**Response to reviewers’ comments**

**Manuscript ID:** EJH-2020-0055

**Title:** Nonmyeloablative matched stem cell transplantation with optional reinforced stem cell infusion for hemoglobinopathies

Dear Editor,

We appreciate the editor and reviewers of “*European journal of Haematology*” for their effort and time in reviewing our manuscript. We have made some corrections and clarifications regarding the following points in the revised manuscript according to the reviewers’ comments. We are pleased that you are interested in our paper and hope that the revised manuscript will be suitable for publication in your journal. Again, we thank you for your constructive comments.

\*The numbers in the parentheses indicate the column in the marked-up version of the revised manuscript.

Sincerely,

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**Reviewer #1**

Q1. The non-myeloablative conditioning of NIH consisted of 300 cGy total-body irradiation. Please, clarify that why did you employ a range of 300–400 cGy total body irradiation? In addition, patients who received the optional reinforced stem cell infusion exposed to an additional dose of 500 cGy. The cumulative toxicities of total body or total nodal irradiation, including hypothyroidism, hepatotoxicity, and cardiomyopathy, should be mentioned.

A1. Although the non-myeloablative conditioning of NIH consisted of 300 cGy total-body irradiation, we employed 300–400 cGy. Because 400 cGy total body irradiation in many conditioning regimens of various hematologic disease was preferred in our transplantation center, we admixed 300 and 400 cGy total body irradiation. On the other hand, currently available literatures of total body irradiation toxicities suggested the dose over 1200 cGy was associated with increased incidence of late sequeles, including neurologic complication, renal complication and secondary malignancies (Crit Rev Oncol Hematol. 2018 Mar;123:138-148). In addition, total nodal irradiation was generally accepted as having less toxicities than total body irradiation. Considering these findings, only small proportion of patients in our cohort experienced late complications followed by total body and total nodal irradiation. We have added corresponding sentences as follows:

Q2. Table 1 should provide additional information: e.g. iron-chelating agents with serum ferritin level for β thalassemia major patients and hydroxyurea administration for sick cell anemia patients.

A2. According to your comment, we have added corresponding information for β thalassemia major and sickle cell anemia patients.

Q3. Gene therapy is another curative therapy for thalassemia and sickle disease. Therefore, the ‘only’ in sentence of ‘Allogeneic stem cell transplantation remains the only curative treatment option ~ hematologic disorders.’ (Page 3, Line 49) should be corrected.

A3. According to your comment, we have removed the ‘only’ in the corresponding sentence (Page 3; Line # 46).

Q4. You should mention that graft was unmanipulated in patients requiring the optional reinforced stem cell infusion.

A4. We have specified that initial and reinforced stem cells were infused without manipulation as follows: ‘, and then administered to patients without manipulation.’ (Page 4; Line # 92–93) and ‘patients received the optional reinforced unmanipulated SC infusion from the same donor after total-nodal irradiation (TNI) at a single dose of 500 cGy.’ (Page 5; Line 99-100)

Q5. Please, clarify whether the same donor or another donor was employed in your report, when patients received the optional reinforced stem cell infusion.

A5. We employed the same donor of initial peripheral blood stem cell infusion, when patients received the optional reinforced stem cell infusion as follows: ‘patients received the optional reinforced unmanipulated SC from the same donors after total-nodal irradiation (TNI) at a single dose of 500 cGy.’ (Page 5; Line # 99).

Q6. Please, describe the detailed parameters for packed red cell transfusional support of patients in your cohort.

A6. Our hemoglobin level threshold for packed red cell transfusion in patients β thalassemia and sickle cell disease was less than 7 g/dL. We have added corresponding sentences as follows: ‘Patients received packed red cell transfusion if their hemoglobin level were less than 7 g/dL.’ (Page 5; Line #115)

Q7. Did receive sickle cell disease patients in your cohort routine penicillin prophylaxis?

A7. Sickle cell disease patients in our cohort did not receive routine penicillin prophylaxis, because of they were more than 18 years old.

Q8. Peripheral blood donor T-cell chimerism at post-transplant day 30 may be not meaningful with your NMA conditioning. Would you consider removing corresponding sentences?

A8. According to your comment, we have removed the sentences of peripheral blood donor T-cell chimerism at post-transplant day 30.

**Reviewer #2**

Q1. Correct the number of patients experiencing graft-versus-host disease in the ‘ABSTRACT’, ‘RESULTS’, and ‘Figure 1’ sections.

A1.

Q2. The result that graft-versus-host disease occurred in three patients who received the optional reinforced infusion after post-transplant one year, whereas two patients who did before post-transplant 6 months did not (may be due to persistent alemtuzumab level) should be emphasized.

A2.

Q3. The paradigm considering that peripheral blood T cell chimerism > 50% should be reached before tapering immunosuppressive agent to avoid graft rejection should be confirmed by larger studies. In fact, several adult patients with sickle cell disease ceased sirolimus earlier than required and did not experience graft rejection with sustained peripheral blood donor chimerism after one year. It should be addressed.

A3.

Q4. Authors should discuss that these data suggest that optional reinforced stem cell infusion should be only proposed for patients with very early chimerism decline, because those who received the procedure had high risk of graft-versus-host disease, but low risk of graft rejection after one year.

Q4.