Revascularization of Transplanted Islets
Can It Be Improved?

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Pancreatic islets are highly vascularized, which is important in their ability to quickly secrete insulin in response to changes in blood glucose. Although pancreatic islets comprise only 1–2% of pancreatic mass, they receive 5–10% of pancreatic blood flow. Blood vessels within pancreatic islets are of a greater density than those in surrounding exocrine tissue and are lined with fenestrated endothelial cells. These specialized features are responsible for the greater partial pressure of oxygen in islets compared with acinar tissue and other organs, which is likely important for normal islet cell function. Islet production of angiogenic factors such as vascular endothelial growth factor-A (VEGF-A) and angiopoietin-1 is critical for creating this highly vascularized state (1,2). During embryonic development, reciprocal endothelial-endocrine cell signaling and the formation of functional blood vessels appear to instruct pancreatic differentiation and morphogenesis (3–5). Development of the islet vasculature is coordinated with islet formation, but blood flow to endocrine cells precedes their final assembly into a mature islet (2).

Pancreatic islet isolation severs the connections between the islet vasculature and the systemic circulation. In contrast with whole-organ transplantation, where organ perfusion is quickly reestablished by reconnection of arterial and venous vessels, the reestablishment of blood flow to transplanted islets requires several days and involves angiogenesis and possibly vasculogenesis. Not only are islets avascular for several days following transplantation, they are less vascularized and have a lower oxygen tension than islets in the pancreas when revascularization is complete (6,7). The death of significant numbers of islets in the days following transplantation results from several factors, but ischemia and inadequate blood supply are likely contributors to islet death in the immediate posttransplant period and may impair islet survival and function long term. Thus, improvements in the revascularization of transplanted islets may enhance islet survival and the outcomes of islet transplantation.

Efforts to improve the revascularization of transplanted islets are hindered because the responsible ligands, receptors, cells, and mechanisms are not well defined. Recent evidence indicates that the endothelial cells creating new capillaries or vessels within the islet graft arise from three sources (Fig. 1). The first source is the endothelial cells from the transplant recipient, which are recruited into the islet graft. A second source is intraislet endothelial cells, which exist in large numbers in isolated islets and may account for up to 40% of the endothelial cells lining capillaries within a revascularized graft (8,9). Interestingly, functional vessels within a revascularized graft are often chimeric, consisting of both endothelial cells from the transplant recipient and donor-derived, intraislet endothelial cells. Bone marrow–derived cells are a third, but likely minor, source of endothelial cells (10,11). The factors produced by transplanted islet cells that stimulate or recruit endothelial cells from the three potential sources in the graft include VEGF-A (2), but other pro- or antiangiogenic molecules could also play a role (Fig. 1). The formation of new vessels also requires vascular remodeling involving the basement membrane, vascular supporting cells such as pericytes, and the extracellular matrix. Little is known about these processes in islet revascularization.

The revascularization of transplanted islets might be enhanced or accelerated by several types of interventions. One approach would be to increase the action of proangiogenic factors or to inhibit antiangiogenic factors and thus stimulate the proliferation, migration, and maturation of endothelial cells into functional vessels. This approach has had some hints of success (12–15), but it is likely that the optimal formation of mature, fully functional islet vasculature will require precise control of the timing, dose, and duration of angiogenic factor action in the posttransplant period. A second approach could directly target endothelial cells or enhance their ability to form mature, functional vessels and might involve the addition of preactivated endothelial cells or some type of endothelial progenitor cell population. These two approaches should be applicable to isolated islets before transplantation or, also, could be used to prepare the transplantation site before transplantation of isolated islets.

In this issue of *Diabetes*, Johansson et al. (16) propose a new approach using tissue engineering to enhance islet revascularization. These investigators provide evidence that the coculture of mesenchymal stem cells (MSCs) and endothelial cells with human islets in vitro before transplantation initiates formation of vessel-like structures that may promote islet engraftment after transplantation. MSCs, multipotent cells usually isolated from bone marrow but also present in other tissues, exhibit a wide range of properties in other settings, properties that might enhance islet survival (17–19). For example, MSCs positively modulate inflammation, tissue regeneration, and

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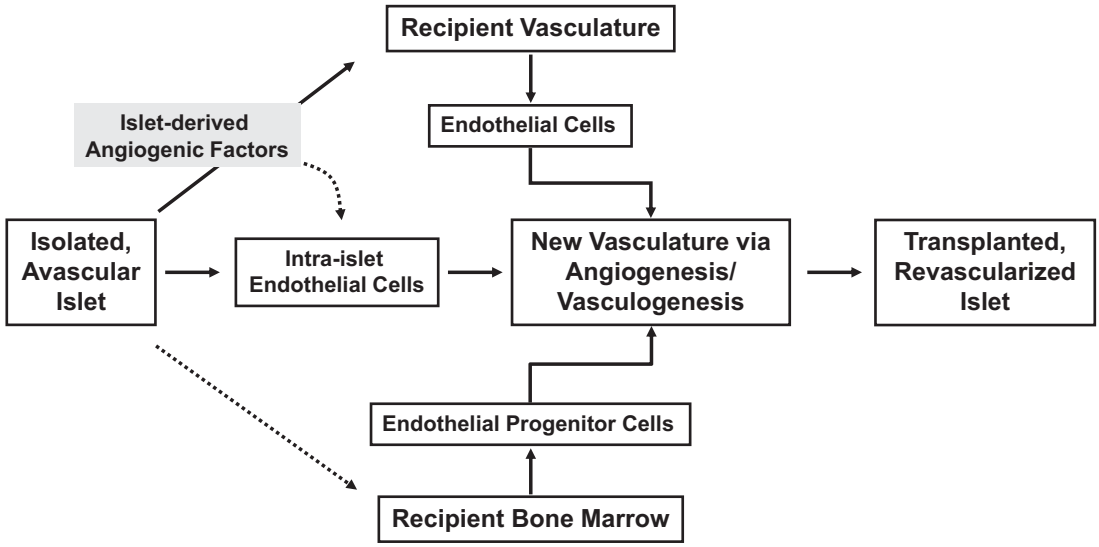

FIG. 1. Model of islet revascularization after transplantation. Endothelial cells from three sources (recipient endothelial cells at the site of transplant, intra-islet endothelial cells, and endothelial progenitor cells from the bone marrow) contribute to revascularization. The dotted line refers to processes or factors not yet defined.

immune attack either through cell-to-cell contact, differentiation into other cell types, or by the local production of factors such as platelet-derived growth factor. Johansson et al. purified MSCs from normal human bone marrow using cell-surface markers and found that MSCs or factors produced by these cells promoted endothelial cell proliferation and migration and the "coating" of cultured islets with endothelial cells (16). Using an in vitro system to study angiogenesis, these investigators demonstrated that this mixture of MSCs, endothelial cells, and islets promoted the migration of exogenous endothelial cells into the cultured islets; the formation of chimeric, vessel-like structures between the endogenous intra-islet endothelial cells and the endothelial cells added to the islet culture; and the formation of new vessel "sprouts" from islets. A unique and possibly critical component in these studies was the microvascular endothelial cells harvested from human dermis, which are likely more receptive to remodeling signals from the MSCs. Such MSCs and microvascular endothelial cells could likely be harvested and expanded from the bone marrow or adipose tissue of humans selected to receive an islet transplant.

So, how did MSCs promote these changes in endothelial cells and promote the formation of new intraislet vascular-like structures, and how might this be translated to islet transplantation? Additional work is needed to define the ligands, receptors, and mechanisms responsible for these effects, but Johansson et al. speculate that proteases from MSCs may degrade the islet extracellular matrix and thus allow the migration of endothelial cells that have been stimulated by growth factors such as VEGF-A produced by MSCs. Identification of these factors should allow one to test whether addition of these factors to cultured islets could substitute for the MSCs. As one considers how to extend these in vitro findings, demonstration that this coculture approach improves the function and survival of transplanted islets using in vivo models is the critical next step. Likewise, the long-term fate and growth potential of MSCs must be determined and may be dependent on the transplantation site microenviroment. Hopefully, as we better understand the molecular events of islet revascularization, delivery of angiogenic factors at the optimal dose, time, and duration combined with tissue engineering approaches such as that described by Johansson et al. will accelerate and enhance the revascularization of transplanted islets and lead to improved islet function and survival.

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