NEO CHEM
CKNAC
(Mod.IFCC method)

KIT NAME	KIT SIZE
NEO CHEM - CK NAC	2 X 10 ml

NEOGENIX
DIAGNOSTICS PVT LTD

INTRODUCTION
Creatine kinase (CK) catalyzes the transfer of phosphate group between creatine phosphate and adenosine diphosphate (ADP). The product of this reaction is adenosine triphosphate (ATP) – molecular source of energy. CK is a dimer, composed of two different subunits called M and B. Three different isoenzymes formed from these subunits are found in brain and smooth muscle (BB), skeletal muscle (MM) and cardiac muscle (MM and MB). Increased level of CK is usually the result of muscle injury, myocardial or pulmonary Infarction.

METHOD PRINCIPLE
Optimized kinetic method according to International Federation of Clinical Chemistry (IFCC).

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

ATP + D-glucose $\xrightarrow{HK}$ ADP + D-glucose-6-P

D-glucose-6-P + NADP $\xrightarrow{G6P\text{-}DH}$ D-gluconate-6-P + NADPH + H+

The rate of absorbance changes at l=340 nm is directly proportional to creatine kinase activity.

KIT CONTENTS

Reagent Name	Pack size
R1 CKNAC reagent	2 x 8 ml
R2 CKNAC reagent	2 x 2 ml

The reagents when stored at 2-8°C are stable up to expiry date printed on the package. The reagents are stable for 3 weeks on board the analyser at 2-10°C. Protect from light and avoid contamination.

WORKING REAGENT PREPARATION AND STABILITY
Assay can be performed with use of separate R1-CKNAC and R2-CKNAC reagents or with use of working reagent. For working reagent preparation mix gently 4 parts of R1-CKNAC with 1 part of R2-CKNAC Avoid Foaming.

Stability of working reagent : 3 weeks at 2-8°C
5 days at 15-25°C

Protect from light and avoid contamination

CONCENTRATION IN TESTS

Imidazole buffer, pH 6.7	100 mmol/l
D-glucose	20 mmol/l
Magnesium acetate	10 mmol/l
EDTA	2 mmol/l
ADP	2 mmol/l
AMP	5 mmol/l
Diadenosinepentaphosphate	10 µmol/l
N-acetylcystein	20 mmol/l
Creatine phosphate	30 mmol/l
NADP	2 mmol/l
Hexokinase (HK)	2.5 KU/l
Glucose-6-phosphate-dehydrogenase	≥ 1.5 KU/l (G6P-DH)

Warnings and Notes
- Product for in vitro diagnostic use only.
- The reagents contain 0.09% sodium azide as a preservative. avoid contact with skin and mucous membranes.
- It is advisable to use instruments with resolving power of Absorbance 0.0001 A.

ADDITIONAL EQUIPMENT
- Automatic analyzer or photometer able to read at 340 nm with resolving power of absorbance 0.0001
- Thermostat at 37°C
- General laboratory equipment

SPECIMEN
Serum, heparinized or EDTA plasma free from hemolysis.
As an anticoagulant for plasma preparation use heparin or EDTA lithium, sodium or ammonium salt CK activity is unstable and is rapidly lost during storage. Probes should be stored tightly closed and protected from light. Specimens can be stored up to 4-8 hours at 15-25°C or 1-2 days at 2-8°C or 1 month at -20°C, but it is recommended to perform the assay with freshly collected samples.

PROCEDURE
These reagents may be used both for manual assay and in several automatic analysers. Programme Sheets are available on request.

Wavelength	340 nm
Temperature	37°C
Cuvette	1 cm

Pipette into the cuvettes:

Reagent	Test (T)
R1 CK NAC reagent	800 µl
R2 CK NAC reagent	200 µl
Bring to assay temperature, then add	
Sample	50 µl

Mix and incubate at adequate temperature. After about 60secs. read the absorbance against air or water. Repeat the reading after exactly 60, 120 and 180 seconds interval. Calculate the mean absorbance change per minute (A/min.).

CALCULATION
CK activity [U/L] = DA/min.(T) x 4127 (factor)

REFERENCE VALUES

Female	25 to 175 U/L
Male	25 to 200 U/L

It is recommended for each laboratory to establish its own reference ranges for local population.

QUALITY CONTROL
To ensure adequate quality control, each run should include assayed normal and abnormal controls. If commercial controls are not available it is recommended that known value samples be aliquoted, frozen and used as controls

PERFORMANCE CHARACTERISTICS

Sensitivity / Limit of Quantitation: 6.5 U/LLinearity : up to 2000 U/L
Specificity / Interferences:
Haemoglobin up to 3.75 g/dl, ascorbate up to 62 mg/l, bilirubin up to 20 mg/dl and triglycerides up to 500 mg/dl do not interfere with the test.

WASTE MANAGEMENT

Please refer to local legal requirements.

LITERATURE

1. DGKC: J. Clin. Chem. Clin. Biochem.: 15, 249-254 (1977).
2. The Committee on Enzymes of The Scandinavian Society for Clinical Chemistry and Clinical Phys.: Scand. J. Clin. Lab. Invest. 36, 1-5 (1979).
3. Lott J.A., Stang J.M.: Clin. Chem. 26/9, 1241-1250 (1980).
4. Commission Enzymologie, Comité de Standardisation, Société Francaise de Biologie Clinique: Ann. Biol. Clin. 40, 138-149 (1981).
5. Tietz N.W., ed. Clinical Guide to Laboratory Tests, 3rd ed. Philadelphia, PA: WB Saunders, 806-6 (1995).
6. Burtis C.A., Ashwood E.R., ed. Tietz Textbook of Clinical Chemistry, 3rd ed. Philadelphia, PA: Moss D. W., Henderson A.R., 652 (1999).

SYSTEM PARAMETERS

Method	Kinetic
Wavelength	340 nm
Zero Setting	Distilled Water
Temperature Setting	37° C
Incubation Temperature	37° C
Incubation Time	----
Delay Time	60 secs
Read Time	180 secs
No. of Reading	3
Interval Time	60 secs
Sample Volume	0.05 ml (50 ul)
Reagent Volume	1.0 ml (1000 ul)
Standard Concentration	----
Units	U/L
Factor	4127
Reaction Slope	Increasing
Linearity	2000 U/L