SPECTRUM AND RISK FACTORS OF ANEMIA IN HIV INFECTED INDIAN CHILDREN PRESENTING TO A TERTIARY LEVEL TEACHING HOSPITAL AT NEW DELHI

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ABSTRACT

Objective: To assess prevalence and risk factors of anemia in HIV infected children

Design: Cross sectional study.

Setting: Anti-Retroviral Therapy clinic at Maulana Azad Medical College and associated Lok Nayak Hospital, New Delhi,India.

Participants: 48 HIV infected children between age of 18 months – 12 years.

Results: Most common hematological abnormality were anemia (75%) followed by Thrombocytopenia and Leucopenia. Most common type of anemia, according to the peripheral smear and red cell indices was, found to be of normocytic normochromic variety seen in 23 (63.9%) children. Malnutrition (under-weight), stunting, symptomatic presentation, fever, low immunological stage, were risk factors for anemia and its severity. No anti-retroviral drug therapy was risk factor for anemia but not severity. Rural Background was risk factor only for severity.

Conclusions: Prevention, early detection and treatment of the risk factors of anemia in HIV infected children can prevent anemia and will improve overall quality of life.

Key words: HIV, anemia, children.

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INTRODUCTION

Globally[1], the HIV epidemic remains a serious challenge specially in children. Ongoing perinatal transmission substantially impacts the incidence of pediatric HIV, adding to the large pool of HIV-infected children in developing countries like India and background co-morbidities compound the problem. Two such major co-morbidities include anemia and poor nutrition, whose detrimental effects are magnified in the context of HIV infection.

From very early in the HIV-pandemic, hematological abnormalities were recognized as a common manifestation of HIV infection. And among them anemia, appeared to be the most common hematological complication in HIV infected individuals[2,3] and a positive association had been reported between the prevalence of anemia and the severity of clinical disease[4]. The pathogenesis of anemia in HIV-infected individuals, although multifactorial, relates primarily to a reduced production of erythrocytes. Often, several mechanisms are operative in a single patient. These are: direct effect of HIV infection of hematopoietic cells or bone marrow stromal cells, effect of opportunistic infection, hemophagocytic syndrome, hematinic deficiency, auto-immune hemolytic anemia, drug toxicity, direct or indirect effect of HIV related lymphomas or others. In-vitro data suggested that HIV itself may diminish erythropoiesis through apoptosis of erythroid precursors or infection of auxiliary cells, by altering cytokine and erythropoietin responses[5]. Children may be more vulnerable to these mechanisms because they differ from adults in their hematopoiesis, increased cytokine responses, or viral loads[6]. Studies have demonstrated that anemia is associated with decreased survival and increased disease progression in HIV infection. In children, it may present with weakness, fatigue, tachypnea and congestive cardiac failure and is associated with poor mental, motor, social–emotional, and neurophysiologic functioning[6].

Most common anemia in HIV infected individuals is anemia of chronic disease. Anemia of chronic disease in AIDS shows the usual characteristics of this condition. The red cells are initially normocytic and normochromic but when infection is chronic or recurrent, red cells may become hypochromic and microcytic. The serum iron, iron-binding capacity, and transferrin concentrations are all low, and serum ferritin concentration is increased. There is increased rouleaux formation, increased background staining on the blood film, and an elevated erythrocyte sedimentation rate. The erythropoietin response to anemia is blunted[7,8] and the reticulocyte count is similarly inappropriately low for the degree of anemia. Tumor necrosis factor, which is an important mediator of the anemia of chronic disease, is elevated in the serum of patients with AIDS and AIDS-related complex but not in the serum of those with asymptomatic infection. Severity of anemia correlates with serum concentration of soluble tumor necrosis factor receptor and interleukin-6.

Compared with adults there is very little information available about the association between HIV infection and anemia in children. Given that the negative impact of anemia is magnified on account of its close relation to overall nutrition and growth, there is limited data from Asian countries, including India, where HIV infection, malnutrition and nutritional deficiencies co-exist[9].

METHODS

A total of 48 children were included in our study from Lok Nayak Hospital, New Delhi. 17 were newly diagnosed and 11 who were diagnosed earlier, but were not on the ART, were included. Rest of the patients, 20 in number, were old cases that were already on ART for minimum duration of 3 months.

TYPE OF STUDY: Cross sectional study.

INCLUSION CRITERIAS: All newly diagnosed cases of HIV infected children during the study period in the age group of 18 months-12 years and HIV infected children on ART for at least 3 months in the age group 18 months-12 years with clinical anemia were included.

All 48 children were subjected to detailed history with special references to symptoms, co-morbidities and different risk factors of anemia. Factors which were assessed in the history: age, sex, religion, socio-economic status, background, dietary intake, duration of enrollment, clinical and immunological stage at time of study, ART and duration, use of zidovudine and co-trimoxazole, compliance, co-infections and opportunistic infections like TB, fever, candidiasis, diarrhea, anthropometry: weight for age, height/length for age, weight for height and BMI. Symptoms at enrollment like diarrhea, fever and conditions like TB, opportunistic infection, candidiasis were included in our study. Whether children are on anti-retroviral therapy, their duration and regimen being used was also taken into account. Concomitant uses of other drugs like co-trimoxazole, anti mycobacterial treatment were also noted from records. Co-infections and their treatment were recorded. Dietary adequacy assessed using age for calories charts and classified as whether adequate or inadequate. Anthropometric assessment were done using WHO charts and classified into moderate and severe: weight for age, height for age, weight for height (wasting), BMI. Those lower than 3SD were classified as severe and between 2 and 3 SD were taken as moderate. Socio-economic status was assessed using Kuppuswamy scale.

All children underwent following investigations to assess for anemia: Complete blood counts (using Sysmex XT-2000i counter and Grabner counters), peripheral smear for type of anemia, various red cell indices –HCT ,MCV, MCH, MCHC, RDW. CD4 count (using BD FACS flow-cytometer): for immunological staging.

Anemia grading done using WHO cut-offs[10]. Other investigations were done when indicated depending upon the type of anemia in peripheral smear and availability of tests.

STATISTICAL ANALYSIS: The data were analysed using appropriate statistical methods to determine the correlates and spectrum of anemia: Chi square, Univariate analysis.

RESULTS

In our study population, (77.1%) were ≥5 years of age and rest (22.9%) were <5 years of age. Males were 32(66.7%) and female were 16(33.3%). Majority were Hindus 43 (89.6%) coming

from rural 28(58.3%), including urban slum, background. According to the Kuppuswamy Scale most number of patients 30 (62.4%) were from lower class.

- Most common hematological abnormality found was anemia, 36 (75%) of the 48 recruited patients were anemic. In order of frequency Anemia > Thrombocytopenia > Leucopenia were the hematological abnormalities noted in children with HIV infection in our study.

- In our study mild anemia was present in 15(31.3%) of total 36 anemic children, and 16(33.3%) were having moderate anemia. Severe anemia was present in 5(10.4%) of cases.The mean hemoglobin in study population was 9.8gms/dl with a range of minimum 2.9 gm/dl and maximum 9.8 gm/dl.

- In our study most common type of anemia, according to the peripheral smear and red cell indices was, found to be of normocytic normochromic variety 23 (63.9%). Microcytic variety was present in 5(13.9%) and macrocytic variety was observed in least number of the patients only in 1(2.8%). Mixed type of anemia in peripheral examination was present in 7(19.4%) children.

- Value of RDW (indicator of anisopoikilocytosis) was in normal range in 33(68.6%) patients. Most of the children, 35(72.9%), were having MCV values between 80 fl and 95 fl meaning either they were non anemic or were having normocytic variety of anemia.

- In our study, Ferritin (most useful indicator of iron deficiency) was low in 7 (14.6%) out of the total 48 subjects and vitamin B12 was low in 6 (12.5%). Normal value of Ferritin was taken as >12ng/ml. In our study Fe deficiency was found to be causing anemia only in 7(14.6%). Among them concomitant B12 deficiency was present in 2 patients.

- Bone-Marrow Aspiration could be done only in 6 patients. Bone-marrow was hypo-cellular in 4(66.6%) and hyper-cellular in 2(33.4%).

- 9(25.0%) anemic patients (total anemic 36) were <5 years of age and 27(75.0%) were ≥5years of age. Age was not found to be a risk factor for the development of anemia.

- Among total 36 with anemia, 25(69.4%) of the males were anemic and 11(30.6%) of the females were having anemia. No relation was observed between anemia and the gender.

- Among the children with anemia 33(91.7%) were Hindus and 3(8.3%) were Muslims. Religion was not found to be risk factor for the development of anemia.

- Among children with anemia, 23(63.9%) were from rural background and 13(36.1%) belonged to urban background. Background is not found to be a risk factor for the development of anemia but background was a significant risk factor for the severity (grading) of anemia in our study.

- In our study majority 30 (62.5%) belonged to the lower class. Among the lower class 20(66.67%) were anemic and in the middle class 16(88.9%) were anemic. Socioeconomic status was not found to be significantly associated with the development of anemia.

- Children were grouped according to the clinical stage and those who were on ART treatment staging were used. 21(43.75%) in stage 1,2 and 15(31.25%) in stage 3,4 were having anemia. Clinical stage was not related to the development of anemia.

- 11(91.7%) of 12 anemic patients had normal or low (but not in severe immunosuppression) CD4 counts. In comparison 23(63.9%) of 36 anemic patients had low for age (severe immunosuppression). Immunological stage was a risk factor for the development and severity of anemia. Hence, more the immunosuppression higher the chances of anemia and of severe grade.

- 8(66.7%) of the 12 non-anemic were on ART and 12 (33.3%) of 36 anemic patients were on ART. Use of ART was found to be protective against development of anemia. But use of ART was not protective against the development of severe anemia. Thus, ART which was protective for the development of anemia was not protective for the severity of anemia. Duration of ART, and the regimen used (Zidovudine and Stavudine) were not significantly associated with occurrence of anemia. Use of Co-trimoxazole and compliance with ART were also statistically found to be insignificant.

- 32 patients were symptomatic at time of enrollment into the study. Among them 28 (87.5%) were anemic. Major symptom was fever and all 14 patients presenting with prolonged fever were anemic. Thus symptoms at enrollment, particularly fever was associated with increased risk of anemia and was risk factor for the severity too.

- In our study 30 (83.3%) of total 36 anemic children were moderate to severe under-weight and 28(77.8%) were moderately to severely stunted. They were found as a significant risk factor for the development of anemia its severity. But wasting or low BMI as well as dietary intake were not statistically significant. It proves that malnutrition is definitely a major risk factor for the development of anemia and of severe grade in the children with HIV infection.

- Using multi-variate analysis, only CD4 count was found to be significantly associated with anemia (with a wide confidence interval) (Table 1).

TABLE 1: RISK FACTORS FOR ANEMIA AND ITS SEVERITY

ANEMIA	SEVERITY OF ANEMIA
Malnutrition (Under-Weight)	Rural Background
Stunting	Malnutrition (Under-Weight)
Symptomatic	Stunting
Fever	Symptomatic
Low Immunological Stage	Fever
Not On ART	Low Immunological Stage

DISCUSSION

In our study, most common hematological abnormality found was in order of frequency Anemia > Thrombocytopenia > Leucopenia. The percentage of anemic in HIV infected children were similar to what was found in the other studies of similar kind[11]. Studies in different parts of the

world have found anemia as the most common hematological abnormality in as much as 70-90% of HIV infected children. Majority, 16(33.3%), were having moderate anemia. Total moderate to severe anemia was seen in 21(43.7%) cases. These findings were similar to the prevalence of severity of the anemia observed in other studies. In a systematic analysis by Calis et al[12] prevalence of mild or moderate anemia in HIV-infected children varied between 22–94 and 3–82%, respectively. In our study most common type of anemia, according to the peripheral smear and red cell indices was, found to be of normocytic normochromic variety 23 (63.9%). Microcytic variety was present in 5(13.9%) and macrocytic variety was observed in least number of the patients only in 1(2.8%). Mixed type of anemia in peripheral examination was present in 7(19.4%) children. It is well known that most common anemia in HIV infected individuals is anemia of chronic disease[11]. The red cells are initially normocytic and normochromic. When infection is chronic or recurrent, red cells may become hypochromic and microcytic. RDW (Red Cell Distribution Width) is an indicator of degree of anisopoikilocytosis in peripheral smear. Higher the value of RDW, higher the chances of being it an iron deficiency anemia. In our study, Value of RDW was in normal range in 33(68.6%) patients. And was increased in 15(31.2%) suggestive of iron deficiency anemia. MCV (Mean Cell Volume) measures average volume of the RBC. A low value indicates microcytic anemia and higher values are indicators of macrocytic anemia in presence of anemia. In our study most of the children, 35(72.9%), were having MCV values between 80fl and 95fl meaning either they were non anemic or were having normocytic variety of anemia. Thus peripheral smear examination along with red cell indices can point towards possible type and etiology of anemia in HIV infected children. Ferritin was low (<12ng/ml) in 7 (14.6%) out of the total 48 subjects and vitamin B12 was low in 6 (12.5%). Ferritin is a positive serological marker of infection[13] and that may explain low ferritin in fewer subjects even though total number of patients having microcytic hypochromic and mixed anemia was 12 out of the total 36 anemic patients. Similar results were observed in a study by Eley et al[14]. In our study we could not use investigations like serum iron status and Soluble Trasferrin Receptor (sTfR) which could have been more sensitive markers for identifying body iron status. Age was not found to be a risk factor for the development of anemia (p 0.552). Children in the pre-school age group are at considerably higher risk for developing anemia, possibly due to their increased growth requirements and higher frequency of gastrointestinal infection. Background was not a risk factor for the development of anemia but was a significant risk factor (p value

0.008) for the severity (grading) of anemia in our study. That means even though rural children are at equal risk for the development of anemia but when they develop it is of more severe grade in comparison to the children of urban background. Study by Shet et al[15] found rural background as a risk factor for the development of anemia. Clinical staging was not but immunological stage (p value=0.001) was a risk factor for the development and severity of anemia. Hence, more the immunosuppression higher the chances of anemia and of severe grade. This association is most likely explained by the increasing viral burden as HIV disease progresses, which could cause anemia by increased cytokine mediated myelosuppression. And higher immunosuppression is also associated with the development of severe and opportunistic infections making children more prone to the development of anemia. Use of ART was found to be protective against development of anemia (p value 0.043) but not against the development of severe anemia. Drugs contribute to the anemia of AIDS by causing either bone marrow suppression or hemolysis among other mechanisms[11]. All the children with moderate and severe anemia were symptomatic at the time of enrollment and fever was the main symptom. In our study 30 (83.3%) of total 36 anemic children were moderate to severe under-weight and 28(77.8%) were moderately to severely stunted. They were found as a significant risk factor for the development of anemia having p values 0.014 and 0.042 (<0.05) respectively. But wasting or low BMI were not statistically significant. It proves that malnutrition is definitely a major risk factor for the development of anemia and of severe grade in the children with HIV infection. Growth failure may be a direct consequence of the HIV infection, secondary to the clinical illness associated with HIV, a function of the child's adverse environment, or a combination of these factors. It is probable that, independent of HIV infection, malnutrition can reduce immunological function and can impair that child's ability to resolve acute infections and can cause anemia.

Once anemia develops intervention is appropriate as it may both improve quality of life and reduce morbidity and mortality. The Anemia in HIV Working Group[16] advised that evidence-based treatment strategies should be implemented for anemia in HIV infected patients. Patients with HIV infection might have a mixture of chronic under-nutrition and infection that further weakens their immune response, leading to altered immune cell populations and a generalized increase in inflammatory mediators. Recombinant human erythropoietin (r-HuEPO) is now recognized as a valued treatment for anemia in HIV-infected patients[17]. Although using rHuEPO might increase the production of red blood cells, this production will not reach the

optimal level without adequate levels of iron, vitamin B12, and folic acid. Optimal therapy for anemia must be directed against the underlying disease, because its severity is linked with disease activity and patients will require high doses of rHuEPO, which is very expensive.

REFERENCES

1. UNAIDS epidemic update. December 2010 http://www.unaids.org/globalreport.

2. SpivakJL, Bender BS, Quinn TC. Hematological abnormalities in AIDS. Am J Med 1984;77:224-8.

3. Coyle TE.Hematological complications of human immunodeficiency virus infection and the acquired immunodeficiency syndrome. Med Clin North Am 1997;81:449-70.

4. Clark TD, Mmiro F, Ndugwa C, Perry RT, Jackson JB, Melikian G, et al. Risk factors and cumulative incidence of anaemia among human immunodeficiency virus-infected children in Uganda. Ann Trop Paediatr 2002;22:11–7.

5. Volberding PA, Baker KR, Levine AM. Human immunodeficiency virus hematology.Hematology Am SocHematolEduc Program 2003:294-313.

6. Walker AS, Doerholt K, Sharland M, Gibb DM. Response to highly active antiretroviral therapy varies with age: the UK and Ireland Collaborative HIV Paediatric Study. AIDS 2004;18:1915–24.

7. Spivak JL, Barnes DC, Fuchs E, Quinn TC. Serum immunoreactive erythropoietin in HIV-infected patients. JAMA 1989;261:3104–7.

8. Kreuzer KA, Rockstroh JK, Jelkmann W, Theisen A, Spengler U, Sauerbruch T. Inadequate erythropoietin response to anaemia in HIV patients: relationship to serum levels of tumour necrosis factor-α, interleukin-6 and their soluble receptors. Br J Haematol 1997;96:235–9.

9. Walker SP, Wachs TD, Gardner JM, Lozoff B, Wasserman GA, Pollitt E, et al. Child development: risk factors for adverse outcomes in developing countries. Lancet 2007; 369:145–57.

10. Iron Deficiency Anaemia Assessment, Prevention, and Control. A guide for programme managers. WHO 2001.

11. Bain B J. Pathogenesis and pathophysiology of anemia in HIV infection. CurrOpinHematol 1999;6:89-102.

12. Calis JC, van Hensbroek MB, deHaan RJ, Moons P, Brabin BJ, Bates I. HIV-associated anemia in children: a systematic review from a global perspective. AIDS 2008,22:1099-112.

13. Feelders RA, Kuiper EPA-Kramer, van Eijk HG. Structure, function and clinical significance of transferrin receptors. ClinChem Lab Med 1999;37:1-10.

14. Eley BS, Sive AA, Shuttleworth M, Hussey GD. A prospective, cross-sectional study of anaemia and peripheral iron status in antiretroviral naïve, HIV infected children in Cape Town, South Africa. BMC Infectious Diseases 2002;2:3.

15. Shet A, Mehta S, Rajagopalan N, Dinakar C, Ramesh E, Samuel NM et al. Anemia and growth failure among HIV-infected children in India: a retrospective analysis. BMC Pediatrics 2009;9:37.

16. Volberding PA, Levine AM, Dieterich D, Mildvan D, Mitsuyasu R, Saag M: Anemia in HIV infection: clinical impact and evidence based management strategies. Clin Infect Dis 2004;38:1454-63.

17. Henry DH, Volberding PA, Leitz G.Epoetin Alfa for Treatment of Anemia in HIV-Infected PatientsPast, Present, and Future. J Acquir Immune DeficSyndr 2004;37:1221–7.