CHLORIDE

PRINCIPLE OF THE METHOD
The quantitative displacement of thiocyanate by chloride from mercuric thiocyanate and subsequent formation of a red mercuric thiocyanate complex is measured colorimetrically:

\[ 2 \text{Cl}^- + \text{Hg(SCN)}^2- \rightarrow \text{HgCl}_2 + 2 \text{SCN}^- \]

\[ \text{SCN}^- + \text{Fe}^{3+} \rightarrow \text{Fe(SCN)}^2- \]

The intensity of the color formed is proportional to the chloride ion concentration in the sample.\(^2,3,4\)

CLINICAL SIGNIFICANCE
It is important clinically the determination of chloride due regulation of osmotic pressure of extra cellular fluid and its significant role in acid-base balance. Increases in chloride ion concentration may be found in severe dehydration, excessive intake of chloride, severe renal tubular damage and in patients with cystic fibrosis. Decrease in chloride ion concentration may be found in metabolic acidosis, loss from prolonged vomiting and chronic pyelonephritis.\(^5,6\)

Clinical diagnosis should not be made on a single test result; it should integrate observations with other laboratory data.

REAGENTS
- Mercuric thiocyanate: 4 mmol/L
- Ferric nitrate: 40 mmol/L
- Mercuric nitrate: 2 mmol/L
- Nitric acid: 45 mmol/L
- Chloride aqueous primary standard 125 mmol/L

PRECAUTIONS
- Mercury(II) Thiocyanate: Harmful (Xn): R20/21/22: Harmful by inhalation, in contact with skin and if swallowed.
- Cholesterol: Dangers of cumulative effects.
- Mercuric thiocyanate: Harmful (Xn): R20/21/22: Harmful by inhalation, in contact with skin and if swallowed. R33: Danger of cumulative effects.

STORAGE AND STABILITY
- All the components of the kit are stable until the expiration date on the label when stored tightly closed at 2-8ºC, protected from light and label when stored tightly closed at 2-8ºC, protected from light and contaminants prevented during their use.
- Calcium lactate solution (50 mmol/L NaCl) is stable up to 1 month when stored tightly closed at 2-8ºC, protected from light and contaminants prevented during their use.

ADDITIONAL EQUIPMENT
- Spectrophotometer or colorimeter measuring at 480 nm.

SPECIAL ATTENTION
- Matched cuvettes 1.0 cm light path.
- General laboratory equipment.

SAMPLES
- Serum, plasma, CSF, sweat and other body fluids: Free of hemolysis and separated from cells as rapidly as possible. Anticoagulants such as oxalate or EDTA are not acceptable if they will interfere with results.
- Urine: Collect 24-hour urine specimen in chloride-free containers. Dilute a sample 1/2 in distilled water. Mix. Multiply results by 2 (dilution factor). Stability of the sample: Chloride is stable 1 week at room temperature (15-25ºC), in refrigerator (2-8ºC) or frozen (-20ºC) temperatures.

PROCEDURE
1. Assay conditions:
   - Wavelength: 440-500 nm
   - Cuvette: 1 cm, light path
   - Temperature: 37ºC / 15-25ºC
2. Adjust the instrument to zero with distilled water.
3. Pipette into a cuvette:
   - R (mL)
   - Standard (mmol/L) µL
   - Sample (µL)

4. Mix and incubate for 5 min at 37ºC / 15-25ºC.
5. Read the absorbance (A) of the samples and Standard, against the Blank. The blank color is stable for at least 30 minutes.

CALCULATIONS
- Conversion factor: mmol/L = mEq/L

QUALITY CONTROL
Control sera are recommended to monitor the performance of assay procedures.
- If control values are found outside the defined range, check the instrument, reagents and calibrator for problems.
- Each laboratory should establish its own Quality Control scheme and corrective actions if controls do not meet the acceptable tolerances.

REFERENCE VALUES
- Serum or plasma: 95-115 mmol/L
- CSF: 95-110 mmol/L
- Urine: 110 - 250 mmol/L
- Sweat: Up to 60 mmol/L

BIBLIOGRAPHY