DIALAB Produktion und Vertrieb von chemisch-technischen Produkten und Laborinstrumenten Gesellschaft m.b.H.
Hondastrasse, Objekt M55, IZ NOE-Sued, 2351 Wiener Neudorf, Austria,
Phone: +43 (0) 2236 660910-0, Fax: +43 (0) 2236 660910-30, e-mail: office@dialab.at

Liquid Reagents – ready to use

CHLORIDE

Mercuric thiocyanate

Single Reagent

Diagnostic reagent for quantitative in vitro determination of chloride in human serum or plasma on photometric systems

Ref.No.	Kit Size	Configuration
D01222B	**1 x 1 L**	Single Reagent
D01223	**5 x 100 ml**	Single Reagent
D01225	**5 x 50 ml**	Single Reagent
D01227	**5 x 25 ml**	Single Reagent
D01228	**5 x 10 ml**	Single Reagent
D61911	**10 x 50 ml**	Single Reagent
D0415917	**9 x 65 ml**	Single Reagent
DA0813	**5 x 50 ml**	Single Reagent
DT1013	**4 x 50 ml**	Single Reagent
DK0713	**5 x 50 ml**	Single Reagent
DE1813	**5 x 20 ml**	Single Reagent

Additionally offered:

D95108	1 x 3 mL	Chloride Standard	
D98485	5 x 3 mL	Calibrator	Diacal Auto
D98485SV	1 x 3 mL	Calibrator	Diacal Auto
D98481	12 x 5 mL	Control normal	Diacon N
D14481	5 x 5 mL	Control normal	Diacon N
D98481SV	1 x 5 mL	Control normal	Diacon N
D98482	12 x 5 mL	Control abnormal	Diacon P
D14482	5 x 5 mL	Control abnormal	Diacon P
D98481SV	1 x 5 mL	Control abnormal	Diacon P

TEST PARAMETERS

Method:	Colorimetric, endpoint, increasing reaction, Mercuric thiocyanate
Wavelength:	436 nm
Temperature:	20 – 25 °C, 37 °C
Sample:	Serum or plasma,
Linearity:	up to 130 mmol/L
Sensitivity:	The lower limit of detection is 1 mmol/L

SUMMARY [1,2]

Chloride is the most important anion in serum besides bicarbonate. Together with sodium it is an essential osmotically active component in plasma which is involved in maintenance of water distribution and anion-cation-balance. Serum concentrations of chloride behave parallel to sodium levels and reciprocally to bicarbonate. Increased chloride values occur in dehydration, metabolic acidosis related with prolonged diarrhoea and bicarbonate loss, renal insufficiencies and endocrinological disorders as reduced or increased adrenal function. Decreased values are observed in metabolic acidosis with increased production of organic acids, salt-losing nephritis and excessive sweating.

TEST PRINCIPLE

This assay method is based on the following reaction sequences:

$$6\ Cl^- + 3\ Hg(SCN)_2 \rightleftarrows 3\ HgCl_2 + 6\ (SCN)^-$$

$$6\ (SCN)^- + 2\ Fe^{3+} \rightleftarrows 2\ Fe(SCN)_3$$

Chloride ions in the sample react with mercuric thiocyanate releasing equivalent quantities of thiocyanate.
Free thiocyanate ions then react with iron ions forming a red coloured complex whose absorbance at 436 nm is proportional to the chloride concentration in the sample.

REAGENT COMPOSITION

COMPONENTS	CONCENTRATION
Mercury (II) thiocyanate	2 mmol/L
Mercury (II) chloride	0.8 mmol/L
Ferric (III) nitrate	20 mmol/L
Nitric acid	28 mmol/L

REAGENT PREPARATION

The reagent is ready to use.

REAGENT STABILITY AND STORAGE

Conditions:	Protect from light. Close immediately after use Avoid contamination. Do not freeze the reagent.
Storage:	at 15 – 25 °C
Stability:	up to the expiration date indicated on labels

SAMPLE STABILITY AND STORAGE

Stability [3]:	at 20 – 25 °C	7 days
	at 4 - 8 °C	7 days
	at -20 °C	at least one year

Freeze only once!
Discard contaminated specimens.

MATERIALS REQUIRED BUT NOT PROVIDED

General laboratory equipment

STANDARD

(not included in the kit – has to be ordered separately)

Concentration:	100 mmol/L
Storage:	2 – 25 °C
Stability:	up to the expiration date

Close immediately after use! Avoid contamination!

MANUAL TEST PROCEDURE

Pipette into test tubes	Blank	Std./Cal.	Sample
Reagent	1000 µL	1000 µL	1000 µL
Sample	-	-	10 µL
Stdandard/Calibrator	-	10 µL	-
Distilled water	10 µL	-	-
Mix, Incubate for 5 min. at 20 – 25 °C / 37 °C. Read absorbance against reagent blank.			

CALCULATION

$$\text{Chloride [mmol/L]} = \frac{\Delta A\ \text{Sample}}{\Delta A\ \text{Std/Cal}} \times \text{Conc. of Std/Cal [mmol/L]}$$

UNIT CONVERSION

mmol/L = mEq/L
mmol x 3.545 = mg/dL

REFERENCE RANGES [1] * [mmol/L]

Adults:		95 – 105
Children:	1 – 7 days	96 – 111
	7 – 30 days	96 – 110
	1 – 6 months	96 – 110
	6 months – 1 year	96 – 108
	> 1 year	96 – 109

* Each laboratory should check if the reference ranges are transferable to its own patient population and determine own reference ranges if necessary.

PERFORMANCE CHARACTERISTICS

LINEARITY, MEASURING RANGE

The test has been developed to determine chloride concentrations within a measuring range from 1 to 130 mmol/L. Samples with chloride ion concentrations higher than 130 mmol/L should be diluted 1+1 with distilled or deionized water and the results multiplied by 2.

PRECISION (at 25°C)

Intra-assay n = 20	Mean [mmol/L]	SD [mmol/L]	CV [%]
Sample 1	96.5	1.85	1.92
Sample 2	110	3.56	3.22
Sample 3	117	2.70	2.31

Inter-assay n = 20	Mean [mmol/L]	SD [mmol/L]	CV [%]
Sample 1	97.5	2.12	2.18
Sample 2	108	2.26	2.08
Sample 3	117	1.70	1.45

SENSITIVITY/LIMIT OF DETECTION
The lower limit of detection is 1 mmol/L

INTERFERING SUBSTANCES
no interference up to:

Ascorbic acid	30 mg/dL
Bilirubin	20 mg/dL
Hemoglobin	500 mg/dL
Triglycerides	250 mg/dL

For further information on interfering substances refer to Young DS [5].

CALIBRATION

The assay requires the use of a Chloride Standard or a Chloride Calibrator.
We recommend the Dialab Chloride Standard or the multi calibration serum **Diacal Auto**. The assigned standard and calibrator values have been made traceable to the NIST-SRM® 999b reference material.

QUALITY CONTROL

All control sera with Chloride values determined by this method can be used.
We recommend the Dialab serum controls **Diacon N** (control serum with values in the normal range) and **Diacon P** (control serum with values in the abnormal range).
Each laboratory should establish corrective action in case of deviations in control recovery.

AUTOMATION

Special applications for automated analysers can be made on request.

WASTE MANAGEMENT

Please refer to local legal requirements.

WARNINGS AND PRECAUTIONS

1. Reagent: Warning.
   H290: May be corrosive to metals.
   H411: Toxic to aquatic life with ling lasting effects.
   P 234: Keep only in original container.
   P 391: Collect spillage.
   P 501: Dispose of contents/container to hazardous or special waste collection point.
2. In very rare cases, samples of patients with gammopathy might give falsified results [6].
3. Please refer to the safety data sheet and take the necessary precautions for the use of laboratory reagents.
4. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.
5. For professional use only!

REFERENCES

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3. Guder WG, Zawta B et al. The Quality of Diagnostic Samples. 1st ed. Darmstadt: GIT Verlag; 2001; p.22-3.
4. Schoenfeld RG, Lewellen CJ. A colorimetric method for determination of serum chloride. Clin Chem 1964; 10: 533-9.
5. Young DS. Effects of Drugs on Clinical Laboratory Tests. 5th ed. Volume 1 and 2. Washington, DC: The American Association for Clinical Chemistry Press 2000.
6. Bakker AJ, Mücke M Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. ClinChemLabMed 2007; 45(9): 1240-1243.