



Montreal Neurological Institute and Hospital
McGill University

CBIG-02-011

PLASMA ISOLATION FROM WHOLE BLOOD

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1. REVISION HISTORY

Version	Summary of revisions	Effective Date
1.0	Initial	2-Oct-2020

2. SCOPE AND APPLICATION

This protocol is to be used for the *in vitro* isolation of human plasma from whole blood.

3. REFERENCE TO OTHER SOP OR DOCUMENTS

When adopting this SOP for local use, please reference *C-BIG Repository: CBIG-02-011 Plasma Isolation from Whole Blood*.

3.1 Reference to Other C-BIG SOPs or Documents

1. C-BIG Repository: CBIG-03-002 RPM Conversion

3.2 Reference to External SOPs or Documents

1. Experimental Therapeutics Program: ETP-P-0011 Plasma Isolation from Whole Blood
2. Article : Clark S, *et al.* Stability of Plasma Analytes after Delayed Separation of whole Blood: Implications for Epidemiological studies. *International Journal of Epidemiology*. 2003. 32:125-130

4. PERSONNEL QUALIFICATION AND RESPONSIBILITIES

To be read by all personnel who process human peripheral blood samples for plasma isolation. All personnel who read this SOP should sign the form found in the reading log binder.

5. ABBREVIATIONS AND DEFINITIONS

Abbreviation	Definition
C-BIG	Clinical Biological Imaging and Genetic Repository
Min	Minutes
mL	Milliliters
PTT	Purple/Lavender Top Tube (vacutainer)
QA	Quality Assurance
QC	Quality Control
RT	Room Temperature
SOP	Standard Operating Procedure
°C	Degree Celsius
µL	Microliters

6. MATERIALS AND EQUIPMENT

The materials and equipment listed below are recommendations only and may be substituted by alternative/equivalent products more suitable for site-specific task or procedure.

NOTE: All disposable items are sterile (gamma-irradiated) unless otherwise specified. All equipment, disposables or reagents can be substitutes with equivalent materials following evaluations and approval, unless specified otherwise.

NOTE: All sample contact materials must be suitable for RNA work (i.e., RNase-free). Use clean gloves at all times to prevent inadvertent RNase contamination during processing.

Material/Equipment	Material/Equipment (site specific)
15 mL polypropylene conical tubes	
Blood collection tube with K2 EDTA, 10 mL, purple/lavender top	BD Diagnostic; Cat # 366643
Cryovials, 1 mL, conical bottom	Nunc Biobanking and Cell Culture Cryogenic Tubes, Thermo Scientific; Cat # 366656
Transfer Pipets	
Centrifuge with swinging bucket	Eppendorf centrifuge 5810; Cat # 022625101, with rotor S-4-104; Cat # 5820759003
Micropipets, range 100 to 1000 µL	
Pipet tips, 1000 µL	
Pipet-aid	
Class II biological safety cabinet	
-80°C Freezer	

7. PROCEDURES

PRECAUTIONS: All biological samples derived from human source are considered to be biohazardous. Use appropriate precautions when working with such samples (i.e. personal protection equipment such as gloves, lab coat and safety glasses). All waste (samples and related contact materials) must be placed in marked biohazardous waste containers and disposed of under hospital guidelines.

NOTE: Best results are obtained when whole blood is processed promptly. When possible, processing should be done the same day as the blood draw.

7.1 Plasma Isolation from Whole Blood

1. Store PTT tube upright at RT and away from direct light for a minimum of 30 min to allow blood to sediment.

NOTE: The above step can be skipped should the sample is received by external courier service.

2. Centrifuge at 4°C for 12 min at 250Xg using a centrifuge with swinging buckets.
3. Quickly transfer supernatant (plasma) to a new 15 mL conical tube using a disposable transfer pipette.
4. Prepare aliquots of 250 µL into 1 mL conical cryovials.

5. Plasma samples can be frozen at -20°C for 2-3 days (temporary storage) and then transferred to a -80°C freezer for long-term storage. Alternatively, transfer plasma samples directly to -80°C.

7.2 Summary Table

Steps
1. Store PTT upright at RTT and away from light for 30 min.
2. Centrifuge at 4°C for 12 min at 250Xg
3. Transfer supernatant to new conical tube.
4. Prepare aliquots.
5. Store temporarily at -20°C for 2-3 days and then at -80°C for long term storage or transfer directly at -80°C.

8. QUALITY CONTROL / QUALITY ASSURANCE

All the equipment used should be monitored, cleaned and calibrated as by their specific SOPs.

Reagents with an expiry date should be monitored and used before this date. If used after expiry date, it should be recorded.

9. APPENDICES/FORMS

9.1 Appendix A – Sample processing form: Plasma