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Supplementary Material

Association between Higher $CD32a^{+}CD4^{+}$ T Cell Count and Viral Load in the Peripheral Blood of HIV-infected Patients

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Abstract:

Background:

The significance of CD32a receptor expression in individuals infected with Human Immunodeficiency Virus (HIV) is currently unclear. Previously, B. Descours *et al*. (2017) concluded that in patients infected with HIV-1, CD32a is expressed on resting T cells that contain HIV DNA. According to the authors, these cells are reservoirs for inducible, replication-competent viruses. However, other studies have reported that CD32a expression is associated with activated T cells and is not a marker of HIV-1 reservoirs. The aims of this study were: to determine the significance of the CD32a marker in HIV infection, to assess its expression on T helper (Th) subpopulations in peripheral blood of HIV-infected individuals and to clarify the relationship between this expression and viral load.

Methods:

For comparative analysis, the following groups were used: 27 HIV-infected patients; 11 individuals with Hepatitis C Virus (HCV) infection; 16 individuals with Hepatitis B Virus (HBV) infection; and 13 healthy donors. Peripheral blood served as the study material. The expression of CD32a receptor on Th cell subpopulations was assessed using flow cytometry. Nonparametric statistical methods were used for data analysis.

Results:

It was found that relative $CD32a^{+}$ Th cell counts in HIV-infected individuals significantly exceeded corresponding values in other groups: healthy individuals ($p<0.0001$), those with HCV infection ($p=0.0008$) and those with HBV infection ($p<0.0001$). Among the Th subpopulations in HIV-infected patients, the CD32a receptor was predominantly expressed on Th1 cells ($p<0.0001$) and Th2 cells ($p<0.0001$), compared with Th17. We found a strong, direct correlation ($r=0.78$; $p<0.0001$) between viral load and $CD32a^{+}CD4^{+}$ T cell count in peripheral blood of HIV-infected individuals.

Conclusion:

Thus, our results provide evidence that the CD32a receptor can serve as a marker of HIV infection, and its expression depends on viral load. Clinical material was used here, for the first time, to show that CD32a is predominantly expressed on Th1 and Th2 cells.

Keywords: CD32a, HIV infection, Th cells, HCV, HBV, Viral load, Flow cytometry.

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Table S1. Study participants.

Code	Gender	Age (years)	Current Treatment	Viral Load, Copies/mL
1008	M	36	ABC/3TC/EFV	≤40
1009	M	30	ABC/3TC/EFV	≤40
1010	F	30	3TC/TDF/EFV	≤40
1011	F	28	ABC/3TC/LPV/r	≤40

(Table S1) contd.....

Code	Gender	Age (years)	Current Treatment	Viral Load, Copies/mL
1012	M	26	ABC/3TC/EFV	≤40
1013	M	41	ABC/3TC/EFV	≤40
1014	F	42	do not receive ART	82900
1015	M	35	3TC/TDF/EFV	≤40
1016	F	49	ABC/3TC/EFV	≤40
1017	F	31	do not receive ART	≤40
1018	M	41	FTC/RPV/TDF	≤40
1019	M	48	ABC/3TC/EFV	≤40
1020	F	32	3TC/TDF/EFV	≤40
1021	F	47	3TC/TDF/LPV/r	1210000
1022	F	24	do not receive ART	93200
1023	M	38	ZDV/3TC,EFV	≤40
1024	F	36	ABC/3TC/EFV	≤40
1025	F	31	ABC/3TC/EFV	≤40
1026	F	29	ABC/3TC/EFV	≤40
1027	F	41	3TC/TDF/ATV 200	62399
1028	F	46	3TC/TDF/EFV	359426
1029	M	40	do not receive ART	15532289
1030	M	53	do not receive ART	682869
1031	M	34	3TC/TDF/EFV	14184
1032	F	32	do not receive ART	932291
1033	M	42	ZDV/3TC/EFV	≤40
1034	M	40	do not receive ART	≤40

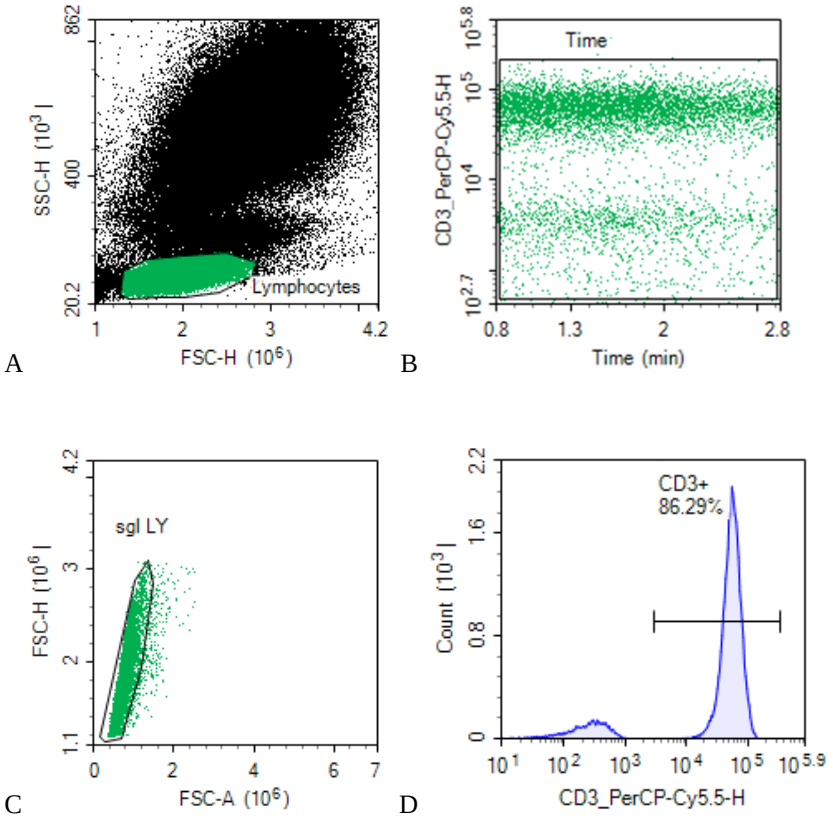

Fig. S1 contd.....

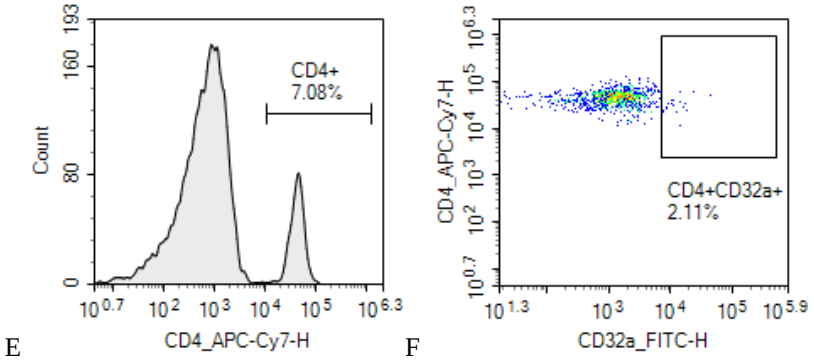

Fig. (S1). CD32a+ T helper cells flow cytometry immunophenotyping gating strategy.
Total lymphocyte population was first gated based on their forward (FSC) and side (SSC) scatter (dot plot **A**); dot plot **B** – artifact exclusion included time gating; then doublets were excluded from the analysis using FS-height and FS-area on dot plot **C**. Next, based on CD3 expression (dot plot **D**) the total T cell subset was identified. T cells were further analyzed by expression of CD4 (dot plot **E**) for identification of T helper cells (CD4+). T helper cells were then gated as CD32a+ lymphocytes (dot plot **F**).