Original Research Paper

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Evaluation of The Efficacy of Anti-Retroviral Therapy in Hiv-Seropositive Individuals in Mumbai – A Relationship Between Viral Load & Cd4+ T-Lymphocyte Count

Medical Science

KEYWORDS : HIV, zidovudine, lamivudine, nevirapine

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ABSTRACT

Both viral load and CD4+ T cell count has a role in the routine laboratory monitoring of patients with HIV infection, for assessing the response to anti-retroviral therapy (ART) over time. In the present study, the sample population consisted of 180 HIV-seropositive individuals from Mumbai in the age group 25 – 75 years. The viral load in ART-naïve individuals significantly dropped while CD4+ T cell count significantly raised after ART initiation (P < 0.0001). Viral load from the ART-naïve and ART-treated individuals exhibited a negative correlation with CD4+ T lymphocyte count (r = -0.2758, P < 0.02 for ART-naïve and r = -0.1404, P < 0.16 for ART-treated samples). The HIV-1 RNA level and CD4+ T cell count achieved at the end of 12 weeks of ART with fixed dose combination of zidovudine, lamivudine and nevirapine were important predictors of subsequent virologic and immunologic outcomes picturing its clinical effectiveness.

INTRODUCTION

Human Immunodeficiency Virus (HIV) is a lentivirus that causes Acquired Immunodeficiency Syndrome (AIDS) leading to failure of immune system in combating life-threatening opportunistic infections. HIV-1 infection leads to low levels of CD4+ T cells through 3 main mechanisms, i.e. cell damage by virus, increased apoptosis in affected cells and CD4+ T cell destruction by CD8+ cytotoxic lymphocytes.

When CD4+ T cell count declines below a critical level, cell mediated immunity is lost and body becomes progressively more susceptible to opportunistic infections. Current treatment for HIV-1 infection consists of highly active ART (HAART) which include these drugs belonging to at least two classes of ART agents. These classes are two nucleotide analogues reverse transcriptase inhibitor with either a protease inhibitor or a non-nucleoside reverse transcriptase inhibitor.

Incorporation of the competitive inhibitors of reverse transcriptase prevents further elongation of the DNA and terminates its activity. Non-nucleoside based reverse transcriptase inhibitors (NNRTIs) exert antiretroviral activity by allosteric mechanism of action. NNRTIs exhibit synergistic antiretroviral activity with nucleoside analogues antiretroviral agents. The tolerability and relatively low cost of manufacture has made nevirapine an important component of regimens designed to reduce maternofoetal transmission of HIV-1.[1] The half-life of the drug allows once daily dosing, in vivo antiviral activity is in the range of 1.5log10 of plasma viral RNA suppression.[2]

Most clinicians recommend initiation of the therapy before the CD4+ T cell count declines to < 350/μL. Most current initial regimens of therapy include at least two nucleoside analogues to provide high genetic barrier for resistance and at least a third drug to maximize potency.[3] Two studies (ACTG 384 and Gilead 903) have demonstrated the utility of these regimens.[4]

AIM AND OBJECTIVE

The aim of the study was to establish a relation between the viral load and CD4+ T lymphocyte count in ART-naïve and ART treated subjects, and to determine the efficacy of the ART regimen (fixed dose combination of zidovudine, lamivudine and nevirapine) 12-weeks after the initiation of ART in ART-naïve HIV-1 seropositive subjects.

MATERIAL AND METHODS

The present study involved 180 HIV-1 seropositive, ART-naïve individuals from Mumbai population in the age group of 25 – 65 years, eligible for ART according to WHO guidelines. The sample consisted of 108 (60%) males and 72 (40%) females. Age and gender-wise distribution of the study subjects is given in Table 1.

EDTA-anticoagulated blood samples were collected aseptically. For viral load determination, sample was prepared by plasmapheresis and extraction of plasma HIV-1 RNA, followed by calibration of Abbott m2000rt system and quantitation of viral load by real time polymerase chain reaction (RT-PCR).

For CD4+ T cell count, sample was prepared by tagging the cells with flurochrome-labelled antibodies specifically to leucocyte surface antigens and estimation of CD4+ T cell count was done on BD FACS Calibur flow cytometry system after its calibration and internal quality control check. The study group was given fixed dose combination of zidovudine (AZT), lamivudine (3TC) and nevirapine (NVP) to be taken twice daily. The viral load determination and CD4+ T cell counts were done before and after 12-weeks of initiation of ART. All the statistical analyses were done on SPSS Statistical Software.

RESULTS

The efficacy of the fixed dose combination was studied by comparing viral load and CD4+ T cell count before and after 12-weeks of ART (Table 2). The viral load was 58164.22 ± 7674.69 copies/ml in ART-naïve individuals which significantly dropped to 5490.90 ± 1354.43 copies/ml after ART (P < 0.0001).

While CD4+ T cell count was 190.98 ± 119.53 cells/μL in ART-naïve subjects and significantly raised to 292.24 ± 152.72 cells/μL after ART initiation (P < 0.0001). Students'

Paired t-test was used for statistical comparison.

Table 1: Age and Gender-wise distribution of study population

Age Group (yrs.)	Males	Females
25 - 35	15 (14%)	22 (31%)
35 - 45	21 (19%)	27 (38%)
45 - 55	45 (42%)	11 (15%)
55 - 65	15 (14%)	6 (8%)
65 - 75	12 (11%)	6 (8%)
Total	108	72

Table 2: Comparison of viral load and CD4+ T cell count in ART-naive and ART-treated individuals

Parameter	Viral Load (copies/ml)		CD4+ T Cell Count (cells/μL)	
	ART-naïve	ART-treated	ART-naïve	ART-treated
Mean ± SD	58164.22 ± 7674.69	5490.90 ± 1354.43	190.98 ± 119.53	292.24 ± 152.72
P-value	0.0001		0.0001	
	(Student's Paired t-test)			

Table 3: Correlation and Regression analysis between viral load and CD4+ T cell count in ART-naive and ART-treated individuals

Parameter	ART-naïve	ART-treated
Pearson's correlation coefficient (r)	-0.2758	-0.1404
r^2	0.0761	0.0197
P-value	0.02	0.16
Regression line	y = -177.10x + 91993.45	y = -12.44x + 9149.07
*'**y**' and '**x**' represents viral load and CD4+ T cell count respectively.*		

Plasma HIV-1 RNA level was correlated with CD4+ T lymphocyte count in ART-naïve (Figure 1) as well as ART-treated patients (Figure 2). Table 3 shows linear regression analysis showing viral load correlated negatively with CD4+ T cell count (r = -0.2758, P < 0.02 for ART-naïve and r = -0.1404, P < 0.16 for ART-treated) (Table 3).

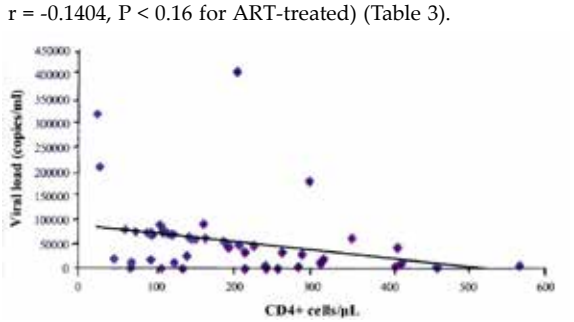

Figure 1: Correlation between Viral Load and CD4+ T cell count in ART-naive individuals

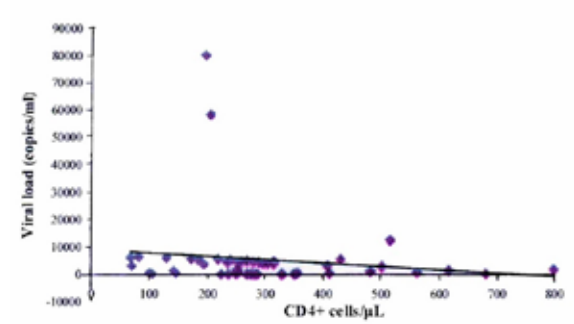

Figure 2: Correlation between Viral Load and CD4+ T cell count in ART-treated individuals

DISCUSSION

HIV-1 RNA concentration were significantly correlated with the corresponding absolute CD4+ T cell counts in a study by Mellors et al[5] although the association was weak which is strikingly similar to our study.

In a study by Piatak et al[6] there was also a significant correlation between PCR-determined HIV-1 RNA levels & decreasing absolute CD4+ T cell counts. A non-linear regression analysis of this data yielded the equation log (RNA) = 4.43 + 1.77exp [-0.0049 x (number of CD4+ T cells)], with r2 = 0.56 (P < 0.0001).

In this study, plasma HIV-1 RNA level and CD4+ T cell count was used as predictors of response to ART. RT-PCR analysis demonstrated that viral load from ART-treated individuals displayed a statistically significant reduction. Flow cytometry revealed statistically significant improvement in CD4+ T cell counts in ART-treated individuals. Thus, these results show that ART effectively tackled HIV-1 RNA in the peripheral blood mononuclear cells.

It is conceivable that ART inhibitory effect may be linked to ART induced decrease of plasma HIV-1 RNA load as a direct consequence of suppression of HIV-1 replication. The increase in CD4+ T cell counts may be due to a combination of redistribution and expansion of T cells. The magnitude of change in plasma HIV-1 RNA level in response to the given ART differed among patients, which could be due to differences in drug adherence, drug metabolism, baseline HIV-1 drug resistance, or other viral factors.

It is noteworthy that the study results are in accord with several studies that previously demonstrated a quantitative decrease of total HIV-1 RNA content when an effective treatment was carried out. A pilot study carried out to assess effectiveness of generic AZT, 3TC and NVP by Zijenah et al[7] exhibited that median CD4+ count for both men and women increased significantly, along with a significant decrease in the viral load.

O'Brien et al[8] have shown that reductions in plasma HIV-1 RNA concentration in response to AZT treatment account for a substantial portion (59%) of the benefit of this drug in delaying the onset of AIDS. In a pilot project by Calmy et al[9] AZT/3TC/NVP was chosen as the initial regimen, and 70% of those treated with it had viral load below 100 copies/mm3 after 6 months. There was a rapid improvement in the mean CD4+ count in the first 3-6 months after initiation of generic FDC formulations of AZT/3TC/NVP, which later reached a plateau in a study by Pujari et al[10]. The difference in the mean CD4+ T cell counts at baseline and at 12 months was significant.

CONCLUSION

The HIV-1 RNA level and CD4+ T cell count achieved at the end of 12 weeks of treatment were important predictors of subsequent virologic and immunologic outcomes. Patients initiating ART experienced marked clinical and immunologic improvement at 12 weeks of treatment along with significant virologic suppression. Hence, this fixed dose combination of zidovudine, lamivudine and nevirapine can be positioned as a good first-line regimen in programmes intended to deliver ART therapy in resource limited settings.

REFERENCES

1. Guay, L.A., Musoke, P., Fleming, T., Bagenda, D., Allen, M., Nakabiito, C., et al (1999), "Intrapartum and neonatal single-dose nevirapine compared with zidovudine for prevention of mother-to-child transmission of

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HIV-1 in Kampala, Uganda: HIVNET 012 randomized trial", Lancet, 354, 795-802.

2. Staszewski, S., Morales-Remirez, J., Tashima, K.T., Rachlis, A., Skiest, D., Stanford, J., et al (1999), "Efavirenz plus zidovudine and lamivudine, efavirenz plus indinavir, and indinavir plus zidovudine and lamivudine in the treatment of HIV-1 infection in adults", N Engl J Med, 341, 1865-73.
3. Larder, B.A., Kemp, S.D., Harrigan, P.R. (1995), "Potential mechanism for sustained antiretroviral efficacy of AZT-3TC combination therapy", Science, 269, 696-9.
4. Robbins, G., Shafer, R., Smeaton, L., DeGruttola, V., Pettinelli, C., Snyder, S., et al (2002), "Antiretroviral strategies in naïve HIV+ subjects: Comparison of sequential 3-drug regimens", Fourteenth International AIDS Conference, Barcelona, Spain, Abst LbOr20a.
5. Mellors, J.W., Rinaldo, Jr. C.R., Gupta, P., White, John, R.M., Todd, A., Kingsley, L.A. (1996), "Prognosis in HIV-1 infection predicted by the quantity of virus in plasma", Science, 272, 1167-70.
6. Piatak, M., Saag, M.S., Yang, L.C., Clark, S.J., Kappes, J.C., Luk, K.C., et al (1993), "High levels of HIV-1 in plasma during all stages of infection determined by competitive PCR", Science, 259, 1749-53.
7. Zijenah, L.S., Kadzirange, G., Rusakaniko, S., Kufa, T., Gonah, N., Tobaiwa, O., et al (2002), "A pilot study to assess the immunologic and virologic efficacy of generic nevirapine, zidovudine and lamivudine in the treatment of HIV-infected women with pre-exposure to single dose nevirapine or short course zidovudine and their spouses in Chitungwiza, Zimbabwe", HIV Clin Trials, 3(3), 177-85.
8. O'Brian, T.R., Blattner, W.A., Waters, D., et al (1996), "Serum HIV-1 RNA levels and time to development of AIDS in the Multicenter Hemophilia Cohort Study", JAMA, 276, 105-10.
9. Calmy, A., Pinoges, L., Szumilin, E., Zachariah, R., Ford, N., Ferradini, L. (2006), "Generic fixed-dose combination antiretroviral treatment in resource-poor settings: Multicentric Observational Cohort", AIDS, 20, 1163-9.
10. Pujari, S., Patel, A., Patel, K., Dravid, A., Patel, J., Mane, A., et al (2003), "Safety and long term effectiveness of generic fixed dose formulations of nevirapine based HAART amongst antiretroviral-naïve HIV-infected patients in India", Second International AIDS Society Conference on HIV Pathogenesis and Treatment, Paris, Abst 110.