



CK - MB (NAC - act.)

E.C.2.7.3.2.

UV Method

Cat.No. 101-0375

Size 19 x 2.5 ml

PRINCIPLE:

CK-MB is composed of the two moieties CK-M and CK-B. A specific antibody inhibits the CK-M moiety without affecting the CK-B moiety.

The CK-B fraction accounts for one half the activity of CK-MB, it is determined by the NAC-activated method.

SAMPLE:

Serum, plasma (heparin, EDTA).

Activity decrease in serum after 24 hours at +2 °C to +8 °C or 1 hour at room temperature 10%. Bring to assay temperature before use.

REAGENTS:

	Concentration in the test
1. Buffer/Glucose (1 x 70 ml)	
Imidazole Buffer, pH 6.7	100 mmol/L
Glucose	20 mmol/L
Mg-Acetate	10 mmol/L
EDTA	2 mmol/L
2. Enzymes/Coenzymes/Substrate/Antibody (19x2.5 ml)	
Ab. Anti-CK-M	2000 U/L
ADP	2 mmol/L
AMP	5 mmol/L
Diadenosine Pentaphosphate	10 µmol/L
NADP	2 mmol/L
HK	2500 U/L
G ₆ P-DH	1500 U/L
N-Acetylcysteine	20 mmol/L
Creatine Phosphate	30 mmol/L
3. Control	
Lyophilized vial for 3 ml	

REAGENT PREPARATION:

Dissolve one tablet in one vial of Buffer R1.

Stable for 8 days at +2 °C to +8 °C or 24 hours at room temperature. Bring to assay temperature before use.

Standard: Reconstitute the contents of one vial with 3 ml of distilled water. Mix gently until complete solution.

PROCEDURE:

Before carrying out the activity of CK-MB in serum, it is necessary to measure total CK activity using the CK-NAC method.

Wavelength: 340 nm, Hg 334 nm, Hg 365 nm
 Cuvette: 1 cm light path
 Temperature: 25 °C, 30 °C, 37 °C
 Zero: air

	Macro	Semi-micro
Working reagent solution	2.5 ml	1.0 ml
Sample	100 µl	40 µl
Mix, and let stand at the appropriate temperature for 10 minutes. Then read absorbance A ₁ . Read absorbance A ₂ exactly 5 minutes later. Calculate $\Delta A = A_2 - A_1$		

CALCULATION:

Calculate the activities **CK-MB** in the sample at: 25/30/37 °C:

$$\begin{aligned} 340 \text{ nm } \Delta A \times 1651 &= \text{U/L} \\ \text{Hg } 334 \text{ nm } \Delta A \times 1683 &= \text{U/L} \\ \text{Hg } 365 \text{ nm } \Delta A \times 2972 &= \text{U/L} \end{aligned}$$

Calculate **CK-B** fraction: at 25/30/37 °C

$$\begin{aligned} 340 \text{ nm } \Delta A \times 825 &= \text{U/L} \\ \text{Hg } 334 \text{ nm } \Delta A \times 841 &= \text{U/L} \\ \text{Hg } 365 \text{ nm } \Delta A \times 1486 &= \text{U/L} \end{aligned}$$

Conversion: 1 µkat = 60 Units
1 U = 16.67 x 10⁻³ µkat

EXPECTED VALUES:

In serum taken from healthy people the CK-MB activities are below :

$$\begin{aligned} 25 \text{ }^\circ\text{C} &< 10 \text{ U/L (167 nkat/L)} \\ 30 \text{ }^\circ\text{C} &< 16 \text{ U/L (267 nkat/L)} \\ 37 \text{ }^\circ\text{C} &< 25 \text{ U/L (417 nkat/L)} \end{aligned}$$

The likelihood of myocardial damage is high when the following triad of factors are present:

	25 °C	30 °C	37 °C	Units
CK (men)	> 80 > 1334	> 130 > 2167	> 195 > 3251	U/L nkat/L
CK-MB (women)	> 70 > 1167	> 110 > 1834	> 170 > 2834	U/L nkat/L
CK-MB	> 10 > 167	> 16 > 267	> 25 > 417	U/L nkat/L

A CK-MB activity between 6 and 25% of the total CK activity.

If MI is suspected but the values obtained are below the specified limits, a fresh infarct may have occurred. In this case, tests should be repeated after 4 hours.

Temperature conversion factors:

Assay temperature	Desired temperature		
	25 °C	30 °C	37 °C
25 °C	1.00	1.53	2.38
30 °C	0.65	1.00	1.56
37 °C	0.42	0.64	1.00

LINEARITY:

Antibody inhibits up to 2000 U/L (33340 nkat/L) of CK-MM.

NOTE:

- Solution 1 (buffer/glucose) contains sodium azide as preservative. Do not swallow. Avoid contact with the skin or mucous membranes.
- Hemolytic sera must not be used for the determination of CK and CK-MB activities.

REFERENCES :

- Rec. GSCC (DGKC); J.Clin.Chem. Clin.Biochem. 15, 255 (1977).
- Szasz, G.Gruber, W. Clin. Chem., 22, 650 (1976)