Reviewer's report

Title: Targeting insulin resistance in type 2 diabetes via immune modulation of cord blood-derived multipotent stem cells (CB-SCs) in stem cell educator therapy: phase I/II clinical trial

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Reviewer: Sigurd Lenzen

Reviewer's report:

Major compulsory Revisions:

1) General: The authors show in this study that in three sub-groups of patients with type 2 diabetes (T2DM) a treatment of the blood of the patients with the Stem Cell Educator and subsequent co-culture of the separated mononuclear cells with adherent CB-SCs causes an improvement of the diabetic state as documented by a decrease of the HbA1C value. The authors explain this improvement of the diabetic state both due to an improvement of insulin sensitivity as well as of beta cell function. The authors argue that this therapy success is due to a reversal of immune cell dysfunction.

2) Major comment: This is a very interesting observation; however, the weakness is that the authors can not really provide a mechanistic explanation for this improvement. In particular it is not clear, how and in which concrete way this Stem Cell Educator causes this reversal of immune cell dysfunction. This must be explained better in the discussion section.

In particular the authors should explain why it is apparently sufficient to treat the mononuclear cells in the blood only to obtain the therapy success. Why is it not necessary to modulate the behaviour of the immune cells which reside in the tissues, for example in the adipose tissue? Is it not that such untreated immune cells from tissues constantly migrate into the circulation and decrease the therapy success with time?

Further comments:

3) Results, Fig. 1: How many of the patients in each group showed an improvement of HbA1C and how many did not? This should be mentioned additionally in the results section.

4) Discussion, Fig. 1: The authors should discuss why the improvement of insulin sensitivity and beta cell function progresses slowly over weeks. Rather one would expect that the observed improvement vanishes again with progression of time after treatment?

5) Results, page 13: The authors found after Stem Cell Educator therapy no change in such important cytokines like IL-1, IL-6 and TNF. The only major improvement they found was a suppression of the cytokine TGF. Can this suppression explain the complete therapy success or is the therapy success also the result of reduction of other cytokines such as IL-17, IL-12, IL-4 and IL-5? This
should be discussed properly in the discussion section and put in relation to references from the literature. It is unlikely that it is only a result of a change of CD-86.

6) Results, page 16: The changes in NO production and its relation to the observed effects should be properly discussed in a mechanistic perspective in the discussion section.

7) Discussion, page 17, 2nd para: Is there experimental evidence that the Stem Cell Educator therapy also reduced chronic inflammation of VAT?

8) Discussion, page 17, 2nd para: The authors consider abnormalities in monocytes/macrophages as of crucial importance for chronic inflammation and insulin resistance in T2DM and they cite in this context references 27 and 30. If this is really the case, these cells should show signs of activation, i.e. expression of proinflammatory cytokines and others mediators. Can the authors provide experimental evidence for this?

9) Page 18, last line: Can the authors please specify, to which restoration of monocyte function they specifically refer here with respect to the therapy success?

10) Page 16, line 5: Typo: co-cultured.

Page 18, line 6: Typo: unique therapy success.

**Quality of written English:** Acceptable

**Statistical review:** No, the manuscript does not need to be seen by a statistician.

**Declaration of competing interests:**

NO competing interests.

I do not perform studies myself.