

# Clinical Chemistry IMPC\_CBC\_003

## Purpose

Clinical chemistry determines biochemical parameters in plasma including enzymatic activity, specific substrates and electrolytes.

Ontological description: MP:0001545 – blood physiology abnormalities.

## Experimental Design

- **Minimum number of animals** : 7M + 7F
- **Age at test**: Week 16
- **Sex**: We would expect the results of this test to show sexual dimorphism

## Equipment

1. Clinical chemistry analyser
2. Vortex
3. Refrigerated centrifuge
4. Eppendorf tubes
5. Pipettes (200-1000 ul)

## Procedure

Set up the clinical chemistry analyser and perform QC analyses of the control reagents in accordance with the equipment guidelines.

### Sample collection and preparation:

- a. Collect the appropriate volume of blood required (160-200l of plasma), for the clinical chemistry analyser being used for assessment, in gel tube containing lithium Heparin with the relevant blood collection procedure (see IMPC protocol Blood collection by retro-orbital puncture). Time of day for collection is in the morning, starting no earlier than 07:30.
- b. Keep whole blood samples in a bag on wet ice until centrifugation. Centrifuge for 10 minutes at 5000 x g in a refrigerated centrifuge set at 8°C. If plasma samples cannot be analysed immediately, keep them in the fridge until analysis.
- c. Analysis of samples is optimally done on the day of collection. When not possible the plasma samples can be stored at 2-8°C. If samples require storage for > 48 hours,

freeze plasma at -20 °C in single aliquots. All samples are allowed to come to room temperature prior to analysis.

- d. Use plasma samples undiluted or diluted to a ratio of 1:2 with deionised water if the volume is insufficient.
- e. Plasma samples that were frozen or stored in the fridge should be vortexed briefly and centrifuged again at ~5000 x g for 2-3 minutes immediately prior to analysis. If necessary, remove fibrin clots using a wooden applicator.

## Analysis:

Samples that produce results that lie outside the linear range for a specific assay have to be re-tested. In some cases it may be necessary to dilute samples with water to bring test results into range.

## Notes

Blood collection for Clinical Chemistry and Hematology is usually performed as a non-fasting, terminal procedure but can be performed as a non-terminal procedure under certain circumstances. Mice from the terminal procedure may be used for subsequent gross pathology and other procedures included in terminal assessments. Whole blood (for Hematology) and plasma (for Clinical Chemistry) require different collection tubes so two independent samples are required from each mouse.

The information about the date of the experiment, that is the date when the measurement is performed, is an important parameter which is to be submitted in the Experiment xml file (dateOfExperiment="2013-02-28").

**Dilution.** Dilution of blood is highly discouraged, but is allowed when the total necessary amount is not obtained. If dilution is necessary then the assays should be done in one run.

**Hemolysis.** Two fields currently exist to capture metadata information about the hemolysis status in the clinical chemistry plasma samples. The first is the LIH Hemolysis severity score which can only be performed by clinics who run one of the Beckman Coulter AU-series of analysers. Such clinics are encouraged to capture and submit the hemolysis score of the LIH test in this field. Clinics who do not have an AU analyser are encouraged to use the second /alternative field which is simply titled Hemolysis. Simply enter "slight", "moderate", or "marked" based on whether the sample is visibly haemolysed or not. Provision of this information is not compulsory and it is suggested that any clinic completes at least one field or the other (not both).

## Data QC

1. Plasma samples must be free of Fibrin clots in order to be analysed.
2. Badly haemolysed samples should be discarded.
3. Each morning, all parameters are tested with control sera (see ESLIM\_015\_001\_Annex\_3: Controls for biochemistry on AU400). Some parameters are tested with control serum level 1 (Beckman Coulter System Reagent, ODC0003)

and control serum level 2 (Beckman Coulter System Reagent, ODC0004), which consists of lyophilised human plasma with a normal and a pathological concentration. Other parameters are tested with specific controls from other suppliers.

4. Controls are thawed and vortexed before utilisation and loaded according to the analyser's display. Control values must lie within the acceptable range indicated by the manufacturer, otherwise the specific tests must be recalibrated and specific measurements repeated. Controls can be stored in 200l aliquots at -20°C for up to 1 week.

## Metadata and examples

| Metadata                                            | Example                                                                                                                                                   |
|-----------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------|
| Equipment ID                                        | ID of the machine used when more than 1 is used having same model and manufacturer. E.g. machine 1, machine 2, machine Minnie, machine Mickey Mouse, etc. |
| Equipment manufacturer                              | Manufacturer of the equipment. E.g. Olympus Diagnostics.                                                                                                  |
| Equipment model                                     | Model of the equipment. E.g. AU400                                                                                                                        |
| Blood collection tubes                              | The tubes used for blood collection. E.g. Sarstedt Li-Heparin gel tubes or Kabe Labortechnik Lithium heparin coated tubes.                                |
| Anaesthesia used for blood collection               | The drug used for anaesthesia during blood collection. E.g. Isoflurane.                                                                                   |
| Method of blood collection                          | Concise description of the method used for blood collection. E.g. retro-orbital puncture.                                                                 |
| Anticoagulant                                       | Anticoagulant drug used for blood collection. E.g. Li-Heparin.                                                                                            |
| Samples kept on ice between collection and analysis | Yes/No.                                                                                                                                                   |
|                                                     |                                                                                                                                                           |

|                                                            |                                                                                                                                                                                          |
|------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Storage temperature from blood collection till measurement | E.g. 2°C                                                                                                                                                                                 |
| Sample status                                              | Indicate if the sample were frozen (analysis on the same day of collection not possible) or fresh (analysis on the same day of collection). E.g Fresh/Frozen.                            |
| Plasma dilution                                            | Dilution is highly discouraged but if necessary indicate here. E.g. "No dilution" or 1:2. Note that results submitted to DCC are assumed to be already corrected for any dilutions made. |
| ID of blood collection SOP                                 | ID of the protocol followed for blood collection. Can be a center specific protocol. E.g. ESLIM_024_001.                                                                                 |
| Date and time of blood collection                          | Time of day for collection is in the morning, starting no earlier than 07:30. E.g. Year, month, day, time.                                                                               |
| Date of measurement                                        | The day of blood analysis. Year, month, day.                                                                                                                                             |
| Hemolysis status                                           | If no AU analyser score is provided, indicate here the gauged degree of hemolysis. E.g. slight/moderate/marked.                                                                          |
| Blood collection experimenter ID                           | An ID of any format to be used coherently both inside the same procedure and for all procedures indicating the experimenter who collected the blood. E.g. Harw_001, or 1/2/3.            |
| Blood analysis experimenter ID                             | An ID of any format to be used coherently both inside the same procedure and for all procedures indicating the experimenter who analyzed the blood. E.g. Harw_001, or 1/2/3.             |
| Date equipment last calibrated                             | Most recent date in which the equipment (or any part of) used in the procedure was subject to a calibration event.                                                                       |
| Date and time of sacrifice                                 | The date and time when the mouse is sacrificed.                                                                                                                                          |

# Parameters and Metadata

## Sodium IMPC\_CBC\_001\_001 | v1.3

simpleParameter

Req. Analysis: false

Req. Upload: false

Is Annotated: true

Unit Measured: mmol/l

Description: sodium

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## Potassium IMPC\_CBC\_002\_001 | v1.3

simpleParameter

Req. Analysis: false

Req. Upload: false

Is Annotated: true

Unit Measured: mmol/l

Description: potassium

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## Chloride IMPC\_CBC\_003\_001 | v1.4

simpleParameter

Req. Analysis: false

Req. Upload: false

Is Annotated: true

**Unit Measured:** mmol/l

**Description:** chloride

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**Urea (Blood Urea Nitrogen - BUN)** IMPC\_CBC\_004\_001 | v1.5

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** mg/dl

**Description:** urea

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**Creatinine** IMPC\_CBC\_005\_001 | v1.5

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** mg/dl

**Description:** creatinine\_enzymatic\_method\_preferred\_

---

**Total protein** IMPC\_CBC\_006\_001 | v1.2

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** g/l

**Description:** total\_protein

---

**Albumin** IMPC\_CBC\_007\_001 | v1.2

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** g/l

**Description:** albumin

---

**Total bilirubin** IMPC\_CBC\_008\_001 | v1.4

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** mg/dl

**Description:** total\_bilirubin

---

**Calcium** IMPC\_CBC\_009\_001 | v1.5

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** mg/dl

**Description:** calcium

---

## Phosphorus IMPC\_CBC\_010\_001 | v1.6

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** mg/dl

**Description:** phosphate

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## Iron IMPC\_CBC\_011\_001 | v1.5

simpleParameter

**Req. Analysis:** false

**Req. Upload:** false

**Is Annotated:** true

**Unit Measured:** mg/dl

**Description:** iron

---

## Aspartate aminotransferase IMPC\_CBC\_012\_001 | v1.2

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** U/l

**Description:** aspartate\_aminotransferase

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## Alanine aminotransferase IMPC\_CBC\_013\_001 | v1.2

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** U/l

**Description:** alanine\_aminotransferase

---

## Alkaline phosphatase IMPC\_CBC\_014\_001 | v1.2

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** U/l

**Description:** alkaline\_phosphatase

---

## Total cholesterol IMPC\_CBC\_015\_001 | v1.4

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** mg/dl

**Description:** total\_cholesterol

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## **HDL-cholesterol** IMPC\_CBC\_016\_001 | v1.4

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** mg/dl

**Description:** hdl\_cholesterol

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## **Triglycerides** IMPC\_CBC\_017\_001 | v1.4

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** mg/dl

**Description:** triglycerides

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## **Glucose** IMPC\_CBC\_018\_001 | v1.5

simpleParameter

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** true

**Unit Measured:** mg/dl

**Description:** glucose

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## **LIH (Hemolysis Severity - available on AU analysers)** IMPC\_ CBC\_019\_001 | v1.3

simpleParameter

**Req. Analysis:** false

**Req. Upload:** false

**Is Annotated:** false

**Description:** lih\_hemolysis\_severity\_available\_on\_au\_analysers\_

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## **Fructosamine** IMPC\_CBC\_020\_001 | v1.2

simpleParameter

**Req. Analysis:** false

**Req. Upload:** false

**Is Annotated:** true

**Unit Measured:** umol/l

**Description:** fructosamine

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## **Lipase** IMPC\_CBC\_021\_001 | v1.1

simpleParameter

**Req. Analysis:** false

**Req. Upload:** false

**Is Annotated:** true

**Unit Measured:** U/l

**Description:** lipase

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## Lactate dehydrogenase IMPC\_CBC\_022\_001 | v1.2

simpleParameter

**Req. Analysis:** false

**Req. Upload:** false

**Is Annotated:** true

**Unit Measured:** U/l

**Description:** lactate\_dehydrogenase

---

## Alpha-amylase IMPC\_CBC\_023\_001 | v1.2

simpleParameter

**Req. Analysis:** false

**Req. Upload:** false

**Is Annotated:** true

**Unit Measured:** U/l

**Description:** alpha\_amylase

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# UIBC (unsaturated iron binding capacity) IMPC\_CBC\_024\_001 | v1.0

simpleParameter

Req. Analysis: false      Req. Upload: false      Is Annotated: true

Unit Measured: umol/l

Description: uibc\_unsaturated\_iron\_binding\_capacity\_

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# LDL-cholesterol IMPC\_CBC\_025\_001 | v1.3

simpleParameter

Req. Analysis: false      Req. Upload: false      Is Annotated: true

Unit Measured: mg/dl

Description: ldl\_cholesterol

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# Free fatty acids IMPC\_CBC\_026\_001 | v1.4

simpleParameter

Req. Analysis: false      Req. Upload: false      Is Annotated: true

Unit Measured: mmol/l

Description: free\_fatty\_acids

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## Glycerol IMPC\_CBC\_027\_001 | v1.4

simpleParameter

Req. Analysis: false

Req. Upload: false

Is Annotated: true

Unit Measured: mmol/l

Description: glycerol

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## Creatine kinase IMPC\_CBC\_028\_001 | v1.2

simpleParameter

Req. Analysis: false

Req. Upload: false

Is Annotated: true

Unit Measured: U/l

Description: creatine\_kinase

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## Uric acid IMPC\_CBC\_029\_001 | v1.2

simpleParameter

Req. Analysis: false

Req. Upload: false

Is Annotated: true

Unit Measured: umol/l

Description: uric\_acid

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## Ferritin IMPC\_CBC\_030\_001 | v1.3

simpleParameter

Req. Analysis: false

Req. Upload: false

Is Annotated: true

Unit Measured: ng/ml

Description: ferritin

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## Transferrin IMPC\_CBC\_031\_001 | v1.2

simpleParameter

Req. Analysis: false

Req. Upload: false

Is Annotated: true

Unit Measured: mg/dl

Description: transferrin

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## C-reactive protein IMPC\_CBC\_032\_001 | v1.0

simpleParameter

Req. Analysis: false

Req. Upload: false

Is Annotated: true

Unit Measured: mg/l

Description: c\_reactive\_protein

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## Equipment ID IMPC\_CBC\_033\_001 | v1.0

procedureMetadata

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** false

**Description:** equipment\_name

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## Equipment manufacturer IMPC\_CBC\_034\_001 | v1.0

procedureMetadata

**Req. Analysis:** true

**Req. Upload:** true

**Is Annotated:** false

**Description:** equipment\_manufacturer

**Options:** Cobas, Olympus Diagnostics, Beckman Coulter, Hitachi, JEOL (Siemens), Roche,

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## Equipment model IMPC\_CBC\_035\_001 | v1.0

procedureMetadata

**Req. Analysis:** true

**Req. Upload:** true

**Is Annotated:** false

**Description:** equipment\_model

**Options:** Integra 400 Plus, AU 400, AU 480, 7020, JCA-BM2250 (Advia 2400), Hitachi 917, AU 680, UniCel 600 Pro, JCA-BM6070, DxC AU 700,

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## Anesthesia used for blood collection IMPC\_CBC\_036\_001 | v1.0

procedureMetadata

**Req. Analysis:** true

**Req. Upload:** true

**Is Annotated:** false

**Description:** anesthesia\_used\_for\_blood\_collection

**Options:** Gas anaesthesia with Isoflurane,  
Injection narcosis with Ketamine (100mg/kg)/Xylazine (10mg/kg),  
Injection narcosis with Ketamine (100mg/kg)/ Xylazine (10mg/kg )/Antipamezole (Antisedan, 1mg/kg),  
Injection narcosis with Ketamine (110mg/kg)/Xylazine (11mg/kg),  
Injection narcosis with Ketamine (110mg/kg)/Xylazine (11mg/kg)/ Antipamezole (Antisedan, 1mg/kg),  
Injection narcosis with Tribromoethanol (Avertin),  
Injection narcosis with Sodium Pentobarbital (Pentobarb, 0.1ml),  
Injection narcosis with Sodium Pentobarbital (Euthatal), No,  
Injection narcosis with Ketamine (137mg/kg)/Xylazine (6.6mg/kg),  
Injection narcosis with Sodium Pentobarbital (Somnopentyl),  
Topical local anaesthesia with EMLA,

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## Method of blood collection IMPC\_CBC\_037\_001 | v1.0

procedureMetadata

**Req. Analysis:** true

**Req. Upload:** true

**Is Annotated:** false

**Description:** method\_of\_blood\_collection

**Options:** Cardiac puncture, Retro-orbital puncture, Heart puncture, Jugular vein, Tail vein,

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## Anticoagulant IMPC\_CBC\_038\_001 | v1.1

procedureMetadata

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** false

**Description:** anticoagulant

**Options:** No, Lithium Heparin, Sodium Heparin, Heparine,

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## Samples kept on ice between collection and analysis IMPC\_

CBC\_042\_001 | v1.1

procedureMetadata

**Req. Analysis:** true

**Req. Upload:** true

**Is Annotated:** false

**Description:** samples\_kept\_on\_ice\_between\_collection\_and\_analysis

**Options:** Yes, No,

---

## Sample status IMPC\_CBC\_043\_001 | v1.1

procedureMetadata

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** false

**Description:** sample\_status

**Options:** Fresh, Frozen, Fresh and frozen,

---

## Sample dilution IMPC\_CBC\_044\_001 | v1.2

procedureMetadata

Req. Analysis: false

Req. Upload: true

Is Annotated: false

Description: plasma\_dilution

Options: Neat plasma, 1:2, Yes (by Equipment, automatically), Varies, Neat serum, 1:3, 1:4, 1:5,

---

## ID of blood collection SOP IMPC\_CBC\_045\_001 | v1.1

procedureMetadata

Req. Analysis: false

Req. Upload: true

Is Annotated: false

Description: id\_of\_blood\_collection\_sop

Options: ESLIM\_024\_001, sop.inv.019, RIKENMPP\_004a\_003, PHENO\_CBC, sop.inv.063, Biochem\_blood SOP,

---

## Date and time of blood collection IMPC\_CBC\_046\_001 | v1.2

procedureMetadata

Req. Analysis: false

Req. Upload: true

Is Annotated: false

Description: date\_and\_time\_of\_blood\_collection

---

## Hemolysis status IMPC\_CBC\_048\_001 | v1.1

procedureMetadata

Req. Analysis: false

Req. Upload: false

Is Annotated: false

Description: hemolysis\_status

Options: Slight, Moderate, Marked, None,

---

## Blood collection experimenter ID IMPC\_CBC\_049\_001 | v1.1

procedureMetadata

Req. Analysis: false

Req. Upload: true

Is Annotated: false

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## Date equipment last calibrated IMPC\_CBC\_050\_001 | v1.2

procedureMetadata

Req. Analysis: false

Req. Upload: false

Is Annotated: false

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## Blood collection tubes IMPC\_CBC\_039\_001 | v1.1

procedureMetadata

**Req. Analysis:** false

**Req. Upload:** false

**Is Annotated:** false

**Options:** Sarstedt Li-Heparin gel tubes, Kabe Labortechnik Lithium heparin coated tubes, Kabe Labortechnik 1000ul Lithium Heparin, BD Microtainer Lithium Heparin/PST Gel Blood Tube, TERUMO CAPIJECT Lithium heparin coated tubes, Eppendorf 1.7ml, BD Microtainer Lithium Heparin Tube, Greiner MiniCollect Lithium Heparin 1ml,

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## **Date and time of sacrifice** IMPC\_CBC\_040\_001 | v1.1

procedureMetadata

**Req. Analysis:** false

**Req. Upload:** true

**Is Annotated:** false

## **Storage temperature from blood collection till measurement** IMPC\_CBC\_041\_001 | v1.3

procedureMetadata

**Req. Analysis:** true

**Req. Upload:** true

**Is Annotated:** false

**Unit Measured:** C

**Options:** 2, 18-22, 4, -80,

---

## **Blood analysis experimenter ID** IMPC\_CBC\_051\_001 | v1.0

procedureMetadata

Req. Analysis: false

Req. Upload: true

Is Annotated: false

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## Glycosilated hemoglobin A1c (HbA1c) IMPC\_CBC\_052\_001 | v1.3

simpleParameter

Req. Analysis: false

Req. Upload: false

Is Annotated: true

Unit Measured: %

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## Thyroxine IMPC\_CBC\_053\_001 | v1.2

simpleParameter

Req. Analysis: false

Req. Upload: false

Is Annotated: true

Unit Measured: ug/dl

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## Magnesium IMPC\_CBC\_054\_001 | v1.5

simpleParameter

Req. Analysis: false

Req. Upload: false

Is Annotated: true

Unit Measured: mg/dl

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**Difficult bleed** IMPC\_CBC\_055\_001 | v1.0

procedureMetadata

Req. Analysis: false      Req. Upload: false      Is Annotated: false

Options: Yes, No,

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**Sample type** IMPC\_CBC\_056\_001 | v1.0

procedureMetadata

Req. Analysis: true      Req. Upload: true      Is Annotated: false

Options: Serum, Plasma,

---

**Fasting** IMPC\_CBC\_057\_001 | v1.0

procedureMetadata

Req. Analysis: true      Req. Upload: true      Is Annotated: false

Options: No, Four hours before bleeding, Sixteen hours before bleeding,

---

**Cholesterol ratio** IMPC\_CBC\_058\_001 | v1.0

simpleParameter

**Req. Analysis:** false

**Req. Upload:** false

**Is Annotated:** true

**Description:** TC/HDLC ratio

**Derivation:** div('IMPC\_CBC\_015\_001', 'IMPC\_CBC\_016\_001')

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**Reagent manufacturer** IMPC\_CBC\_059\_001 | v1.0

procedureMetadata

**Req. Analysis:** true

**Req. Upload:** false

**Is Annotated:** false

**Options:** Beckman Coulter, Microgenics, Wako and Sekisui,

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