ALPHA AMYLASE (SINGLE REAGENT)

Cat. No.	Pack Name	Packaging (Content)
BLT00006	AMY SINGLE 100	R1: 5 x 20 ml

EN

CE IVD

INTENDED USE
Diagnostic reagent for quantitative *in vitro* determination of alpha-Amylase in human serum and plasma.

CLINICAL SIGNIFICANCE
α-Amylase is derived mainly from the salivary glands and the exocrine pancreas. α-Amylase catalyses the hydrolysis of α-1-4 glucosidic linkages of starch and other related polysaccharides to produce maltose and other oligosaccharides. The enzyme is a relatively small molecule which is rapidly cleared by the kidneys and excreted in the urine.
α-Amylase is most frequently measured in the diagnosis of acute pancreatitis when serum levels may be grossly elevated. In acute pancreatitis α-amylase starts to rise approximately 4 hours after the onset of pain, reaches a peak at 24 hours and remains elevated for 3-7 days. Hyperamylasamia is also associated with other acute abdominal disorders, biliary dysfunction, salivary gland disorders, ruptured ectopic pregnancy and macroamylasamia.

PRINCIPLE
2-Chloro-4-nitrophenol-β -1- 4 galactopyranosylmaltotrioside (CNP-G) is a direct substrate for determination of α-amylase activity, which does not require the presence of ancillary enzymes. The rate of 2-chloro-4-nitrophenol formation can be monitored at (400-420) nm and is proportional to the α-amylase activity.

Gal – G2 - α-CNP —(Amylase)→ Gal – G2 + CNP

REAGENT COMPOSITION
R1

MES buffer	50 mmol/l
Calcium Chloride	3.81 mmol/l
Sodium Chloride	300 mmol/l
Potassium Thiocyanate	450 mmol/l
Sodium Azide	13.85 mmol/l
CNPG	0.91 mmol/l

REAGENT PREPARATION
Reagent is liquid, ready to use.

STABILITY AND STORAGE
The unopened reagents are stable till the expiry date stated on the bottle and kit label when stored at 2–8°C.

SPECIMEN COLLECTION AND HANDLING
Use serum, plasma (heparin, EDTA), urine.
It is recommended to follow NCCLS procedures (or similar standardized conditions).
Stability

in serum/plasma:	7 days	at 20–25°C
	7 days	at 4–8°C
	1 year	at -20°C
in urine:	2 days	at 20–25°C
	10 days	at 4–8°C
	3 weeks	at -20°C

Discard contaminated specimens.

CALIBRATION
Calibration with calibrator XL MULTICAL, Cat. No. XSYS0034 is recommended.

QUALITY CONTROL
For quality control ERBA NORM, Cat. No. BLT00080 and ERBA PATH, Cat. No. BLT00081 are recommended.

UNIT CONVERSION
U/l x 0.017 = µkat/l

EXPECTED VALUES [7]
at 37°C
Serum: up to 80 U/l
Urine: up to 500 U/l
It is recommended that each laboratory verify this range or derives reference interval for the population it serves.

PERFORMANCE DATA
Data contained within this section is representative of performance on ERBA XL systems. Data obtained in your laboratory may differ from these values.
Limit of quantification: 10.8 U/l
Linearity: 1500 U/l
Measuring range: 10.8 – 1500 U/l

PRECISION

Intra-assay precision Within run (n=20)	Mean (U/l)	SD (U/l)	CV (%)
Sample 1	247.1	2.5	1.0
Sample 2	260.8	2.6	1.0

Inter-assay precision Run to run (n=20)	Mean (U/l)	SD (U/l)	CV (%)
Sample 1	58.3	1.3	2.2
Sample 2	142.9	2.3	1.6

COMPARISON
A comparison between XL-Systems Amylase (y) and a commercially available test (x) using 40 samples gave following results:
y = 0.973 x - 4.80 U/l
r = 0.989

INTERFERENCES
Following substances do not interfere:
haemoglobin up to 2.5 g/l, bilirubin up to 40 mg/dl, triglycerides up to 2000 mg/dl.
Note:
Saliva and skin contain alpha-amylase therefore never pipette reagents by mouth and avoid contamination of samples and reagents. Even trace contamination can affect results.

WARNING AND PRECAUTIONS
For *in vitro* diagnostic use. To be handled by entitled and professionally educated person.
Reagents of the kit are not classified like dangerous but contain less than 0.1% sodium azide - classified as very toxic and dangerous substance for the environment.

WASTE MANAGEMENT
Please refer to local legal requirements.

ASSAY PROCEDURE
Wavelength: 405 (400 – 420) nm
Cuvette: 1 cm

Working solution	1000 µl
Sample	20 µl

Mix, incubate 1 min. at 37°C and then measure the initial absorbance of calibrator and sample against reagent blank. Measure the absorbance change exactly after 1, 2 and 3 min. Calculate 1 minute absorbance change (ΔA/min).

CALCULATION
1. Amylase activity (U/l) = $\frac{\Delta A_{sam}/\text{min.}}{\Delta A_{cal}/\text{min.}} \times C_{cal}$

C_{cal} = calibrator concentration

2. Using factor:
Amylase activity (U/l) = f x ΔA/min
f = factor
f = 3128 (at 405 nm)

Applications for automatic analysers are available on request.

ASSAY PARAMETERS FOR PHOTOMETERS

Mode	Kinetic
Wavelength 1 (nm)	405
Sample Volume (µl)	10/20
Working Reagent Volume (µl)	500/1000
Lag time (sec.)	60
Kinetic interval (sec.)	60
No. of readings	3
Kinetic factor	3128
Reaction temperature (°C)	37
Reaction direction	Increasing
Normal Low (U/l)	0
Normal High (U/l)	80
Linearity Low (U/l)	10.8
Linearity High (U/l)	1500
Blank with	Water
Absorbance limit (max.)	0.14
Units	U/l

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SYMBOLS USED ON LABELS

REF Catalogue Number	Manufacturer	See Instruction for Use
LOT Lot Number	CE Mark - Device comply with the Directive 98/79/EC	Storage Temperature
Expiry Date	IVD In Vitro Diagnostics	CONT Content

QUALITY SYSTEM CERTIFIED
ISO 9001 ISO 13485

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