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Efficacy of Autologous Bone Marrow–Derived Stem Cell Transplantation in Patients With Type 2 Diabetes Mellitus

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Progressive and inexorable β-cell dysfunction is the hallmark of type 2 diabetes mellitus (T2DM) and β-cell regeneration using stem cell therapy may prove to be an effective modality. A total of 10 patients (8 men) with T2DM for >5 years, failure of triple oral antidiabetic drugs, currently on insulin (≥0.7 U/kg/day) at least for 1 year, and glutamic acid decarboxylase antibody negative were included. Patients on stable doses of medications for past 3 months were recruited. Primary end points were reduction in insulin requirement by ≥50% and improvement in glucagon-stimulated C-peptide levels at the end of 6 months of autologous bone marrow–derived stem cell transplantation (SCT), while secondary end points were a change in weight and HbA1c and lipid levels as compared to baseline. Seven patients were responders and showed a reduction in insulin requirement by 75% as compared to baseline. Mean duration to achieve the primary objective was 48 days. Three patients were able to discontinue insulin completely, although it was short-lived in one. Mean HbA1c reduction was 1% and 3 of the 7 responders had HbA1c value <7%. A significant weight loss of 5.5 kg was noted in the responders, whereas, nonresponders gained 2.2 kg of weight. However, weight loss did not correlate with reduction in insulin requirement ($r = 0.68$, $P = 0.06$). There was a significant improvement in both fasting and glucagon-stimulated C-peptide level in the group ($P = 0.03$) and responders ($P = 0.03$). HOMA-B increased significantly in the whole group ($P = 0.02$) and responders ($P = 0.04$) whereas, HOMA-IR did not change significantly ($P = 0.74$). Reduction in insulin doses correlated with stimulated C-peptide response at the baseline ($r = 0.83$, $P = 0.047$) and mononuclear cell count of infused stem cells ($r = 0.57$, $P = 0.04$). No serious adverse effects were noted. Our observations indicate that SCT is a safe and effective modality of treatment to improve β-cell function in patients with T2DM. However, further large-scale studies are needed to substantiate these observations.

Introduction

Type 2 diabetes mellitus (T2DM) is characterized by 2 defects namely progressive and inexorable β-cell dysfunction superimposed on insulin resistance [1]. The consequent hyperglycemia results in micro- and macrovascular complications with high morbidity and mortality [1]. At diagnosis of T2DM, almost 50% of β-cell mass is lost and it further declines with the increasing duration of disease, whereas insulin resistance remains fairly constant [2–4]. Currently available therapeutic regimens either target insulin resistance or insulin deficiency [5]. None of these modalities modulate the course of the disease and eventually the patients require insulin therapy for optimal glycemic control [6]. Insulin therapy is not physiological as there is no hepatic "first-pass" metabolism of insulin [7], which is required to knock down hepatic glucose output. Even with intensive insulin therapy, patients exhibit variable periods of hyperglycemia or hypoglycemia when studied using 24-hour glucose profile [8] leading to detrimental cardiovascular and psychological effects. Improvement in endogenous β-cell reserve can overcome these deficiencies in existing modalities.

There are animal and experimental data to suggest augmentation of β-cell mass with the use of thiazolidinediones (TZDs), glucagons-like polypeptide-1 (GLP-1) analogues, and dipeptidyl peptidase-IV (DPP-IV) inhibitors [9–11]; however,

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such data in human beings are lacking. Therefore, it is imperative to explore new strategies for β-cell replacement.

Existing strategies for β-cell replacement include simultaneous pancreatic or islet cell transplantation, which are expensive, labor intensive, limited, and restricted to a few specialized centers [12,13]. Their experience in patients with T2DM is also limited. Stem cells are primitive cells capable of self renewal and differentiation into cells of other lineage. Stem cell therapy offers a new paradigm in the management of T2DM after its success in various experimental and animal studies [14–17] and in an elegant study by Voltarelli et al. [18], though in patients with type 1 diabetes mellitus (T1DM). Bone marrow is an important and easily accessible source of adult stem cells. Use of autologous bone marrow–derived stem cells is safe and devoid of any ethical issues.

We hypothesized that autologous bone marrow–derived stem cell transplantation (SCT) into the pancreas of patients with T2DM will lead to reduction or abolition of insulin requirement.

Materials and Methods

Patients

A total of 90 patients were screened from November 2007 to August 2008 out of which 42 were eligible for this study. Finally, 10 patients who fulfilled the criteria and consented for the study were included, giving a power of 95% at the confidence interval of 95% to our study. The protocol was approved by the ethics committee of the institute.

The inclusion criteria were patients with T2DM between 30 and 75 years of age, duration of diabetes >5 years, failure of triple oral antidiabetic drugs (a combination of sulfonylurea, metformin, and TZD in optimal doses), requiring insulin for optimal glycemic control in a dose of ≥0.7 U/kg/day [19,20] at least for 1 year, having glutamic acid decarboxylase (GAD) antibody negative status, and willingness to participate in the study. At the time of entry into the study, all the patients were on stable dose of insulin, metformin (2 g/day), and pioglitazone (30–45 mg/day) for the past 3 months. A run-in period of 2 weeks was given after entry during which patients were counseled about the procedure and investigations were carried out. Patients were excluded from the study if they had T1DM, serum creatinine >1.5 mg/dL, abnormal liver function tests, active infections, malignancy, or acute coronary syndrome in the previous 3 months.

Clinical examination

A detailed clinical examination was carried out at the baseline, which entailed blood pressure (BP), body mass index (BMI), waist line, body fat estimation (Omron, HBF-302, Japan), and workup for micro- and macrovascular complications. Baseline investigations included complete hemogram, liver and renal function tests, serum electrolytes, and lipid profile. HbA1c level was measured using Bio-Rad D10 (Bio-Rad Laboratories Inc, Hercules, CA, USA; normal range: 3.8%–5.9%). Glucagon-stimulated C-peptide level was estimated in a fasting state [21] after intravenous administration of glucagon (1 mg; GlucaGen, Novo Nordisk, Denmark) and drawing blood samples at –15, 0 (mean taken as fasting value), and 6 min (stimulated value) of injection. Estimation was done using Elecsys 2010 (Roche Diagnostics, GmbH, Mannheim, Germany; normal range: 1.1–4.4 ng/mL). GAD antibody status was assessed using an immunoblot assay. During the 2-week run-in period patients were asked to keep a record of self-monitored blood glucose (SMBG) values including at least a 5-point profile (fasting, three 2-h postmeal and 3 AM values) for 3 days. Fasting plasma insulin was assayed in a sample drawn after omitting NPH insulin for at least 36 h and regular insulin for at least 18 h. Homeostatic model assessment was applied for the assessment of HOMA-IR (insulin resistance) and HOMA-B (β-cell function) [22].

Stem cell harvesting

Stem cells were harvested using autologous bone marrow obtained through posterior superior iliac spine approach under local anesthesia. Approximately 150 mL of bone marrow was harvested. Mononuclear cells (MNCs) were separated by ultracentrifugation after layering on density-gradient medium (Ficoll–Hypaque), washed using phosphate-buffered saline, and resuspended in heparinized normal saline (final volume: 5 mL). Aliquots were taken from the above sample for total nucleated cell count, MNC count, viability testing using trypan blue, CD 34+ cell count by flow cytometry and sterility testing.

Stem cell transplantation

A 5F catheter was navigated through transfemoral route into gastroduodenal artery beyond the origin of cystic artery and superselective injection of stem cells was carried out. After SCT, a diagnostic run was taken to look for the patency of gastroduodenal artery. After the procedure the patients were observed for 72 h for any procedure-related complications (local bleeding, hematoma formation, examination of distal pulses), serial estimation of hemoglobin, 5-point profile monitoring of blood glucose values, and insulin requirement.

Follow up

Following discharge, patients were followed up at 2 weekly intervals for first 2 months and, thereafter, monthly for next 4 months. Patients were asked to perform SMBG (at least 15 values in a month including a 5-point profile) on various days during the period of follow up. Target plasma glucose values were as follows: fasting: between 70 and 130 mg/dL, postmeal: <180 mg/dL, and HbA1c: <7% [23]. Patients were asked to contact a single investigator (VU) telephonically for insulin dose adjustment. A standard counseling regarding diet control and regular exercise was given to all patients on all the visits. The tests carried out at 3 months of follow up were routine biochemistry, basal C-peptide in a fasting state, lipid profile, and HbA1c estimation. At 6 months of follow up, following tests were performed: biochemistry, glucagon-stimulated C-peptide level, fasting plasma insulin, lipid profile, HbA1c, 24-h urinary protein, and microalbumin estimation.

Outcomes

Primary end points were a reduction in insulin requirement by ≥50% and improvement in glucagon-stimulated C-peptide levels at the end of 6 months of SCT, while,

Table 1. Baseline Clinical and Biochemical Parameters

SN	*Age (yrs)*	*Sex*	*Duration of DM (yrs)*	*Duration of insulin therapy (yrs)*	*Dose of insulin U/d (U/kg/d)*	*Duration of HTN (yrs)*	*Weight (kg)*	*BMI (kg/m²)*	*Waist (cm)*	*Body fat (%)*	*FPG (mg/dL)*	*HbA1C (%)*	*Triglycerides (mg/dL)*	*HDL-C (mg/dL)*	*LDL-C (mg/dL)*
1	50	M	15	5.00	80 (1.1)	15	75.00	28.20	97.00	31.70	149	8.9	209	40	45
2	58	M	10	3.00	75 (1.3)	4	59.00	20.40	79.00	25.70	127	8.0	185	40	80
3	54	M	17	10.00	65 (0.78)	17	83.00	24.80	93.50	23.80	105	8.4	70	36	77
4	66	M	5	4.50	65 (0.86)	0	76.00	26.60	92.00	34.60	136	8.6	100	42	83
5	55	F	7	3.00	59 (0.76)	3	77.30	26.68	104.00	42.10	118	8.2	240	39	127
6	60	M	23	4.00	58 (0.89)	6	65.00	31.80	93.00	32.60	131	8.7	104	28	77
7	68	F	12	11.50	44 (0.80)	3	55.00	22.60	82.00	36.70	161	9.3	78	49	108
8	53	M	27	7.00	54 (0.70)	25	77.10	26.70	92.00	31.70	121	8.1	73	47	47
9	59	M	22	5.00	120 (1.35)	20	88.90	28.40	102.00	29.60	125	7.2	131	43	60
10	52	M	8	3.00	74 (0.83)	8	88.80	30.40	97.00	25.20	192	7.9	217	44	144
Mean ± SD	57.5 ± 5.9		14.6 ± 7.5	5.6 ± 3	69.4 ± 6.6* (0.93 ± 0.2)	10.1 ± 2.7*	74.5 ± 11.6	26.5 ± 3.4	93.2 ± 7.8	31.4 ± 5.7	136.5 ± 25.1	8.4 ± 0.6	140.7 ± 65.5	40.8 ± 5.9	84.8 ± 32.7

*Mean ± SEM.

Abbreviations: BMI, body mass index; DM, diabetes mellitus; FPG, fasting plasma glucose; HDL-C, high-density lipoprotein cholesterol; HTN, hypertension; LDL-C, low-density lipoprotein cholesterol; MPG, mean plasma glucose; SN, subject number.

secondary end points were a change in weight, HbA1c, and lipid levels as compared to baseline.

Statistical analysis

The statistical program for the Social Sciences (Release 10.01, PC Windows; SPSS Inc., Chicago, IL) was used for the data analysis. Data were expressed as mean ± SD, until otherwise specified. Baseline and post treatment data were compared using Friedman test and Wilcoxon signed ranks test for tests of significance and linear regression analysis was used to find correlation between independent variables. A probability (P) value of <0.05 was regarded statistically significant.

Results

A total of 10 patients (8 men) were included in this study. Their baseline demographic and biochemical parameters are summarized in Table 1. At presentation, 9 (90%) patients had neuropathy, 8 (80%) retinopathy, and 4 (40%) patients had nephropathy [microalbuminuria in 3 (75%) and overt proteinuria in 1 (25%)]. Nine (90%) patients had hypertension, whereas none had any macrovascular complications. The mean dose of bone marrow harvested for SCT was 156.0 ± 4.9 mL (Table 2), which yielded a mean of $3.5 \pm 1.4 \times 10^8$ MNC. In one patient, gastroduodenal artery could not be cannulated due to an atherosclerotic plaque obscuring the origin of celiac axis and stem cell infusion was carried out in the superior mesenteric artery.

After SCT, 7 (70%) patients fulfilled the primary objective, that is, reduction of insulin requirement by ≥50% (henceforth called responders), while the remainder 3 (30%) were nonresponders. The mean duration (±SEM) required for 50% reduction in insulin requirement was 47.7 ± 15.8 (range: 7–104) days. Salient clinical and biochemical parameters 6 months after SCT are enumerated in Table 3.

There was a significant reduction in the insulin requirement with a mean of 41.2 (59.5%) units/day in the whole group ($P = 0.007$) and 56.4 (75%) units/day in the responders ($P = 0.02$; Table 4). Three patients were able to discontinue insulin completely on days 7, 41, and 111. However, in the third patient it had to be restarted after 3 weeks of discontinuation, though at a lower dose. HbA1c showed a significant decline from 8.1 ± 0.2% to 7.3 ± 0.4% in the responders ($P = 0.04$) and a nonsignificant reduction in nonresponders (from 8.9 ± 0.2% to 7.5 ± 0.2%; $P = 0.11$). At the end of the study, 3 patients (all responders) had HbA1c values of <7%. Fasting plasma glucose (FPG) reduced nonsignificantly in the whole group from 136.5 ± 25.1 to 119.1 ± 21.3 ($P = 0.059$), in the responders from 133.9 ± 28.8 to 112.3 ± 4.6 ($P = 0.063$) as well as in the nonresponders from 142.7 ± 16.1 to 134.7 ± 37.8 ($P = 0.59$). Data of individual patients are represented in Figure 1. Both fasting and glucagon-stimulated C-peptide levels rose significantly in the responders ($P = 0.03$) but not in nonresponders ($P = 0.99$). Parameters of lipid profile showed an apparent improvement but failed to achieve a statistical significance, nor did 24-h urinary protein excretion or microalbuminuria. HOMA-B increased significantly both in the entire group from 46.1 ± 14.0 to 174.5 ± 52.9 ($P = 0.02$) and in the responders from 37.7 ± 18.4 to 154.5 ± 74.3 ($P = 0.043$), whereas it did not achieve a statistical significance in the nonresponders from 67.3 ± 9.8 to 224.5 ± 2.8 ($P = 0.18$). HOMA-IR did not change significantly after SCT (Table 4).

Table 2. Details of the Procedure

Parameter	*Comment*
Volume of marrow aspirated (mL)	156.0 ± 4.9
Aspiration	
Unilateral	8
Bilateral	2
MNC count ($\times 10^8$ cells/mL)	3.5 ± 1.4
CD 34+ cell count ($\times 10^6$ cells/mL)	3.1 ± 1.4
Procedure time (min)	67.3 ± 25.8
Artery in which stem cells were injected	
Pancreaticoduodenal	9
Superior mesenteric	1
Catheter change required	2

Abbreviation: MNC, mononuclear cell.

At the end of the study, there was a weight loss of 3.1 kg in the whole group ($P = 0.012$) and 5.5 kg in the responders ($P = 0.018$), whereas nonresponders gained 2.2 kg of weight ($P = 0.11$). However, no correlation was observed with weight loss either at the end of the study ($r = 0.68$, $P = 0.06$) or till achievement of primary endpoint ($r = 0.59$, $P = 0.07$). Moreover, 50% decrease in insulin requirement preceded weight loss in 4 patients (within 10 days in 3). Waistline ($P = 0.04$) and body fat ($P = 0.02$) also decreased significantly in the responders. Systolic but not diastolic blood pressure reduced significantly ($P = 0.04$) in the entire group; however, it was not significant in the subgroup analysis. Another, interesting observation made was the reduction in dosage of the antihypertensive drugs in 5 of 9 hypertensive subjects.

Using linear regression analysis, predictors of response were stimulated C-peptide response at baseline ($r = 0.83$, $P = 0.047$) and MNC count ($r = 0.57$, $P = 0.04$). A trend toward negative correlation was observed with the dose of insulin/kg body weight at the baseline ($r = -0.40$, $P = 0.051$).

Adverse events

Adverse events noted were transient self-limiting nausea in 6 patients (in 4 following glucagon injection and in 2 after SCT) and vomiting occurred in 1 patient after SCT. One patient developed injection site hematoma after SCT. A drop of hemoglobin from 14.7 to 12.7 g/dL was seen in one at the end of first month and from 12.6 to 10.7 g/dL in another patient in the third month, improving spontaneously in both within a month. None of the patients developed major hypoglycemia, whereas, 76 episodes of minor hypoglycemia were noted. Of these, 18 were seen in responders and 58 in nonresponders ($P = 0.012$). Self-limiting upper respiratory tract infection was seen in one patient.

Discussion

This study examines the benefit of targeted autologous bone marrow–derived SCT as β-cell replacement therapy in patients with T2DM. It showed that after single administration, SCT was able to bring down the daily requirement of insulin dosage by 75% in 70% of patients. SCT resulted in reduction in HbA1c by 1.1% and improvement in glucagon-stimulated C-peptide level. A significant weight loss was also noted; however, decrease in insulin requirement did

Table 3. Clinical and Biochemical Parameters 6 Months Following Stem Cell Transplantation

SN	Dose of insulin U/d (U/kg/d)	Weight (kg)	BMI (kg/m²)	Waist (cm)	Body fat (%)	FPG (mg/dL)	HbA1C (%)	Triglycerides (mg/dL)	HDL-C (mg/dL)	LDL-C (mg/dL)
1	0 (0)	68.2	25.67	87	30.5	116	8.1	96	37	30
2	0 (0)	54.8	18.96	74	22.8	106	7.6	155	46	76
3	10 (0.14)	72.6	21.68	82	18.4	112	8.2	104	48	83
4	52 (0.67)	77.3	27.06	97	36.9	123	7.2	116	45	76
5	18 (0.24)	73.5	30.20	94	40.8	113	5.9	173	48	88
6	66 (0.97)	68.0	26.56	96	36.1	104	7.7	103	33	96
7	32 (0.56)	57.3	23.55	80	39.4	177	7.6	134	48	76
8	20 (0.29)	68.4	23.67	81	31.1	120	6.3	107	47	44
9	57 (0.65)	87.4	27.90	104	26.7	110	6.8	162	52	30
10	27 (0.32)	85.6	29.27	96	24.2	109	7.7	101	41	86
Mean ± SD	28.2 ± 7.4* (0.38 ± 0.1)	71.3 ± 10.6	25.5 ± 3.5	89.1 ± 9.6	30.7 ± 7.6	119.1 ± 21.3	7.3 ± 0.8	125.1 ± 28.7	44.5 ± 5.8	68.5 ± 24.7

*Mean ± SEM.

Abbreviations: BMI, body mass index; FPG, fasting plasma glucose; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; MPG, mean plasma glucose; SN, subject number.

not correlate with weight loss. Other effects observed concurrently were decrease in doses of antihypertensive agents and nonsignificant improvement in lipid profile.

Previously reported studies indicate that bone marrow–derived stem cells (BMSC) can differentiate into islet or pancreatic ductal cells and can lead to reduction in blood glucose in animal models rendered diabetic by the use of streptozotocin [14–16,24]. Banerjee et al. have shown reversal of diabetes and appearance of new pancreatic islets after multiple (≥3) administrations of SCT in mice [17]. A study in newly diagnosed T1DM also shows that autologous nonmyeloablative hematopoietic SCT along with high-dose immunosuppression resulted in prolonged insulin independence in majority of patients [18]. However, these studies have used either experimental models or patients of T1DM, which is fundamentally a different disorder from T2DM. T-cell-mediated autoimmune response against β-cells is the main disease mechanism in T1DM, whereas insulin resistance is the key metabolic abnormality that is superimposed on genetically susceptible β-cells in T2DM. However, regardless of inciting events, the final pathway of β-cell injury is common to both these forms of diabetes resulting from glucotoxicity, lipotoxicity, and cytotoxic cytokines [25]. This leads to similar damage and local microenvironment in the pancreas in both the disorders. Improvement with anakinra, interleukin-1-receptor antagonist, in patients with T2DM has further strengthened this hypothesis [26]. Hence, use of SC in patients with T2DM should be as effective as in those with T1DM. Data of SCT in animal models of T2DM are lacking. Unpublished reports of use of SC in patients with T2DM [27,28] are available on the World Wide Web, which further strengthens our hypothesis.

There was a significant reduction in daily insulin requirement in the group as a whole and responders. Although fasting plasma glucose level declined, it did not reach statistical significance. Still, a significant reduction was observed in HbA1c (−1.1%) and 3 of the 7 responders achieved a target HbA1c of <7%, which can be attributed to decreased glycemic excursions as a result of improved β-cell reserve, evidenced by improvement in C-peptide response similar to that described in the literature [18] and HOMA-B level, in our patients. Improvement in β-cell reserve could have occurred either due to β-cell regeneration by various mechanisms described later and/or functional recovery of residual β-cells as reduction in insulin doses correlated with higher stimulated C-peptide values at baseline of the responders and frequent monitoring and lifestyle modification could have resulted in improvement in residual β-cell function as a result of improved metabolic profile in them.

Decrease in insulin doses did not correlate with weight loss; in fact, a 50% reduction in insulin doses occurred within 10 days following SCT in 3 patients, early enough to be attributed to any lifestyle modification. Weight loss in the responders can be explained by reduction in insulin requirement, whereas, nonresponders gained weight due to the anabolic effects of insulin, recurrent hypoglycemia, and defensive eating to prevent hypoglycemia [29,30]. Any treatment modality that can improve glycemic control while leading to weight loss can be seen as a major boon and SCT scores well along with incretin-based therapies [11] in this respect.

Incidence of hypoglycemia was higher in the nonresponders ($P = 0.012$), which is likely to have contributed to weight gain that is similar to the data of DCCT and UKPDS in the intensive treatment group [29,30]. Less hypoglycemia in responders is ascribed both to reductions in insulin dosage and improved endogenous insulin secretion, corroborated by improvement in C-peptide response.

Our observation of improvement in the lipid profile and reduction in dosage of antihypertensives can be attributed to weight loss, lifestyle modification, and improved glycemic control in these subjects, though the study was not designed to examine these parameters.

Most of the available studies have injected SC [17,18] in a peripheral intravenous line, whereas, targeted approach of direct injection of SC into the celiac axis has been described to be effective leading to homing of stem cells into pancreas in one animal study [31]. We and others (unpublished reports) [27,28] have shown good results following a targeted approach, which has the theoretical advantage of delivering a bolus dose of SC and various growth factors to the affected site, thereby providing maximum stimulation to the various regenerative mechanisms; however, at present it is unclear whether peripherally administered SC can give similar results or which of the 2 approaches is superior in patients with T2DM, as such studies are not available.

Bone marrow contains various subpopulation of stem cells including hematopoietic and nonhematopoietic SC including mesenchymal stem cells (MSCs) and endothelial

Table 4. Comparison of Clinical and Biochemical Parameters Before and After Stem Cell Transplantation

Parameter	*Baseline*	*3 months*	*6 months*	*P value*[a]
Weight (kg)				
Group ($n = 10$)	74.5 ± 11.6	72.3 ± 10.6	71.3 ± 10.6	0.047
Responders ($n = 7$)	78.4 ± 10.3	74.1 ± .11.2	72.9 ± 11.1	0.02
Nonresponders ($n = 3$)	65.3 ± 10.5	68.2 ± 9.8	67.5 ± 10	0.11
BMI (kg/m^2)				
Group	26.5 ± 3.4	25.7 ± 3.4	25.5 ± 3.3	0.06
Responders	27.2 ± 3.8	25.7 ± 4.0	25.3 ± 4.2	0.02
Nonresponders	24.9 ± 2.1	26.0 ± 1.8	25.7 ± 1.9	0.11
Waist (cm)				
Group	93.2 ± 7.8	89.7 ± 9.5	89.1 ± 9.6	0.11
Responders	94.9 ± 8.2	88.3 ± 9.1	88.3 ± 10.3	0.04
Nonresponders	89.0 ± 6.1	86.3 ± 12.5	91.0 ± 9.5	0.29
Body fat (%)				
Group	31.4 ± 5.6	31.5 ± 6.7	30.7 ± 7.6	0.44
Responders	30.0 ± 6.2	29.1 ± 6.6	27.8 ± 7.2	0.02
Nonresponders	34.6 ± 2.1	37.1 ± 2.5	37.5 ± 1.7	0.11
SBP				
Group	130.0 ± 11.6	127.0 ± 12.5	120.8 ± 8.3	0.04
Responders	127.5 ± 11.6	126.3 ± 9.1	119.5 ± 8.7	0.11
Nonresponders	130.0 ± 17.3	123.3 ± 23.1	113.3 ± 5.7	0.18
DBP				
Group	80.1 ± 4.7	78.3 ± 6.3	75.0 ± 5.2	0.1
Responders	78.5 ± 4.6	77.3 ± 4.8	75.7 ± 5.3	0.32
Nonresponders	83.3 ± 5.8	76.7 ± 6.7	73.3 ± 5.8	0.18
Insulin requirement/day (U)[b]				
Group	69.4 ± 6.6	33.0 ± 8.8	28.2 ± 7.4	0.007
Responders	75.3 ± 8.2	27.3 ± 11.3	18.9 ± 7.4	0.02
Nonresponders	55.7 ± 6.2	46.3 ± 11.9	50.0 ± 9.9	0.29
FPG (mg/dL)				
Group	136.5 ± 25.1	126.1 ± 35.1	119.1 ± 21.3	0.059
Responders	133.9 ± 28.8	111.4 ± 11.9	112.3 ± 4.6	0.063
Nonresponders	142.7 ± 16.1	160.3 ± 51.1	134.7 ± 37.8	0.59
HbA1C (%)[b]				
Group	8.4 ± 0.6	7.4 ± 1.0	7.3 ± 0.8	0.009
Responders	8.1 ± 0.2	7.2 ± 0.4	7.3 ± 0.4	0.04
Nonresponders	8.9 ± 0.2	8.0 ± 0.4	7.5 ± 0.2	0.11
Fasting C-peptide (ng/mL)[b]				
Group	0.6 ± 0.1	0.9 ± 0.2	1.1 ± 0.2	0.03
Responders	0.7 ± 0.1	1.2 ± 0.1	1.4 ± 0.2	0.03
Nonresponders	0.5 ± 0.4	0.3 ± 0.2	0.6 ± 0.4	0.99
Stimulated C-peptide (ng/mL)[b]				
Group	1.1 ± 0.2		2.1 ± 0.3	0.03
Responders	1.2 ± 0.1		2.6 ± 0.3	0.03
Nonresponders	0.8 ± 0.7		0.9 ± 0.6	0.99
HOMA-IR				
Group	4.74 ± 1.4		3.37 ± 0.95	0.74
Responders	5.14 ± 1.94		3.86 ± 1.27	0.89
Nonresponders	3.75 ± 1.75		2.15 ± 0.75	0.66
HOMA-B				
Group	46.1 ± 14.0		174.5 ± 52.9	0.02
Responders	37.7 ± 18.4		154.5 ± 74.3	0.043
Nonresponders	67.3 ± 9.8		224.5 ± 2.8	0.18

[a]*P* value comparison between baseline and 6 month value.

[b]Mean ± SEM.

Abbreviations: BMI, body mass index; DBP, diastolic blood pressure; FPG, fasting plasma glucose; SBP, systolic blood pressure.

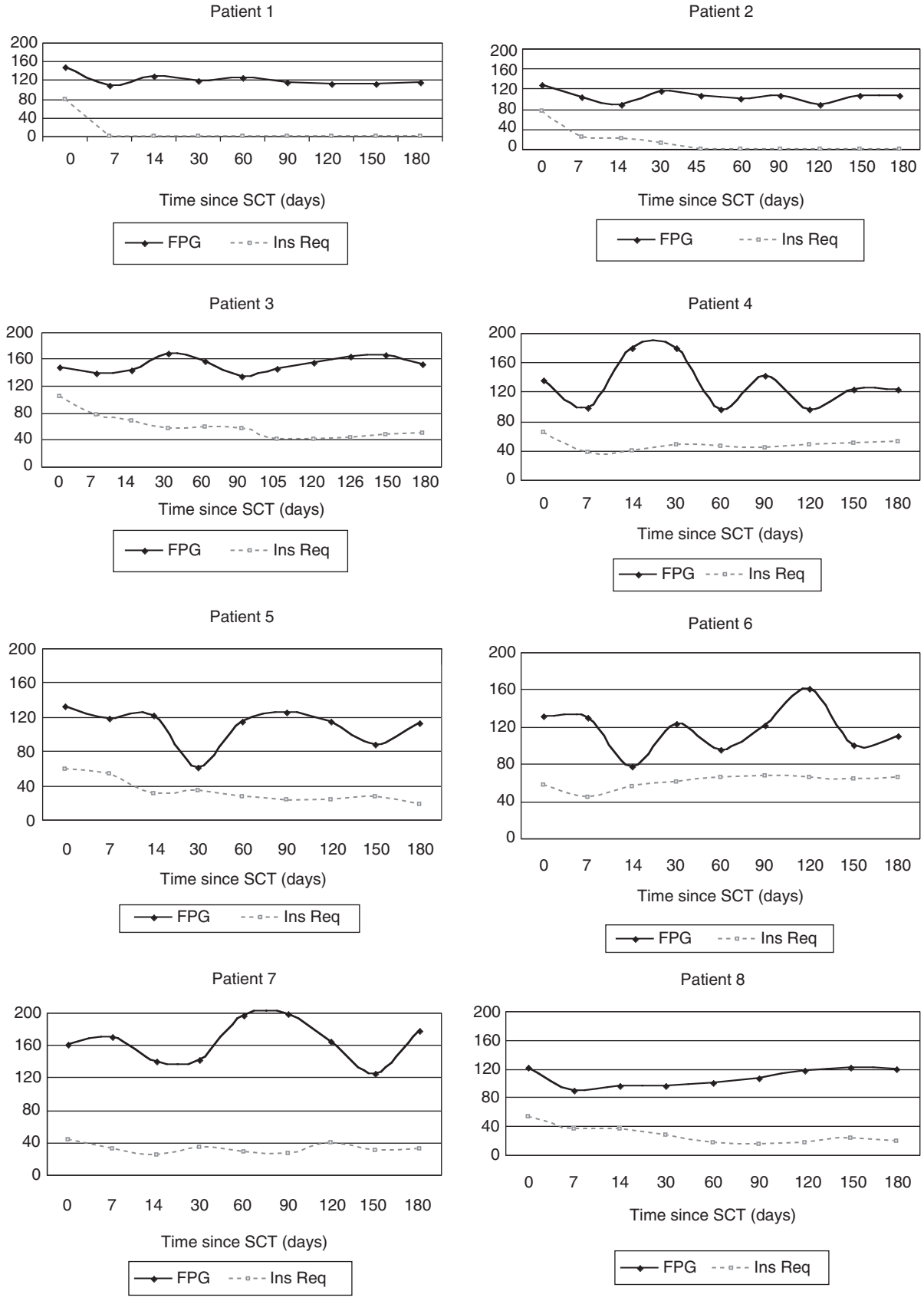

FIG. 1. Fasting plasma glucose profile and insulin requirement in patients with T2DM who underwent stem cell transplantation (SCT). Abbreviations: FPG, fasting plasma glucose in mg/dL; Ins Req, insulin requirement in units per day. (*Continued*)

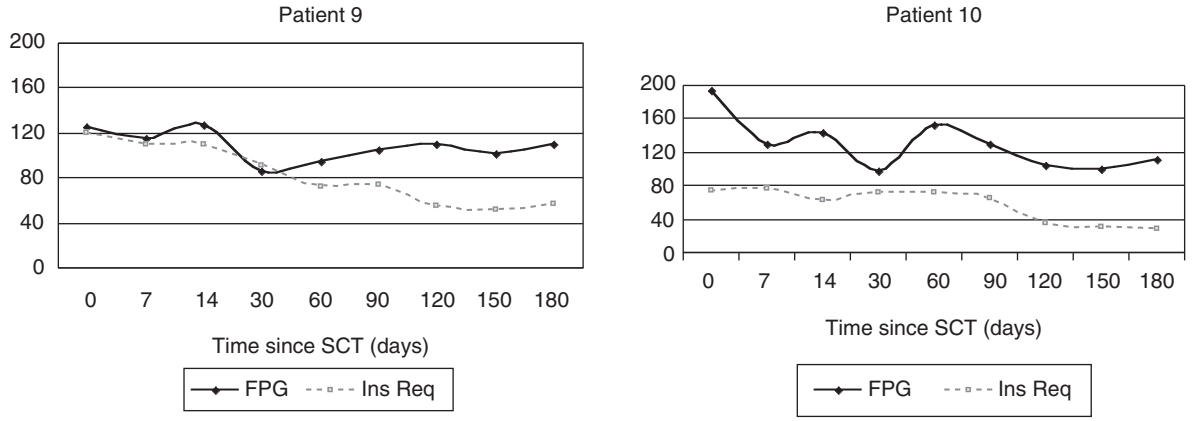

FIG. 1. (*Continued*)

progenitor cells [32]. MSCs exhibit a property whereby they can transform into cells of various lineage called plasticity [32]. MSCs and other stem cells are contained in the MNC fraction of the bone marrow. Reduction in insulin doses correlated with MNC count of infused stem cells in our study and probably a higher dose would have been more effective. Delivery of higher doses of stem cells can be achieved either by their repeated infusion in multiple sittings [17] or by mobilizing more marrow using recombinant human granulocyte colony–stimulating factor (rh-GCSF) [33]. Though it seems to be an attractive option, rh-GCSF can lead to preferential mobilization of stem cells of hematopoietic lineage [33] and may not give desired results. However, some unpublished observation suggests that mobilized stem cells using rh-GCSF can lead to reduction in blood glucose level in diabetic mice [34]. Hence, use of this novel agent for SC mobilization needs to be addressed by future studies. Difference in the quality of stem cells or some other confounding variables can also explain differences in individual outcome, especially in one of the nonresponder who had a baseline stimulated C-peptide response and MNC count comparable to the responders.

MSCs have the capability of migrating and homing to damaged pancreas [24], possibly guided by its altered cytokine milieu following which they secrete various growth factors such as hepatocyte growth factor, vascular endothelial growth factors, and others. These lead to angiogenesis and stimulation of growth, survival, and differentiation of β-cells through expression of PDX-1, which is a key regulator of pancreatic development [24]. Transdifferentiation of stem cells into β-cells contributes to a small fraction of such cells (~3%) as assessed by demonstration of green fluorescent protein positive (GFP+) donor cells in recipient's pancreas using fluorescent histochemistry [35,36]. Regeneration of small islets from pancreatic stem cells around the pancreatic ducts in animal model has also been demonstrated [36]. Though the literature is replete with histomorphometric evidence suggesting regeneration of β-cells after the use of BMSC in animal models [14,17,36], these hypotheses cannot be substantiated by any morphometric studies in humans.

To summarize, this study shows that SCT is an effective and safe option of β-cell regeneration leading to reduction in insulin requirement in patients with T2DM requiring high doses of insulin. However, further large-scale and long-term studies are required to fully substantiate the role of SCT in the management of T2DM.

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Author Disclosure Statement

Authors have nothing to declare.

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