

## Prevention of opportunistic infections in HIV infection by pentoxiphylline

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**Background & objectives:** Levels of tumour necrosis factor (TNF) are increased in patients with HIV infection leading to increased apoptosis and reduced CD4 cell life. Pentoxiphylline is a TNF inhibitor with properties that might make it useful for the treatment of HIV infection. These include improved cell mediated immunity and inhibition of viral replication. We carried out this study to determine the therapeutic utility of pentoxiphylline in improving constitutional manifestations, preventing opportunistic infections and sustaining CD4 counts among asymptomatic HIV infected individuals (*i.e.*, those with no opportunistic infection).

**Methods:** Individuals with HIV infection who were over 18 yr of age and free of opportunistic infections were recruited in the study and followed up 4 weekly. CD4 counts were measured using a flowcytometer using anti-human CD4 intervals. Pentoxiphylline was prescribed in a dose of 400 mg thrice daily.

**Results:** Thirty three (18 males) patients with HIV infection were studied. During their follow up (mean  $12.5 \pm 5.6$  months) one patient each developed cryptococcal meningitis and fibrocavitary tuberculosis. Weight increased from  $51.3 \pm 7.4$  kg at baseline to  $55.3 \pm 7.4$  kg ( $P < 0.05$ ). Malaise, fatigue and appetite improved in all those with these complaints, except the two with opportunistic infections. Mean CD4 counts were  $184 \pm 36.4/\mu\text{l}$  at baseline and increased to  $210 \pm 28.6/\mu\text{l}^3$  at four weeks ( $P < 0.05$ ). The patients had stable CD4 counts over the follow up period since then, *i.e.*, within 25 per cent of the previous levels.

**Interpretation & conclusion:** Pentoxiphylline therapy in HIV infected individuals, who were free of opportunistic infections, improved their body weight, minimized opportunistic infections, increased and sustained CD4 counts. Given the low cost of the drug it could be recommended for the use in individuals who are at a high risk of developing opportunistic infections.

**Key words** Anti-TNF therapy - HIV - opportunistic infections - pentoxiphylline

As of end 2004, there were approximately 39.4 million individuals infected with HIV world-wide<sup>1</sup>. The current cost of antiretroviral therapy is prohibitive for the majority of patients in most developing countries and there is an urgent need

for effective, safe and cheap antiretrovirals<sup>2</sup>. The only treatment modalities available for therapy for the masses are those for opportunistic infections that this does not take adequate care of the immunodeficient state induced by the virus itself.

Tumour necrosis factor- $\alpha$  (TNF $\alpha$ ) levels rise in patients with HIV infection<sup>3-5</sup>. TNF $\alpha$  has significant role in apoptosis, one of the major mechanisms of cell death in AIDS<sup>6</sup>. It markedly enhances the *in vitro* spread of HIV-1 by increasing viral RNA synthesis<sup>7</sup>. It may also be involved in several complications associated with clinical AIDS such as wasting, haematological, renal and neurological disorders<sup>8</sup>. It has been suggested that blockade of TNF production and/or receptor sites would benefit individuals with HIV infection<sup>9</sup>. Anti-TNF agents include phosphodiesterase inhibitors that inhibit TNF synthesis<sup>9</sup>. Pentoxiphylline, a phosphodiesterase inhibitor, blocks the accumulation of TNF $\alpha$  mRNA<sup>10</sup>.

Pentoxiphylline has been used earlier in the therapy of patients with HIV infection and some studies have shown benefit with respect to some immunological parameters. Heinkelein *et al*<sup>11</sup> showed that pentoxiphylline might inhibit cytotoxicity and cytokine secretion among patients with HIV infection. It has been shown to improve cell-mediated immunity and reduce plasma viraemia in asymptomatic HIV seropositive persons<sup>12</sup>. It has also been suggested to inhibit acute HIV-1 replication in human T cells by a mechanism not involving inhibition of TNF synthesis or nuclear factor-kappa B activation<sup>13</sup>. In standard doses of 400 mg thrice daily the cost of the drug is less than Rs 10 per day in India. We studied the therapeutic benefit of pentoxiphylline in the prevention of opportunistic infections and improvement in constitutional manifestations in HIV infected Indian patients.

### Material & Methods

This study was carried out among HIV positive individuals attending the Medical Outpatients Department at Postgraduate Institute of Medical Education and Research, Chandigarh, between 1999 and 2002. All the patients who were willing and eligible for the study were included. In order to avoid confounding variables among various parameters, individuals with active opportunistic infection were not included in the study. The following were the inclusion criteria: age >18 yr, no opportunistic infection or neoplasm, CD4 counts between 50 and 500/ $\mu$ l and willingness to participate in the trial. The following were the exclusion criteria: age <18 yr, CD4 count more than 500 or less than 50/ $\mu$ l, Karnofsky score <70, therapy with antiretrovirals, pregnant or lactating

women, hemoglobin <9 g/dl, absolute granulocyte count <1000/ $\mu$ l, platelet count <25,000/ $\mu$ l, uric acid >8 mg/dl, derangement in hepatic or renal functions (urea and creatinine) to more than two times normal, active and untreated infection, pregnant or lactating women, intolerance to pentoxiphylline and refusal to give informed consent. At baseline, a complete physical examination was performed, along with routine tests for haemogram, liver functions, X-ray chest and CD4 count. The drug was prescribed in a dose of 400 mg thrice daily. Follow up was made every four weeks with clinical assessment, including body weight, and CD4 counts. Efforts to procure the drug and an identical placebo from manufacturers of the drug in India were unsuccessful; hence, this was an open label study. No patient received co-trimoxazole prophylaxis.

At each 5 ml visit blood samples were collected. Serum was separated and stored at -20°C for future analysis. Informed consent was taken at the time of recruitment in the trial. The study was carried out after obtaining permission from the Institute's Ethics Committee.

CD4 counts were measured on a flowcytometer (Becton-Dickinson, USA) anti-human CD4-FITC (Sigma Immunochemicals, USA). The results were analyzed by two-way analysis of variance (ANOVA). Pearson's correlation coefficient was calculated to determine correlation;  $P < 0.05$  was considered significant.

### Results

Thirty three (18 males) patients with HIV infection were studied (Table I). Mean follow up of these patients was  $12.5 \pm 5.6$  months (range 6-20 months). Two patients developed infections during the course of follow up. One patient developed cryptococcal meningitis within four weeks and another developed fibrocavitary tuberculosis six months after initiation. The former was diagnosed on cerebrospinal fluid culture and the latter by positive acid-fast bacilli in the sputum. The patient who developed cryptococcal meningitis had CD4 count of 0/ $\mu$ l and was subsequently started on triple drug antiretroviral therapy. Six months after start of this therapy his CD4 count was 64/ $\mu$ l. The other patient was treated with standard anti-tubercular therapy and was maintaining well one year later. Weight increased from  $51.3 \pm 7.4$  kg at baseline to

55.3 ± 7.4 kg ( $P < 0.05$ ), the last recorded weight. There has been return to gainful employment in eight cases. Malaise, fatigue and appetite have improved in all those with these complaints, except the two with opportunistic infections. Mean CD4 counts were 184 ± 36.4/μl at baseline and increased to 210 ± 28.6/μl at four weeks ( $P < 0.05$ ). They have since remained stable over the follow up period since then, *i.e.*, within 25 per cent of the previous levels. The drug was well tolerated with minimal side effects. Four patients felt mild abdominal discomfort but none of the patients had reason to discontinue the therapy.

### Discussion

In this study, the use of pentoxiphylline led to amelioration of constitutional symptoms in most patients. Weight gain was sustained in virtually all the patients. This led to gainful employment in eight cases. Weight loss is usually progressive among HIV infected individuals<sup>14</sup>. This process was halted and in most of them original weight was restored. These improvements had a positive influence on the patients and improved their quality of life. At a price of less than a quarter dollar per day, this may be a useful form of therapy for HIV infected individuals. There were only two opportunistic infections in our patients despite the fact that individuals with CD4 count less than 500/μl are known to be at an increased risk of development of opportunistic infections<sup>15</sup>. CD4 counts showed an increase and have been sustained over the period of follow up. This is significant, as these counts are indicative of the likelihood of the development of opportunistic infections in the near future<sup>16</sup>. CD4 count of 500 was taken as the cut-off since at the time of start of study that was the indication of initiation of

antiretroviral therapy. The cost of pentoxiphylline is far less than the cheapest triple drug regimen (Table II). In a poor country like India this is an important consideration. A useful corollary to this is to institute this form of therapy and sustain the CD4 count for as long as possible and, thereby, delay the time at which antiretroviral therapy needs to be initiated. This alone could reduce the long-term cost of treatment and reduce mortality and morbidity by reducing opportunistic infections.

Several studies have documented the mechanisms of action of pentoxiphylline in patients with HIV infection. TNF-α was suppressed at four weeks with this therapy<sup>17</sup>. Since it is responsible for weight loss, its suppression is the likely explanation for weight gain in our patients. With four weeks of this therapy, level of nitric oxide production decreased significantly, as demonstrated by its surrogate markers, nitrite and citrulline<sup>17</sup>. In another study, there was a decline in beta-2 microglobulin (B2M) levels after four weeks of use of pentoxiphylline<sup>18</sup>. This assumes significance in view of the importance of both immune defects, as shown by decreased CD4 counts, and immune stimulation, as shown by elevated B2M, in predicting disease course and prognosis<sup>19</sup>. Immune stimulation occurs very early in the course of HIV infection, as evidenced by the demonstration of HIV antibodies in the serum of these patients<sup>20</sup>. There is additional importance of demonstrating immune stimulation in these patients as these can predict the future rate of decrease in CD4 count for at least two to three years<sup>20</sup>. Elevated B2M can have predictive value at normal, intermediate or markedly reduced CD4 counts<sup>19</sup>. The possible mechanism of its action beyond TNF inhibition needs to be elucidated. However, markers of apoptosis, namely, caspase 1 and caspase 8 were decreased significantly following four weeks of pentoxiphylline<sup>21</sup>.

**Table I.** Clinical profile of patients with HIV infection

|                                  |   |
|----------------------------------|---|
| Number                           | 33  |
| Age (yr) (range)                 | 32.5 ± 5.9 (21-45)  |
| Sex ratio: m : f                 | 18 : 15   |
| Route of transmission            | heterosexual 30, blood transfusion 2, IVDU* 1   |
| Past AIDS defining illness       | 8   |
| Chief complaints at presentation | Loss of weight 28, malaise or fatigue 20, loss of appetite 18, fever 12, enlarged lymph nodes 7 |
| Hb, g/dl (range)                 | 9.2 ± 0.8 (8.9 - 11.6)  |
| *IVDU, intravenous drug user     |   |
| Values are mean ± SD             |   |

**Table II.** Relative monthly costs (US\$) of antiretroviral combinations and pentoxiphylline in India in July 2005

| Regimen  | Cost (US\$) |
|--|-------------|
| Protease inhibitor based                                   | >120        |
| Efavirenz  | 60          |
| Nevirapine based ( <i>e.g.</i> , triple drug combinations) | 30          |
| Pentoxiphylline  | 12          |

*Source: MIMS India 2005; 25 : 5*

An obvious pitfall in this study was that neither patient nor the physician has been blinded to the therapy and there was no placebo control. Efforts to procure the drug and an identical placebo from manufacturers of the drug in India were unsuccessful.

There have been four other patients outside this clinical trial who have been administered this drug for sometime and on gaining weight have discontinued it. With this they lost weight and appetite that was gained earlier. On restarting pentoxiphylline both these parameters improved in four weeks time.

In conclusion, a study with a large sample size and a longer follow up needs to be carried out with a placebo control arm, to see if the use of highly active antiretroviral therapy (HAART) can be delayed. An ancillary benefit of this drug is increase in body weight. This too can be a pointer for improvement as the natural course of HIV infection is such that either weight remains the same or declines over a period of time.

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