

Lifetime sexual violence exposure in women compromises systemic innate immune mediators associated with HIV pathogenesis: A cross-sectional analysis

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Abstract

Objectives: Violence and HIV/AIDS syndemic highly prevalent among women impairs HIV prevention efforts. Prolonged exposure to violence results in physical trauma and psychological distress. Building on previous findings regarding genital immune dysregulation following sexual abuse exposure, we investigate here whether systemic changes occur as well.

Methods: Using the Women's Interagency HIV Study repository, 77 women were stratified by HIV serostatus and categorized into four subgroups: (1) no sexual abuse history and lower depression score (Control); (2) no sexual abuse history but higher depression score (Depression); (3) high sexual abuse exposure and lower depression score (Abuse); (4) high sexual abuse exposure and higher depression score (Abuse + Depression). Inflammation-associated immune biomarkers (TNF- α , IL-6, IL-1 α , IL-1 β , TGF- β , MIP-3 α , IP-10, MCP-1, and Cathepsin-B) and anti-inflammatory/anti-HIV biomarkers (Secretory leukocyte protease inhibitor, Elafin, human beta-defensin-2 (HBD-2), alpha-defensins 1-3, Thrombospondin, Serpin-A1, and Cystatin-C) were measured in plasma using enzyme-linked immunosorbent assay. Within each HIV serostatus, differences in biomarker levels between subgroups were evaluated with Kruskal–Wallis and Dunn's test with Bonferroni correction. Spearman correlations between biomarkers were assessed for each subgroup.

Results: Compared to the Control and Depression groups, Abuse + Depression was associated with significantly higher levels of chemokines MIP-3 α and IP-10 ($p < 0.01$) and lower levels of inflammatory cytokine IL-1 β ($p < 0.01$) in the HIV-uninfected population. Human beta-defensin-2 was lowest in the Abuse + Depression group ($p < 0.05$ versus Depression). By contrast, among HIV-infected, Abuse and Abuse + Depression were associated with lower levels of MIP-3 α ($p < 0.05$ versus Control) and IP-10 ($p < 0.05$, Abuse versus Control). Inflammatory cytokine IL-6 was higher in both Abuse groups ($p < 0.05$ versus Control), while Elafin was lowest in the Abuse + Depression group ($p < 0.01$ versus Depression).

Conclusion: We report compromised plasma immune responses that parallel previous findings in the genital mucosa, based on sexual abuse and HIV status. Systemic biomarkers may indicate trauma exposure and impact risk of HIV acquisition/transmission.

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Introduction

The epidemics of violence against women (VAW) and HIV/AIDS act synergistically to adversely impact women's health (reviewed).^{1–3} Globally, lifetime prevalence of violence in women ranges from 6% to 59% with higher percentages in areas of the world where women are disproportionately affected by HIV/AIDS.⁴ Studies in US women as part of the Women's interagency HIV Study (WIHS) cohort report high percentage of abuse experience associated with increased risk of acquisition of sexually transmitted infections (STI).^{5,6} Recognizing that unhealthy social conditions impart stress on vulnerable populations and expose them to disease clusters, syndemic theory examines the spread of disease in the context of interrelated biological, psychological, social, and political factors.⁷ Exposure to violence can significantly increase the risk of HIV acquisition/transmission in women due to inability to negotiate safe sex, refuse unwanted sex, fear of disclosure of their status, and access to antiretroviral drugs.^{8–14} Whereas it is established that violence exposure can lead to severe psychological stress and depression, there exists a paucity of information regarding systemic immunological dysregulation in women exposed to violence that may increase their risk of acquiring or transmitting HIV.

VAW includes physical, emotional, and sexual violence and can be committed by partners (intimate partner violence, IPV) or non-partners (domestic violence). All forms are associated with negative impact on health and well-being, individually and in combination.¹⁵ Whereas experiencing violence can be an isolated event, long-term exposure to any type of violence is defined as abuse, with cumulative (or chronic) lifetime exposure being highly predictive of worse health outcomes beyond that accounted for by a single type of event.^{15,16} Exposure to abuse-associated stressors early in life has been associated with the development of cardiovascular complications and high morbidity in women later in their lives.^{17–20} In addition, co-occurrence of childhood sexual abuse, adult sexual abuse, IPV, and sexual harassment has been shown to be predictive of post-traumatic stress disorder (PTSD).¹⁶ In HIV-infected women, exposure to violence has been associated with faster rates of disease progression.⁸

Psychological stress or perception of stress associated with violence/abuse can impact the immune system in a complex manner.^{21,22} Studies have reported higher ratios and greater activation of CD4+ and CD8+ T cells, lower activity of natural killer (NK) cells, compromised immune

responses to viral infections, vaccines, and delayed wound-healing in women exposed to abuse.^{21,23–29} In a healthy functioning immune system, acute short-term stress (for example, from wound or infection), typically results in a protective immune response. This is characterized by a tightly regulated inflammatory response followed by the activation of the hypothalamic–pituitary–adrenal (HPA) axis and release of cortisol, the primary glucocorticoid in humans, which then downregulates the inflammation and returns the immune response to baseline.³⁰ However, prolonged stress, (as in the case for concurrent or cumulative exposure to multiple types of abuse), can result either in a chronic inflammatory phenotype or a chronic immunosuppressive phenotype, both of which are detrimental to the health of the individual.^{31–33} A study by Dhabar and McEwen reported enhanced immune response in a rat model when acute stress was administered prior to antigenic challenge.³⁴ However, the same study also reported, when subjected to chronic stress, the immune response was significantly dampened and was correlated with attenuated glucocorticoid response.³⁴

Co-occurrence of elevated levels of psychological stress and depressive symptoms associated with violence/abuse is known to dysregulate the immune system.³⁵ Depression is a disorder of both immune activation and immune suppression.³⁶ Depressive symptoms have not only been associated with higher levels of inflammatory cytokines^{37–39} but also decreased functionality of T cells and NK cells.³⁶ Furthermore, chronic stress and inflammation have been associated with depression and linked to dysregulation of the HPA axis and resistance to cortisol.³⁶ Although causal links between depression and immunity are still unclear, the relationship is clinically relevant. A recent systemic review by Kappelmann et al.⁴⁰ indicates that treatment to suppress inflammatory cytokines can reduce depressive symptoms. In HIV-infected women where exposure to violence/abuse is high,^{12,41} depression is highly prevalent and associated with increased substance abuse, decreased adherence to antiretroviral therapy, chronic inflammation/immune activation, faster disease progression, and increased mortality.^{42–48}

We have previously reported lifelong sexual abuse exposure to be associated with increased inflammation-associated cytokine/chemokine expression and impaired wound-healing pathways in genital tract samples selected from the WIHS repository.⁴⁹ Building on those findings and understanding that sexual abuse markedly affects several psychological factors, we ask here whether systemic changes occur that parallel genital tract responses to

violence. Our working hypothesis is that women with a history of chronic depression and repeated sexual violence exposure, compared to women with no sexual violence exposure and no depression, will have alterations in their systemic and genital immune environment that predispose them to an increased risk of acquiring/transmitting HIV. These studies are essential to determine if blood contains surrogate markers for chronic genital trauma and whether HIV-associated immune mediators are impacted in this population.

Methods

Ethical statement

The WIHS protocol and this study were conducted according to the principles expressed in the Declaration of Helsinki. All sites with direct participant contact received approval by the respective participating institution's review board. Study staff at each site obtained written informed consent for the collection and use of data and specimens from each research participant including consent to the future use of their data and their specimens in the repository. The IRB approval number for the Washington DC WIHS site is 1993-007. The George Washington University (GWU) site only received existing, fully de-identified, data and samples with no access to code link. Therefore, it was determined to not meet the definition of human subjects' research by The GWU IRB.

Study participants and demographics

WIHS is an ongoing prospective observational cohort study of HIV-infected and socio-demographically matched uninfected women in the United States. Study methods, baseline cohort characteristics, and long-term retention have been previously described.^{50–52}

For this cross-sectional study, we included participants who enrolled in the WIHS during either 1994–1995 or 2001–2002 and had complete baseline and longitudinal childhood and adult abuse data.⁵³ Participants were stratified by HIV serostatus and categorized based on their self-reported sexual abuse histories. Lifetime chronic sexual abuse group was defined by those with the highest level of sexual abuse exposure, past and current (childhood and adult sexual abuse including any reported transactional sex). The comparison control group was selected from those who reported no sexual abuse. As depression can be comorbid and a potential confounder in this population, we further stratified based on the level of self-reported depressive symptoms at the time of visit (CES-D scale).^{54,55} Using the clinically significant cut-off of 16, scores were categorized as indicating low (<16) or high (>16) depression.

A total of 77 women were selected to form four HIV-infected and four HIV-uninfected groups (n=8–11 per

group) in the following categories: (1) no sexual abuse history and lower depressive symptom score (Control); (2) no sexual abuse history but higher depression score (Depression); (3) chronic sexual abuse exposure and lower depression score (Abuse); and (4) chronic sexual abuse exposure and higher depression score (Abuse + Depression). Plasma viral load and CD4 counts in HIV-infected women were obtained from WIHS database. All HIV-infected women were on highly active antiretroviral therapy (HAART) at the sampling visit. Data on genital immune biomarkers have been previously published.⁴⁹ In this sub-study, we analyzed plasma immune biomarkers from the same cohort.

Measurement of cytokines, chemokines, and anti-HIV mediators in plasma

Immune mediators that have been defined to play a role in HIV infection and pathogenesis were selected based on published literature. Plasma samples from visits that matched our inclusion/exclusion criteria⁴⁹ were selected from the WIHS repository and stored at -80°C until assayed. We performed enzyme-linked immunosorbent assay (ELISA) for cytokines: TNF- α , IL-6, IL-1 α , IL-1 β , TGF- β ; chemokines: MIP-3 α , IL-8, MCP-1, IP-10; anti-inflammatory/antiproteases with HIV inhibitory activity: secretory leukocyte protease inhibitor (SLPI), Elafin, human beta-defensin 2 (HBD-2), human alpha-defensins 1-3 (HNP 1-3), Thrombospondin (TSP-1), Serpin A1, and Cystatin-C; endogenous protease, Cathepsin-B. All except HBD-2 were purchased from R&D Systems (Minneapolis, MN), and assays were performed according to the manufacturer's protocols. HBD-2 was assayed using an ELISA test kit from PeproTech (Rocky Hill, NJ). All immune mediators were quantified based on standard curves obtained using a Microplate Reader (Biotek, Winooski, VT). All biomarkers were analyzed in triplicate. Biomarker concentrations below the lower limit of detection were reported as the mid-point between the lowest concentration measured and zero,⁵⁶ after which concentrations were log₁₀ transformed.

Power analysis

An a priori power analysis was conducted for detecting mean differences between each pair of groups using two-sample *t* tests with alpha=0.20 (two-sided), unadjusted for multiple comparisons. This yielded n=10 per group to detect a standardized effect size of 1.0 with 80% power. The high alpha of 0.20 was chosen to err on the side of avoiding Type II errors (false negative results) for this preliminary investigation. At the analysis stage, because of the large number of parameters being tested, it was decided that it would be more prudent to reduce the Type I error rate modestly using alpha=0.05 and the screening procedure described below.

Table 1. Levels of soluble immune biomarkers in plasma comparing HIV-uninfected women with history of abuse, depression, or both to controls.

HIV-uninfected					
Variable (Log ₁₀ pg/mL)	Percent detectable	Control median (IQR)	Depression median (IQR)	Abuse median (IQR)	Abuse + depression median (IQR)
N		10	11	10	8
TNF- α	7.7	-1.60 (-1.60, -1.60)	-1.60 (-1.60, -1.60)	-1.60 (-1.60, -1.60)	-1.60 (-1.60, -1.60)
IL-6	12.8	-1.01 (-1.01, -1.01)	-1.01 (-1.01, -1.01)	-1.01 (-1.01, -0.79)	-1.01 (-1.01, -1.01)
IL-1 α	0.0	0.42 (0.42, 0.42)	0.42 (0.42, 0.42)	0.42 (0.42, 0.42)	0.42 (0.42, 0.42)
IL-1 β	74.4	0.85 (0.80, 0.87)	0.83 (0.78, 0.87)	0.75 (-0.18, 0.78)	-0.48 (-0.48, -0.48) ***††
TGF- β	35.9	-0.48 (-0.48, -0.48)	-0.48 (-0.48, 1.98)	-0.48 (-0.48, 1.97)	1.37 (-0.48, 2.07)
MIP-3 α	30.8	-0.67 (-0.67, -0.67)	-0.67 (-0.67, -0.67)	0.68 (-0.67, 2.10)	2.07 (2.04, 2.08) ***†††
IL-8	25.6	0.38 (0.38, 0.38)	0.38 (0.38, 2.06)	0.38 (0.38, 1.89)	0.38 (0.38, 0.38)
MCP-1	35.9	0.02 (0.02, 0.02)	0.02 (0.02, 1.92)	0.02 (0.02, 1.77)	1.38 (0.02, 1.70)
IP-10	30.8	1.24 (1.24, 1.24)	1.24 (1.24, 1.24)	1.24 (1.24, 1.93)	2.00 (1.91, 2.04) ***†††
SLPI	100.0	4.82 (4.75, 4.89)	4.81 (4.70, 4.97)	4.77 (4.71, 4.84)	4.87 (4.82, 4.95)
Elafin	100.0	4.35 (4.28, 4.39)	4.40 (4.24, 4.44)	4.27 (4.23, 4.29)	4.28 (4.21, 4.41)
HBD-2	74.4	2.57 (2.48, 2.79)	2.59 (2.54, 2.63)	2.55 (1.54, 2.90)	1.23 (1.23, 1.23) †
HNPI-3	100.0	3.68 (3.59, 3.90)	3.81 (3.70, 4.04)	4.01 (3.72, 4.16)	3.88 (3.77, 4.04)
TSP-1	100.0	2.55 (2.11, 3.31)	3.31 (2.83, 3.38)	3.22 (3.03, 3.43)	3.23 (2.76, 3.39)
Serpin A1	100.0	8.92 (8.90, 8.97)	8.88 (8.79, 8.94)	8.90 (8.81, 8.92)	8.87 (8.81, 8.89)
Cystatin-C	100.0	5.73 (5.63, 5.81)	5.66 (5.56, 5.79)	5.62 (5.52, 5.64)	5.51 (5.46, 5.68)
Cathepsin-B	100.0	4.39 (4.35, 4.56)	4.64 (4.55, 4.67)	4.53 (4.46, 4.61)	4.57 (4.44, 4.68)

SLPI: secretory leukocyte protease inhibitor. HBD2: human beta-defensin-2. TSP-1: thrombospondin-1. HNPI-3: human alpha defensin 1-3. Percent detectable column shows percentage of samples with detectable levels of biomarker. All comparisons based on pairwise Dunn's test with Bonferroni correction. Significant p values are denoted in bold: ***p < 0.001; **p < 0.01; *p < 0.05 versus Control; †††p < 0.001; ††p < 0.01; †p < 0.05 versus Depression.

Statistical analysis

Within each HIV serostatus stratum, biomarker concentrations were compared across the four exposure groups. When performing significance tests for a large number of biomarkers, especially with a modest sample size, it is possible that some results will not be reproducible. To reduce this likelihood, we used a two-step approach. First, we compared biomarker concentrations across all four exposure groups using the Kruskal-Wallis test. Only if this result was statistically significant ($\alpha < 0.05$), we followed with pairwise Dunn's tests, Bonferroni-corrected for six comparisons per biomarker.

In addition, we measured Spearman correlations between each pair of biomarkers, stratified by analysis group. All analyses were conducted in R version 4.0.3.

Results

Differences in systemic immune mediators by abuse or depression status in HIV-uninfected women

A total of 77 samples were analyzed (n=8–11 per group). Baseline cohort characteristics and long-term retention have been previously described.^{50–52}

Median log levels of two chemokines known to attract HIV target cells, MIP-3 α and IP-10, were highest in the Abuse + Depression group ($p < 0.01$ versus Control, Depression) and mostly undetectable in Control and Depression groups, with intermediate concentrations in the Abuse-only group (Table 1, Figures 1(a) and (b)). In contrast, the pro-inflammatory cytokine IL-1 β was mostly undetectable in the Abuse + Depression group ($p < 0.01$ versus Control, Depression) with higher levels in all other groups (Table 1, Figure 1(c)). Among the anti-inflammatory/anti-HIV mediators, HBD-2 levels were mostly undetectable in Abuse + Depression group and higher in other groups, but the only statistically significant pairwise difference was between Depression and Abuse + Depression ($p < 0.05$) (Table 1, Figure 1(d)). Pro-inflammatory cytokine IL-1 α was undetectable in all samples, and TNF- α and IL-6 were detectable only in 8% and 13% of samples, respectively.

Differences in systemic immune mediators by abuse or depression status in HIV-infected women

In HIV-infected women, the relative concentrations of biomarkers followed a different pattern. MIP-3 α was lower in the Abuse ($p < 0.05$ versus Control, Depression) and Abuse + Depression ($p < 0.05$ versus Control) groups

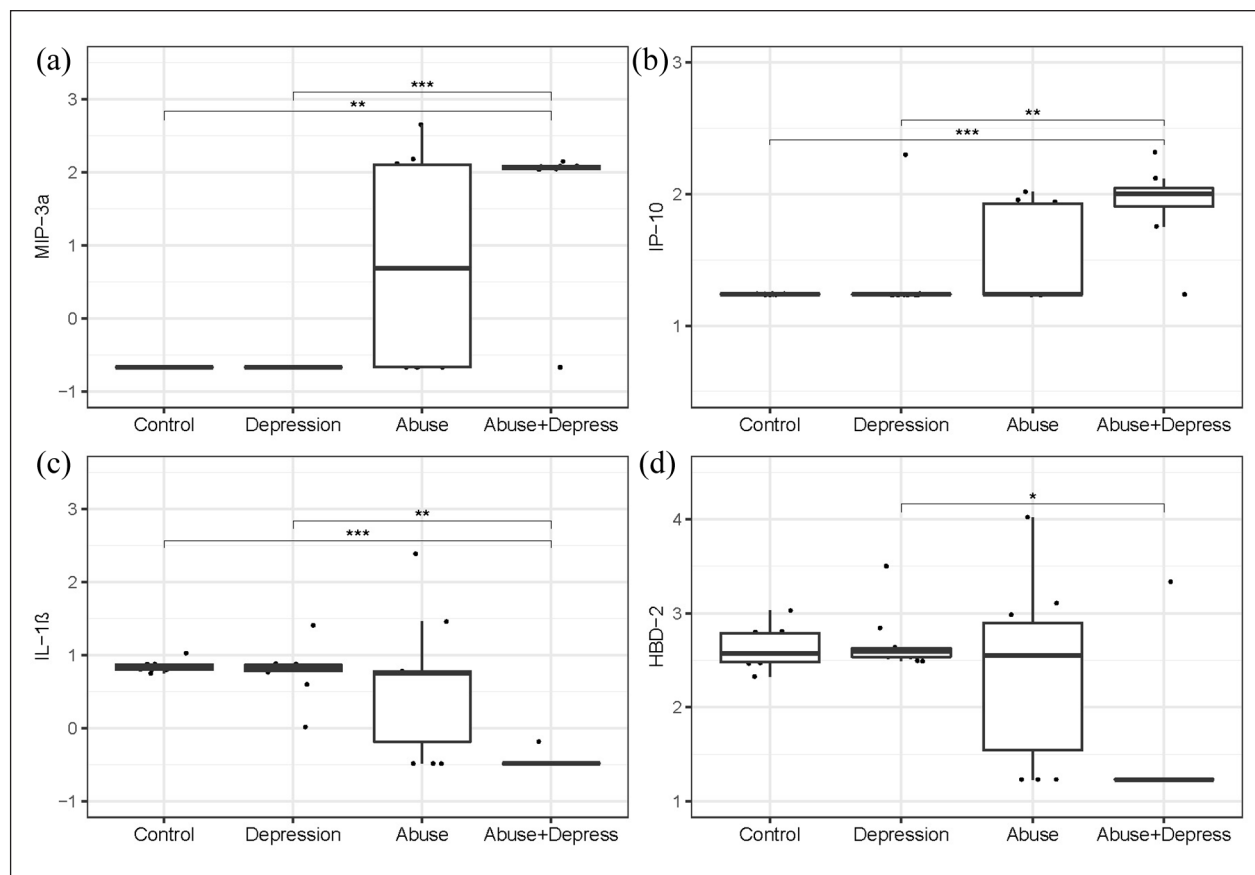


Figure 1. Differences in systemic immune mediators by abuse status in HIV-uninfected women.

Plasma from control women with no history of sexual abuse or depression ($n = 10$), women with current depression ($n = 11$), women with history of chronic sexual abuse ($n = 10$), and women with the history of chronic sexual abuse plus current depression ($n = 8$) were tested for levels of (a) MIP-3 α , (b) IP-10, (c) IL-1 β , and (d) HBD-2, by standard ELISA assays. Bars depict median, in log pg/mL of protein. Asterisks indicate significant p values as follows: *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$.

(Table 2, Figure 2(a)), the reverse of what was seen for uninfected women. IP-10 followed a similar pattern, but the adjusted pairwise difference was significant only for the Abuse group ($p < 0.05$ versus Controls) (Table 2, Figure 2(b)). In contrast, levels of pro-inflammatory cytokine IL-6 were significantly higher in the Abuse ($p < 0.05$) and Abuse + Depression ($p < 0.001$) groups compared to Controls, all of whom had undetectable levels (Table 2, Figure 2(c)). For the anti-inflammatory/anti-HIV mediator Elafin, samples from Control, Depression, and Abuse groups had higher levels of Elafin compared to Abuse + Depression, but the difference was statistically significant only between Depression and Abuse + Depression ($p < 0.01$) (Table 2, Figure 2(d)). As with the HIV-uninfected women, IL-1 α was undetectable in all samples, and TNF- α and IL-1 β were detectable in only 5% of samples each.

Compared to the HIV-uninfected, HIV-infected samples had higher detectable levels of the pro-inflammatory cytokine IL-6 (55% versus 13%), three chemokines, MIP-3 α (71% versus 31%), MCP-1 (58% versus 36%), IP-10 (53% versus 31%), and lower detectable levels of cytokines

IL-1 β (5% versus 74%), TGF- β (13% versus 36%), chemokine IL-8 (11% versus 26%), and antimicrobial/anti-HIV Elafin (76% versus 100%) and HBD-2 (34% versus 74%) (Tables 1 and 2).

Differences in immune biomarker (cytokine/chemokine/antimicrobial) network

We conducted Spearman correlation analyses to evaluate clustering between inflammatory and anti-inflammatory immune biomarkers by depression, abuse, and HIV status. However, unlike our previous findings in the genital tract,⁴⁹ no significant clustering patterns among the groups were observed in the plasma samples (data not shown).

Discussion

Syndemic theory seeks to understand the biological, psychological, social, and political interactions that influence the spread of disease.⁷ Here we examined the biological mechanisms by which physical and psychological stress resulting from long-term sexual abuse and depression

Table 2. Levels of soluble immune biomarkers in plasma comparing HIV-infected women with history of abuse, depression, or both to controls.

HIV-infected					
Variable (Log ₁₀ pg/mL)	Percent detectable	Control median (IQR)	Depression median (IQR)	Abuse median (IQR)	Abuse + depression median (IQR)
N		10	10	8	10
TNF- α	5.3	-1.60 (-1.60, -1.60)	-1.60 (-1.60, -1.60)	-1.60 (-1.60, -1.60)	-1.60 (-1.60, -1.60)
IL-6	55.3	-1.01 (-1.01, -1.01)	-0.56 (-1.01, 0.79)	0.45 (-0.06, 1.36)*	0.68 (0.61, 0.82)***
IL-1 α	0.0	0.42 (0.42, 0.42)	0.42 (0.42, 0.42)	0.42 (0.42, 0.42)	0.42 (0.42, 0.42)
IL-1 β	5.3	-0.48 (-0.48, -0.48)	-0.48 (-0.48, -0.48)	-0.48 (-0.48, -0.10)	-0.48 (-0.48, -0.48)
TGF- β	13.2	-0.48 (-0.48, 1.74)	-0.48 (-0.48, -0.48)	-0.48 (-0.48, -0.48)	-0.48 (-0.48, -0.48)
MIP-3 α	71.1	2.09 (2.05, 2.13)	2.09 (1.96, 2.12)	-0.67 (-0.67, 1.14)**†	0.72 (-0.67, 1.85)*
IL-8	10.5	0.38 (0.38, 0.38)	0.38 (0.38, 0.38)	0.38 (0.38, 0.52)	0.38 (0.38, 0.38)
MCP-1	57.9	1.82 (0.02, 1.88)	1.80 (0.29, 2.18)	0.02 (0.02, 1.91)	2.17 (0.02, 2.57)
IP-10	52.6	2.14 (2.06, 2.26)	1.96 (1.24, 2.13)	1.24 (1.24, 1.24)*	1.24 (1.24, 2.14)
SLPI	100.0	4.96 (4.87, 5.02)	4.89 (4.85, 5.00)	4.92 (4.88, 4.97)	5.00 (4.85, 5.03)
Elafin	76.3	4.19 (4.14, 4.43)	4.43 (4.27, 4.49)	3.56 (3.41, 4.50)	3.60 (3.41, 3.94)††
HBD-2	34.2	1.23 (1.23, 1.23)	1.23 (1.23, 2.39)	2.63 (1.23, 2.74)	1.38 (1.23, 3.47)
HNPI-3	100.0	3.77 (3.73, 3.86)	3.85 (3.73, 4.07)	3.79 (3.60, 4.06)	4.06 (3.82, 4.11)
TSP-1	100.0	2.59 (2.32, 3.19)	2.88 (2.51, 3.04)	3.05 (2.04, 3.21)	2.86 (2.73, 3.03)
Serpin-A1	100.0	8.81 (8.74, 8.83)	8.86 (8.79, 8.92)	8.79 (8.77, 8.86)	8.84 (8.77, 8.86)
Cystatin-C	100.0	5.79 (5.75, 5.84)	5.73 (5.68, 5.81)	5.62 (5.51, 5.78)	5.72 (5.59, 5.79)
Cathepsin-B	100.0	4.50 (4.44, 4.52)	4.46 (4.33, 4.60)	4.67 (4.42, 4.79)	4.51 (4.46, 4.55)

Percent detectable column shows percentage of samples with detectable levels of biomarker. SLPI: secretory leukocyte protease inhibitor; HBD2: human beta-defensin-2; TSP-1: thrombospondin-1; HNPI-3: human alpha defensin 1-3. All comparisons based on pairwise Dunn's test with Bonferroni correction. Significant p values are denoted in bold: ***p < 0.001; **p < 0.01; *p < 0.05 versus Control; ††p < 0.001; †p < 0.01; †p < 0.05 versus Depression.

contributes to the acquisition and spread of HIV. Following up on our previous findings on genital immune dysregulation in women exposed to lifetime chronic sexual violence, in this study, we evaluated systemic immune parameters in the same cohort. As there is a paucity of information regarding immune dysregulation and HIV risk in women experiencing violence, this study was designed as hypothesis-generating with the goal of screening immune biomarkers that are relevant to HIV immuno-pathogenesis in women. Of the 17 outputs analyzed, we found significant differences by abuse/depression status for four biomarkers in HIV-uninfected and four in HIV-infected, with two that were common to both groups. Our key findings point to specific and selective dysregulation of systemic immune mediators in those exposed to chronic abuse. Although we did not find statistically significant differences between Depression only and Controls, often, the most pronounced differences were noted in women with both abuse and depression risk factors. In addition, we found immune changes to be distinct based on HIV status of the individual.

Two potential immuno-biological pathways that may enhance HIV acquisition/transmission risk in this population are local immune changes in the genital mucosa^{23,49} and systemic immune changes as a function of chronic stress exposure. Exposure to psychological stressors has been shown to have a detrimental effect on immune

responses (and hence overall health), including poor responses to infections and vaccines.^{28,57-60} Depression, which frequently co-occurs with violence exposure, can interact with psychological stressors and has been linked to inflammation and immune dysfunction.^{36,61}

We observed a pattern of increased chemokines and reduced pro-inflammatory cytokines and anti-HIV mediators in HIV-uninfected women. Chemokines MIP-3 α and IP-10, positively associated with Abuse + Depression status, are potent chemo-attractors of immune cells, particularly activated T cells which are targets for HIV.⁶² Systemic upregulation of activated T cells has been reported in women experiencing IPV.^{27,29} Our study was not designed to evaluate the cellular immune activation but the increased IP-10 and MIP-3 α that we observed could potentially be associated with the activated immune phenotype observed by Kalokhe et al.²⁷ We also observed reduced pro-inflammatory cytokine IL-1 β to be significantly associated with Abuse + Depression status. Suppression of inflammatory pathways has been reported in conditions of chronic stress.^{31,34} In particular, IL-1 β has been reported to be associated with stress and depression.^{36,61,63} In HIV-infected women, the chemokine pattern was reversed, and reduced MIP-3 α and IP-10 were associated with Abuse and/or Abuse + Depression status. Abuse status was also associated with increased pro-inflammatory mediator IL-6, both with and without depression. As all the HIV-infected women in our cohort were on HAART at the

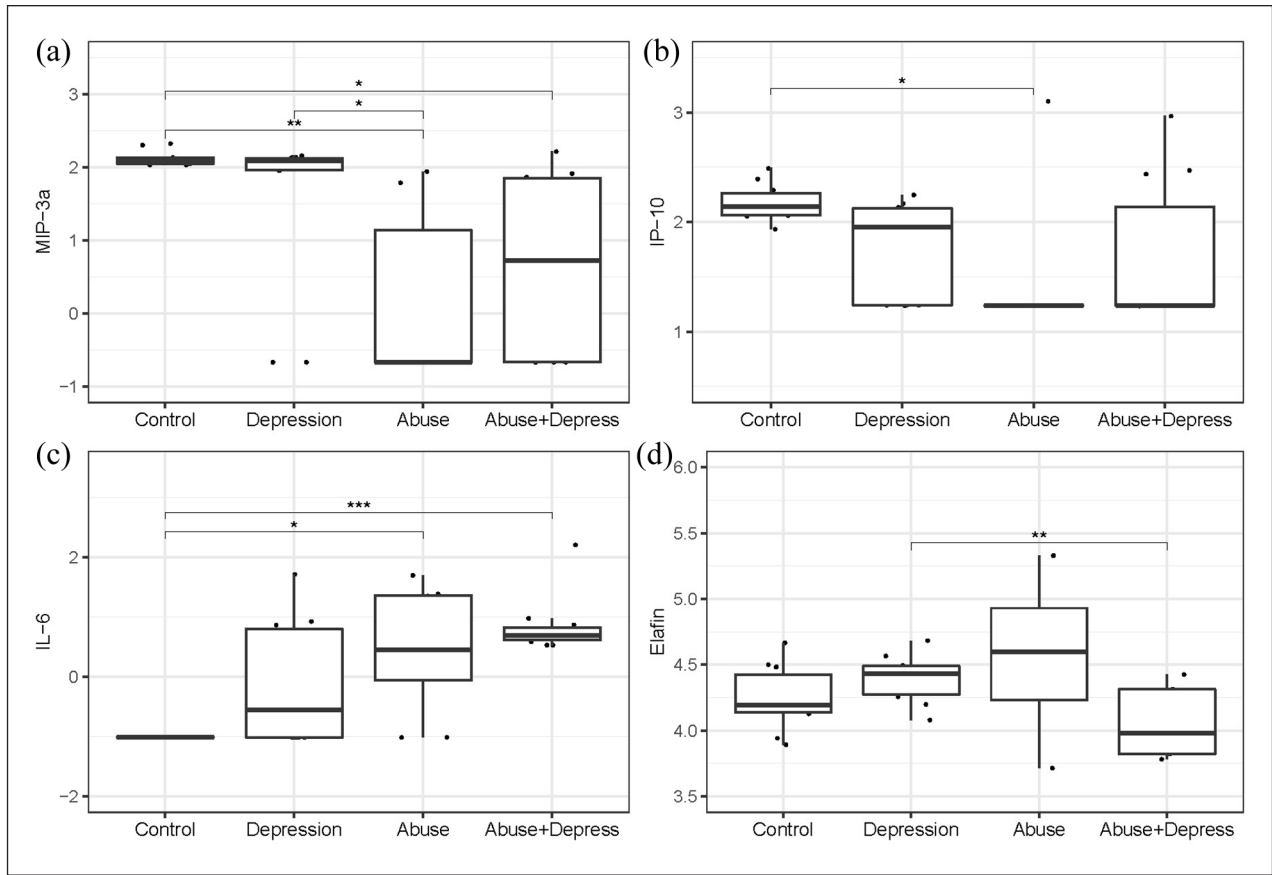


Figure 2. Differences in systemic immune mediators by abuse status in HIV-infected women.

Plasma from control women with no history of sexual abuse or depression ($n=10$), women with current depression ($n=10$), women with history of chronic sexual abuse ($n=8$), and women with history of chronic sexual abuse plus current depression ($n=10$) were tested for levels of (a) MIP-3 α , (b) IP-10, (c) IL-6, and (d) Elafin by standard ELISA assays. Bars depict median, in log pg/mL of protein. Asterisks indicate significant p values as follows: *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$.

time of sampling, it is unclear as to what extent the observed immune dysregulation may be attributable to selective immune modulation of the antiretroviral medications.⁶⁴⁻⁶⁶ In fact, we did observe distinct patterns of detectability of biomarkers in HIV-uninfected versus HIV-infected samples. Further studies are needed to understand the impact of HAART on immune modulation in women experiencing violence and depression.

In regard to depression status, we found that the levels of biomarkers were, in general, not significantly different from the Control group. However, we did observe significantly reduced anti-inflammatory/anti-HIV biomarkers HBD-2 (in HIV-uninfected samples) and Elafin (in HIV-infected samples) in Abuse + Depression compared to Depression only group. To our knowledge, plasma levels of these biomarkers have not been previously shown to be associated with depression. However, some studies indicate that the anti-inflammatory properties of HBD-2 can extend to the modulation of neuroimmune functions and neurodegeneration.⁶⁷

The effects of HIV infection in reorganizing immune networks into more rigid clustering have been previously

described.^{68,69} Our previous publication with this cohort demonstrated distinct clustering patterns in the genital tract.⁴⁹ However, we did not observe any significant patterns in plasma samples by depression, abuse, or HIV status.

Comparison of the findings in this study with the previously published analysis of genital immune dysregulation in the same cohort⁴⁹ indicates that whereas some immune biomarkers showed similar patterns of dysregulation in genital tract as in plasma, others were distinct. For example, in HIV-uninfected women, increase in chemokines that can attract HIV target cells (MIP-3 α and IP-10) was associated with exposure to chronic sexual abuse in both plasma and genital tract samples. In HIV-infected women, increased pro-inflammatory cytokine IL-6 and decreased chemokine MIP-3 α in both plasma and genital tract compartments were associated with chronic sexual abuse. This shows that certain immune biomarkers measured in the systemic compartment may function as surrogate markers for genital trauma. In contrast, clear compartmentalization was observed for other immune mediators. Whereas significant changes in IL-1 α and TNF- α in the genital tract

were associated with abuse exposure,⁴⁹ both biomarkers were mostly undetectable in plasma, irrespective of abuse, depression, or HIV status. The reduced pattern of anti-HIV mediators (HBD-2, Elafin) observed in plasma is a novel finding and was not observed in the genital tract. Although numerous publications to date have pointed to an association between violence exposure and increased HIV acquisition/transmission, women who experience sexual violence remain severely underrepresented in research studies with biological endpoints. As this was a pilot study, the sample size was small which prevented us from conducting extensive statistical analyses. However, we were able to identify important biomarker changes in women exposed to chronic sexual violence, which points to the need for future research in this arena. Furthermore, we were unable to control for some parameters including hormonal status (estrogen, progesterone, and cortisol), which can impact inherent variability of immune biomarkers, comorbidities (such as diabetes, hypertension), and overall health indicators (such as BMI). Other data on cohort characteristics, including socioeconomic status, smoking, alcohol use, and high-risk sexual behavior, have been reported in our previous publication.⁴⁹ Finally, the cross-sectional design of the study does not allow us to make causal inferences.

Conclusion

The strength of this study lies in the novel characterization of the plasma secretome in the context of chronic violence exposure in HIV-uninfected and -infected women. Although a handful of recent studies has evaluated cellular immune activation and suppression^{27,29} in women experiencing violence, to our knowledge, dysregulation in secreted immune biomarkers in plasma has not been reported. Building on our previous studies that demonstrated genital mucosal changes in response to sexual violence exposure, we show here that systemic changes also occur and may be further evaluated as surrogate markers of chronic genital trauma. Understanding how the immune system is affected by long-term exposure to sexual violence is likely to promote trauma-informed care and impact recommendations for post-exposure prophylaxis for HIV and STI in emergency care settings.^{70–72}

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Supplemental material

Supplemental material for this article is available online.

References

- Meyer JP, Springer SA and Altice FL. Substance abuse, violence, and HIV in women: a literature review of the syndemic. *J Womens Health* 2011; 20(7): 991–1006.
- Pyra M, Weber K, Wilson TE, et al. Sexual minority status and violence among HIV infected and at-risk women. *J Gen Intern Med* 2014; 29(8): 1131–1138.
- Pyra M, Weber KM, Wilson TE, et al. Sexual minority women and depressive symptoms throughout adulthood. *Am J Public Health* 2014; 104(12): e83–e90.
- World Health Organization. *Violence against women: a problem of epidemic proportions*. Geneva: WHO, 2013.
- Decker MR, Seage GR 3rd, Hemenway D, et al. Intimate partner violence perpetration, standard and gendered STI/HIV risk behaviour, and STI/HIV diagnosis among a clinic-based sample of men. *Sex Transm Infect* 2009; 85(7): 555–560.
- Geller RJ, Decker MR, Adedimeji AA, et al. A prospective study of exposure to gender-based violence and risk of sexually transmitted infection acquisition in the women's interagency HIV study, 1995–2018. *J Womens Health* 2020; 29(10): 1256–1267.
- Singer M, Bulled N, Ostrach B, et al. Syndemics and the biosocial conception of health. *Lancet* 2017; 389(10072): 941–950.
- Jewkes R, Dunkle K, Jama-Shai N, et al. Impact of exposure to intimate partner violence on CD4+ and CD8+ T cell decay in HIV infected women: longitudinal study. *PLoS ONE* 2015; 10(3): e0122001.
- Wingood GM, DiClemente RJ and Seth P. Improving health outcomes for IPV-exposed women living with HIV. *J Acquir Immune Defic Syndr* 2013; 64(1): 1–2.
- Gielen AC, O'Campo P, Faden RR, et al. Women's disclosure of HIV status: experiences of mistreatment and violence in an urban setting. *Women Health* 1997; 25(3): 19–31.
- Zierler S, Cunningham WE, Andersen R, et al. Violence victimization after HIV infection in a US probability sample of adult patients in primary care. *Am J Public Health* 2000; 90(2): 208–215.
- Cohen MH, Benekigeri C and Fabri MR. Gender-based violence and HIV in Rwanda: respecting women's voices. *J Womens Health* 2015; 24(9): 784–785.
- Durevall D and Lindskog A. Intimate partner violence and HIV in ten sub-Saharan African countries: what do the demographic and health surveys tell us. *Lancet Glob Health* 2015; 3(1): e34–e43.
- Li Y, Marshall CM, Rees HC, et al. Intimate partner violence and HIV infection among women: a systematic review and meta-analysis. *J Int AIDS Soc* 2014; 17: 18845.
- Scott-Storey K. Cumulative abuse: do things add up? An evaluation of the conceptualization, operationalization, and methodological approaches in the study of the phenomenon of cumulative abuse. *Trauma Violence Abuse* 2011; 12(3): 135–150.
- Campbell R, Greeson MR, Bybee D, et al. The co-occurrence of childhood sexual abuse, adult sexual assault, intimate partner violence, and sexual harassment: a mediational model of posttraumatic stress disorder and physical health outcomes. *J Consult Clin Psychol* 2008; 76(2): 194–207.
- Bertone-Johnson ER, Whitcomb BW, Missmer SA, et al. Inflammation and early-life abuse in women. *Am J Prev Med* 2012; 43(6): 611–620.
- Thurston RC, Chang Y, Derby CA, et al. Abuse and subclinical cardiovascular disease among midlife women: the study of women's health across the nation. *Stroke* 2014; 45(8): 2246–2251.
- Liu YZ, Wang YX and Jiang CL. Inflammation: the common pathway of stress-related diseases. *Front Hum Neurosci* 2017; 11: 316.
- Janusek LW, Tell D, Gaylord-Harden N, et al. Relationship of childhood adversity and neighborhood violence to a proinflammatory phenotype in emerging adult African American men: an epigenetic link. *Brain Behav Immun* 2017; 60: 126–135.
- Campbell JC, Baty ML, Ghandour RM, et al. The intersection of intimate partner violence against women and HIV/AIDS: a review. *Int J Inj Contr Saf Promot* 2008; 15(4): 221–231.
- Bauer ME, Wieck A, Lopes RP, et al. Interplay between neuroimmunoendocrine systems during post-traumatic stress disorder: a mini review. *Neuroimmunomodulation* 2010; 17(3): 192–195.

23. Tsuyuki K, Cimino AN, Holliday CN, et al. Physiological changes from violence-induced stress and trauma enhance HIV susceptibility among women. *Curr HIV/AIDS Rep* 2019; 16(1): 57–65.
24. Bauer ME. Chronic stress and immunosenescence: a review. *Neuroimmunomodulation* 2008; 15(4–6): 241–250.
25. Bicanic IA, Postma RM, Sinnema G, et al. Salivary cortisol and dehydroepiandrosterone sulfate in adolescent rape victims with post traumatic stress disorder. *Psychoneuroendocrinology* 2013; 38(3): 408–415.
26. Woods SJ, Wineman NM, Page GG, et al. Predicting immune status in women from PTSD and childhood and adult violence. *ANS Adv Nurs Sci* 2005; 28(4): 306–319.
27. Kalokhe AS, Ibegbu CC, Kaur SP, et al. Intimate partner violence is associated with increased Cd4+ T-cell activation among hiv-negative high-risk women. *Pathog Immun* 2016; 1(1): 193–213.
28. Garcia-Linares MI, Sanchez-Lorente S, Coe CL, et al. Intimate male partner violence impairs immune control over herpes simplex virus type 1 in physically and psychologically abused women. *Psychosom Med* 2004; 66(6): 965–972.
29. Swaims-Kohlmeier A, Haddad LB, Li ZT, et al. Chronic immune barrier dysregulation among women with a history of violence victimization. *JCI Insight* 2019; 4(10): 126097.
30. Malek H, Ebadzadeh MM, Safabakhsh R, et al. Dynamics of the HPA axis and inflammatory cytokines: insights from mathematical modeling. *Comput Biol Med* 2015; 67: 1–12.
31. Dhabhar FS. Enhancing versus suppressive effects of stress on immune function: implications for immunoprotection and immunopathology. *Neuroimmunomodulation* 2009; 16(5): 300–317.
32. Dhabhar FS and McEwen BS. Enhancing versus suppressive effects of stress hormones on skin immune function. *Proc Natl Acad Sci USA* 1999; 96(3): 1059–1064.
33. Takahashi A, Flanigan ME, McEwen BS, et al. Aggression, social stress, and the immune system in humans and animal models. *Front Behav Neurosci* 2018; 12: 56.
34. Dhabhar FS and McEwen BS. Acute stress enhances while chronic stress suppresses cell-mediated immunity in vivo: a potential role for leukocyte trafficking. *Brain Behav Immun* 1997; 11(4): 286–306.
35. World Health Organization. *Gender disparities in mental health*. Geneva: WHO, 2013.
36. Blume J, Douglas SD and Evans DL. Immune suppression and immune activation in depression. *Brain Behav Immun* 2011; 25(2): 221–229.
37. Pitychoutis PM and Papadopoulou-Daifoti Z. Of depression and immunity: does sex matter. *Int J Neuropsychopharmacol* 2010; 13(5): 675–689.
38. Zorrilla EP, Luborsky L, McKay JR, et al. The relationship of depression and stressors to immunological assays: a meta-analytic review. *Brain Behav Immun* 2001; 15(3): 199–226.
39. Miller AH. Depression and immunity: a role for T cells. *Brain Behav Immun* 2010; 24(1): 1–8.
40. Kappelmann N, Lewis G, Dantzer R, et al. Antidepressant activity of anti-cytokine treatment: a systematic review and meta-analysis of clinical trials of chronic inflammatory conditions. *Mol Psychiatry* 2016; 23: 335–343.
41. Decker MR, Benning L, Weber KM, et al. Physical and sexual violence predictors: 20 years of the women's interagency HIV study cohort. *Am J Prev Med* 2016; 51(5): 731–742.
42. Illangasekare SL, Burke JG, McDonnell KA, et al. The impact of intimate partner violence, substance use, and HIV on depressive symptoms among abused low-income urban women. *J Interpers Violence* 2013; 28(14): 2831–2848.
43. Ickovics JR, Hamburger ME, Vlahov D, et al. Mortality, CD4 cell count decline, and depressive symptoms among HIV-seropositive women: longitudinal analysis from the HIV epidemiology research study. *JAMA* 2001; 285(11): 1466–1474.
44. Morrison MF, Petitto JM, Ten Have T, et al. Depressive and anxiety disorders in women with HIV infection. *Am J Psychiatry* 2002; 159(5): 789–796.
45. Cook JA, Grey D, Burke J, et al. Depressive symptoms and AIDS-related mortality among a multisite cohort of HIV-positive women. *Am J Public Health* 2004; 94(7): 1133–1140.
46. Evans DL, Ten Have TR, Douglas SD, et al. Association of depression with viral load, CD8 T lymphocytes, and natural killer cells in women with HIV infection. *Am J Psychiatry* 2002; 159(10): 1752–1759.
47. Rivera-Rivera Y, Vazquez-Santiago FJ, Albino E, et al. Impact of depression and inflammation on the progression of HIV disease. *J Clin Cell Immunol* 2016; 7(3): 423.
48. Leserman J. Role of depression, stress, and trauma in HIV disease progression. *Psychosom Med* 2008; 70(5): 539–545.
49. Ghosh M, Daniels J, Pyra M, et al. Impact of chronic sexual abuse and depression on inflammation and wound healing in the female reproductive tract of HIV-uninfected and HIV-infected women. *PLoS ONE* 2018; 13(6): e0198412.
50. Barkan SE, Melnick SL, Preston-Martin S, et al. The women's interagency HIV study: WIHS Collaborative Study Group. *Epidemiology* 1998; 9(2): 117–125.
51. Bacon MC, von Wyl V, Alden C, et al. The women's interagency HIV study: an observational cohort brings clinical sciences to the bench. *Clin Diagn Lab Immunol* 2005; 12(9): 1013–1019.
52. Hessel NA, Weber KM, Holman S, et al. Retention and attendance of women enrolled in a large prospective study of HIV-1 in the United States. *J Womens Health* 2009; 18(10): 1627–1637.
53. Cohen M, Deamant C, Barkan S, et al. Domestic violence and childhood sexual abuse in HIV-infected women and women at risk for HIV. *Am J Public Health* 2000; 90(4): 560–565.
54. Radloff L. The CES-D scale: a self-report depression scale for research in the general population. *Appl Psychol Meas* 1977; 1: 385–401.
55. Radloff LS. The use of the center for epidemiologic studies depression scale in adolescents and young adults. *J Youth Adolesc* 1991; 20(2): 149–166.
56. Kyongo JK, Jespers V, Goovaerts O, et al. Searching for lower female genital tract soluble and cellular biomarkers: defining levels and predictors in a cohort of healthy Caucasian women. *PLoS ONE* 2012; 7(8): e43951.
57. Campbell J, Jones AS, Dienemann J, et al. Intimate partner violence and physical health consequences. *Arch Intern Med* 2002; 162(10): 1157–1163.

58. Sanchez-Lorente S, Blasco-Ros C, Coe CL, et al. Recovery of immune control over herpes simplex virus type 1 in female victims of intimate partner violence. *Psychosom Med* 2010; 72(1): 97–106.
59. Glaser R and Kiecolt-Glaser JK. Stress-induced immune dysfunction: implications for health. *Nat Rev Immunol* 2005; 5(3): 243–251.
60. Djuric Z, Bird CE, Furumoto-Dawson A, et al. Biomarkers of psychological stress in health disparities research. *Open Biomark J* 2008; 1: 7–19.
61. Slavich GM and Irwin MR. From stress to inflammation and major depressive disorder: a social signal transduction theory of depression. *Psychol Bull* 2014; 140(3): 774.
62. Reinhart TA, Qin S and Sui Y. Multiple roles for chemokines in the pathogenesis of SIV infection. *Curr HIV Res* 2009; 7(1): 73–82.
63. Farooq RK, Asghar K, Kanwal S, et al. Role of inflammatory cytokines in depression: focus on interleukin-1beta. *Biomed Rep* 2017; 6(1): 15–20.
64. Osuji FN, Onyenekwe CC, Ahaneku JE, et al. The effects of highly active antiretroviral therapy on the serum levels of pro-inflammatory and anti-inflammatory cytokines in HIV infected subjects. *J Biomed Sci* 2018; 25(1): 88.
65. Wada NI, Jacobson LP, Margolick JB, et al. The effect of HAART-induced HIV suppression on circulating markers of inflammation and immune activation. *AIDS* 2015; 29(4): 463–471.
66. Theron AJ, Anderson R, Rossouw TM, et al. The role of transforming growth factor beta-1 in the progression of HIV/AIDS and development of non-AIDS-defining fibrotic disorders. *Front Immunol* 2017; 8: 1461.
67. Williams WM, Castellani RJ, Weinberg A, et al. Do beta-defensins and other antimicrobial peptides play a role in neuroimmune function and neurodegeneration? *Sci World J* 2012; 2012: 905785.
68. Lisco A, Introini A, Munawwar A, et al. HIV-1 imposes rigidity on blood and semen cytokine networks. *Am J Reprod Immunol* 2012; 68(6): 515–521.
69. Vanpouille C, Introini A, Morris SR, et al. Distinct cytokine/chemokine network in semen and blood characterize different stages of HIV infection. *AIDS* 2016; 30(2): 193–201.
70. Brezing C, Ferrara M and Freudenreich O. The syndemic illness of HIV and trauma: implications for a trauma-informed model of care. *Psychosomatics* 2015; 56(2): 107–118.
71. Sales JM, Swartzendruber A and Phillips AL. Trauma-informed HIV prevention and treatment. *Curr HIV/AIDS Rep* 2016; 13(6): 374–382.
72. Espino SR, Fletcher J, Gonzalez M, et al. Violence screening and viral load suppression among HIV-positive women of color. *AIDS Patient Care STDS* 2015; 29(Suppl. 1): S36–S41.