# **Clinical Chemistry IMPC\_CBC\_002**

## 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.
- 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 2001 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. Isofluorane.
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 sacrified.

### **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		
<b>Potassium</b> IMPC_CBC simpleParameter	=_002_001  v1.3	
Req. Analysis: false	Req. Upload: false	Is Annotated: true

Unit Measured: mmol/l

**Description:** potassium

#### 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		

### 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\_

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### 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

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

simpleParameter

Req. Analysis: false Req. Upload: true

Unit Measured: mg/dl

Description: calcium

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

simpleParameter

Req. Analysis: false	Req. Upload: true	Is Annotated: true
Unit Measured: mg/dl		
Description: phosphate		

#### **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

Unit Measured: U/I

Description: aspartate\_aminotransferase

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

simpleParameter

Req. Analysis: false	Req. Upload: true	Is Annotated: true	
Unit Measured: U/I			
Description: alanine_ar	ninotransferase		

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

simpleParameter

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

Unit Measured: U/I

Description: alkaline\_phosphatase

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

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

Unit Measured: mg/dl

Description: total\_cholesterol

#### 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		

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

simpleParameter

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

 Unit Measured: mg/dl
 Is Annotated: true
 Is Annotated: true

**Description:** triglycerides

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

Req. Analysis: false	Req. Upload: true	Is Annotated: true
Unit Measured: mg/dl		
Description: glucose		

#### 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\_

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

#### simpleParameter

Req. Analysis: false Req. Upload: false

Is Annotated: true

Unit Measured: umol/l

**Description:** fructosamine

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

Req. Analysis: false Req. Upload: false Is Annotated: true Unit Measured: U/I **Description:** lipase Lactate dehydrogenase IMPC\_CBC\_022\_001 | v1.2 simpleParameter Req. Analysis: false Req. Upload: false Is Annotated: true Unit Measured: U/I Description: lactate\_dehydrogenase \_\_\_\_\_ Alpha-amylase IMPC\_CBC\_023\_001 | v1.2 simpleParameter

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

 Unit Measured: U/I
 V/I

Description: alpha\_amylase

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

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Req. Analysis: false	Req. Upload: false	Is Annotated: true
Unit Measured: umol/l		
Description: uibc_unsaturated	d_iron_binding_capacity_	
LDL-cholesterol IMF simpleParameter	PC_CBC_025_001   v1.3	
Req. Analysis: false	Req. Upload: false	Is Annotated: true
Unit Measured: mg/dl		
Description: Idl_cholesterol		
Free fatty acids IMP simpleParameter	C_CBC_026_001   v1.4	
Req. Analysis: false	Req. Upload: false	Is Annotated: true
Unit Measured: mmol/l		
Description: free_fatty_acids		

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

simpleParameter

Req. Analysis: false	Req. Upload: false	Is Annotated: true
Unit Measured: mmol/l		
Description: glycerol		
Creatine kinase IMP simpleParameter	C_CBC_028_001   v1.2	
Req. Analysis: false	Req. Upload: false	Is Annotated: true
Unit Measured: U/I		
Description: creatine_kinase		
Uric acid IMPC_CBC_0 simpleParameter	29_001   v1.2	
Req. Analysis: false	Req. Upload: false	Is Annotated: true
Unit Measured: umol/l		
Description: uric_acid		

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

Req. Analysis: false	Req. Upload: false	Is Annotated: true
Unit Measured: ng/ml		
Description: ferritin		
Transferrin IMPC_CBC simpleParameter	C_031_001   v1.2	
Req. Analysis: false	Req. Upload: false	Is Annotated: true
Unit Measured: mg/dl		
Description: transferrin		
C-reactive protein simpleParameter	IMPC_CBC_032_001   v1.0	
Req. Analysis: false	Req. Upload: false	Is Annotated: true
Unit Measured: mg/l		
Description: c_reactive_prote	ein	

### 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

Pog Analysis: true	Pog Upload: true	Is Annotated: false
Req. Analysis: true	Req. Upload: true	is Annotateu. Taise

**Description:** equipment\_manufacturer

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

#### 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,

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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_u	sed_for_blood_collection	
Options: Gas anaesthesia	with Isofluorane,	
Injection narcosis with Keta	mine (100mg/kg)/Xylazine	(10mg/kg),
Injection narcosis with Keta	mine (100mg/kg)/ Xylazine	(10mg/kg )/Antipamezole (Antisedan,
1mg/kg),		
Injection narcosis with Keta	mine (110mg/kg)/Xylazine	(11mg/kg),
Injection narcosis with Keta	mine (110mg/kg)/Xylazine	(11mg/kg)/ Antipamezole (Antisedan,
1mg/kg),		
Injection narcosis with Tribr	omoethanol (Avertin),	
Injection narcosis with Sodi	um Pentobarbital (Pentoba	rb, 0.1ml),
Injection narcosis with Sodi	um Pentobarbital (Euthatal)	), No,
Injection narcosis with Keta	mine (137mg/kg)/Xylazine	(6.6mg/kg),
Injection narcosis with Sodi	um Pentobarbital (Somnop	entyl),

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

procedureMetadata

Req. Analysis: true Req.	Upload: true	Is Annotated: false
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**Description:** method\_of\_blood\_collection

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

## 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,	
Samples kept on ic CBC_042_001   v1.1 procedureMetadata	e between collection	on and analysis IMPC_
Req. Analysis: true	Req. Upload: true	Is Annotated: false
Description: samples_kept_o Options: Yes, No,	n_ice_between_collection_and	I_analysis
Sample status IMPC_ procedureMetadata	_CBC_043_001   v1.1	
Req. Analysis: false	Req. Upload: true	Is Annotated: false
Description: sample_status		
Options: Fresh, Frozen, Fresh	n 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

Reg. Analysis: false Reg. 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

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

procedureMetadata

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

#### 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,

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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

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

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Req. Analysis: false	Req. Upload: false	Is Annotated: true	
Unit Measured: %			
Thyroxine IMPC_CBC_ simpleParameter	_053_001   v1.2		
Req. Analysis: false	Req. Upload: false	Is Annotated: true	
Unit Measured: ug/dl			
Magnesium IMPC_CBC_054_001   v1.5			
simpleParameter	C_054_001   V1.5		
Req. Analysis: false	Req. Upload: false	Is Annotated: true	
Unit Measured: mg/dl			

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

procedureMetadata

Req. Analysis: false	Req. Upload: false	Is Annotated: false
Options: Yes, No,		