards from stock solutions, as well as dilution and reanalysis of unknown samples exceeding the calibration range.

Introduction

Ion chromatographs are widely used for the determination of ionic components in aqueous solutions. They are also widely adopted in official test methods for various regulations, particularly in the environmental field, where highly reliable analytical results are required. Therefore, it is important to establish an analytical environment that minimizes human errors in measurement operations and data processing while ensuring consistent analytical performance. In addition, improved operational efficiency is required for routine analysis involving a large number of samples. This article presents an example of how IC Solution, dedicated software for ion chromatography, can streamline the analytical workflow and improve the reliability of analytical results.

Analytical Workflow Using IC Solution

Fig. 1 shows the analytical workflow using IC Solution. The software provides a variety of functions to streamline each step of the workflow, from pre-analysis preparation to report generation. For example, the startup function automates pre-analysis procedures such as purging the pump and autosampler and equilibrating the column. The automatic dilution function also enables automatic preparation of calibration curves and automatic dilution and reanalysis of unknown samples with concentrations exceeding the calibration range. These functions simplify analytical operations, reduce operator workload, and contribute to establishing a stable and efficient analytical workflow.

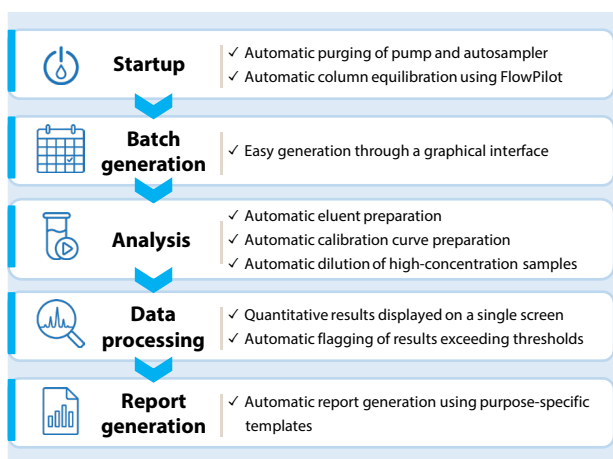


Fig. 1 Analytical Workflow Using IC Solution

Target Compounds and Analytical Conditions

In this article, a mixed standard solution containing representative anions commonly analyzed by ion chromatography was used as a model sample (analytical conditions: Table 1). Nexera IC (Fig. 2) was used as the ion chromatograph. Nexera IC has a height of 50 cm or less to facilitate the installation of eluent bottles. With a width of approximately 50 cm, the instrument also has a compact

footprint, making it suitable for laboratories with limited space. A conductivity detector (CDD) is equipped as standard, enabling highly sensitive ion analysis when combined with a suppressor. An optional absorbance detector (UV) can also be installed on top of the instrument for selective detection of ions with UV absorption, such as nitrite ions.

Table 1 Target Compounds and Analytical Conditions

System	: Nexera IC with ICDS-Ai anion suppressor
Sample	: F ⁻ (5 mg/L) : Cl ⁻ (5 mg/L) : NO ₂ ⁻ (20 mg/L) : Br ⁻ (20 mg/L) : NO ₃ ⁻ (20 mg/L) : PO ₄ ³⁻ (50 mg/L) : SO ₄ ²⁻ (20 mg/L)
Mobile phase	: 1.8 mmol/L Sodium Carbonate / 1.7 mmol/L Sodium Hydrogen Carbonate in water
Column	: Shim-pack IC-SA2 (250 mm × 4 mm I.D., 9 μm) ^{*1}
Guard column	: Shim-pack IC-SA2 (G) (10 mm × 4.6 mm I.D., 9 μm) ^{*2}
Analytical conditions	
Flow rate	: 1.5 mL/min
Injection Vol.	: 50 μL (partial-loop injection)
Loop size	: 200 μL
Needle stroke	: 47 mm (4 mL vial), 45 mm (1 mL vial)
Vial for mixing	: Shimadzu Vial, LC, 1 mL, Polypropylene ^{*3}
Vial for standard	: Shimadzu Vial, LC, 4 mL, Polypropylene ^{*4}
Vial for sample	: Shimadzu Vial, LC, 4 mL, Polypropylene ^{*4}
Sample solvent	: Ultrapure water
Column Temp.	: 40 °C
Detection	: Conductivity

*1 P/N : 228-38983-91, *2 P/N : 228-38983-92

*3 P/N : 228-31600-91, *4 P/N : 228-31537-91



Fig. 2 Nexera™ IC

■ Startup (System Stabilization Before Analysis)

The startup function of IC Solution automates pre-analysis procedures such as purging the pump and autosampler and equilibrating the column. During column equilibration, the FlowPilot function (Fig. 3) automatically adjusts the flow rate according to the column oven temperature, enabling column equilibration without applying excessive pressure to the column (red circle in Fig. 3). Fig. 4 shows the actual screens for purging the pump and autosampler, as well as the pressure changes during column equilibration using FlowPilot. By executing the startup function in advance, manual purging and waiting time for column equilibration can be reduced, thereby improving operational efficiency.

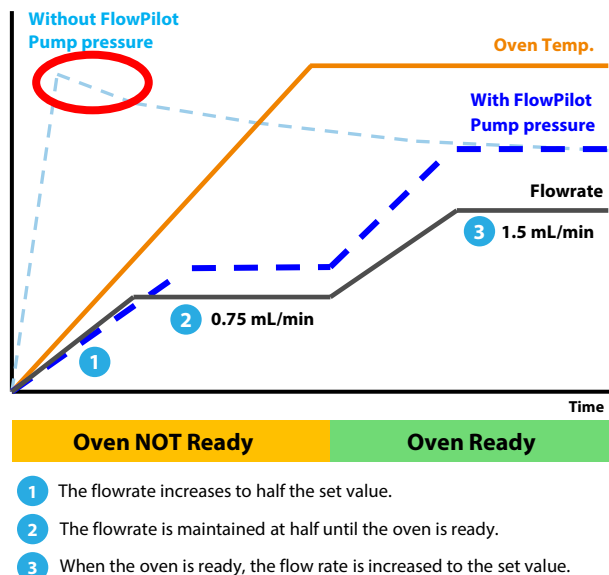


Fig. 3 Column Protection Using FlowPilot

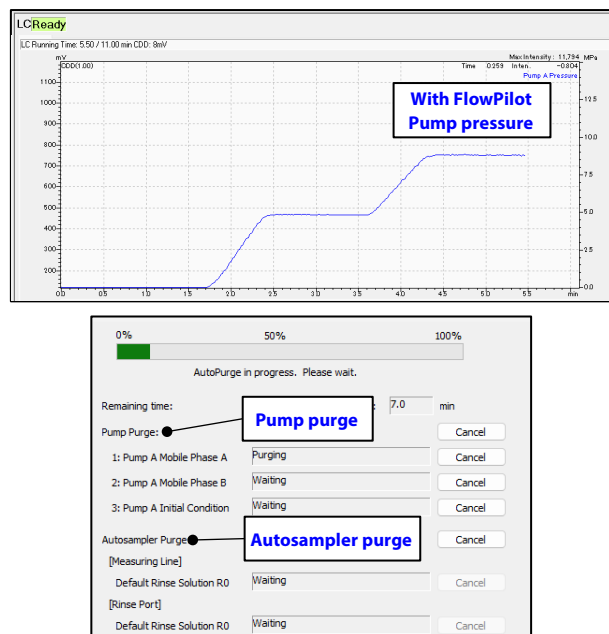


Fig. 4 Pump Pressure During FlowPilot (Upper)
Automatic Purging Screens for Pump and Autosampler (Lower)

■ Batch Generation

IC Solution enables easy creation of batch schedules through an intuitive graphical user interface (Fig. 5, steps (1)–(3)). It also allows automatic preparation of calibration standards from stock solutions using the autosampler's dilution function. For each analysis row, automatic dilution can be performed simply by specifying the stock solution tray/vial number and dilution factor (Fig. 5, step (3)). In addition, when an additional sample requiring urgent analysis arises during an ongoing sequence, it can be inserted immediately after the current analysis.

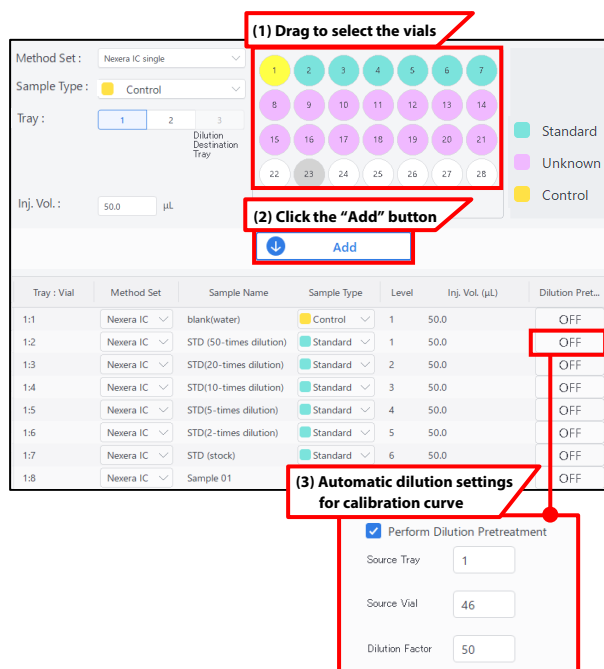


Fig. 5 Batch Generation Screen in IC Solution

■ Automatic Eluent Preparation

Nexera IC enables automatic eluent preparation using an eluent switching valve and a dedicated mixer. A concentrated eluent and ultrapure water are alternately delivered at predetermined time intervals and mixed using the dedicated mixer to automatically prepare the eluent by dilution (Fig. 6). The dilution factor can be selected from 2-, 5-, and 10-fold. The automatic eluent preparation function not only reduces the workload associated with manual preparation but also minimizes variations in eluent preparation, thereby enabling stable analysis without fluctuations in retention times.

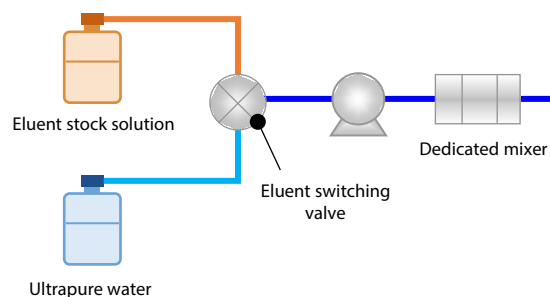


Fig. 6 Overview of the Automatic Eluent Preparation

Fig. 7 shows a comparison of chromatograms obtained using automatically prepared eluent (10-fold dilution) and manually prepared eluent. Table 2 summarizes the reproducibility (%RSD) of the retention times and peak areas for each anion over six replicate analyses using the automatic eluent preparation function (chromatograms: Fig. 8). The results confirmed that the automatic eluent preparation provided a separation pattern comparable to that obtained with manual preparation. Good reproducibility was also obtained for all anions, demonstrating that the automatic eluent preparation function provides sufficient mixing performance.

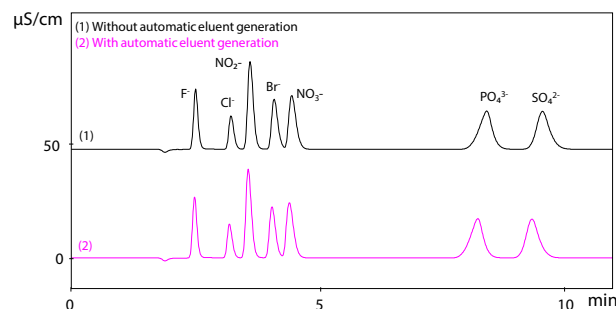


Fig. 7 Comparison of Chromatograms With and Without the Automatic Eluent Preparation Function

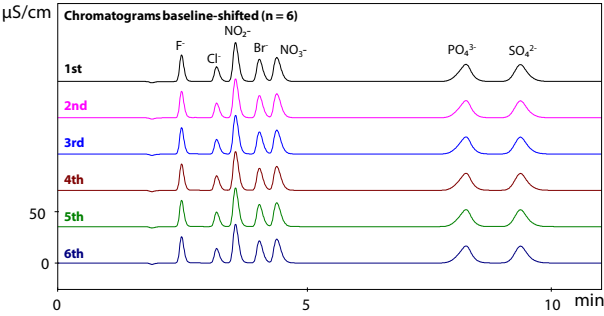


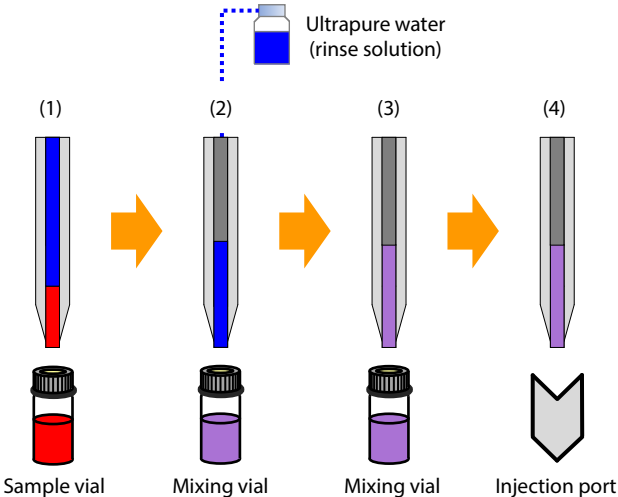
Fig. 8 Chromatograms from Replicate Analyses (n = 6)
(obtained using the automatic eluent preparation function)

Table 2 Reproducibility (%RSD) of Each Anion over Six Replicate Analyses

	Retention time (min)	Peak area
F ⁻	0.14	0.21
Cl ⁻	0.11	0.20
NO ₂ ⁻	0.12	0.22
Br ⁻	<0.01	0.20
NO ₃ ⁻	0.08	0.20
PO ₄ ³⁻	<0.01	0.25
SO ₄ ²⁻	<0.01	0.18

■ Automatic Calibration Curve Preparation

In typical calibration curve preparation, standard solutions must be manually diluted to the required concentrations using pipettes or other tools. This process is time-consuming and labor-intensive, making it a challenge for improving the efficiency of analytical operations. IC Solution automates the preparation of the standard solutions required for calibration curve by using its automatic dilution function. This function enables samples to be automatically diluted at the desired dilution factors. The workflow of the automatic dilution function is shown in Fig. 9.



- (1) Aspirate the sample
- (2) Dispense the sample and ultrapure water into the mixing vial
- (3) Mix in the vial by aspirating and dispensing with the needle
- (4) Inject the mixed sample

Fig. 9 Automatic Dilution Workflow Using IC Solution

The automatic dilution function was applied to the anion mixed standard solution (Table 1) at dilution factors of 2, 5, 10, 20, and 50 from the stock solution to automate calibration curve preparation (six-point calibration curve including the stock solution). A 1-mL polypropylene vial was used as the mixing vial, and a 4-mL polypropylene vial was used for the stock solution vial. Ultrapure water, also used as the rinse solution, was used as the diluent. Fig. 10 shows the chromatogram of the anion mixed standard solution diluted 50-fold.

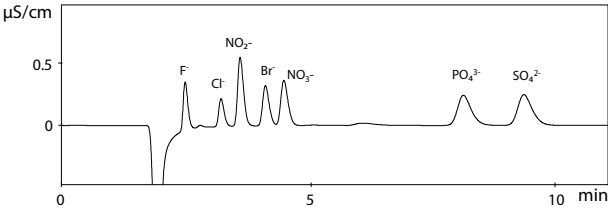


Fig. 10 Chromatogram of Anion Standard Solutions Prepared by Automatic Dilution (dilution factor: 50-fold)

■ Reproducibility and Calibration Curve Coefficient of Determination

Table 3 shows the reproducibility (%RSD) of the retention times and peak areas for each anion over three replicate analyses of an anion mixed standard solution automatically diluted 50-fold. Table 4 shows the coefficients of determination (r^2) for the calibration curves of each anion prepared at dilution factors of 2, 5, 10, 20, and 50 from the stock solution. Good reproducibility and coefficients of determination ($r^2 > 0.9999$) were obtained for all anions.

Table 3 Reproducibility (%RSD) of Each Anion over Three Replicate Analyses (dilution factor: 50-fold)

	Retention time (min)	Peak area
F ⁻	<0.01	1.02
Cl ⁻	0.06	0.87
NO ₂ ⁻	<0.01	0.82
Br ⁻	0.05	1.07
NO ₃ ⁻	<0.01	0.43
PO ₄ ³⁻	0.02	0.41
SO ₄ ²⁻	0.02	0.50

Table 4 Coefficients of Determination (r^2) for Each Anion (n = 1)
(Quadratic calibration curve)

	Calib. range (mg/L)	Coefficient of determination (r^2)
F ⁻	0.1-5	>0.9999
Cl ⁻	0.1-5	>0.9999
NO ₂ ⁻	0.4-20	>0.9999
Br ⁻	0.4-20	>0.9999
NO ₃ ⁻	0.4-20	>0.9999
PO ₄ ³⁻	1-50	>0.9999
SO ₄ ²⁻	0.4-20	>0.9999

■ Comparison of Peak Areas with Manual Dilution

Table 5 shows the accuracy of the peak areas obtained by automatic dilution (dilution factor: 50-fold), with the peak areas obtained by manual dilution set to 100% as a reference. The results confirmed that good agreement was obtained compared with manual preparation.

Table 5 Accuracy of Peak Areas Obtained by Automatic Dilution (n = 1)

	50-fold dilution
F ⁻	97.90%
Cl ⁻	97.31%
NO ₂ ⁻	98.71%
Br ⁻	99.17%
NO ₃ ⁻	96.31%
PO ₄ ³⁻	97.43%
SO ₄ ²⁻	97.43%

■ Automatic Dilution Function for High-Concentration Samples

In general, when quantifying unknown samples using a calibration curve, it is important to ensure that the concentration of the unknown sample falls within the calibration range to obtain reliable quantitative results. Therefore, when analysis reveals that the concentration of an unknown sample is outside the calibration range, the sample must be manually diluted by an appropriate factor and reanalyzed. This process requires not only manual dilution but also repeated QC analysis, increasing the workload associated with routine analysis. When the analytical result of an unknown sample exceeds the calibration range, IC Solution automatically dilutes the sample in the autosampler and reanalyzes it. This eliminates the need for manual dilution and reanalysis, thereby improving analytical workflow efficiency. The threshold for triggering automatic dilution can be set individually for each ion. In addition, to minimize carryover caused by high-concentration samples, flow path rinsing and a blank check can be automatically performed before reanalysis. Fig. 11 shows the procedure for setting up automatic dilution of unknown samples. As an example, Cl^- was selected as the target ion for automatic dilution. The maximum calibration point concentration (peak area at Level 6) was set as the threshold for triggering automatic dilution, while 0.2 times the minimum calibration point concentration was set as the threshold for the blank check. Fig. 12 shows the sequence of operations when a high-concentration unknown sample exceeding the calibration range (Table 4) was analyzed using IC Solution, and Fig. 13 shows the chromatograms obtained during this sequence. As shown in Fig. 12, because the Cl^- concentration in the unknown sample exceeded the upper limit of the calibration range, flow path rinsing and a blank check were automatically performed, followed by reanalysis after automatic dilution. Chromatogram (4) in Fig. 13 shows the result obtained from the reanalysis after automatic dilution. IC Solution automatically set the dilution factor to 18-fold (red box in Fig. 12), and the Cl^- concentration after automatic dilution was confirmed to be within the calibration range (Fig. 14). Thus, even when the analytical result of an unknown sample exceeds the upper limit of the calibration range, IC Solution can automatically determine that the result is outside the calibration range and perform dilution and reanalysis. This eliminates the need for manual dilution and reanalysis, thereby contributing to improved analytical workflow efficiency. In this article, the maximum calibration point concentration was set as the threshold. On the other hand, the peak area of each ion can also be used as the threshold. This enables automatic reanalysis when the measured peak area falls outside the specified range for samples whose peak areas are known based on previous analytical results or other information.

No	Status	Automation	Tray : Vial	Method Set	Sample Name	Sample ID	Dilution ...
1	Completed	High-concentration sample detection		DilutionRinse			1.0000...
2	Running	Auto rinse of flow path		AutoRinse	AutoRinse		1.0000...
3	Ready	Blank injection		Blank	Blank		1.0000...
4	Ready	Re-injection after automatic dilution		DilutionRinse			18.000

Fig. 12 Sequence from High-Concentration Sample Detection to Automatic Dilution

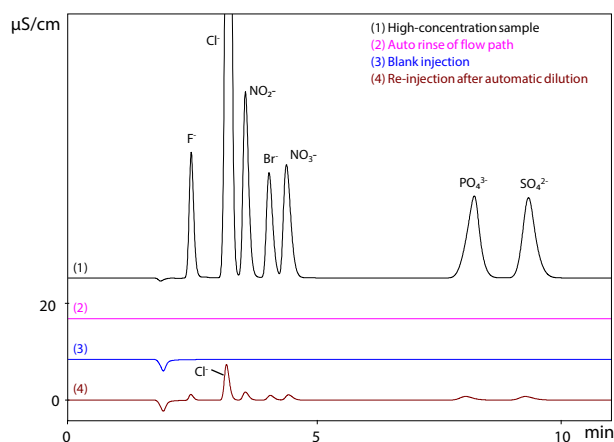


Fig. 13 Chromatograms Obtained from High-Concentration Unknown Sample Detection to Automatic Dilution

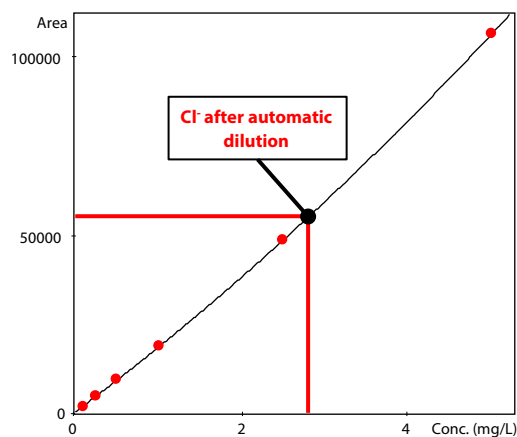


Fig. 14 Calibration Curve for Cl^- (concentration range: 0.1–5 mg/L)

General Automation

☒ Perform Automatic Rinsing ☒ Perform Automatic Dilution and Reinjection

High Conc. Sample Detection Automatic Rinsing Blank Check Automatic Dilution and Reinjection

High Conc. Sample Detection Condition Blank Sample Automatic Dilution and Reinjection Settings

Next Sample Analysis

(1) Select the ions for automatic dilution

(2) Set the threshold for automatic dilution

(3) Set the threshold for the blank check

High Conc. Criteria Settings

Blank Criteria Settings

Fig. 11 Procedure for Setting Up Automatic Dilution of Unknown Samples Exceeding the Calibration Range

■ Data Analysis and Report Generation Using IC Solution

Fig. 15 shows the data analysis results obtained using IC Solution. The quantitative results for each ion are displayed on a single screen, allowing the results to be easily reviewed (red box in Fig. 15). IC Solution also features a flagging function that automatically evaluates analytical results based on predefined thresholds and highlights results that fall outside the specified thresholds in red. This function facilitates the identification of abnormal results. For example, it can be used to determine whether the concentrations of target ions in each sample fall within user-defined criteria. Thresholds can be set for parameters such as upper and lower limits of quantitative values, calibration curve error, and S/N, and results that do not meet the specified criteria can be automatically highlighted. Thresholds can also be configured for each sample type, including standard, QC, and unknown samples. In Fig. 15, the ion exceeding the upper limit of the calibration range is highlighted in red by the flagging function. This function facilitates the review of large volumes of analytical results, helping to reduce review time and minimize errors during result verification. An example of a report generated

by IC Solution, namely a quantitative results report, is shown in Fig. 16. The report provides a list of quantitative results for each ion, along with the corresponding calibration curves and chromatograms. When data are simultaneously acquired using the CDD and UV detectors, chromatograms from both detectors can also be included in the same report.

■ Conclusion

This article demonstrated that IC Solution, dedicated software for ion chromatography, can significantly streamline the entire workflow from analysis to data processing. The eluent preparation process can be automated using an eluent switching valve and a dedicated mixer. In addition, the autosampler's dilution function enables automatic preparation of calibration standards from stock solutions, as well as dilution and reanalysis of unknown samples exceeding the calibration range. Furthermore, the flagging function facilitates the review of large volumes of analytical results, helping to reduce review time and minimize errors during result verification. These functions enable more efficient and standardized analytical operations and contribute to an efficient workflow that is less dependent on chromatography experience.

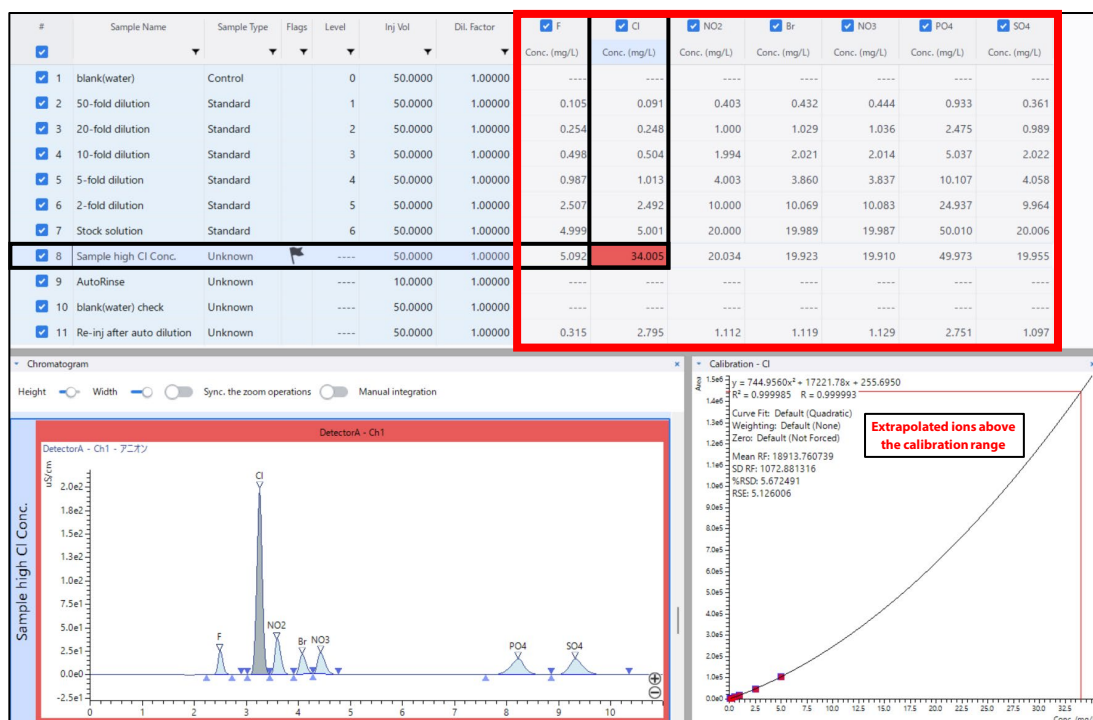


Fig. 15 IC Solution Data Analysis Screen

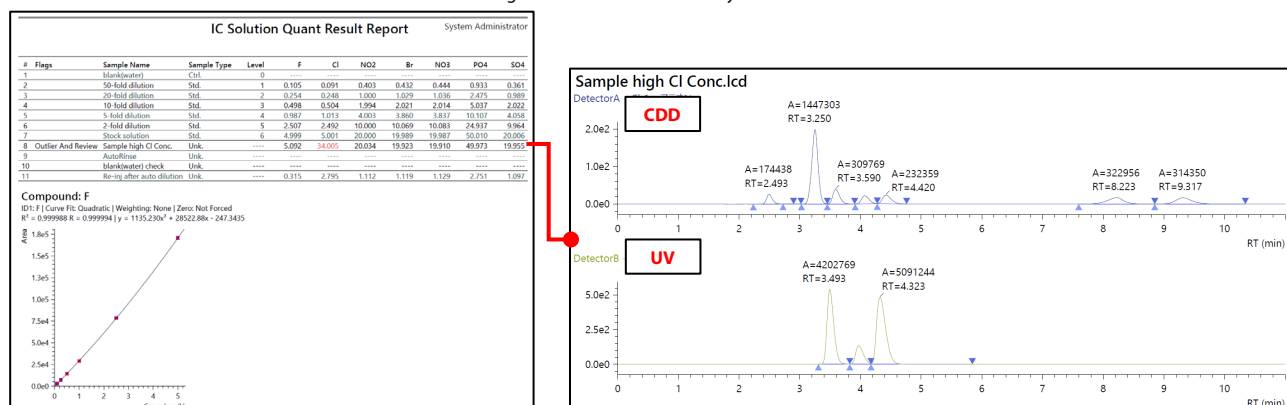


Fig. 16 Quantitative Results Report Generated by IC Solution

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