

Chapter 2

Stem Cell Therapy for Type-1 Diabetes Mellitus

Umang G. Thakkar, Aruna V. Vanikar, and Hargovind L. Trivedi

Abbreviations

AD-MS	Adipose tissue-derived mesenchymal stem cell
BM	Bone marrow
DC	Dendritic cells
DCCTR	Diabetes control and complication trial research
DCDM	Diagnostic criteria for DM

U.G. Thakkar, MBBS, DCH (✉)

Department of Regenerative Medicine and Stem Cell Therapy and Department of Pediatrics,
G.R. Doshi and K.M. Mehta Institute of Kidney Diseases & Research Centre (IKDRC)-
Dr. H.L. Trivedi Institute of Transplantation Sciences (ITS), Civil Hospital Campus,
Asarwa, Ahmedabad, Gujarat 380016, India
e-mail: umangpaedia@yahoo.co.in

A.V. Vanikar

Department of Regenerative Medicine and Stem Cell Therapy and Department of Pediatrics,
G.R. Doshi and K.M. Mehta Institute of Kidney Diseases & Research Centre (IKDRC)-
Dr. H.L. Trivedi Institute of Transplantation Sciences (ITS), Civil Hospital Campus,
Asarwa, Ahmedabad, Gujarat 380016, India

Department of Pathology, Laboratory Medicine, Transfusion Services and
Immunohematology, G.R. Doshi and K.M. Mehta Institute of Kidney Diseases & Research
Centre (IKDRC)- Dr. H.L. Trivedi Institute of Transplantation Sciences (ITS),
Gujarat 380016, India

H.L. Trivedi

Department of Regenerative Medicine and Stem Cell Therapy and Department of Pediatrics,
G.R. Doshi and K.M. Mehta Institute of Kidney Diseases & Research Centre (IKDRC)-
Dr. H.L. Trivedi Institute of Transplantation Sciences (ITS), Civil Hospital Campus,
Asarwa, Ahmedabad, Gujarat 380016, India

Department of Nephrology and Transplantation Medicine, G.R. Doshi and K.M. Mehta
Institute of Kidney Diseases & Research Centre (IKDRC)- Dr. H.L. Trivedi Institute of
Transplantation Sciences (ITS), Gujarat 380016, India

DM	Diabetes mellitus
ESC	Embryonic stem cell
EV	Extracellular vesicles
HbA1c	Glycosylated hemoglobin
HSC	Hematopoietic stem cell
IPC	Insulin-producing cell
iPSC	Induced pluripotent stem cells
IPSC	Insulin-producing stem cell
ISC	Insulin-secreting cell
MSC	Mesenchymal stem cell
MODY	Maturity onset diabetes of the young
SCT	Stem cell therapy
T1DM	Type-1 diabetes mellitus
T2DM	Type-2 diabetes mellitus
UCB	Umbilical cord blood
WHO	World Health Organization

2.1 Review

2.1.1 Introduction of Disease

Diabetes mellitus (DM) is a metabolic disease characterized by chronic hyperglycemia resulting from defects in insulin secretion, insulin action, or both. The abnormalities are found in carbohydrate, lipid, and protein metabolism in diabetics, because of deficient action of insulin on target organs. If ketones are present in blood/urine, treatment is required urgently, because ketoacidosis can evolve rapidly.

2.1.2 Historical Witnesses of Diabetes Mellitus and Discovery of Insulin

DM was described for the first time as “Madhumeha” (sweet-tasting urine) by noted Indian physician Charaka and surgeon Sushruta in the fifth and sixth century AD who observed that urine of diabetics attracted ants and was sticky to touch. The Egyptian physicians also described it about 3500 years ago. The word “diabetes” (“to go through”) was coined by Apollonius of Memphis around 250 BC. These patients drained more fluid than the amount consumed; hence, this term was coined (Mac Cracken et al. 1997). Then, in the second century AD, the term “diabetes” was once again adopted by Aretaeus of Cappadocia who stated that it was the fault of the kidney by which diabetes developed. This theory was also accepted by a Roman physician named Galen (AD 131–201). He proposed an alternative name to

diabetes, “diarrhea urinosa” for excessive urinary output and “dipsakos” for excessive thirst and drinking (Pickup and Willium 2003). However, the Indian description of diabetes was more scholastic, differentiating two types of diabetes: one affecting older and obese persons and the second affecting younger and thin persons. They also had observed that young diabetics had shorter life span. This description is followed as of today also: as type-1 (T1) and type-2 (T2) DM (Kahn 1994). In 1809, John Rollo, an apothecary and chemist, was the first to use the term “mellitus,” the Latin and Greek root for honey. He documented high level of sugar in blood and in urine (MacCracken and Hoel 1997). It took about 60 years for further research in diabetes when Paul Langerhans noticed small clusters of cells in the pancreas in the year 1869 and described these structures without speculating about their possible function. In 1893, Edouard Laguesse suggested that these clusters of cells in the pancreas might constitute endocrine tissue of the pancreas, which he named as “islets of Langerhans” (after Paul Langerhans) (Pickup and Willium 2003). In 1909, a Belgian physician, Jean de Meyer isolated glucose-lowering hormone from pancreatic islet cells and named it as insulin (Latin, insula=Island) (De Meyer 1904). In 1921, Frederick G. Banting and Charles Herbert Best (his student under Macleod’s patronization) proved that insulin is an active substance of the pancreas associated with hypoglycemia in diabetic dogs. On 1st January 1922, the insulin extract made by Banting and Best was injected for the first time in history, into Leonard Thompson, a 14-year-old boy with DM. This was repeated by Collip. Thompson’s blood sugar returned to normal in about 24 h along with disappearance of his glycosuria and ketonuria (MacCracken and Hoel 1997)! In 1922, Banting treated Elizabeth Evans Hughes. Her recovery from hyperglycemia due to DM was hailed around the world as a true medical miracle. Banting and Macleod won the Nobel Prize for the discovery of insulin in 1923. The first commercial product of human insulin was developed by recombinant DNA technology in 1979 by Goeddel et al. (Ullrich et al. 1977), and human insulin was first prepared by Graham Bell et al. in 1980. In July 1996, the Food and Drug Administration of the USA approved the first recombinant DNA human insulin analogue, Lispro (Humalog). Currently more than 300 insulin analogues have been identified, including 70 from animals, 80 chemically modified, and 150 biosynthetic insulin preparations (Drjer 1992). This historical review shows that most of the advancement in discovery, treatment, and management of diabetes occurred in the twentieth century and that has given the greatest benefit to mankind in the form of longer life with better quality.

2.1.3 Classification of Diabetes Mellitus

The etiological classification was recommended by the “American Diabetes Association” (Diagnosis and Classification of Diabetes Mellitus 2009) and “WHO” expert committee on the classification and diagnosis of diabetes (World Health Organization WHO/NCD/NCS/99.2. 1999). Classification with minor modification is as below.

- I. Type-1 diabetes (β -cell destruction, usually leading to absolute insulin deficiency)
 1. Immune mediated
 2. Idiopathic
- II. Type-2 diabetes (may range from predominantly insulin resistance with relative insulin deficiency to a predominantly secretory defect with insulin resistance)
- III. Other specific types
 1. Genetic defects of β -cell function
 2. Genetic defects in insulin action
 3. Diseases of the exocrine pancreas
 4. Endocrinopathies
 5. Drug or chemical induced
 6. Infections
 7. Uncommon forms of immune-mediated diabetes
 8. Other genetic syndromes sometimes associated with diabetes

Genetic defects of β -cell function or insulin action, formerly termed “maturity onset diabetes of the young” (MODY), was originally described as a disorder with the following characteristics: onset before 25 years of age, autosomal dominant inheritance, and nonketotic diabetes mellitus (Fajans et al. 2001; Murphy et al. 2008).

IV. Gestational diabetes mellitus

(Patients with any form of DM may require insulin treatment at some stage of their disease. Such use of insulin does not, of itself, classify the patient.)

2.1.4 Criteria for Diagnosis of Diabetes

Diagnostic criteria for DM are based on blood glucose measurements and the presence or absence of symptoms (DCDM 2009; WHO/NCD/NCS/99.2. 1999). Three ways to diagnose DM are possible and each, in the absence of unequivocal hyperglycemia, must be confirmed, on a subsequent day, by any one of the three methods given in Table 2.1.

2.1.5 Spectrum of DM

T1DM, T2DM, and T1.5DM are differentiated from each other in Table 2.2 (Islets of Hope 2006; Zimmet et al. 2001; Leroith et al. 2003; Yoon and Jun 2005; Daneman 2006; Wilkin 2008).

Table 2.1 Criteria for the diagnosis of diabetes mellitus

1. Symptoms of diabetes plus casual plasma glucose concentration ≥ 11.1 mmol/L (200 mg/dL) ^a Casual is defined as any time of day without regard to time since last meal. Or
2. Fasting plasma glucose ≥ 7.0 mmol/L (≥ 126 mg/dL). Fasting is defined as no caloric intake for at least 8 h. Or
3. 2-h post-load glucose ≥ 11.1 mmol/L (≥ 200 mg/dL) during an OGTT. The test should be performed as described by WHO (86), using a glucose load containing the equivalent of 75 g anhydrous glucose dissolved in water or 1.75 g/kg of body weight to a maximum of 75 g (65)

^aCorresponding values (mmol/L) are ≥ 10.0 for venous whole blood and ≥ 11.1 for capillary whole blood and ≥ 6.3 for both venous and capillary whole blood

Or

HbA1C $\geq 6.5\%$. The test should be performed in a laboratory using a method that is NGSP certified and standardized to the DCCT assay (In the absence of unequivocal hyperglycemia, criteria 1–3 should be confirmed by repeat testing)

Table 2.2 Spectrum of diabetes

Feature	T1DM (IDDM)	T2DM (NIDDM)	T1.5DM (LADA)
Nature of onset	Usually rapid onset	Onset is months/	Onset is slow. Occurs
Age of diagnosis	Occurs in usually childhood age	years. Occur mainly in older adults	in 35–40 years/earliest at 25 years
Genes, triggers factors	Autoimmune, idiopathic, genetic	Hereditary, sedentary lifestyle, obesity, etc.	Autoimmune
ICA (islet cell antibodies)	ICA – 80%	ICA – no	ICA – positive
IAA (insulin autoantibodies)	IAA – often detected	IAA – no	IAA – yes, often
IA2 (islet antigen 2)	IA2 – 50–70%	IA2 – no	IA2 – often
GAD (GAD65-AAGAD)	GAD – positive	GAD – negative	GAD – positive
HLA	HLA – yes	HLA – no	HLA – yes, often
C-peptide	C-peptides – always low	C-peptides – normal to high	C-peptides – low
Treatment	Insulin	Oral hypoglycemic agents \pm insulin	Insulin \pm OHA
Prognosis and Complications	No complete cure	No complete cure	No complete cure

2.1.6 Epidemiology of T1DM

Occurrence of T1DM is increasing internationally, and it accounts for an approximate of 5–10% of all cases of DM (Daneman 2006) or 11–22 million globally (<http://ghr.nlm.nih.gov/condition/type-1-diabetes> 2013). According to Diabetes Control and Complications Trial (DCCT), the prevalence of T1DM in 2010 was 0.1–0.5% worldwide among general population, more than six million patients (1 out of 100–300 newborns), and its incidence was 30–50 new patients per every 100,000 individuals, with a 3% increase yearly, mainly in developing nations acquiring a Western lifestyle and diet (DCCTR 1993; Zimmet et al. 2001; Leroith et al. 2003, Lippincott and Williams, Yoon and Jun 2005, Daneman 2006; Wilkin

2008). T1DM is one of the most common chronic diseases of childhood; however, it can be diagnosed at any age (Gale 2005). Peak in presentation of T1DM usually noticed in between 5 and 7 years of age and at or near puberty (Harjutsalo et al. 2008). Whereas most autoimmune disorders disproportionately affect female, T1DM is slightly more common in male (Ostman et al. 2008). The incidence of T1DM varies with seasonal changes and birth month. More cases are diagnosed in autumn and winter (Moltchanova et al. 2009), and being born in the spring is associated with a higher chance of having T1DM (Kahn et al. 2009). Development of T1DM-associated autoimmunity (i.e., formation of islet autoantibodies) in the months or years before onset of symptoms also shows some seasonal synchronization (Kukko et al. 2005). The incidence of T1DM has been increasing globally for several decades (Dabelea 2009); still its incidence and prevalence vary substantially (Fig. 2.1) (Maahs et al. 2010). T1DM is most common in Finland (>60 cases/100,000 people each year) and Sardinia (around 40 cases/100,000 people each year) (Patterson et al. 2009). By contrast, the disorder is uncommon in China, India, and Venezuela (around 0.1 cases per 100,000 people each year) (Thunander et al. 2008). A plethora of environmental influences have been purported to affect the epidemiology of T1DM (Maclaren and Atkinson 1992), with infant and adolescent diets (Knip et al. 2010), vitamin-D and its pathway constituents (Svoren et al. 2009; Blanton et al. 2011; Cooper et al. 2011), and viruses receiving the most focus (Yeung et al. 2011; Stene and Rewers 2012).

2.1.7 Management of T1DM

The discovery of insulin in 1921–1922 was the most significant therapeutic event in the history of T1DM; however, exogenous insulin administration is not always necessary to provide the metabolic regulation to avoid disease related-complications. Because of selective destruction of the insulin-producing β cells within the pancreatic islets by triggers resulting in complete insulin deficiency, which causes hyperglycemia eventually leading to acute (ketoacidosis) and chronic (retinopathy, nephropathy, neuropathy) complications, hypercoagulability, dyslipidemia, and accelerated atherosclerosis, increased cardiovascular diseases, and reduced life expectancy (DCCTR 1993; Zimmet et al. 2001; Leroith et al. 2003). The impact of DM involves $\approx 10\%$ of total health-care budget in developed nations with over US \$100 billion spent every year in the USA alone and over US \$200 billion worldwide. The treatment of choice in association with tailored diet and physical exercise programs is daily exogenous insulin administration. Novel insulin formulations (e.g., glargine and lispro analogues) together with infusion-pump and glucose-sensor technologies have improved metabolic control with limited benefits (DCCTR 1993; Zimmet et al. 2001; Leroith et al. 2003; Daneman 2006). Treatment targets to achieve better glycemic control with T1DM in children and adolescents by plasma glucose and glycosylated hemoglobin (HbA1c) established in the

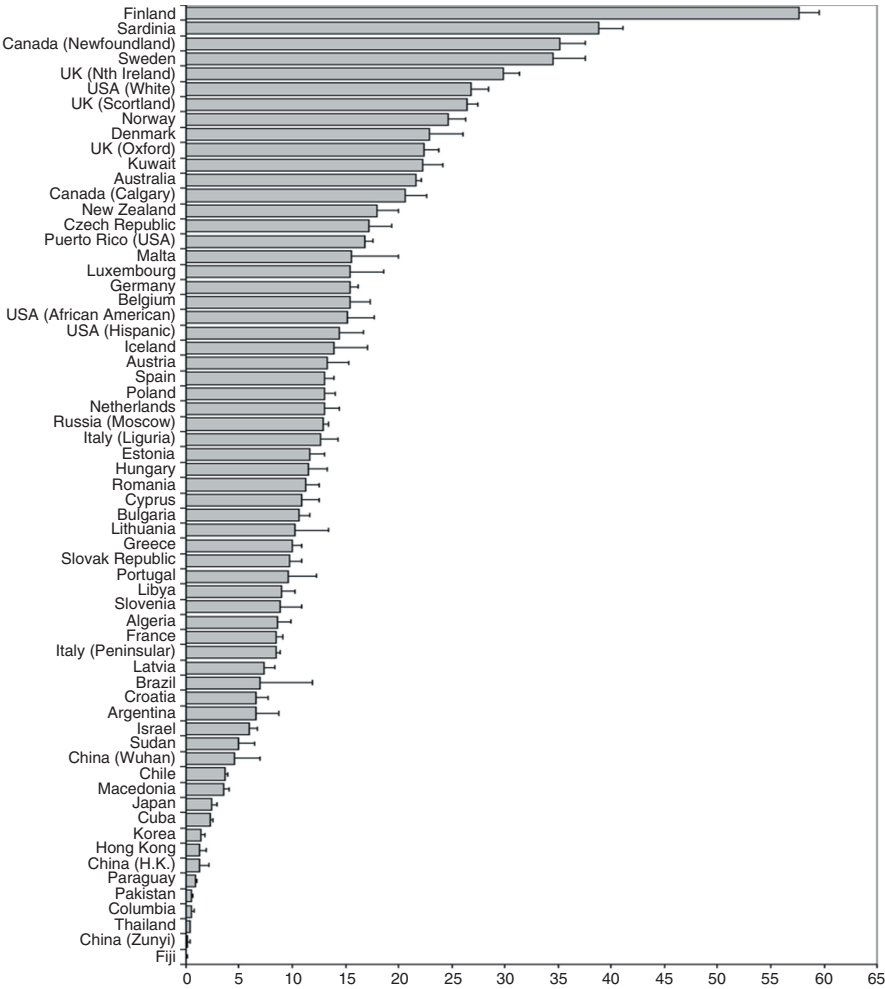


Fig. 2.1 Global incidence and prevalence of T1DM

evidence-based 2003 clinical practice guidelines of the Canadian Diabetes Association (2003) (Table 2.3).

Currently no approved agents are available to arrest the autoimmune destruction of β cells. The interest in reversing T1DM has grown in the last 5 years (Greenbaum and Atkinson 2011). In addition to preserving C-peptide production, the key goal is to induce immune tolerance against β cells. Majority of the approaches involve provision of self-antigens like vaccination with specific islet-cell proteins like insulin or glutamic acid decarboxylase (GAD) or immune suppression (Table 2.4).

Disappointingly, after promising phase 1–2 trials in patients with recent-onset T1DM and detectable endogenous insulin production, phase 3 trials of anti-CD3 antibodies (otelixizumab and teplizumab) and Diamyd vaccine (GAD-alum

Table 2.3 Glycemic and HbA1c targets by age for children and adolescents with T1DM

Age (years)	Plasma glucose (mmol/L)	HbA1c (%)	Considerations
<5	6.0–12.0	≤9.0	Careful avoidance of hypoglycemia in this age group due to risk of cognitive impairment
5–12	4.0–10.0	8.0	Adapt targets to patient's age
13–18	4.0–7.0	7.0	Appropriate for most patients
>18	4.0–6.0	6.0	Only if targets can be achieved safely

Source: 2003 clinical practice guidelines of the Canadian Diabetes Association (Canadian Diabetes Association 2003)

HbA1c hemoglobin A1c

Table 2.4 Agents assessed as immunomodulatory therapy to reverse T1DM

Study	Study phase and year	Main finding
Insulin APL (NBI-6042)	Phase 2; 2009	No change in metabolic response (i.e., C-peptide preservation) 135
Anti-CD20 (rituximab)	Phase 2; 2011	Preservation of C-peptide concentrations at 1 year, but no difference from placebo at 2 years 136
Anti-CD3 (teplizumab)	Phase 3; 2011	Although phase 2 studies showed preservation of C-peptide concentrations, phase I trials (Protégé study) 137 showed no change in metabolic response and the study stopped early
CTLA4, (abatacept) immunoglobulin fusion protein	Phase 2; 2011	T-cell co-stimulatory modulation slowed reduction in β -cell function over 2 years, although preservation of C-peptide was seen for 9–6 months 138
Anti-CD3 (otelixizumab)	Phase 3; 2011	Although phase 2 studies showed preservation of C-peptide concentrations, a phase 3 trial showed no change in metabolic response 139
GAD65 protein (Diamyd)	Phase 3; 2012	Phase 2 studies reported preserved C-peptide concentration, with no improvements in insulin needs. Two phase 3 trials did not meet end points 140,141
HSP60 (DiaPep277)	Phase 3; 2012	Phase 2 trials suggested increased C-peptide concentrations; a phase 3 trial noted C-peptide preservation at 1 year but only in adults (age 16–45 years) with type-1 diabetes 142

immunotherapy) did not meet with primary end points (Walter et al. 2009; Pescovitz et al. 2009; Sherry et al. 2011; Orban et al. 2011; Bach 2011; Ludvigsson et al. 2012; Wherrett et al. 2011). Administration of DiaPep277, a synthetic immunomodulator at 3-month intervals, resulted in less decline in stimulated C-peptide concentrations at 1 year in adults with T1DM than in the cohort that received placebo (Buzzetti et al. 2011; Schloot et al. 2007). Other phase 2 studies of immunomodulators showed evidence of therapeutic efficacy in settings of recent-onset T1DM; however, even with continued use, majority did not show long-lasting effects. For example, the fusion protein CTLA4-Ig (abatacept) preserved stimulated C-peptide

concentration for only 9 months despite continuous intravenous administration for 2 years (Orban et al. 2011). These results imply that single-agent immunosuppression alone might be insufficient to completely control the autoimmune destruction of β cells, or more specific and targeted therapies are required.

Glucose homeostasis requires finely regulated insulin secretion by pancreatic β cells present in islets of Langerhans (Mering and Minkowski 1889). In healthy person, insulin is secreted at a rate of ~ 2 pmol/kg/min, under fasting basal conditions (Eaton et al. 1980; Polonsky et al. 1984), and it increases by rate of ~ 5 to tenfold, after meal ingestion (Meier and Butler 2005). To accomplish this requirement, normally functioning β cells, and adequate number of β cells, means β -cell mass should be present. The human pancreas contains ≈ 1 million islets, each containing approximately 2000 β cells in healthy individual (Langerhans 1869; Stefan et al. 1982; Rahier et al. 1983; Bonner-Weir 1991). Thus, the β cells constitute $\sim 1.5\%$ of the total pancreas (1–2 g in total) (Bonner-Weir 1991).

The pancreatic islets of Langerhans containing β cells are functionally complex endocrine structures which produce insulin, that detect minimal changes in blood glucose levels and other metabolites, and also maintain metabolic homeostasis by a fine real-time secretion of specific hormones. A single β cell with a size of 15 μm can store about 10,000 insulin granules, and a single insulin granule of the size of 300 nm contains approximately 200,000 molecules of the crystallized insulin (Halban 2004). This is a well-orchestrated process, which is initially triggered by the glucose intake at the cell membrane and then eventually ends up with the glucose-responding insulin secretion (Ball and Barber 2003). Therefore, the generation of reliable pancreatic β cells is quite a difficult task.

These studies imply that a combination of therapies targeting multiple pathogenic pathways and improving β -cell viability is needed to preserve endogenous insulin production in this group of patients.

2.1.8 Pancreatic Transplantation

After ≈ 40 years of unsuccessful attempts by various researchers to control DM using partial pancreas transplantation, an English surgeon Charles Pybus (1882–1975) made a statement in 1924 that resonates even today: “Not much can be said about the principles of grafting, but it seems that until we are able to understand them (and I feel we do not understand them at present, especially the chemical factors), then we must continue to fail in such operations, although they may appear the most rational treatment for the diseases for which they are attempted” (Benedum 1999; Pybus 1924). The very first pancreatic transplant was surgically operated in 1966 by Kelly and colleagues (1967), and in the same year, pancreatic transplantation for T1DM was achieved at the University of Minnesota by Lillehei and co-workers (Jahansouz et al. 2011). Afterward, $>25,000$ pancreatic transplants have been performed worldwide (Gruessner and Sutherland 2005). The apparent easiest solution was deceased donors’ pancreatic transplantation to replace diseased/

destroyed β cells of islet of Langerhans, in T1DM. The pancreas can be grafted heterotopically under strict immunosuppressive regimen to avoid immune rejection. Data from the International Pancreas Transplant Registry shows encouraging results of metabolic control following whole pancreas transplantation with consequential discontinuation of the exogenous insulin administration. However, there are two major drawbacks with this approach, significant morbidity related to major surgery, and life-long immunosuppression requirement (Cefalu 2012).

As compared to kidney transplantation, pancreas transplantation has higher risk of surgical complications; also non-immunological complications of pancreas transplantation (including thrombosis and graft pancreatitis) account for graft loss in 5–10% of cases. These usually occur within 6 months of transplantation and are as important as etiology of pancreas graft loss in simultaneous pancreas and kidney transplantation (Ciancio et al. 1996b; Gruessner et al. 1996; Gruessner and Sutherland 2001, 2005).

All these problems related to pancreatic surgery led to introduce alternative approach of islet transplantation (Ballinger and Lacy 1972; Scharp et al. 1990). First clinical practice for pancreatic islet transplant was reported in 1977 to treat diabetic patients (Lakey et al. 2003). With further developments of the Ricordi method in 1989 for islet extraction (Ricordi et al. 1989), Edmonton Protocol in 2000 (Shapiro et al. 2000), and recent refinements, islet transplantation is the feasible option to treat T1DM, but potential immunosuppression is needed to alleviate the graft survival.

2.1.9 Islet Cell Transplantation

Islets of Langerhans are clusters of different endocrine cells scattered throughout the pancreas, each type secreting a specific hormone: α cells (glucagon), β cells (insulin and amylin), δ cells (somatostatin), and PP cells (pancreatic polypeptide). It is estimated that a normal human pancreas hosts about one million islet cells; however, the number varies with age, sex, weight of the donor, organ size, and with functional integrity (Ricordi 1992; Leroith et al. 2003; Cabrera et al. 2006; Leibiger and Berggren 2008). In 2000, a breakthrough protocol was developed for islet transplantation without the use of glucocorticoids for immune suppression; (Shapiro et al. 2000) the initially promising results deteriorated so that at 5 years, only 10% of patients remained independent of exogenous insulin (Ryan et al. 2005). Therefore, islet transplantation still remains an experimental procedure with ongoing research focusing on new methods using biomaterials (e.g., encapsulation), immunomodulation, site of delivery, and improved vascularization (Gibly et al. 2011). There are many reasons for poor outcome of clinical trials of islet transplantation, and only some of them are identified. In particular, during islet infusion, an intravascular instant blood-mediated inflammatory reaction (IBMIR) is believed to be responsible for destroying 50–70% of the infused β cells. An upregulation of tissue factor and other molecules on islet cell surface after isolation process is capable of

triggering innate immunity via activation of coagulation, complements, inflammation, and natural antibodies thereby destroying the islets. Peri-transplant anticoagulant prophylaxis with heparin can counteract this reaction (Moberg et al. 2002; Johansson et al. 2005; Eich et al. 2007).

Progressive decline in functioning of active β -cell mass from retrieval to the grafting procedure and immediate posttransplant time period is due to immune and nonimmune factors including activation of coagulation cascade and hostile microenvironment (Balamurugan et al. 2014; Nilsson et al. 2011). Calcineurin inhibitor/mTOR inhibitor and the process of islet isolation itself may contribute to low islet viability (Posselt et al. 2010; Balamurugan et al. 2010; Webb et al. 2012). The main hindrance to achieve consistent positive results is diabetogenic effect of corticosteroids and CNIs on β -cell function and survival, as well as on the development of peripheral insulin resistance. Posttransplant DM occurs in more than 50% solid organ transplant recipients, and the incidence increases with dose and duration of immunosuppressive therapy. Moreover, drug-dependent increment of lipids is associated with increased allograft loss and toxicity. Glucolipotoxicity may cause β -cell dysfunction and loss (Subramanian and Trencce 2007; Vantyghem et al. 2007; Poitout and Robertson 2008). These immunosuppressants result in both local and systemic toxicity that invariably impairs survival and functional competence of the grafted islet cells. Thus both pancreas and islet transplantation require immunosuppressive therapy, which carry the threat of recurrence of autoimmunity. Experiments were carried out in Japan for using living donors, but these may lead to clinical complications jeopardizing the donor to a risk of developing DM (Matsumoto et al. 2005, 2006).

Recipient- and graft-related complications post-islet transplantation include intra-abdominal bleeding, pleural/abdominal effusion, peripheral portal vein branch thrombosis, transient transaminitis, intrahepatic focal steatosis and amyloid deposits, common or opportunistic infections, profound neutropenia, pneumonia, ovarian cysts, viral reactivation, dyslipidemia, and renal toxicity. Tacrolimus may cause acute vasomotor vasculopathy with tubular necrosis and/or chronic fibrotic vasculopathy with glomerulosclerosis and interstitial fibrosis. Moreover, sirolimus may induce acute renal dysfunction and/or chronic proteinuria by increasing glomerular permeability and injury or by suppressing the compensatory renal cell proliferation and repair capacity. Papillary thyroid carcinomas, squamous and basal-cell skin carcinomas, ovarian and breast cancer, and pulmonary nodule have also been reported (Mineo et al. 2010).

Thus surgeons/researchers must consider the chance of infections (zoonotic diseases) into the hosts from the donating animal species by xenotransplantation, and also the limited availability of islet for transplantation from donated organs have driven efforts to introduce other potential sources of glucose-responsive insulin-producing tissue, as well as the use of stem cells (SC) (human embryonic SCs, non-pancreatic SCs, pancreatic SCs, and induced pluripotent SCs) as insulin-producing surrogates for β cells will provide a therapeutically meaningful advance.

Thus, therapeutic interventions to cure T1DM mainly focus on preservation of residual β cells, restoration of glucose-responsive, insulin-producing β cells using replacement or regeneration strategies, protection of replaced β cells from allo-/

autoimmune destruction and/or restoration of β -cell-specific unresponsiveness in the absence of chronic immunosuppression (Chhabra and Brayman 2013).

The need for unlimited supply, of a substitute for β cells of primary human islet of Langerhans, has led to a research on the suitability of stem or progenitor cells to generate insulin-secreting cells (ISC), in replacement therapies for DM. Other than downregulating the immune system for subsequently preserving residual β cells, another way is to offer a cell-based therapy, with differentiation of SCs into functional insulin-secreting β cell or a β -like cell, as the use of SCs to treat T1DM has been proposed for many years.

The main goal of stem cell therapy (SCT) is to achieve stable and normalized glycemic control with the absence of severe hypoglycemic attacks and improving quality of life, preventing long-term complications related to T1DM, and reducing procedure \pm immunosuppression-related adverse effects. Insulin independence is not necessarily the primary goal of SCT, although desirable; however, a reduction in insulin requirement and restoration of C-peptide secretion should be desired and with beneficial effects.

2.1.10 Alternative Approach to Islet/Pancreas Transplantation

In 1925, Nobel Prize winner Banting described in his lecture that, “Insulin is not a cure for diabetes; it is a treatment” (Banting 1965). Advances in clinical transplantation of pancreas/pancreatic islets of Langerhans have some limitations to generate insulin-producing cells from renewable SCs to treat DM. A recently reported work in Nature Biotechnology strengthens the evidence that SCs can give rise to cells that secrete insulin in a glucose-responsive manner, which is the characteristic of pancreatic β cells. This encourages hope of curing T1DM patients with cell therapy.

Cell therapy in actual sense involves “immunological resetting” by SC rescue through its reproducibility under strict proliferating control to generate sufficient quantity of tissue by differentiation into desired cell type(s) with surrounding tissue integrity and survival even after transplantation via proper function throughout the life of recipients without any untoward effects. The replaced cells must have the ability to synthesize, store, and release insulin in response to ambient glycemia and to avoid development of hyper-insulinemic hypoglycemia from induction of insulin-producing cells (IPCs) (pancreatic β cells) either by differentiation of SCs in vivo or transplantation of ex vivo differentiated cells in the pancreas in T1DM.

2.1.11 Stem Cells Transplantation

Canadian scientists Ernest A. McCulloch and James Till from the Ontario Cancer Institute in Toronto, with their colleagues Andy Becker and Lou Siminovitch, reported the presence of self-renewing cells within the bone marrow (BM) of mice

and postulated that these cells were regenerative SCs. In 1924, cell morphologist Alexander A. Maximow has been the first to discover a type of cell within the mesenchyme that develops into various types of blood cells. However, McCulloch and Till were the first to reveal the clonal nature of marrow cells (Becker et al. 1963; Siminovitch et al. 1963; Zhang et al. 1999), now identified as the first described adult SCs, the hematopoietic SCs (HSC).

Then, SCs were reported in 1963 by Becker and colleagues (1963) and are defined by two characteristics; (I) they can differentiate into many cell types in response to appropriate signals, (II) in undifferentiated state, and SCs have the ability to regenerate themselves by cell division. SCs can be divided into two subtypes: embryonic SCs and adult SCs. Embryonic SCs are pluripotent cells derived from inner cell mass of blastocysts and have the ability to differentiate into any of the three germ cell types (Thomson et al. 1998). These were initially derived from embryos of mouse in 1981 (Evans and Kaufman 1981; Martin 1981), and in 1998 James Thomson and colleagues successfully cultivated and continuously cultured embryonic SCs from human blastocysts (Thomson et al. 1998). On the other hand, adult SCs are self-renewed and undifferentiated cells found in adult organ niches and also known as somatic SCs. Adult SCs function as repair cells to regenerate damaged tissues. For example, mesenchymal stem cells (MSC) (capable of generating bones, fats, and cartilage), HSCs (derived from mesoderm, give rise to adult blood lineages), mammary SCs, intestinal SCs, endothelial SCs, neural SCs, and testicular cells.

2.1.12 Strategies for β -Cell Repair by Stem Cells

SC-based strategies represent significant therapeutic potential owing to both the intrinsic regenerative capacity and the immunomodulatory potential of SCs to restore glycol-metabolic and immune homeostasis (Fig. 2.2). This regenerative capacity can be harnessed to make available a self-replenishing supply of glucose-responsive insulin-producing cells, and the immunomodulatory properties help in arresting β -cell destruction, preserving residual β -cell mass, facilitating endogenous β -cell regeneration, ameliorating innate/alloimmune graft rejection, and preventing the recurrence of autoimmunity (Fiorina et al. 2011; Barcala Tabarozzi et al. 2013; Fandrich and Ungefroren 2010; Sims and Evans-Molina 2012). Thus, SCs with immunomodulatory properties can potentially be used to reverse hyperglycemia, alone or in combination with β -cell replacement strategies (Madec et al. 2009; Jurewicz et al. 2010; Rackham et al. 2011).

SCs obtained from a different sources, have been tested for their β -cell regenerative capacity and ability to restore immune homeostasis or promote longitudinal islet graft survival. These include embryonic SCs (ESCs), induced pluripotent SCs (iPSCs), BM-HSCs and umbilical cord blood-derived MSCs (UCB-MSCs), adipose tissue-derived MSCs, and pancreas-derived multipotent precursor cells, as well as

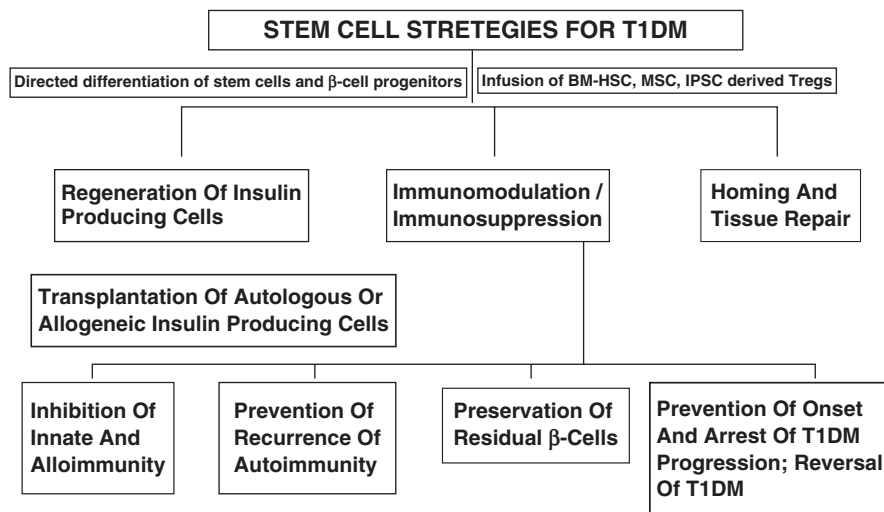


Fig. 2.2 Stem cell-based strategies for β -cell regeneration and immunomodulation

pancreatic β -cell progenitors that reside in the ductal epithelium, exocrine tissue, and within islet proper, neural progenitor cells, and facultative β -cell progenitors from spleen, liver, and the endometrium.

2.2 Role of Stem Cells in Treatment of Diabetes

2.2.1 Embryonic Stem Cells

The continued need for an alternate of replacing β cells in T1DM patients has fostered scientific and public interest in ESC as a potential therapy. ESCs have characteristic of pluripotency and self-renewal ability, which allowed researchers to explore the application of SCs in a number of medical conditions contributing to destructive etiology. T1DM fits in category of such diseases. Islet transplantation has revealed significant potential to achieve insulin independence (Shapiro et al. 2006). Theoretically, ESCs can be differentiated into any cell line, including pancreatic β cell over an appropriate time span and when exposed to appropriate signal in correct sequence (Martin 1981). It is also hypothesized that if pancreatic islet cells have been developed from ESCs differentiation, the shortage of β cell of the pancreas would be overcome in diabetics. The discovery of methods of isolation and growth of human ESCs in 1998 renewed the hopes of researchers, clinicians, and

diabetic patients and their families to cure the T1DM, and perhaps T2DM as well may be within striking distance.

First in vitro attempt to produce islet cells from mouse ESCs was reported in 2000; Soria and team were able to achieve a degree of control in hyperglycemia which lasted for few months (Soria et al. 2000). Bernat Soria and his colleagues added DNA containing part of the insulin gene to ESC from mice. The insulin gene was linked to another gene which rendered the mice resistant to an antibiotic drug. In the presence of antibiotics, only activating insulin promoter cells were able to survive. These cells were cloned and then cultured under different conditions. Cells were cultured in low concentrated glucose medium and responded accordingly with changes in glucose concentration by increasing insulin secretion nearly seven-fold. Unfortunately, production of insulin-positive cells was low due to contamination of non-islet insulin-producing cells and selection of cells before full differentiation. Other results have also been reported in different experiments over the next few years with variable degrees of successes. These experiments were carried out on human (Assady et al. 2001; Segev et al. 2004) and mouse (Lumelsky et al. 2001; Blyszczuk et al. 2004) ESCs, but factors like differentiated cells, immaturity (Segev et al. 2004), low glucose-insulin response (Assady et al. 2001), low number of insulin-producing cells (Hori et al. 2005), and by low cell homogeneity (Lumelsky et al. 2001) limit the success of the strategy. Ron McKay and his colleagues described many experiments, in which they induced mouse ESC to differentiate into insulin-secreting structures, which resembled pancreatic islets (Lumelsky et al. 2001). Several research groups are trying to apply McKay's results with mice, to induce human ESCs to differentiate into insulin-producing islets. All these studies made the researchers to rethink the existing strategies for cell differentiation techniques. Kubo's method (Kubo et al. 2004) to retain culture conditions to convert mouse ESCs into definitive endoderm was updated and refined by D'Amour et al. (2005) to achieve to nearly 100% output to produce pure definitive endoderm cell population. This research work was continued until a five stage in vitro process was introduced after in vivo development of the pancreas (D'Amour et al. 2006).

According to Jon Odorico from the University of Wisconsin in Madison, ESCs can differentiate and express the insulin gene. Itskovitz-Eldor and colleague further characterized insulin-producing β cells ~1–3% from embryoid bodies (Assady et al. 2001). However as compared to previous experiments, there was no insulin secretion found in response to glucose even when C-peptide was released in response to stimuli like KCl and cAMP. The cells resembled to 6–9 weeks of embryo; in vitro differentiation was stopped at that time to achieve more specific and successful results (Kroon et al. 2008). These differentiated cells were transplanted into epididymal fat pad in immune-deficient mice. Insulin secretion was measured in glucose-dependent manner. Posttransplantation C-peptide secretion levels were low at 1 month, and at the 90th day, it was reached at those levels where they could be seen with 3000–5000 human islets transplantation.

2.2.2 *Obstacles in Application of Embryonic Stem Cells in Clinics*

ESCs-based β -cell replacement therapy for T1DM has several major obstacles that must be overcome before this approach can be considered as a therapeutic option.

Ethical and religious sensitivities: There are ethical and religious sensitivities concerning with the use of human embryos which still hamper its use for research purposes, and many governments ban or at least highly restrict association with this research (Watson 2003; Gruss 2003). The Roman Catholic Church has repeatedly demanded an international ban on the use of human ESC for research purpose, because it requires the destruction of embryos who has all the moral rights and protection as any other human being (Copland 2004; Oakley 2002). Owing to these moral concerns, the United States Congress has enacted a broad ban on federal funding for human ESC research. Later, this ban was loosened to allow research on human SC lines that already existed. A highly debated question is whether SCs from human embryos should be given to use for generation of SC lines, which were unsuitable for fertilization programs and discarded. The US President George W. Bush stated that, "There is no such thing as a spare embryo" to the religious authorities on this matter (New York Times, May 26, 2005). Landry and Zucker (2004) pointed out that death before the onset of neural development, a significant fraction of these human embryos will be found to be "organismically" dead. So, using such embryos to generate human SC lines would not contradict the ethics of the Catholic Church and other religious authorities. Recently, novel techniques to derive mouse SCs without affecting the subsequent development of the embryo have been described (Chung et al. 2006; Meissner and Jaenisch 2006). These types of techniques may resolve some of the ethical concerns regarding the generation of ESC line.

There were some potential risks imposed by continued replication of ESCs-derived transplants including loss of cell cycle control and the induction of tumor cell growth. Teratomas have been observed in iPSC lines derived from ESCs consistent with it (Fujikawa et al. 2005). To provide an ultimate cure for diabetes, human ESCs-derived islets or β cells would either need to maintain their ability to proliferate or the transplantation procedure would have to be repeated at regular intervals. Therefore, maintaining of a physiologic balance between replication and cell death appears to be a major challenge for β -cell replacement therapies based on ESCs.

Functional β -cell mass requires more than 18 months to establish in developing humans (Bouwens et al. 1997; Kassem et al. 2000), which is not yet clear if it will be possible to drive ESCs to a useful mass of β cells or β and other cell-type aggregates, ex vivo tend to undergo senescence and differentiation within days in culture thereby losing their pluripotency (Halvorsen et al. 2000).

2.3 Adult Stem Cells

2.3.1 *Adult Pancreatic Stem Cells*

Islet comprises functional cells of four types: glucagon-producing α cells, insulin-producing β cells, somatostatin-producing δ cells, and pancreatic polypeptide-producing cells (Liu et al. 2013). Adult pancreatic SCs can also be another source for pancreatic β cells as they carry the characteristics of multipotency and clonogenic potential. By applying pancreatic duct ligation model of injury, differentiation and proliferation characteristics of ductal cells of the pancreas have been proposed as the major source for β cells for regeneration of tissues (Wang et al. 1995). Experiments on rats have revealed that β cells and pancreatic tissues would be regenerated expeditiously if 90% of the pancreas undergo resection (Bonner-Weir et al. 1993). β -cells mass was reported after the activation of duct lining NGN3+ endocrine precursors in adult mice (Xu et al. 2008). Advanced strategies are needed to be developed to isolate and grow adult pancreatic cells and to differentiate into β cells. Epithelial cells of pancreatic duct were harvested and induced in vitro to become functional islets (Ramiya et al. 2000). Clonal characteristics of multiple progenitor cells from the pancreas of adult mouse were reported; at differentiation stage, endocrine, exocrine, neuronal, and glial cell populations were produced by clonal colonies. Produced β cells express the insulin secretion and glucose-dependent reactivity (Seaberg et al. 2004). To identify pancreatic SCs, clonal analysis was performed which is capable of differentiating the pancreatic SCs into pancreatic endocrine and exocrine cells (Suzuki et al. 2004). Future targets will be to tackle all challenges of harvesting, purifying, and growing various populations of pancreatic progenitor cells and also for inducing the β -cell differentiation without genetic mutations.

2.3.2 *Adult Non-pancreatic Stem Cells*

MSCs and HSCs have the capability to proliferate and refill the damaged or dead tissues and cells as these possess multipotency.

2.3.3 *Mesenchymal Stem Cells*

MSCs were first identified by Friedenstein and his colleagues (Friedenstein et al. 1966). They described it as bone-forming progenitor cells from rat BM. MSCs can be harvested from adipose tissues, BM, and other organs, but the richest source remains BM. They are reported as pericytes which were localized in the blood

vessels' wall (Meirelles Lda and Nardi 2009; Masoud et al. 2012). MSCs have high potential to self-renew and to differentiate *in vitro* and *in vivo*. Immunomodulatory properties of MSCs inhibit several components of immune systems *in vitro* (Hoogduijn et al. 2010). Hence, severe refractory diseases have been treated using MSCs in human (Dazzi and Marelli-Berg 2008). Several lines of evidence have shown that under appropriate environments, MSCs are able to differentiate into mesodermal, endodermal, and even ectodermal cells. MSCs are known as hypoinmunogenic cells because of its properties like escaping immune recognition and inhibiting immune responses. Therefore, MSCs appear to be a very promising tool for regenerative and immunoregulatory cell therapy. The Mesenchymal and Tissue Stem Cell Committee of the International Society for Cellular Therapy has proposed a minimal set of four criteria to define human MSC (Dominici et al. 2006):

1. MSCs have to be plastic adherent when maintained under standard culture conditions.
2. MSC must have the ability for osteogenic, adipogenic, and chondrogenic differentiation.
3. MSC must express CD73, CD90, and CD105.
4. MSC must lack expression of the hematopoietic lineage markers c-kit, CD14, CD11b, CD34, CD45, CD19, CD79 α , and human leukocyte antigen (HLA)-DR.

MSCs obtained from BM, adipose tissue, and umbilical cord were same for its morphology, immune phenotype, success rate of isolating MSC, colony frequency, and differentiation capacity. (Kern et al. 2006; Izadpanah et al. 2006). Adipose tissue is the most attractive source for generation of MSC for researchers and clinicians of nearly all medical subspecialties as it requires simple surgical procedure and uncomplicated enzyme-based isolation procedures, and easy and repeatable access to the subcutaneous adipose tissue is possible (Casteilla et al. 2005; Oedayrajsingh-Varma et al. 2006). Therefore, ADSC do represent an alternative source of autologous adult SCs which can be obtained repeatedly in large quantities under local anesthesia with a minimum patient discomfort.

These advantage and properties of MSCs triggered the researchers to find out the effects of MSCs in autoimmune diseases like T1DM. Differentiation of MSCs gives rise to mesodermal tissues, but other tissues including iPSCs have also been reported (Davani et al. 2009). Many protocols have been devised to differentiate MSCs into iPSCs. Some researchers used pancreatectomy model to release unknown pancreas regenerative factors. MSCs were grown in this extract of regenerative factors transformed to insulin-producing islet-like clusters which were responsive to glucose concentration (Jahr and Bretzel 2003). Another research group used defined culture conditions and successfully differentiated the BM-derived stromal cells to insulin-producing islet-like aggregates which resemble to β cells in gene expression pattern as well as active production of insulin (Oh et al. 2004). Serial differentiation of BM-derived MSCs in a three step differentiation mechanism using endocrine differentiation inducers and primed with stromal-derived factor-1- α to enhance its therapeutic potential and survival has been reported, and the resultant iPSCs were found to respond to glucose tolerance test as well as lowered blood glucose levels

when transplanted in streptozotocin-induced diabetic rats (Tariq et al. 2013). In animal models of T1DM alone (Lee et al. 2006; Fiorina et al. 2009) and in combination with HSCs (Urban et al. 2008), MSCs exhibit beneficiary effects in glycemic control. The HSCs were used along with AD-MSC because HSC transplantation with immunosuppression conditioning is believed to create active and passive tolerance by clonal deletion/T-cell suppression and helps in angiogenesis. Researchers also used them to revive nonfunctioning pancreatic β cells. Co-infusion of HSCs with Ad-MSC-ISCs that was examined on five patients with T1DM showed a decrease in exogenous insulin requirement and an increase in C-peptide level (Trivedi et al. 2008). In another trial on mice, researchers injected a mixture of MSCs and HSCs into the BM of mice with STZ-/radiation-induced injury, and the normal blood glucose level was regained successfully. It is reported that MSCs have the capability to inhibit the proliferation of pancreatic β -cell-specific T-cells, ultimately reducing the damage induced by T-cells on new β cells (Liu et al. 2013). Although it is impossible to elaborate all mechanisms in detail, BM SCs can stimulate the regeneration of damaged pancreatic β cells. Without any doubt, BM SCs have therapeutic effects on DM, and these are ideal adjuvants for cell treatment and therapy in future.

2.3.4 MSC-Derived Extracellular Vesicles as Novel Immunomodulator in T1DM

Depending on the cellular sources, extracellular vesicles (EV) may induce immune cell activation or inhibition because of their different immunomodulatory functions (Théry et al. 2009). The EVs have emerged as paracrine factors of MSC actions. In fact, MSC-derived EVs released proteins and nucleic acids, capable to mimic the effect of originating cells. Antigen-presenting cells/B lymphocytes-released vesicles activate the T cells by direct peptide-MHC complex presentation to transfer antigen and/or peptide-MHC complex, from dendritic cells (DCs), natural killer (NK) cells, macrophages, and B cells and to induce DC maturation (Théry et al. 2009; Robbins and Morelli 2014). Conversely, EV released from tumor cells or from MSC exhibited inhibitory functions on T cells, NK cells, and DCs (Robbins and Morelli 2014). Moreover, EVs of MSCs promoted the regulatory T-cell activity and induced the monocyte differentiation into myeloid-derived suppressor cells (MDSCs). In an autoimmune encephalomyelitis experiment, MSC-derived EVs inhibited autoreactive lymphocyte proliferation and induced tolerogenic signaling, via PD-L1, TGF- β , IL-10, and CD4+ CD25+ Foxp3+ Treg cells (Mokarizadeh et al. 2012).

EV released from heterologous human BM-MSC mimic the immunomodulatory characteristic of the cells in T1DM by inducing a shift toward an anti-inflammatory and regulatory T-cell. After an integrin-mediated EV internalization in patient's peripheral blood mononuclear cells (PBMCs), a downregulation of Th1 responses, observed as IFN- γ production, number of Th17 cells, and levels of pro-inflammatory IL-17 was observed (Favaro et al. 2014). These Th17 effector cells participate in T1DM pathways paralleling Th1 cells, and their secreted IL-17 contributes to β -cell death.

T-cells have shown to produce PGE2 and TGF- β , involved in immunomodulation which is the property of MSC (Spaggiari et al. 2009). TGF- β conveyed as protein and as mRNA within and on the surface of EV (Pap et al. 2011). It also inhibits the lymphocyte proliferation and promotes the Treg generation (Bruno et al. 2015). Thus, EV-associated functional miRNAs can be transferred to target cells (Ratajczak et al. 2006; Deregibus et al. 2007; Valadi et al. 2007). MSC-derived EV expressed miRNAs, miR-21, known to enhance TGF- β signaling. RNA depletion of EV reduced the TGF- β transcript in PBMCs that suggested increasing the TGF- β production by transfer of TGF- β mRNA or miRNA. Hence, EV may increase the TGF- β activation pathway and its release, in a paracrine/autocrine way in T-lymphocytes. MSC-derived EVs may restore Th1/Th2 balance and preserve Treg cells in T1DM. In fact, EV enhanced the production of IL-10 and induced higher frequencies of Foxp3+ Treg cells (Favaro et al. 2014). The production of IL-6 was increased in the presence of EV by PBMCs, which is known to suppress maturation of inflammatory DC and mediate β -cell repair (Boumaza et al. 2009). Researchers also observed that MSC-derived EV mimics the effect of MSC on DC maturation, impairing antigen presentation.

2.3.5 MSCs as Cellular Vehicle for Insulin-Producing Gene Therapy

MSCs are a promising tool for cell-based gene therapy against a variety of different diseases (Hamada et al. 2005). Their high self-renewal potentiality makes them strong candidates for delivering genes and restoring function of organs and tissues. The ability to genetically modify MSCs provides durable expression of therapeutic genes.

Human insulin gene is located on chromosome 11p15.5 (Ohneda et al. 2000). Insulin synthesis and its release from islet β cells is complex and tightly regulated mechanism. Glucose affects insulin at all levels, including transcription, translation, and release. Mature insulin results from a processing pathway starting from the rough endoplasmic reticulum and ending at the Golgi apparatus. Translation of insulin mRNA yields pre-pro-insulin, cleaved by endoproteases PC1 and PC2/PC3 to give pro-insulin first and mature insulin + C-peptide subsequently. In the secretory granule, six insulin molecules are coordinated by a Zn atom, seen under microscope by dithizone stain.

With better assays for SCs and improving the vector biology, gene transfer efficiency into MSCs has been increased. The transfected MSCs expressed the insulin gene and secreted insulin in culture media consistently. Xu et al. reported that diabetes could be relieved effectively for up to 6 weeks in mice model by intrahepatic transplantation of BM-derived murine MSCs infected with the recombinant retrovirus carrying human insulin gene. In further study, implantation of engineered cells using diabetic animal models and observed its therapeutic effect with more tests of

efficacy and safety of engineered human MSCs as surrogate β cells (Xu et al. 2007). Further work was also carried out for modified herpes I virus as a vector for the human insulin gene (Calne 2005). The theoretical advantages of the herpes I virus are (i) its large capacity to accommodate a construct; (ii) its ability to infect primary and secondary cell lines in vitro; (iii) even though its entry in to the nucleus not integrate with the host DNA and functions separately as an episome which is not likely to unmask ontogenesis; (iv) most of the patients have already had contacts with the herpes I virus, which normally resides in a quiescent state in neurological tissue; (v) relatively mild immune reaction against the virus; and (vi) antiviral treatment against the herpes virus is established and available. Thus, herpes I virus could serve as a new vector for human insulin gene delivery into MSCs.

2.3.6 Minimum Requirements for Replacement β Cells

IPCs act as replacement β cells for cell therapy of T1DM which could be generated either by trans-differentiation of MSCs or by delivery of insulin gene into MSCs. These MSC-derived IPCs may solve the problem of donor shortage for islet cell transplantation and provide a cure for T1DM. Any options for primary islets of Langerhans will require some minimum essentiality. The basic requirements for surrogate β cells are:

1. Large numbers of replacement β cells will be required for significant therapeutic impact. Current transplantation protocols use up to 1×10^6 primary human islets per recipient, equivalent to approximately $2-4 \times 10^9$ β cells. As a result, the ability of MSCs to replicate and to differentiate toward pancreatic endocrine phenotype makes them attractive candidates for producing replacement β cells.
2. Replacement β cells must have synthesize, store, and release ability of insulin in response to changes in the ambient glycemia. Understanding β -cell function at the molecular level will likely facilitate the manufacturing of physiologically competent IPCs from MSCs.
3. Proliferative capacity of replacement β cells must be tightly controlled to avoid development of hyperinsulinemic hypoglycemia because of its expansion can occur in vivo. Excluding proliferative cells from the transplant material will help to overcome this problem. In the case of insulin gene transferred MSCs, the possibility of tumor formation has to be considered.
4. The transplanted replacement β cells must not be destroyed by the recipient's immune system (Burns et al. 2004).

With appropriate immunosuppressive medication, autologous MSCs-derived IPCs transplantation will circumvent the immune-rejection dilemma. On the other hand, Burt et al. indicated that HSC transplantation may reintroduce tolerance to islet cells in T1DM (Burt et al. 2002). Thus, co-transplantation of MSCs-derived IPCs and HSC from the same donor (autologous/allogeneic) could evade the risks of recurring autoimmunity. Furthermore, the pathways of β -cell differentiation were

different in “in vitro” and “in vivo” (Houard et al. 2003). However, in vitro differentiation protocols can generate surrogate β cells having some phenotypic and functional similarity to authentic β cells, which is actually not the β cells. Since, MSCs-derived IPCs are developmentally and immunologically distinct from primary β cells, who may escape the recipient’s autoimmune assault.

2.3.7 Hematopoietic Stem Cells

Multipotent SCs that give rise to all the other blood cells located in red BM and are derived from mesoderm are called HSC. BM is an important source of relatively easily accessible adult SCs. BM transplantation is considered to be effective for the treatment of autoimmune T1DM. However, there is a great debate on the issue of the fate of transplanted BM-SC. Based on animal models, autoimmune diseases have been effectively treated with combination of HSC and high-dose immunosuppression (Sykes and Nikolic 2005; Burt et al. 2008), and the very first patient was treated with HSC for autoimmune diseases in 1996 (Voltarelli et al. 2011). To date, approximately 1500 patients carrying autoimmune disease have been treated (Passweg and Tyndall 2007; Vanikar et al. 2012) with HSC transplantation because of low risk of complications. After decades of clinical use of HSC transplantation for severe and stubborn autoimmune diseases, it can be speculated that this approach may also be useful for treating T1DM. In an experiment in nonobese diabetic mice, clinically observable T1DM has been easily precluded by allogeneic HSC transplantation and not by autologous HSC. Results could be predicted by the genetic nature of the disease in this animal model (Atkinson and Leiter 1999). However, the clinically observable T1DM in nonobese diabetic mice cannot be reversed only by the allogeneic HSC transplantation but also require efficient source of pancreatic β cells (Sykes and Nikolic 2005; Kang et al. 2005). Allogeneic HSC can restore tolerance to pancreatic β cells but cannot restore the cells pool if once destroyed by autoimmune system.

The pancreatic duodenal homeobox-1 (PDX-1) gene-modified MSCs derived from the human BM can be induced to differentiate into functional IPCs (Li et al. 2007; Karnieli et al. 2007). In addition, Sun et al. demonstrated that BM-derived MSCs can differentiate into IPCs under appropriate conditions in vitro in diabetics. This study provides the information regarding the feasibility of using autologous BM-MSCs as a source of IPCs for β -cell replacement therapy (Sun et al. 2007).

2.3.8 Umbilical Cord Blood Stem Cells

SCs from umbilical cord can also be used for treating DM (Kucia et al. 2006). These SCs have higher regenerative potency and have low rate of rejection than BM cells after allogeneic transplantation. In human UCB, the presence of MSCs was reported,

when cells isolated from UCB exhibited the characteristic immunophenotype and differentiation of BM-MSC (Lu et al. 2006; Kern et al. 2006). In animal models to prevent or reverse T1DM, MSC (Wang et al. 2011), cord blood mononuclear cells (Ende et al. 2004), combination of T-regulatory cells, and UCB-SC (McGuckin and Forraz 2008) have been used. Cord blood MSCs have been obtained from cord walls with greater efficiency than the cord blood, and these cells have the capability to treat T1DM (Anzalone et al. 2011). In an experiment, in Florida University, no significant improvements have been reported as compared to control group when 15 T1DM patients were injected with autologous mononuclear cord blood cells (Haller et al. 2009). In 1988, after first productive transplantation of UCB (Gluckman et al. 1989), it has been recognized as the ultimate source for HSC to treat blood diseases and genetic disorders. In addition to human UCB, Wharton's jelly of the UCB is rich in MSC. UCB-SC has gained substantial attention for the therapeutic options in regenerative medicine.

2.3.9 Hepatic Stem Cells

In developmental biology, endoderm gives rise to the pancreas and liver, and these contain similar progenitor cells, so it has been proposed that liver cells would be an ultimate source for β cells because of their easy availability by biopsy and their strong regenerative capability (Zaret and Grompe 2008). By using adenoviral transduction, pancreatic endocrine and exocrine gene expressions have been reported in liver cells when NGN3 and PDX1 have been expressed in mouse liver (Liu et al. 2013). Newly grown pancreatic tissues form clusters around the central veins of the liver secreting insulin and without disturbing the normal liver functions. Moreover, insulin-producing cells maintain STZ-induced hyperglycemia and glucose levels for 8-month time period (Ber et al. 2003; Yechoor et al. 2009). To date, no in vitro evidence has been reported to show that modified liver cells can be proliferated in vitro conditions. In the future, efficient methodology needs to be developed for getting in vitro expressions of trans-differentiated cells from liver.

2.3.10 Induced Pluripotent Stem Cells

The process of formation of pluripotent SC (PSC) from non-pluripotent cells is referred to as induced pluripotency. Somatic cells can be transformed to PSC under specific conditions, and such cells are called iPSCs. This ground breaking discovery led to an outburst in the studies of reprogramming of cells (Takahashi and Yamanaka 2006). Induced PSCs have the same characteristics like ESCs. These have high telomerase activity as well as gene promoters are also hypo-methylated. Human iPSCs obtained by reprogramming of human somatic cells (skin fibroblasts) can represent an alternate to human ESCs and eliminate the ethical issues pertaining to

ESCs. Therefore, these are ethically more acceptable. Induced PSCs can be the preferred cell type for the treatment of DM for autologous cell transplantation. Autoimmunity in T1DM may lead to the immune rejection of transplanted autologous iPSCs. However, this strategy may be successful for T2DM where autologous SCT is required. Second problem pertaining to iPSCs is same as with ESCs: the formation of teratoma. These PSCs are formed due to the expression of some specific factors which involve the use of genome-integrating viruses. In iPSCs, the use of DNA-based reprogramming strategy could lead to the insertional mutagenesis and the use of oncogenic reprogramming factors could add to the risk of tumor formation (Okita et al. 2013). Reprogramming of cells using nonintegrating approach may be required before it moves toward clinics. The problem of genome integration was addressed by the study of Stadtfeld et al., by introduction of four factor of pluripotency using nonintegrating adenoviral vectors (Stadtfeld et al. 2008).

2.3.11 Induction of Insulin-Producing Cells from Stem Cells by Protein Transduction Technology

Protein transduction technology has been recently emerged for induction of IPCs from SCs. A variety of peptides, like protein transduction domains (PTDs) or cell-penetrating peptides (CPPs), have been characterized for their ability to translocate into live cells. When proteins and peptides synthesized as recombinant fusion proteins or covalently cross-linked to PTDs, these can be directly internalized into cells. Biologically active full-length proteins and peptides have been delivered to cells both in vitro and in vivo. The homeodomain transcription factors like Antennapedia (Antp), HSV type-1 protein VP22, and HIV-1 trans-activator TAT protein are most commonly studied PTDs. The involved mechanism is endocytosis followed by passage from the vesicle into the cytoplasm for PTD-mediated protein transduction (Noguchi and Matsumoto 2006). This technology facilitates the differentiation of SCs into IPCs, so it can cure the T1DM. Two pancreatic endocrine transcription factors, PDX-1 protein and BETA2/NeuroD protein, have a PTD sequence in their structure. Noguchi et al. observed that PDX-1 (Noguchi et al. 2003) or BETA2/NeuroD (Noguchi et al. 2005) protein induced insulin expression in pancreatic ductal progenitor cells. Similarly, Domínguez-Bendala et al. (2005) reported that in vitro pancreatic endocrine differentiation was stimulated by TAT-mediated neurogenin-3 (ngn3) protein transduction. Gräslund's group (Kilk et al. 2001) showed that the third helix of the homeodomain of transcription factor Isl-1 internalized into cells. Thus, delivery of exogenous transcription factors (PDX-1, BETA2/Neuro D, ngn3, Isl-1, etc.) by protein transduction technology could be a novel strategy for generating IPCs from SCs/progenitor cells without requiring gene transfer technology. Thus, MSC is the strong candidate for this emerging modality.

2.3.12 Our Experience with Insulin-Secreting Cells from Adipose Tissue-Derived MSC for T1DM

In 2008, Trivedi et al. reported safe and effective treatment in five insulinopenic diabetes using intra-portal infusion of insulin-secreting AD-MSC with BM-HSC. No xenogeneic material was used in this study. There was 30–50% fall in insulin requirement with 4- to 26-fold rise in serum C-peptide levels. This effect was found to be sustained for 3 years. No further follow-up was available. No immunosuppression was used. Subtotal lymphoid irradiation of 200 cGy for 5 days followed by rabbit antithymocyte globulin, 1.5 mg/kgBW, was used for conditioning, before infusing the SC in liver, subcutaneous tissue, and thymus. No infective episodes or graft versus host disease were observed (Trivedi et al. 2008). Vanikar et al., in 2010, have generated in vitro MSC from human adipose tissue (Vanikar et al. 2010), which qualify the definition standardized by the Mesenchymal and Tissue Stem Cell Committee of the International Society for Cellular Therapy (Yañez et al. 2006). Our cells fulfilled these criteria. They were further differentiated into insulin-secreting cells (ISC) under defined culture conditions; these cells were phenotypically identical to pancreatic β cells (Trivedi et al. 2008). These cells expressed transcription factors *pdx-1*, *pax-6*, and *isl-1*; all three are central controlling genes capable of reprogramming non-pancreatic cells to surrogate β -cell functions. Further infusion of AD-MSC-ISC with BM-HSC was carried out safely in 11 diabetics with 1 to 24 years of disease duration. Over a mean follow-up of 23 months, all patients responded, with a decreased mean exogenous insulin requirement from 1.14 to 0.63 units/kg BW/day, Hb1Ac dropped from 8.47% to 7.39%, serum C-peptide levels increased from 0.1 to 0.38 ng/mL, and all of them became free of diabetic ketoacidosis events.

In 2015, Thakkar et al. have treated 20 T1DM patients, 10 with autologous SCT and 10 with allogeneic SCT (Thakkar et al. 2015). Both groups received exactly similar treatment of AD-MSC-ISC with BM-HSC into the liver via intra-portal route, thymic circulation, and subcutaneous fat pad. Thymic infusion was carried out to achieve central tolerance (Sprent and Kishimoto 2001), and portal circulation was performed to take advantage of tolerogenicity of the liver and for better grafting (Starzl 2001). Subcutaneous tissue is an immunologically privileged site; hence, we decided to inject part of the cells into abdominal subcutaneous tissue so that it could serve as a “back-up reservoir” for insulin supply (Prokhorova et al. 2009). Results in both groups were compared, autologous SCT was found to be better than allogeneic source vis a vis long-term control of hyperglycemia. This study also established one important fact that patients with any type of DM need not search for donors; they can use their own fat reservoir for treating their own disease.

2.3.13 Lessons Learned

Still some researchers and clinicians are disappointed because of limited benefits with recent advancement of T1DM research and therapy. Large investments in terms of time, finances, and patient resources have been required for SCT, islet-cell transplantation, genetics, primary/secondary prevention, and reversal of T1DM (Greenbaum and Atkinson 2011). It is difficult to decide whether the goal is disease prevention or reversal even with therapeutic interventions using conventional or experimental agents (Staeva et al. 2013). T1DM is a multifactorial etiology that overcomes its autoimmune nature; permanent cure of the disease is not achieved and till now requires more intense research. Similarly, islet-cell transplantation depends on overcoming recurrent autoimmunity and averting alloimmunity (Vendrame et al. 2010). Even islet/pancreas transplantation have their limitations/hurdles as described previously in this review, so investigators are focusing now on xenotransplantation, encapsulation, novel sites for cell delivery (e.g., eye), and development of surrogate insulin-producing cells (Gibly et al. 2011). T1DM has polygenic nature, in which more than 40 loci have been identified with disease susceptibility/resistance (Concanon et al. 2009), combined with environmental factors suggesting the unpredictable pathogenesis of the disease. Experiments are going on to improve understanding of the genetic risk for T1DM by genotyping at multiple susceptibility loci (Winkler et al. 2012). Genetic involvement for disease development is complex in nature regarding the immune response in T1DM. Mechanism of selective destruction/loss of pancreatic β cells remains unclear apart from antigen-specific immune response till now for this disease (Atkinson et al. 2011). Many researchers have described the putative role for adaptive rather than innate immune responses in the disease pathogenesis which is crucial for the development of improved management. Luckily, trial networks like NIH TrialNet and Immune Tolerance Network and registries like T1D Exchange can judge the ability of therapeutic agents and effects in terms of improvement of patient recruitment and increase the precision of disease prediction (Sosenko et al. 2012). Modification/changes in clinical trial design like adaptive trial design, utilization of animal model (Atkinson 2011a, b), identifying more practical therapies, and better defining disease heterogeneity (Pozzilli 2012) could improve the applicability of T1DM research and which will be more effective. If the therapeutic intervention is applied during the natural history of the disease with silent or asymptomatic state, this may be more effective.

2.3.14 Road Map for Future Generation

The most pressing questions are: Will the recipients' immune response to infused cells destroy them eventually? Whether more number of SCs are required or repeated stem cell infusion cycles are required? Is there any better engraftment

technique available? Whether requirement of more potent/supporting cells like regulatory T/B cells? Are SCs capable for producing long-term immunological tolerance? For predicting disease development, can improved markers be obtained? Can replication and neogenesis of β cell induce safely in humans? Is it possible to develop safe and effective closed-loop therapy system?

These questions make a road map of investigations for the next generation and if properly addressed, should result in substantial improvements in the quality of life of T1DM patients.

2.3.15 Current Challenges and Future Perspectives

SCs are the potentially unlimited source of functioning surrogate β cells of the pancreas; still its research is in fundamental stage (Hansson et al. 2004; Dor et al. 2004). Induced surrogate β cells from the SCs by differentiation and its infusion technique should be improved and maintained them functionally in a specialized microenvironment termed as SC niche. The niches maintain the SCs quantum, and multiple signals are required to maintain a balanced control of SC self-renewal (Hou and Singh 2008; Singh et al. 2007; Singh and Hou 2008, 2009; Scheres 2007; Fuchs 2009; Yamashita 2009; Meirelles and Nardi 2009; Discher et al. 2009). Advance technology is required for successful transplantation of β cells into suitable niches to achieve maximum therapeutic effects.

SCT has some technical and clinical limitations. Various cytoprotective strategies and agents are under investigation for improving the SC yield and outcome. Patients should get the benefit of insurance and/or reimbursement for the therapy.

Several pathological conditions have regulatory mechanism via microRNAs (Liu et al. 2008; Mishra et al. 2009; Wang and Wu 2009). This experience suggests that they may have a role in mechanisms underlying differentiation of SCs into β cells. Adenovirus genes for cell engineering reduce cell immunogenicity, allowed successful transplantation across allogeneic barriers, without immunosuppression/immune-isolation. Genetic modification can be applied in the future to cultured human islets, to derive a universal donor β cells because of current use of adenovirus genes in cell engineering. Genetically engineered β cells hold the promise of replacing exogenous administration of insulin as an accurate, convenient, and safe way for long-term control of euglycemia in T1DM (Orlando et al. 2014). Thus in vitro generated ISC from SCs to treat T1DM appears extremely promising, with bona fide hope for a complete cure.

2.3.16 Unanswered Questions

The key goal of research in T1DM is detection of ex vivo islet autoreactive T cells and their functions. This may provide the markers for detection of patients at risk and to design intervention strategies to preserve surrogate β cells.

Why islet β cells are target specific for destruction/elimination and do inherent processes for developing the disease? Is the clinical dilemma involving the autoimmune issue? How can control continuously generated autoantibodies against β cells? Understanding the innate and adaptive immune response helps in improving the therapies.

This is a review of change with respect to understanding of the epidemiology, current ongoing research in management, and prospects for curing T1DM with cell-based therapy. In hindsight, many long-held goals once thought readily achievable have been difficult to realize, and concepts regarded as dogmas have proven to be flawed.

2.3.17 *Raised Issue*

The major raised question is whether it is reasonable to expose diabetics to such type of therapies with some/minimal degree of untoward effects with risk, when the therapeutic option of exogenous insulin administration is effectively available.

2.4 Conclusion

SCT is a better alternative to islet/pancreas transplantation. It is safe, viable, and easily reproducible treatment modality for T1DM which has recently achieved successful graft function, with long-term better metabolic control without any untoward effects. However, intense work needs to be addressed properly before pushing the cell-based therapy from bench to bedside.

Acknowledgment Author acknowledges the immense help received from the scholars whose articles are cited and included in the references of this review chapter. The authors are also grateful to the author/editors/publishers of all those articles, journals, and books from where the literature for this chapter has been reviewed and discussed.

References

- Anzalone R, Iacono ML, Loria T et al (2011) Wharton's jelly mesenchymal stem cells as candidates for beta cells regeneration: extending the differentiative and immunomodulatory benefits of adult mesenchymal stem cells for the treatment of type 1 diabetes. *Stem Cell Rev Rep* 7:342–363
- Assady S, Maor G, Amit M, Itskovitz-Eldor J, Skorecki KL, Tzukerman M (2001) Insulin production by human embryonic stem cells. *Diabetes* 50:1691–1697
- Atkinson MA (2011a) It's time to consider changing the rules: the rationale for rethinking control groups in clinical trials aimed at reversing type 1 diabetes. *Diabetes* 60:361–363

- Atkinson MA (2011b) Evaluating preclinical efficacy. *Sci Transl Med* 3:96cm22
- Atkinson MA, Leiter EH (1999) The NOD mouse model of type 1 diabetes: as good as it gets? *Nat Med* 5:601–604
- Atkinson MA, Bluestone JA, Eisenbarth GS et al (2011) How does type 1 diabetes develop?: the notion of homicide or beta-cell suicide revisited. *Diabetes* 60:1370–1379
- Bach JF (2011) Anti-CD3 antibodies for type 1 diabetes: beyond expectations. *Lancet* 378:459–460
- Balamurugan AN, Breite AG, Anazawa T, Loganathan G, Wilhelm JJ et al (2010) Successful human islet isolation and transplantation indicating the importance of class 1 collagenase and collagen degradation activity assay. *Transplantation* 89:954–961
- Balamurugan AN, Naziruddin B, Lockridge A et al (2014) Islet product characteristics and factors related to successful human islet transplantation from the collaborative islet transplant registry (CITR) 1999–2010. *Am J Transplant* 14:2595–2606
- Ball SG, Barber TM (2003) Molecular development of the pancreatic beta cell: implications for cell replacement therapy. *Trends Endocrinol Metab* 14:349–355
- Ballinger WF, Lacy PE (1972) Transplantation of intact pancreatic islets in rats. *Surgery* 72:175–186
- Banting FG (1965) Diabetes, insulin, nobel lectures, physiology or medicine 1922–1941. Elsevier, Amsterdam. <http://nobelprize.org/nobelprizes/medicine/laureates/1923/banting-lecture.html>
- Barcala Tabarrozzi AE, Castro CN, Dewey RA (2013) Cell-based interventions to halt autoimmunity in type 1 diabetes mellitus. *Clin Exp Immunol* 171:135–146
- Becker AJ, McCulloch EA, Till JE (1963) Cytological demonstration of the clonal nature of spleen colonies derived from transplanted mouse marrow cells. *Nature* 197:452–454
- Bell GI, Pictet RL, Rutter WJ, Cordell B, Tischer E, Goodman HM (1980) Sequence of the human insulin gene. *Nature* 284:26–32
- Benedum J (1999) The early history of endocrine cell transplantation. *J Mol Med* 77:30–35
- Ber I, Shternhall K, Perl S et al (2003) Functional, persistent, and extended liver to pancreas trans-differentiation. *J Biol Chem* 278:31950–31957
- Blanton D, Han Z, Bierschenk L et al (2011) Reduced serum vitamin D-binding protein levels are associated with type 1 diabetes. *Diabetes* 60:2566–2570
- Blyszczuk P, Asbrand C, Rozzo A et al (2004) Embryonic stem cells differentiate into insulin-producing cells without selection of nestin-expressing cells. *Int J Dev Biol* 48:1095–1104
- Bonner-Weir S (1991) Anatomy of the islet of Langerhans. In: Samols E (ed) *The endocrine pancreas*. Raven Press, New York, pp 15–27
- Bonner-Weir S, Baxter LA, Schuppin GT, Smith FE (1993) A second pathway for regeneration of adult exocrine and endocrine pancreas: a possible recapitulation of embryonic development. *Diabetes* 42:1715–1720
- Boumaza I, Srinivasan S, Witt WT, Feghali-Bostwick C, Dai Y, Garcia-Ocana A, Feili-Hariri M (2009) Autologous bone marrow-derived rat mesenchymal stem cells promote PDX-1 and insulin expression in the islets, alter T cell cytokine pattern and preserve regulatory T cells in the periphery and induce sustained normoglycemia. *J Autoimmun* 32:33–42
- Bouwens L, Lu WG, De Krijger R (1997) Proliferation and differentiation in the human fetal endocrine pancreas. *Diabetologia* 40:398–404
- Bruno S, Deregibus MC, Camussi G (2015) The secretome of mesenchymal stromal cells: role of extracellular vesicle in the immunomodulation. *Immunol Lett* 168:154–158. pii: S0165-2478 (15)00105-4
- Burns CJ, Persaud SJ, Jones PM (2004) Stem cell therapy for diabetes: do we need to make beta cells? *J Endocrinol* 183:437–443
- Burt RK, Oyama Y, Traynor A, Kenyon NS (2002) Hematopoietic stem cell therapy for type 1 diabetes: induction of tolerance and islet cell neogenesis. *Autoimmun Rev* 1:133–138
- Burt RK, Loh Y, Pearce W et al (2008) Clinical applications of blood-derived and marrow-derived stem cells for non-malignant diseases. *JAMA* 299:925–936

- Buzzetti R, Cernea S, Petrone A et al (2011) C-peptide response and HLA genotypes in subjects with recent-onset type 1 diabetes after immunotherapy with DiaPep277: an exploratory study. *Diabetes* 60:3067–3072
- Cabrera O, Berman DM, Kenyon NS et al (2006) The unique cyto-architecture of human pancreatic islets has implications for islet cell function. *Proc Natl Acad Sci U S A* 103:2334–2339
- Calne R (2005) Cell transplantation for diabetes. *Philos Trans R Soc Lond B Biol Sci* 360:1769–1774
- Canadian Diabetes Association (2003) Clinical practice guidelines for the prevention and management of diabetes in Canada. *Can J Diabetes* 27(suppl 2):S21–S23
- Casteilla L, Planat-Benard V, Cousin B et al (2005) Plasticity of adipose tissue: a promising therapeutic avenue in the treatment of cardiovascular and blood diseases. *Arch Mal Coeur Vaiss* 98:922–926
- Cefalu WT (2012) American diabetes association-European association for the study of diabetes position statement: due diligence was conducted. *Diabetes Care* 35:1201–1203
- Chhabra P, Brayman KL (2013) Stem cell therapy to cure type 1 diabetes: from hype to hope. *Stem Cells Transl Med* 2:328–336
- Chung Y, Klimanskaya I, Becker S, Marh J, Lu SJ, Johnson J, Meisner L, Lanza R (2006) Embryonic and extraembryonic stem cell lines derived from single mouse blastomeres. *Nature* 439:216–219
- Ciancio G, Burke GW, Viciano AL et al (1996) Destructive allograft fungal arteritis following simultaneous pancreas-kidney transplantation. *Transplantation* 61:1172–1175
- Comparison of clinical features between (juvenile) type 1 diabetes, type 2 diabetes and LADA (2006) Islets of Hope
- Concanon P, Rich SS, Nepom GT (2009) Genetics of type 1A diabetes. *N Engl J Med* 360:1646–1654
- Cooper JD, Smyth DJ, Walker NM et al (2011) Inherited variation in vitamin D genes is associated with predisposition to autoimmune disease type 1 diabetes. *Diabetes* 60:1624–1631
- Copland PS (2004) The roman catholic church and embryonic stem cells. *J Med Ethics* 30:607–608
- D'Amour KA, Agulnick AD, Eliazar S, Kelly OG, Kroon E, Baetge EE (2005) Efficient differentiation of human embryonic stem cells to definitive endoderm. *Nat Biotechnol* 23:1534–1541
- D'Amour KA, Bang AG, Eliazar S et al (2006) Production of pancreatic hormone-expressing endocrine cells from human embryonic stem cells. *Nat Biotechnol* 24:1392–1401
- Dabelea D (2009) The accelerating epidemic of childhood diabetes. *Lancet* 373:1999–2000
- Daneman D (2006) Type 1 diabetes. *Lancet* 367:847–858
- Davani B, Arieli S, Ikonomou L, Oron Y, Gershengorn MC (2009) Human islet-derived precursor cells can cycle between epithelial clusters and mesenchymal phenotypes. *J Cell Mol Med* 13:2570–2581
- Dazzi F, Marelli-Berg FM (2008) Mesenchymal stem cells for graft-versus-host disease: close encounters with T cells. *Eur J Immunol* 38:1479–1482
- De Meyer J (1904) Sur la signification physiologique de la secretion interne du pancreas. *Zbl Physiol* 18:S826
- Deregibus MC, Cantaluppi V, Calogero R, Lo Iacono M, Tetta C, Biancone L, Bruno S, Bussolati B, Camussi G (2007) Endothelial progenitor cell derived microvesicles activate an angiogenic program in endothelial cells by a horizontal transfer of mRNA. *Blood* 110:2440–2448
- Discher DE, Mooney DJ, Zandstra PW (2009) Growth factors, matrices, and forces combine and control stem cells. *Science* 324:167–1677
- Domínguez-Bendala J, Klein D, Ribeiro M, Ricordi C, Invernardi L, Pastori R, Edlund H (2005) TAT-mediated neurogenin 3 protein transduction stimulates pancreatic endocrine differentiation *in vitro*. *Diabetes* 54:720–726
- Dominici M, Le Blanc K, Mueller I et al (2006) Minimal criteria for defining multipotent mesenchymal stromal cells. The International Society for Cellular Therapy position statement. *Cytotherapy* 8:315–317

- Dor Y, Brown J, Martinez OI, Melton DA (2004) Adult pancreatic beta cells are formed by self-duplication rather than stem-cell differentiation. *Nature* 429:41–46
- Drjer K (1992) The bioactivity of insulin analogues from in vitro receptor binding to in vivo glucose uptake. *Diabetes Metab Rev* 8:259–285
- Eaton RP, Allen RC, Schade DS, Erickson KM, Standefer J (1980) Prehepatic insulin production in man: kinetic analysis using peripheral connecting peptide behavior. *J Clin Endocrinol Metab* 51:520–528
- Eich T, Eriksson O, Lundgren T (2007) Nordic network for clinical islet transplantation. Visualization of early engraftment in clinical islet transplantation by positron-emission tomography. *N Engl J Med* 356:2754–2755
- Ende N, Chen R, Reddi AS (2004) Effect of human umbilical cord blood cells on glycemia and insulinitis in type 1 diabetic mice. *Biochem Biophys Res Commun* 325:665–669
- Evans MJ, Kaufman MH (1981) Establishment in culture of pluripotent cells from mouse embryos. *Nature* 292:154–156
- Fajans SS, Bell GI, Polonsky KS (2001) Molecular mechanisms and clinical pathophysiology of maturity-onset diabetes of the young. *N Engl J Med* 345:971–980
- Fandrich F, Ungefroren H (2010) Customized cell-based treatment options to combat autoimmunity and restore beta-cell function in type 1 diabetes mellitus: current protocols and future perspectives. *Adv Exp Med Biol* 654:641–665
- Favaro E, Carpanetto A, Lamorte S, Fusco A, Caorsi C, Deregibus MC, Bruno S, Amoroso A, Giovarelli M, Porta M, Perin PC, Tetta C, Camussi G, Zanone MM (2014) Human mesenchymal stem cell-derived microvesicles modulate T cell response to islet antigen glutamic acid decarboxylase in patients with type 1 diabetes. *Diabetologia* 57:1664–1673
- Fiorina P, Jurewicz M, Augello A et al (2009) Immunomodulatory function of bone marrow-derived mesenchymal stem cells in experimental autoimmune type 1 diabetes. *J Immunol* 183:993–1004
- Fiorina P, Voltarelli J, Zavazava N (2011) Immunological applications of stem cells in type 1 diabetes. *Endocr Rev* 32:725–754
- Friedenstein AJ, Piatetzky-Shapiro II, Petrakova KV (1966) Osteogenesis in transplants of bone marrow cells. *J Embryol Exp Morphol* 16:381–390
- Fuchs E (2009) Finding one's niche in the skin. *Cell Stem Cell* 2009(4):499–502
- Fujikawa T, Oh SH, Pi L, Hatch HM, Shupe T, Petersen BE (2005) Teratoma formation leads to failure of treatment for type 1 diabetes using embryonic stem cell-derived insulin-producing cells. *Am J Pathol* 166:1781–1791
- Gale EA (2005) Type 1 diabetes in the young: the harvest of sorrow goes on. *Diabetologia* 48:1435–1438
- Gibby RF, Graham JG, Luo X, Lowe WL Jr, Hering BJ, Shea LD (2011) Advancing islet transplantation: from engraftment to the immune response. *Diabetologia* 54:2494–2505
- Gluckman E, Broxmeyer HA, Auerbach AD et al (1989) Hematopoietic reconstitution in a patient with Fanconi's anemia by means of umbilical-cord blood from an HLA-identical sibling. *N Engl J Med* 321:1174–1178
- Greenbaum C, Atkinson MA (2011) Persistence is the twin sister of excellence: an important lesson for attempts to prevent and reverse type 1 diabetes. *Diabetes* 60:693–694
- Gruessner AC, Sutherland DE (2001) Report from the international pancreas transplant registry 2000. *Transplant Proc* 33:1643–1646
- Gruessner AC, Sutherland DER (2005) Pancreas transplant outcomes for United States (US) and non-US cases as reported to the United Network for Organ Sharing (UNOS) and the International Pancreas Transplant Registry (IPTR) as of June 2004. *Clin Transpl* 19:433–455
- Gruessner RW, Burke GW, Stratta R et al (1996) Multicenter analysis of the first experience with FK506 for induction and rescue therapy after pancreas transplantation. *Transplantation* 61:261–273
- Gruss P (2003) Human ES cells in Europe. *Science* 301:1017
- Halban PA (2004) Cellular sources of new pancreatic beta cells and therapeutic implications for regenerative medicine. *Nat Cell Biol* 6:1021–1025

- Haller MJ, Wasserfall CH, McGrail KM et al (2009) Autologous umbilical cord blood transfusion in very young children with type 1 diabetes. *Diabetes Care* 32:2041–2046
- Halvorsen TL, Beattie GM, Lopez AD, Hayek A, Levine F (2000) Accelerated telomere shortening and senescence in human pancreatic islet cells stimulated to divide in vitro. *J Endocrinol* 166: 103–109
- Hamada H, Kobune M, Nakamura K, Kawano Y, Kato K, Honmou O, Houkin K, Matsunaga T, Niitsu Y (2005) Mesenchymal stem cells (MSC) as therapeutic cytoreagents for gene therapy. *Cancer Sci* 96:149–156
- Hansson M, Tonning A, Frandsen U et al (2004) Artifactual insulin release from differentiated embryonic stem cells. *Diabetes* 53:2603–2609
- Harjutsalo V, Sjöberg L, Tuomilehto J (2008) Time trends in the incidence of type 1 diabetes in Finnish children: a cohort study. *Lancet* 371:1777–1782
- Hoogduijn MJ, Popp F, Verbeek R et al (2010) The immunomodulatory properties of mesenchymal stem cells and their use for immunotherapy. *Int Immunopharmacol* 10:1496–1500
- Hori Y, Gu X, Xie X, Kim SK (2005) Differentiation of insulin-producing cells from human neural progenitor cells. *PLoS Med* 2:e103
- Hou SX, Singh SR (2008) *Germline stem cells*. Springer, New York, p 450
- Houard N, Rousseau GG, Lemaigre FP (2003) HNF-6-independent differentiation of mouse embryonic stem cells into insulin producing cells. *Diabetologia* 46:378–385
- Izadpanah R, Trygg C, Patel B et al (2006) Biologic properties of mesenchymal stem cells derived from bone marrow and adipose tissue. *J Cell Biochem* 99:1285–1297
- Jahansouz C, Kumer SC, Ellenbogen M, Brayman KL (2011) Evolution of beta-cell replacement therapy in diabetes mellitus: pancreas transplantation. *Diabetes Technol Ther* 13:395–418
- Jahr H, Bretzel RG (2003) Insulin-positive cells in vitro generated from rat bone marrow stromal cells. *Transplant Proc* 35:2140–2141
- Johansson H, Lukinius A, Moberg L et al (2005) Tissue factor produced by the endocrine cells of the islets of Langerhans is associated with a negative outcome of clinical islet transplantation. *Diabetes* 54:1755–1762
- Jurewicz M, Yang S, Augello A et al (2010) Congenic mesenchymal stem cell therapy reverses hyperglycemia in experimental type 1 diabetes. *Diabetes* 59:3139–3147
- Kahn CR (1994) Insulin action, diabetogene and the cause of type II diabetes: banting lecture. *Diabetes* 43:1066–1084
- Kahn HS, Morgan TM, Case LD et al (2009) Association of type 1 diabetes with month of birth among US youth: the SEARCH for diabetes in youth study. *Diabetes Care* 32:2010–2015
- Kang EM, Zickler PP, Burns S et al (2005) Hematopoietic stem cell transplantation prevents diabetes in NOD mice but does not contribute to significant islet cell regeneration once disease is established. *Exp Hematol* 33(6):699–705
- Karnieli O, Izhar-Prato Y, Bulvik S, Efrat S (2007) Generation of insulin-producing cells from human bone marrow mesenchymal stem cells by genetic manipulation. *Stem Cells* 25: 2837–2844
- Kassem SA, Ariel I, Thornton PS, Scheimberg I, Glaser B (2000) Beta-cell proliferation and apoptosis in the developing normal human pancreas and in hyperinsulinism of infancy. *Diabetes* 49:1325–1333
- Kelly WD, Lillehei RC, Merkel FK, Idezuki Y, Goetz FC (1967) Allotransplantation of the pancreas and duodenum along with the kidney in diabetic nephropathy. *Surgery* 61:827–837
- Kern S, Eichler H, Stoeve J et al (2006) Comparative analysis of mesenchymal stem cells from bone marrow, umbilical cord blood, or adipose tissue. *Stem Cells* 24:1294–1301
- Kilk K, Magzoub M, Pooga M, Eriksson LE, Langel U, Gräslund A (2001) Cellular internalization of a cargo complex with a novel peptide derived from the third helix of the islet-1 homeodomain. Comparison with the penetratin peptide. *Bioconj Chem* 12:911–916
- Knip M, Virtanen SM, Akerblom HK (2010) Infant feeding and the risk of type 1 diabetes. *Am J Clin Nutr* 91:1506–1513
- Kroon E, Martinson LA, Kadoya K et al (2008) Pancreatic endoderm derived from human embryonic stem cells generates glucose-responsive insulin-secreting cells in vivo. *Nat Biotechnol* 26:443–452

- Kubo A, Shinozaki K, Shannon JM et al (2004) Development of definitive endoderm from embryonic stem cells in culture. *Development* 131:1651–1662
- Kucia M, Halasa M, Wysoczynski M et al (2006) Morphological and molecular characterization of novel population of CXCR4+; SSEA-4+; Oct-4+; very small embryonic-like cells purified from human cord blood—preliminary report. *Leukemia* 21:297–303
- Kukko M, Kimpimaki T, Korhonen S et al (2005) Dynamics of diabetes-associated autoantibodies in young children with human leukocyte antigen-conferred risk of type 1 diabetes recruited from the general population. *J Clin Endocrinol Metab* 90:2712–2717
- Lakey JR, Burridge PW, Shapiro AM (2003) Technical aspects of islet preparation and transplantation. *Transpl Int: Off J Eur Soc Organ Transplant* 16:613–632
- Landry DW, Zucker HA (2004) Embryonic death and the creation of human embryonic stem cells. *J Clin Invest* 114:1184–1186
- Langerhans P (1869) *Beiträge zur mikroskopischen Anatomie der Bauchspeicheldrüse*. Inaug.-Diss, Berlin
- Lee RH, Seo MJ, Reger RL et al (2006) Multipotent stromal cells from human marrow home to and promote repair of pancreatic islets and renal glomeruli in diabetic NOD/scid mice. *Proc Natl Acad Sci U S A* 103:17438–17443
- Leibiger IB, Berggren PO (2008) Insulin signaling in the pancreatic β -cell. *Annu Rev Nutr* 28:233–251
- Leroith D, Taylor SI, Oefky JM (eds) (2003) *Diabetes mellitus. A fundamental and clinical text*, 3rd edn. Lippincott, Williams Wilkins, Philadelphia
- Li Y, Zhang R, Qiao H, Zhang H, Wang Y, Yuan H, Liu Q, Liu D, Chen L, Pei X (2007) Generation of insulin-producing cells from PDX-1 gene-modified human mesenchymal stem cells. *J Cell Physiol* 211:36–44
- Liu Z, Sall A, Yang D (2008) MicroRNA: an emerging therapeutic target and intervention tool. *Int J Mol Sci* 9:978–999
- Liu X, Wang Y, Li Y, Pei X (2013) Research status and prospect of stem cells in the treatment of diabetes mellitus. *Sci China Life Sci* 56:306–312
- Lu LL, Liu YJ, Yang SG et al (2006) Isolation and characterization of human umbilical cord mesenchymal stem cells with hematopoiesis-supportive function and other potentials. *Haematologica* 91:1017–1026
- Ludvigsson J, Krisky D, Casas R et al (2012) GAD65 antigen therapy in recently diagnosed type 1 diabetes mellitus. *N Engl J Med* 366:433–442
- Lumelsky N, Blondel O, Laeng P, Velasco I, Ravin R, McKay R (2001) Differentiation of embryonic stem cells to insulin-secreting structures similar to pancreatic islets. *Science* 292:1389–1394
- Maahs DM, West NA, Lawrence JM, Mayer-Davis EJ (2010) Epidemiology of type 1 diabetes. *Endocrinol Metab Clin N Am* 39:481–497
- MacCracken J, Hoel D (1997) From ants to analogues: puzzles and promises in diabetes management. *Postgrad Med* 101(4):138–140. commentaries
- Maclaren N, Atkinson M (1992) Is insulin-dependent diabetes mellitus environmentally induced? *N Engl J Med* 327:348–349
- Madec AM, Mallone R, Afonso G et al (2009) Mesenchymal stem cells protect NOD mice from diabetes by inducing regulatory T cells. *Diabetologia* 52:1391–1399
- Martin GR (1981) Isolation of a pluripotent cell line from early mouse embryos cultured in medium conditioned by teratocarcinoma stem cells. *Proc Natl Acad Sci U S A* 78:7634–7638
- Masoud MS, Anwar SS, Afzal MZ, Mehmood A, Khan SN, Riazuddin S (2012) Pre-conditioned mesenchymal stem cells ameliorate renal ischemic injury in rats by augmented survival and engraftment. *J Transl Med* 10:243
- Matsumoto S, Okitsu T, Iwanaga Y et al (2005) Insulin independence after living-donor distal pancreatectomy and islet allotransplantation. *Lancet* 365:1642–1644
- Matsumoto S, Okitsu T, Iwanaga Y et al (2006) Follow-up study of the first successful living donor islet transplantation. *Transplantation* 82:1629–1633
- McGuckin CP, Forraz N (2008) Potential for access to embryonic-like cells from human umbilical cord blood. *Cell Prolif* 41:31–40

- Meier JJ, Butler PC (2005) Insulin secretion. In: Endocrinology. Elsevier Saunders, Philadelphia
- Meirelles Lda S, Nardi NB (2009) Methodology, biology and clinical applications of mesenchymal stem cells. *Front Biosci* 14:4281–4298
- Meissner A, Jaenisch R (2006) Generation of nuclear transfer-derived pluripotent ES cells from cloned Cdx2-deficient blastocysts. *Nature* 439:212–215
- Mering JV, Minkowski O (1889) Diabetes mellitus nach Pankreasexstirpation. *Zschr klin Med* 14:404–423
- Mineo D, Ciancio G, Burke GW, Alejandro R, Ricordi C (2010) Islet and pancreas transplantation. In: Efrat S (ed) Stem cell therapy for diabetes. Stem cell biology and regenerative medicine. Humana Press, New York, pp 41–83. doi:10.1007/978-1-60761-366-4_2
- Mishra PK, Tyagi N, Kumar M, Tyagi SC (2009) MicroRNAs as a therapeutic target for cardiovascular disease. *J Cell Mol Med* 13:778–789
- Moberg L, Johansson H, Lukinius A et al (2002) Production of tissue factor by pancreatic islet cells as a trigger of detrimental thrombotic reactions in clinical islet transplantation. *Lancet* 360:2039–2045
- Mokarizadeh A, Delirezh N, Morshedi A, Mosayebi G, Farshid AA, Mardani K (2012) Microvesicles derived from mesenchymal stem cells: potent organelles for induction of tolerogenic signaling. *Immunol Lett* 147:47–54
- Moltchanova EV, Schreier N, Lammi N, Karvonen M (2009) Seasonal variation of diagnosis of type 1 diabetes mellitus in children worldwide. *Diabet Med* 26:673–678
- Murphy R, Ellard S, Hattersley AT (2008) Clinical implications of a molecular genetics classification of monogenic β -cell diabetes. *Nat Clin Pract Endocrinol Metab* 4:200
- Nilsson B, Ekdahl KN, Korsgren O (2011) Control of instant blood-mediated inflammatory reaction to improve islets of Langerhans engraftment. *Curr Opin Organ Transplant* 16:620–626
- Noguchi H, Matsumoto S (2006) Protein transduction technology offers a novel therapeutic approach for diabetes. *J Hepato-Biliary-Pancreat Surg* 13:306–313
- Noguchi H, Kaneto H, Weir GC, Bonner-Weir S (2003) PDX-1 protein containing its own antennapedia-like protein transduction domain can transduce pancreatic duct and islet cells. *Diabetes* 52:1732–1737
- Noguchi H, Bonner-Weir S, Wei FY, Matsushita M, Matsumoto S (2005) BETA2/neuro D protein can be transduced into cells due to an arginine- and lysine-rich sequence. *Diabetes* 54:2859–2866
- Oakley J (2002) Democracy, embryonic stem cell research, and the roman catholic church. *Med Ethics* 28:22856
- Oedayrajsingh-Varma M, van Ham S, Knippenberg M et al (2006) Adipose tissue-derived mesenchymal stem cell yield and growth characteristics are affected by the tissue-harvesting procedure. *Cytotherapy* 8:166–177
- Oh SH, Muzzonigro TM, Bae SH, LaPlante JM, Hatch HM, Petersen BE (2004) Adult bone marrow-derived cells trans-differentiating into insulin-producing cells for the treatment of type I diabetes. *Lab Invest* 84:607–617
- Ohneda K, Ee H, German M (2000) Regulation of insulin gene transcription. *Semin Cell Dev Biol* 11:227–233
- Okita K, Yamakawa T, Matsumura Y et al (2013) An efficient nonviral method to generate integration-free human-induced pluripotent stem cells from cord blood and peripheral blood cells. *Stem Cells* 31:458–466
- Orban T, Bundy B, Becker DJ et al (2011) Co-stimulation modulation with abatacept in patients with recent-onset type 1 diabetes: a randomised, double-blind, placebo-controlled trial. *Lancet* 378:412–419
- Orlando G, Gianello P, Salvatori M, Stratta RJ, Soker S, Ricordi C et al (2014) Cell replacement strategies aimed at reconstitution of the beta-cell compartment in type 1 diabetes. *Diabetes* 63:1433–1444

- Ostman J, Lonnberg G, Arnqvist HJ et al (2008) Gender differences and temporal variation in the incidence of type 1 diabetes: results of 8012 cases in the nationwide diabetes incidence study in Sweden 1983–2002. *J Intern Med* 263:386–394
- Pap E, Pállinger E, Falus A (2011) The role of membrane vesicles in tumorigenesis. *Crit Rev Oncol Hematol* 79:213–223
- Passweg J, Tyndall A (2007) Autologous stem cell transplantation in autoimmune diseases. *Semin Hematol* 44:278–285
- Patterson CC, Dahlquist GG, Gyürüs E, Green A, Soltész G, EURODIAB Study Group (2009) Incidence trends for childhood type 1 diabetes in Europe during 1989–2003 and predicted new cases 2005–20: a multicentre prospective registration study. *Lancet* 373:2027–2033
- Pescovitz MD, Greenbaum CJ, Krause-Steinrauf H et al (2009) Rituximab, B-lymphocyte depletion, and preservation of beta-cell function. *N Engl J Med* 361:2143–2152
- Pickup JC, William G (2003) The history of diabetes mellitus, *Textbook of diabetes*, vol 1, 3rd edn. Blackwell Science Limited, Oxford
- Poitout V, Robertson RP (2008) Glucolipotoxicity: fuel excess and β -cell dysfunction. *Endocr Rev* 29:351–366
- Polonsky KS, Pugh W, Jaspan JB, Cohen DM, Karrison T, Tager HS, Rubenstein AH (1984) C-peptide and insulin secretion. Relationship between peripheral concentrations of C-peptide and insulin and their secretion rates in the dog. *J Clin Invest* 74:1821–1829
- Posselt AM, Szot GL, Frassetto LA, Masharani U, Tavakol M et al (2010) Islet transplantation in type 1 diabetic patients using calcineurin inhibitor-free immunosuppressive protocols based on T-cell adhesion or costimulation blockade. *Transplantation* 90:1595–1601
- Pozzilli P (2012) Type 1 diabetes mellitus in 2011: heterogeneity of T1DM raises questions for therapy. *Nat Rev Endocrinol* 8:78–80
- Prokhorova TA, Harkness LM, Frandsen U, Ditzel N, Schroder HD, Burns JS et al (2009) Teratoma formation by human embryonic stem cells is site dependent and enhanced by the presence of Matrigel. *Stem Cells Dev* 18:47–54
- Pybus FC (1924) Notes on suprarenal and pancreatic grafting. *Lancet* 204:550–551
- Rackham CL, Chagastelles PC, Nardi NB et al (2011) Co-transplantation of mesenchymal stem cells maintains islet organisation and morphology in mice. *Diabetologia* 54:1127–1135
- Rahier J, Goebbels RM, Henquin JC (1983) Cellular composition of the human diabetic pancreas. *Diabetologia* 24:366–371
- Ramiya VK, Maraist M, Arfors KE, Schatz DA, Peck AB, Cornelius JG (2000) Reversal of insulin-dependent diabetes using islets generated in vitro from pancreatic stem cells. *Nat Med* 6:278–282
- Ratajczak J, Miekus K, Kucia M, Zhang J, Reca R, Dvorak P, Ratajczak MZ (2006) Embryonic stem cell-derived microvesicles reprogram hematopoietic progenitors: evidence for horizontal transfer of mRNA and protein delivery. *Leukemia* 20:847–856
- Ricordi C (ed) (1992) 1892–1992. One century of transplantation for diabetes: pancreatic islet cell. Transplantation. R.G. Landes Company, Austin, p 291
- Ricordi C, Lacy PE, Scharp DW (1989) Automated islet isolation from human pancreas. *Diabetes* 38:140–142
- Robbins PD, Morelli AE (2014) Regulation of immune responses by extracellular vesicles. *Nat Rev Immunol* 14:195–208
- Ryan EA, Paty BW, Senior PA et al (2005) Five-year follow-up after clinical islet transplantation. *Diabetes* 54:2060–2069
- Scharp DW, Lacy PE, Santiago JV et al (1990) Insulin independence after islet transplantation into type I diabetic patient. *Diabetes* 39:515–518
- Scheres B (2007) Stem-cell niches: nursery rhymes across kingdoms. *Nat Rev Mol Cell Biol* 8:345–354
- Schlott NC, Meierhoff G, Lengyel C et al (2007) Effect of heat shock protein peptide DiaPep277 on beta-cell function in paediatric and adult patients with recent-onset diabetes mellitus type 1: two prospective, randomized, double-blind phase II trials. *Diabetes Metab Res Rev* 23:276–285

- Seaberg RM, Smukler SR, Kieffer TJ et al (2004) Clonal identification of multipotent precursors from adult mouse pancreas that generate neural and pancreatic lineages. *Nat Biotechnol* 22:1115–1124
- Segev H, Fishman B, Ziskind A, Shulman M, Itskovitz-Eldor J (2004) Differentiation of human embryonic stem cells into insulin-producing clusters. *Stem Cells* 22:265–274
- Shapiro AM, Lakey JR, Ryan EA et al (2000) Islet transplantation in seven patients with type 1 diabetes mellitus using a glucocorticoid-free immunosuppressive regimen. *N Engl J Med* 343:230–238
- Shapiro AM, Ricordi C, Hering BJ et al (2006) International trial of the Edmonton protocol for islet transplantation. *N Engl J Med* 355:1318–1330
- Sherry N, Hagopian W, Ludvigsson J et al (2011) Teplizumab for treatment of type 1 diabetes (Protege study): 1-year results from randomised, placebo-controlled trial. *Lancet* 378: 487–497
- Siminovitch L, McCulloch EA, Till JE (1963) The distribution of colony-forming cells among spleen colonies. *J Cell Physiol* 62:327–336
- Sims E, Evans-Molina C (2012) Stem cells as a tool to improve outcomes of islet transplantation. *J Transplant* 2012:736491
- Singh SR, Hou SX (2008) Lessons learned about adult kidney stem cells from the malpighian tubules of *Drosophila*. *J Am Soc Nephrol* 19:660–666
- Singh SR, Hou SX (2009) Multipotent stem cells in the Malpighian tubules of adult *Drosophila melanogaster*. *J Exp Biol* 212:413–423
- Singh SR, Liu W, Hou SX (2007) The adult *Drosophila* malpighian tubules are maintained by multipotent stem cells. *Cell Stem Cell* 1:191–203
- Soria B, Roche E, Berná G, Leon-Quinto T, Reig JA, Martin F (2000) Insulin-secreting cells derived from embryonic stem cells normalize glycemia in streptozotocin induced diabetic mice. *Diabetes* 49:157–162
- Sosenko JM, Skyler JS, Mahon J et al (2012) The application of the diabetes prevention trial-type 1 risk score for identifying a preclinical state of type 1 diabetes. *Diabetes Care* 35:1552–1555
- Spaggiari GM, Abdelrazik H, Becchetti F, Moretta L (2009) MSCs inhibit monocyte-derived DC maturation and function by selectively interfering with the generation of immature DCs: central role of MSC-derived prostaglandin E2. *Blood* 113:6576–6583
- Sprent J, Kishimoto H (2001) The thymus and central tolerance. *Philos Trans R Soc Lond B Biol Sci* 356:609–616
- Stadtfeld M, Nagaya M, Utikal J, Weir G, Hochedlinger K (2008) Induced pluripotent stem cells generated without viral integration. *Science* 322:945–949
- Staeva TP, Chatenoud L, Insel R, Atkinson MA (2013) Recent lessons learned from prevention and recent-onset type 1 diabetes immunotherapy trials. *Diabetes* 62:9–17
- Starzl TE (2001) The “privileged” liver and hepatic tolerogenicity. *Liver Transpl* 7:918–920
- Stefan Y, Orsi L, Malaisse-Lagae F, Perrelet A, Patel Y, Unger RH (1982) Quantitation of endocrine cell content in the pancreas of non-diabetic and diabetic humans. *Diabetes* 31:694–700
- Stene LC, Rewers M (2012) Immunology in the clinic review series; focus on type 1 diabetes and viruses: the enterovirus link to type 1 diabetes: critical review of human studies. *Clin Exp Immunol* 168:12–23
- Subramanian S, Trencle DL (2007) Immunosuppressive agents: effects on glucose and lipid metabolism. *Endocrinol Metab Clin N Am* 36:891–905
- Sun Y, Chen L, Hou XG, Hou WK, Dong JJ, Sun L et al (2007) Differentiation of bone marrow derived mesenchymal stem cells from diabetic patients into insulin-producing cells in vitro. *Chin Med J* 120:771–776
- Suzuki A, Nakauchi H, Taniguchi H (2004) Prospective isolation of multipotent pancreatic progenitors using flow-cytometric cell sorting. *Diabetes* 53:2143–2152
- Svoren BM, Volkening LK, Wood JR, Laffel LM (2009) Significant vitamin D deficiency in youth with type 1 diabetes mellitus. *J Pediatr* 154:132–134
- Sykes M, Nikolic B (2005) Treatment of severe autoimmune disease by stem-cell transplantation. *Nature* 435:620–627

- Takahashi K, Yamanaka S (2006) Induction of pluripotent stem cells from mouse embryonic and adult fibroblast cultures by defined factors. *Cell* 126:663–676
- Tariq M, Masoud MS, Mehmood A, Khan SN, Riazuddin S (2013) Stromal cell derived factor-1 α protects stem cell derived insulin-producing cells from glucotoxicity under high glucose conditions in-vitro and ameliorates drug induced diabetes in rats. *J Transl Med* 11:115
- Thakkar UG, Trivedi HL, Vanikar AV, Dave SD (2015) Insulin-secreting adipose-derived mesenchymal stromal cells with bone marrow-derived hematopoietic stem cells from autologous and allogenic sources for type 1 diabetes mellitus. *Cytotherapy* 17:940–947
- The American Diabetes Association (2009) Diagnosis and classification of diabetes mellitus. *Diabetes Care* 32:S62–S67
- The Diabetes Control and Complications Trial Research Group (1993) The effect of intensive treatment of diabetes on the development and progression of long-term complications in insulin-dependent diabetes mellitus. *N Engl J Med* 329:977–986
- Théry C, Ostrowski M, Segura E (2009) Membrane vesicles as conveyors of immune responses. *Nat Rev Immunol* 9:581–593
- Thomson JA, Itskovitz-Eldor J, Shapiro SS et al (1998) Embryonic stem cell lines derived from human blastocysts. *Science* 282:1145–1147
- Thunander M, Petersson C, Jonzon K et al (2008) Incidence of type 1 and type 2 diabetes in adults and children in Kronoberg, Sweden. *Diabetes Res Clin Pract* 82:247–255
- Trivedi HL, Vanikar AV, Thakker U et al (2008) Human adipose tissue-derived mesenchymal stem cells combined with hematopoietic stem cell transplantation synthesize insulin. *Transplant Proc* 40:1135–1139
- Ullrich A, Shrine J, Chirgwin J (1977) Rat insulin genes construction of plasmids containing the coding sequence. *Science* 196:1313–1319
- Urban VS, Kiss J, Kovacs J et al (2008) Mesenchymal stem cells cooperate with bone marrow cells in therapy of diabetes. *Stem Cells* 26:244–253
- Valadi H, Ekström K, Bossio A, Sjöstrand M, Lee JJ, Lötvall JO (2007) Exosome-mediated transfer of mRNA and micro RNAs is a novel mechanism of genetic exchange between cells. *Nat Cell Biol* 9:654–659
- Vanikar AV, Dave SD, Thakkar UG, Trivedi HL (2010) Co-transplantation of adipose tissue-derived insulin-secreting mesenchymal stem cells and hematopoietic stem cells: a novel therapy for insulin-dependent diabetes mellitus. *Stem Cells Int* 2010 :5. doi:10.4061/2010/582382 Article ID 582382
- Vanikar AV, Trivedi HL, Patel RD, Kanodia KV, Modi PR, Shah VR (2012) Allogenic hematopoietic stem cell transplantation in pemphigus vulgaris: a single-center experience. *Indian J Dermatol* 57:9–11
- Vantyghem MC, Marcelli-Tourvielle S, Pattou F et al (2007) Effects of non-steroid immunosuppressive drugs on insulin secretion in transplantation. *Ann Endocrinol* 68:21–27
- Vendrame F, Pileggi A, Laughlin E et al (2010) Recurrence of type 1 diabetes after simultaneous pancreas-kidney transplantation, despite immunosuppression, is associated with autoantibodies and pathogenic autoreactive CD4 T-cells. *Diabetes* 59:947–957
- Voltarelli JC, Couri CE, Rodrigues MC et al (2011) Stem cell therapies for type 1 diabetes mellitus. *Indian J Exp Biol* 49:395–400
- Walter M, Philotheou A, Bonnici F, Ziegler AG, Jimenez R (2009) No effect of the altered peptide ligand NBI-6024 on beta-cell residual function and insulin needs in new-onset type 1 diabetes. *Diabetes Care* 32:2036–2040
- Wang V, Wu W (2009) MicroRNA-based therapeutics for cancer. *Bio Drugs* 23:15–23
- Wang RN, Kloppel G, Bouwens L (1995) Duct- to islet-cell differentiation and islet growth in the pancreas of duct-ligated adult rats. *Diabetologia* 38:1405–1411
- Wang HS, Shyu JF, Shen WS et al (2011) Transplantation of insulin-producing cells derived from umbilical cord stromal mesenchymal stem cells to treat NOD mice. *Cell Transplant* 20:455–466
- Watson R (2003) Euro MPs threaten UK stem cell research. *BMJ* 326:838

- Webb MA, Dennison AR, James RF (2012) The potential benefit of non-purified islets preparations for islet transplantation. *Biotechnol Genet Eng Rev* 28:101–114
- Wherrett DK, Bundy B, Becker DJ et al (2011) Antigen-based therapy with glutamic acid decarboxylase (GAD) vaccine in patients with recent-onset type 1 diabetes: a randomised double-blind trial. *Lancet* 378:319–327
- Wilkin TJ (2008) Diabetes 1 and 2, or one and the same? Progress with the accelerator hypothesis. *Pediatr Diabetes* 9:23–32
- Winkler C, Krumsiek J, Lempainen J et al (2012) A strategy for combining minor genetic susceptibility genes to improve prediction of disease in type 1 diabetes. *Genes Immun* 13: 549–555
- World Health Organisation (1999) Definition, diagnosis and classification of diabetes mellitus and its complications. Part 1: diagnosis and classification of diabetes mellitus. WHO/NCD/NCS/99.2. Geneva. Ref Type: Report
- Xu J, Lu Y, Ding F, Zhan X, Zhu M, Wang Z (2007) Reversal of diabetes in mice by intrahepatic injection of bone-derived GFP murine mesenchymal stem cells infected with the recombinant retrovirus-carrying human insulin gene. *World J Surg* 31:1872–1882
- Xu X, D'Hoker J, Stange G et al (2008) β -cells can be generated from endogenous progenitors in injured adult mouse pancreas. *Cell* 132:197–207
- Yamashita YM (2009) Regulation of asymmetric stem cell division: spindle orientation and the centrosome. *Front Biosci* 14:3003–3011
- Yañez R, Lamana ML, García-Castro J, Colmenero I, Ramírez M, Bueren JA (2006) Adipose tissue-derived mesenchymal stem cells have in vivo immunosuppressive properties applicable for the control of the graft-versus-host disease. *Stem Cells* 24:2582–2591
- Yechoor V, Liu V, Espiritu C et al (2009) Neurogenin3 is sufficient for transdetermination of hepatic progenitor cells into neo-islets in vivo but not transdifferentiation of hepatocytes. *Dev Cell* 16:358–373
- Yeung WC, Rawlinson WD, Craig ME (2011) Enterovirus infection and type 1 diabetes mellitus: systematic review and meta-analysis of observational molecular studies. *BMJ* 342:35
- Yoon JW, Jun HS (2005) Autoimmune destruction of pancreatic β -cells. *Am J Ther* 12:580–591
- Zaret KS, Grompe M (2008) Generation and regeneration of cells of the liver and pancreas. *Science* 322:1490–1494
- Zhang J, Shehabeldin A, da Cruz LA, Butler J, Somani AK, McGavin M et al (1999) Antigen receptor-induced activation and cytoskeletal rearrangement are impaired in Wiskott-Aldrich syndrome protein-deficient lymphocytes. *J Exp Med* 190:1329–1342
- Zimmet P, Alberti KG, Shaw J (2001) Global and societal implications of the diabetes epidemic. *Nature* 414:782–787

Pancreas, Kidney and Skin Regeneration

Pham, P.V. (Ed.)

2017, XI, 326 p. 29 illus., 23 illus. in color., Hardcover

ISBN: 978-3-319-55686-4