Association of Selected Phenotypic Markers of Lymphocyte Activation and Differentiation with Perinatal Human Immunodeficiency Virus Transmission and Infant Infection

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This study of a subset of women and infants participating in National Institutes of Health Pediatric AIDS Clinical Trials Group protocol 185 evaluated lymphocyte phenotypic markers of immune activation and differentiation to determine their association with the likelihood of human immunodeficiency virus (HIV) transmission from the women to their infants and the potential for early identification and/or prognosis of infection in the infants. Lymphocytes from 215 human immunodeficiency virus type 1 (HIV)-infected women and 192 of their infants were analyzed by flow cytometry with an extended three-color panel of monoclonal antibodies. Women who did not transmit to their infants tended to have higher CD4+ T cells. Most notably, levels of total CD8+ T cells and CD8+ CD38+ cells made significant independent contributions to predicting the risk of mother-to-child transmission. Adjusting for HIV-1 RNA level at entry, a one percentage-point increase in these marker combinations was associated with a nine percent increase in the likelihood of maternal transmission. Total as well as naïve CD4+ T cells were significantly higher in uninfected than infected infants. Total CD8+ cells, as well as CD8+ cells positive for HLA-DR+, CD45 RA+ HLA-DR+, and CD28+ HLA-DR+ were elevated in infected infants. Detailed immunophenotyping may be helpful in predicting which pregnant HIV-infected women are at increased risk of transmitting HIV to their infants. Increasing differences in lymphocyte subsets between infected and uninfected infants became apparent as early as six weeks of age. Detailed immunophenotyping may be useful in supporting the diagnosis of HIV infection in infants with perinatal HIV exposure.

CD4+ T-lymphocyte count is recognized universally as a standard indicator in the medical management of human immunodeficiency virus (HIV) infection. More detailed phenotypic characterization of lymphocyte subsets has contributed to the understanding of disease pathogenesis and progression in HIV-infected patients in a number of clinical centers where multiparameter flow cytometry can be performed. Such studies have shown that lymphocyte activation is characteristic of HIV infection, and that CD8+ cells lacking activation markers can be an independent indication of good prognosis in men. Several studies have identified lymphocyte subsets associated with increased risk of mother-to-child transmission of HIV or with early identification of infected children. Historically, one of the challenges in assessing the immunopathogenesis of HIV in pregnant women has been separating out the effects of the infection from the generalized effects of immunological suppression during pregnancy. A number of studies have now provided clarification for some of these differences. In addition, it also has been a challenge to differentiate immunological changes in infants caused by HIV from those due to the normal maturation of the immune system that occurs rapidly over the first few months and years of life. Several studies have provided normal values for lymphocyte subsets in healthy infants and young children, and some also have identified characteristic differences in HIV-infected infants. This study confirms those findings in a much larger number of women and infants than studied previously, and reports a new correlate of mother-to-child transmission.

PACTG 185 was a multicenter, randomized phase III controlled clinical trial, conducted between October 1993 and March 1997 at 53 clinical sites in the United States mainland and Puerto Rico. The trial was designed to evaluate whether prophylaxis with hyperimmune anti-HIV-1 intravenous immune globulin (HIVIG) would lower the risk of perinatal HIV-1 transmission relative to that achieved with intravenous...
infusions of standard immune globulin (IVIG) in HIV-infected pregnant women with CD4+ T-lymphocyte counts of 500/µl or below who receive zidovudine treatment and prophylaxis (32). Women were followed for two years after delivery (37), and children born to these mothers were followed for a period of 18 months postpartum to assess the safety of the administration of these treatments and to assess HIV infection status. Zidovudine therapy and prophylaxis reduced mother-to-child transmission risk to rates inadequate to provide sufficient power to determine whether HIVIG contributed to further transmission reduction, and the study was stopped early for that reason. Further analyses of virologic data from this trial showed that HIV transmission increases with increasing viral load, although mother-to-child transmission can occur at almost any level (21).

We report here the results of a Pediatric AIDS Clinical Trials Group (PACTG) 185 substudy analysis of detailed lymphocyte phenotype that was conducted on all study subjects from clinics served by three PACTG advanced flow cytometry laboratories. The overall objectives of this substudy were to evaluate, in HIV-infected pregnant women, whether selected lymphocyte immunophenotypic markers of activation and differentiation were associated with perinatal HIV transmission or the prevention of mother-to-child transmission, to evaluate, in perinatally exposed newborns, whether selected lymphocyte immunophenotypic markers of activation and differentiation were associated with infant HIV infection, and to assess whether HIVIG or IVIG administered to the mothers and newborns had any effect on these markers.

(Preliminary findings have been presented in part [6th Conference on Retroviruses and Opportunistic Infections, Chicago, Illinois, February 1, 1999, Abstract 240].)

MATERIALS AND METHODS

Study design. A total of 215 HIV-infected pregnant women (eight of whom transmitted HIV to their infants) and 192 of their infants (six of whom were HIV infected) were evaluated. Among the 215 women, 109 received HIVIG and 106 received IVIG. Among the 192 children, 96 received HIVIG and 96 received IVIG. All study subjects enrolled in PACTG 185 at the three sites performing the PACTG 185 substudy (Baylor College of Medicine, Rush Medical College, and UCLA School of Medicine) were included. The median age of the 215 women included in this analysis was 26.6 years (25th and 75th percentiles, 22.3 and 30.7). Race was reported as 11% white, 42% black, 46% Hispanic, and 1% other.

Immunophenotyping of lymphocyte subset. Fresh whole blood was transported to the three laboratories at ambient temperature and processed within 2 h of collection. Enumeration of lymphocyte phenotypes was performed according to standard methods (7, 22), which are used in the consensus protocol of the AIDS Clinical Trial Group. Specific lymphocyte subsets were identified by three-color flow cytometry using combinations of monoclonal antibodies to lymphocyte surface antigens conjugated to fluorochromes fluorescein isothiocyanate, phycoerythrin, and energy coupled dye or peridinin chlorophyll protein. A seven-tube panel was used to assess the combinations CD4/CD19/CD8, CD8/CD3, CD19/CD62L/CD3, HLA-DR/CD38/CD3, HLA-DR/CD38/CD4, CD45RA/CD45RO/CD4, and CD57/CD56 16/CD3 in women. A six-tube panel was used to assess the combinations CD45/CD14/CD19, CD8/CD4/CD3, CD45RA/HLA-DR/CD4, CD45RA/HLA-DR/CD8, CD28/HLA-DR/CD8, and CD23/CD62L/CD19 in infants. Stained cells were enumerated using either a Coulter EPICS XL or Becton Dickinson FACScan laser flow cytometer. The three laboratories where testing was performed demonstrated approved performance in the NIH Division of AIDS Immunology Quality Assurance Program and had undergone further assessment for assay consistency using a detailed immunophenotyping panel during the course of the substudy.

Statistical analysis. Repeated measures longitudinal analysis was performed to compare different flow cytometric markers of lymphocyte activation and memory according to treatment group, maternal HIV transmission status, and infant HIV infection status. The repeated measures model was used to estimate the mean proportion (and its standard error) of selected lymphocyte immunophenotypic markers and the overall difference between groups, both over individuals within groups and over multiple time points within individuals. The model yields the P value associated with the test of whether the difference in group effects differs significantly from zero. The “least squares mean” option was used to calculate mean values. This method adjusts for trends over time and, if desired, covariates. Models for women were fit to the data with and without adjustment for baseline viral load (log10 HIV-1 RNA).

Separate logistic regression analyses were conducted to evaluate predictive associations between prenatal maternal lymphocyte immunophenotypic marker combinations and transmission risk. The odds ratio, estimating the magnitude of the increase or decrease in mother-to-child transmission risk associated with a one percentage-point increase in each marker combination, is presented along with the 95 percent confidence interval for the estimate. Models were fit to the data with and without adjustment for baseline viral load (log10 HIV-1 RNA).

RESULTS

Lymphocyte immunophenotyping results for individual subjects expressed as relative counts over time are shown in Fig. 1 in transmitting compared with non-transmitting women for those marker combinations associated with HIV transmission or protection from transmission (CD3+CD4+, CD4+DR+, CD4+CD45RO+, CD3+CD8+, CD8+CD38+, and CD8+CD38+DR+) and in Fig. 2 for infected compared with uninfected infants for those marker combinations associated with HIV infection or lack of infection (CD3+CD4+, CD4+CD45RA+, CD3+CD8+, CD8+DR+, CD8+CD45RA-DR-, CD8+CD28-DR+), illustrating the direction and magnitude of differences over time and at each study evaluation time point. Marker combinations that demonstrated no association with protection from or risk of transmitting or acquiring HIV are not shown (CD3+, CD3+CD62L+, CD8+CD45RA-, CD19-, CD19+CD23-, CD19+CD23+CD62L-, CD3-CD16+/56+).

Tables 1 and 2 display the comparison of maternal markers by transmission status and treatment arm. Statistically significant differences between non-transmitters and transmitters were observed for the combinations of CD3+CD4+ (P = 0.046) and for CD8−CD38− (P = 0.026) (Table 1). Adjusting for baseline HIV-1 RNA level, the combination of CD3+CD4+ no longer differed significantly between non-transmitters and transmitters, while differences for the combinations of CD4+DR+ and CD8−CD38− were of borderline significance (P = 0.052). The maternal comparison of those who received HIVIG versus IVIG shows statistically significant differences for CD3+CD8− (P = 0.039) and for CD3+ cells (P = 0.016) in the unadjusted models (Table 2). After adjusting for HIV-1 RNA at entry, statistically significant differences remained for
FIG. 1. Maternal lymphocyte immunophenotypic marker combinations demonstrating associations with HIV transmission or protection from transmission. Open circles denote nontransmitting women and closed circles denote transmitting women, with regression lines and 95 percent confidence intervals depicted for each group.
CD8⁺ Maternal T-lymphocytes

CD8⁺CD38⁺ Maternal T-lymphocytes

CD8⁺CD38⁺DR⁺ Maternal T-lymphocytes

FIG. 1—Continued.
FIG. 2. Infant lymphocyte immunophenotypic marker combinations demonstrating associations with HIV infection or lack of infection. Open circles denote uninfected infants and closed circles denote infected infants, with regression lines and 95 percent confidence intervals depicted for each group.
FIG. 2—Continued.

**CD8⁺DR⁺ Infant T-lymphocytes**

**CD8⁺RA⁺DR⁺ Infant T-lymphocytes**

**CD8⁺CD28⁺DR⁺ Infant T-lymphocytes**
and test for differences between groups.

The percent difference was calculated based on the mean proportion for nontransmitters minus the mean proportion for transmitters.

Table 3 summarizes the results of logistical regression analyses estimating the risk of perinatal transmission of HIV associated with a one percentage-point increase in selected prenatal maternal flow cytometric markers of lymphocyte activation and memory. In the unadjusted models, the odds ratios for the marker combinations of CD3^+ CD4^+ (P = 0.025) and CD4^+ CD45RO^+ (P = 0.040) differed significantly from zero; a one-percentage increase in these marker combinations was associated with a modest decrease in the likelihood of mother-to-child transmission. The marker combinations of CD3^+ CD8^+ (P = 0.024), CD8^+ CD38^+ (P = 0.007) and CD8^+ CD38^+ DR^+ (P = 0.023) were associated with modest but significant increases in risk of perinatal transmission. However, with adjustment for maternal entry HIV-1 RNA level, only the combinations CD3^+ CD8^+ and CD8^+ CD38^+ made a significant independent contribution to predicting the risk of mother-to-child transmission; adjusting for HIV-1 RNA level at entry, a one-percentage-point increase in these marker combinations was associated with a nine percent increase in the likelihood of mother-to-child transmission.

Lymphocyte subsets in children. Table 4 shows the longitudinal comparison of markers assessed in infants by infection status and treatment arm. Statistically significant differences by infection status were observed for the following combinations: CD3^+ CD4^+ (P < 0.001); CD4^+ CD45 RA^+ (P = 0.006); CD3^+ CD8^+ (P = 0.0001); CD8^+ DR^+ (P = 0.0001); CD8^+ CD45RA^+ DR^+ (P = 0.011); and CD8^+ CD28^+ DR^+ (P = 0.001). Uninfected infants had higher levels of CD3^+ CD4^+ and CD4^+ CD45RA^+ cells than infected infants, whereas all of the CD8^+ T-cell combinations were increased in the infected infants. In the analyses comparing different combinations of lymphocyte markers between those who received HIVIG ver-

### Table 1. Comparison of relative counts of selected prenatal maternal immunophenotypic markers of activation and differentiation by HIV perinatal transmission statusa

| Marker combination | Unadjusted analyses | | | Adjusted analyses | | |
|-------------------|---------------------|----------------|----------------|---------------------|----------------|
|                   | Nontransmitter mean (SE), % | Transmitter mean (SE), % | Percent difference | P | Nontransmitter mean (SE), % | Transmitter mean (SE), % | Percent difference | P |
| CD3^+ CD4^+       | 23.47 (0.64) | 16.98 (3.18) | 6.50 | 0.046 | 23.26 (0.59) | 18.95 (2.93) | 4.31 | 0.15 |
| CD4^+ CD38^+      | 13.61 (0.38) | 11.87 (1.95) | 1.73 | 0.38 | 13.54 (0.37) | 12.76 (1.88) | 0.79 | 0.68 |
| CD4^+ DR^+        | 3.57 (0.11) | 2.59 (0.53) | 0.97 | 0.07 | 3.57 (0.10) | 2.51 (0.53) | 1.05 | 0.052 |
| CD8^+ CD38^+ DR^+ | 2.14 (0.07) | 1.66 (0.34) | 0.47 | 0.18 | 2.15 (0.07) | 1.58 (0.34) | 0.57 | 0.11 |
| CD4^+ CD45 RO^+   | 13.48 (0.40) | 10.20 (2.01) | 3.28 | 0.11 | 13.42 (0.37) | 11.25 (1.87) | 2.17 | 0.26 |
| CD4^+ CD45 RA^A   | 9.25 (0.35) | 7.27 (1.72) | 1.98 | 0.26 | 9.21 (0.33) | 8.15 (1.66) | 1.05 | 0.53 |
| CD3^+ CD8^+       | 51.41 (0.64) | 56.66 (3.18) | 5.25 | 0.11 | 51.46 (0.63) | 56.32 (3.17) | 4.86 | 0.13 |
| CD8^+ CD38^+      | 37.51 (0.76) | 44.37 (3.80) | 6.86 | 0.026 | 36.04 (0.73) | 43.32 (3.65) | 7.28 | 0.052 |
| CD8^+ DR^+        | 24.87 (0.67) | 28.62 (3.31) | 3.75 | 0.27 | 25.00 (0.64) | 28.04 (3.21) | 3.04 | 0.35 |
| CD8^+ CD38^+ DR^+ | 19.80 (0.60) | 23.91 (3.03) | 4.11 | 0.19 | 20.07 (0.58) | 23.42 (2.91) | 3.35 | 0.26 |
| CD19^+            | 10.39 (0.39) | 8.89 (2.07) | 1.50 | 0.48 | 10.48 (0.39) | 8.78 (2.06) | 1.70 | 0.42 |
| CD3^+            | 80.01 (0.58) | 81.01 (2.85) | 1.00 | 0.73 | 79.79 (0.56) | 81.44 (2.80) | 1.66 | 0.56 |
| CD3^+ CD62L^+     | 43.17 (0.77) | 39.45 (4.02) | 3.72 | 0.36 | 43.10 (0.71) | 41.34 (3.75) | 1.76 | 0.65 |
| CD3^+ CD16^+/56^+ | 5.87 (0.29) | 7.02 (1.44) | 1.15 | 0.44 | 5.90 (0.28) | 6.64 (1.39) | 0.73 | 0.61 |

a See Table 1, footnotes a to c.
sus IVIG treatment, the comparison was significant only for the marker combination of CD4+CD45RA+ (P = 0.02). It should be noted that the observed differences in lymphocyte subsets for the six infected infants became more pronounced after 10 weeks of age (data not shown).

**DISCUSSION**

This study’s most novel finding is the predictive association with transmission of prenatal markers of lymphocyte activation in HIV-infected pregnant women. However, the individual values for these markers do not show discrete values that are predictive, i.e., a substantial number of women with similar subset values did not transmit HIV to their infants. We found significant reductions in total and memory CD4+ T-lymphocytes, and increases in total and activated CD8+ T-lymphocytes in transmitting compared with non-transmitting women. However, when adjustments were made for the contribution of HIV viral load, only CD4+DR- and CD8+CD38+ lymphocytes maintained borderline significance. High levels of CD8+CD38+ cells have previously been associated with poor disease prognosis in HIV-infected men (10). In a study of 75 HIV-infected pregnant women, Plaeger et al. did not find a significant difference in these subsets in the nine transmitting women, possibly due to the smaller number of subjects in the study (25).

Studies concurrent with PACTG 185 indicated an increased risk of transmission with increasing viral load but showed that mother-to-child transmission of HIV can occur at almost any level (21). Thus, an adjustment for viral load permitted us to identify activated lymphocyte subsets that might be an indicator for intensification of antiretroviral treatment or obstetrical intervention. Monitoring such markers during pregnancy, either where viral load measurements are not available or as a complement to viral load, to assess mother-to-child transmission risk may be useful in managing HIV-infected pregnant women. In particular, identifying women at higher risk of transmission by lymphocyte immunophenotyping may afford the opportunity to administer additional treatment interventions in pregnancy to women who might be assumed to be at reduced risk because of low viral load.

Of note, when comparing the HIVIG with the IVIG treated arm, there were significant differences in CD3+CD8- and CD3+ lymphocyte subsets, which persisted when adjusted for viral load. At the same time, there was no detectable effect on the subsets defined by activation markers. Thus, HIVIG may exert an effect on pregnant women, independent of viral load, by an as yet unexplained immunological mechanism. However, similar differences were not observed among infants in this study. Likewise, use of prenatal IVIG in other disorders is not associated with lymphocyte activation or premature maturation of the neonatal immune system (26).

### TABLE 3. Estimated risk (odds ratio and 95% confidence interval [CI]) of perinatal transmission of HIV infection associated with a one-percentage-point change in relative counts of selected prenatal maternal immunophenotypic markers of activation and differentiation

| Marker combination | Unadjusted odds ratio (95% CI) | Adjusted odds ratio (95% CI) |
|--------------------|-------------------------------|----------------------------|
| CD3+ CD4+         | 0.92 (0.85–0.99)              | 0.93 (0.86–1.01)            |
| CD4+ CD38-        | 0.91 (0.81–1.01)              | 0.93 (0.83–1.03)            |
| CD4+ DR-          | 0.58 (0.31–1.09)              | 0.58 (0.32–1.06)            |
| CD4+ CD45 RA+     | 0.57 (0.31–1.09)              | 0.53 (0.24–1.18)            |
| CD4+ CD45 RO+     | 0.86 (0.75–0.99)              | 0.88 (0.76–1.03)            |
| CD4+ CD45 RA+     | 0.88 (0.75–1.02)              | 0.90 (0.77–1.05)            |
| CD8+ CD8-         | 1.09 (1.01–1.18)              | 1.09 (1.004–1.18)           |
| CD8+ CD38-        | 1.10 (1.03–1.18)              | 1.09 (1.02–1.17)            |
| CD8+ DR-          | 1.06 (1.00–1.13)              | 1.05 (0.98–1.12)            |
| CD8+ CD38-DR+     | 1.07 (1.01–1.14)              | 1.06 (0.995–1.144)          |
| CD19+             | 0.97 (0.83–1.14)              | 0.97 (0.82–1.14)            |
| CD3+              | 1.00 (0.91–1.09)              | 1.01 (0.93–1.11)            |
| CD3+ CD62L-       | 0.99 (0.93–1.05)              | 1.00 (0.94–1.07)            |
| CD3+ CD16+/56+    | 1.01 (0.87–1.19)              | 0.98 (0.84–1.16)            |

* Adjusted for baseline viral load (log10 transformed).

### TABLE 4. Comparison of relative counts of selected immunophenotypic markers of activation and differentiation among perinatally exposed infants by HIV infection status and by treatment arm

| Marker combination | Uninfected mean (SE), % | Infected mean (SE), % | Percent difference | p  |
|--------------------|-------------------------|-----------------------|-------------------|----|
| CD3+ CD4+         | 46.56 (0.53)            | 34.40 (3.21)          | 12.16             | 0.0002 |
| CD4+ CD45 RA-     | 33.03 (0.51)            | 24.38 (3.11)          | 8.65              | 0.006  |
| CD4+ DR-          | 1.76 (0.06)             | 1.74 (0.47)           | 0.02              | 0.97  |
| CD4+ CD45 RA- DR+ | 0.96 (0.04)             | 0.70 (0.32)           | 0.26              | 0.41  |
| CD3+ CD8-         | 17.24 (0.37)            | 26.33 (2.28)          | −9.09             | 0.0001 |
| CD8+ CD45 RA+     | 15.17 (0.33)            | 15.68 (1.99)          | −0.51             | 0.80  |
| CD8+ DR-          | 1.74 (0.15)             | 9.67 (0.98)           | −7.93             | 0.0001 |
| CD8+ CD45 RA- DR+ | 1.04 (0.08)             | 2.27 (0.47)           | −1.23             | 0.011  |
| CD8+ CD28-        | 11.03 (0.29)            | 13.83 (1.92)          | −2.80             | 0.15  |
| CD8+ CD38- DR+    | 0.51 (0.05)             | 3.15 (0.29)           | −2.64             | 0.0001 |
| CD19+             | 7.56 (0.27)             | 6.07 (1.65)           | 1.49              | 0.37  |
| CD19+ CD23+ CD62L+| 5.14 (0.24)             | 4.30 (1.49)           | 0.83              | 0.58  |

| HIVIG mean (SE), % | IVIG mean (SE), % | Percent difference | p  |
|--------------------|-------------------|-------------------|----|
| 45.90 (0.75)       | 46.56 (0.75)      | −0.65             | 0.54 |
| 31.58 (0.70)       | 33.89 (0.70)      | −2.31             | 0.02 |
| 1.78 (0.08)        | 1.75 (0.08)       | 0.03              | 0.88 |
| 0.97 (0.06)        | 0.95 (0.06)       | 0.01              | 0.89 |
| 17.46 (0.54)       | 18.02 (0.53)      | −0.56             | 0.46 |
| 14.89 (0.45)       | 15.57 (0.45)      | −0.68             | 0.29 |
| 2.05 (0.25)        | 1.99 (0.25)       | 0.05              | 0.88 |
| 1.23 (0.12)        | 1.05 (0.12)       | 0.17              | 0.32 |
| 11.31 (0.41)       | 11.32 (0.40)      | −0.01             | 0.99 |
| 21.99 (0.62)       | 20.95 (0.63)      | 1.07              | 0.22 |
| 7.43 (0.37)        | 7.53 (0.37)       | −0.11             | 0.84 |
| 4.91 (0.34)        | 5.25 (0.33)       | −0.34             | 0.48 |

* A repeated measures longitudinal model (SAS PROC MIXED) was used to estimate the mean proportions (and standard error [SE]), the overall percentage difference and test for differences between groups.

* The percent difference was calculated based on the mean proportion for the nontransmitters minus the mean proportion for transmitters.

* The percent difference was calculated based on the mean proportion for the HIVIG arm minus the mean proportion for IVIG arm.
While specific immunological effects of HIVIG in pregnancy have not been studied, IVIG treatment in pregnancy does not appear to affect levels of CD3+CD8+ or CD3+ lymphocytes (17, 30), but does reduce peripheral blood NK cells (15). IVIG appears to exert its therapeutic effects in autoimmune and alloimmune disorders by down-regulation of costimulatory molecules associated with modulation of cytokine secretion, with resultant inhibition of autoreactive and alloreactive T-cell activation and proliferation (2); by reduction of intact B cell and monocyte cell numbers, modulation of surface molecule expression on B cells, and deletion of B and T cells by apoptosis with resultant inhibition of optimal T-cell activation (34); and by inhibition of cytokine-induced NF-κB activation and blocking of FcγRIII on cell membranes (11, 12).

Our study also showed differences in markers of lymphocyte activation and differentiation in children born to HIV-infected women, and that these markers appear to distinguish infected from uninfected infants. One of the challenges in the interpretation of CD4+ and CD8+ T-cells in children is that changes may represent both a maturational effect (changing with increasing age), as well as an immune perturbation effect secondary to the presence of HIV. Gallagher et al. evaluated HIV infected and uninfected infants, and followed changes in CD4+ and CD8+ cells similar to this study (9). They found that both infected and uninfected infants had a decline in the percentages and numbers of CD4+ cells beginning at two weeks of age, and that the decline was greater in the HIV infected group. Activation and differentiation of CD8+ T-cells in the HIV-infected infants were demonstrated by a significant increase in CD45RA−/CD45RO+ CD8+ cells by six weeks of age (see also reference 1) and by increases in CD8+ S6F1CD3+ cells and HLA-DR+CD38+ CD8+ cells by two weeks of age. Rich et al. have also reported that infants with positive HIV cultures at day seven of life had a higher percentage of CD8+DR+ T-cells (suggesting intrauterine transmission) versus those who are infected intrapartum (27). The data from these studies suggest that although there are differences in T-cell subsets between HIV-infected and uninfected infants, the ability to discriminate using detailed immunophenotyping is not feasible before six weeks of age for any of the parameters evaluated. Thus, detailed lymphocyte subset evaluation could be used as an adjunct to the diagnosis of HIV infection in children, and possibly for estimating the risk of disease progression or response to treatment of these children. However, longer follow-up of children who are not on antiretroviral therapy would be needed to assess this possibility.

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