DEPARTMENT OF HEALTH AND HUMAN SERVICES

Centers for Disease Control and Prevention Model Performance Evaluation Program

T-Lymphocyte Immunophenotyping ($CD4^+$ T-Cell Determinations)

Report of Results for the Performance Evaluation Survey Conducted in October 2002

CENTERS FOR DISEASE™
CONTROL AND PREVENTION

PUBLIC HEALTH PRACTICE PROGRAM OFFICE
DIVISION OF LABORATORY SYSTEMS
ATLANTA, GEORGIA

Analysis of the October 2002 T-Lymphocyte Immunophenotyping Results ($CD4^+$ T-cell Determinations) Provided by Participant Laboratories in the Centers for Disease Control and Prevention (CDC) Model Performance Evaluation Program

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Table of Contents

Tables

Figures

Centers for Disease Control and Prevention (CDC)
Model Performance Evaluation Program for $CD4^+$ T-Cell Determinations

Table 1. Donor Identification for October 2002 Shipment Specimens

Panel Letter	Participant Laboratory Vial Label	CDC Donor Number	Donor Information (HIV-1* status)
A	A1	05	HIV-1 Antibody-Positive
	A2, A3	02	HIV-1 Antibody-Positive
	A4	01	HIV-1 Antibody-Negative
	A5	03	HIV-1 Antibody-Negative
B	B1, B3	04	HIV-1 Antibody-Positive
	B2	01	HIV-1 Antibody-Negative
	B4	05	HIV-1 Antibody-Positive
	B5	03	HIV-1 Antibody-Negative
C	C1, C2	07	HIV-1 Antibody-Positive
	C3	09	HIV-1 Antibody-Negative
	C4	10	HIV-1 Antibody-Positive
	C5	08	HIV-1 Antibody-Negative
D	D1	10	HIV-1 Antibody-Positive
	D2	08	HIV-1 Antibody-Negative
	D3, D5	06	HIV-1 Antibody-Positive
	D4	09	HIV-1 Antibody-Negative

* Human immunodeficiency virus type 1

Analysis of the October 2002 Performance Evaluation Testing Results for $CD4^+$ T-Cell Determination Program Reported to the Centers for Disease Control and Prevention by Participating Laboratories

Introduction

This report analyzes testing results reported by laboratories participating in the Centers for Disease Control and Prevention (CDC) Model Performance Evaluation Program (MPEP) after they tested the $CD4^+$ T-cell determination ($CD4^+$T-cell) performance evaluation specimens sent on October 8 and October 15, 2002. Of the 284 laboratories receiving specimen panels, 274 (96.5%) reported testing results. Of the 10 nonreporting laboratories, one laboratory was unable to report results due to inadvertently storing the specimens in the refrigerator upon arrival; two had discontinued testing, and seven provided no explanation.

Materials and Methods

Each laboratory received a total of five whole blood specimens collected in K_3EDTA, three HIV-1 antibody-positive and two HIV-1 antibody-negative specimens. One of the HIV-1 antibody-positive whole blood specimens was sent to the participant laboratories in duplicate. Not all laboratories received the same panel of specimens. Table 1, page 4, contains the specimen numbers and donor information for each performance evaluation specimen.

Laboratories were notified a month in advance of the date they would be receiving specimens. An air-bill tracking number was included in these preshipment letters, enabling the laboratories to locate the specimens in the event the shipment was not received by noon on the scheduled date of specimen receipt. These shipment notifications also allowed the laboratories to minimize within-institution delivery delays. Participant laboratories were instructed to process and test the MPEP $CD4^+$ T-cell specimens as they would patient specimens they routinely receive in their laboratory.

The result reporting booklet used for the October 2002 specimen shipment was designed to be consistent with the CDC guidelines for $CD4^+$ T-cell testing (MMWR, vol. 46, no. RR-2, January 10, 1997). Laboratories were encouraged by the MPEP to use these guidelines in performing $CD4^+$ T-cell determinations on patient specimens. According to these guidelines, specimens should be processed for hematologic testing and flow cytometric immunophenotyping within 30 hours of collection.

Methods used to derive the cell marker-specific absolute cell count were classified as either multi-platform or single-platform. Multi-platform methods are those which use the results from the flow cytometer (cell marker percentages) in combination with the results from a hematology analyzer (white blood cell count, percent lymphocytes, and absolute lymphocyte count) to calculate the specific absolute cell count. Single-platform methods are those methods whereby the absolute cell count is derived on a single instrument (e.g., FACSCount, TruCount, or Flow-Count) or in a single procedural assay (e.g., Coulter manual CD4, or Zymmune).

All cell marker percentage results reported by the laboratories were grouped according to the cell marker of interest, regardless of the flow cytometer or monoclonal antibody combination used to derive the specific result, e.g., $CD4^+$ results were grouped from laboratories using

CD3/CD4, CD3/CD4/CD8, CD45/CD3/CD4, and CD45/CD3/CD4/CD8. Similarly, regardless of the method used to obtain the absolute cell count (single-platform or multi-platform), all results for $CD4^+$ and $CD8^+$ absolute cell counts were grouped. These results were used to calculate 95% confidence limits for each donor and cell marker using the SAS procedure PROC GLM. Before calculation, data were analyzed for possible outliers. Only 234 (2.2%) of 10,813 results were considered to be outliers. These outlier results were removed before we calculated the 95% confidence limits shown in Table 3. However, no data from any laboratory were removed from the aggregate results table comparing values obtained by the laboratories against the 95% confidence limits.

Because of insufficient data, 95% confidence limits could not be calculated for $CD3^-/CD16^+$ or $CD3^-/CD56^+$. Table 3 shows the entire range of laboratory results (maximum and minimum) reported for these two cell markers.

Summary of Results

In general, most laboratories performed well on the donor specimens in the October 2002 shipment. The percentages of participating laboratory results within the 95% confidence limits established for the cell marker percentage results, the marker specific absolute cell counts, white blood cell count, lymphocyte percentage, and absolute lymphocyte count are shown in Table 2 below:

Table 2. Total percentage of participant laboratory results within or outside the established 95% confidence limits

	Cell Marker Percentage		Absolute Cell Counts			Hematology Results		
Cell Marker	Within 95% Confidence Limits	Outside 95% Confidence Limits	Within 95% Confidence Limits	Outside 95% Confidence Limits			Within 95% Confidence Limits	Outside 95% Confidence Limits
$CD3^+$	93.9%	6.1%	93.2%	6.8%		White Blood Cell Count	93.3%	6.7%
$CD4^+$	93.7%	6.3%	92.3%	7.7%		Lymphocyte Percentage	93.7%	6.3%
$CD8^+$	93.9%	6.1%				Absolute Lymphocyte Count	93.5%	6.5%
$CD14^+$	96.6%	3.4%						
$CD19^+$	96.1%	3.9%						
$CD45^+$	96.5%	3.5%						
$CD(56+16)^+$	95.2%	4.8%						

The types of laboratories participating in the October 2002 $CD4^+$ T-cell determinations shipment are shown in Figure 1 below.

Figure 1. Primary classification of laboratories participating in the October 2002 shipment.

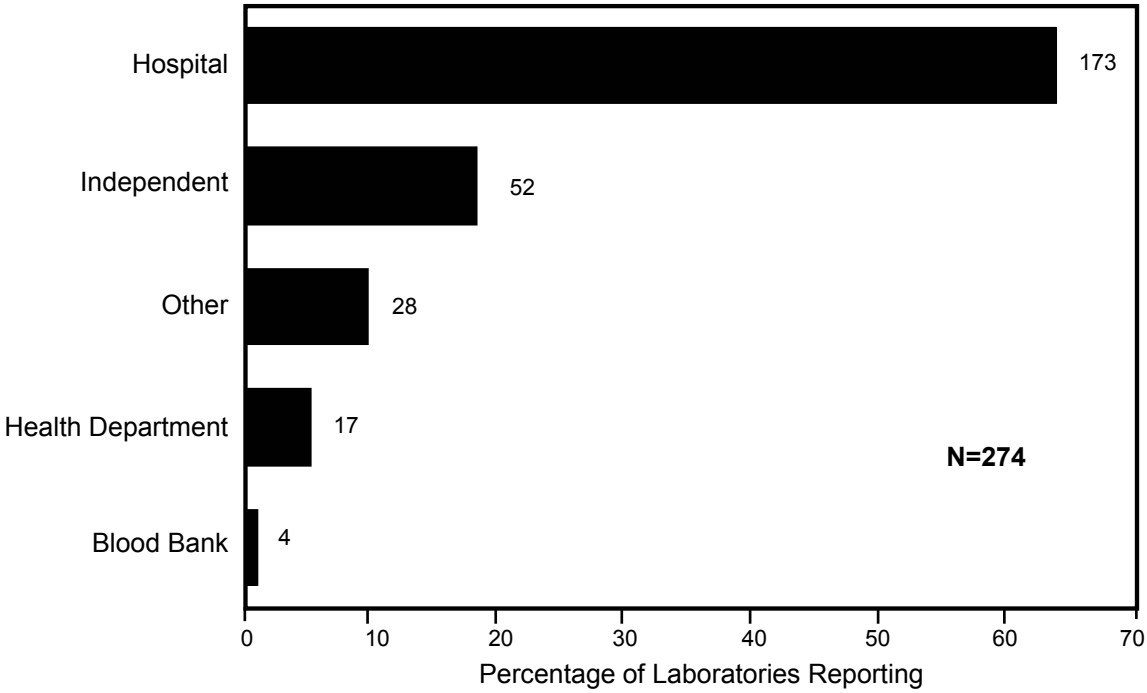
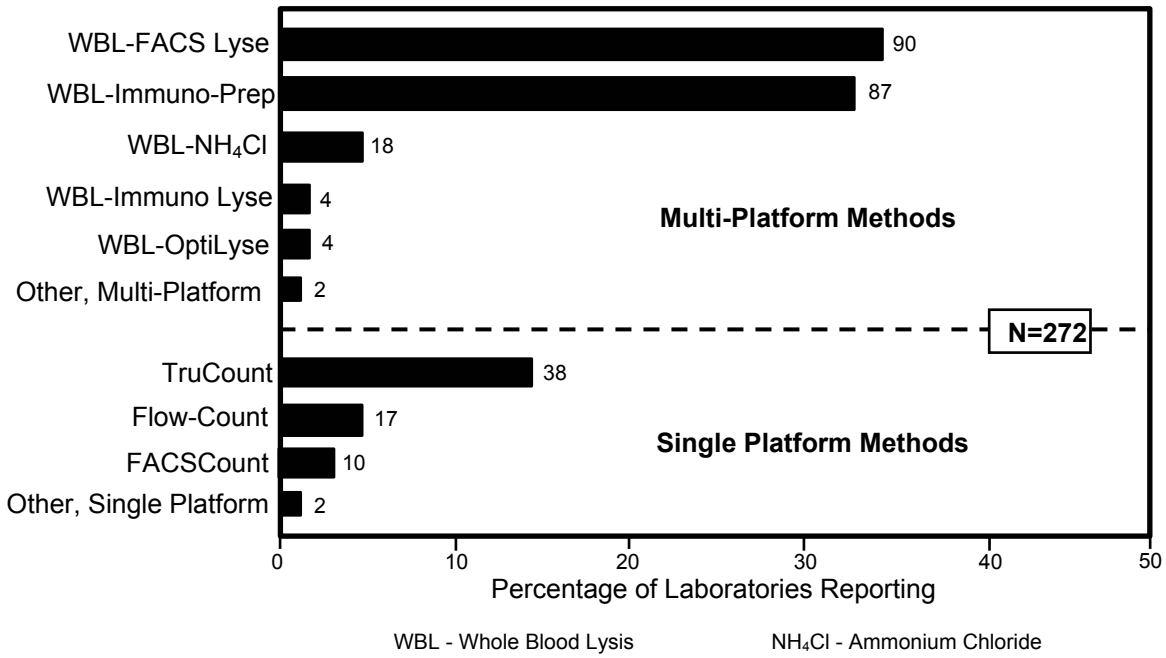


Figure 2 below shows the methods used by the laboratories to prepare specimens for $CD4^+$ T-cell determinations. All of the laboratories performing multi-platform methods reported using a method of whole blood lysis to prepare specimens for $CD4^+$ T-cell (including 2 methods described as "Other"). The frequency of preparation methods specific for single-platform methods is also reflected in this figure.

Figure 2. Methods used to prepare specimens for $CD4^+$ T-cell determinations reported by participant laboratories



"Other" multi-platform methods were described as TQ Prep and Cal-Lyse (CalTag). "Other" single-platform methods were described as FACS Calibur Volumetric Particles and Coulter Tetra-One.

Figure 3 below shows the methods used by the laboratories to fix their $CD4^+$ T-cell specimens before flow cytometric analysis. Of laboratories reporting testing results, 33 (12.4%) of 267, specifically stated that they did not fix their $CD4^+$ T-cell specimens before analyzing them, even though the panel sent to the laboratories contained known HIV antibody-positive specimens.

Figure 3. Methods used to fix specimens for $CD4^+$ T-cell determinations reported by participant laboratories

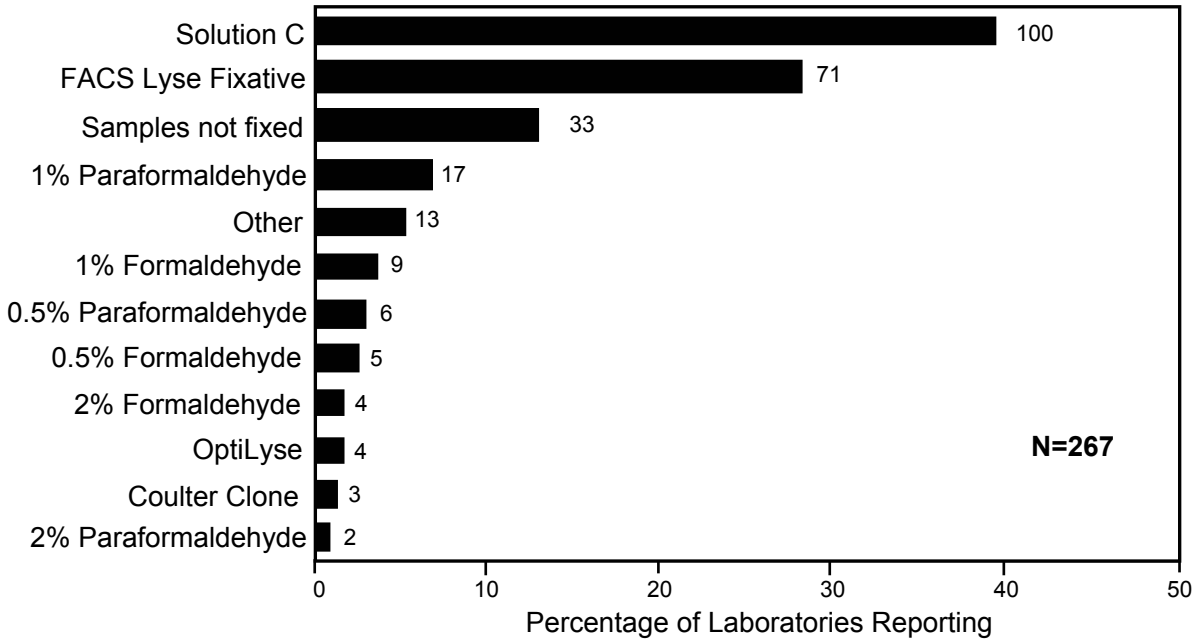
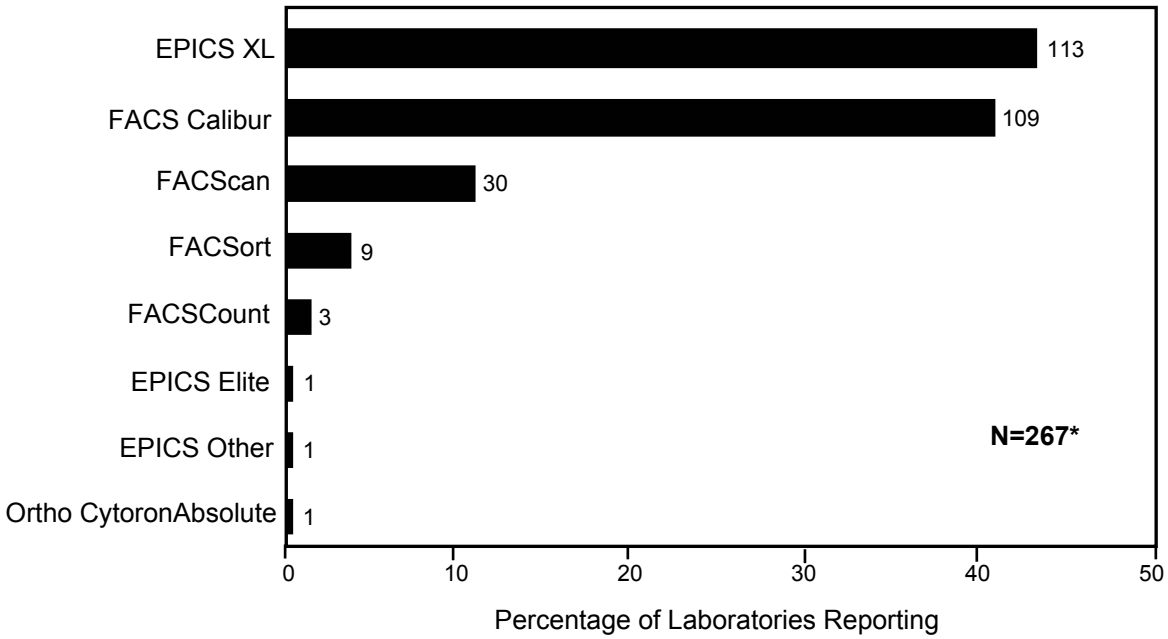


Figure 4. Types of flow cytometers used for $CD4^+$ T-cell determinations reported by participant laboratories



* Not all laboratories reported the type of flow cytometer used

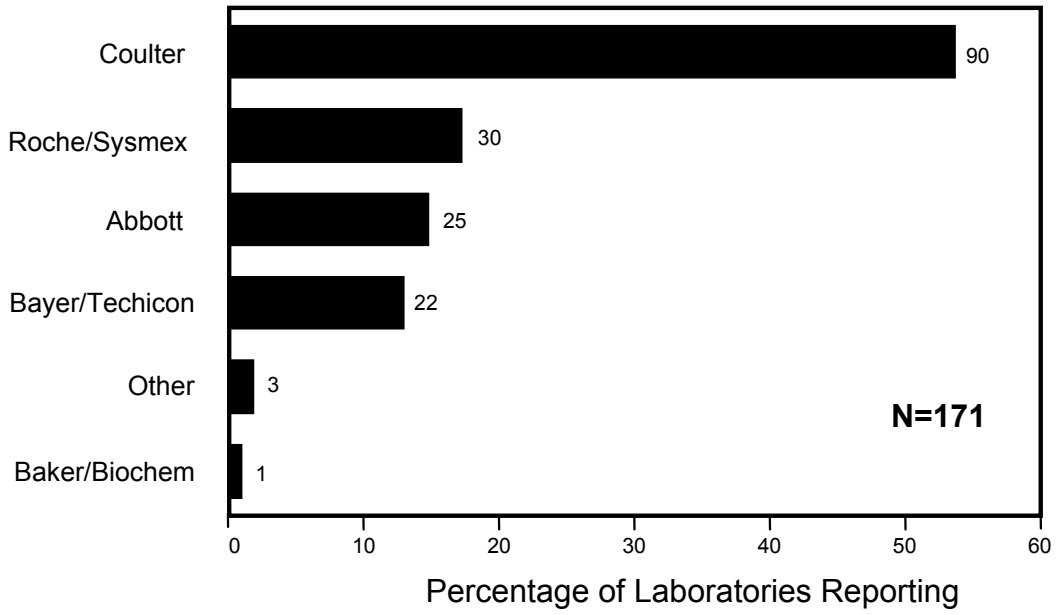
Among the 274 laboratories reporting results, 219 reported absolute cell counts. Of these, 152 (69.4%) of 219 used only a multi-platform method to derive marker-specific absolute cell counts. Sixty-seven (30.6%) of 219 laboratories, used only a single-platform method. Table 3 below shows the numbers and percentages of laboratories reporting the use of single-platform methods has increased over a six-year period.

Table 3. Laboratories reporting use of single-platform methods for absolute cell counts

Shipments	Sept. 1997	March 1998	Sept./Oct. 1998	April 1999	Oct. 1999	April 2000	Oct. 2000	April 2001	Oct. 2001	April 2002	Oct. 2002
Total # of Labs Reporting	162	188	188	208	205	198	206	205	210	215	219
# of Labs using Single-Platform	30	36	35	42	42	51	51	57	57	67	67
% of Labs using Single-Platform	18.5	19.1	18.6	20.2	20.5	25.8	24.7	27.8	27.1	31.2	30.6

Of the 274 participant laboratories, 171 (62.4%) provided information regarding the manufacturer of the hematology instrument in use in their laboratory. The manufacturers of hematology instruments used by the laboratories are shown in Figure 5 below.

Figure 5. Hematology instruments, by manufacturer, used for $CD4^+$ T-cell determinations reported by participant laboratories



Cell Marker Statistical Calculations and Results

Table 4, pages 12 – 17, contains the frequency of participant laboratory lymphocyte immunophenotyping percentage results, by donor and cell marker, within, above, or below the 95% confidence limits established using results from all laboratories, regardless of the monoclonal antibody combination or manufacturer of flow cytometry instrument used to obtain these percentage results. This table also contains the frequency of participant laboratory hematology results (white blood cell count, percentage of lymphocytes and absolute lymphocyte count) and absolute cell count results for $CD4^+$ and $CD8^+$, within, above, or below the statistically established 95% confidence limits.

Table 4. Participant Laboratory Aggregate Results, October 2002 Shipment

Donor Number 1 - Donor Status: HIV-antibody Negative

Cell Marker	Percentage Results Range	No.	Absolute Counts Range	No.
CD45	> 100	0		
	91 - 100	27		
	< 91	1		
CD14	> 2	0		
	0 - 2	25		
	< 0	0		
CD4	> 67	6	> 2,058	2
	57 - 67	117	572 - 2,058	99
	< 57	2	< 572	1
CD8	> 28	5	> 874	3
	23 - 28	118	233 - 874	97
	< 23	3	< 233	1
CD19	> 7	2		
	3 - 7	95		
	< 3	2		
CD56+16	> 7	0		
	3 - 7	58		
	< 3	4		
CD3 Average	> 95	2		
	82 - 95	87		
	< 82	5		
CD56	1 - 26	37		
CD16	1 - 5	4		

Hematology Results

Hematology Parameter	Range	No.
WBC	> 10,487	0
	5,191 - 10,487	71
	< 5,191	1
% Lymphs	> 40	3
	19 - 40	67
	< 19	1
Absolute Lymphs	> 3,608	1
	1,127 - 3,608	69
	< 1,127	2

Legend:
95% Confidence limits highlighted
"No." represents number of laboratories reporting in these ranges.
No confidence limits established for CD56 and CD16 - maximum and minimum values reported

Donor Number 2 - Donor Status: HIV-antibody Positive

Cell Marker	Percentage Results Range	No.	Absolute Counts Range	No.
CD45	> 100	0		
	96 - 100	26		
	< 96	2		
CD14	> 1	1		
	0 - 1	25		
	< 0	0		
CD4	> 35	9	> 1,821	2
	25 - 35	112	841 - 1,821	105
	< 25	3	< 841	5
CD8	> 35	8	> 1,626	6
	21 - 35	117	821 - 1,626	101
	< 21	1	< 821	3
CD19	> 18	2		
	11 - 18	93		
	< 11	5		
CD56+16	> 30	2		
	21 - 30	61		
	< 21	1		
CD3 Average	> 66	7		
	53 - 66	90		
	< 53	2		
CD56	9 - 35	42		
CD16	26 - 27	2		

Hematology Results

Hematology Parameter	Range	No.
WBC	> 13,562	1
	5,910 - 13,562	72
	< 5,910	5
% Lymphs	> 53	2
	39 - 53	70
	< 39	4
Absolute Lymphs	> 5,968	2
	2,837 - 5,968	73
	< 2,837	3

Table 4. Participant Laboratory Aggregate Results, October 2002 Shipment

Donor Number 3 - Donor Status: HIV-antibody Negative

Cell Marker	Percentage Results Range	No.	Absolute Counts Range	No.
CD45	> 100	0		
	93 - 100	30		
	< 93	0		
CD14	> 2	2		
	0 - 2	25		
	< 0	0		
CD4	> 54	11	> 1,207	7
	43 - 54	116	766 - 1,207	94
	< 43	0	< 766	2
CD8	> 21	4	> 478	7
	17 - 21	123	294 - 478	93
	< 17	1	< 294	2
CD19	> 15	2		
	10 - 15	95		
	< 10	4		
CD56+16	> 24	0		
	17 - 24	60		
	< 17	4		
CD3 Average	> 74	9		
	61 - 74	85		
	< 61	0		
CD56	3 - 21	37		
CD16	17 - 21	4		

Hematology Results

Hematology Parameter	Range	No.
WBC	> 6,715	2
	5,640 - 6,715	68
	< 5,640	3
% Lymphs	> 39	4
	28 - 39	68
	< 28	0
Absolute Lymphs	> 2,508	5
	1,649 - 2,508	68
	< 1,649	0

Legend:
95% Confidence limits highlighted
"No." represents number of laboratories reporting in these ranges.
No confidence limits established for CD56 and CD16 - maximum and minimum values reported

Donor Number 4 - Donor Status: HIV-antibody Positive

Cell Marker	Percentage Results Range	No.	Absolute Counts Range	No.
CD45	> 100	0		
	95 - 100	32		
	< 95	0		
CD14	> 3	0		
	0 - 3	28		
	< 0	0		
CD4	> 40	4	> 1,151	4
	34 - 40	123	700 - 1,151	86
	< 34	3	< 700	4
CD8	> 68	0	> 1,947	3
	44 - 68	124	787 - 1,947	86
	< 44	6	< 787	5
CD19	> 5	1		
	2 - 5	101		
	< 2	0		
CD56+16	> 11	1		
	6 - 11	62		
	< 6	1		
CD3 Average	> 91	3		
	83 - 91	84		
	< 83	1		
CD56	1 - 11	32		
CD16	8 - 12	6		

Hematology Results

Hematology Parameter	Range	No.
WBC	> 7,850	2
	5,892 - 7,850	62
	< 5,892	4
% Lymphs	> 45	5
	28 - 45	62
	< 28	1
Absolute Lymphs	> 3,050	3
	2,036 - 3,050	61
	< 2,036	4

Table 4. Participant Laboratory Aggregate Results, October 2002 Shipment

Donor Number 5 - Donor Status: HIV-antibody Positive

Cell Marker	Percentage Results Range	No.	Absolute Counts Range	No.
CD45	> 100	0		
	91 - 100	30		
	< 91	0		
CD14	> 3	0		
	0 - 3	27		
	< 0	0		
CD4	> 18	3	> 301	7
	12 - 18	124	139 - 301	94
	< 12	0	< 139	2
CD8	> 57	6	> 988	4
	43 - 57	121	453 - 988	94
	< 43	1	< 453	4
CD19	> 30	0		
	14 - 30	99		
	< 14	2		
CD56+16	> 8	4		
	1 - 8	60		
	< 1	0		
CD3 Average	> 80	2		
	63 - 80	92		
	< 63	0		
CD56	0 - 47	37		
CD16	4 - 7	4		

Hematology Results

Hematology Parameter	Range	No.
WBC	> 4,512	3
	3,514 - 4,512	67
	< 3,514	3
% Lymphs	> 49	4
	28 - 49	67
	< 28	1
Absolute Lymphs	> 1,975	6
	1,104 - 1,975	66
	< 1,104	1

Legend:
95% Confidence limits highlighted
"No." represents number of laboratories reporting in these ranges.
No confidence limits established for CD56 and CD16 - maximum and minimum values reported

Donor Number 6 - Donor Status: HIV-antibody Positive

Cell Marker	Percentage Results Range	No.	Absolute Counts Range	No.
CD45	> 100	0		
	85 - 100	34		
	< 85	0		
CD14	> 3	2		
	0 - 3	32		
	< 0	0		
CD4	> 51	5	> 954	7
	42 - 51	121	626 - 954	99
	< 42	0	< 626	0
CD8	> 28	5	> 525	6
	23 - 28	119	345 - 525	97
	< 23	0	< 345	1
CD19	> 17	0		
	10 - 17	105		
	< 10	5		
CD56+16	> 18	1		
	6 - 18	57		
	< 6	2		
CD3 Average	> 80	3		
	67 - 80	88		
	< 67	1		
CD56	3 - 12	38		
CD16	7 - 14	8		

Hematology Results

Hematology Parameter	Range	No.
WBC	> 6,463	3
	5,319 - 6,463	80
	< 5,319	3
% Lymphs	> 34	5
	23 - 34	79
	< 23	2
Absolute Lymphs	> 2,026	5
	1,351 - 2,026	81
	< 1,351	0

Table 4. Participant Laboratory Aggregate Results, October 2002 Shipment

Donor Number 7 - Donor Status: HIV-antibody Positive

Cell Marker	Percentage Results Range	No.	Absolute Counts Range	No.
CD45	> 100	0		
	96 - 100	32		
	< 96	0		
CD14	> 1	0		
	0 - 1	32		
	< 0	0		
CD4	> 29	2	> 637	5
	23 - 29	134	392 - 637	116
	< 23	10	< 392	5
CD8	> 50	0	> 1,094	3
	41 - 50	134	721 - 1,094	115
	< 41	12	< 721	8
CD19	> 22	0		
	17 - 22	109		
	< 17	1		
CD56+16	> 10	3		
	6 - 10	78		
	< 6	1		
CD3 Average	> 75	0		
	68 - 75	77		
	< 68	3		
CD56	3 - 11	30		
CD16	NA			

Hematology Results

Hematology Parameter	Range	No.
WBC	> 4,640	2
	3,804 - 4,640	80
	< 3,804	4
% Lymphs	> 55	3
	43 - 55	81
	< 43	2
Absolute Lymphs	> 2,378	0
	1,715 - 2,378	82
	< 1,715	4

Legend:
95% Confidence limits highlighted
"No." represents number of laboratories reporting in these ranges.
No confidence limits established for CD56 and CD16 - maximum and minimum values reported

Donor Number 8 - Donor Status: HIV-antibody Negative

Cell Marker	Percentage Results Range	No.	Absolute Counts Range	No.
CD45	> 100	0		
	94 - 100	30		
	< 94	3		
CD14	> 2	3		
	0 - 2	30		
	< 0	0		
CD4	> 48	2	> 922	5
	39 - 48	126	589 - 922	109
	< 39	8	< 589	2
CD8	> 31	3	> 592	7
	23 - 31	126	350 - 592	106
	< 23	6	< 350	2
CD19	> 15	0		
	10 - 15	107		
	< 10	3		
CD56+16	> 14	1		
	9 - 14	69		
	< 9	1		
CD3 Average	> 79	1		
	71 - 79	81		
	< 71	4		
CD56	1 - 17	34		
CD16	2 - 13	4		

Hematology Results

Hematology Parameter	Range	No.
WBC	> 8,270	2
	7,048 - 8,270	81
	< 7,048	3
% Lymphs	> 27	4
	19 - 27	82
	< 19	0
Absolute Lymphs	> 2,085	3
	1,386 - 2,085	81
	< 1,386	2

Table 4. Participant Laboratory Aggregate Results, October 2002 Shipment

Donor Number 9 - Donor Status: HIV-antibody Negative

Cell Marker	Percentage Results Range	No.	Absolute Counts Range	No.
CD45	> 100	0		
	92 - 100	30		
	< 92	3		
CD14	> 2	1		
	0 - 2	32		
	< 0	0		
CD4	> 52	1	> 1,397	6
	43 - 52	128	656 - 1,397	108
	< 43	7	< 656	2
CD8	> 21	2	> 538	7
	15 - 21	128	242 - 538	106
	< 15	5	< 242	2
CD19	> 20	0		
	15 - 20	106		
	< 15	4		
CD56+16	> 17	1		
	10 - 17	69		
	< 10	1		
CD3 Average	> 73	1		
	64 - 73	81		
	< 64	4		
CD56	6 - 16	34		
CD16	1 - 16	4		

Hematology Results

Hematology Parameter	Range	No.
WBC	> 9,746	1
	8,163 - 9,746	80
	< 8,163	5
% Lymphs	> 34	5
	15 - 34	80
	< 15	1
Absolute Lymphs	> 3,038	5
	1,321 - 3,038	80
	< 1,321	1

Legend:
95% Confidence limits highlighted
"No." represents number of laboratories reporting in these ranges.
No confidence limits established for CD56 and CD16 - maximum and minimum values reported

Donor Number 10 - Donor Status: HIV-antibody Positive

Cell Marker	Percentage Results Range	No.	Absolute Counts Range	No.
CD45	> 100	0		
	94 - 100	31		
	< 94	2		
CD14	> 1	1		
	0 - 1	32		
	< 0	0		
CD4	> 11	3	> 225	5
	7 - 11	130	129 - 225	110
	< 7	4	< 129	1
CD8	> 65	6	> 1,329	8
	50 - 65	123	892 - 1,329	105
	< 50	6	< 892	2
CD19	> 29	1		
	20 - 29	102		
	< 20	7		
CD56+16	> 9	4		
	4 - 9	67		
	< 4	0		
CD3 Average	> 73	6		
	63 - 73	79		
	< 63	1		
CD56	1 - 10	34		
CD16	4 - 8	4		

Hematology Results

Hematology Parameter	Range	No.
WBC	> 6,341	4
	5,423 - 6,341	80
	< 5,423	2
% Lymphs	> 37	3
	29 - 37	83
	< 29	0
Absolute Lymphs	> 2,214	2
	1,700 - 2,214	81
	< 1,700	3

As can be seen in Table 5 below, the range of results reported for absolute $CD4^+$ and $CD8^+$ T-cell counts was different depending on the method used to obtain the result, i.e., single-platform or multi-platform. **Note: These are inclusive ranges (lowest value to highest value) and are not 95% confidence limits as presented in the results in the previous tables.**

Table 5. Inclusive* Range of Absolute T-cell Counts Reported, Single-Platform vs. Multi-Platform Derived

		$CD4^+$ T-cell Count		$CD8^+$ T-cell Count		Absolute Lymphocyte Count
Vial Label	Donor Identification	Single-Platform	Multi-Platform	Single-Platform	Multi-Platform	
A4, B2	1	732 - 1221	505 - 2344	284 - 1601	200 - 999	200 - 3843
A2, A3	2	853 - 1642	348 - 2043	905 - 1454	261 – 2554	551 - 7084
A5, B5	3	716 - 1053	818 - 1812	279 - 451	274 - 694	1700 - 3856
B1, B3	4	581 - 1193	7 - 2248	480 - 1726	337 - 3078	748 – 5765
A1, B4	5	124 - 251	133 – 1079	423 - 897	416 – 1750	885 - 3182
D3, D5	6	657 - 861	644 – 1500	372 – 488	343 – 860	1410 – 3200
C1, C2	7	371 – 924	304 – 1052	673 – 1368	537 – 1135	1065 – 2365
C5, D2	8	559 – 1367	482 – 1171	365 – 840	282 – 905	1159 – 3016
C3, D4	9	786 – 1607	473 – 3000	309 – 561	176 – 1213	1010 – 6383
C4, D1	10	142 – 303	105 – 516	872 – 1569	605 – 1873	1226 - 2901

* Inclusive ranges – smallest to largest value, not 95% confidence limits

The Model Performance Evaluation Program for $CD4^+$ T-cell determinations is interested in the total testing process, including errors made in reporting due to errors in mathematical calculation. In general, the multi-platform ranges were larger than the corresponding single-platform ranges for both $CD4^+$ and $CD8^+$ absolute T-cell counts. The ranges of multi-platform results were affected by the magnitude of the ranges of the absolute lymphocyte count results (last column), which were often quite large (e.g., Donors 2, 4, and 9). The magnitude of some of the ranges may be caused by simple reporting errors on the part of the laboratories. For example, one laboratory for one specimen tested reported a lymphocyte count result that was in error by nearly a factor of 10 (i.e., the laboratory reported a WBC of 6800 and a lymphocyte percent of 26, which should have yielded a lymphocyte count of 1768; however, the laboratory reported a lymphocyte count of 200). There were a total of eight laboratories that reported lymphocyte counts that were greater than 5% different than the true calculated lymphocyte count (WBC X Lymphocyte percent) on at least one specimen. Of these eight, two laboratories reported inaccurately calculated lymphocyte counts (greater than 5% difference between true and reported) on all 5 specimens tested.

Discussion

Specimen panel receipt was delayed one day for five laboratories due to problems related to the overnight carrier (FedEx). Eight laboratories reported a one-day delay, two laboratories reported a two-day delay, and two laboratories reported a 6-day delay in receiving their specimens due to delivery problems within their institution. Additionally, 35 (12.8%) of 274 laboratories reported they did not process the MPEP $CD4^+$ T-cell specimens on the day they were received (32 laboratories, one-day delay; two laboratories, two-day delay; one laboratory, 5-day delay). These delays may have affected the testing results from these laboratories.

Differences in laboratory performance of cell marker analysis may be related to:

- the use of the CDC $CD4^+$ T-cell testing guidelines
- the use of multi-platform versus single-platform procedures
- the use of different flow cytometer, hematology instrument, and reagent manufacturer combinations
- factors associated with specimen preparation (including specimen fixation before analysis and delay in preparing specimens for analysis), and
- reporting errors on the part of the laboratories.

Those laboratories performing $CD4^+$ T-cell determinations using a single-platform method should follow the recently published CDC *Guidelines for Performing Single-Platform Absolute CD4+ T-Cell Determinations with CD45 Gating for Persons Infected with Human Immunodeficiency Virus* [MMWR 2003 January 31; 52(RR-2):1-13].