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# Selective destruction of mouse islet beta cells by human T lymphocytes in a newly-established humanized type 1 diabetic model

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ABSTRACT

Type 1 diabetes (T1D) is caused by a T cell-mediated autoimmune response that leads to the loss of insulin-producing β cells. The optimal preclinical testing of promising therapies would be aided by a humanized immune-mediated T1D model. We develop this model in NOD-scid IL2rγ$^{null}$ mice. The selective destruction of pancreatic islet β cells was mediated by human T lymphocytes after an initial trigger was supplied by the injection of irradiated spleen mononuclear cells (SMC) from diabetic nonobese diabetic (NOD) mice. This resulted in severe insulitis, a marked loss of total β-cell mass, and other related phenotypes of T1D. The migration of human T cells to pancreatic islets was controlled by the β cell-produced highly conserved chemokine stromal cell-derived factor 1 (SDF-1) and its receptor C-X-C chemokine receptor (CXCR) 4, as demonstrated by *in vivo* blocking experiments using antibody to CXCR4. The specificity of humanized T cell-mediated immune responses against islet β cells was generated by the local inflammatory microenvironment in pancreatic islets including human CD4$^+$ T cell infiltration and clonal expansion, and the mouse islet β-cell-derived CD1d-mediated human iNKT activation. The selective destruction of mouse islet β cells by a human T cell-mediated immune response in this humanized T1D model can mimic those observed in T1D patients. This model can provide a valuable tool for translational research into T1D.



## 1. Introduction

Immune cell infiltration and destruction of β cells in pancreatic islets are the hallmark features of T1D[1], both in diabetic patients and animal models [1]. Among animal T1D models, the nonobese diabetic (NOD) mice and biobreeding (BB) rats spontaneously develop an immune-mediated T1D resembling that in humans. They have been widely used for exploring T1D pathogenesis and drug development [2–5]. However, increasing evidence demonstrates that these rodent models have limited usefulness due to key differences between the human and rodent immune system [6,7]. Therefore, the establishment of a human immunocyte-mediated disease model would provide unique and reliable tool for modern translational medicine to promote the development of new therapies [4–7], such as human blood stem cell-based therapies [8–10] and umbilical cord blood stem cell-modulated regulatory T cell (Treg) immunotherapy [1]. Here, we have successfully established a type 1 diabetic mouse model in NOD-scid IL2rγ$^{null}$ mice that is mediated by human T lymphocytes from diabetic patients.

## 2. Materials and methods

### 2.1. Establishment of a humanized T1D model

We established a humanized immune system in mice by administering human PBMC intraperitoneally into NOD-scid IL2rγ$^{null}$ mice with a dosage of 2–5 × 10$^7$ cells/mouse in 300 μl PBS with a 27-gauge needle. One week later, we established a humanized

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[1] *Abbreviations used:* CXCR4, C-X-C chemokine receptor 4; IPGTT, intraperitoneal glucose tolerance test; iNKT, invariant Natural Killer T cells; MHC, major histocompatibility complex; NOD-scid IL2r$^{null}$ mice, nonobese diabetic-severe combined immunodeficient interleukin 2 receptor $^{null}$ mice; PBMC, peripheral blood mononuclear cells; SDF-1, stromal cell-derived factor-1; SMC, spleen mononuclear cells; T1D, type 1 diabetes.

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immune-mediated T1D model by administering the irradiated diabetic NOD mouse spleen mononuclear cells (SMC, $1 \times 10^7$ cells/mouse in 300 μl PBS, i.p.) (Supplementary data: Scheme 1). SMC were isolated from diabetic NOD mouse spleens as previously described [1], followed by irradiation with $^{60}$Co γ rays at a dose of 5000 rad. Mice were monitored twice a week for development of diabetes. To explore the kinetics of diabetes development, mice were sacrificed on day 1, 5, 10, and 14 respectively after administering the irradiated SMC, according to a protocol approved by the Animal Care Committee (ACC) of University of Illinois at Chicago.

## 3. Results and discussion

### 3.1. Establishment of a humanized immune system in NOD-scid IL2rγ$^{null}$ mice

To establish a humanized immune system, peripheral blood mononuclear cells (PBMC) isolated from healthy donors ($n = 12$) were transplanted into NOD-scid IL2rγ$^{null}$ mice. Three weeks later, mouse spleen, blood, and pancreatic lymph nodes were examined to evaluate the proportion of human immune cells by using anti-human monoclonal antibodies (mAbs). Initially, we assessed spleen mononuclear cells (SMC). Flow analysis showed that a substantial fraction of total SMC (80%) was positive for human T cells antigens including CD4$^+$ and CD8$^+$ T cells (Fig. S1A, left panel). The ratio of human CD4/CD8 T cells was 1.5 to ~2, consistent with that in the peripheral blood of healthy donors (Fig. S1A, right panel). Next, we examined other types of immune cells by using human mAbs against different cell lineage markers. We found the percentages of monocytes (CD14$^+$) and dendritic cells (DC, positive for CD11c and CD83) were significantly increased in humanized NOD-scid IL2rγ$^{null}$ mice when compared with those in the peripheral blood of humans; there were no significant differences in CD1a$^+$ DC and MΦ (positive for CD11b$^+$ and CD163$^+$). In contrast, human B lymphocyte markers CD19 and CD20 were down-regulated in humanized NOD-scid IL2rγ$^{null}$ mice compared with cells from the peripheral blood of humans (Fig. S1B). The proportion of conventional CD4$^+$ CD25$^+$ regulatory T cells (Tregs) in humanized mouse SMC was less than that shown in human PBMC (Fig. S1C). Next, we examined the expression of co-stimulating molecules on antigen-presenting cells including DC and MΦ. Results demonstrated that humanized mouse SMC expressed higher levels of antigen-presenting function-related molecules including CD80, CD86, and HLA-DR; but decreased CD40 expression, with equivalent expression of HLA-DQ compared with that in human PBMC (Fig. S1D). Flow cytometric analysis of pancreatic lymph nodes and peripheral blood demonstrated the expression of the above markers equivalent to those expressed in the spleen site. Thus, these data indicate that a humanized immune system can be established in NOD-scid IL2rγ$^{null}$ mice by administering PBMC that retain their phenotype and function following transplant. Due to the difference between humans and mice, the transplanted human PBMC can naturally and physiologically adapt to the new environment with some phenotypic changes.

### 3.2. Priming of human T cells requires the irradiated diabetic NOD SMC to destroy mouse pancreatic islet β cells

To establish humanized immune system of T1D patients, PBMC isolated from type 1 diabetic donors were transplanted into NOD-scid IL2rγ$^{null}$ mice. Due to the constraints of HLA (human leukocyte antigen) molecules, autoimmune-mediated diabetes cannot be adoptively transferred from T1D patients to NOD-scid IL2rγ$^{null}$ mice by administering human PBMC (Fig. 1A, top right panel; Fig. S2A, red line and Fig. S2B, red bar). Additionally, pancreatic islet β cells display a very low level of MHC-class I antigen (Fig. S2C, top panels). Therefore, the low immunogenicity of islet β cells cannot directly evoke human immune cell responses against islet β cells (Fig 1A, top right panel; Fig. S2A, red line and Fig. S2B, red bar). In order to initiate the immune response, we infused irradiated spleen mononuclear cells (SMC) from diabetic NOD mice which can target the pancreatic islet β cells of NOD-scid IL2rγ$^{null}$ mice. These irradiated cells could not divide and expired within 24 h, but retained sufficient function to migrate through the basement membrane of pancreatic islets and causing peri-insulitis (Fig. 1A, bottom left panel). However, adoptive transfer with these irradiated diabetic SMC alone could not cause diabetes in NOD-scid IL2rγ$^{null}$ mice (Fig. S2A, green line and Fig. S2B, green bar). Notably, if adoptively transferred the irradiated diabetic SMC + human PBMC in NOD-scid IL2rγ$^{null}$ mice, pancreatic histology revealed profound insulitis with dramatic leukocyte infiltration into the islets (Fig. 1A, bottom right panel). To further demonstrate that the diabetogenic mouse T cells (including CD4$^+$ T cells and CD8$^+$ T cells) retained their potential to trigger the human T cell-mediated immune responses against mouse islet β cells after irradiation, we utilized 3-week-old (non-diabetic) NOD mouse SMC as a control group, which have few diabetogenic T cells [2]. These SMC were irradiated at the same irradiation dose and then transplanted at the same cell dose as those performed using diabetic NOD mouse SMC. After transplantation for two weeks, IPGTT results demonstrated that all mice ($n = 5$) in the group treated with 3-week-old NOD mouse SMC displayed normal glucose responses (Fig. 1B, red line) comparable with normal non-diabetic control mice (Fig. 1B, green line); however, all mice ($n = 5$) in the group treated with diabetic NOD mouse SMC showed impaired glucose tolerance and maintained high levels of glucose after 1.5 h (Fig. 1B, black line). Therefore, the data confirmed that diabetogenic T cells play a key role in initiating the immune destruction of islet β cells and retained their diabetogenic potential after a high dose of irradiation.

To document the migration of human immune cells into islets, we performed pancreatic histological analysis in conjunction with human cell markers, and scored insulitis two weeks after administering the irradiated diabetic NOD mouse SMC. The insulitis score demonstrated that ~80% of islet β cells (profound insulitis) were destroyed in humanized NOD-scid IL2rγ$^{null}$ mice; 17% of islets displayed severe insulitis and 3% of islets showed moderate insulitis, with no islets free of infiltration of inflammatory cells (Fig. 1C).

To verify that these infiltrated immune cells were derived from human lymphocytes, we performed double staining with anti-human mAbs to different T and B cell markers including CD4, CD8, Vα 24 Jα 18 TCR (for invariant NK T cells, iNKT cells), and CD20 in combination with insulin staining. Results demonstrated that a very large proportion of infiltrated cells were human CD4$^+$ (Fig. S3A), CD8$^+$ positive T cells (Fig. S3B), and Vα 24 Jα 18 TCR-positive iNKT cells (Fig. S3C) (a few CD8$^+$ Vα 24 Jα 18 TCR$^+$ iNKT cells, data not shown), with only a few CD20$^+$ positive B cells (Fig. S3D). These data imply that human T cells play a more important role than B cells in the β-cell destruction.

### 3.3. Pancreatic islet-released chemokine SDF-1 promotes the recruitment of human T cells to pancreatic islets and molecular mechanisms on β-cell destruction

To dissect the molecular nature preferentially controlling the migration of human CD4$^+$ T cells, CD8$^+$ T cells, and iNKT cells into mouse pancreatic islets, we focused on the chemokine stromal cell-derived factor (SDF-1) and its receptor CXCR4, which is known to control human CXCR4$^+$ cells homing to pancreatic islets and up-regulated during inflammation [10]. Immunostaining demonstrated that pancreatic islets of humanized diabetic NOD-scid IL2rγ$^{null}$ mice strongly expressed SDF-1 (Fig. 2A), but failed to

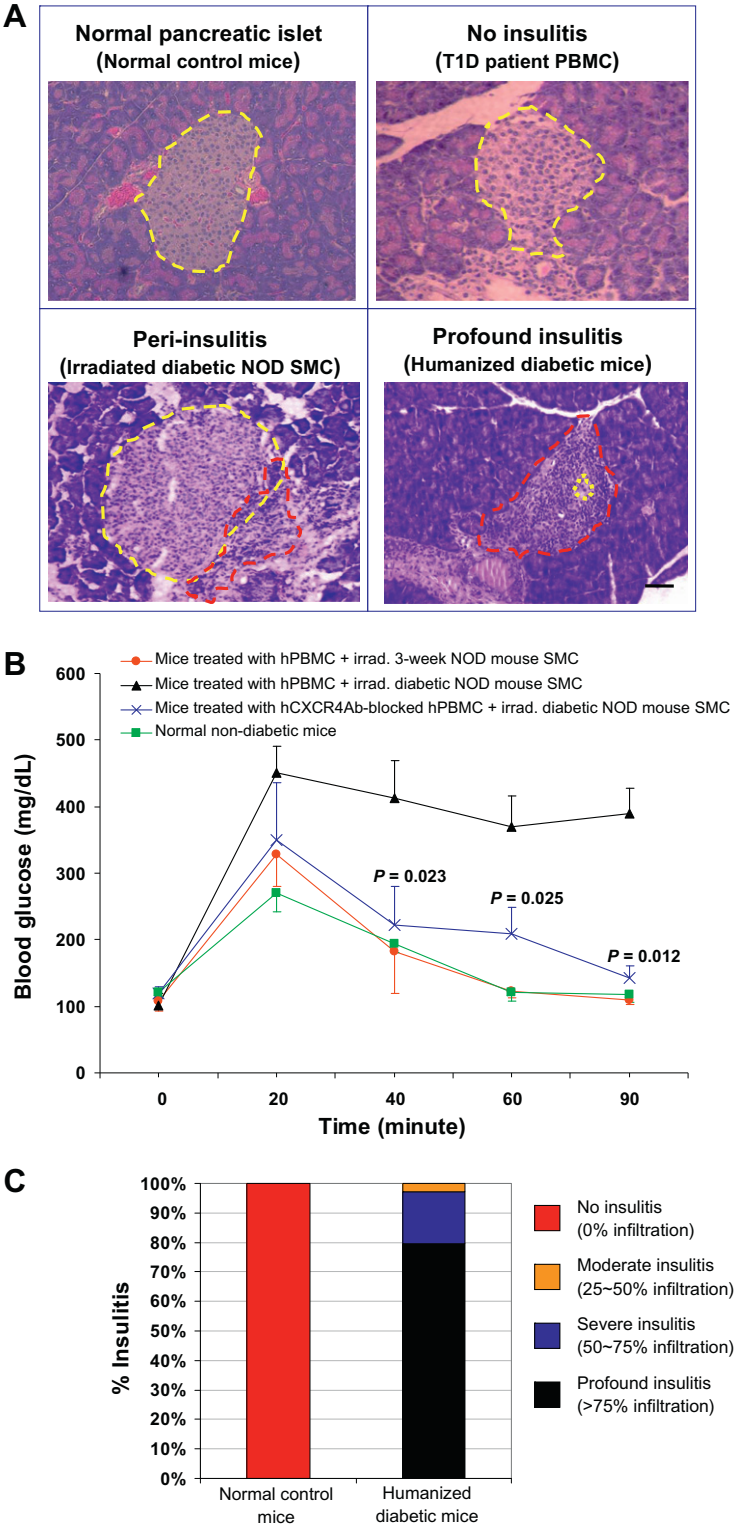


**Fig. 1.** Pancreatic insulitis caused by the infiltration of human T cells. (A) Hematoxylin and eosin (H&E) staining. No insulitis was showed in pancreata of normal NOD-scid IL-2rγ$^{null}$ mice (top left panel) and those of mice transplanted with T1D patient-derived PBMC (top right panel). Mice transplanted with the irradiated diabetic SMC displayed peri-insulitis (bottom left panel). After administering T1D patient-derived PBMC, mice transplanted with the irradiated diabetic SMC displayed profound insulitis (bottom right panel). Dashed yellow line indicates the normal area of pancreatic islets; dashed red line reveals the infiltration of inflammatory leukocytes. Scale bar, 50 μm. (B) Intraperitoneal glucose tolerance testing (IPGTT). Spleen mononuclear cells (SMC) were isolated from 3-week-old NOD mice and then irradiated at the same dose (5000 rad) as those isolated from diabetic NOD mice; Consequently, the irradiated SMC were transplanted into humanized NOD-scid IL-2rγ$^{null}$ mice ($1 \times 10^7$ cells/mouse, i.p., $n = 5$ for each group). To test SDF-1/CXCR4 action, human PBMC were incubated with functional grade purified anti-human CXCR4 mAb (20 μg/ml, eBioscience) and then transplanted into NOD-scid IL-2rγ$^{null}$ mice ($n = 5$). After two weeks, mice were challenged with high glucose (2 mg/kg of body weight, i.p.) for IPGTT. Normal NOD-scid IL-2rγ$^{null}$ mice served as control (green line, $n = 5$). Data are shown as mean ± s.d. of mouse blood glucose levels. (C) Scoring of insulitis. Pancreatic islets were scored for% mononuclear cell infiltration after immunostaining for insulin and counter-staining with hematoxylin as described in methods. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

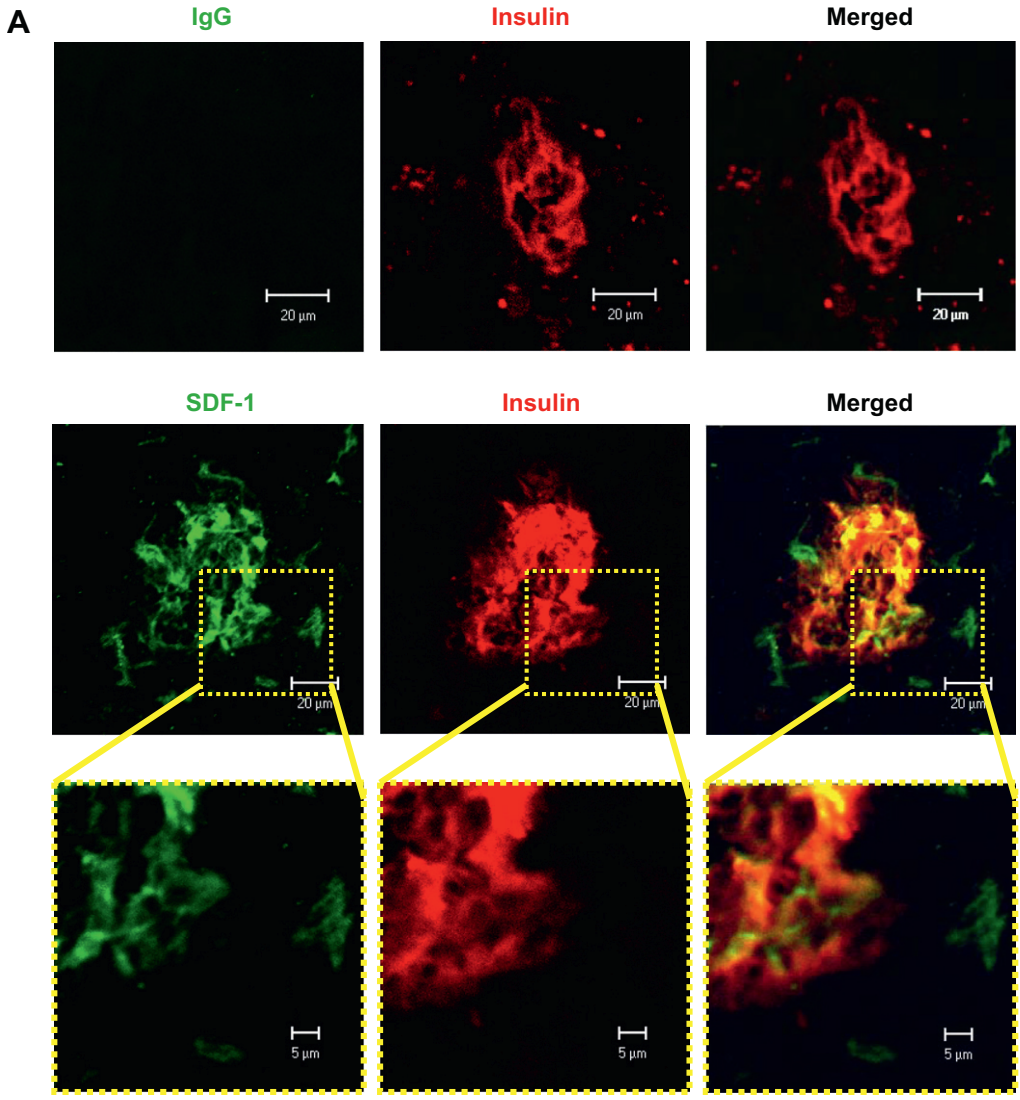


**Fig. 2.** Molecular mechanisms underlying the recruitment of human T cells. (A) Pancreatic islets display a chemokine SDF-1. Scale bar, 20 μm. High magnification, scale bar 5 μm. (B) Pancreatic islets fail to display monocyte chemoattractant protein-1 (MCP-1). (C) Expression of CD1d on mouse pancreatic islets. Isotype-matched IgG served as negative control. Scale bar 50 μm. (D) Migration of human Vα 24 Jα18$^+$ iNKT cells into pancreatic islets (scale bar 50 μm), with directly contacting between insulin-producing β cells (red) and iNKT cells (green) (scale bar 10 μm). Isotype-matched mouse $IgG_1$ served as a negative control. Representative images of those obtained from six experiments. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

express monocyte chemoattractant protein-1 (MCP-1) (Fig. 2B). SDF-1 is a highly conserved chemokine with 100% similarity between human and mouse SDF-1 protein sequences (FASTA Program, UniProt Knowledgebase, http://www.ebi.ac.uk/Tools/fasta33/index.html). To directly address the role of SDF-1/CXCR4, we performed *in vivo* blocking experiment using anti-human CXCR4 mAb to neutralize CXCR4 on human T cells. IPGTT results revealed that, as before, all mice ($n = 5$) in hPBMC + irrad. diabetic NOD mouse SMC group showed significantly impaired glucose tolerance testing and maintained hyperglycemia during the test period. However, in 5/5 of mice treated with hCXCR4 mAb-blocked hPBMC + irrad. diabetic NOD mouse SMC, glucose levels were markedly lowered after 40 min ($p = 0.023$) and similar to control mice; 3/5 of mice achieved euglycemia after 90 min, with average blood glucose level (143.2 ± 18.22 mg/dL) significantly lower than that in the hPBMC + irrad. diabetic NOD mouse SMC group (390 ± 37.8 mg/dL, $p = 0.012$) (Fig. 1B, blue line). Additionally, we compared body weight between two groups and found a marked difference (23.48 ± 0.15 g in hCXCR4 mAb blocking group vs 18.73 ± 1.41 g in non-blocking group, $p = 0.00054$). Thus, these data indicate that blocking CXCR4 on human T cells significantly decreases the diabetic induction. Pancreatic islet-derived chemokine SDF-1 and its receptor CXCR4 on human T cells [11] can be one of major mechanisms for homing to diabetic islets.

We found that mouse islets strongly displayed CD1d molecule (Fig. 2C) related to antigen presentation to NKT cells. Human NKT cells can recognize mouse CD1d due to the similarity of CD1d between mouse and human [12]. Immunostaining demonstrated that a large proportion of human iNKT cells migrated into pancreatic islets (Fig. 2D, top panels) and directly targeted on islet β cells (Fig. 2D, bottom panels). To determine molecular mechanisms underlying the destruction of islet β cells, pancreatic slides were immunostained with anti-human perforin (Fig. 3A) and Granzyme B (Fig. S4) mAbs. Results indicated that human perforin was strongly expressed in pancreatic islets that resulted in the β-cell death. Additionally, we examined another important cytotoxic cytokine interferon γ (IFNγ) produced by CD8$^+$ T cells and/or iNKT cells [13]. Triple immunostaining demonstrated that there was a

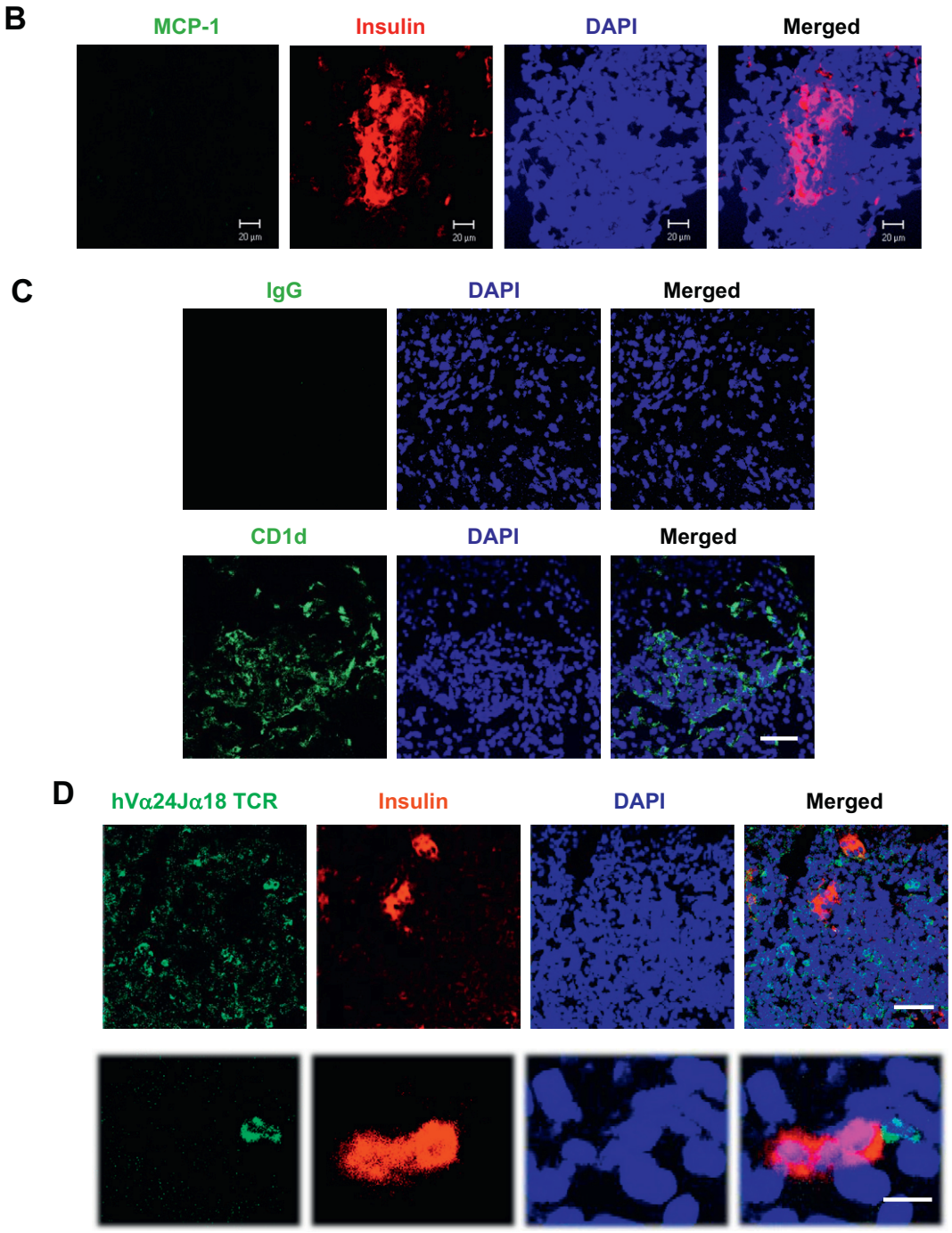


**Fig. 2** (*continued*)

strong expression of IFNγ that was mainly produced by iNKT cells in this model (Fig. 3B). Thus, iNKT cells became innate immune cells critical for the first line of killing β cells in this model.

To clarify when human T cells were recruited to pancreatic islets, humanized NOD-scid IL2rγ$^{null}$ mice were sacrificed for histological analysis at different time points (including days 1, 3, and 5) after administering the irradiated diabetic SMC. We found that a predominant population of human CD4$^+$ T cells had entered the pancreas and accumulated around islets on day 1 (Fig. 3C, bottom panels), and strongly expressed a cell proliferation nuclear marker Ki67 (Fig. 3D). However, there were a low percentage of human CD11b$^+$ MΦ and CD83$^+$ DC (or CD205$^+$ DC, data not shown) among the infiltrated leukocytes (<0.2%) (Fig. 3C, top panels for CD11b; Fig. S5 for CD83$^+$ DC). Thus, these data indicate that human CD4$^+$ T cells, not MΦ and DC, firstly entered pancreatic islets via SDF-1/CXCR4 mechanism after triggering with the irradiated diabetic SMC and play a key role in the initiation of humanized immune responses against islet β cells.

### *3.4. Disappearance of the irradiated NOD mouse SMC in humanized diabetic NOD-scid IL2rγ$^{null}$ mice*

To determine the cell fate of irradiated diabetic NOD mouse SMC, we initially assessed their cell viability in *vitro*. Flow analysis

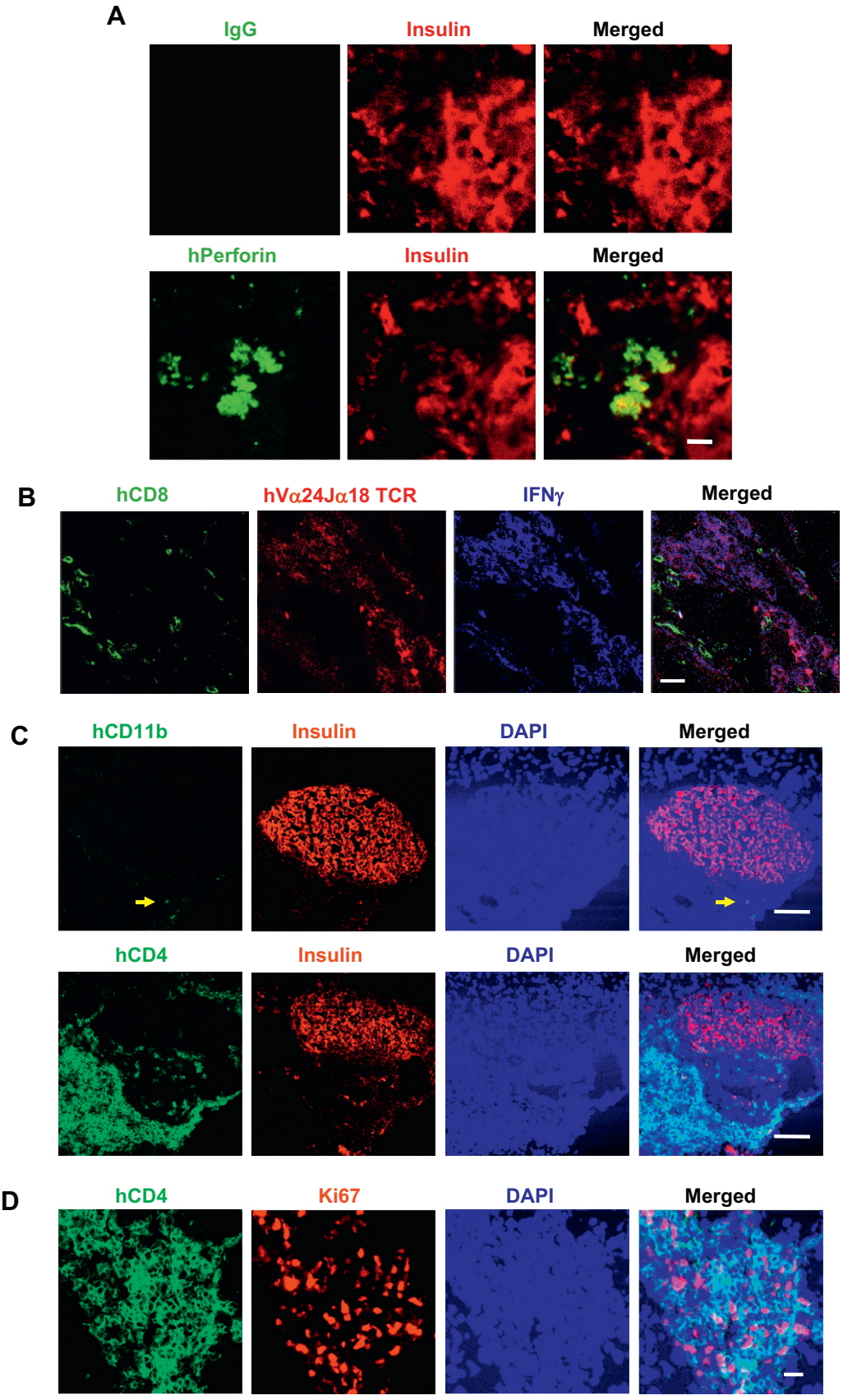


**Fig. 3.** Molecular mechanisms underlying the β-cell destruction. (A) Human T cell-released perforin in pancreatic islets, as demonstrated by immunostaining with anti-human perforin mAb (scale bar 10 μm). (B) Expression of human IFNγ (blue) in pancreatic islets by triple immunostaining for human $CD8^+$ T cells (green) and $V\alpha 24\ J\alpha 18^+$ iNKT cells (red) (scale bar 20 μm). (C and D) show the data from humanized NOD-scid IL-$2r\gamma^{null}$ mice after adoptively-transferred irradiated diabetic SMC for 1 day. (C) Migration of human $CD4^+$ T cells into pancreas and accumulation around pancreatic islets (bottom panels). A few $CD11b^+$ human MΦ present in a serial section (top panels). Scale bar, 50 μm. (D) Human $CD4^+$ T cells (green) display a cell proliferation nuclear marker Ki67 (red), scale bar, 20 μm. Isotype-matched mouse $IgG_1$ served as a negative control. Representative images of those obtained from six experiments. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

showed that 75% of freshly-irradiated SMC failed to express apoptosis marker Annexin V and cell necrosis marker propidium iodide (PI) (Fig. S6A, left panel), suggesting they were viable. However, 99% of cells died after in *vitro* culture for 24 h, as demonstrated by flow cytometry with 93% of cells positive for PI, 6% of cells double positive for Annexin V and PI (both apoptotic and necrotic cells at late stage), and 0.1% of cells positive for Annexin V (apoptotic cells) (Fig. S6A, right panel). However, phenotypic analysis revealed that both CD4 and CD8 T cells maintained their expression of chemokine SDF-1 receptor CXCR4 after irradiation (Fig. S6B). Using Chemotaxis Cell Migration Assay (3 μm pore size), we have tested the migration potential of the irradiated diabetic NOD mouse T cells in comparison with viable (non-irradiated) diabetic NOD mouse T cells in the presence of recombinant mouse chemokine SDF-1α (10 ng/ml). After overnight culture, microscopic analysis confirmed that the irradiated diabetic NOD mouse T cells can migrate through the membrane. Spectrometric quantification of migratory cells revealed a 3.7-fold reduction of migration potential for the irradiated diabetic NOD mouse T cells relative to that of the non-irradiated diabetic NOD mouse T cells.

Next, we explored the migration of irradiated mouse CD4 and CD8 T cells to pancreatic islets. Immunostaining results demonstrated that the infiltration of both mouse CD4 and CD8 T cells to pancreatic islets had occurred after being adoptively transferred into humanized NOD-scid IL2rγ$^{null}$ mice for one day (Fig. S6C and D). However, after 5 days, neither mouse CD4 nor CD8 T cells were detected among the infiltrated cells, even in the whole pancreatic sections (Fig. S6E and F), with only an occasional evidence of a small amount of cell debris distributed inside of the inflammatory islets displaying residual mouse CD4 and CD8 antigens (Fig. S6E). Thus, these data indicate that both mouse CD4 and CD8 T cells, as major players for adoptively transferring diabetes [14,15], can maintain their diabetogenic potential for a short period (<24 h) after high dose irradiation.

### 3.5. Metabolic abnormalities in humanized diabetic NOD-scid IL2rγ$^{null}$ mice

To determine the destruction of islet β cells, we subjected pancreata to histological analysis and evaluated total β-cell mass followed by immunostaining with insulin Ab on serial pancreatic sections. Morphometric analysis demonstrated that 90% of total β-cell mass was decreased in humanized diabetic NOD-scid IL2rγ$^{null}$ mice compared with normal non-diabetic NOD-scid IL2rγ$^{null}$ mice (Fig. 4A, $p = 0.0064$). With 10% of residual total β-cell mass, humanized diabetic NOD-scid IL2rγ$^{null}$ mice maintained euglycemia (92.3 ± 20.4 mg/dL) prior to high glucose challenge within the time period observed (45 days post administering diabetic NOD mouse SMC), similar to that of non-diabetic NOD-scid IL2rγ$^{null}$ mice (99.6 ± 17.9 mg/dL, $p = 0.42$). However, 24/25 of humanized diabetic NOD-scid IL2rγ$^{null}$ mice displayed an impaired intraperitoneal glucose tolerance test (IPGTT) and maintained high glucose levels (>400 mg/dL) without any observable down-regulation (Fig. 4B). Moreover, blood insulin levels were monitored, both prior to IPGTT and following IPGTT. Results revealed that insulin level was significantly decreased in the sera of humanized diabetic NOD-scid IL2rγ$^{null}$ mice prior to IPGTT compared with that of normal non-diabetic NOD-scid IL2rγ$^{null}$ mice ($p = 0.0055$). After glucose challenge, insulin level was markedly increased to a level that was about 2 times higher than before IPGTT in the sera of normal non-diabetic NOD-scid IL2rγ$^{null}$ mice ($p = 0.03$). In contrast, humanized diabetic NOD-scid IL2rγ$^{null}$

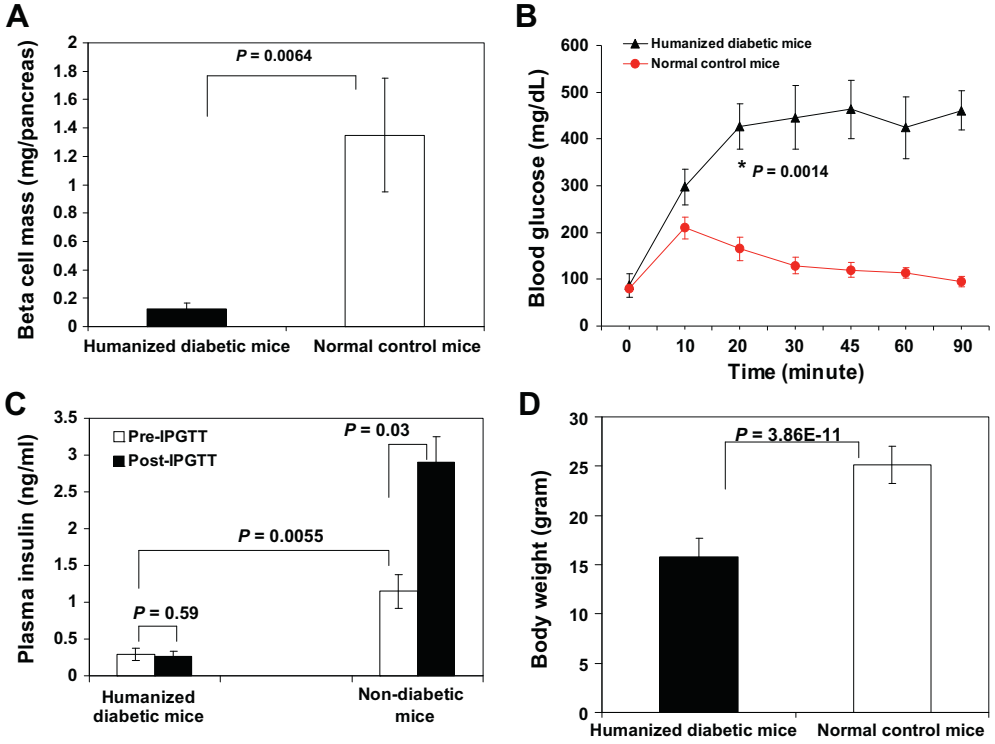


**Fig. 4.** Measurement of type 1 diabetic phenotypes in humanized diabetic NOD-scid IL-2rγ$^{null}$ mice. (A) Total β-cell mass. (B) Intraperitoneal glucose tolerance testing (IPGTT). Eight-week old NOD-scid IL-2rγ$^{null}$ mice ($n = 5$) served as normal controls for humanized diabetic NOD-scid IL-2rγ$^{null}$ mice ($n = 10$). (C) Determination of blood insulin levels by ELISA. Blood samples were collected from tail veins of humanized diabetic mice ($n = 10$) before glucose challenge. After glucose challenge for 30 min, blood was collected again. (D) Mouse body weight. Two weeks later following adoptive transfer with the irradiated SMC, body weight was tested in comparison with normal control.

mice failed to show any up-regulation after IPGTT (Fig. 4C, $p = 0.59$). These mice had to be sacrificed because of severe loss of body weight in comparison with normal non-diabetic NOD-scid IL2r$\gamma^{null}$ mice (>20%, Fig. 4D, $p = 3.86E{-}11$) according to the protocol approved by the Animal Care Committee.

To elucidate whether the destruction of pancreatic islets was β-cell specific, we performed double-immunostaining. Notably, we found that the insulin-producing β cells had disappeared from the pancreatic islets (42/56 of islets) of humanized diabetic NOD-scid IL2r$\gamma^{null}$ mice two weeks after transfer irradiated SMC (Fig. S7A, middle panel). In contrast, the glucagon-producing α cells were found distributed around islets or present in clusters after islet structures had been completely destroyed (Fig. S7A, middle panel). These results confirm that the β cells were selectively killed by humanized immune system without influencing the α cells, which are similar to those observed in diabetic NOD mouse model (Fig. S7A, bottom panel) and T1D patients (Fig. S7B, bottom panel). In addition, we assessed the infiltration of human leukocytes into other organs including the liver and lung. Histological examination demonstrated that mice transplanted with human PBMC + irradiated diabetic NOD SMC failed to display infiltration after 2 weeks of observation. This was similar to the un-transplanted control group (Fig. S8). Thus, these data indicate that destruction of pancreatic islet β cells caused by human T cell-mediated immune responses was specific in humanized diabetic NOD-scid IL2r$\gamma^{null}$ mice.

This model is well suited to explore the trafficking of human T lymphocytes toward islets and understanding the disease process that will help to develop novel interventional strategies (e.g., immunomodulators and trafficking/chemokine inhibitors) and translate into clinical care. The selective destruction of islet β cells by a human T cell-mediated immune response in this humanized T1D model can mimic the features observed in T1D patients. This model has overcome the limitations of current rodent T1D models and will facilitate translational research to find a cure for T1D.

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## Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bbrc.2010.07.128.

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