



PRINCIPLE

Alpha Amylase hydrolyses the p-Nitrophenol maltoheptoside producing Glucose polymers and p- nitrophenol oligosaccharides. These will be further hydrolyzed by glucoamylase and alpha glucosidase to release p-Nitrophenol. The rate of increase in absorbance at 405nm is proportional to Alpha Amylase activity.



CLINICAL SIGNIFICANCE

Amylase is produced by the pancreas and released into the intestinal tract. Amylase in the blood is eliminated through the kidneys and excreted into urine. Any elevation of the levels is reflected in the rise of urinary amylase activity. Measurement of alpha amylase in serum and urine is mainly used for the diagnosis of pancreatic disorders as well as (or detecting any complications.

In acute pancreatitis the blood amylase activity increases rapidly after onset of abdominal pain peaks after 12hrs and returns to values within reference range at the latest after 5 days. The specificity of Alpha amylase is for pancreatic disorders is not very high as elevated levels are observed also in various non pancreatic disorders like parotitis and renal insufficiency. Therefore for confirmation of an acute pancreatitis measurement of Lipase should be additionally performed.

SAMPLE COLLECTION & STORAGE

1. Non-homolysed serum sample is preferred
2. Plasma from heparin tubes may be used
3. Anticoagulants like citrate, and EDTA should be avoided

PRECAUTIONS

- * α Amylase Reagent is for in vitro diagnostic use only
- * Bring all reagents to room temperature before use

KIT CONTENTS & STORAGE

Reagent (CNPG3 + Buffer + Stabilizers) - 10ml 5 vials

All reagents are to be stored at 2-8⁰c and stable till expiry date mentioned.

REAGENT PREPARATION

Reagent is ready to use. Reagent to be discarded if the initial OD is > 0.60 at 405nm

SYSTEM PARAMETERS

Reaction type	Kinetic
Wavelength	405nm
Flow Cell Temp	37 ⁰ C
Reagent Volume	1.0ml
Sample Volume	25 μl
Delay time	60Sec
Interval	60Sec
No of Readings	2
Factor	4640
Units	IU/L
Zero Setting	Distilled Water

PROCEDURE

Pipette in a clean dry test tubes labeled as test(T)

Procedure	Test
Reagent	1.0ml
Sample	25ml

Mix well & read absorbance at 405nm after 60 seconds and then second reading at 90 seconds, third at 120 seconds and obtain the change of absorbance per minute($\Delta A/\text{Min}$)

CALCULATION

Activity of Amylase in sample(IU/L) $\Delta A/\text{Min} \times$
Factor (4640)

QUALITY CONTROL

It is recommended to include assayed quality control (serum level 1&2) with each assay batch to verify the performance of the procedure. Failure to obtain the proper range of values in the assay of control sera may indicate reagent deterioration, instrument malfunction or procedural errors.

LINEARITY

This method is linear up to 2000 IU/L . For sample values exceeding the linearity limit, dilute the sample suitable with normal and repeat the assay. Apply proper dilution factor while calculation.

NORMAL RANGE

Serum Amylase activity is : 25 - 125 IU/L

Urine : 1 - 17 IU/L

Due to variation in inter-laboratory assay conditions, instruments and demography, it is recommended that each laboratory should establish its own normal range.

Bibliography

1. Ramson, JHC *Cutt Prob Surg.* 1979-16:1
2. Henry RJ, Chlamor N. *Clin Chern* 1960:6:436