Lifechem™ CK-MB

(Modf.IFCC Method)

CLINICAL SIGNIFICANCE :

Creatine kinases are dimeric molecules composed of M and B subunits and exist as the isoenzymes MM,MB and BB.The sub uints M and B are immunologically distinct .CK-MM and CK-MB are distributed primarily in the skeletal muscle and heart muscle respectively while CK-BB is present mainly in the brain and in tissues composed of smooth muscle.After acute myocardial infarction , CK-MB activity increases significantly and this elevation is highly specific for the laboratory diagnosis of myocardial infarction**.**

TEST PRINCIPLE :

The sample is incubated in the CK-MB reagent which includes the anti-CK-M antibody. The activity ot the noninhibited CK-B is then determined by series of reactions.

Creatine Kinase catalyses the reactions between creatine phosphate and ADP with formation of creatine and ATP. The ATP formed in presence of Glucose and Hexokinase (HK) gives ADP and glucose-6-phosphate. Glucose-6-phosphate, in presence of Glucose-6-phosphate dehydrogenase (G_6PDH) reacts with NADP forming 6-phosphogluconate and NADPH. The increase in absorbance due to the reduction of NADP to NADPH measured at 340 nm is proportional to the activity of CK-MB in the sample. The presence of N-Acetylcysteine (NAC) in the reaction mixture allows the optimal activation of the enzyme.

$$\text{Creatine Phosphate} + \text{ADP} \xrightarrow{\text{Creatine Kinase (CK)}} \text{Creatine} + \text{ATP}$$

$$\text{ATP} + \text{Glucose} \xrightarrow{\text{Hexokinase (HK)}} \text{ADP} + \text{Glucose-6-phosphate}$$

$$\text{Glucose-6-phosphate} + \text{NADP}^+ \xrightarrow{\text{G6PDH}} \text{6- phosphogluconate} + \text{NADPH} + \text{H}^+$$

NORMAL RANGE :

Serum: Up to 24 IU/L
It is recommended that laboratories should establish their own normal ranges.

KIT CONTENTS :

	CODE No. **KMB1** **20ml**
1. REAGENT – A	2ml
2. REAGENT – B	18ml

SPECIMEN :

Un-hemolysed serum

REAGENT PREPARATION :

Mix 1 volume of Reagent **A** with 9 volumes of Reagent **B** according to the requirement.
0.1 ml (100µl) of Reagent **A** and 0.9 ml (900µl) of Reagent **B** are mixed for preparing 1 ml of working reagent.

WORKING REAGENT IS STABLE FOR 15 DAYS AT 2-8°C.
Reagent solution should be protected from light.

PROCEDURE :

Allow the working reagent to reach room temperature before use.
Perform the assay as given below :

Working Reagent	1.0 ml
Serum Sample	0.040 ml (40µl)

Mix well and aspirate. Read the Initial absorbance after 10 minutes. Repeat the absorbance readings exactly after 1, 2 and 3 minutes. Calculate the mean of Δ A / min.

CALCULATIONS :

CK- MB conc. in IU / L = Δ A / min x 6666

QUALITY CONTROL :

It is recommended to include Assayed Quality Control Serum (Level 1 & II) with each assay batch to verify the performance of the procedure. Failure to obtain the proper range of values in the assay of control sera may indicate either reagent deterioration, instrument malfunction or procedural errors.

SYSTEM PARAMETERS :

Reaction Type	:	Kinetic
Wavelength	:	340 nm
Flow cell temperature	:	37°C
Blank	:	Distilled water
Factor	:	6666
Reaction slope	:	Increasing
Delay time	:	600 seconds
Measuring time	:	180 seconds
Reagent volume	:	1000 µl
Sample volume	:	40 µl
Units	:	IU / L
Low Normal	:	0
High Normal	:	24
Linearity	:	1000

NOTE :

1. Do not leave reagents at room temperature when not in use.
2. Use always new tips for working reagent preparation in order to avoid contamination of reagents.
3. No interference of Bilirubin upto 20 mg/dl and Hemoglobin upto 10 gm/L

LINEARITY :

Linearity of the kit is up to 1000 IU/L

BIBLIOGRAPHY :

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