

<b>EN</b>	<b>REF A53704</b>	<b>Enzymatic colorimetric determination of pancreatic amylase (AMY-P) according to the IFCC mod. recommendations in serum and plasma</b>	<b>IVD</b>
	<b>Pancreatic Amylase</b>	<b>REAGENT 1: 2 x 40 mL - REAGENT 2: 2 x 11 mL</b> <b>SENTINEL</b> DIAGNOSTICS distributed by: <b>Beckman Coulter, Inc.</b>	<b>CE</b>
<b>NOTE:</b> This package insert must be read carefully prior to product use. Package insert instructions must be followed accordingly. Reliability of assay results cannot be guaranteed if there are any deviations from the instructions in this package insert.			

**INTENDED USE**

The Pancreatic Amylase assay is an *in vitro* diagnostic test used for the determination on AU480, AU680, AU5800 and DxC 700 AU automatic analyzers of the pancreatic amylase in serum and plasma by enzymatic colorimetric method. The assay is intended for professional use only.

**SUMMARY**

Alpha-amylases are hydrolytic enzymes which break down starch into maltose. In the human body, alpha-amylases originate from various organs that give the corresponding name to the enzyme. The pancreatic alpha-amylase is produced almost exclusively by the pancreas and released into the intestinal tract; the salivary alpha-amylase, mainly synthesized in the salivary glands, is secreted into saliva and is also present in tears, sweat and amniotic fluid. Pancreatic alpha-amylase assays are suitable for monitoring acute pancreatitis and acute attacks during chronic pancreatitis.

**PRINCIPLE**

The enzymatic colorimetric assay for pancreatic alpha-amylase determination is carried out in two successive steps. In the first incubation step, the activity of the human salivary alpha-amylase is inhibited using two different monoclonal antibodies with no effect on the pancreatic alpha-amylase. In the second reaction step the pancreatic alpha-amylase catalyzes the hydrolysis of the EPS substrate (Ethylidene Protected Substrate) p-nitrophenyl-maltoheptaoside 4,6-ethylidene-blocked (ethylidene-G7PNP) forming 2 ethylidene-G5 + 2 G2PNP + 2 ethylidene-G4 + 2 G3PNP + ethylidene-G3 + G4PNP. The  $\alpha$ -glucosidase hydrolyses all fragments of G2PNP, G3PNP and G4PNP into p-nitrophenol (PNP) and glucose (G). The increase of absorbance, due to PNP formation, is proportional to the activity of pancreatic alpha-amylase in the examined sample.

**REAGENTS**

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

**REAGENT 1:** HEPES\* buffer 52.5 mmol/L pH 7.15, sodium chloride 87 mmol/L, magnesium chloride 12.6 mmol/L,  $\alpha$ -glucosidase  $\geq 4$  kU/L, monoclonal antibodies anti salivary alpha-amylase  $\geq 30$  mg/L, sodium azide  $< 0.1\%$

**REAGENT 2:** HEPES\* buffer 52.5 mmol/L pH 7.15, 4,6-ethylidene-G7PNP  $\geq 4$  mmol/L, sodium azide  $< 0.1\%$

\* HEPES: 2-[4-(2-hydroxyethyl)-1-piperazinyl]-ethane sulfonic acid

**Test Estimated Tests per Kit°**

Pancreatic Amylase 640

° = calculation is based on minimum reagent fill volume per kit

**NOTES AND LIMITATIONS**

- A slight yellow colour of REAGENT 2 does not influence the product performance.
-  **CAUTION:** REAGENT bubbles may interfere with proper detection of reagent level causing insufficient reagent aspiration that could impact results.

**PREPARATION OF REAGENTS FOR USE**

- REAGENT 1 and REAGENT 2 are liquid and ready to use.
- Invert vials gently before removing screw caps.
- Remove screw caps from the reagent vials.
- Check each compartment for bubbles and remove any bubbles present.

**ON BOARD STABILITY OF REAGENTS**

30 days, if contamination is avoided.

**CALIBRATION**

For the calibration, use the following material:

**Clin Chem Cal** REF 16550 4 x 3 mL

Multiparametric lyophilised calibration serum. For use, follow the instructions contained in the kit.

**Calibration Stability:** 14 days.

Repeat the calibration at any variation in the reagent lot.

**Calculation:** the concentration is calculated using a linear calibration model.

**QUALITY CONTROL**

As appropriate, refer to your laboratory Standard Operating Procedures, Quality Assurance Plan for additional Quality Control Requirements and potential Corrective Actions.

- Two levels of Controls (normal and abnormal) are to be run every 24 hours.
- If more frequent control monitoring is required, follow the Quality Control procedures established by your laboratory.
- If Quality Control results do not meet the acceptance criteria defined by your laboratory, patient values may be suspect. Follow the Quality Control procedures established by your laboratory.
- Review Quality Control results and acceptance criteria following a change of reagent lot.

Use the following Beckman Coulter control materials to verify test accuracy:

**Control Serum 1 \*** REF ODC0003 \* 20 x 5 mL

**Control Serum 2 \*** REF ODC0004 \* 20 x 5 mL

For use, follow the instructions contained in the kit. For the target values refer to the concentration table (TcA) posted on web site:

[https://intranet.sentinel.it/BCI\\_Applications/](https://intranet.sentinel.it/BCI_Applications/)

\*: not available in all countries.

**STANDARDIZATION**

Enzymatic method according to IFCC (International Federation of Clinical Chemistry and Laboratory Medicine) mod. recommendations.

**SPECIMEN COLLECTION AND PREPARATION**

Serum or plasma (Li-heparin, Na-heparin). Collect samples in accordance with the NCCLS H3-A5 procedure<sup>1</sup>.

**Serum:** Use serum collected by standard venipuncture techniques. Ensure that complete clot formation has taken place prior to centrifugation. When processing samples, separate serum from blood cells according to the specimen collection tube's manufacturer instructions. Some specimens, especially those from patients undergoing anticoagulant or thrombolytic therapy, may take longer to complete the clotting process.

**Plasma:** Use plasma collected by standard venipuncture techniques. When processing samples, separate plasma from blood cells according to the specimen collection tube's manufacturer instructions.

**SAMPLE PREPARATION**

Sample preparation is not required.

**STABILITY OF THE SAMPLE<sup>8</sup>**

7 days at 2-8 °C or 12 months at -20 °C.

**PRECAUTIONS AND WASTE MANAGEMENT**

- For *in vitro* diagnostic use.
- Do not use components beyond the expiration date.
- Do not mix materials from different kit lot numbers.
- Safety Data Sheets are available at [https://intranet.sentinel.it/BCI\\_Applications/](https://intranet.sentinel.it/BCI_Applications/) or contact your local representative.

-  **CAUTION:** This product requires the handling of human specimens. It is recommended that all human sourced materials be considered potentially infectious and be handled in accordance with

the OSHA Standard on Bloodborne Pathogens<sup>2</sup>, Biosafety Level 2<sup>3</sup> or other appropriate biosafety practices<sup>4,5</sup> should be used for materials that contain or are suspected of containing infectious agents.

- This product contains sodium azide; for a specific listing, refer to the CONTENTS section of this package insert. Contact with acids liberates very toxic gas. This material and its container must be disposed of in a safe way.

#### MATERIALS PROVIDED

- Pancreatic Amylase REF A53704 R1:2x40mL, R2:2x11 mL

#### INSTRUMENTATION AND MATERIALS REQUIRED BUT NOT PROVIDED

- General laboratory equipment
- Normal Saline (NaCl 0.9 %)
- AU480, AU680 and AU5800 automatic analyzers

#### ANALYTICAL PROCEDURE

Refer to the instrument specific chemistry parameters.

#### RESULTS

Refer to the Instrument Specific Operations manual for information on results calculations.

#### CONVERSION FACTOR

The factor to convert U/L to  $\mu\text{kat/L}$  is: 0.01667  
(U/L x 0.01667 =  $\mu\text{kat/L}$ ).

#### REFERENCE VALUES<sup>10</sup>

Adults: **8 - 53 U/L**

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.

### PERFORMANCES on BECKMAN COULTER AU480 INTEGRATED SYSTEM

#### ANALYTICAL SENSITIVITY:

**Limit of Detection (LOD): 0.5 U/L**

LOD was determined on 20 replicates of normal saline x 3 runs and reported as the "mean zero value + 3 SD".

**Limit of Quantification (LOQ): 4.7 U/L**

LOQ was calculated on 8 decreasing levels obtained by dilutions. Result is reported as lowest concentration giving CV%  $\leq \pm 17.9\%$ .

#### INTER-ASSAY PRECISION:

was determined from 10x2x3 tests (day x run x rep) on 2 commercial controls (L1/L2) and on a human sera pool (L3). The results were as follows:

	Mean (U/L)	Total Imprecision		Between Days		Repeatability	
		SD	CV (%)	SD	CV (%)	SD	CV (%)
L1	36.8	0.51	1.4	0.39	1.0	0.27	0.7
L2	105.8	1.31	1.2	1.03	1.0	0.81	0.8
L3	24.5	0.43	1.8	0.25	1.0	0.28	1.1

#### ACCURACY:

this test (y) performed on Beckman Coulter AU 480 was compared on Abbott c8000 instrument (x). The results were as follows:

**N = 240, r = 0.999, y = 1.02x - 5.28**

#### INTERFERENCES

Interference studies were conducted using an acceptance criteria of  $\pm 10\%$  or 5.3 U/L deviation, whichever is greater, from the target value. The test is not affected by the presence of unconjugated bilirubin up to **66 mg/dL**, conjugated bilirubin up to **68 mg/dL** ascorbic acid up to **90 mg/dL**, haemoglobin up to **0.8 g/dL**, total proteins up to **14 g/dL**, and triglycerides up to **1000 mg/dL**.

#### MEASURING RANGE

**4.7 - 2000 U/L.**

Samples with results over the range must be confirmed by manual dilution with saline, and rerun of test. The result must be multiplied by the dilution factor.

#### WASTE MANAGEMENT

Reagents must be disposed of in accordance with local regulations.

#### ADDITIONAL INFORMATION

For more detailed information on AU Systems, refer to the appropriate system manual. Since Beckman Coulter does not manufacture the reagent or perform quality control or other tests on individual lots, Beckman Coulter cannot be responsible for the quality of the data obtained which is caused by performance of the reagent, any variation between lots of reagent, or protocol changes by the manufacturer.

#### SHIPPING DAMAGE

Please notify your Beckman Coulter Clinical Support Centre if this product is received damaged.

#### FOR OTHER LANGUAGES AND TECHNICAL INFORMATION VISIT

[https://intranet.sentinel.it/BCI\\_Applications/](https://intranet.sentinel.it/BCI_Applications/)

#### BIBLIOGRAPHY

- 1) NCCLS Procedures for the Collection of Diagnostic Blood Specimens by Venipuncture; Approved Standard - Fifth Edition (H3-A5). Wayne, PA: The National Committee for Clinical Laboratory Standards, 2003.
- 2) US Department of Labor, Occupational Safety and Health Administration. 29 CFR Part 1910.1030. Bloodborne Pathogens.
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- 4) World Health Organization. Laboratory Biosafety Manual, 3rd ed. Geneva: World Health Organization, 2004.
- 5) Sewell DL, Bove KE, Callihan DR, et al. Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline - Third Edition (M29-A3). Wayne, PA: Clinical and Laboratory Standards Institute, 2005.
- 6) Pesce, A.J., Kaplan, L.A.: "Methods in Clinical Chemistry", Mosby Ed. (1996).
- 7) Burtis C.A., Ashwood E.R.: "Tietz Textbook of Clinical Chemistry", W.B. Saunders Company Ed. (3<sup>rd</sup> edition, 1999).
- 8) Guder W.G.: "The Quality of Diagnostic Sample". Recommendations of the Working Group on Preanalytical Quality of the German Society for Clinical Chemistry and the German Society for Laboratory Medicine. (1<sup>st</sup> Edition - 2001).
- 9) Jakobs, D.S., Kasten, Jr., B.L., DeMott, W.R., Wolfson, W.L.: "Laboratory Test Handbook", Lexi-Comp and Williams & Wilkins Ed. (2<sup>nd</sup> Edition - 1990).
- 10) Junge W, Waldenström J, Bouman A et al. Evaluation of the Assays for Total and Pancreatic  $\alpha$ -Amylase based on 100% Cleavage of Et-G7-PNP at 6 European Clinical Centres (Poster Medlab 97). Basel, Switzerland: 12th IFCC European Congress of Clinical Chemistry, August 17-22, 1997.

#### Explanation of symbols

##### REAGENT / STANDARD / CALIBRATOR / CONTROL

The terms refers to the: single reagent / standard / calibrator / control

**IVD**

In vitro Diagnostic Medical Device

**REF**

Catalogue number

**LOT**

Batch code

**Cont.**

Contents of kit

**distributed by**

Distributed by



Caution, consult accompanying documents



Consult instructions for use



Use by



xxx

Contains sufficient for <n> tests



Temperature limitation



Manufacturer

**Note: changes in comparison to the previous version are indicated by a vertical bar in the text margin.**