Manual
Procedure

MEDICHEM MIDDLE EAST
Clinical Chemistry Reagents
Liquid Stable Reagents

Cat. No. 13261 R1 1 x 20 ml
For 40 tests R2 1 x Powder for 4 ml

Cholinesterase (CHE)

Kinetic colorimetric method,
based on DGKC recommendations

Powder Reagent

Test principle

Butyrylthiocholine + H_2O $\xrightarrow{Cholinesterase}$ Thiocholine + Butyrate

2 Thiocholine + 2 OH^- + 2 $[Fe(CN)_6]^{-3}$ ⟶ Dithiobis (Choline) + H_2O + 2$[Fe(CN)_6]^{-4}$

Cholinesterase hydrolyzes butyrylthiocholine to give thiocholine and butyrate. The reaction between thiocholine and ferricyanide ions in an alkaline medium gives dithiobis (choline) a yellow compound measured at 405 nm. The color intensity is proportional to the concentration of cholinesterase present.

Concentrations in the test

Reagent R1		
Phosphate buffer, PH = 7.6	92	mmol/L
Potassium hexacyanoferrate	2.7	mmol/L
Reagent R2		
Butyrylthiocholin	126	mmol/L

Stability of reagent

Reagent R1: *Liquid, ready to use.*
Reagent R2: *powder.*
Dissolve content of bottle R2 with 4.0 ml distilled water.
Stable for 6 months at +2 ➔ +8 °C.

All reagents are stable up to expiry date given on the label when stored at 2 - 8 °C.

Note: Don't use if reagent R1 is turbid.

Specimen collection and handling

Serum, heparinized or EDTA plasma.
Stability: 7 days at 2 – 8 °C.
Specimens submitted to evaluate possible pesticide toxicity should be separated and frozen promptly until analyzed. Otherwise, in vitro destruction of the enzyme inhibitor may occur leading to falsely normal values.

Calibrator

MediCal U Cat. No. 15011

Quality control

Meditrol N Cat. No. 15171
Meditrol P Cat. No. 15181

Procedure

Wavelength	Hg 405 (400 - 440 nm)
Spectrophotometer	405 nm
Cuvette	1 cm light path
Temperature	37°C
Measurement	against air or distilled water
Reaction	kinetic – decrease

Assay

Sample	10 µl
Reagent R1	500 µl
Incubate for 5 min. at 37 °C.	
Reagent R2	100 µl
Mix, incubate for 90 Sec. at 37°C. Read change in the absorbance per 1 min. for 3 min. Determine the mean absorbance change per 1 min. (ΔA/min).	

Calculation

Cholinesterase activity = ΔA/min. X Factor at 405 nm

Factors

Units	U/L	KU/L
Factors at 37°C	65800	65.8

Note: It is recommended that each laboratory (as per instrument performance) could make its own factor (F) by the use of a calibrator according to the following formula:

$$F = \frac{\text{Conc.}_{\text{Clibrator}}}{\Delta/\text{min}_{\text{Calibrator}}}$$

Linearity

Up to 32900 U/L (548.3 µkat/L, 32.9 kU/L).
If the result exceeds 32.9 kU/L repeat the test using diluted sample (1+3) with sodium chloride solution (0.9 %) and multiply the result by 4.
Note: 1 kU/L = 1000 U/L = 16.67 µkat/L

Interferances

1. Fluride interferes with the test.
2. Pregnancy, estrogens therapy, and oral contraceptives decrease test values.
3. Drugs that may cause decreased values include atropine, caffeine, codeine, morphine sulfate, neostigmine, phenothiazines, theophylline and vit K.
4. See Young *et. al*. for a list of other interfering substances.

Reference range

(m,w > 40 yr.) Children / Adults		5300 - 12900	U/L
w (16 – 39 yr.) Not pregnant,Not taking oral Contraceptive		4300 - 11200	U/L
w (18 – 40 yr.) Pregnant or taking oral Contraceptives		3600 - 9100	U/L
Adults	women	3900 - 10800	U/L
	men	4600 - 11500	U/L

References

1. Tietz, N. W. Textbook of clinical chemistry, Philadelphia, W.B. Saunders Company, pp. 725 - 734 (1986).
2. Laurence Lepuge. Clin. Chem. 31/4.546 - 550 (1985).
3. Hiroaki Okabe. Clin. Chem. Acta, 80 (1977) 87 - 94.
4. Young, DS., Effects of Drugs on Clinical Laboratory Tests, fifth edition 2000, AACC Press, Washington, D.