Central Laboratory Data

Data generated at the central laboratory were transmitted electronically to NERI; there were no paper forms. Therefore, the documentation files (listed in Table 1) for each Laboratory assay are similar to the Clinical Center Data Collection Forms Documentation except that there are no Forms or QxQ's.

One of the primary uses of central laboratory data was to investigate short to medium term changes in various laboratory values after transfusion. For some laboratory values, long term changes were also of interest. Each test was performed on a designated subset of blood draws. For each type of laboratory result, it would have been ideal for all samples from a given patient to be tested in one laboratory run, so that there would be no run-to-run variability in a given patient's results. However, for many laboratory values this would not have allowed testing to proceed until the end of the patient's time on study. Furthermore, for certain tests, samples from a single patient could exceed the maximum size of a test batch. To allow testing to proceed in a timely fashion over the course of the study, but ensure that "before and after" samples for the most important changes were run in the same laboratory batch, each testing protocol grouped related samples together. These protocols differed from test to test, and are shown in Table 2. For example, for HIV-RNA PCR on a given patient, the pre-transfusion sample from the T1 transfusion episode and the four weekly post-transfusion samples from that episode would all be tested in one laboratory run. The pre-transfusion and post-transfusion samples from the T2 transfusion would also be tested in one laboratory run, but possibly not the same run as the T1 samples. Quarterly samples from the first 8 quarterly visits would be tested together, and the rest of the quarterly samples would also be tested together.

Table 1. Central Laboratory Assays: Codebooks and Data Files.

Assays	Codebook File	Data File[1]
Plasma HIV RNA PCR	CL01.DOC	CL01DATA
Total lymphocyte count, $CD3^+$ count, $CD3^+CD4^+$ count and percent	CL02.DOC	CL02DATA
$CD3^+CD8^+$ count and percent	CL03.DOC	CL03DATA
CD38% (percent of $CD8^+$ cells)	CL04.DOC	CL04DATA
HLA-DR% (percent of $CD8^+$ cells)[2]	CL05.DOC	CL05DATA
Beta2 Microglobulin	CL06.DOC	CL06DATA
Tumor Necrosis Factor alpha (TNFa)	CL07.DOC	CL07DATA
Tumor Necrosis Factor Receptor type II (TNFrII)	CL08.DOC	CL08DATA
Interleukin 6 (IL-6)	CL09.DOC	CL09DATA
Donor WBC Survival	CL10.DOC	CL10DATA
Post-Filtration Residual WBC	CL11.DOC	CL11DATA
HIV-1 EIA	CL12.DOC	CL12DATA
CMV EIA (recipient)	CL13.DOC	CL13DATA
Qualitative Plasma CMV DNA PCR	CL14.DOC	CL14DATA
bDNA CMV[3]	not included	
EPO EIA	CL16.DOC	CL16DATA
Quantitative Plasma CMV DNA PCR	CL17.DOC	CL17DATA
CMV EIA (donor)	CL18.DOC	CL18DATA

[1] Associated data file, after expansion of SAS Transport File.
[2] HLA-DR assay discontinued after 04/23/98. Only partial data available.
[3] bDNA CMV assay discontinued very early during the study. Data not included.

Table 2: Testing Protocols for VATS Central Laboratory Testing.

Data file	Test	Test Groups (samples batched together)	Designated type of sample
CL01DATA	HIV-RT PCR	T1PT, T107, T114, T121, T128 T2PT, T207, T214, T221, T228 QU03 – QU24 QU27 - QU42	ACD (yellow top) plasma
CL02DATA	CD4	T1PT, T107, T128, QU 03 T2PT, T207, T228 QU06 – QU24 QU27 - QU42	EDTA (lavender top) whole blood with DMSO
CL03DATA	CD8	T1PT, T107, T128, QU 03 T2PT, T207, T228	EDTA (lavender top) whole blood with DMSO
CL04DATA	CD38	T1PT, T107, T128, QU 03 T2PT, T207, T228	EDTA (lavender top) whole blood with DMSO
CL05DATA	HLA-DR	T1PT, T107, T128, QU 03 T2PT, T207, T228	EDTA (lavender top) whole blood with DMSO
CL06DATA	Beta-2	T1PT, T107 T2PT, T207	Clot (red top) serum
CL07DATA	TNF alpha EIA	T1PT, T107 T2PT, T207	EDTA (lavender top) plasma
CL08DATA	TNFrII EIA	T1PT, T107 T2PT, T207	Clot (red top) serum
CL09DATA	IL-6	T1PT, T107 T2PT, T207	EDTA (lavender top) plasma
CL10DATA	Donor WBC survival (female patients)	T1PT, T107, T114, T121, T128 T2PT, T207, T214, T221, T228 QU03 QU06 – QU39 for selected patients	EDTA (lavender top) whole blood
CL11DATA	Residual WBC (leukoreduced donor units)	Individual donor units	Post-leukoreduction aliquot
CL12DATA	HIV-1 EIA	T1PT	Clot (red top) serum
CL13DATA	CMV EIA (recipients)	T1PT	Clot (red top) serum
CL14DATA	CMV Qual. PCR	T1PT, T107, T114, T121, T128 T2PT, T207, T214, T221, T228 QU03 – QU24	ACD (yellow top) plasma

(continued on next page)

Table 2: Testing Protocols for VATS Central Laboratory Testing (continued)

Data file	Test	Test Groups (samples batched together)	Designated type of sample
CL16DATA	EPO EIA	T1PT	Clot (red top) serum
CL17DATA	CMV Quant. PCR	T1PT, T107, T114, T121, T128 T2PT, T207, T214, T221, T228 QU03 – QU24 (only sets with at least one qualitative positive in CL14DATA were tested)	ACD (yellow top) plasma
CL18DATA	CMV EIA (donor units)	Individual donor RBC units	Segment

Table 3. Assays used at Central Laboratory

Measurement	Assay	References
HIV-1 RNA PCR	Amplicor HIV Monitor assay - Reverse transcriptase polymerase chain reaction (PCR) assay with lower limit of quantification of 200 copies/mL (Roche Molecular Systems, Branchburg, NJ).	
Total lymphocyte count and lymphocyte subsets (CD3+CD4+, CD3+CD8+, proportion of CD8+ cells expressing CD38+, proportion of CD8+ cells expressing HLA-DR+)	Flow cytometry on thawed whole blood samples that had been cryopreserved in 10% dimethylsulfoxide using directly conjugated monoclonal antibodies (Becton Dickinson Immunocytometry, San Jose, CA)	Fiebig et al, 1997; Lee et al, 1998
Beta2 Microglobulin	Elisa (Coulter, Hialeah, FL; R&D Systems, Minneapolis, MN)	
Tumor necrosis factor alpha (TNFa)	Elisa (R&D Systems, Minneapolis, MN)	
TNF type II receptor (TNFrII)	Elisa (R&D Systems, Minneapolis, MN)	
Interleukin-6 (IL-6)	Elisa (R&D Systems, Minneapolis, MN)	
Donor WBC Survival	Quantitative PCR of Y chromosome sequence	Lee et al, 1999
Residual WBC	Quantitative PCR of a generic HLA DQ-alpha sequence	Lee et al, 1994; Lee et al, 1998
HIV-1 Antibody	EIA (Abbott)	
CMV antibody	EIA (Abbott)	
CMV Qualitative DNA PCR	AMPLICOR CMV Monitor Assay. (Roche Molecular Systems, Alameda, CA)	Long et al, 1998
CMV Quantitative DNA PCR	AMPLICOR CMV Monitor Assay with lower quantification limit of 400 copies/mL. (Roche Molecular Systems, Alameda, CA)	Pellegrin et al, 1999
EPO	EIA	