ISE S.r.l.
CUSTOMISED SOLUTIONS FOR YOUR LABORATORY

CK NAC - Instruction for use

CE

IVD

R1: 4 x 50 mL REF A-R0200000301
R2: 4 x 20 mL

INTENDED USE
In addition to the possible risks regarding the reactive components, the product may contain non-reactive components such as preservatives (i.e. sodium azide or other) and detergents. The total concentration of these components is lower than the limits reported by the 67/548/EEC and 1999/45/EC directives and modifications and amendments regarding classification, labelling and packaging of dangerous preparations (reagents) have been made accordingly. However, it is recommended that this product be handled carefully, that ingestion and contact with eyes, skin and mucous membranes be avoided and that laboratory reagents are used according to good laboratory practice.

INTRODUCTION
Creatine kinase (CK) exists in 3 cytoplasmic forms: CK-MM (instriated and cardiac muscle), CK-MB (in cardiac muscle only),and CK- BB (especially in brain).Determination of CK is used for the diagnosis and the follow-upof muscular diseases (especially muscular dystrophies) and oflesions of the cardiac muscle. In myocardial infarction, the ratesof total CK and CK-MB increase quickly until reaching a peak 10-24h after the onset of the infarction. The levels return to normalwithin 3 to 4 days. Certain Central nervous system diseases and hypothyroidismalso trigger CK activity increase.

PRINCIPLE
The creatine kinase catalyses the reaction 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. The glucose-6phophate formed in presence of glucose-6-phosphate dehydrogenase (G6P-DH), reacts with ß-$NADP^+$ forming 6-phosphogluconate and ß-NADPH. The increase in absorbance due to the reaction ß-$NADP^+$ ---> ß-NADPH is proportional to the activity of CK in the sample. The presence of N-acetyl-L-cysteine (Nac) in the reaction mixture allows the optimal activation of the CK enzyme.

REAGENTS
Kit R1: 4 x 50 mL R2: 4 x 20 mL code A-R0000000301
Reagent 1: no. 4 vials x 40 mL
Reagent 2: no. 4 vials x 10 mL

Concentrations :

Reagent 1: R1	
Imidazole buffer, pH 6.10	125 mmol/L
D-Glucose	25 mmol/L
N-Acetyl-L-Cysteine	25 mmol/L
Magnesium acetate	12.5 mmol/L
NADP	2.4 mmol/L
EDTA	2.0 mmol/L
Hexokinase	≥ 6 800 U/L
Reagent 2: R2	
Creatine phosphate	250 mmol/L
ADP	15.2 mmol/L
AMP	25 mmol/L
Diadenosine pentaphosphate	103 µmol/L
G-6-PDH	≥ 8 800 U/L

Classification
According to regulation (EC) N°1272/2008

GHS08
Repr. 1B H360D May demage the unborn child.

Storage and stability
The components of the kit, stored at 2-8 °C in unopened vials, are stable up to the expiry date indicated on the package. Components of the kit and concentration of reactive ingredients

STANDARDIZATION
IFCC (International Federation of Clinical Chemistry and Laboratory Medicine).

SAMPLE COLLECTION

SAMPLE
Serum free from hemolysis (specimen recommended by IFCC). Heparinized plasma free from hemolysis.

STABILITY OF THE SAMPLE
Samples must be analyzed immediately or stored protected fromair and light for 8 hours at room température, 2 days at 2-8 °C or 1 month at -20 °C

Precaution
All human samples must be handled and disposed of as potentially infectious materials.

PROCEDURE
Quality control
Human control serum with known levels of CKnac is commercially available for quality control purposes. Data sheets are included, listing the values and the confidence limits. Normal and abnormal control sera are available from I.S.E. S.r.l. "Normal control serum" code R0400000006 and "Pathological control serum" code R0400000106. Obtained values must be within the range of acceptability. If erratic results occur, the following points should be checked:

- Cleanliness of glassware.
- Wavelength setting.
- Expiration date of reagents.

Automation
This kit, though developed and manufactured to be used as manual assay and with I.S.E. S.r.l. analyzer, can be used also with other analyzers able to meet the specifications indicated in section "Reaction conditions - Test procedure" Application sheets are available for automatic instruments.
All applications not explicitly approved by I.S.E. S.r.l. cannot be guaranteed in terms of performance, and must therefore be established by the operator.

Calibration
For calibration use the "Multicalibrator" I.S.E. S.r.l. code R0300000006.

Traceability:
The CKnac value is reported in the package insert supplied with the "Multicalibrator".

Calibration Stability
For the instrumentation series Miura, the calibration is recommended to be done every 2 weeks. Repeat the calibration at any variation in the reagent lot.

Method for automated instrumentation

Analyzer:	**I.S.E. Miura**		
Analyte Name :	**CKnac**	Ref.:	**A-R0200000301**
Method Code:	**CKnac**		
Type:	**Kinetic – Substrate start**		
Unit:	**UI/L**		
Filter F1:	**340**		
Blank in:	**Not Used**		
Step	**Reaction volume**		**U.M.**
Volume reagent R1:	**160**		µL
Sample volume:	**8**		µL
Incubation R1,S→R2	**300**		Sec.
Volume reagent R2:	**40**		µL
Final Incubation:	**120**		Sec.
Kinetic reading	**192**		Sec.

Reagents included in the kit
The reagent is described above.

Materials required but not supplied in the kit
Calibrators and controls.

CONVERSION FACTOR
CK [U/L] x0. 01667 = CK [µkat/L]

REFERENCE VALUES
Male: < **171** U/L
Female: < **145** U/L

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It is recommended that each laboratory establish its own expected range. For diagnostic purposes, results obtained should always be evaluated taking into consideration the patient's history and all other clinical findings.

ANALYTICAL CHARACTERISTICS / PERFORMANCE
SENSITIVITY:
Determined according to SFBC protocol, the detection limit is equal to 2 U/L.

ANALYTICAL RANGE:
8-1700 U/L.
Samples with values higher than 880U/L must be diluted 1:10 with normal saline and the result multiplied by 10.

INTRA-ASSAY PRECISION:

U/L	L1	L2	L3
Mean	75	187	562
SD	0.91	2.14	1.34
CV %	1.2	0.7	0.6

INTER-ASSAY PRECISION:

U/L	L1	L2	L3
Mean	92	168	496
SD	1.21	3.92	5.43
CV %	3.1	2.0	0.6

ACCURACY:
this test (y) was compared with a commercially available method (x). The results were as follows:
N = 60, r = 0.999, y = 1.0193 x +3 U/L

INTERFERENCES:
the test is not affected by the presence of ascorbic acid up to 100 mg/dL, haemoglobin up to 50 mg/dL and lipids up to 1000 mg/dL.
In haemolysed samples, the presence of haemoglobin can cause erroneously low bilirubin results; it is therefore recommended not to use haemolysed samples.

Disposal of reagent
Disposal of reagents must be performed in accordance with the EC regulations regarding waste, or the local national or regional legislation.
The product is in conformity with D.L: 8 September 2000, no. 332 "Actuation of the directive 98/79/EC regarding in vitro medical diagnostic devices".

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Symbols used on labels and packaging

IVD = In vitro diagnostic medical device
REF = Catalog Number
LOT = Lot Number
= Manufacturer
= Expiration date
= Temperature limitation
= Instruction for use

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