HEALTH HOLDING

HAFER ALBATIN HEALTH CLUSTER
MATERNITY AND CHILDREN HOSPITAL

| | | | |
|---|---|---|---|
| **Department:** | Laboratory and Blood Bank ( Chemistry ) | | |
| **Document:** | Internal Policy and Procedure | | |
| **Title:** | Analysis of Amylase Level | | |
| **Applies To:** | All Laboratory Staff | | |
| **Preparation Date:** | January 06, 2025 | **Index No:** | LB-IPP-153 |
| **Approval Date:** | January 20, 2025 | **Version :** | 2 |
| **Effective Date:** | February 20, 2025 | **Replacement No.:** | LB-IPP-153(1) |
| **Review Date:** | February 20, 2028 | **No. of Pages:** | 05 |

## 1. PURPOSE:

1.1 The purpose of this policy and procedure is to provide all information related to the analysis of amylase level in blood (serum/plasma) on DimensionEXL200 ,Synchron DXC700 and Atelica CI .machines.

## 2. DEFINITONS:

2.1 The a-amylases (1,4-a-D-glucanohydrolases, EC 3.2.1.1) catalyse the hydrolytic degradation of polymeric carbohydrates.

## 3. POLICY:

3.1 This policy provides instructions for performing the quantitative determination of amylase in human serum plasma or urine at PH7 on DimensionEXL200 ,Synchron DXC700 and Atelica CI .machines.

3.2 The amylases (1,4-a-D-glucanohydrolases, EC 3.2.1.1) catalyse the hydrolytic degradation of polymeric carbohydrates such as amylose, amylopectin and glycogen by cleaving 1,4-a-glucosidic bonds.

3.3 Serum or plasma amylase level is increased in acute pancreatitis, pancreatic neoplasm, perforated duodenal ulcer, intestinal obstruction, acute cholecystitis, diabetic ketosis, salivary gland disorder (notablymumps), ascites, burns, carcinoma of oesophagus, ruptured ectopic pregnancy.

3.4 Sample of urine and other body fluids can also be used.

## 4. PROCEDURE:

4.1 **Specimen:**

4.1.1 Type:

4.1.1.1 Serum, plasma, or urine sample collected at PH 7 and refrigerated.

4.1.2 Tube Type:

4.1.2.1 Gel tube, Plain tube, Li-Heparin

4.1.3 Amount Required:

4.1.3.1 2.0 to 3.0 ml

4.1.4 Delivery Arrangements:

4.1.4.1 Sample to be delivered to the lab as soon as possible.

4.1.5 If the sample is serum should be ensuring complete clot formation before centrifugation. Some specimens, especially those from patients receiving anticoagulant or thrombolytic therapy, may exhibit increased clotting time. If the specimen is centrifuged before a complete clot forms, the presence of fibrin may cause erroneous results.

4.1.6 Temperature Restrictions:

4.1.6.1 At room temperature

4.1.7 Unacceptable Specimen:

4.1.7.1 See sample rejection criteria policy

4.1.8 Specimen Retention:

4.1.8.1 Period of retention: up to one week after separation of the sample.

4.1.8.2 Storage condition: store at 2-8 $^{0}$C.

4.1.9 Safety Precaution:

4.1.9.1 Treat all samples as potentially infectious and handle in accordance with the OHSA standard on blood borne pathogen.

4.2 **Principle:**

4.2.1 A-Amylase catalyzes the hydrolysis of a defined synthetic substrate 2-choloro-4-nitrophenyl-a-D maltotrioside (CNPG3), to yield 2- choloro-4-nitrophenol (CNP),2-cholro-4-nitrophenyl-a-D-maltoside (CNPG2) ,maltotriose (G3) and glucose .

4.2.2 CNPG3 -----amylase---------- CNP +CNPG2 + G3 +glucose

4.2.3 The colour intensity of the p-nitrophenol formed is directly proportional to the a-amylase activity.

4.2.4 It is determined by measuring the increase in absorbance.

4.3 **Method:**

4.3.1 See policy of loading sample on machine (Ref: Operative Manuals' of DimensionEXL200 ,Synchron DXC700 and Atelica CI machines.

4.4 **Calculation:**

4.4.1 Instrument system automatically calculates the Analytic activity and gives results in the form of print outs.

4.5 **Format:**

4.5.1 Numeric

4.6 **Reference range:**

4.6.1 Serum/plasma 25 — 115 U/ L

4.6.2 Urine: 24-hour urine 59 — 401 U/ 24 hr

4.6.3 AMY/CREA Ratio 1.3 — 4.3 %

4.7 **Dilution information:**

4.7.1 Specimens with values exceeding the linearity range are flagged and may be diluted with automatic dilution or manual dilution

4.7.2 Manual Dilution should be performed as follows:

4.7.2.1 Use saline (0.85% to 0.90%) to dilute the sample

4.7.2.2 The operator must enter the dilution factor in the patient order screen. The system uses this

4.7.2.3 If the operator does not enter the dilution factor, the result must be multiplied by the appropriate dilution factor before reporting the result.

4.7.2.4 If a diluted sample result generates a Linear Low (LL) result error code, do not report the result. Prepare an appropriate dilution/concentration and rerun.

4.8 **Test Limitation:**

4.8.1 Recognizing:

4.8.1.1 Haemolysed sample

4.8.1.2 Lipaemia: because of absorbance flagging > 600 mg/di

4.8.1.3 Icterus: bilirubin > 80 mg/dl

4.8.1.4 Ammonium ions may cause erroneously elevated results

4.8.2 Avoiding Error:

4.8.2.1 Following acceptance criteria of the sample

4.8.2.2 By following the maintenance protocol. Daily, weekly, month

4.8.2.3 Run control before starting the tests

4.8.3 Error Correction:

4.8.3.1 Look for a fibrin clot or air bubbles

4.8.3.2 Repeat the sample from the original tube

4.8.3.3 Ask for another sample

4.9 **Specific Performance Characteristics:**

4.9.1 Linearity: Amylaseis linear up to 650 U/L

4.9.2 Limit of Detection: The Limit of Detection is 0

4.10 **Procedure Note:**

4.10.1 For in vitro diagnostic use
4.10.2 Do not use component beyond expiration date
4.10.3 Do not mix components from different kit lot numbers
4.10.4 Protect from strong light for optimum stability
4.10.5 Remove air bubbles that may interfere with proper reagent level detection

## 5. MATERIALS AND EQUIPMENT:

5.1 **Reagents:**

5.1.1 Mylase flex contains 6 wells with the following ingredients:

| Reactive Ingredients | Ingredients Concentration |
|---|---|
| Liquid1-6 wells) | |
| CNPG3 j | 1.24 mmol/L |

5.1.2 Reagent Preparation:

5.1.2.1 Mixing and diluting are automatically performed by the Dimension system
5.1.2.2 Estimated test per cassette, 60 tests

5.1.3 Analytical Range:

5.1.3.1 Serum/plasma 0-650 U/L

5.1.4 Reagents retention:

5.1.4.1 The unopened reagents are stable until the expiration date when stored at 2-8 °C. Reagent stability is 30 days if the reagent is unopened and for 3 days if the reagent is opened.

5.2 **Calibration:**

5.2.1 Calibration is stable approximately 30 days and required with each change in reagent lot number. Verify calibration curve with at least two levels of controls according to the established Quality Control requirements for your laboratory.

5.2.2 Calibration must be done when:

5.2.2.1 A complete change of reagents that affects the range used to report patient results or QC value.
5.2.2.2 A reagent kit with new lot number is used.
5.2.2.3 A new assay file that requires a calibration is installed.
5.2.2.4 QC fails to meet the established criteria.
5.2.2.5 After major maintenance or service.
5.2.2.6 When recommended by the manufacturer.
5.2.2.7 Documentation accompanying a new version of an existing file states calibration is required.
5.2.2.8 At least every 6 months.

5.2.3 Calibrator retention 24 h at 2 - 8 °C:

5.2.3.1 Instability or deterioration should be suspected if there are visible signs of leakage, extreme turbidity microbial growth or if calibration does not meet the appropriate package insert and/or instrument-operation manual criteria.

5.2.4 Calibration Procedure:

5.2.4.1 Calibration is performed by running Distilled Water and 3 levels of enzyme verifier calibrator for Dimension machines.
5.2.4.2 Verity that the correct calibrator values have been entered into the calibration file. For details refer to Operator Guide of DimensionEXL200 ,Synchron DXC700 and Atelica CI .machines.
5.2.4.3 Allow calibrator to come to room temperature.
5.2.4.4 Mix bottle 10 times by inversion.
5.2.4.5 Open bottle, place a minimum of 300 ul of each level in separate sample cup, and place on the assigned positions.
5.2.4.6 Cap bottle tightly and store at 2-8°C. Immediately after use.

5.2.4.7 Perform calibration as indicated in Operator Guide of DimensionEXL200 ,Synchron DXC700 and Atelica CI . machines.

5.2.5 Calibration Expected Values:

5.2.5.1 Refer to enzyme verifier calibrator for Dimension.

5.2.5.2 Refer to Synchron calibrator leaflet

5.3 **Quality Control:**

5.3.1 Normal and pathological control. One time in 24 hours (once per day).

5.3.2 If more frequent control monitoring is required, follow the established quality control procedures your laboratory.

5.3.3 If quality control results do not fall within an acceptable range defined by your laboratory, may be affected and corrective action should be taken.

5.3.4 Quality Control retention:

5.3.4.1 Unopened control vial is stable up to expiry date printed on the label when stored at cold room.

5.3.4.2 Opened control vial is stable for: After reconstituting and tightly capped at 2 — 8 °C. All analytes will be stable for 7 days except Bilirubin (Direct) for 4 days.

5.3.5 QC Procedure:

Verify that the correct QC values have been entered into the QC file. For details refer to Operator Guide of DimensionEXL200 ,Synchron DXC700 and Atelica CI .machines.

5.3.5.1 Allow QC to come to room temperature.

5.3.5.2 Gently remove the stopper to avoid loss of the lyophilized pellet and add exactly 5.0 ml distilled or de-ionized water.

5.3.5.3 Leave to stand for 20 minutes.

5.3.5.4 Mix bottle several times by inversion to allow homogeneity.

5.3.5.5 Gently invert just prior to use. Avoid foaming.

5.3.5.6 Open bottle, place a minimum of 1000 ul of each level in separate sample cup, and place on the assigned positions.

5.3.5.7 Cap bottle tightly and store at 2-8°C. Immediately after use.

5.3.5.8 Perform QC as indicated in Operator Guide of DimensionEXL200 ,Synchron DXC700 and Atelica CI . machines.

5.3.6 QC Expected Values: Refer to the BioradLyphochek assayed chemistry controls value sheet for DimensionEXL200 ,Synchron DXC700 and Atelica CI .machines.

## 6. RESPONSIBILITIES:

6.1 Chemistry shift in charge is responsible for, running calibration and control and samples of Amylase.

6.2 Chemistry staff are responsible for running Amylase samples all over the day.

## 7. APPENDICES:

7.1 N/A

## 8. REFERENCES:

8.1 Tietz Text Book of clinical chemistry and molecular diagnostics 4th Edition,2006.

8.2 Company Leaflets of reagents.

## 9. APPROVALS:

| | Name | Title | Signature | Date |
|---|---|---|---|---|
| **Prepared by:** | Dr. Talal Abdelgawad | Clinical Pathologist | | January 06, 2025 |
| **Reviewed by:** | Dr. Kawther M. Abdou | Consultant & Lab. Medical Director | | January 13, 2025 |
| **Reviewed by:** | Ms. Noora Melfi Alanizi | Laboratory & Blood Bank Director | | January 13, 2025 |
| **Reviewed by:** | Mr. Abdulelah Ayed Al Mutairi | QM&PS Director | | January 13, 2025 |
| **Reviewed by:** | Dr. Tamer Mohamed Naguib | Medical Director | | January 13, 2025 |
| **Approved by:** | Mr. Fahad Hazam Alshammari | Hospital Director | | January 20, 2025 |