

Case Report

Autologous Adipose Tissue Derived Insulin-Secreting Mesenchymal Stem Cell Transplantation In Late Onset of Autoimmune Mediated Diabetes Mellitus (LADA)

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Abstract

Latent autoimmune diabetes of adults (LADA) is a term describing patients with a type 2 diabetic phenotype combined with islet antibodies and slowly progressive β -cell failure. Insulin therapy is the treatment of choice. We report a 36-years-male with LADA since 4 years, treated with insulin-secreting human adipose tissue derived mesenchymal stem cells (IS-ADMSC). He was glutamic acid decarboxylase antibody positive (26 IU/ml). He was on insulin, 65 International units (IU) and 4 OHG/day. Autologous IS-ADMSC + bone marrow-derived haematopoietic stem cells (HSC) were infused into subcutaneous, portal and thymic circulation with conditioning of bortezomibe and rabbit-antithymoglobulin. Over follow-up of 18 months he is doing well with sustained fall of glycosylated haemoglobin from 12.7 to 6.7% and with sustained 60% decreased insulin requirement with sustained control of the plasma blood glucose level. Thus long-term control of LADA with co-transplantation of IS-ADMSC+HSC can be achieved safely and effectively.

Keywords: Haematopoietic stem cells; insulin; glutamic acid decarboxylase antibody; glycosylated haemoglobin

1. Introduction

The term latent autoimmune diabetes of adults (LADA) was coined by Tuomi *et al.* in 1993 to describe slow-onset type 1 autoimmune diabetes in adults. The experts define LADA as “a condition in which type 1 diabetes develops in adults”. LADA, also known as diabetes type 1.5, is slow-onset type 1 autoimmune, genetically-linked, hereditary disorder in adults and is also a term describing patients with a type 2 diabetic phenotype combined with anti-islet antibodies and slowly progressive β -cell failure. Among patients with phenotypic type 2 diabetes, LADA occurs in 10% of individuals older than 35 years and in 25% below that age. Glutamic acid decarboxylase (GAD) antibodies tests are used for differential diagnosis between LADA and type 1 diabetes mellitus. Uncontrolled LADA can lead to diabetic retinopathy, neuropathy, nephropathy, vasculopathy eventually leading to organ hypo-function and failure. Therapeutic options are regular exercise, dietary modification, oral hypoglycemic agents and exogenous insulin.

We report a 36-years-male with uncontrolled LADA on oral hypoglycemic agents and exogenous insulin since 4 years, treated with insulin-secreting cells generated from autologous human adipose tissue derived mesenchymal stem cells after approval from Institutional Review Board.

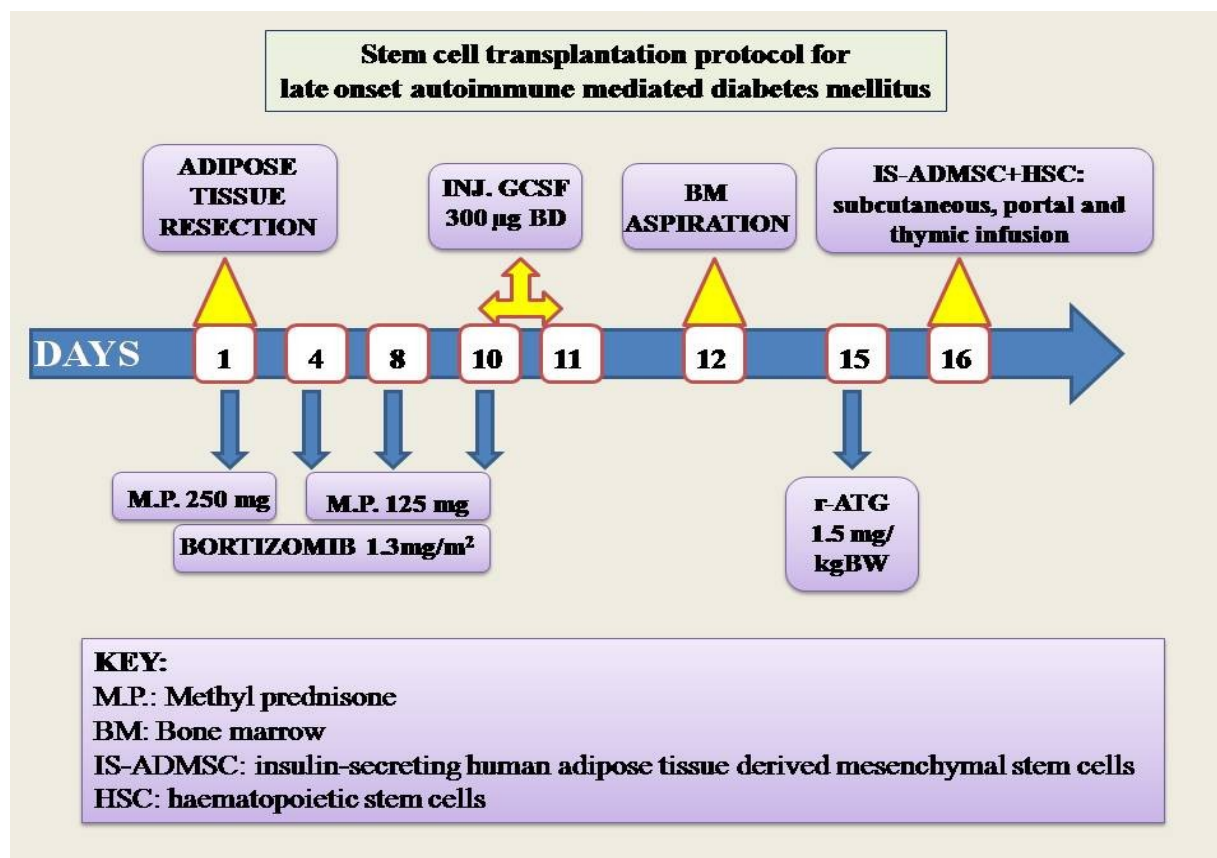
2. Case History

A 36-years-male with LADA since 4 years presented in March,'11 with weakness, fatigue, weight loss and uncontrolled blood sugar for last 7 months. He was on Insulin, 65 International units (IU) and 4 OHG/ day [T. glymepride (2 mg), T. Acarbose (50 mg), T. Pioglitazone (30 mg), T. Rosuvastatin (5 mg)].

His clinical examination was unremarkable with 71.5 kg body weight, 174 cm height and body surface area, 1.8 m². His fasting blood sugar (FBS) and postprandial blood sugars (PPBS), (FBS)/PPBS) were 268 mg/dL and 349 mg/dL on admission, serum C-peptide was 7.09 ng/ ml, glycosylated hemoglobin (HbA1c), 12.7 % and urine sugar was +4. Serum acetone was absent, GAD antibody was 26 IU/ ml (normal range: <10 IU/ ml), anti-islet cell antibody was absent (by immunofluorescent assay) and insulin antibody was 8.87 U/ml (normal range: < 12 U/ ml).

He was subjected for stem cell transplantation (SCT). Insulin doses were subsequently adjusted on sliding scale requirement. SCT protocol consisted of bortezomib, 1.3 mg/m² on days 1, 4, 8 and 10 along with 250 mg methyl prednisone on day 1 and 125 mg methyl prednisone on days 4, 8 10 followed by administration of granulocyte colony stimulating factor, 300 ug subcutaneously twice daily on days 10 and 11. Ten gram of autologous adipose tissue was resected from anterior abdominal wall on day 1 and subjected to *in vitro* MSC generation and further differentiation into insulin-secreting stem cells after harvesting MSC on day 11. Bone marrow (150 ml) was aspirated from posterior superior iliac crest under local anesthesia on day 12 for *in vitro* generation of haematopoietic stem cells (HSC). Rabbit antithymocyte globulin (rATG), 1.5 mg/kgBW was administered on day 15. On day 16, IS-ADMSC (4 ml- cell counts, 2 x 10²/ μL; CD90⁺,77.76% ; CD73⁺,29.17%) and HSC (147 ml- cell counts, 6.6 x 10⁴/μL; CD34⁺,0.15%) were injected into subcutaneous, portal (via omental vein) and thymic circulation under short general anesthesia by mini-laparotomy approach (Figure 1). SCT was uneventful.

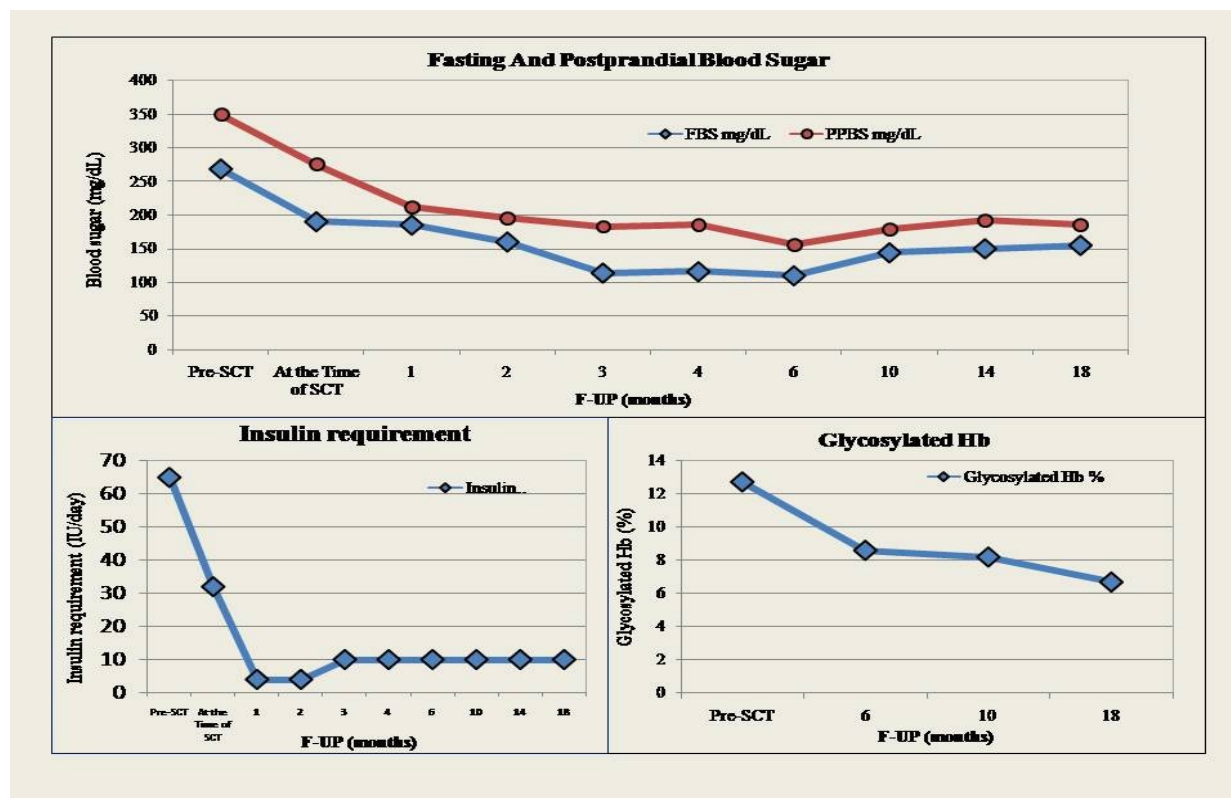
Figure 1: Stem cell transplantation (SCT) protocol for late onset autoimmune mediated diabetes mellitus



Immediate postoperative monitoring included 6 hourly blood sugar levels. He was discharged after 6 days.

Over follow-up of 18 months, patient is doing well. His insulin requirement decreased to 10 IU/day, he is still continuing 4 oral hypoglycemic agent(OHG). His HbA1C decreased to 6.7 % with 60% decreased insulin requirement, S.C-Peptide level of 6.9 ng/ml and maintaining FBS and PPBS, 114 mg/dL and 163 mg/dL respectively (Figure 2).

Figure 2: Post stem cell transplantation (SCT) Fasting & Postprandial Blood Sugar, Glycosylated Hemoglobin, Insulin Requirement per day



3. Discussion

LADA is a rather common and often under-recognized form of diabetes whose clinical presentation falls somewhere between type 1 and type 2 diabetes. From a patho-physiological perspective, it is more closely related to type 1, but based on etiology frequently adults with LADA are initially misdiagnosed and treated as having type 2 diabetes¹.

Human leukocyte antigen genes associated with type 1 diabetes are seen in LADA but not in type 2 diabetes. Islet cell, insulin, and GAD antibodies testing are used for differential diagnosis between LADA, type 1 and type 2 diabetes. Testing for anti-GAD in adult-onset non-obese diabetic patients helps to detect latent insulin-dependency at the earliest possible stage, since this assay can assist in the correct classification of diabetes, and more appropriate therapy².

LADA can have long-term devastating complications same as for those with type 1 and with type 2. Uncertainties concern almost all aspects of this disease, including the nomenclature, diagnostic criteria, pathogenesis with genetic, metabolic, and immunological aspects. Unfortunately, as a consequence, currently there is no established clear management strategy for it, in terms of therapy and prevention.

Although there are a good proportion of patients with LADA, surprisingly there are only a few studies that have evaluated interventions for this group. Treatment to prevent progression of β -cell destruction is still in experimental phase. Oral hypoglycemic agents seemed to provide poorer glycemic control than insulin alone and caused earlier insulin dependence. Daily exogenous insulin injections will be required for management of LADA when blood glucose can no

longer be managed through lifestyle. Some of the studies conducted in LADA patients have shown that insulin treatment is associated with better outcome in terms of metabolic control, insulin secretion, autoimmune responses against pancreatic β -cells and better long-term outcome by preserving β -cells and endogenous c-peptide secretion. Patients receiving insulin monotherapy had improved markers of autoimmunity and glycemic control was significantly improved.

MSCs are able to serve as a cellular vehicle for the expression of human insulin gene and have promising therapeutic role in the correction of metabolic derangement of DM and also in controlling and reverting complications of DM, like cardiac dysfunction, diabetic cardiomyopathy/ nephropathy/ polyneuropathy and wounds in diabetic patients.³ The most effective protocols till date have produced cells that express insulin and have molecular characteristics that closely resemble bonafide insulin-secreting cells; however, these cells are often unresponsive to glucose, which is also the most crucial characteristic concern in therapeutic approach while using these cells. It needs to be solved before finding a definite clinical application. To our knowledge this is first report of using IS-ADMSC in management of LADA. MSC showed beneficial effects in glycemic control, either isolated or combined to HSC. Previously we have treated 12 type 1 patients with our *in vitro* generated IS-ADMSC, who have sustained control of type 1 with raised c-peptide levels and controlled Hb1Ac. Hence we decided to explore this protocol which has already given sustained benefits without any adverse effects. *In vitro* functionality of these cells was also confirmed by insulin production and release in a glucose-responsive manner.⁴⁻⁶ We decided to infuse the cells in portal circulation since liver is the most tolerogenic organ⁷. LADA is believed to be an auto-immune disorder and once the cells are infused in to thymus, it helps in preventing the rejection of these cells by inducing central tolerance⁸. Hence we decided to infuse part of inoculum in to thymus also. Considering the autoimmune pathogenesis, this is the first case report to our knowledge where co-transplantation of IS-h-AD-MSc with HSC was successfully carried out and successful control of the plasma blood glucose level as a treatment of LADA is achieved.

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