

## Roles of Toll-like Receptors on Islets in Allogeneic Islet Transplantation

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**Purpose:** Toll-like receptors (TLRs) are involved in rejection of organ allografts. We investigated the roles of toll-like receptors on pancreatic islet cells in allogeneic islet transplantation together with roles of toll-like receptors on the recipient side.

**Methods:** After islet cells were stimulated by polyinosinic:polycytidylic acid (poly I:C) and lipopolysaccharide (LPS), expression of chemokines, cytokines, and procoagulants in islet cells were measured using reverse transcriptase polymerase chain reaction (RT-PCR). In order to assess roles of TLRs on recipient or donor islets, allogeneic islet transplantation models were used using MyD88, TLR4, or Trif knockout (KO) mice with or without anti-CD154 antibodies (MR-1). Next, LPS (100 mcg i.p.; D1,3,5,7,9) was administered to simulate TLRs in vivo with or without MR-1. Recipients were rendered diabetic using streptozotocin (STZ, 180–200 mg/Kg, i.p.). Islets (400 IEQ) were transplanted under the kidney capsule of the recipients. Blood glucose levels were monitored to assess islet graft survival.

**Results:** Murine islet cells expressed TLR3 and TLR4 at resting status. Various chemokines (RANTES, IP-10, MCP-1) and tissue factor were upregulated in islets in response to TLR ligands, especially PolyI:C. iNOS upregulated higher by LPS than PolyI:C. These inductions by TLR activation were abrogated in experiments using TLR4, MyD88, Trif, and IPS-1 KO mice. When islets from MyD88 KO, Trif KO, or TLR4 KO C57BL6/J donor mice were transplanted to diabetic Balb/C mice without MR-1, their islet allograft survivals were not better than the wild type group. There was also no significant difference in islet allograft survival between MyD88 KO, Trif KO, and wild type recipients without MR-1, although some of MyD88 KO recipients obtained long-term allograft survival. When MR-1 0.25mg was added on transplantation day 0, MyD88 deficiency of recipients prolonged islet allograft survival (Log-rank test,  $p=0.015$ ). However, MyD88 deficiency of donors did not influence islet allograft survival with MR-1. TLR4 stimulation on donor showed shorter islet allograft survival (Log-rank test,  $p=0.003$ ).

**Conclusion:** TLRs on both recipient and donor can contribute to islet allograft rejection.

**Key Words:** Toll-like receptor, Islet transplantation