STANDARD OPERATING PROCEDURE								
A Ragon Institute of MGH, MIT and Harvard	Ragon Institute Protocol f HLA/KIR t	Ü	SOP#	PL_	PL_SOP-004			
	Originated by:	Ildiko Toth	Date:		25 April 17			
	Laboratory:	Processing Laboratory	Pages:		1 of 2			
	Approved by:	Alicja Trocha						

## I. PURPOSE:

The purpose of this procedure is to establish and outline the process for sending DNA samples for HLA/KIR typing to Carrington lab.

## II. PROCEDURE:

- 1. On the first working day of every month the Manager of the PL sends out an email asking for DNA samples to be sent off for HLA/KIR typing to the Carrington Lab. The e-mail contains the latest template to be used.
- 2. The researchers have 5 working days to complete the template-manifest and sending it back to the Manager.
- 3. The other option is to save the red blood fraction after the Ficoll step of blood processing, treat it with Red Blood Cell Lysis buffer, followed by centrifugation to remove the lysed RBC fraction.
- 4. After adding 5.0 ml of Cell Lysis Buffer to the granulocytes fraction the sample can be brought to the PL for further DNA isolation by members of the PL. Only 5.0 ml lysate in a 50.0 ml conical that labeled correctly with PID and isolation date will be accepted.
- 5. The manifest will contain the following pieces of information:
  Ragon ID (RAGN191447), ethnicity (Hispanic, non-Hispanic), race (white, black....), gender (male, female..), sample drawn date (11/12/2016), 260/280 OD (best is 1.8) concentration (50 ug/ul), DNA volume (100 ul), DNA amount (5 ug), Principal Investigator (Bruce Walker), requestor (Ildiko Toth), funds (224134), HLA class I., II, KIR, cohort (chronic HIV), protocol number (2010P002463), source of DNA (granulocytes), is this resubmission (no). The template has drop down menus and is very easy to use.
- 6. Only completed manifests will be accepted within the 5 working days.
- 7. The ideal concentration of DNA is 50 ng/ul, 100 ul with purity of 260/280=1.80. <50 ng/ul or >100 ng/ul is not acceptable. In general 2-3 ug is sufficient for HLA typing but if KIR typing also needed 5 ug is the best.
- 8. Only samples collected under an already approved protocol will be accepted. The Regulatory Manager will work with the researchers to make the necessary changes.
- 9. If the sample has no Ragon ID, yet, the CID Team will assign one based on the provided information.
- 10. The Manager forwards the completed manifest to the CIDB Team, the Regulatory Manager and the Business Person for quality control.
- 11. The Business Person sends the payment to the Carrington Lab.
- 12. The manager sends the list to the Carrington Lab and will receive labels with the Ragon ID and also the Carrington ID.
- 13. The DNA is transferred into a special bottom-barcoded vial, the labels attached and ready to ship.
- 14. When the payment was received the Manager sends the shipment to the Carrington Lab by FedEx.
- 15. It takes about three months to generate the HLA I, II and KIR results that are sent to the Ragon Institute CIDB Team.

STANDARD OPERATING PROCEDURE								
A Ragon Institute of MGH, MIT and Harvard	Ragon Institute Protocol for sending DNA for HLA/KIR typing		SOP#	PL_SOP-004				
	Originated by:	Ildiko Toth	Date:		25 April 17			
	Laboratory:	Processing Laboratory	Pages:		2 of 2			
	Approved by:	Alicja Trocha						

## III. REFERENCES/ ADDITIONS/ NOTES:

N/A