

674 Association between Immune Activation and HIV-1 RNA Genital Tract Shedding



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Background

HIV-1 RNA is the most significant determinant of cervical HIV-1 shedding. Shedding has also been related to STDs and cervical inflammation. The mechanism by which this occurs is poorly understood. There is evidence that systemic immune activation promotes viral entry and replication, and HIV disease progression. However, the impact of systemic immune activation (CD8+CD38+DR+ and CD4+CD38+DR+) on local genital HIV shedding has not been characterized.

Objectives

To assess for differences in immune activation in relation to HIV RNA levels in the genital tract.
 To determine which factors contribute to immune activation and higher genital HIV RNA levels.

Materials and Methods

Design: Prospective cohort study

Subjects: Subjects were a subset of 225 HIV infected women enrolled in WIHS. Women were selected who had paired plasma and cervicovaginal (CVL) specimens available for evaluation.

Procedures: Clinical and lab evaluations were performed at study entry and at 6-month intervals. Demographic, virologic, behavioral, and clinical characteristics were obtained at the first visit at which genital HIV RNA and immune marker data were collected.

Lab Procedure: HIV-1 genital shedding was defined as ≥ 176 copies/ml (NucliSense). For plasma, the lower limit of detection was 4000 copies/ml. Immune activation was defined as CD8+CD38+DR+ and CD4+CD38+DR+. CD4 and CD8 subset analyses were performed real-time using standard flow cytometric techniques in laboratories that were certified by the NIAID Immunology Quality Assurance Program as previously reported. A subset of women were evaluated for expression of activation CD4+ and CD8+ T cells using either fresh whole blood collected in EDTA tubes using three-color flow cytometry or frozen PBMCs using three or four-color flow cytometry. The following fluorochrome-conjugated antibodies were used anti-(anti- CD3, -CD4, -CD8, -HLA-DR, -CD38, -CD45RO, -CD45RA, -IlgG.

Results

Table 1. Characteristics and Demographics

Characteristics	Subjects (n=225)
Age, years, median (range)	38.5 (25.0- 60.3)
Race	
White, n (%)	42 (18.6)
African-American, n (%)	125 (55.5)
Hispanic, n (%)	54 (24.0)
Other, n (%)	4 (2.0)
Marital Status	
Married/Living with Partner, n (%)	76 (33.7)
Divorced/Widowed/Separated, n (%)	77 (34.2)
Single/Other, n (%)	66 (29.3)
CD4 cell count, cells/uL, median (range)	373.5 (0- 1356.0)
Initial HIV RNA Viral Load, copies/ml median (range)	4,800 (80-610,000)
ARV Therapy	
None, n (%)	87 (38.6)
Mono Combo, n (%)	56 (24.8)
HAART, n (%)	81 (36.0)
Risk Factors	
IVDU, n (%)	85 (37.8)
Heterosexual contact, n (%)	98 (43.6)
Not identified, n (%)	41 (18.2)
Vaginal pH, median (range)	5.0 (3.5- 9.5)
HPV status	
≥ 1 subtype detected, n (%)	79 (35.1)
No subtypes detected, n (%)	70 (31.1)
Inflammation Present, n (%)	23 (10.2)
T. Vaginalis Present, n (%)	19 (8.4)

Table 2. Univariate association of immune markers and HIV-1 Genital Shedding

Immune Marker	OR	95% CI (n=225)	P-value
CD8+CD38+DR- <32 ≥ 32	1.00 0.59	(0.38, 0.91)	0.0170
CD8+CD38+DR+ <34 ≥ 34	1.00 3.17	(1.76, 5.69)	0.0001
CD8+CD38-DR- <17 ≥ 17	1.00 0.42	(0.24, 0.74)	0.003
CD8+CD38-DR+ <7 ≥ 7	1.00 0.49	(0.31, 0.76)	0.0015
CD4+CD38+DR- <49 ≥ 49	1.00 1.02	(0.67, 1.56)	0.92
CD4+CD38+DR+ <10 ≥ 10	2.83 1.00	(1.74, 4.62)	<0.0001
CD4+CD38-DR- <28 ≥ 28	1.00 0.38	(0.24, 0.62)	<0.0001
CD4+CD38-DR+ <6 ≥ 6	1.00 0.92	(0.62, 1.36)	0.67

Table 3. Multivariate association of Immune markers with HIV-1 Genital Shedding

Immune Markers	Adjusting for CD4 cell count			Adjusting for HIV RNA		
	OR	95% CI	P-value	OR	95% CI	P-value
CD8+CD38-DR- <32 ≥ 32	1.00 0.82	(0.52, 1.31)	0.41	1.00 0.83	(0.53, 1.30)	0.41
CD8+CD38+DR+ <34 ≥ 34	1.00 1.94	(1.01, 3.75)	0.047	1.00 1.68	(0.92, 3.06)	0.09
CD8+CD38-DR- <17 ≥ 17	1.00 0.53	(0.28, 1.00)	0.050	1.00 0.63	(0.31, 1.27)	0.19
CD8+CD38-DR+ <7 ≥ 7	1.00 0.61	(0.38, 0.98)	0.041	1.00 0.71	(0.42, 1.19)	0.19
CD4+CD38-DR- <49 ≥ 49	1.00 1.51	(0.95, 2.38)	0.08	1.00 1.24	(0.77, 1.98)	0.38
CD4+CD38+DR+ <10 ≥ 10	1.00 1.72	(1.01, 2.94)	0.0470	1.00 1.66	(0.97, 2.83)	0.06
CD4+CD38-DR- <28 ≥ 28	1.00 0.48	(0.29, 0.79)	0.004	1.00 0.51	(0.30, 0.87)	0.013
CD4+CD38-DR+ <6 ≥ 6	1.00 0.77	(0.49, 1.20)	0.24	1.00 0.92	(0.60, 1.42)	0.71

Adjusted for ARV therapy, genital infection and CD4 or HIV Viral load

•225 HIV + women had approximately 550 genital evaluations over the course of the study, accounting for 157 shedding visits. Clinical and demographic characteristics are described in Table 1.

•Immune markers of interest were analyzed in both univariate and multivariate models.

•Systemic Immune activation markers, CD8+CD38+DR+ and CD4+CD38+DR+ were univariately associated with HIV genital shedding. (Table 2)

•In the multivariate model (table 3) adjusting for CD4 cell count and other cofactors, genital HIV shedding was associated with increased systemic immune activation:

- Increased CD8+CD38+DR+ OR 1.9 95% CI 1.0-3.8
- Increased CD4+CD38+DR+ OR 1.7 95% CI 1.0- 2.9

•Lack of systemic immune activation was associated with decreased likelihood of HIV genital shedding:

- Increased CD4+CD38-DR- OR 0.5 95% CI 0.3- 0.8
- This held true when adjusting for HIV viral load; women with higher levels of CD4+CD38-DR-, were less likely to have HIV genital shedding.

Conclusions

1. Systemic immune activation is significantly associated with HIV RNA-1 genital shedding
2. Lack of systemic immune activation was protective for HIV RNA-1 genital shedding
3. The pathophysiologic mechanism of HIV genital shedding may be related to systemic immune activation.
4. Systemic immune activation may enhance local HIV RNA-1 replication in genital secretions and thus increase transmission and acquisition of HIV infection.

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