

Multi Anion Standard II  
UME CRM 2282

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## ABBREVIATIONS

$\alpha$	Significance level
CRM	Certified reference material
GUM	Guide to the Expression of Uncertainty in Measurement
HDPE	High density polyethylene
IC	Ion chromatography
HP-IC	High Performance Ion Chromatography
ISO	International Organization for Standardization
LTS	Long term stability
$MS_{between}$	Mean square between-bottle from ANOVA
$MS_{within}$	Mean square within-bottle from ANOVA
$n$	Number of replicates per unit
PRS	Primary reference standard
RSD	Relative standard deviation
$s$	Standard deviation
$s_{bb}$	Between-bottle standard deviation
SGT	Single Grubbs' test
SI	International System of Units
STS	Short term stability
$s_{wb}$	Within-bottle standard deviation
$U_{bb}$	Standard uncertainty related to possible between-bottle heterogeneity
$U_{bb}^*$	Standard uncertainty of heterogeneity that can be hidden by method repeatability
$U_{char}$	Standard uncertainty related to characterization
$U_{char,rel}$	Relative standard uncertainty related to characterization
$U_{lts}$	Standard uncertainty related to long term stability
$U_{lts,rel}$	Relative standard uncertainty related to long term stability
$U_{sts}$	Standard uncertainty related to short term stability
$U_{sts,rel}$	Relative standard uncertainty related to short term stability
$V_{MS_{within}}$	Degrees of freedom for $MS_{within}$

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## ABSTRACT

This report describes the production of UME CRM 2282 Multi Anion Standard II Certified Reference Material (CRM) from NaF (sodium fluoride), KBr (potassium bromide) and KH<sub>2</sub>PO<sub>4</sub> (potassium di hydrogen phosphate) Primary Reference Standards (PRS) and certification of F<sup>-</sup>, Br<sup>-</sup> and PO<sub>4</sub><sup>3-</sup> mass fractions in the CRM. The reference material production process consists of gravimetric CRM preparation, homogeneity, short-term stability, long-term stability and characterization stages. UME CRM 2282 certification studies were carried out in accordance with the requirements of the ISO 17034:2016 Standard and the ISO Guide 35:2017 document. In the chemical measurements carried out in this context, TS EN ISO/IEC 17025:2017 requirements were met. Uncertainties of certified values were calculated in accordance with JCGM 100:2008 Guide to the Expression of Uncertainty in Measurement (GUM).

Planning of all stages of this reference material production project, coordination of activities, execution of experiments and evaluation of all data obtained were carried out by TÜBİTAK UME experts using the same Institute infrastructure.

UME CRM 2282 certified reference material is intended to be used as a calibration standard in the determination of F<sup>-</sup>, Br<sup>-</sup> and PO<sub>4</sub><sup>3-</sup> anions.

## INTRODUCTION

Inorganic anions are controlled in wide variety of fields such as municipal water, drinking water, groundwater, surface water, wastewater, various food products and food additives, in accordance with both national and international legislations. In European Union countries, anion concentrations in various waters are controlled by directives 91/676/EEC, 98/83/EC, 2006/118/EC and 2009/54/EC. In parallel with these directives implemented in the European Union, in our country, spring waters, drinking water, drinking and utility water are regulated by the "Regulation on Water Intended for Human Consumption" [1] published in the Official Gazette No. 25730 dated 17.02.2005 and  $F^-$ ,  $NO_2^-$ ,  $NO_3^-$ ,  $F^-$ ,  $SO_4^{2-}$  and  $PO_4^{3-}$  concentrations in natural mineral waters are controlled within the scope of the "Regulation on Natural Mineral Waters" [2] published in the Official Gazette No.25657 dated 01.12.2004. In addition,  $F^-$  and  $SO_4^{2-}$  concentrations in groundwater have been regulated by the "Regulation on the Protection of Groundwater against Pollution and Deterioration" published in the Official Gazette No. 28257 dated 07.04.2012 [3]. Within the scope of the "Regulation on the Protection of Water from Agricultural Nitrate Pollution" published in the Official Gazette No. 29779 dated 23.07.2016,  $NO_3^-$  concentration in surface waters have been regulated. Within the scope of these regulations  $Br^-$ ,  $F^-$ ,  $NO_2^-$ ,  $NO_3^-$ ,  $F^-$ ,  $SO_4^{2-}$  and  $PO_4^{3-}$  determinations are made in various waters by Public Health Laboratories, State Hydraulic Works (DSI) Laboratories and many private laboratories. In the European Union, food additives are subject to legal regulations through Regulation (EC) No 1333/2008 [5] of the European Parliament and of the Council, and the Commission Regulation (EU) No 231/2012 [6] dated 09.03.2012, which determines the specifications for food additives listed in Annex II and Annex III of the Regulation (EC). In our country, the "Turkish Food Codex Food Additives Regulation" [7], published in the Official Gazette dated 30.06.2013, No. 28693, determines the conditions for the use of food additives (maximum quantities, limitations, etc.). Additionally, the qualities of food additives are specified in the "Regulation on the Specifications of Food Additives of the Turkish Food Codex," published in the Official Gazette dated 03.04.2017, No. 30027 (Duplicate) [8]. Within the scope of these regulations,  $NO_2^-$ ,  $NO_3^-$ ,  $F^-$ ,  $SO_4^{2-}$  and  $PO_4^{3-}$  determinations are conducted in various food products by public or private food control laboratories.

There are many public and private laboratories all over the world, conducting inorganic anion analyses on various food products and/or waters. Many of these laboratories are accredited and need internationally recognized calibration standards to ensure traceable measurement results.

With this project, SI traceable calibration standards have been produced to meet the calibration standard needs of laboratories conducting anion analysis. The aim is to ensure the metrological traceability of measurements conducted to the SI unit system through TÜBİTAK UME (National Metrology Institute of Türkiye) multi anion standards.

The nominal concentrations of  $F^-$ ,  $Br^-$  and  $PO_4^{3-}$  are 1000 mg/kg in UME CRM 2282 Multi Anion Standard II reference material and the mass fractions of each anions were assigned after the certification process. The mass fractions were determined using both gravimetric solution preparation with high-purity salts (Primary Reference Standards), whose purity were determined by TÜBİTAK UME, and High-Performance Ion Chromatography (HP-IC) [9] analysis of the solution. NaF, KBr and  $KH_2PO_4$  Primary Reference Standards that were used both for the preparation of UME CRM 2282 Multi Anion Standard II and the preparation of calibration standards for HP-IC measurements, were dried before use.

The certification process was carried out in accordance with the requirements of ISO 17034:2016 [10] standard and ISO Guide 35:2017 [11] document. The measurements in the certification process were performed in compliance with the requirements of ISO/IEC 17025:2017 [12] standard. The uncertainties

associated with the certification values were calculated in accordance with the JCGM 100:2008 Guide to the Expression of Uncertainty in Measurement (GUM) [13].

UME CRM 2282 Multi Anion Standard II certified reference material, which was produced by TÜBİTAK UME, is intended to be used as a calibration standard for the determination of F<sup>-</sup>, Br<sup>-</sup> and PO<sub>4</sub><sup>3-</sup> anions.

## PARTICIPANTS

Information on the organization involved in material processing, homogeneity, stability and characterization studies and other activities of the project for the production of certified reference materials is presented in Table 1.

**Table 1.** Organization Participating in the Production and Certification Processes

Activity	Laboratory/Organization
Material Processing	
Homogeneity Study	
Stability Studies	TÜBİTAK UME, National Metrology Institute, Gebze - Kocaeli, Türkiye
Characterization Study	
Project Management and Data Evaluation	

## MATERIAL PROCESSING

### *Material Supply*

The materials used in the production of UME CRM 2282 Multi Anion Standard II certified reference material are primary reference standards (PRS) of NaF, KBr, KH<sub>2</sub>PO<sub>4</sub> and ultrapure water.

### *Material Preparation*

The PRSs used in the production of the UME CRM 2282 Multi Anion Standard II certified reference material were dried under the conditions specified in Table 2 and stored in a desiccator (on a calcium sulfate desiccant) until the CRM preparation.

**Table 2.** Drying Conditions of Primary Reference Standards

PRS	Drying Temperature, °C	Drying Time, hours
NaF	500	15
KBr	500	15
KH <sub>2</sub> PO <sub>4</sub>	150	3

### *Gravimetric Preparation*

The nominal value for F<sup>-</sup>, Br<sup>-</sup> and PO<sub>4</sub><sup>3-</sup> in the Multi Anion II certified reference material was set at 1000 mg/kg. The amounts of PRSs required to achieve the targeted mass fractions were determined and

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weighing for each analyte was performed by substitution weighing. In substitution weighing procedure a standard and an unknown weight are compared to determine the average difference between the two weights. In this way, the calibration bias of the balance is eliminated since the balance is only used as a comparator. Traceability to the International System of Units (SI) is achieved directly via the mass standards when substitution weighing is used, thus reducing the uncertainty of the measurement result. Since the mass difference between the mass standards and the sample is very small, the linearity component of the balance uncertainty is negligible.

The weighing of the PRSs were carried out using the Sartorius MSA524S balance (calibrated by TÜBİTAK UME Mass Laboratory). OIML E2 class mass set (calibrated by TÜBİTAK UME Mass Laboratory) was utilized to weigh each PRS into separate beakers.

The 60 L HDPE carboy in which the reference material had been prepared was cleaned, checked for contamination and dried in a laminar flow cabinet. Empty weighing of the cleaned carboy (with its lid) was performed by substitution weighing by TÜBİTAK UME Mass Laboratory. After weighing, carboy was filled with 40 L of ultrapure water (18.2 M $\Omega$ -cm) from a water purification system with a point of use micro filter (0.22  $\mu$ m) and UV lamp in an ISO 7 class clean room. The pre weighed PRSs in the beakers were separately transferred into the carboy, the beakers were rinsed with ultrapure water at least 10 times and these washings were also transferred to the carboy. The carboy was filled to approximately 60 kg with ultrapure water, the lid was closed and transferred to TÜBİTAK UME Mass Laboratory. Before the final weighing, the solution completed to the target weight with ultra-pure water, was homogenized in the laboratory for at least 17 hours where the weighing would take place, allowing the CRM to reach the ambient temperature. The substitution weighing of the homogenized material was conducted the following day.

### ***Homogenization and Bottling***

The filling of the reference material into bottles was carried out in an ISO 7 class clean laboratory. Previously cleaned, subjected to contamination control, dried, and labeled using an automated labelling machine (Farmatek, Türkiye), total of 591 HDPE bottles (125 mL) were manually filled with CRM (100  $\pm$  5) mL in each bottle. Each bottle was placed in a labeled (with the same sequence number) aluminum sachet and sachet was closed by heating. The 591 bottles that placed inside the sachets (hereafter referred to as "unit") were transferred to the storage area under the specified storage conditions.

## **HOMOGENEITY**

For homogeneity tests, 10 units were selected according to the random stratified sample selection principle from the materials numbered according to the filling order. Random sample selection was performed using the "TRaNS" software developed by TÜBİTAK UME. This ensured that the selected samples were representative of the total number of samples. Homogeneity tests were performed by preparing 3 subsamples from each of the 10 units (19, 63, 140, 214, 272, 333, 398, 464, 522, and 568). The measurements for the homogeneity test were performed using the DIONEX ICS 3000 IC system and validated HP-IC method. The measurements were performed in random order to allow independent observation of any trends that may have emerged during filling and analysis.

The statistical evaluations of the mass fraction values for each parameter were performed using one-way analysis of variance (ANOVA). Prior to statistical evaluation with ANOVA, data distributions were examined. For this purpose, results obtained within units and between units results were checked for unimodal distribution using histograms. In addition, the Shapiro-Wilk test indicated that the obtained data generally followed normal distribution. These statistical tests were performed using Microsoft

Excel® templates that were created and validated by TÜBİTAK UME Chemistry Group. The assessments revealed that the distribution of 10 units (between units) obtained from the averages of each unit for F<sup>-</sup> and Br<sup>-</sup> could be considered as normal distribution, for PO<sub>4</sub><sup>3-</sup> could not be considered as normal distribution.

The measurement results were statistically evaluated for the presence of outliers and any trends depending on the analytical measurement and/or filling sequence and the results are given in Table 3. No trend has been detected for any parameter based on the filling sequence or analytical sequence.

When one-sided and two-sided Grubbs' tests were applied, no outliers were encountered for F<sup>-</sup>. The first and the second parallel measurements of unit 272 were observed as outliers for Br<sup>-</sup> and PO<sub>4</sub><sup>3-</sup>, respectively. Outliers were not included in the evaluation since they occurred in one parallel measurement of the units. After discarding the outlier for PO<sub>4</sub><sup>3-</sup>, the distribution was found to be a normal distribution.

**Table 3.** Statistical Evaluation of Homogeneity Test Results

Parameter	Any Trend?		Any Outlier?		Distribution
	Analytical Sequence	Filling Sequence	All Data	Unit Averages	Normal / Unimodal
F <sup>-</sup>	No	No	No	No	Yes / Yes
Br <sup>-</sup>	No	No	Yes	No	Yes / Yes
PO <sub>4</sub> <sup>3-</sup>	No	No	Yes	No	Yes / Yes

To calculate the within-unit ( $s_{wb}$ ) and between-unit ( $s_{bb}$ ) standard deviation using ANOVA, Equation (1) and Equation (2) are applied, respectively [14]:

$$s_{wb} = \sqrt{MS_{within}} \quad (1)$$

$MS_{within}$  : Mean squares of within-unit,

$s_{wb}$  : Equivalent to the s of the method, provided that subsamples are representative for the whole unit.

$$s_{bb} = \sqrt{\frac{MS_{between} - MS_{within}}{n}} \quad (2)$$

$MS_{between}$  : Mean squares between-unit,

$N$  : Number of replicates per unit.

In cases where the method repeatability is not good enough to determine the homogeneity of the material or due to random fluctuations during the measurement, the  $MS_{between}$  may be less than  $MS_{within}$ . Since  $s_{bb}$  cannot be calculated in these cases, the highest heterogeneity uncertainty,  $u^{*bb}$ , including method repeatability, is calculated using Equation (3).



$$u_{bb}^* = \frac{s_{wb}}{\sqrt{n}} \sqrt[4]{\frac{2}{v_{MS_{within}}}} \quad (3)$$

$v_{MS_{within}}$ : Degrees of freedom of  $MS_{within}$ .

The results obtained from the homogeneity study are given in Table 4. In the calculations performed by applying ANOVA, the larger of  $s_{bb}$  and  $u_{bb}^*$  values was taken as the homogeneity uncertainty component  $u_{bb}$ .

**Table 4.** Results of the Homogeneity Study

Parameter	$s_{wb,rel}$ (%)	$s_{bb,rel}$ (%)	$u_{bb,rel}^*$ (%)	$u_{bb,rel}$ (%)
F <sup>-</sup>	0.11	$MS_{between} < MS_{within}$	0.04	0.04
Br <sup>-</sup>	0.06	0.05	0.02	0.05
PO <sub>4</sub> <sup>-3</sup>	0.13	0.06	0.04	0.06

The heterogeneity-based uncertainty values obtained for all parameters to be certified in the candidate CRM were found to be below the targeted maximum uncertainty value of 0.2%. The graphs of the data obtained in homogeneity tests are presented in Annex 1.

## STABILITY

Stability studies were carried out by creating similar conditions in the laboratory environment to the environmental conditions that may occur during shipment of the certified reference material to the user (short-term stability, STS) and storage conditions (long-term stability, LTS). The 27 units for the short-term stability test and 21 units for the long-term stability test were determined by random stratified sample selection using TRaNS software. Stability measurements of the anions to be certified were carried out with the DIONEX ICS 3000 IC system using the HP-IC method.

For short-term stability studies, the test temperatures were +25 °C and +45 °C, and the durations were 1, 2, 3 and 4 weeks. For each time period at both temperatures, 3 units were placed in the test chamber/oven at that temperature. In the stability test, 3 units were reserved for the reference point and these units were placed directly to the reference temperature of +4 °C. At the end of each test period, three units from each of the two temperature environments were transferred to the reference temperature. At the end of the four-week test period, all units that were transferred to the reference temperature were analyzed isochronously along with the reference units.

The long-term stability test was performed using the classical design at +4 °C. All selected test samples were placed at the reference temperature (+4 °C). The initial value was determined by 3 parallel analyses from each 3 reference units at the beginning of the study. Then, 3 units were taken from the LTS samples at the reference temperature on days 71, 177, 246, 345, 490 and 603 and analyzed in 3 parallels from each unit.

In order to distinguish between possible trends due to filling or test time and trends due to analytical sequence, the measurements were conducted with an analytical measurement sequence in random order of test time and fill order for both stability tests. Statistical calculations of the obtained data were

performed using Microsoft Excel® templates created and validated by TÜBİTAK UME Chemistry Group experts.

### Short Term Stability Study Results

The isochronous measurement results of the short-term stability study were first grouped according to the same time points and evaluated for each time point. These evaluations were performed separately for both temperatures.

The measured values obtained for each time period were evaluated for outliers at 95% and 99% confidence levels by applying Grubb's test to determine their compatibility with other values in that temperature group. Outliers were found in F<sup>-</sup>, Br<sup>-</sup> and PO<sub>4</sub><sup>3-</sup> data. The third parallel measurement of unit 254 (*t* = 1 week) in the +45 °C test for F<sup>-</sup> and Br<sup>-</sup>, the third parallel measurement of unit 328 (reference) and the second parallel measurement of unit 585 (*t* = 1 weeks) in the +25 °C test for PO<sub>4</sub><sup>3-</sup> were identified as outliers. Outliers were not included in the evaluation as they occurred in one parallel measurement.

In the evaluation of the short-term stability data, the values calculated for each time point were plotted against time and the relationship between the variables was examined to determine whether there was any significant change in concentration values against time (regression analysis). Linear graphs were plotted for each anion and the t-test (two-tailed *t*<sub>crit</sub> value) was used to test whether the slopes were significantly different from zero at 95% confidence level ( $\alpha = 0.05$ ). The slopes are not significantly different from zero except for Br<sup>-</sup>. This observed slope for Br<sup>-</sup> was included in the uncertainty. The short-term stability test results are given in Table 5 and the graphs are given in Annex 2.

Uncertainties for short-term stability test were calculated using Equation (4), taking into account the uncertainty of the slope and the maximum exposure time [15].

$$u_{sts,rel} = \frac{RSD}{\sqrt{\sum(t_i - \bar{t})^2}} \times t \quad (4)$$

where,

*RSD* : Relative standard deviation of the points on the regression line,

*t<sub>i</sub>* : Time point for each replicate,

$\bar{t}$  : Mean of all time points,

*t* : Maximum time suggested for transfer (2 weeks).

**Table 5.** Results of Short Term Stability Tests and Uncertainty Values for 2 Weeks

Parameter	Number of outliers in 95% confidence level		Is there a significant trend in 95% confidence interval?		+25 °C 2 weeks <i>u</i> <sub>sts,rel</sub> (%)	+45 °C 2 weeks <i>u</i> <sub>sts,rel</sub> (%)
	+25 °C	+45 °C	+25 °C	+45 °C		
F <sup>-</sup>	-	1	No	No	0.03	0.03
Br <sup>-</sup>	-	1	Yes	Yes	0.04	0.04 <sup>[1]</sup>
PO <sub>4</sub> <sup>3-</sup>	1	1	No	No	0.05	0.06

[1] *u*<sub>sts</sub>, calculated taking into account the degradation (*u*<sub>sts</sub>=Slope/ $\sqrt{3}$ )

The certified reference material was found to be stable at both +25 °C and +45 °C for 2 weeks. As a result of this study, it was concluded that the samples can be dispatched to the end user without any

cooling, provided that the temperature does not exceed +45 °C and the duration does not exceed 2 weeks. The uncertainties given in the certificate include the STS uncertainties calculated considering the maximum transfer temperature of +45 °C and the transfer time of 2 weeks.

### **Long Term Stability Study Results**

The shelf life of the produced CRMs are determined according to the results of long-term stability studies. As mentioned above, 3 units were used for each time point and 3 independent parallel samples were prepared from each unit and long-term stability test measurements were performed. In this study, the test period was designed to cover a period of 20 months with a total of 7 time points. The graphs of the results are given in Annex 3. The error lines at each time point were calculated as the standard deviation of the three results obtained for each of the three units.

Outliers were observed in F<sup>-</sup> (95% and 99%), Br<sup>-</sup> (95% and 99%) and PO<sub>4</sub><sup>3-</sup> (95% and 99%) data by applying one-way and two-way Grubb's test at 95% and 99% confidence levels. One of the three parallel measurements for the units 70 and 239 for F<sup>-</sup>, 70 and 299 for Br<sup>-</sup>, 44, 70, 239, 299 and 322 for PO<sub>4</sub><sup>3-</sup> were identified as outliers. Since all outliers were only in one parallel measurement of the units, these values were not included in the data set.

The values were analyzed by plotting the regression line on the concentration versus time graph. At the temperature tested, (+4 ± 3) °C, the t-test at 95% confidence level for the plotted regression lines showed that the slope for Br<sup>-</sup> was not significantly different from zero, but slopes for F<sup>-</sup> and PO<sub>4</sub><sup>3-</sup> were different from zero.

Long-term stability is one of the four parameters that contribute to the total uncertainty. The uncertainty value due to long-term stability of the certified reference material was calculated using Equation (5) [14].

$$u_{lts,rel} = \frac{RSD}{\sqrt{\sum(t_i - \bar{t})^2}} \times t \quad (5)$$

where,

*RSD* : The relative standard deviation of the points on the regression line,

*t<sub>i</sub>* : Time point for each replicate,

$\bar{t}$  : Mean of all time points,

*t* : Proposed shelf life at +4 °C: 24 months.

The calculated results are given in Table 6 and the graphs are presented in Annex 3. The uncertainty contribution *u<sub>lts</sub>* was calculated for 24 months at +4°C. To ensure stability beyond the shelf life, reassessments will be made periodically based on the results of post-certification monitoring (PCM).

**Table 6.** Long Term Stability Test Results and Uncertainty Values for 24 Months

Parameter	Number of outliers in 95% confidence level	Is the slope different from zero at 95% confidence level?	+4 °C, 24 ay $u_{lts,rel}$ (%)
F <sup>-</sup>	2	Yes	0.23 <sup>[1]</sup>
Br <sup>-</sup>	2	No	0.09
PO <sub>4</sub> <sup>-3</sup>	5	Yes	0.23 <sup>[1]</sup>

[1]  $u_{lts}$ , calculated taking into account the degradation ( $u_{lts} = \text{Slope} / \sqrt{3}$ )

## CHARACTERIZATION

According to ISO 17034 standard, the characterization study can be carried out in different ways. One of these designs is a characterization study using a reference method in a single laboratory. Another design is based on a weighted or unweighted average of the results of different methods. In this production process, characterization was performed using the average of gravimetric solution preparation and HP-IC analysis results for all anions.

In the gravimetric solution preparation, the weighing of the salts, the weighing of the empty carboy and final solution were carried out metrologically. The HP-IC measurement of the characterization study was carried out in 6 units randomly selected among the units using the TRaNS program, on two different days (3 separate units each day and 3 parallel measurements from each unit). The measurements were performed using the validated HP-IC method. Data from the homogeneity study were also included in the characterization study and the average of the two-day characterization study data and homogeneity data were used. NIST 3100 series standard solutions were used as verification solutions to check the accuracy and the standards used are given in Table 7.

**Table 7.** Standards Used for Method Verification

Parameter	Name	Code	Traceability
F <sup>-</sup>	F <sup>-</sup> Anion Standard Solution	NIST SRM 3183	SI
Br <sup>-</sup>	Br <sup>-</sup> Anion Standard Solution	NIST SRM 3184	SI
PO <sub>4</sub> <sup>-3</sup>	PO <sub>4</sub> <sup>-3</sup> Anion Standard Solution	NIST SRM 3186	SI

Uncertainty calculations for gravimetric solution preparation and HP-IC were performed in accordance with "Guide to the Expression of Uncertainty in Measurements (GUM)" [13] and EURACHEM/CITAC "Guide Quantifying Uncertainty in Analytical Measurement". Characterization uncertainties were combined using equations (6) – (8), which were given by M. S. Levenson et al. (2000) [16].

$$u(B) = \frac{|X_{m1} - X_{m2}|}{2\sqrt{3}} \quad (6)$$

$$u(X) = \sqrt{\left(\frac{1}{2}\right)^2 u^2(m_1) + \left(\frac{1}{2}\right)^2 u^2(m_2)} \quad (7)$$

$$u_{char} = \sqrt{u^2(X) + u^2(B)} \quad (8)$$

where,

- $u(B)$  : Standard uncertainty due to the difference in the results of the two methods,  
 $u(X)$  : Standard uncertainty obtained by combining the uncertainty of the two methods,  
 $u_{char}$  : Characterization standard uncertainty.

## PROPERTY VALUE AND UNCERTAINTY ASSIGNMENT

The assignment of certified property values and their associated uncertainties was carried out by evaluating contributions from data obtained in characterization studies for each parameter, along with the accompanying uncertainty values. Additionally, uncertainties calculated from the data obtained in homogeneity and stability tests were considered in this process.

The property values to be given in the certificate were calculated by taking the arithmetic mean of the value obtained from the gravimetric solution preparations and the HP-IC measurement data of the characterization studies (Equation (9)).

$$C_{CRM} = (C_{m1} + C_{m2})/2 \quad (9)$$

Where  $C_{CRM}$  stands for the certified value.

In the calculation of the uncertainty of the property values, the contributions of characterization, homogeneity, short-term and long-term stability uncertainties are combined as given in Equation (10).

$$u_{CRM} = \sqrt{u_{char}^2 + u_{bb}^2 + u_{sts}^2 + u_{its}^2} \quad (10)$$

Where  $u_{CRM}$  is the combined standard uncertainty of the property value. This value is converted into the expanded uncertainty of the certified value by multiplying it with the coverage factor  $k$  as shown in Equation (11).

$$U_{CRM} = k \cdot u_{CRM} \quad (11)$$

The uncertainty values given with the certified values are the expanded uncertainties, which were calculated using a coverage factor  $k = 2$ , corresponding to a confidence level of approximately 95%. The certified values and the uncertainties for each parameter are given in Table 8. The contributions of uncertainty components to the combined uncertainty are given in Table 9.

**Table 8.** Certified Values and Uncertainties

Parameter	Certified Value (mg/kg)	$U_{CRM}$ (mg/kg) ( $k = 2$ )	$U_{CRM}$ Relative (%) ( $k = 2$ )
F <sup>-</sup>	993.8	5.3	0.53
Br <sup>-</sup>	998.5	2.8	0.28
PO <sub>4</sub> <sup>-3</sup>	998.9	5.2	0.52

**Table 9.** Percent Contribution of Uncertainty Components to the Combined Uncertainty

Parameter	$u_{char}$ (%)	$u_{bb}$ (%)	$u_{lts}$ (%)	$u_{sts}$ (%)
F <sup>-</sup>	22.6	2.0	73.8	1.6
Br <sup>-</sup>	30.2	12.3	47.7	9.7
PO <sub>4</sub> <sup>-3</sup>	12.3	4.8	78.2	4.7

## INFORMATIVE VALUE

Density measurements of UME CRM 2282 were made on 3 different units and 3 parallel measurements from each unit. The result and corresponding uncertainty value are given in Table 10 for informational purposes.

**Table 10.** Density for Informative Purposes

Parameter	Value	$U$ ( $k = 2$ )
Density (20 °C)	1002.625 kg/m <sup>3</sup>	0.044 kg/m <sup>3</sup>

## TRACEABILITY

In this study, validated methods were used for all certification measurements including homogeneity and stability tests. Samples were prepared gravimetrically for both preparation and measurements of the reference material. Weighings were performed using calibrated balances traceable to national measurement standards and the balances were checked with appropriate mass set weights. The balances and mass set weights used were traceable to national measurement standards, realizing units defined in the International System of Units (SI). In the characterization, homogeneity and stability studies, SI traceability was ensured by using Primary Reference Standards (PRS) whose purity was determined and certified by TÜBİTAK UME using the method of determination of impurities. Primary Reference Standards that were used as traceability source in certification studies are given in Table 11.

**Table 11.** Primary Reference Standards Used as Traceability Sources in Certification Studies

Parameter	Name	Purity and Uncertainty, % ( $k = 2$ )	Traceability
F <sup>-</sup>	NaF Primary Reference Standard	99.456 ± 0.032	SI
Br <sup>-</sup>	KBr Primary Reference Standard	99.9810 ± 0.0038	SI
PO <sub>4</sub> <sup>3-</sup>	KH <sub>2</sub> PO <sub>4</sub> Primary Reference Standard	99.9926 ± 0.0021	SI

## INSTRUCTIONS FOR USE

### Intended Use

This material is intended to be used as a calibration standard for the determination of F<sup>-</sup>, Br<sup>-</sup> and PO<sub>4</sub><sup>3-</sup> anions.

### Storage Conditions

The material should be stored at (+2 - +8) °C before and after use. TÜBİTAK UME cannot be held responsible for any changes that may occur in the material due to non-compliance with the storage conditions and instructions for use.

### Safety Precautions

The material is intended for laboratory use only. General laboratory precautions should be followed when storing and handling the material. It is recommended to use and dispose the material according to current safety rules. The Safety Data Sheet (SDS) should be read carefully before using the material.

### Material Handling

The bottle should be allowed to reach equilibrium with the ambient temperature before opening. The bottle should be shaken before opening. All precautions must be taken to prevent contamination and evaporation of the material during opening and use of the material.

### Minimum Sample Intake

The homogeneity of the solution for F<sup>-</sup>, Br<sup>-</sup> and PO<sub>4</sub><sup>3-</sup> has been demonstrated by within and between bottle homogeneity tests. The minimum sample intake amount should be determined by the end user based on the measurement capability, taking into account the effect on the uncertainty of the working solution that will be prepared.

## REFERENCES

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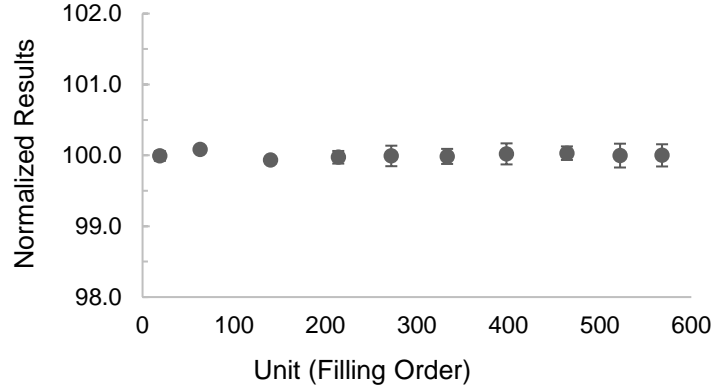
## REVISION HISTORY

Date	Remarks
12.04.2023	First Issue.

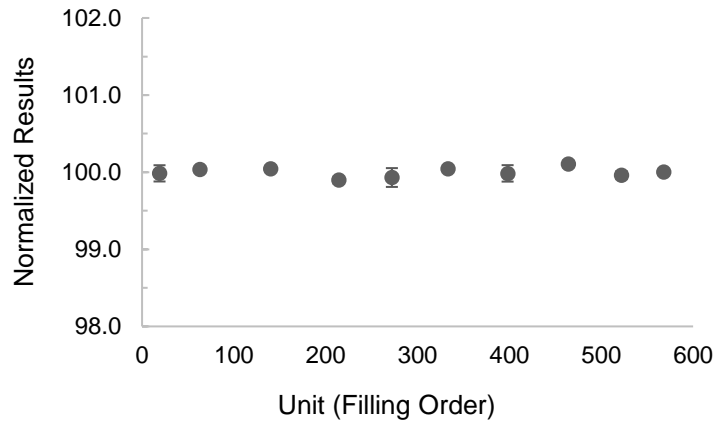


## ANNEXES

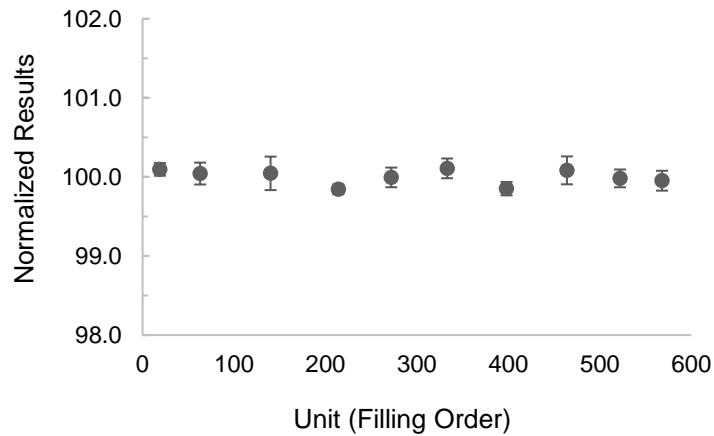
### Annex 1. Graphs for Homogeneity Studies



**Figure A1.1.** Homogeneity Plot for F<sup>-</sup>

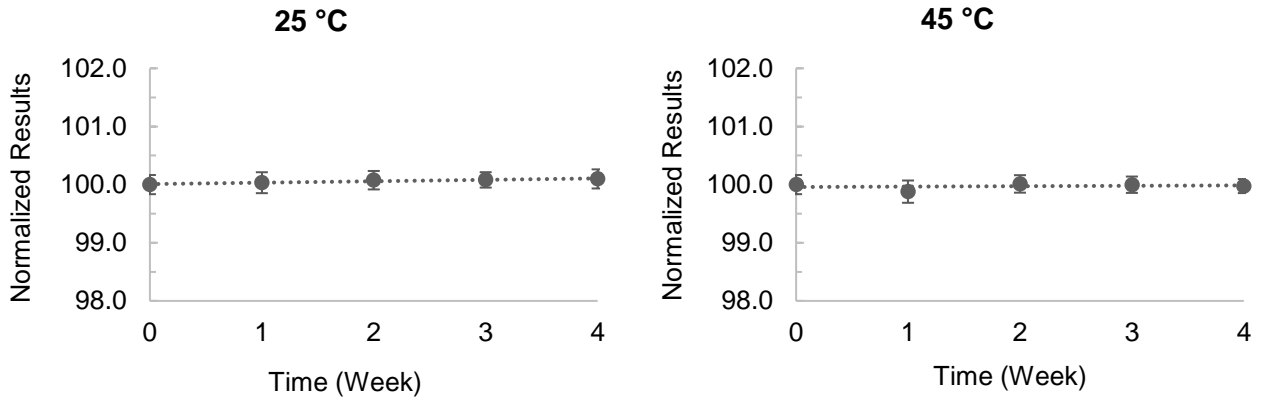


**Figure A1.2.** Homogeneity Plot for Br<sup>-</sup>

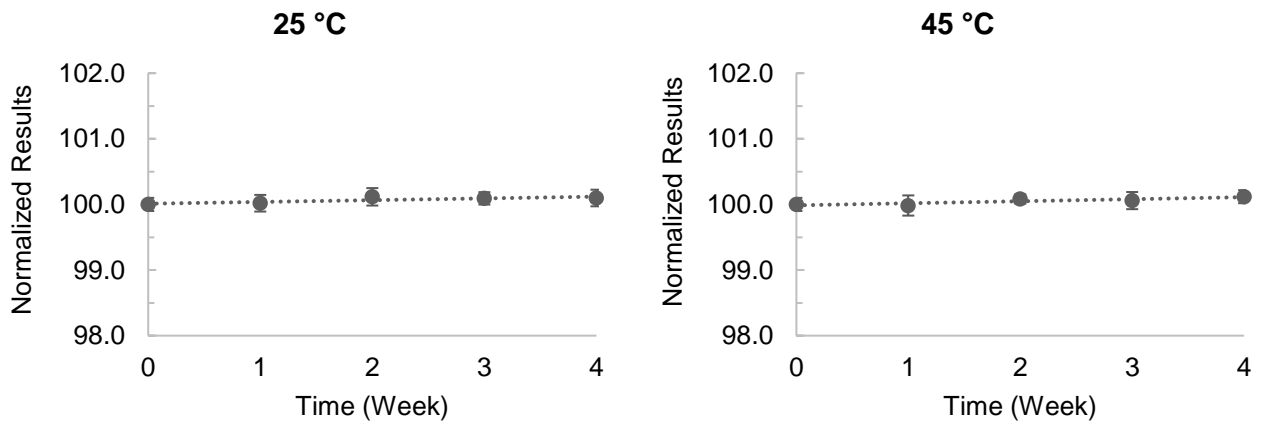


**Figure A1.3.** Homogeneity Plot for PO<sub>4</sub><sup>3-</sup>

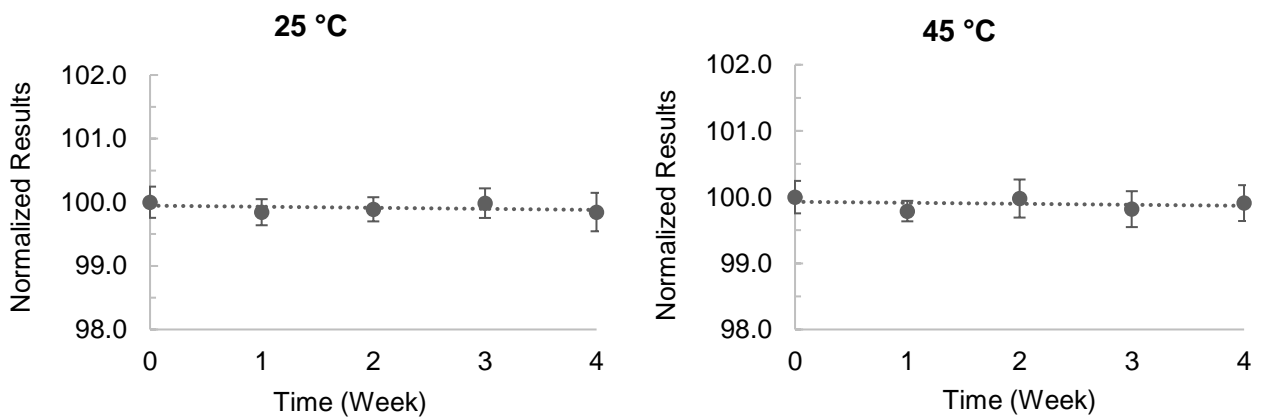
## Annex 2. Graphs for Short Term Stability Studies



**Figure A2.1.** STS Plots for  $F^-$  at +25 °C and +45 °C

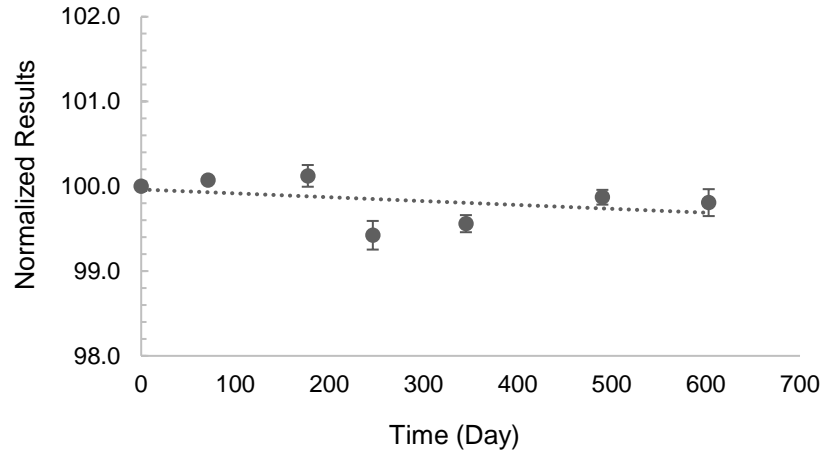


**Figure A2.2.** STS Plots for  $Br$  at +25 °C and +45 °C

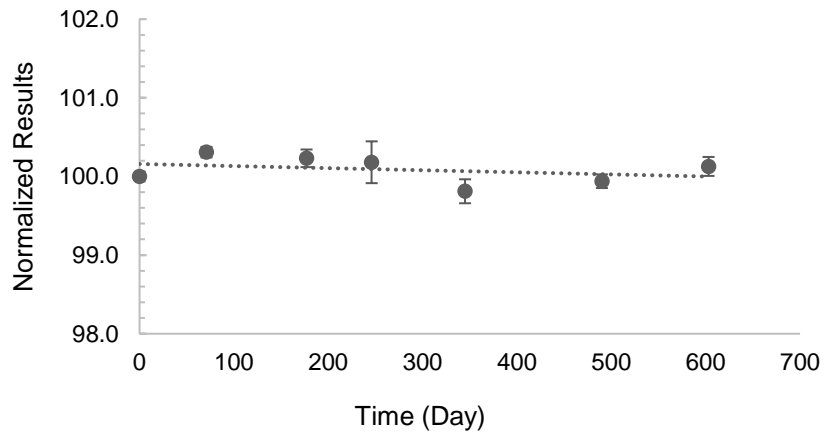


**Figure A2.3.** STS Plots for  $PO_4^{3-}$  at +25 °C and +45 °C

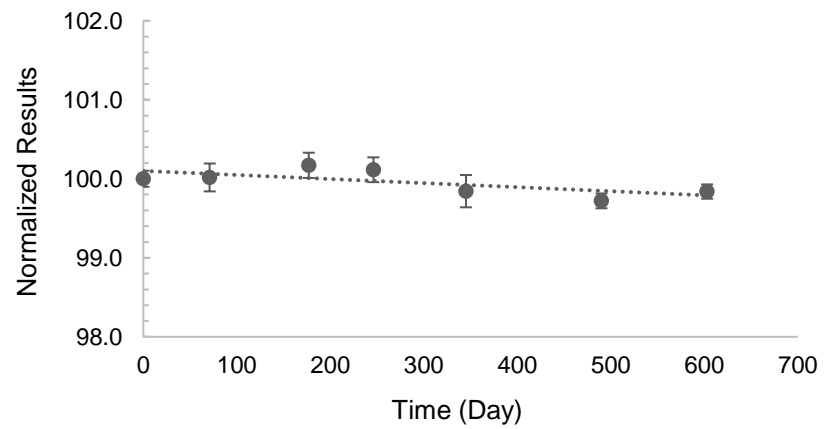
### Annex 3. Graphs for Long Term Stability Studies



**Figure A3.1.** LTS Plot for F<sup>-</sup> at +4 °C



**Figure A3.2** LTS Plot for Br at +4 °C



**Figure A3.3.** LTS Plot for PO<sub>4</sub><sup>3-</sup> at +4 °C