Laboratory Processing Chart
IMPAACT 2001

A Phase I/II Trial of the Pharmacokinetics, Tolerability, and Safety of Once-Weekly Rifapentine and Isoniazid in HIV-1-infected and HIV-1-uninfected Pregnant and Postpartum Women with Latent Tuberculosis Infection

Protocol Version: 1.0 with LoA #1-2 and CM # 1-3 **LPC Version Date:** 24 May 2016 **LPC Edited/Updated:** 14 Feb 2018

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Section 1: Protocol-Required Non-Standard Reagents and Supplies		
Evaluation	**Reagent or supply**	**Order information**
IGRA (QGIT)	QuantiFeron TB Gold(In Tube method) tubes	**Tuberculosis and Control Antigen Blood Collection Tubes** **Catalog Number: 0590 0301** 1. Nil Control (Grey cap with white ring) 100 x tubes 2. TB Antigen (Red cap with white ring) 100 x tubes 3. Mitogen Control (Purple cap with white ring) 100 x tubes **High Altitude (HA) Blood Collection Tubes *(for use above 3,350 feet)*** **Catalog Number: T0590 0505 (refer to Section 5)** 4. Nil Control (Grey cap with yellow ring) 100 x tubes 5. TB Antigen (Red cap with yellow ring) 100 x tubes 6. Mitogen Control (Purple cap with yellow ring) 100 x tubes <u>Tubes are also available in other configurations:</u> 100 x Nil Control, 100 x TB Antigen tubes **(Catalog Number: 0590 0201)** 100 x Mitogen Control tubes **(Catalog No. 0593 0201)**
	QuantiFeron TB Gold(In Tube method) ELISA test kit	ELISA Components – **Catalog Number: 0594 0201**
Intensive and Sparse PK	EDTA (Spray-dried) vacutainer tubes	Whole Blood Tube; Size: 13x75 mm x 2.0 mL K2 EDTA (spray dried) Plastic; Label: Paper; Closure: Hemogard™; Color: Lavender (**Catalog Number: 367842**) or Lavender (**Catalog Number: 367841**)

Appendix I-A: Maternal Schedule of Evaluations

Evaluation	SCR (*up to 2 weeks prior to entry*)	Entry/Intensive PK Sampling (*Days 0-3*)	Weekly (7 ± 2 days apart) (*weeks 1-11*)	Postpartum Visit *(Within 3 days of delivery; if study drug regimen completed prior to delivery only)*	Monthly (Every 4 weeks ±2 weeks) (*until 24 weeks after delivery*)	Study Exit/ Early Discontinuation
Gene Xpert, shielded chest x-ray or sputum microscopy (if needed)	X					
IGRA (3-4mL) (if needed and not available per medical record)	3-**4**mL					
Complete blood count (CBC)	3mL		3mL (every 4 weeks)	3mL (if clinically indicated)	3mL	
Liver Function tests	3mL		3mL (every 4 weeks)		3mL	
HIV-1 test (confirmatory tests as needed)	6mL					
CD4 count (HIV-1-infected only)		3mL				
Coagulation profile	5mL		5mL (at one visit at >/= 34 weeks gestational age only)	5mL (if clinically indicated)	5mL (if clinically indicated)	
HBsAG, HBsAb, HCV Ab		6mL				
Intensive PK sample collection		22mL				
Sparse PK sample collection			8mL (last dose)			
Plasma sample collection (breast milk PK eligible only)			2-4 mL (first and second week postpartum)			
Breast milk PK sample collection (if eligible)			X (first and second week postpartum and last dose)			
TOTAL BLOOD	17-21mL	28-31mL	11-23mL	8mL	6-11mL	

Note: NIH recommendations for maximum pediatric and adult blood draw volumes will be followed in this study. For women, the volume of blood drawn shall not exceed 10.5 mL/kg or 550 mL, whichever is smaller, over any eight week period.
The priority order of sample collections will be as follows: samples needed for clinical safety assessments/AEs will be collected first (specific tests as determined by investigator), followed by samples needed for PK analysis.

Prioritization of sample collection of blood tubes and processing for insufficient draws, by visit		
Visit	**Tube type**	**Purpose**
Screening (up to 2 weeks prior to entry)	3mL EDTA	Complete Blood Count
	3mLRed top or SST	Liver Function Tests
	5mL Blue top Sodium Citrate	Coagulation profile
	3-4mL QGIT tubes (Nil, Mitogen and TB antigen tube(s) each)	IGRA
	6ml Red top or SST or EDTA	HIV-1 Test (confirmatory test as needed)
Entry/Intensive PK sampling (Days 0-3)	2mL **K2** EDTA (**spray dried)**	Intensive PK
	3mL EDTA	CD4 count
	6ml Redtop or SST	HbsAg,HbsAb,HCV Ab
Weekly Visits (Weeks 1-11)	3mL EDTA	Complete Blood Count
	3mL Red top or SST	Liver Function Tests
	5mL Blue top Sodium Citrate	Coagulation profile (Prothrombin Time; INR if available at the site, as per Sections 8.1.7 and 8.1.8)
	2mL **K2** EDTA **(spray dried)**	Sparse PK
	2mL **K2** EDTA (**spray dried**)	Plasma (PL1) for PK(1.2 ml minimum blood required)
Postpartum Visit (Within 3 days of delivery)	3mL EDTA	Complete Blood Count
	5mL Blue top Sodium Citrate	Coagulation profile (Prothrombin Time; INR if available at the site, as per Sections 8.1.7 and 8.1.8)
Monthly Visits (Every 4 weeks, until 24 weeks after delivery)	3mL EDTA	Complete Blood Count
	3mL Red top or SST	Liver Function Tests
	5mL Blue top Sodium citrate	Coagulation profile (Prothrombin Time; INR if available at the site, as per Sections 8.1.7 and 8.1.8)

Section 2: Safety/Clinical Laboratory Evaluations					
Evaluation	**Tube Type**	**Tests**		**DMC Test Code**	**CRF #**
Interferon Gamma Release Assay (IGRA)	Nil, TB antigen, Mitogen	Quantiferon Gold In-tube Test(QGIT)		MTBIGRA	TBW0034
Complete Blood Count (CBC)	EDTA	White blood count, Hemoglobin and Platelets		N/A	PE6813
Liver Function Test	SST or NON	Albumin, AST,ALT, and Total bilirubin		N/A	PE6818
HIV-1 test (confirmatory test as needed)	SST or NON or EDTA	*As per protocol section 4.1.4*		N/A	
CD4 count (HIV-1 infected only)	EDTA	CD4 cell counts and percentages	Dual platform labs only must also have a WBC and diff.	CD4CD8	LBW0054
Coagulation profile	Sodium citrate	Prothrombin time (PT), and INR if INR is available at the site, as per Sections 8.1.7 and 8.1.8.		N/A	PE6813
HbsAG,HbsAb,HCV Ab	SST or NON	Blood will be collected at Entry visit for later testing of HbsAG, HbsAb, HCV Ab		HEPSER	F3008 & SR0010

Section 3: Specimen Processing. Refer to Section 4 for tube types and collection volumes					
Evaluation	**Tube Type**	**Special Collection Notes**	**CRF # DMC Test Code**	**Processing**	**Shipping**
Intensive PK sample collection (day 0 and 72 hours following) Collection Timepoints: prior to first dose(t0), then 0.5, 1, 2, 4, 5, 8, 12, 24, 48, and 72hrs after the dose	**K2** EDTA **(spray dried)** (Polypropylene tube recommended)	Process within one hour of collection.Place plasma aliquots in crushed ice as INH is sensitive to temp.Storage must be within 1 hr at -70^{0}C.	PKW0392 PKINT	*Process within one hour of collection.* Spin blood at 1500 xg for 10min. Remove plasma and prepare 2 equal aliquots. Freeze at approx. -70°C LDMS spec. code: BLD/EDT/PL1 *Min: 2 x 0.2ML*	Ship to University of Cape Town, South Africa upon the protocol team's request.
Sparse PK sample collection at 1, 4, 24, and 48 hrs after the dose	**K2** EDTA **(spray dried)** (Polypropylene tube recommended)	Process within one hour of collection.	PKW0392 PKRAN	*Process within one hour of collection.* *Spin blood at 1500xg for 10min.* Remove plasma and prepare 2 equal aliquots. Freeze at approx.-70°C LDMS spec. code: BLD/EDT/PL1 *Min: 2 x 0.2ML*	Batch ship to University of Cape Town, South Africa at end of the study or may be along with intensive PK samples shipped at that time.
Plasma sample collection (Breast milk PK eligible only) At 1st week after delivery-3 h after drug dose At 2nd week after delivery-6h after drug dose	**K2** EDTA **(spray dried)** (Polypropylene tube recommended)	Process within one hour of collection.	PKW0394 PKRAN	*Spin blood at 1500xg for 10min.* Remove plasma and prepare 2 equal aliquots. <u>Store two plasma aliquots in separate cryoboxes.</u> Freeze at -70°C LDMS spec. code: BLD/EDT/PL1 *Min: 2 x 0.2ML*	Batch ship to University of Cape Town, South Africa at end of the study
Breast milk PK collection At 1st week after delivery-3 h after drug dose At 2nd week after delivery-6h after drug dose Last dose visit-24h after drug dose	15 mL sterile polypropylene conical tube or sterile plastic container	Place breast milk aliquots in crushed ice as INH is sensitive to temp.Storage must be within 1 hr at -70^{0}C.	PKW0394 PKRAN	Collect 5 mL of breast milk either from one or both breasts. Refer IMPAACT 2001 MOP for details. Prepare 2 x 1.5 mL aliquots or more, stored in cryovials and keep in crushed ice until freeze at -70^{0}C. LDMS spec. code: BMK/NON/BMW	Batch ship to University of Cape Town, South Africa at end of the study.

Section 3: Specimen Processing. Refer to Section 4 for tube types and collection volumes					
Evaluation	**Tube Type**	**Special Collection Notes**	**CRF # DMC Test Code**	**Processing**	**Shipping**
Cord blood PK collection (Optional) Only from women who have received study drug dose within 72 h prior to delivery *Collect 2 mL of Cord Blood if available at sites with capacity (e.g. during normal business hours)*	**K2** EDTA **(spray dried)** (Polypropylene tube recommended)	Process within one hour of collection.Place plasma aliquots in crushed ice as INH is sensitive to temp.Storage must be within 1 hr at -70^{0}C	PKW0393 PKRAN	*Process within one hour of collection.* Spin blood at 1500 xg for 10min. Remove plasma and prepare 2 equal aliquots. Freeze at approx. -70°C LDMS spec. code: CRD/EDT/PL1 Min: 2 x 0.2ML See IMPAACT 2001 MOP for instructions on collection, processing, and storage of Cord Blood.	Batch ship to University of Cape Town, South Africa at end of the study.
HIV-1 test (confirmatory test as needed)	SST or NON or EDTA	Invert tube 8-10 times gently. Send to processing lab ambient.		Process within 6 hours. For HIV RNA PCR: Spin blood at 800xg for 10 min.Remove plasma. Re-spin plasma at 800xg for 10 min. Freeze 2x 1.8mL aliquots at -70°C or colder. LDMS spec. code: BLD/EDT/PL2	Refer to Section 4.1.4 **For U.S. sites**: Sample #2 must be tested in a CAP/CLIA approved laboratory. **For Non-U.S.sites**: Sample #2 must be tested in a lab that operates according to GCLP and participates in an EQA program.

Section 4: Evaluations by Visit						
Screening: Must be completed up to 2 weeks prior to study entry						
LDMS Visit Code = 0/SCR						
Evaluation	**Patient Subset**	**Specimen**	**CRF**	**Aliquots**	**LDMS Code**	**Special Notes**
IGRA (Quantiferon-TB Gold In Tube Test- QGIT)		Nil, TB antigen and Mitogen 3mL	TBW0034	Two equal aliquots from each tube	BLD/QTF/PL1/ATG BLD/QTF/PL1/MIT BLD/QTF/PL1/NIL	If not available per medical records
Complete Blood Count (CBC)		EDTA 3mL	PE6813	N/A	N/A	Laboratory values should be obtained within **14** days prior to enrollment
Liver Function Tests (albumin, AST ALT, and total bilirubin)		SST or NON 3mL	PE6818	N/A	N/A	Laboratory values should be obtained within **14** days prior to enrollment
HIV-1 test		SST or NON or EDTA 6mL		N/A	N/A	Confirmatory test as needed
Coagulation profile		Sodium citrate 5mL	PE6813	N/A	N/A	**For laboratories who are not performing coagulation profile testing locally, the maternal specimens collected at screening should be processed and frozen according to the MOP. These specimens should be batch shipped to the testing laboratory *at least every three months* beginning at the time of first enrollment at the site or upon request by the protocol team.**

Entry/ Intensive PK Visit					
Must be completed within Days 0-3 LDMS Visit Code= 0/Ent					
Evaluation	**Specimen**	**CRF**	**Aliquots**	**LDMS Code**	**Special Notes**
Intensive PK	2mL **K2** EDTA **(spray dried)** at each timepoint Collection Timepoints: 0, 0.5, 1, 2, 4, 5, 8, 12, 24, 48 and 72 hrs post-dose.	PKW0392	Prepare 2 equal aliquots.	BLD/ EDT/PL1	Must be initiated on the same day as the first dose of study regimen and conducted over the course of 72 hours according to the collection timepoints. RPT PK will be performed on all intensive PK samples, after which t0 and 2h or 4h samples will be stored for INH PK testing.
CD4 Count (HIV-1 infected only)	EDTA 3mL	LBW0054	N/A	N/A	
HBsAg, HBsAb, HCV Ab	SST or NON 6mL	F3008	Prepare 2 equal serum aliquots or as per site requirement	BLD/SST or NON/SER	Blood will be collected for later testing of HBsAG, HBsAb, HCV Ab as determined by protocol team.

Weekly (7+/-2 days apart) **(Weeks 1-11)**					
LDMS Visit Code = X/WK					
Evaluation	**Specimen**	**CRF**	**Aliquots**	**LDMS Code**	**Special Notes**
Sparse PK	2m**L K2** EDTA **(spray dried)** at each timepoint	PKW0392	Prepare 2 equal aliquots	BLD/EDT/PL1	Sample collection will be conducted on the visit of women's last dose of study drug regimen at 1h, 4h, 24h and 48h post dose
Plasma sample collection (Breast milk PK participants only)	**K2** EDTA **(spray dried)** (Polypropylene tube recommended)	PKW0394	Prepare 2 equal aliquots	BLD/EDT/PL1	At first week (3h post dose) and second week (6h post dose) postpartum if participant is taking study drug regimen
Complete Blood Count (CBC)	EDTA 3mL	PE6813	N/A	N/A	Every four weeks
Liver Function Test (albumin, AST, ALT, and total bilirubin)	SST or NON 3mL	PE6818	N/A	N/A	Every four weeks
Coagulation profiles	Sodium citrate 5mL	PE6813	N/A	N/A	Collected at one visit for women who are ≥34 weeks gestational age only **For laboratories who are not performing coagulation profile testing locally, ship samples <u>in real-time</u>.**

Weekly (7+/-2 days apart) (Weeks 1-11)					
LDMS Visit Code = X/WK					
Evaluation	**Specimen**	**CRF**	**Aliquots**	**LDMS Code**	**Special Notes**
Breast milk PK	15 mL sterile polypropylene conical tube or sterile plastic container	PKW0394	Prepare 2 x 1.5 ml aliquots or more	BMK/NON/BMW	At first and second week postpartum and on the visit of last dose of study drug regimen

Postpartum Visit **(Within 3 days of delivery; if study drug regimen completed prior to delivery only)**					
LDMS Visit Code = X/WK					
Evaluation	**Specimen**	**CRF**	**Aliquots**	**LDMS Code**	**Special Notes**
Complete Blood Count (CBC)	EDTA 3mL	PE6813	N/A	N/A	If clinically indicated
Coagulation profiles	Sodium citrate 5mL	PE6813	N/A	N/A	If clinically indicated **For laboratories who are not performing coagulation profile testing locally, ship samples in real-time.**
Maternal Monthly (Every 4 weeks +/- 2 weeks) **(Until 24 weeks after delivery)**					
LDMS Visit Code = X/Mo					
Evaluation	**Specimen**	**CRF**	**Aliquots**	**LDMS Code**	**Special Notes**
Complete Blood Count (CBC)	EDTA 3mL	PE6813	N/A	N/A	
Liver Function Test (albumin, AST, ALT, and total bilirubin)	SST or NON 3mL	PE6818	N/A	N/A	
Coagulation profiles	Sodium citrate 5mL	PE6813	N/A	N/A	If clinically indicated **For laboratories who are not performing coagulation profile testing locally, ship samples in real-time.**

Appendix I-B: Infant Schedule of Evaluations

Evaluation	Newborn Visit (Within 3 days of life)	Monthly (Every 4 weeks+/- 2 weeks) (Until 24 weeks after delivery)
Single PK sample collection (if eligible)	2mL	
Coagulation profile (if eligible)	2mL	2mL (if clinically indicated)
Complete blood count (if clinically indicated)		1mL
Liver Function tests (if clinically indicated)		2mL
Cord blood (optional if eligible, during mother's delivery)	X	
TOTAL BLOOD	**2-4mL**	**1-5mL**

For infants, the volume of blood drawn at any study visit should not exceed 5 mL/kg in a single day and 9.5 mL/kg over any eight-week period.

Newborn Visit **(Within 3 days of life)**					
LDMS Visit Code = XX/Bth					
Evaluation	**Specimen**	**CRF**	**Aliquots**	**LDMS Code**	**Special Notes**
Single PK sample collection (if eligible)	**K2** EDTA**(spray dried)** (Polypropylene tube recommended)	PKW0393	Prepare 2 equal plasma aliquots	BLD/EDT/PL1	Single PK sample will be obtained within the first 3 days of life. Only collected if mother's most recent dose was taken within 72 hours of infants's blood collection.
Coagulation profiles (if eligible)	Sodium citrate 2mL	PE6813	N/A	N/A	Only if the mother is on the study drug regimen. **For laboratories who are not performing coagulation profile testing locally, the specimens should be processed and frozen according to the MOP. Specimens should be batch shipped to the testing laboratory along with the maternal specimens for coagulation profile collected at the maternal screening visit *at least every three months* or upon request by the protocol team.**
Cord blood (optional if eligible, during mother's delivery)	**K2** EDTA**(spray dried)**	PKW0393	Prepare 2 equal plasma aliquots	CRD/EDT/PL1	Eligibility for cord blood PK sampling must be confirmed prior to collection.
Infant Monthly **(Every 4 weeks +/- 2 weeks)** **(Until 24 weeks after delivery)**					
LDMS Visit Code = XX/Mo					
Evaluation	**Specimen**	**CRF**	**Aliquots**	**LDMS Code**	**Special Notes**
Coagulation profiles	Sodium citrate 2mL	PE6813	N/A	N/A	if clinically indicated **For laboratories who are not performing coagulaiotn profile testing locally, ship samples <u>in real-time</u>.**

Complete blood count (if clinically indicated)	EDTA 1mL	PE6813	N/A	N/A	If indicated based on symptoms
Liver Function tests (includes albumin, AST, ALT, and total bilirubin, if clinically indicated)	SST or NON 2mL	PE6818	N/A	N/A	If indicated based on symptoms

Section 5: Helpful Links and Shipping Addresses

ACTG/IMPAACT Laboratory Manual, Shipping Information and other useful information: http://www.hanc.info/labs/labresources/Pages/informationActgImpaactLabs.aspx

Intensive and Sparse PK, plasma storage, breast milk and cord blood shipment University of Cape Town

Attention: Jennifer Norman/Shameema Witbooi
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Observatory
7925, Cape Town, South Africa.
Telephone: +27 21 404 7695
Email Jennifer.Norman@uct.ac.za
LDMS number 499.

Specimen Repository Facility – NIAID Sites

Biomedical Research Institute
Biomedical Research Institute
9410 Key West Avenue, First Floor
Rockville, M D 20850
Phone : (301) 881-7636
Fax : (301) 770-9811
Email : brirepository@afbr-bri.com
LDMS lab code: 999

Specimen Repository Facility – NICHD Sites

Fisher Bioservices
625 Lofstrand Lane
Rockville, MD 20850 USA
Attention: Maria Wolff
Phone (301) 340-1620
Fax (301) 838-9753
Email maria.wolff@thermofisher.com
LDMS lab code: 243

Section 6: Revision History			
Protocol Version	LPC Change Date	Page(s)	Description
1.0	07 Sept 2016	6,9	Added 'special note' for coagulation profile at screening visit.
1.0	06 Nov 2016	4	Revised blood volume for IGRA test ie. 3- 4 ml and total blood volume to 21ml
		5,6	Added 'Prothrombin Time; INR if available at the site, as per Sections 8.1.7 and 8.1.8 in Coagulation profile
		9	Revised Laboratories values window to 14 days at screening visit
1.0	12 Jan 2017	All relevant pages	Updated K2 EDTA(spray dried) tubes for K3 EDTA tubes
1.0	**24 May 2017**	**14**	**Added note for infant coagulation profile at Newborn Visit**
1.0	**14 Feb 2018**	**All relevant pages**	**Added Special Note to indicate when real-time shipping of coagulation profile should occur for laboratories not doing local testing. Coagulation profile samples at monthly visits revised to "if clinically indicated" in Special Notes.**