Transplantation of Human dgHPSCs Overexpressing Human Insulin/ERRγ Genes Can Not Only Decrease the Glucose Levels but Also Decrease the Hypertension in T2D Patient (Case #1-B)

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ABSTRACT

Transplantation of directly-generated human pluripotent stem cells (dgHPSCs) overexpressing human insulin (INS) and/or estrogen-related receptor γ (ERRγ) genes can efficiently improve the symptoms of type 2 diabetes (T2D), such as the glucose (GLU) and glycosylated haemoglobin (HbA1c) levels, diabetes-derived complications, etc. Therefore, human stem cell transplantation might be a potential strategy to replace exogenous INS administration, and further to prevent the patients from developing diabetic complications. However, little is known concerning the efficacy duration of the transplanted stem cells. Here, we reported the following-up visits of one patient for a year. We found that the patient not only maintained almost normal blood glucose levels, but also his blood pressure decreased to normal range without taking anti-hypertensive drugs. Thus, our data indicated that the efficacy duration of transplanted stem cells can last about two years. At the same time, the transplanted stem cells can not only decrease the blood glucose levels, but also reduce the hypertensions in T2D patient.

Keywords
dgHPSCs, ERRγ, INS, Human T2D, Hypertension.

Introduction

With the widespread of diabetes, particularly type 2 diabetes (T2D), it becomes a great threat for human health and lifespan. Even worse, the current therapies for diabetes including insulin (INS) administration only have limited effects in preventing the processing of diabetes-derived complications [1,2]. So far, many fundamental investigations in human stem cell biology already substantially performed, such as the induction of human pluripotent stem cells (iPSCs) [3], the successful production of human pancreatic β-like cells from human embryonic stem cells (ESCs) and iPSCs, etc. [4,5], the major concerns regarding tumour...
formation and immune rejection of transplanted human stem cells greatly impede the translation from human stem cell biology into clinical medicine [6,7].

Previously, we reported that transplantation of directly generated human pluripotent stem cells (dgHPSCs) overexpressing INS and/or ERRγ genes can efficiently decrease the blood glucose (GLU) and glycosylated haemoglobin (HbA1c) levels, and significantly improve the physical and mental conditions of T2D patients [8-10]. But how long is the efficacy duration of these transplanted stem cells? Although little is known, it is a critical question we must answer.

To investigate the efficacy duration of transplanted stem cells, we request the recipient patient to monitor his fasting capillary blood glucose (FFT-CBG) daily, and also keep his close eyes on the other possible changes of his health. The follow-up visits revealed that the efficacy duration for controlling the GLU levels can last almost two years. More surprisingly, after about one and a half years of transplantation, the patient’s hypertension decreased to normal range without taking anti-hypertensive drugs. The patient will continue to monitor his GLU levels and hypertension changes subsequently. Our data demonstrated that the efficacy duration of transplanted stem cells can last at least about two years. And besides decreasing the blood GLU levels, the stem cells can also reduce the hypertensions in T2D patient.

Materials and Methods

Statement of Ethical Approval
The treatments for the patients and the use of human stem cells were approved by the Ethics Committee of Interventional Hospital of Shandong Red Cross Society (Shengjieyi 2003, No. 26) in compliance with Helsinki Declaration. The Ethics Committee of Interventional Hospital of Shandong Red Cross Society approved this clinical study and treatments. The participants provided their written confirmed consent to participate the clinical study and treatments. The Ethics Committee of Interventional Hospital of Shandong Red Cross Society approved this consent procedure. All the treatments for the patients and use of human stem cells were performed in accordance with the guidelines established in Interventional Hospital of Shandong Red Cross Society approved by the Ethics Committee. After traditional daily INS injection for about four years, the patient agreed to try the stem cell therapy with overexpression of INS and ERRγ genes in our hospital to treat and cure his diabetes and coronary heart disease complications. All these stem cells were isolated and proliferated with the written confirmed consent of the participants [8,10].

Patient Case
The detailed description of this patient, T. S. D., was described in References [8,10]. After he finished the stem cell transplantation therapy at June 30 of 2018, he kept monitoring his FFT-CBG levels daily (Table 1), and tried his best to keep diabetic diet and exercise physically. He felt his body was full of strength. Sometimes, he would walk about 10,000 to 15,000 steps daily. Some other times, he would do other exercises, such as whipping a top for one to two hours. From June 1st of 2019 on, he stopped taking anti-hypertensive drugs, and his blood pressure was kept around the normal range (Tables 2 and 3).
The decrease of blood pressure after human stem cell transplantations without taking anti-hypertensive drugs.

**Table 1:** Daily monitoring of FFT-CBG levels (from 23/10/2018 to 15/10/2019).

<table>
<thead>
<tr>
<th>Date (dd/mm/yyyy)</th>
<th>Blood pressure (Relax/contract)</th>
<th>Date (dd/mm/yyyy)</th>
<th>Blood pressure (Relax/contract)</th>
<th>Date (dd/mm/yyyy)</th>
<th>Blood pressure (Relax/contract)</th>
</tr>
</thead>
<tbody>
<tr>
<td>15/09/2019</td>
<td>70/110</td>
<td>16/09/2019</td>
<td>70/120</td>
<td>17/09/2019</td>
<td>65/105</td>
</tr>
<tr>
<td>18/09/2019</td>
<td>65/110</td>
<td>19/09/2019</td>
<td>75/120</td>
<td>20/09/2019</td>
<td>70/110</td>
</tr>
<tr>
<td>27/09/2019</td>
<td>70/110</td>
<td>28/09/2019</td>
<td>65/100</td>
<td>29/09/2019</td>
<td>60/95</td>
</tr>
<tr>
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<td>60/100</td>
<td>01/10/2019</td>
<td>60/100</td>
<td>02/10/2019</td>
<td>75/125</td>
</tr>
<tr>
<td>03/10/2019</td>
<td>70/120</td>
<td>04/10/2019</td>
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</tr>
<tr>
<td>06/10/2019</td>
<td>70/110</td>
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<td>70/120</td>
</tr>
<tr>
<td>09/10/2019</td>
<td>80/140</td>
<td>10/10/2019</td>
<td>75/125</td>
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<tr>
<td>15/10/2019</td>
<td>78/135</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Table 2:** The decrease of blood pressure after human stem cell transplantations without taking anti-hypertensive drugs.

**Cell preparation**

Refer to the references [8-10].

**Lentivirus vector (LV) construction, production and infection**

Refer to the references [8-15].
After transplantation demonstrated that the transplanted human stem cells not only can decrease blood pressure to normal range (Tables 2 and 3). These data suggested that the transplanted human stem cells can maintain the blood GLU levels around the normal range, but also can improve the patient’s cardiovascular function and decrease the hypertension.

The follow-up visits of the patient
His body weight keeps around 68kg, and he feels strong. He adheres to diabetic diet fairly well. He persists in exercising every day, sometimes, he walked about 10,000 to 15,000 steps daily, at other times, he did some other exercises, such as whisking a top. If he decreased his exercises due to the weather, etc., the next day fasting blood glucose would increase a little bit. Or when he caught a cold and had less exercises, his fasting glucose also would go up somewhat. When he had a feast with his friends and had more food, his fasting glucose levels would also rise up to some extent. Mostly, he could keep his fasting blood glucose levels within normal range (Table 1). As mentioned above, he stopped taking anti-hypertensive drugs from June 1 of 2019 until present, and his blood pressure kept within normal range (Tables 2 and 3). The comparison between before stem cell transplantation and after transplantation was listed in Table 3.

Results
The efficacy duration of transplanted dgHPSCs-INS/ERRγ cells can last almost two years.

In order to investigate the efficacy duration of transplanted dgHPSCs overexpressing human INS and/or ERRγ genes, the patient was requested to monitor his FFT-CBG levels daily (Table 1) [8-10]. And from June 1 of 2019 on, he stopped taking anti-hypertensive drugs. From September 15 of 2019 on, he began to measure is blood pressure every day (Tables 2 and 3).

Follow-up visits
In order to investigate the efficacy duration of the transplanted human stem cells overexpressing human INS and/or ERRγ genes, the patient was requested to monitor his FFT-CBG levels daily (Table 1) [8-10]. And from June 1 of 2019 on, he stopped taking anti-hypertensive drugs. From September 15 of 2019 on, he began to measure is blood pressure every day (Tables 2 and 3).

<table>
<thead>
<tr>
<th>Disease and complications</th>
<th>Treatment before transplantation</th>
<th>After transplantation</th>
</tr>
</thead>
<tbody>
<tr>
<td>T2D</td>
<td>24 IU INS were injected daily, 12 IU in the morning, 12 IU at evening [8,10].</td>
<td>Totally stopped INS injection.</td>
</tr>
<tr>
<td>Diabetes-derived coronary heart disease and myocardial infarct</td>
<td>Three cardiac stents were implanted [8,10].</td>
<td>Myocardial Infarct disappeared, coronary heart disease improved significantly.</td>
</tr>
<tr>
<td>Hypertension</td>
<td>Taking Benazepril Hydrochloride Tablets, 10 mg/Tablet, half tablet per day.</td>
<td>Stopped taking Benazepril Hydrochloride Tablets, the blood pressure became normal.</td>
</tr>
</tbody>
</table>

Refer to references [8,10].

References
10. Wang T, Wang X, Zhang B, et al. Transplantation of human dgHPSCs overexpressing insulin and ERRγ can efficiently decrease the glucose and HbA1c levels, increase the secretion of C-peptide, and repair the complications of coronary heart


