Author’s response to reviews

Title: Magnitude of Elevated iron stores and Risk associated in Steady state Sickle Cell Anemia Congolese children: a cross sectional study

Authors:

Jean-Robert Makulo (jrmakulo2016@gmail.com)
Karen Itokua (karenitokua@yahoo.fr)
Rosette Lepira (rosettelepira@gmail.com)
Gloria Bundutidi (gloriadea03@gmail.com)
Michel Aloni (Michelaloni2003@yahoo.fr)
Réné Ngiyulu (Relyse2000@yahoo.fr)
Jean-Lambert Gini (giniehungu@gmail.com)
François Lepira (lepslepira@gmail.com)

Version: 1 Date: 06 Jul 2018

Author’s response to reviews:

We thank the editor and all reviewers for the comments and proposals made. We took it into account to improve the article.

Editor Comments:

1. Dr. Makulo et al raised a concern that iron overloading has been underestimated after several transfusions in SCA children in Congolese. Can the authors suggest/discuss a few methods to potentially solve this problem? For example, educate the local physicians, perform serum ferritin test in SCA patients, and use iron chelator if possible, etc.

We added this proposal in the final version of the text (Discussion section, last paragraph, line 45 page 6):

Considering the results of this study, local physicians should be sensitized to avoid iterative transfusions by respecting the indications for blood transfusion in SCA, to perform serum ferritin test, in particular from more than 3 blood transfusions during the year. Indications of iron
chelation therapy must be known and systematized to prevent comorbidities associated with iron overload.

Minor comments:

1. In Abstract, please spell out abbreviations (e.g. HU, CRP) at the first time using them.

   We corrected this in the final version of the text:

   Democratic Republic of the Congo (DRC) line 27, page 3

   C Reactive Protein (CRP) line 13 page 4

   Lactate dehydrogenase (LDH) line 13 page 4

   Hydroxyurea (HU) line 48 page 4.

2. On page 4, under Materials and Methods, please use the same unit of ng/ml for serum iron/ferritin levels throughout the manuscript (and page 5 under Discussion).

   We corrected this in the final version of the text:

   500 ng/ml line 38 page 5.

3. On page 4, under Statistical analysis, (P should be < 0.05).

   We corrected this in the final version of the text:

   p < 0.05 line 43 page 4.

4. On page 5, there is no Table 2, please revise it.

   Table 2 was reported at the end of the first paragraph line 5 page 5.

5. Figure 1 is not clear, please define the meaning of the symbols, e.g. *55, *4, etc.

   Symbols *55, *4, corresponded to the patients in the group who had marginal results.

   The figure 1 has been corrected.
Reviewer reports:

Victor Gordeuk (Reviewer 1): This paper addresses the problem of increased iron stores in patients with sickle cell disease, an important topic.

1. The authors should use the term "increased iron stores" rather than iron overload throughout the paper.

We corrected this in the final version of the text. Thus the title was even modified as follows:

Magnitude of increased iron stores and Risk associated in Steady state Sickle Cell Anemia Congolese children: a cross sectional study.

2. What is the total number of sickle cell children treated at the centers included in this study? How many were excluded because of the exclusion criteria?

All SCA children between 2 and 18 years of age consecutively followed in the two centers were selected for the period from 15 June, 2014 to 30 August, 2014. Children who were transfused, hospitalized and had a major vaso-occlusive crisis within the last two months before the study had been excluded. Children with a history of liver disease or chronic alcoholism were also excluded. So, children excluded from the inclusion criteria were 24.

However, 4 children who met the inclusion criteria were excluded because of missing data.

Sickle cell children who had consulted outside the study period did not concern our sample. We do not have a precise number of children followed in these centers during a year; in DR Congo, many families do not come to medical appointments.

3. The number for each group should be included in Figure 1 and Table 2.

In the final version of the text, we added the number of patients in each subgroup.

4. On page 5 the text states: "median values of serum ferritin levels were 125 (IQ 25 and 75: 99-185) ng / ml in the group 1, 222 (IQ 25 and 75: 102-453) ng / ml in the group 2, and 301 (IQ 25 and 75: 123-747) ng / ml in the group 3 (p = 0.010)." However, Figure 1 shows a median ferritin of close to 1000 ng/ml for group 3. These presentations should be reconciled.

The figure 1 has been corrected.
5. Also on page 5, the text states: "Compared to the group 1, the probability of having a serum ferritin level > 500 ng/ml was slightly higher in the group 2 [aOR 3.08 (95% CI: 1.06-8.97)] and in group 3 [aOR 6.17 (95% CI: 1.81-20.96)]." However, odds ratios of 3 and 6 are not slightly higher but substantially higher. The text should be revised to indicate this.

We agree with the reviewer. The text is corrected as follows:

Compared to the group 1, the probability of having a serum ferritin level > 500 ng/ml was substantially higher in the group 2 [aOR 3.08 (95% CI: 1.06-8.97)] and in group 3 [aOR 6.17 (95% CI: 1.81-20.96)].

6. The authors could do an analysis of the relationship of serum ferritin with baseline characteristics using serum ferritin as a continuous variable and multiple regression analysis.

Even with multiple linear regression, the number of transfusions per year is maintained in the model. The advantage with logistic regression, ferritinemia has been evaluated at the threshold of 500 ng / ml which has an indication to give an iron chelator.

7. The strong conclusion of this paper is that increased serum ferritin is most strongly related to blood transfusion.

We agree with the reviewer. The conclusion has been modified as follows:

In SCA children, increased serum ferritin is most strongly related to blood transfusion. This situation concerned almost one in five children in present study; this shows the magnitude of the problem which is underestimated.