Cat. No 4-520

DIAGNOSTIC KIT FOR DETERMINATION OF CREATINE KINASE ACTIVITY

HC – CK

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 dimmer, 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 $\xleftrightarrow{CK}$ creatine + ATP

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

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

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

REAGENTS
Package

1-Reagent	6 x 87.5 ml
2-Reagent	6 x 18.5 ml

The reagents when stored at 2-8°C are stable up to expiry date printed on the package. The reagents are stable for 12 weeks on board the analyser at 2-10°C. Do not freeze reagents. Protect from light and avoid contamination!

Concentrations in the test

1-Reagent	
imidazole buffer	100 mmol/l
glucose	20 mmol/l
N-acetylcysteine	20 mmol/l
magnesium acetate	10 mmol/l
EDTA	2 mmol/l
NADP	2 mmol/l
ADP	2 mmol/l
AMP	5 mmol/l
HK	> 2.5 U/ml
2-Reagent	
diadenosinepentaphosphate	10 µmol/l
glucose-6-phosphate-dehydrogenase (G6P-DH)	> 1.5 U/ml
creatine phosphate	30 mmol/l

Warnings and notes
- Product for in vitro diagnostic use only.
- The reagents contain sodium azide (< 0.1%) as a preservative. Avoid contact with skin and mucous membranes.
- Do not use reagents past the expiry date.
- Do not interchange caps among reagents.

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 15-25ºC or 1-2 days at 2-8ºC or 1 month at -20ºC
Nevertheless it is recommended to perform the assay with freshly collected samples.

PROCEDURE
The reagents are ready to use.
These reagents may be used in automatic analyser Hitachi 911/912.
Application should be entered using handheld barcode scanner and attached barcodes sheet, according to procedure described below:
1. Delete previous version of application and calibrators assigned to it and restart the analyser.
2. Enter codes of calibrators according to the attached list.
3. Enter barcoded application and assign proper values to calibrators.
4. To activate entered application go to the tab UTILITY | APPLICATION | RANGE and change value of field DATA MODE from INACTIVE to ON BOARD. Confirm the change using UPDATE button.
5. Put reagents on board the analyser – they will be assigned to relevant tests automatically. Perform also measurement of level of reagents inside the bottles.
6. After calibration analyser is ready to use.

REFERENCE VALUES [8]

serum / plasma	37ºC	
female	< 167 U/l	< 2.78 µkat/l
male	< 190 U/l	< 3.17 µkat/l

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

QUALITY CONTROL
For internal quality control it is recommended to use the CORMAY SERUM HN (Cat. No 5-172) and CORMAY SERUM HP (Cat. No 5-173) with each batch of samples.
For the calibration of automatic analysers systems the CORMAY MULTICALIBRATOR LEVEL 1 (Cat. No 5-174; 5-176) and LEVEL 2 (Cat. No 5-175; 5-177) are recommended. **Calibrators and 0.9% NaCl** should be used for calibration.
The calibration curve should be prepared every 12 weeks, with change of reagent lot number or as required e.g. quality control findings outside the specified range.

PERFORMANCE CHARACTERISTICS
These metrological characteristics have been obtained using the automatic analyser Hitachi 912. Results may vary if a different instrument or a manual procedure is used.

- **Sensitivity:** 8.21 U/l (0.137 µkat/l).

- **Linearity:** up to 2770 U/l (46.17 µkat/l).
  Samples with higher CK activity dilute 1:1 with 0.9% NaCl and repeat the assay. Multiply the result by 2.

- **Specificity / Interferences**
  Haemoglobin up to 0.125 g/dl, bilirubin up to 0.644 mg/dl, ascorbate up to 62 mg/l and triglycerides up to 1000 mg/dl do not interfere with the test.

- **Precision**

Repeatability (run to run) n = 10	Mean [U/l]	SD [U/l]	CV [%]
level 1	41.54	1.13	2.72
level 2	1536.71	8.12	0.53

Reproducibility (day to day) n = 10	Mean [U/l]	SD [U/l]	CV [%]
level 1	147.90	3.57	2.41
level 2	522.75	5.58	1.07

- **Method comparison**

A comparison between CK values determined at Hitachi 912 (y) and at Advia 1650 (x) using 59 samples gave following results:
y = 0.9831 x - 5.2185 U/l;
R = 1.0000 (R – correlation coefficient)

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).
7. Alan H.B. Wu: Tietz Clinical Guide to Laboratory Tests, 4th ed. WB Saunders, 634, (2006).
8. Dembińska-Kieć A., Naskalski J.W.: Diagnostyka laboratoryjna z elementami biochemii klinicznej, Volumed, 786, (1998).

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