
Application Bulletin

Of interest to: General analytical chemistry B 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16

Validation of Metrohm VA instruments using Standard Operating Procedures

Summary

GLP (Good Laboratory Practice) requirements include the periodic check of analytical instruments for reproducibility and accuracy using **standard operating procedures (SOP)**.

The user is advised to validate the Metrohm VA instruments as a complete, integrated voltammetry system, i.e. to perform a voltammetric determination using standard solutions of known content and critically assess the results using statistical methods.

Checking of the electronic and mechanical components of measuring instruments can and should be undertaken by qualified personnel of the manufacturing company as part of regular servicing. All Metrohm instruments are provided with start-up test routines. Each time the instrument is switched on, these test routines check whether the relevant assemblies are working correctly. If no error message is displayed, it can be assumed that the instrument is functioning faultlessly. Metrohm instruments are also supplied with built-in diagnostic programs, which enable the user to check the functioning of certain components in the event of malfunctions or erratic behaviour and to localise the fault. Diagnostic programs may also be integrated in a validation procedure.

The procedure described below is meant as a guideline for the preparation of standard operating procedures for checking a voltammetry system comprising the VA instrument and possibly a dispensing unit or an Autosampler. The limiting values specified must be considered as recommendations. Specific limiting values must be defined in the particular standard operating procedure taking into account in-house requirements concerning the accuracy of the analysis system.

Application range

These test specifications are applicable to the following Metrohm VA instruments:

693 VA Processor with 694 VA Stand
746 VA Trace Analyzer with 747 VA Stand
757 VA Computrace

Test intervals

Annually repeated tests of VA instruments appear appropriate. A special validation is advisable whenever one or more components of the VA system have been replaced.

Internal instrument test routines

The Metrohm VA instruments are equipped with an internal instrument start-up test and test routines. In the start-up test of processor-controlled instruments, the display elements are checked and the contents of the program memories are tested by means of a checksum test. Proper functioning of the data memory area is tested with a write/read test. In the case of the 693 VA Processor or 746 VA Trace Analyzer the presence and operational readiness of a 685 Dosimat, 700 Dosino, or 695 VA Autosampler is verified.

If the VA instruments are regularly serviced, it is generally possible to dispense with the specific validation of the instrument electronics.

Maintenance/Service

Careful maintenance and cleaning are indispensable requirements for the GLP-conforming operation of all instruments used in the laboratory. Particular attention should also be paid to the correct handling of such instruments. The instructions for use supplied with the instrument should be accessible to all workers in the laboratory. We also recommend regular servicing of all measuring instruments involved once a year. Many Metrohm agencies offer favourably priced service contracts for your instruments.

Method

In many cases the daily work involves only a few specific voltammetric methods. For the validation of the VA instrument, it is advisable to select a voltammetric method as similar as possible to those used in one of the frequently employed methods. In addition, it should be possible to eliminate any error sources due to the method.

Generally, Metrohm recommends validating the instrument in two steps:

1. Electronic validation without the electrodes, using the built in dummy cell.
In this step the complete electrical part of the voltammetry system is tested. If the result of the tests fulfil the specifications defined below, perfect functioning has been proved.

2. Chemical validation including the electrodes, applying the method described below using the Pb ion standard solution (6.2301.100) and the KCl electrolyte (6.2308.020). Both solutions are delivered with the instrument.

This step confirms the correct functioning of the entire voltammetry system including the electrodes.

VA Systems with Dosimats, Dosinos, or a Autosampler

If dispensing units are used for automatic addition of auxiliary solutions or standard solutions or if a Autosampler is present, it is recommended to validate the VA system including these additional peripheral instruments. Instead of introducing the necessary solutions manually into the measuring vessel, the dispensing units should be used to add the solutions automatically.

By this procedure, the accuracy of the dispensing units ist tested as well.

Validation of older Metrohm VA instruments

The following older Metrohm VA instruments can be validated in a similar way to the procedures described:

506 and 626 Polarecord with 663 VA Stand
646 VA Processor with 647 VA Stand

Electronic validation (dummy cell test)

Older instruments only have a dummy cell for a linear test. It can be built in or is delivered separately. For those instruments Metrohm recommends use of the polarogram simulator (6.496.8380). The polarogram simulator also gives peak-shaped voltammetric signals. In addition it is possible to simulate standard additions.

Using the dummy cell and the polarogram simulator, the electronic validation of the VA instrument can be performed as in the methods described below.

Detailed procedures for a dummy cell test are described in the manual of the instrument concerned and in the manual of the polarogram simulator.

Chemical validation with ion standard solution

The procedure for chemical validation can easily be transferred to older Metrohm VA equipment. The same composition of the measurement solution and same voltammetric parameters can be used for the test procedure.

Chemicals required

- Ultrapure water
- Pb ion standard solution (6.2301.100)
- KCl electrolyte (6.2308.020).

Requirements

The pipettes should be validated first.

New reference electrodes should be prepared as follows: Fill the reference system with KCl and soak the completely prepared electrode in the electrolyte for several hours. The dry diaphragm needs some time to be saturated completely with electrolyte.

Part 1: 693 VA Processor / 746 VA Trace Analyzer

Accessory required

- Pipettes for 20 mL, 0.5 mL, and 100 µL.

Electronic validation (dummy cell test)**Procedure:**

1. Attach the electrode cables of the VA Stand at the connectors of the dummy cell as follows:
2. Cable AE → Connector AE
3. Cable RE → Connector RE
4. Load the method „Test694.mth“ or „Test747.mth“ from the Method Storage.
5. Start the method and follow the instructions appearing on the screen.
6. Connect cable WE → Connector WE-L.
7. A linearity test will be done and a diagonal line will be registered.
8. Connect cable WE → Connector WE-D.
9. A peak will be registered.
10. The registered voltammograms will be printed.

Voltammetric settings:

Linearity test (connection WE-L)		Peak test (connection WE-D)	
Electrode Type	RDE	Electrode Type	RDE
Measurement Mode	DCT	Measurement Mode	AC1
Start Potential	-200 mV	Start Potential	-200 mV
End Potential	+200 mV	End Potential	-800 mV
voltage step	6 mV	Voltage step	10 mV
voltage step time	0.1 sec	Voltage step time	0.2 sec
		U amplitude	25 mV
		Phase angle	0°
		Modul. frequency	25 Hz
		Prep. cycles	0
		Meas. cycles	2

The detailed procedure is described in the manual of the instrument:

693 VA Processor: chapter 7.7.1.
(Method „Test694.mth“)

746 VA Trace Analyzer: chapter 7.7.1.
(Method „Test747.mth“)

Interpretation of the results

The two diagrams recorded should be assessed as follows:

Linearity Test (Diagonal):

The diagonal must be straight and smooth:

Current at -200 mV	-2 ± 0.4 µA
Current at +200 mV	+2 ± 0.4 µA

Peak test:

A symmetrical, bell-shaped peak should be obtained. The evaluation must provide a result for the peak voltage and the peak current, which are printed out in the full report. Most important are the peak potential and the shape of the peak. The line must be smooth.

Peak voltage	-500 ± 50 mV
Peak current	+250 nA ... +1250 nA

In case one of the tests exceeds the defined tolerances, the instrument is probably not functioning properly. Please contact your local Metrohm representative.

Chemical validation with ion standard solution

Preparing the sample solution

1. Add 20 mL of ultrapure water into the measuring vessel.
2. Add 0.5 mL of KCl electrolyte ($c(\text{KCl})=3 \text{ mol/L}$) into the measuring vessel.
3. Add 100 μL of Pb ion standard solution ($c(\text{Pb})=1 \text{ g/L}$) into the measuring vessel.

Perform the measurement

1. Load the method „TestPb.mth“ from the Method Storage.
2. Start the method.
3. The solution will be degassed and the polarogram will be registered three times.
4. Add 100 μL of Pb ion standard solution ($c(\text{Pb})=1 \text{ g/L}$) into the measuring vessel and press <Enter>.
5. The polarogram of the first standard addition will be registered three times.
6. Add 100 μL of Pb ion standard solution ($c(\text{Pb})=1 \text{ g/L}$) into the measuring vessel and press <Enter>.
7. The polarogram of the second standard addition will be registered three times.
8. The full report and the voltammograms will be printed.

Voltammetric settings:

Electrode Type	DME
Measurement Mode	DP
Start Potential	-200 mV
End Potential	-550 mV
voltage step	6 mV
voltage step time	0.4 sec

The detailed procedure is described in the manual of the instrument:

Chapters 7.7.2. to 7.7.5.

(Method „TestPb.mth“)

Interpretation of the results

The relevant parameters for the validation of measuring instruments are the accuracy and the scatter of the result.

Both values are calculated automatically by the microprocessor-controlled Metrohm VA instruments 693 VA Processor and 746 VA Trace Analyzer.

To assess the recorded lead determination, the results printed in the full report for the concentration of lead and its total scatter are used. The limit values of these two results depend greatly on the care taken in the preparation of the analysis solution and in the dispensing of the standard addition solutions.

If the procedure is carried out properly and carefully, the following results should be obtained:

Accuracy	95% ... 105%	Final Result	1 ± 0.05 g/L
Scatter	≤ ±3%	Res. Dev.	≤ ±0.03 g/L (± 3%)

Part 2: 757 VA Computrace

Accessory required

- Pipettes for 20 mL, 0.5 mL, and 100 µL.

Electronic validation (dummy cell test)

Procedure

1. Attach the electrode cables of the VA Computrace at the connectors of the dummy cell as follows:
2. Cable AE → Connector AE
3. Cable RE → Connector RE
4. Connect cable WE → Connector WE-L.
5. Load the method „Test757_L“ from the directory „methods“.
6. Start the method.
7. A linearity test will be done and a diagonal line will be registered and printed.
8. Connect cable WE → Connector WE-D.
9. Load the method „Test757_D“ from the directory „methods“.
10. A peak will be registered and printed.

Voltammetric settings:

Linearity test (connection WE-L)		Peak test (connection WE-D)	
Electrode Type	RDE	Electrode Type	RDE
Measurement Mode	DCT	Measurement Mode	DP
Start Potential	-200 mV	Start Potential	-200 mV
End Potential	+200 mV	End Potential	-800 mV
voltage step	6 mV	Voltage step	10 mV
voltage step time	0.1 sec	Voltage step time	0.4 sec
		Pulse amplitude	50 mV
		Pulse time	40 ms

The detailed procedure is described in chapter 7.9. of the manual of the software.

„Perform a linearity test with the dummy cell“ (Method „Test757_L“)

„Perform a peak test with the dummy cell“ (Method „Test757_D“).

Interpretation of the results

Two diagrams are recorded and should be assessed as follows:

Linearity Test (Diagonal):

The diagonal must be straight and smooth:

Current at -200 mV	$-2 \pm 0.4 \mu\text{A}$
Current at +200 mV	$+2 \pm 0.4 \mu\text{A}$

Peak test:

A symmetrical, bell-shaped peak should be obtained. The evaluation must provide a result for the peak voltage and the peak current, which are printed out in the full report.

Peak voltage	$-500 \pm 50 \text{ mV}$
Peak current	$-3 \pm 1 \mu\text{A}$

In case one of the tests exceeds the defined tolerances, the instrument is probably not functioning properly. Please contact your local Metrohm representative.

Chemical validation with ion standard solution:
Preparing the sample solution

1. Add 20 mL of ultrapure water into the measuring vessel.
2. Add 0.5 mL of KCl electrolyte ($c(\text{KCl})=3 \text{ mol/L}$) into the measuring vessel.
3. Add 100 μL of Pb ion standard solution ($c(\text{Pb})=1 \text{ g/L}$) into the measuring vessel.

Perform the measurement

1. Load the method „Test Pb in ion standard solution.mth“ from the directory „methods“.
2. Start the method.
3. The solution will be degassed and the polarogram will be registered three times.
4. Add 100 µL of Pb ion standard solution (c(Pb)=1 g/L) into the measuring vessel and press <Enter>.
5. The polarogram of the first standard addition will be registered three times.
6. Add 100 µL of Pb ion standard solution (c(Pb)=1 g/L) into the measuring vessel and press <Enter>.
7. The polarogram of the second standard addition will be registered three times.
8. The full report and the voltammograms will be printed.

Voltammetric settings:

Electrode Type	DME
Measurement Mode	DP
Start Potential	-200 mV
End Potential	-550 mV
voltage step	6 mV
voltage step time	0.4 sec

The detailed procedure is described in the manual of the instrument:

Chapter 7.9. „Perform a GLP test“

(Method „Test Pb in ion standard solution.mth“)

Interpretation of the results

The relevant parameters for the validation of measuring instruments are the reproducibility (precision, scatter) and the accuracy of the measurement results.

Both values are calculated automatically by the PC software of the 757 VA Computrace.

To assess the recorded lead determination, the results printed in the full report for the concentration of lead and its total scatter are used. The limit values of these two results depend greatly on the care taken in the preparation of the analysis solution and in the dispensing of the standard addition solutions.

If the procedure is carried out properly and carefully, the following results should be obtained:

Accuracy	95% ... 105%	Final Result	1 ± 0.05 g/L
Scatter	≤ ±3%	Res. Dev.	≤ ±0.03 g/L (± 3%)

Part 3: General Information

Recommendations for troubleshooting

For troubleshooting see also the manuals of the particular instruments. Additional information is also available in the Metrohm Monograph "First aid for polarography and voltammetry" (8.693.1073). The following lists are not complete and only show examples for possible causes for errors.

Possible error sources

Measuring vessel	contaminated
Pipettes	contaminated, inaccurate
Water, electrolyte	contaminated
Working Electrode	capillary blocked or defective
	needle dirty or deformed
	Hg oxidized
	loose contact at connector
Reference Electrode	electrode empty
	blocked diaphragm
	loose contact at connector
Auxiliary Electrode	loose contact at connector

With systematic deviation too high (accuracy unsatisfactory)

- Check ultrapure water for impurities.
- Clean pipettes.
- Check pipettes for accuracy and reproducibility.
- Clean measuring vessel.
- Check electrodes and voltammetry parameters.
- Check electrolyte for impurities.
- Check nitrogen for purity.
- Increase degassing time.

With rel. standard deviation too high (poor reproducibility))

- Clean pipettes.
- Check pipettes for accuracy and reproducibility.
- Check pressure gauge for accuracy.
- Check electrodes and voltammetry parameters.
- Check gas cylinder for remaining minimum pressure.

Procedure with values not conforming to specifications

All non-conforming values must be commented on in the validation record and the subsequent procedure noted.

If excessive deviations are found, the different points under the section "Recommendations for troubleshooting" must be carefully checked and the interferences eliminated. It is essential to repeat the validation. If unsatisfactory results are still obtained when the test series is repeated, the validation must be performed again by a different person.

If doubt exists regarding the precision of the dispensing unit, this can be checked separately (see Metrohm Application Bulletin No. 238).

Literature

Further information on voltammetry can be found in the following publications:

- Metrohm Application Bulletin No. 238, Check of Dosimat according to GLP/ISO
- Metrohm Monograph "First Aid in Polarography and Voltammetry"

On the following pages you will find an example of a validation.

The last page can be used as a master for copies of the validation record.

Validation Record		Company :	<i>Metrohm Ltd</i>
VA Instrument		Division :	<i>Application lab</i>
Date:	<i>14.01.1999</i>	User:	<i>U. Loyall</i>
Time:	<i>17:00</i>		
Instrument :	<i>757 VA Computrace</i>	Serial number:	<i>02135</i>

Electronic Validation (Dummy Cell Test)

Linearity Test:	Measured values	Tolerances	Test passed	
Current at -200 mV	<i>-2 μA</i>	<i>-1.6 μA ... -2.4 μA</i>	yes <input checked="" type="checkbox"/>	no <input type="checkbox"/>
Current at +200 mV	<i>+2 μA</i>	<i>+1.6 μA ... +2.4 μA</i>	yes <input checked="" type="checkbox"/>	no <input type="checkbox"/>

Peak Test:	Measured values	Tolerances	Test passed	
Peak voltage	<i>-497 mV</i>	<i>-450 mV ... -550 mV</i>	yes <input checked="" type="checkbox"/>	no <input type="checkbox"/>
Peak current	<i>-2.89 μA</i>	<i>746:+250 nA...+1250 nA</i>		
		<i>757: -2 μA ... -4 μA</i>	yes <input checked="" type="checkbox"/>	no <input type="checkbox"/>

Chemical Validation

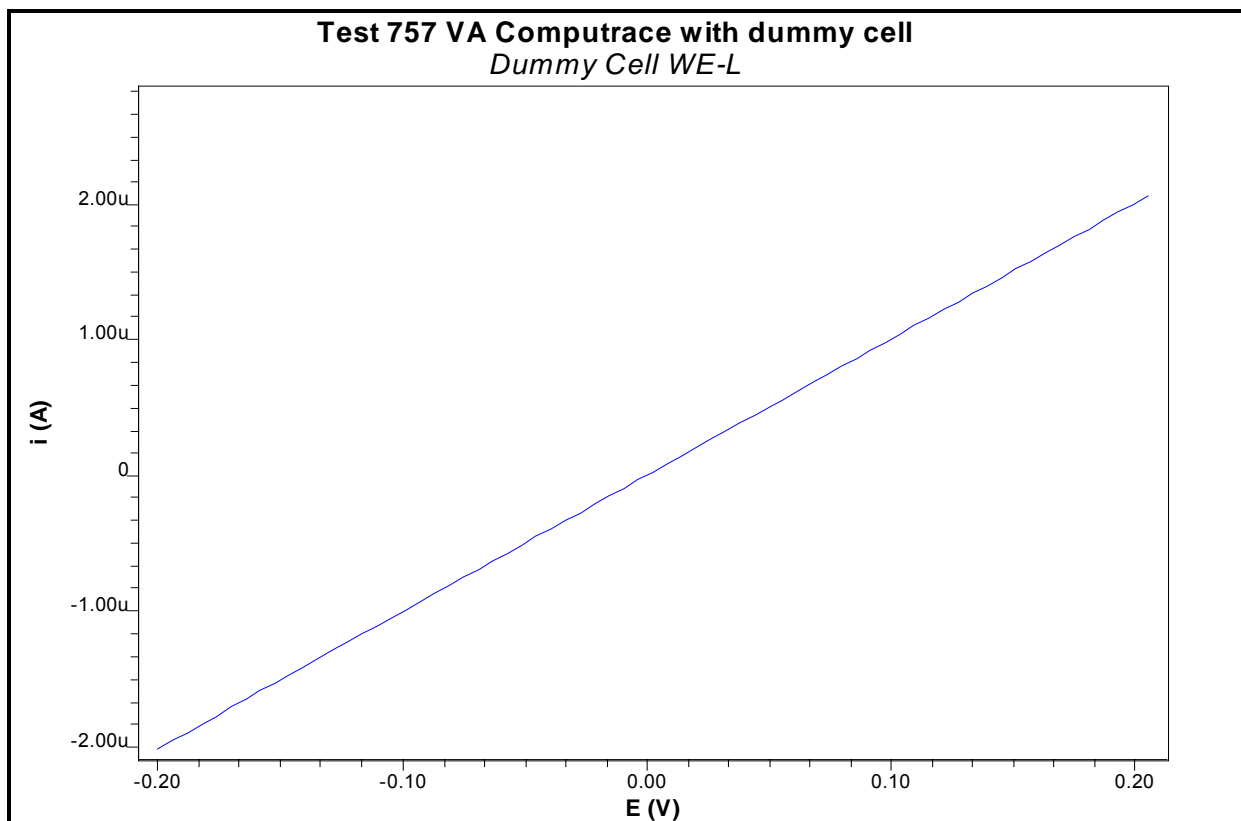
Measuring solution:	<i>20 mL water + 0.5 mL KCl + 100 μL Pb standard</i>
Electrolyte:	<i>c(KCl) = 3 mol/L</i>
Standard Solution:	<i>?(Pb) = 1 g/L</i>

	Measured values	Tolerances	Test passed	
Final Result:	<i>1.034 g/L</i>	<i>0.95 ... 1.05 g/L</i>	yes <input checked="" type="checkbox"/>	no <input type="checkbox"/>
Res. Dev.:	<i>1.199 %</i>	<i>= \pm 0.03 g/L (\pm 3%)</i>	yes <input checked="" type="checkbox"/>	no <input type="checkbox"/>

Validation passed: yes no

Signature: 	Visa: 
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Linearity Test



Peak Test

Determ. : Test757_D.dth
 Date : 1998-03-12
 Modified : ---
 Time: 10:18:06
 User: Cell volume: 10.000 ml

Ident : Dummy Cell WE-D
 Sample volume : 10.000 ml

Method : Test757_D.mth
 Title : Test 757 VA Computrace with dummy cell
 Remark1 : connect to WE-D
 Remark2 : connect AE and RE

Substance : No.1
 Mass conc.: n/a
 MC.dev. : n/a
 Comments

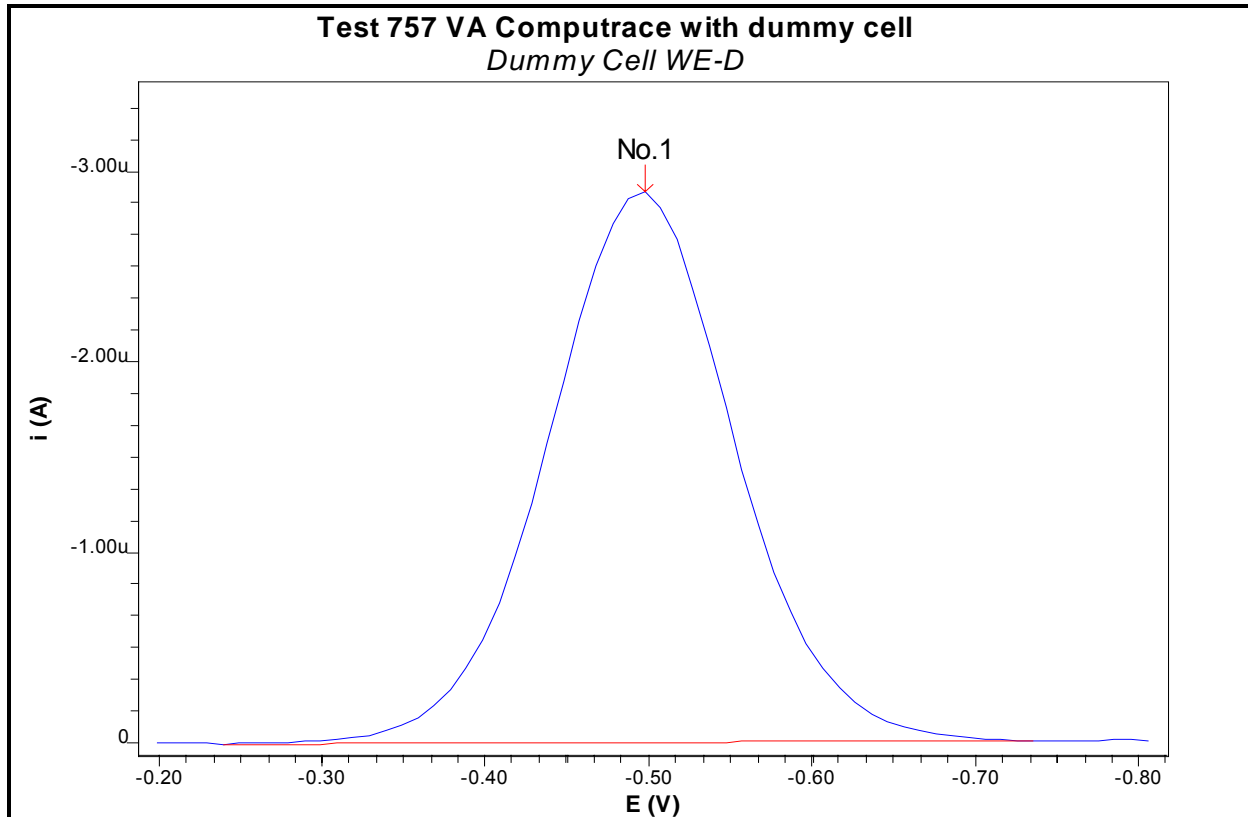
VR	V	uA	i.mean	Std.Dev.	i.delta	Comments
1-1	-0.497	-2.886	-2.886	---	---	---

Substance	Calibr.	Y.reg/offset	Slope	Nonlin.	Mean deviat.
No.1	std.add.	---	---	---	---

Solutions

No.	Content	Vol. (ml)	Predose (ml)
---	---	---	---

Final results	+/-	Res. dev.	%	Comments
No.1 =	n/a	g/l	---	---



Report of Chemical Validation

Determ. : Test Pb in standard solution.dth
 Date : 1998-03-12 Time: 09:53:29
 Modified : --- User: Cell volume: 20.500 ml

Ident : Pb standard Sample volume
 0.100 ml

Method : Test Pb in standard solution.mth
 Title : Determination of Lead in Ion Standard Solution
 Remark1 : 20 ml water + 0.5 ml KCl (3 mol/l) + 100 µl Pb ion standard solution
 Remark2 : c(Pb ion standard solution) = 1 g/l

Substance	: Pb					Comments
Mass conc.	: 5.044 mg/l					
MC.dev.	: 0.060 mg/l	(1.20%)			
Mass	: 103.393 µg					
Add.mass	: 100.000 µg					

	VR	V	uA	i.mean	Std.Dev.	i.delta	Comments
1-1	-0.349	-0.388	-0.389	0.001			
1-2	-0.349	-0.390					
1-3	-0.349	-0.390					
2-1	-0.349	-0.764	-0.765	0.003	-0.375		
2-2	-0.349	-0.768					
2-3	-0.349	-0.762					
3-1	-0.349	-1.118	-1.132	0.012	-0.367		
3-2	-0.349	-1.139					
3-3	-0.349	-1.137					

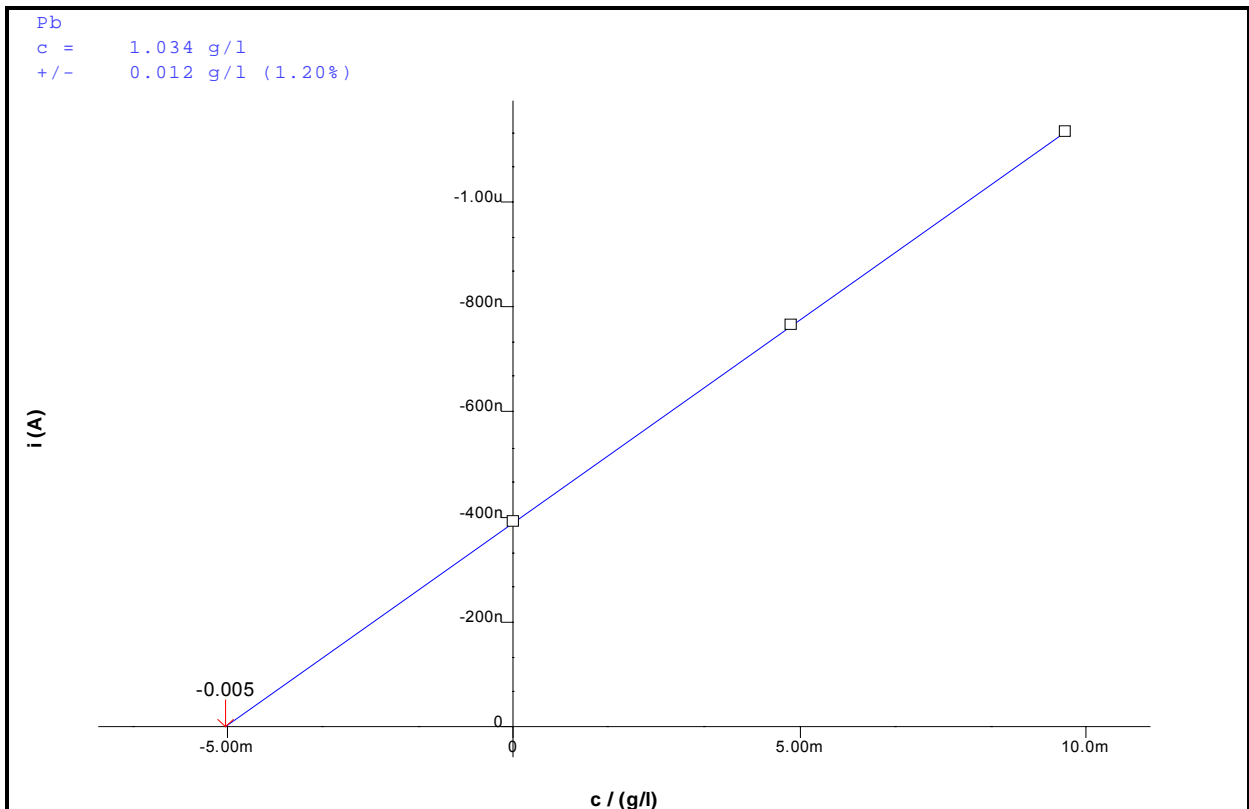
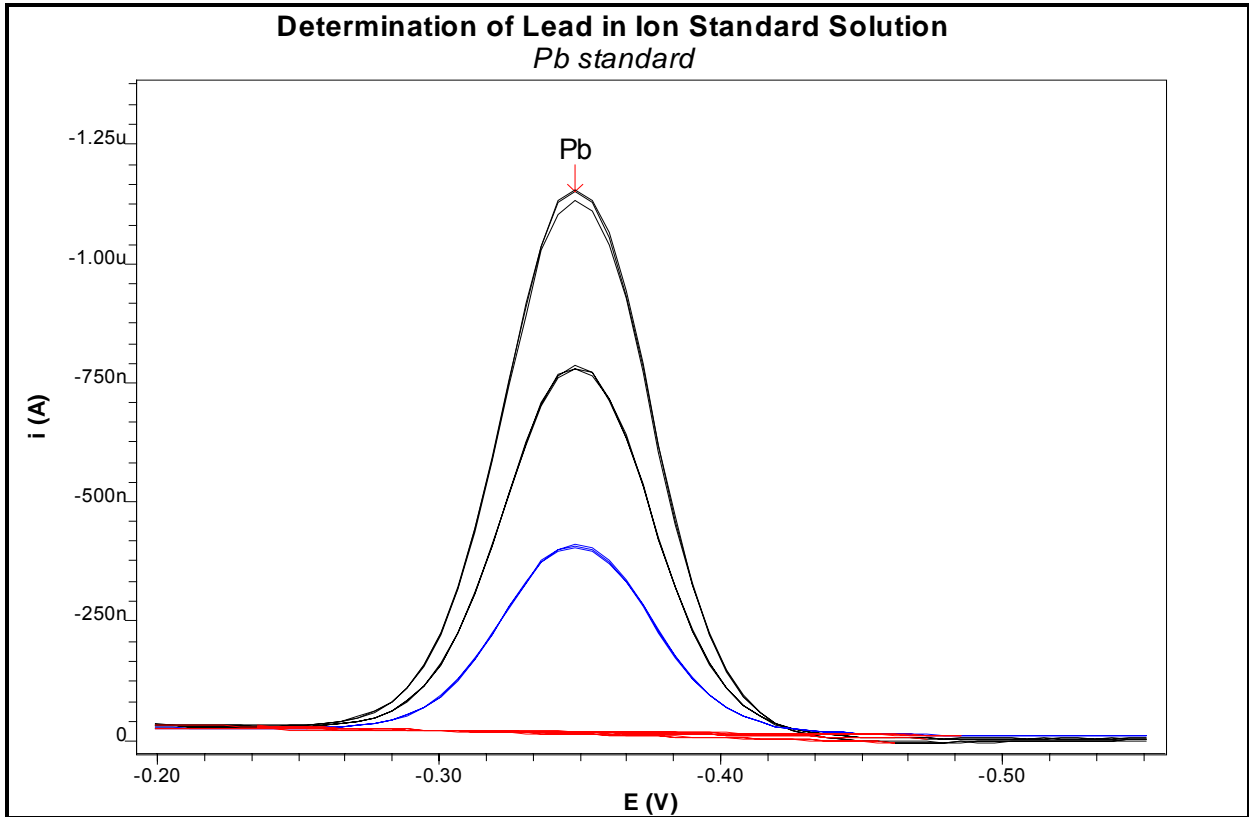
Substance	Calibr.	Y.reg/offset	Slope	Nonlin.	Mean deviat.
Pb	std.add.	-3.894e-007	-7.722e-005		1.429e-009


Solutions

No.	Content	Vol. (ml)	Predose (ml)
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Final results		+/-	Res. dev.	%	Comments
Pb =	1.034 g/l	0.012		1.199	

Voltammogram and Calibration Curve of Chemical Validation



Validation Record		Company :	
VA Instrument		Division :	
Date:		User:	
Time:			
Instrument :		Serial number:	
<u>Electronic Validation (Dummy Cell Test)</u>			
Linearity Test:	Measured values	Tolerances	Test passed
Current at -200 mV		-1.6 μ A ... -2.4 μ A	yes <input type="checkbox"/> no <input type="checkbox"/>
Current at +200 mV		+1.6 μ A ... +2.4 μ A	yes <input type="checkbox"/> no <input type="checkbox"/>
Peak Test:	Measured values	Tolerances	Test passed
Peak voltage		-450 mV ... -550 mV	yes <input type="checkbox"/> no <input type="checkbox"/>
Peak current		746: +250 nA...+1250 nA	
		757: -2 μ A ... -4 μ A	yes <input type="checkbox"/> no <input type="checkbox"/>
<u>Chemical Validation</u>			
Measuring solution:			
Electrolyte:			
Standard Solution:			
	Measured values	Tolerances	Test passed
Final Result:		0.95 ... 1.05 g/L	yes <input type="checkbox"/> no <input type="checkbox"/>
Res. Dev.:		= \pm 0.03 g/L (\pm 3%)	yes <input type="checkbox"/> no <input type="checkbox"/>
Validation passed: yes <input type="checkbox"/> no <input type="checkbox"/>			
Signature: 		Visa: 