CK-NAC

IFCC

Intended use:

In vitro test for the quantitative determination of Creatine Kinase (CK) in human serum and plasma.

Summary:

Creatine kinase (CK) is a dimeric enzyme occurring in four different forms: mitochondrial isoenzyme and the cytosolic isoenzymes CK-MM (muscle type), CKBB (brain type) and CK-MB (myocardial type). The determination of CK and CK-isoenzyme activities is utilized in the diagnosis and monitoring of myocardial infarction and myopathies such as the progressive Duchenne muscular dystrophy. Following injury to the myocardium, such as occurs with acute myocardial infarction. CK is released from the damaged myocardial cells. In early cases, a rise in the CK activity can be found just 4 hours after an infarction, the CK-activities reaches a maximum after 12-24 hours and then falls back to the normal range after 3-4 days. Myocardial damage is very likely when the total CK activity is above 190 U/l, the CK-MB activity is above 24 U/l (+37°C) and the CK-MB activity fraction exceeds 6% of the total. The assay method using creatine phosphate and ADP was first described by Oliver, modified by Rosalki and further improved for optimal test conditions by Szasz. CK is rapidly inactivated by oxidation of the sulfhydryl groups in the active center. The enzyme can be reactivated by the addition of acetylcysteine (NAc). Interference by adenylate kinase is prevented by the addition of diadenosine pentaphosphate and AMP. Standardized methods for the determination for CK using the "reverse reaction" and activation by NAc were recommended by the German Society for Clinical Chemistry (DGKC) and the International Federation of Clinical Chemistry (IFCC) in 1977 and 1990 respectively. This assay meets the recommendations of the IFCC and DGKC (Standard method 94).

Test principle:

UV Test
- Sample and addition of R1
- Addition of R2 and start of reaction:

$$\text{Creatine phosphate + ADP} \xrightarrow{\text{CK}} \text{Creatine + ATP}$$

$$\text{ATP + glucose} \xrightarrow{\text{HK}} \text{Glucose-6-phosphate + ADP}$$

$$\text{Glucose-6-P + NADP} \xrightarrow{\text{G6PDH}} \text{Gluconate-6-P + NADPH+H}_{+}$$

Equimolar quantities of NADPH and creatine are formed at the same rate. The photometrically measured rate of formation of NADPH is proportional to the CK activity.

Reagent Concentration:

R1:	
Imidazole Buffer, pH 6.7	110 mmol/l
Glucose	21 mmol/l
Mg-Acetate	11 mmol/l
EDTA	2,1 mmol/l
NADP	2,4 mmol/l
N-Acetylcysteine	24 mmol/l
Hexokinase (HK) ³	2,5 U/l
Preservatives/Stabilizers	<1 %
R2:	
Tris Buffer, pH 9.1	50 mmol/l
ADP	2,4 mmol/l
AMP	6 mmol/l
Diadenosinpentaphosphate	12 µmol/l
G–6–P–DH ³	1,7 U/l
Creatinphosphate	186 mmol/l

Preparation and stability:

Unopened kid components: Up to the expiry date at +2°C to +8°C.

Substrate start:

R1: Ready for use
R2: Ready for use

Onboard stability: R1: 28 days
R2: 28 days

Specimen:

Collect serum using standard sampling tubes. Heparinized- or EDTA-plasma
Stability: 7 days at + 4°C to + 8°C
2 days at +20°C to +25°C

Limitation interference:

Criterion: Recovery within ± 10% of initial values.
Icterus: No significant interference up to an index I of 46 (approximate bilirubin concentration: 46 mg/dl).
Hemolysis: No significant interference up to an index H of 1100 (approximate haemoglobin concentration: 1100 mg/dl).
Lipemia (Intralipid): No significant interference up to an index L of 625 (approximate triglycerides concentration: 1250 mg/dl). There is poor correlation between turbidity and triglycerides concentration.

Testing procedure:

Applications for automated systems are available on request.

Materials provided
- Working solutions as described above

Additional materials required
- Calibrators and controls as indicated below
- 0.9% NaCl

Manual Testing Procedure: for sunstrate start	
Wavelength:	340 nm
Reaction temperature:	+37°C
Cuvette:	1 cm light path
Zero adjustment	air or distilled water
Cuvette	Normal
R1	800 µl
sample	40µl
Mix and incubate for 30 sec. at 37°C. Then add:	
R2	200 µl
Mix and incubate for 3 minutes. Measure the absorbance increase per minute for another 3 minutes.	

Measuring / reportable range:

5 – 1300 U/l

Determine samples with higher activities via the rerun function. On instruments without rerun function, manually dilute these samples with 0.9% NaCl or distilled/deionized water (e.g. 1 + 10). Multiply the result by the appropriate dilution factor (e.g. 11).

Expected values:

On the basis on the optimized the IFCC method.
Myocardial infarction: There is high probability of myocardial damage when the following three conditions are fulfilled:

	25°C	30°C	37°C
CK Men	10-65 U/l	15-105 U/l	38-174 U/l
CK Women	7-55 U/l	10-80 U/l	26-140 U/l

* Calculated values

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 hours. The following factors were used for converting the reference values from 25°C: 1.53 (+30°C) and 2.38 (+37°C).
CK varies with physical activity level and race in healthy individuals.
Each laboratory should investigate the transferability of the expected values to its own patient population and if necessary determine its own reference range. For diagnostic purposes the CK results should always be assayed in conjunction withthe patient's medical history, clinical examinations and other findings.

Analytical sensitivity (lower detection limit)

Normal Cuvette: 5 U/l
The lower detection limit represents the lowest measurable CK concentration that can be distinguished from zero.

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Imprecision:
Reproducibility was determined using controls. The following results were obtained

	Within run		
Sample	Mean (U/l)	SD (U/l)	CV (%)
Control serum 1	165.08	1,77	1,07
Control serum 2	323.40	5.00	1,55
Control serum 3	481.93	4,80	1,00

	Betwen day		
Sample	Mean (U/l)	SD (U/l)	CV (%)
Control serum 1	167,53	3,25	1,94
Control serum 2	320,63	10,34	1,55
Control serum 3	493,93	8,23	1,00

Method comparison:
A comparison of the BIOANALYTIC CK-NAc (y) with a commercial obtainable assay (x) gave the following result:
y = 0.9724 x – 0.6476; r = 0.9997

Quality Control:
Control Serum:

BIOCON N	5 x 5 ml	#B10814
BIOCON P	5 x 5 ml	#B10817
BIOANALYTIC CK, CK-MB CONTROL	3 x 1 ml	#B10821

The control intervals and limits must be adapted to the individual laboratory and country-specific requirements. Values obtained should fall within established limits. Each laboratory should establish corrective measures to be taken if values fall outside the limits.

Calibration:
S1: 0.9% NaCl
S2: BIOCAL H 5 x 3 ml #B11895

Calibration frequency:

It is suggested to use Calibrator products produced by Bioanaliytic. It is suggested to use supplementary calibrator (pure water or 0.9% NaCl) to conduct 2-point calibration. The calibration curve is formed automatically. When lot number is changed or QC is invalid, calibration shall be conducted again. Recalibrate the assay every 30 days under ideal conditions,
or when the following occur:
Change in reagent lot or significant shift in control values;
Major preventative maintenance was performed on the analyser or a critical part was replaced(Halogen Lamp)

Order information (Cat No.) :

CC370	AB371	B21097	B27095	B30096	B33097	B37096
CC371	CR370	B22095	B27096	B30097	B34095	B80095
OL370	BCKN250	B24095	B27097	B31095	B34096	B80096
OL371	BCKN125	B24096	B28095	B32095	B35095	B80097
KL370	BCKN50	B25095	B28096	B32096	B36095	B80098
KL371	B21095	B25096	B28097	B33095	B36096	
AB370	B21096	B25097	B30095	B33096	B37095	

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Manufacturer

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SYMBOLS

IVD	for in vitro diagnostic use only
LOT	lot of manufacturing
REF	code number
	storage at temperature interval
	expiration date (year/month)
	warning, read enclosed documents
	Read the directions

ISO 9001:2015
ISO 13485:2016