Case report

Pediatric Active Tuberculosis in a Context of Multidrug Resistant HIV-1 in Cameroon Suggests a Cross-Linkage Between Both Pathogens and Complexity in Clinical Management.

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ABSTRACT
We describe active tuberculosis among two children harboring Multi-Drug Resistant HIV (MDRHIV) in Cameroon: A male of age 13, TB-treated in 2009, diagnosed with MDRHIV on antiretroviral therapy intensification. A male of age 15, admitted for intensive care with an atypical persistent pneumonia and weight loss, harbouring MDRHIV with drug treatment failure to all antiretrovirals available nationwide. Specific investigations and guidelines would be relevant.

RÉSUMÉ
Nous décrivons deux cas de tuberculose (TB) actives chez des enfants porteurs de VIH multirésistants aux antirétroviraux (ARV) au Cameroun : Un garçon de 13 ans traité de la TB en 2009 et porteur de virus multirésistants aux ARV sous traitement antiretroviral intensif. Un adolescent de 15 ans admis en soins intensifs pour cause de pneumonie atypique persistante couplée de perte poids, et portant des virus multirésitants à tous les ARV disponibles localement. Des investigations et recommandations spécifiques seraient nécessaires.

INTRODUCTION

Acquired Immuno-Deficiency Syndrome (AIDS) is characterized by the emergence of opportunistic infections among which *Mycobacterium tuberculosis (MTB)* is the leading cause of death among people living with human immunodeficiency virus (HIV) [1]. Among the 2 billion people infected with *MTB* worldwide [2], children account for 10–15% of the global TB cases[3]. The re-emergence of *MTB* and progression toward clinical manifestations in sub-Saharan Africa is significantly associated to the HIV/AIDS epidemic. Furthermore, children infected with HIV have up to 24-fold higher risk of developing TB compared to their HIV-negative peers[4]. Interestingly, diagnosis, treatment and prevention of pediatric TB remains a major challenges. Indeed, CD4 T cells count and HIV viral load are relevant surrogate markers for evaluating HIV progression but not for mapping co-infection with *MTB*. Staging the progression of the *MTB* infection in the context of pediatric HIV would be of great asset in preventing the advent of TB disease and subsequently, in decreasing the burden of co-infections among children. Achieving such goals may require in-depth understanding of biomarkers responsible of interactions between MTB and HIV. These motivated our research focus to these two cases of pediatric TB development in the context of multidrug resistant HIV (MDRHIV) in Cameroon, one of the most highly resource-limited settings with high prevalence of both TB and HIV infections.

CASE PRESENTATION

Case 1 : We report the case of a male child, of age 13, and living with HIV since birth. He had been treated for pulmonary tuberculosis (TB) in 2009 and harbored MDRHIV, with reduced drug susceptibility to the ARV regimens locally available.

His CD4+ result showed 04 CD4+ T-cell/µl both in October 2004 and in April 2005, he was enrolled on a highly active antiretroviral therapy with a regimen containing didanosine, lamivudine, plus efavirenz (ddI+3TC+EFV) from January 2005 to June 2007, afterwards he was switched (due to poor CD4 recovery, and treatment change without a genotypic resistance testing) to a second line regimen containing DDI, abacavir (ABC), and ritonavir boosted lopinavir (LPV/r) in January 2008 up to September 2011. Several treatment interruptions were reported due to non-adherence to ART. A slight increase of CD4 T-cell count (15 CD4+ T-cell/ µl) was reported in May 2009 along with the pulmonary TB infection aforementioned. An increased HIV viral load (from 3 log RNA copies/ml in March 2010 to 4.88 log RNA copies/ml in September 2011) was recorded, and was in correlation with a declining CD4 T-cell count drop within a similar period (from 04 CD4+ T-cell/ µl in March 2010, 02 CD4+ T-cell/ µl in October 2011, to 01 CD4+ T-cell/ µl in November 2011). Following HIV-1 polymerase gene genotyping in January 2012, phylogenetic analysis revealed CRF02_AG as viral clade, with the presence of drug-resistance associated mutations in both the protease and reverse-transcriptase regions: L10V, K20I, M36I, M46I, I47V, L63R, L76V, V82F and L89I were found in the protease, with important reductions in viral susceptibility to protease inhibitors (exception made with ritonavir boosted saquinavir); A62V, L74I, V75T, M184V, A98G, V179E, and P225H were found in the reverse-transcriptase, with important reductions in drug susceptibility to NRTIs (3TC, emtricitabine [FTC] ABC, stavudine [d4T], ddI), and NNRTIs (EFV and nevirapine [NVP]). Based on the viral genotypic profile, a more effective and aggressive HAART was recommended (AZT [zidovudine] + TDF [tenofovir] + 3TC + LPV/r + SQV/r) in order to maximize the likelihood of viral suppression, then simplifying the regimen after what.

Case 2: The second case was a 15 years old male child admitted under emergency condition at the intensive care unit for 35 days and was reported with an atypical pneumonia along with a persistent cough coupled with weight loss. Regardless of a negative *MTB* culture testing, anti-tuberculosis regimen (Isoniazid [INH] and Rifampin [RMP]) was administered to the child, based on his critical clinical condition and his profound immunological deterioration and anti-HIV therapeutic failure. This patient was under Duovir (3TC+AZT) and NVP regimen since January 2008, followed by a switch to Duovir and Aluvia (LPV/r) combination. Based on genotypic resistance testing made in December 2008, phylogenetic analysis also revealed CRF02_AG as the viral strain, with the following drug resistance-associated mutations: L10V, K20I, M36I, M46L/M, T74S, L89I and L90M, found in the protease region, with a minor or major decreased drug susceptibility to protease inhibitors; V75I and M184V were found in the reverse-transcriptase, conferring major resistance to 3T, FTC, and d4T, and lesser to ABC, ddI. CD4+ T-cells recovery was not effective, decreasing from 465 (16%) CD4+ T cell/ µl in December 2008 to a chronic and severe immune-compromised state [107 (6%), 28 (4%), 43 (3%), and 58 (5%) CD4+ T cell/ µl], respectively in September 2009, April 2010, December 2010 and April 2011. The viral load remained high (5,45 log in September 2009 and 4,41 log in December 2010), favored by reported series of non-adherence.

DISCUSSION

The relation between TB and HIV has been extensively demonstrated in several previous works. However, considering the fact that both reported pediatric cases harbored MDRHIV with a pulmonary disease, further highlights the impact of a HAART failing regimens on the progression toward AIDS, and especially TB. Additionally, these cases may raise the awareness on a potential contribution of MDRHIV to the rapid reactivation of TB. On the other hand, it may be of relevance to understand the potential role played by MTB in the selection of HIV multi-resistant strains amongst children. Previous evidences have clearly established a positive association between the number of activated CD4+T cells and HIV replicative fitness and evolution [5, 6]. High viral loads are inversely proportional to the peripheral CD4 T-cell counts and correlate with susceptibility to several opportunistic infections [7, 8, and 9]. In addition, HIV infected people with <200 CD4 T cells/µl (severe immunocompromised) count were found more susceptible to TB than those with >500 CD4 T cells/µl (immunocompetent) count, irrespective to HAART [10]. Thus, the immunological and virological failures of these reported cases would have rendered them more susceptible to pulmonary TB or pulmonary -TB-like diseases. In contrast, HIV negative and immunocompetent children can as well develop TB, but to a lesser extend, and with less severity as compared to their HIV-infected peers. Furthermore, TB treatment has no direct evident effect on HIV viremia, [11] thereby supporting the idea that the peripheral viral load does not itself expressed susceptibility to TB [12]. Thus, a decrease in CD4 T-cell counts together with a high viral titer would not be enough to completely explain the evolution of TB in these two

HIV infected children. Other factors, such as the chronicity of the co-infection, may have favoured the viral evolution and the observed rapid emergence of resistant variants, which in turns accelerates the co-progression of both pathogens. Within this framework, several studies have already identified target sites of *MTB* infection known to promote HIV replication, thereby reducing the ability of these targets in containing both pathogens [12-13.] Our report supports the hypothesis by which pediatric *MTB* promotes selection of MDRHIV strains, which in turns accelerates progression to AIDS, with TB as the main opportunistic infection. Conclusively, TB-HIV co-infection remains a major challenge, especially in the pediatrics, and more importantly in a context of limited resources. The possible cross-linking between both microbial agents warrants further investigations, for a better management, prevention and the implementation of control strategies for this co-infection.

REFERENCES

1. Getahun H, Gunneberg C, Granich R, and Nunn P: *HIV infection associated tuberculosis: the epidemiology and the response*. Clin. Infect. Dis. 50(Suppl. 3):S201–S207, 2010.

2. Global Tuberculosis Control: WHO Report 2010 [WHO website]. Available at: http://whqlibdoc.who.int/publications/2010/9789241564069_eng

3. Sun L, Xiao J, Miao Q et al: *Interferon gamma release assay in diagnosis of pediatric tuberculosis: a meta-analysis.* FEMS Immunology & Medical Microbiology. 63(2): 165-173, 2011.

4. Hesseling AC, Cotton MF, Jenning T, et al: *High incidence of tuberculosis among HIV infected infants. South African population-based study*. Clin Infect Dis. 48:108-114, 2009.

5. Schneider T, Jahn HU, Schmidt W, Riecken EO, Zeitz M and Ullrich R . *Loss of CD4 T lymphocytes in patients infected with human immunodeficiency virus type 1 is more pronounced in the duodenal mucosa than in the peripheral blood. Berlin Diarrhea/Wasting Syndrome Study Group.* Gut. 37:524–529, 1995.

6. Veazey RS, Marx PA, and Lackner AA. *The mucosal immune system: primary target for HIV infection and AIDS.* Trends Immunol. 22:626–633, 2001.

7. Anonymous. *Antiretroviral therapy and medical management of pediatric HIV infection and 1997 USPHS/IDSA report on the prevention of opportunistic infections in persons infected with human immunodeficiency virus.* Pediatrics. 102(4 Pt. 2):999–1085, 1998.

8. Bucy RP, and Kilby JM. *Perspectives on inducing efficient immune control of HIV-1 replication—a new goal for HIV therapeutics?* AIDS. 15(Suppl. 2):S36–S42: 2001.

9. Merchant RH, and Quadir ZA. *Management of opportunistic infections in pediatric HIV*. Indian J. Pediatr. 69:973–977, 2002.

10. Lawn SD, Myer L, Edwards D, Bekker LG, and Wood R. *Short-term and long-term risk of tuberculosis associated with CD4 cell recovery during antiretroviral therapy in South Africa.* AIDS. 23:1717–1725, 2009.

11. Kizza HM, Rodriguez B, Quinones-Mateu M et al. *Persistent replication of human immunodeficiency virus type 1 despite treatment of pulmonary tuberculosis in dually infected subjects.* Clin. Diagn. Lab. Immunol. 12:1298–1304, 2005.

12. Diedirch CR and Flynn JL. *HIV-1/Mycobacterium tuberculosis coinfection immunology: How Does HIV-1 exacerbate tuberculosis?* .Infection and Immunity. 79: 1407–1417, 2011.

13. Toossi Z, Jonhson JL, Kanost RA et al. *Increased replication of HIV-1 at sites of Mycobacterium tuberculosis infection: potential mechanisms of viral activation.* J. Acquir. Immune Defic. Syndr. 28:1–8, 20