Clinical Blood Chemistry HAS_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

Sex: Females only

Age of animals: 28,53 and 80 weeks

Sexual dimorphism: Yes for some of the parameters.

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 a Sarstedt 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 on 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 performed as a non-fasting, terminal procedure, with some mice being used for subsequent gross pathology and other clinic-specific parameters 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.

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. 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 HAS_CBC_001_001 | v1.2

simpleParameter

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

Unit Measured: mmol/l

Description: sodium

Potassium HAS_CBC_002_001 | v1.4

simpleParameter

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

Unit Measured: mmol/l

Description: potassium

Chloride HAS_CBC_003_001 | v1.2

simpleParameter

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

Unit Measured: mmol/l

Description: chloride

Urea HAS_CBC_004_001 | v1.5
simpleParameter

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

Unit Measured: mmol/l

Description: urea

Creatinine HAS_CBC_005_001 | v1.4
simpleParameter

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

Unit Measured: umol/l

Description: creatinine_enzymatic_method_preferred_

Total protein HAS_CBC_006_001 | v1.2
simpleParameter

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

Unit Measured: g/l

Description: total_protein

Albumin HAS_CBC_007_001 | v1.2

simpleParameter

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

Unit Measured: g/l

Description: albumin

Total bilirubin HAS_CBC_008_001 | v1.3

simpleParameter

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

Unit Measured: umol/l

Description: total_bilirubin

Calcium HAS_CBC_009_001 | v1.4

simpleParameter

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

Unit Measured: mmol/l

Description: calcium

Inorganic phosphorous HAS_CBC_010_001 | v1.6

simpleParameter

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

Unit Measured: mmol/l

Description: phosphorus

Iron HAS_CBC_011_001 | v1.2

simpleParameter

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

Unit Measured: umol/l

Description: iron

Aspartate aminotransferase HAS_CBC_012_001 | v1.2

simpleParameter

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

Unit Measured: U/l

Description: aspartate_aminotransferase

Alanine aminotransferase HAS_CBC_013_001 | v1.2
simpleParameter

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

Unit Measured: U/l

Description: alanine_aminotransferase

Alkaline phosphatase HAS_CBC_014_001 | v1.2
simpleParameter

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

Unit Measured: U/l

Description: alkaline_phosphatase

Total cholesterol HAS_CBC_015_001 | v1.4
simpleParameter

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

Unit Measured: mmol/l

Description: total_cholesterol

HDL-cholesterol HAS_CBC_016_001 | v1.4
simpleParameter

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

Unit Measured: mmol/l

Description: hdl_cholesterol

Triglycerides HAS_CBC_017_001 | v1.4
simpleParameter

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

Unit Measured: mmol/l

Description: triglycerides

Glucose HAS_CBC_018_001 | v1.4
simpleParameter

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

Unit Measured: mmol/l

Description: glucose

LIH (Hemolysis Severity - available on AU analysers) HAS_C
BC_019_001 | v1.2

procedureMetadata

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

Description: lih_hemolysis_severity_available_on_au_analysers_

Options: N, /, +, ++, +++, ++++, +++++, ABN,

Fructosamine HAS_CBC_020_001 | v1.1
simpleParameter

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

Unit Measured: umol/l

Description: fructosamine

Lipase HAS_CBC_021_001 | v1.0
simpleParameter

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

Unit Measured: U/l

Description: lipase

Lactate dehydrogenase HAS_CBC_022_001 | v1.1
simpleParameter

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

Unit Measured: U/l

Description: lactate_dehydrogenase

Alpha-amylase HAS_CBC_023_001 | v1.1
simpleParameter

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

Unit Measured: U/l

Description: alpha_amylase

UIBC (unsaturated iron binding capacity) HAS_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_

LDL-cholesterol HAS_CBC_025_001 | v1.3
simpleParameter

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

Unit Measured: mmol/l

Description: ldl_cholesterol

Free fatty acids HAS_CBC_026_001 | v1.1
simpleParameter

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

Unit Measured: mmol/l

Description: free_fatty_acids

Glycerol HAS_CBC_027_001 | v1.2

simpleParameter

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

Unit Measured: umol/l

Description: glycerol

Creatine kinase HAS_CBC_028_001 | v1.1

simpleParameter

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

Unit Measured: U/l

Description: creatine_kinase

Uric acid HAS_CBC_029_001 | v1.1

simpleParameter

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

Unit Measured: umol/l

Description: uric_acid

Ferritin HAS_CBC_030_001 | v1.1

simpleParameter

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

Unit Measured: mg/ml

Description: ferritin

Transferrin HAS_CBC_031_001 | v1.0

simpleParameter

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

Unit Measured: mg/dl

Description: transferrin

C-reactive protein HAS_CBC_032_001 | v1.0

simpleParameter

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

Unit Measured: mg/l

Description: c_reactive_protein

Equipment ID HAS_CBC_033_001 | v1.0

procedureMetadata

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

Description: equipment_name

Equipment manufacturer HAS_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 HAS_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, AU680,

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Anesthesia used for blood collection HAS_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 Isofluorane,
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,

Method of blood collection HAS_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 HAS_CBC_038_001 | v1.0

procedureMetadata

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

Description: anticoagulant

Options: No, Lithium Heparin, Sodium Heparin, Heparin,

Samples kept on ice between collection and analysis HAS_CBC_039_001 | v1.0

procedureMetadata

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

Description: samples_kept_on_ice_between_collection_and_analysis

Options: Yes, No,

Sample status HAS_CBC_040_001 | v1.0

procedureMetadata

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

Description: sample_status

Options: Fresh, Frozen,

Plasma dilution HAS_CBC_041_001 | v1.0

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

Description: plasma_dilution

Date of procedure HAS_CBC_043_001 | v1.2

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

Description: date_and_time_of_blood_collection

Date of analysis HAS_CBC_044_001 | v1.2

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

Unit Measured: Date

Description: date_of_measurement

Blood collection PIL number HAS_CBC_046_001 | v1.1

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

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Date equipment last calibrated HAS_CBC_047_001 | v1.0

procedureMetadata

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

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Blood collection tubes HAS_CBC_048_001 | v1.0

procedureMetadata

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

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Blood analysis experimenter ID HAS_CBC_051_001 | v1.0

procedureMetadata

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

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Diluent HAS_CBC_042_001 | v1.0

procedureMetadata

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

Options: None, PBS, Saline, Water,

General comments HAS_CBC_049_001 | v1.2

simpleParameter

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

Description: general_comments_about_the_mouse

Barcode HAS_CBC_064_001 | v1.0

simpleParameter

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

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Procedural comments HAS_CBC_052_001 | v1.0

simpleParameter

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

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Body weight HAS_CBC_045_001 | v1.0

simpleParameter

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

Unit Measured: g

Description: body_weight

Ketone bodies HAS_CBC_050_001 | v1.3

simpleParameter

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

Unit Measured: mmol/l