Advanced Drug Delivery Reviews 67-68 (2014) 93–110

Contents lists available at ScienceDirect

Advanced Drug Delivery Reviews

journal homepage: www.elsevier.com/locate/addr

Oxygen supply to encapsulated therapeutic cells ☆

Clark K. Colton

Department of Chemical Engineering, Massachusetts Institute of Technology, 77 Massachusetts Avenue, Cambridge, MA 02139, USA

ARTICLE INFO

Available online 27 February 2014

Keywords:
Encapsulation
Oxygen supply
Immunobarrier
Immunoisolation
Hypoxia
Implanted devices
Cell transplantation

ABSTRACT

Therapeutic cells encapsulated in immunobarrier devices have promise for treatment of a variety of human diseases without immunosuppression. The absence of sufficient oxygen supply to maintain viability and function of encapsulated tissue has been the most critical impediment to progress. Within the framework of oxygen supply limitations, we review the major issues related to development of these devices, primarily in the context of encapsulated islets of Langerhans for treating diabetes, including device designs and materials, supply of tissue, protection from immune rejection, and maintenance of cell viability and function. We describe various defensive measures investigated to enhance survival of transplanted tissue, and we review the diverse approaches to enhancement of oxygen transport to encapsulated tissue, including manipulation of diffusion distances and oxygen permeability of materials, induction of neovascularization with angiogenic factors and vascularizing membranes, and methods for increasing the oxygen concentration adjacent to encapsulated tissue so as to exceed that in the microvasculature. Recent developments, particularly in this latter area, suggest that the field is ready for clinical trials of encapsulated therapeutic cells to treat diabetes.

Contents

1. Introduction

Tissue-engineered immunoisolation devices, which provide a semi-permeable barrier to protect transplanted cells from the recipient's

☆ This review is part of the *Advanced Drug Delivery Reviews* theme issue on "Cell encapsulation and drug delivery".

E-mail address: ckcolton@mit.edu.

http://dx.doi.org/10.1016/j.addr.2014.02.007
0169-409X/

immune system without immunosuppressive drugs, have the potential to treat a large range of diseases, including diabetes [1], hemophilia [2], anemia [3], parathyroid disease [4], chronic pain [5], Parkinson's disease [6], Huntington's disease [7], and amyotrophic lateral sclerosis [8]. The therapeutic cells constantly produce a therapeutic protein required for the treatment of most of the diseases listed above. For diabetes treatment, the cells secrete insulin in response to changes in the blood glucose concentration in a feedback-controlled manner.

Cells can be encapsulated at a high, tissue like density or dispersed in an extracellular gel matrix, such as agar, alginate, chitosan, or other hydrogels. The immunobarrier ideally prevents access of immune cells and of the larger humoral immune components but permits passage of smaller secreted products such as insulin. There must be sufficient access to oxygen and nutrients such as glucose and removal of secreted metabolic waste products, such as lactic acid, carbon dioxide, hydrogen ions, and nitrogenous products of metabolism. Encapsulated cells must be supplied with oxygen and nutrients by diffusion from the nearest blood supply, through surrounding tissue, the immunobarrier membrane, and the graft tissue itself.

The issue of oxygen supply to encapsulated therapeutic cells is critically important to their viability and function. Because this issue intersects with all areas of design and performance of devices, we begin this review with a brief discussion of major issues related to the performance of immunobarrier devices, especially as they relate to oxygen supply limitations: (1) device design and materials, (2) supply of tissue, (3) protection from immune rejection, and (4) maintenance of cell viability and function. Most of the literature cited in this review deals with immunobarrier devices used in transplanting islets of Langerhans for type 1 diabetes treatment. The concepts and methods described also apply to other cell therapy applications, especially those involving metabolically active cells. After discussing the major issues, we focus on (1) the existence of oxygen limitations in islet transplantation and its deleterious effect on islet viability and function, and (2) a comprehensive discussion of diverse approaches investigated with encapsulated cell therapy in order to overcome these oxygen limitations. We conclude with the author's perspective on what we have learned about oxygen supply limitations and the challenges that remain for the future. The material presented here builds on previous reviews of the field by the author [9–15] and others [16–23] and highlights recently reported work.

2. Issues in encapsulated cell therapies

2.1. Device designs and materials

Therapeutic cells have been encapsulated in intravascular devices and in extravascular implants.

2.1.1. Intravascular devices

Blood flows from artery to vein through the lumen of a tube encased in a chamber. The tube is an immunoisolating membrane, on the outside of which cells or tissues are cultured, thereby maximizing the effectiveness of oxygen, nutrient, waste, and therapeutic protein transport because diffusion distances are relatively short. The presence of blood flow in close proximity to tissue provides a good arrangement for oxygen supply from the bloodstream. On the other hand, this device is an arteriovenous shunt that disrupts the patient's vascular system, thereby leading to a greater risk of complications such as thrombosis.

Work with intravascular devices began more than four decades ago by Dr. William Chick together with a multidisciplinary team, culminating in the first paper that demonstrated normalization of a disease state (hyperglycemia) with a tissue-engineered device containing encapsulated therapeutic cells [24]. In that study, islets were cultured outside of very small diameter hollow fibers through which anticoagulated blood flowed in an ex vivo arrangement. Subsequently, a single large-diameter tube was used to eliminate the need for systemic anticoagulation.

Additional papers using this approach have been described in an earlier review [10]. In one study [25] in which two devices were implanted in each of 10 diabetic dogs, exogenous insulin was completely supplanted in six animals. Even with the favorable oxygen transport conditions arising from blood flow through the tube, each device contained more than 50% of nonviable islets when examined after explantation. In these experiments, islet surface density was about 2670 islets/cm^2 tubular surface area and dose was 19,400 islets/kg body weight, which was nearly four times the 5000 islets/kg required for satisfactory blood glucose control in a canine autotransplant [26]. Because canine islets have an average diameter of 122 μm compared to 150 μm for an islet equivalent (IE, volume 1.77×10^{-6} cm^3), the volume of canine islets is reduced by a factor of 1.86 compared to an IE. When put on the same volumetric basis of a human IE, the surface density and dose were about 1440 IE/cm^2 and 10,400 IE/kg, respectively.

Work on the intravascular approach progressed successfully to the point where more than 360 successful implantations in diabetic dogs had been carried out, and a human clinical trial of a tubular intravascular device for islet transplantation was being planned. The planning was suspended by the FDA because of a mechanical failure of a cannula in one dog. By the time the cannula was redesigned, resources of the small company that championed this approach were depleted, and the work has never been resumed. This type of device is not currently under study. Surprisingly, designs involving blood flow through hollow fibers [27], akin to the original concept [24], and blood flow through narrow channels [28] have been proposed in the past decade without recognition of their clinical impracticality for chronic use because of the need for chronic systemic anticoagulation.

2.1.2. Extravascular implants

Extravascular implants differ in geometrical characteristics and also in the way the effects of oxygen supply limitations are manifested. Diffusion in a spherical geometry is privileged because, at any radial position away from the center of a sphere, the available surface area for diffusion relative to the volume of tissue contained within that radius is the highest; conversely, the surface to volume ratio is the lowest for the slab. In addition, the presence of an external layer, such as fibrotic tissue arising from the foreign body response, through which diffusion must also occur decreases the available driving force for diffusion within the tissue. This decrease has the smallest relative effect for a spherical geometry, whereas the effect is largest for a planner slab. The mass transfer resistance of a layer outside a sphere reaches a maximum asymptotic value as the thickness of the layer increases. Conversely, as the region outside of a slab thickens, its mass transfer resistance grows without bound. For these reasons, the mass transfer effectiveness of each geometry decreases in the order sphere > cylinder > planar slab.

2.1.2.1. Microcapsules. Beginning with the work of Lin and Sun with islets microencapsulated in alginate [29], small spherical hydrogel beads containing usually 1 or 2 islets and ranging in diameter from 200 μm for thin coatings to 1000 μm or more have become the most extensively studied approach for encapsulated islet transplantation to treat diabetes. The larger beads [30–33], sometimes called macrocapsules or macrobeads, have received less attention. The peritoneal cavity is the most common implantation site because there is ample space for the capsules, and, if the capsules are able to float freely, immune responses are less aggressive than at other implantation sites, such as the subcutaneous space where immune cells can more easily attach to the surface. Microcapsules may aggregate and may be located far from the blood supply; under these conditions, the islets have very limited oxygen supply, which further degrades tissue survival. The feasibility of intraportal injection (used for naked human islet transplantation) has been examined to enhance microcapsule proximity to the blood supply, but there is an increased immune response, which requires short-term immunosuppression [34].

The most common material for microcapsules is still alginate, which forms a viscous solution at low concentration in water and gels in the presence of a divalent (e.g., calcium or barium) or trivalent (e.g., gadolinium) ion without causing damage to cells. Because it is a naturally derived product, its properties are batch and source dependent, and impurities can have detrimental effects. Most typical is alginate cross-linked with barium ions [1,35] or alginate cross-linked with calcium and then used uncoated [36] or coated with poly-L-lysine [37,38] or poly-L-ornithine [39,40] to form a permselective barrier and enhance capsule stability. Because the water content is very high, the diffusivity of small molecules such as oxygen in alginate is only modestly reduced from its value in water. However there is a surprisingly large effect of decreasing protein diffusivity (e.g. albumin) with increasing alginate concentration [41]. Alternate materials investigated include synthetic Tetronic polymers that thermally gel and chemically cross-link to form more stable gels while still maintaining gentle processing steps [42]. The capsule formation process for the Tetronic polymers is adaptable to the machinery developed for making alginate capsules [43]. Reviews are available for cell and islet microencapsulation [43,44] and materials used for islet encapsulation [45,46].

Experiments in primates and exploratory clinical trials in diabetic patients with microencapsulated islet allografts and xenografts [47–52] have yielded a variety of mixed results. In only one study with polylysine-coated microcapsules [47] did all subjects become normoglycemic for many months, and that striking work was never repeated. In all of these studies, very large doses of islets (20,000 to 40,000/kg) were used, which is far in excess of the dose actually needed and implies a very large loss of viability and function of the implanted islets. It is impossible to retrieve all of the microcapsules following implantation. This is a major limitation that has, in part, driven interest in macroencapsulation devices that can be retrieved.

2.1.2.2. Macroencapsulation devices. Macroencapsulation devices are usually in the form of a planar diffusion chamber (or slab) or a tubular hollow fiber membrane in which tissue is contained, often encased in a hydrogel encapsulant. An important feature of these devices is the ability to be retrieved because of their macroscopic dimensions. Devices of this type are often implanted subcutaneously or within a physiological space or cavity of the body. They contain tissue alone or tissue embedded in a hydrogel within the device interior surrounded by an immunoisolating membrane. Delivery of oxygen and nutrients requires diffusion from the surrounding tissue to the device, across the immunobarrier membrane, and then through the interior of the device itself to the tissue. The amount of tissue that can be included within the device is constrained by oxygen supply limitations in the interior of the device. These limitations are further aggravated by overgrowth of fibrotic tissue, which adds another transport barrier and which also consumes some of the oxygen (if there are cells present) that would normally be delivered to the transplanted tissue [11,12]. Macroencapsulation devices are usually made of materials that are stable after implantation in the body and can be retrieved.

Macroencapsulation devices have been studied for several applications, including diabetes [53] and diseases of the central nervous system [54]. The first macroencapsulation devices were wide bore (internal diameter 0.5 to 5 mm) hollow fibers prepared from polyacrylonitrile-co-vinyl chloride [55–59] and polyethersulfone [60].

These hollow fiber devices received intensive investigation and use in small animal in vivo studies, and they achieved some success when a low tissue density was used, usually with a very high islet dose, but suffered large viability losses with high tissue density. A critical flaw with hollow fibers became apparent in the mid 1990s [61] for applications that require a large amount of tissue, such as diabetes. Oxygen diffusion limitations dictate a maximum diameter that can be filled with respiring cells without development of central necrosis [62], which is in the range of several hundred μm or more depending upon tissue density. Because of this constraint and the large tissue mass required in islet transplantation, the large length of hollow fiber needed to provide sufficient volume becomes impractical for implantation. This conclusion is illustrated by an example: Consider a hollow fiber filled completely with islet tissue, which, because of oxygen diffusion requirements, has a maximum internal diameter of, say, 200 μm, an optimistic estimate [10–12]. Encapsulation of 250,000 IE would require a length of about 1700 cm. Implantation of such a long fiber in a convenient location with provision for suitable oxygen supply and prevention of fibrous overgrowth would be a daunting challenge. Even more pessimistic estimates have been reported [63]. This constraint, in turn, limits the use of hollow fibers to applications requiring small numbers of cells such as neurological diseases [8]. As a consequence, recent efforts with macroencapsulation devices are limited primarily to slab configurations.

Planar slab devices began to appear in the late 1990s and can be categorized in terms of their mechanical rigidity. The most flexible is the so-called islet sheet composed of a layer of islets embedded in a thin (250 μm) sheet of alginate [64,65]. Elegant in its simplicity and its ability to be used as a patch, it has remained largely conceptual. A related approach is the thin monolayer cellular device [66,67] that was also developed to improve oxygen supply, the construction of which is best discerned from a patent [68]. It consists of sequential layers of human acellular collagen matrix, islets, a polyester filter, and high mannuronic acid alginate, 3% (w/v). Although correction of diabetes in primates for up to 6 months was reported with a dose of 15,000 IE/kg, subsequent studies [69,70] verified that oxygenation of tissue within the device was insufficient to maintain islet viability and function without some form of oxygen supply enhancement. A different approach with a flexible device is use of living cells as an immunoisolation membrane. In one study, a multilayer structure of alternating sheets of chondrocytes and islets was fabricated and maintained in vitro for 3 months but with a declining rate of insulin secretion [71].

The discovery at Baxter Healthcare Corp. of polytetrafluoroethylene (PTFE) microporous membranes that induce neovascularization [72] led to incorporation of these materials as part of the outer envelope of structures in which tissue was placed without the use of an encapsulant. These planar diffusion chambers were subsequently marketed by Theracyte Inc. They are less flexible than the alginate slabs, but the outer envelope is deformable so that the device can accommodate primary tissue as well as proliferating cells. Devices of this type have been used with genetically engineered human fibroblasts [73], insulinomas [74], parathyroid tissue [75,76], and rat islets [77,78]. In one study with a 4.5 μl Theracyte device, 1000 rat islets in diabetic athymic mice [40,000 IE/kg, 900 IE/cm^2 surface area (one side)] normalized blood glucose, but 500 islets did not. The device can be prevascularized by implantation prior to islet transplantation [79], which lowers the dose of islets required closer to that of naked transplantation [80].

Rigid planar slab devices have been fabricated using alumina [81] and nanoporous micromachined silicon-based membranes [53], which can result in more stringent control over the exact pore size of the immunobarrier. An interesting rigid composite structure has been described [63,82,83] composed of a precisely perforated metal scaffold coated with a reinforced amphiphilic co-network immunobarrier. These sophisticated designs do not address the issue of oxygen supply limitations.

Another type of rigid planar diffusion chamber arose from the notion that supply of exogenous oxygen at increased concentration directly to the tissue can maintain viability and function of transplanted islets. This concept was explored in a series of papers that made use of oxygen generation by electrochemical decomposition of water [13,84], algae photosynthesis [85,86], and direct injection of air or oxygen-enriched gas [87–92] using devices produced by Beta-O_2 Technologies. These studies demonstrated feasibility of allografts in rats, xenografts in pigs, and allografts in a human with modest islet doses and with islet densities that can be increased to high levels by appropriate modification of

exogenous oxygen concentration. These studies are discussed in more detail in the subsequent section on elevated oxygen concentration adjacent to tissue.

2.2. Supply of tissue

There are four types of sources: (1) primary human tissue, (2) primary xenogeneic tissue, (3) cell lines, and (4) differentiated pluripotent stem cells. Human tissue is desirable because, as described in the next section, it should be easier to provide immunoprotection to allografts as compared to xenografts. The main drawback of primary human tissue is the limited supply of cadaveric donors. In addition, procedures used to isolate primary islet tissue cause substantial damage and loss of viable tissue. Xenogeneic tissue has the advantage that its supply can be made more plentiful. However, there is a risk of retroviral disease transmission, immunobarrier requirements are thought to be more stringent, and damaging tissue isolation procedures are still required. As described in the next section, the presence of dying tissue in xenografts is even more harmful than in an allograft.

Islet transplantation in humans requires 10,000 IEQ/kg body weight (about 700,000 islets for a 70 kg patient), usually from two or three cadaveric donors in order to achieve insulin independence [93]. In contrast, about 10–20% [94,95] of the roughly one million endogenous islets [96] are needed to maintain normal blood glucose concentration in a normal non-diabetic person. This large disparity is caused by islet losses at each stage of processing and use. A substantial fraction of islets are damaged during isolation and die prior to transplantation [97]. Only about two-thirds of the islet dose survives the implantation and early engraftment period in mice [98], and an even smaller fraction survives clinical islet transplantation [99,100]. The hypoxic conditions to which islets are exposed after implantation in the liver, the currently-employed transplantation site, is thought to be a major contributor to this loss of tissue [101]. Similarly, oxygen supply limitations to encapsulated islets would further exacerbate this problem. If oxygen supply to encapsulated tissue could be sufficiently enhanced, it is conceivable that the loss of viable tissue could be minimized so as to be substantially smaller than in naked islet transplantation.

Cell lines could provide indefinite supply of a particular cell type, and there are no isolation requirements. When immunobarrier devices are used to deliver a desired product at a continuous rate, as is typically desired for gene therapy strategies, the use of cell lines has proven to be an adequate cell source [54]. The main drawback for diabetes treatment is that the cell line must secrete insulin under the same feedback-control mechanisms as the islet, but this type of cell line has yet to be developed, although efforts have been made in this direction [102,103].

Stem cells also offer great promise as an unlimited source of cells for immunobarrier devices, but understanding and control of the differentiation process to a greater extent than now available is required in order to generate cells with the desired phenotype for a particular application. Current protocols for differentiation of pluripotent (including embryonic) stem cells to beta cells follow a five-stage procedure that recapitulates embryonic stages of development [104]. Only the first four stages have been carried out successfully in vitro. The fifth stage, which involves maturation to glucose-responsive insulin-secreting beta cells and other islet cells, can only be carried out by implantation in vivo with current methods.

Viacyte Inc., a pioneer in developing a β-cell differentiation protocol, has successfully used the Theracyte device in order to carry out in vivo maturation without allograft rejection in immunocompetent mice made diabetic chemically [105]. As described on its website, Viacyte has also developed a device of its own design that functions in a manner similar to the Theracyte. The Theracyte device has been used without prevascularization in immunodeficient diabetic mice with human islets (216 IE/kg) and human β-cell precursors (fetal pancreatic islet-like cell clusters) [106]. The human adult islets exhibited poor survival and function, whereas the human fetal β-cells matured and ameliorated diabetes. The young islets underwent significant cell death immediately post transplant, followed by recovery of cell mass, which suggests an important role for replication in their performance. The same phenomenon would also benefit differentiated stem cells encapsulated in the Theracyte device.

2.3. Protection from immune rejection

Immune rejection is not normally associated with oxygen supply limitations. However, prevention of cell death by providing sufficient oxygen supply to encapsulated islets may provide an important benefit to immunobarrier protection so that true immunoisolation of encapsulated allogeneic or xenogeneic islets can be attained, thereby eliminating the need for immunosuppression [10].

Allograft rejection is mediated primarily by the direct pathway, which requires activation of the cellular immune response by direct contact between donor-derived antigen-presenting cells (APCs) and host-derived T cells. Direct cell contact is prevented by the use of an intact immunobarrier, such as a microporous membrane or encapsulation in a hydrogel. This may be the only requirement for immunoisolation of allogeneic tissue, at least for periods up to several months [107].

Even with all cell contact prevented, the indirect pathway can mediate rejection of xenografts and may also be operative with allografts [108]. In the indirect pathway, host APCs take up graft proteins and peptides released from the encapsulated cells and present donor-derived peptides on host MHC molecules to host T-cells, thereby initiating the immune response. The extracellular domain of many cell surface proteins is normally released by proteolysis to produce immunogenic shed donor antigen [109,110], some of which may be small enough to diffuse across an immunobarrier material. The rate of antigen generation is sharply increased when peptides are released from the interior of dying cells [111–113], a phenomena that may be involved in initiating autoimmune type 1 diabetes [114–116].

It has been hypothesized [117] that antigens shed from only dead or dying cells are important in immune recognition. The presence of dying cells in encapsulated donor islets is likely to generate a large immunogenic stimulus, especially with xenografts, that triggers the indirect pathway in the host, sensitizes the host to donor antigens, and ultimately leads to attack by components of the humoral immune response that should be kept out of the donor tissue compartment. These components include large molecules such as C1q and newly formed antibodies to immunogenic antigens that have leaked across the barrier. In addition, activated immune cells attracted by the antigen leaking from the dying tissue can form a florid response around the implant that consumes oxygen, further lowering the local pO_2. These cells also release small free radicals and cytokines and lymphokines (e.g., interleukin-1), which are relatively small proteins that can permeate across the immunobarrier and can have deleterious effects on β cells. The possible routes by which the indirect pathway of immune rejection can function with encapsulated cells are illustrated in Fig. 1.

An immunobarrier that must allow permeation of insulin, a small protein, will not have selectivity to prevent passage of the low molecular weight cytokines. The only way this scenario can be prevented is to stop the enhanced release of antigens that accompanies dying cells, which, in turn, requires sufficient oxygen supply in order to maintain cell viability. By maintaining the viability of virtually all islets encapsulated in the device, development of this immunogenic stimulus to the indirect pathway is prevented, and immunoisolation can be attained.

Experimental studies with encapsulated islet xenografts are consistent with the scenario described above. Planar slab devices that used microporous membranes to prevent cell contact but permit relatively unhindered diffusion of dissolved molecules prevented allograft rejection [75,78,118] and autoimmune rejection in the rat [119]. Conversely, xenograft rejection was not prevented in any study with these devices [118,120–124] and was invariably attributed to antigen shedding. The same mechanism of antigen shedding was suggested with uncoated

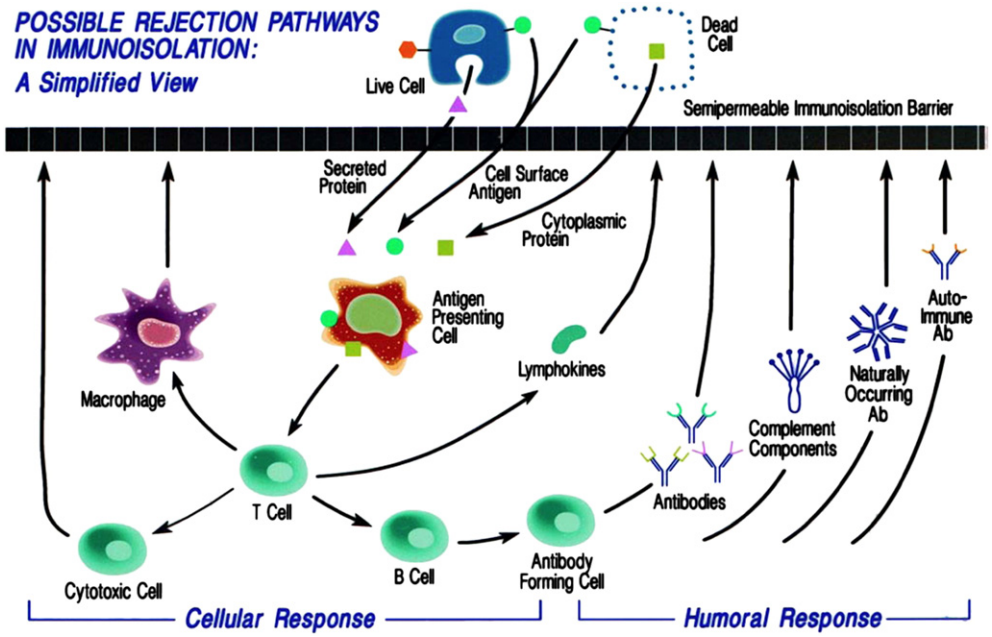

Fig. 1. Possible immune rejection pathways in the presence of a semipermeable immunobarrier membrane that prevents cell–cell contact. The release of antigenic polypeptides from dead or dying cells that permeate across the membrane can markedly stimulate the immune response.
Reproduced, with permission, from [15].

but not coated hydrogel microencapsules [36,123,125,126], for which rejection was also attributed to incomplete encapsulation of parts of the islet graft [123,127], and with hollow fiber diffusion chambers [59]. Studies with uncoated alginate planar slabs have also demonstrated immune response to xenografts [67,69,128–131] although the role of shed antigen in these studies was not recognized. In all of these studies, the extent of xenograft immune reaction should depend on the shed antigen contributions from live and dead or dying cells and the immunobarrier diffusive permeability to these polypeptides, none of which is known in these studies. Islet xenograft survival and function without immunosuppression was reported for a tubular perfusion device in dogs [132], polylysine-coated alginate microcapsules in monkeys [47], and a planar diffusion chamber with exogenous oxygen in pigs [89]. Success in the tubular perfusion device may result, in part, from shear forces associated with blood flow, which may prevent adherence of activated immune cells to the immunobarrier surface. Experience with the planar diffusion chamber using exogenous oxygen is discussed subsequently.

2.4. Maintenance of cell viability and function

Maintenance of cell viability and function is essential and is limited by the supply of nutrients and oxygen. Diffusion limitations of oxygen in tissue in vivo are far more severe than those of glucose because the molar concentration of glucose in tissue is manyfold higher [133]. The requirements of specific tissues for other small molecules and for large macromolecules are poorly understood or have not yet been quantified, and the possible existence of transport limitations for large molecules is unknown and highly dependent on immunobarrier membrane properties, whereas oxygen limitations are always serious. Moreover, a low value of oxygen partial pressure (pO_2) that is sufficient to keep cells alive can nonetheless have deleterious effects on cell functions that require higher pO_2 in order to maintain high cellular ATP concentrations, for example, ATP-dependent insulin secretion [134]. Supply of oxygen to immunoisolated tissue is therefore the critical component that determines tissue survival and function.

2.4.1. Oxygen supply in islet transplantation

Islet transplantation has the potential to cure type 1 diabetes. The fraction of patients achieving initial insulin independence has improved, but longer term outcomes remain poor compared to those for whole pancreas transplants. The diverse set of factors and mechanisms that can lead to graft failure has recently been reviewed [135] and includes issues related to the donor, pancreas retrieval and transportation, islet isolation, instant blood-mediated inflammatory reaction, immunosuppressive medication, delayed immune attack, and hypoxia. Oxygen supply limitations are important in all islet processing and culture and can be particularly severe in vivo after transplantation, especially prior to revascularization, as has been confirmed by direct measurement [136–138]. In rats, roughly one-third of islets implanted in the kidney capsule are lost acutely. The limited revascularization that occurs more than a week after transplantation of naked islets does not resolve the initial oxygen limitation [139].

2.4.2. Oxygen supply to encapsulated islets

The problem of hypoxia in naked islet tissue can be even more severe with encapsulated islets. Oxygen supply to encapsulated cells depends in a complicated way on many factors, including (1) site of implantation and the local pO_2 in the blood, (2) spatial distribution of host blood vessels in the vicinity of the implant surface, (3) oxygen permeability of the membrane or encapsulant, (4) oxygen consumption rate of the encapsulated tissue, (5) geometric characteristics of the implant device, and (6) tissue density and spatial arrangement of the encapsulated cells or tissues. Despite encouraging results with various tissues and applications [18], the problem of oxygen transport limitations is a critical hurdle in all encapsulated cell therapies. It poses constraints on various device geometries, plays a predominant role in maintaining viability and function, and holds the key to achieving true immunoisolation. The maximum pO_2 in the microvasculature available for extravascular devices limits the steady-state amount of viable tissue that can be supported in terms of its volumetric or surface density.

Islets lose their vascular system from enzymatic and mechanical processing and are particularly prone to oxygen supply limitations because they have a relatively high oxygen consumption rate [134,140].

In the normal physiologic state they are highly vascularized and are supplied with blood at arterial pO_2. When cultured in vitro under ambient normoxic conditions, islets develop a necrotic core, the size of which increases with increasing islet size, as is to be expected as a result of oxygen diffusion and consumption within the islet [133]. Central necrosis of the encapsulated islets occurs after islet microcapsule transplantation, resulting in reduction in transplant volume when only a small fraction (about 10%) of the capsules have fibrotic overgrowth [141]. The fact that necrotic tissue is at the center instead of the periphery of the islet indicates that it is likely a nutrient or oxygen supply limitation causing the necrosis and not a mechanism associated with the immune system.

The reduction of pO_2 to hypoxic levels was first measured within encapsulated islets cultured in vitro nearly 30 years ago [142], and its potential importance in vivo was recognized at about the same time [9,10,143]. Hypoxia causes encapsulated islets in culture to become necrotic and to up-regulate inducible nitric oxide synthase (iNOS), which indicates that islets are producing NO that can cause damage to themselves. Hypoxia also causes islets to up-regulate monocyte chemoattractant protein 1 (MCP-1), which can attract macrophages and hence also induce islet damage postimplantation [144]. Thus, in addition to its direct deleterious effect on viability and function, hypoxia leads to an intensification of the immune response separate from the release of immunogenic peptides from dead cells. These early publications established that oxygen transport limitations exist within transplanted islet microcapsules and could have serious effects on islet survival.

An improved understanding of the interplay of oxygen diffusion and consumption rate in devices containing encapsulated islet tissue has come from development of theoretical mathematical models for a wide range of situations and configurations [11–13,62,145–150]. Simple geometries in the form of slabs, cylinders, and spheres with oxygen consumption kinetics expressed as the limiting cases of zero and first-order in oxygen concentration have been amenable to analytical solutions that have wide generality. For a specific set of operating parameters, these analyses provide an estimate of the pO_2 as a function of position throughout the device from which the viability and function can be estimated. More complicated geometries or use of more realistic nonlinear kinetic expressions (e.g., Michaels–Menten) for oxygen consumption rate require numerical solutions of the governing equations, typically by the use of finite element methods, to obtain the oxygen concentration field directly. The recent availability of software packages that incorporate finite element methods in formats that are easy to use has made this capability widely available. These analyses show that all geometries are feasible to use when tissue density is low. As density increases, the relative advantages and disadvantages of each geometry become apparent. As described in an earlier section, in general a spherical geometry is most favored and a slab least favored purely in terms of mass transfer considerations. The opposite is true for practical packaging considerations, i.e., a slab can be increased in any of three dimensions, and it may be possible to encapsulate all of the islets required for a human in a single slab configuration, as described below, whereas a sphere can only be increased in one dimension, and a single large microbead containing 250,000 islets is impractical from many standpoints. The cylindrical configuration in the form of a small diameter hollow fiber is not usable for large quantities of tissue for the reasons given in a previous section.

In addition to solving for the pO_2 field in islet tissue, the mathematical models have been used to estimate viability and function of islet tissue in specific configurations. For this purpose, estimates of certain viability and function parameters are needed. The critical pO_2 at which cells lose viability is unknown and has been estimated at about 0.1 mm Hg, whereas the Michaels constant for mitochondrial respiration is 0.44 mm Hg. In perfusion measurements [134], the highest medium pO_2 at which second-phase insulin secretion began to be reduced was about 60 mm Hg, and an apparent half-maximal insulin secretion rate occurred at 27 mm Hg. A model for the intrinsic effect of pO_2 on an individual cell is unknown. Using a hyperbolic function akin to Michaels–Menten kinetics, the half-maximal inhibition constant was evaluated as 2.9 mm Hg by fitting the raw data [134] to a diffusion reaction model [151]. An equally good fit was obtained with a function that dropped linearly from its maximal value at 5.1 mm Hg. A much more complicated model with many adjustable parameters has been described [150]; the general applicability of the parameters and their numerical values is unknown. Examples of predicted pO_2 profiles in spherical capsules and planar slabs that were generated using numerical methods and used to estimate the viability and function of encapsulated islet tissue are provided subsequently in Section 4.1.

As O_2 diffuses radially inward from the islet surface, it is consumed by the cells and its concentration decreases toward the center of the islet. A very useful rule of thumb is revealed from analysis of a single spherical IE of human origin, containing an average of 1560 cells [152] and having a diameter of 150 μm: the outer islet surface should be at an oxygen partial pressure of about 35–40 mm Hg to maintain full viability and about 45–50 mm Hg to maintain full functionality of all cells [10,14,15,134,149,152].

At the higher values of pO_2 where zero-order kinetics apply for the oxygen consumption rate, an important dimensionless parameter arises from the differential equations that comprise the mathematical models referred to above [e.g., 11–13]:

$$\Phi^2 = \frac{Vh^2}{\alpha D P_s} \tag{1}$$

where V is the constant oxygen consumption rate per unit volume of the system under consideration, h is the radius R of a sphere or cylinder or the half thickness of a slab L, α is the Bunsen solubility coefficient for oxygen, D is the diffusivity of oxygen, and P_s is the oxygen partial pressure at the outer surface of the system. The system could be a single islet or, for example, a suspension of islets in a planar slab of alginate. In the latter situation the average reaction rate in the tissue compartment is related to the rate in islet tissue V_t by

$$V = V_t(1-\varepsilon) \tag{2}$$

where ε is the void fraction (non-tissue in the compartment). Φ^2 is known as the Thiele module and represents the ratio of the time scale for oxygen diffusion divided by the time scale for oxygen consumption in the tissue compartment. For each geometry, a fixed value of Φ corresponds to a unique operating condition such as the situation where the pO_2 at the center of an islet is a specific value, say 3 mm Hg. Eqs. (1) and (2) show how each parameter must relate to each other. For example, if Φ is fixed, then an increase in h by a factor of 2 requires that P_s increase by a factor of 4 to maintain the same pO_2 profile in the islet.

3. Defensive measures to enhance β cell survival

One approach to dealing with the effects of hypoxia is to strengthen the ability of β cells to respond to the stresses of transplantation, of which hypoxia is a major component. Strategies for enhancement of β-cell defense properties against hypoxia include gene therapy [153]. While most of these measures have been aimed at improving native islet transplantation, they are applicable not only to islets but to other cells when encapsulated as well. These diverse measures are briefly reviewed here.

Gene therapy, through overexpression or silencing of genes, has been applied to prevent β cell apoptosis, inhibit rejection, and improve islet function [154–157]. Nongenetic methods to modify β cell behavior include delivery of deferoxamine to increase HIF-α levels, which protects against adverse transplantation outcomes [158], and co-delivery of CXCL12 and Exendin-4 in alginate capsules to promote the survival, function, and proliferation of β cells [159].

An entirely different approach is preconditioning of islets. In one study, dynamic modulation of glucose concentration and pO_2 to hypoxic levels improved glucose-stimulated insulin secretion under hypoxic conditions [160]. In another study, enhanced resistance to toxic agents and hypoxia was observed in islets that underwent three repeated rounds of encapsulation, xenotransplantation, and explantation [161].

Macrophages play an active role in the immune response to immunoisolated tissues, and they are also activated by MCP secreted by beta cells under hypoxia. Neutralization or trapping of toxic molecules released by macrophages, such as nitric oxide, has been examined to enhance graft survival by including a nitric oxide scavenger such as hemoglobin within microcapsules to inhibit islet cell death through nitric oxide-mediated mechanisms. The inclusion of live erythrocytes, fixed erythrocytes, or cross-linked hemoglobin in alginate microcapsules enhances islet survival with exposure to activated macrophages or nitric oxide [162,163].

In a related study with the same methods [164], an increase in the length of islet survival after transplant was attributed to enhanced oxygen permeability. However, that mechanism is unlikely compared to trapping of nitric oxide because high mobility of hemoglobin is required in order for it to facilitate oxygen transport [165]. Similar conclusions apply to a later study [166]. Rather than rely on endogenous hemoglobin, another study used induction of cytoglobin, an intracellular oxygen binding protein, by genetic means. These modified islets underwent a reduction of ischemic cell death in culture [167] and promoted islet cell survival and insulin synthesis and secretion following transplantation with hollow fibers in rats [168]. The authors speculated that the mechanism was protection of islets from the deleterious effects of reactive oxygen species in a chronically hypoxic environment.

4. Enhancement of oxygen transport to encapsulated tissue

There are three broad areas in which research has been carried out in order to enhance oxygen supply to encapsulated therapeutic cells: (1) manipulation of diffusion distances of the device and host tissue and the oxygen permeability of encapsulating material; (2) induction of neovascularization adjacent to the immunoisolation device in order to bring blood flow close to the tissue; and (3) provision of exogenous oxygen to produce a direct increase in oxygen concentration adjacent to the tissue.

4.1. Diffusion distances and oxygen permeability

Oxygen transport to encapsulated islets can be enhanced by reducing diffusion distances through the use of smaller tissue entities or thinner membranes. Use of smaller tissue entities can be particularly effective because of the squared dependence on length scale in Eq. (1). There are drawbacks to reducing the diffusion distance because free radicals released from immune system effector cells may not become inactivated prior to reaching the encapsulated tissue, and the amount of shed antigens from the encapsulated tissue can be increased, thereby enhancing the recipient's immune response to the transplant.

Alginate capsules made using an electronic droplet generator can be made smaller than 500 μm. There are also techniques in which only a thin coherent membrane is used to coat the islets. Examples are the use of an emulsion procedure to form calcium alginate PLO microcapsules [169] or by centrifuging an islet alginate cell suspension in a discontinuous gradient that contains a barium chloride layer [170]. Because of its purported biocompatibility, polyethylene glycol and its derivatives have also been employed to prepare extremely thin coatings around islets with a layer-by-layer encapsulation technique [171] and other methods [172,173]. Methods for preparing conformal coatings have been reviewed [174]. A hydrogel layer around an islet reduces the availability of oxygen, but the magnitude of the effect can be relatively limited. Hence, the use of conformal coatings around islets is not motivated strictly on the basis of improving oxygen supply but also for the purpose of reducing the total volume of the implanted capsules, which increases with the cube of the capsule radius.

Enhanced oxygen delivery within encapsulated tissue can be achieved two ways: (1) increase the effective oxygen solubility of the encapsulant material and thus increase the oxygen permeability and the rate at which oxygen can be delivered to the tissue, for example, by combination of a highly concentrated perfluorocarbon (PFC) emulsion with alginate (PFC alginate); and (2) reduce islet tissue size so as to decrease the diffusion distance within the tissue, for example, by dispersing the islets into single cells followed by reaggregation into cell clusters smaller than the original islet.

Components that can be used for enhancing oxygen permeability are organic compounds with high oxygen solubility, such as perfluorocarbons, silicone oils, or soybean oils. Perfluorocarbon emulsions have been developed as blood substitutes and can be incorporated into the encapsulation material to increase its oxygen permeability. Perfluorocarbons and silicone oils have been used to enhance oxygen transfer in bioreactors [175,176], and including a perfluorocarbon emulsion in islet culture medium enhances islet function [177,178]. Perfluorocarbons have also been used in the storage of the pancreas prior to islet isolation to increase islet yield and storage time [179]. Low concentrations of perfluorocarbon emulsion in alginate have demonstrated beneficial effects on hepatic cells [180,181] and on islets with perfluorocarbon grafted onto alginate [182], but a study at higher concentration with insulinoma cells produced only limited beneficial effect [183]. Inclusion of hemoglobin in alginate microcapsules increases the length of islet survival after transplantation [164], but this enhancement likely results from a mechanism that does not involve enhanced oxygen permeability such as trapping of nitric oxide. Examination of emulsification parameters and physical properties in perfluorocarbon emulsion revealed the importance of particle size for enhancing oxygen transfer [184,185].

In a recent study [149], a theoretical reaction–diffusion model was used to predict the three-dimensional distribution of oxygen partial pressure in a spherical microcapsule and a planar slab containing islet tissue, from which the loss of cell viability and the reduction in insulin secretion rate was estimated. Numerical simulations were carried out for normal alginate and PFC alginate to examine the effect of capsule surface oxygen partial pressure, capsule diameter, slab thickness, diameter of tissue sphere, tissue loading, and volumetric or surface density of dispersed islet tissue on viability and function. Some of the results of that study are summarized in Figs. 2 through 8. The spatial distribution of pO_2 in capsules is shown as contour plots (Fig. 2) and radial profiles (Fig. 3). The distribution is driven by the oxygen consumption of islet tissue, and gradients develop to provide the required diffusion rate in the capsule. Reduction of tissue size leads to marked increase in oxygen levels, and use of PFC alginate leads to a modest increase. Increasing tissue loading steepens gradients and lowers oxygen levels, distorting contours in both domains at very high tissue densities because of crowding effects. Increasing capsule size at constant, high tissue loading (Fig. 2, Group 3 versus Group 2) produces the counterintuitive result that pO_2 values are higher in the larger capsules. This surprising result occurs because the tissue density and the mean consumption rate decrease with the cube of capsule radius, and the crowding effect is thereby substantially reduced, whereas diffusion distances increase with the first power of radius. The effect of capsule size is also examined in terms of the fractional second phase insulin secretion rate in Figs. 4 and 5. The benefit of increased capsule size is not observed with a single, centrally located islet but is pronounced at very high loadings with arrays of multiple tissue spheres. The oxygen distribution for an islet or an array of multiple tissue spheres within a planar slab is illustrated in Figs. 6 and 7. Similar qualitative trends are observed. Increased slab thickness is not investigated, but counterintuitive effects with increased slab thickness would not occur. A comparison between the two geometries at equivalent volumetric densities is shown in Fig. 8. Both geometries benefit from increased encapsulant permeability and reduction in tissue

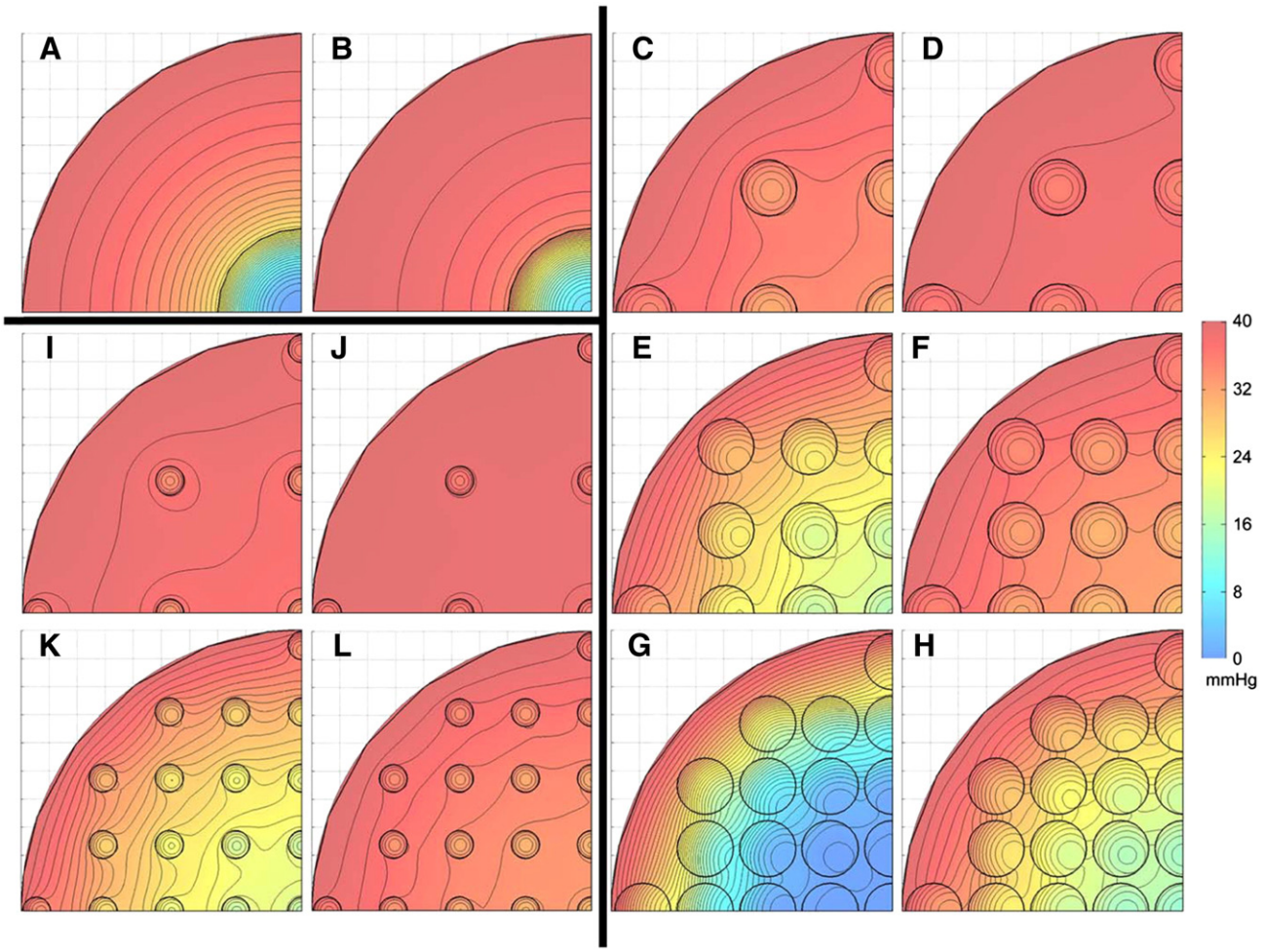

Fig. 2. Contours of constant pO_2 in a capsule containing one centrally located islet (A, B) or a multiplicity of aggregates in a cubic array plotted in a plane of symmetry that contains the maximum number of tissue spheres along the axes (C–L). Independent parameters varied include diameter of encapsulated tissue sphere(s), diameter of capsule, tissue loading, and use of normal or PFC alginate. Maximum oxygen consumption rate used in calculations is 4×10^{-8} mol/cm^3/s [97,239]; other parameters are summarized elsewhere [149]. Reproduced, with permission, from [149].

size. Under all conditions at high density, a capsule suffers less from oxygen supply limitations than a slab. However, the slab configuration is more suitable for enhancing oxygen supply via neovascularization and use of exogenous oxygen as described in subsequent sections.

The effect of enhancing encapsulant oxygen permeability was examined experimentally [186] with two model systems: (1) barium alginate and (2) barium alginate containing a 70% (w/v) perfluorocarbon (PFC) emulsion that is a far higher concentration than in prior studies and would be expected to provide a higher effective oxygen permeability of the encapsulating material. Mitochondrial function was assessed by oxygen consumption rate measurements. After low oxygen culture for 2 days, islets in normal alginate lost substantial viable tissue and displayed necrotic cores, whereas most of the original oxygen consumption rate was recovered with PFC alginate, and little necrosis was observed. These findings suggested that enhancement of oxygen permeability of the encapsulating material with a concentrated PFC emulsion improved survival of encapsulated islets under hypoxic conditions.

Small islet cell aggregates were also studied [187] to determine if their survival and function were superior to intact islets within microcapsules because of reduced oxygen supply limitation and inflammatory mediators. Islet cell aggregates were generated by dispersing rat islets into single cells and allowing them to re-aggregate in culture. Rat islets and islet cell aggregates were encapsulated in barium alginate capsules and studied when cultured in low or normal oxygen or transplanted into mice. Encapsulated islet cell aggregates were able to survive and function better than intact islets in terms of oxygen consumption rate, nuclei counts, insulin-to-DNA ratio, and glucose-stimulated insulin secretion. The islet cell aggregates also had reduced expression of pro-inflammatory genes and reduced tissue necrosis in an immunodeficient transplant model. A much greater proportion of diabetic xenogeneic transplant recipients receiving islet cell aggregates had reversal of hyperglycemia than recipients receiving intact islets. These aggregates were superior to intact islets in terms of survival and function in low-oxygen culture. They demonstrated substantially improved performance following transplantation and provided more efficient utilization of islet tissue.

4.2. Induced vascularization

Insufficient blood supply in close proximity to an immunoisolated macroencapsulation device exacerbates the problem of oxygen supply. Approaches to improve oxygenation by increasing blood supply to the vicinity of the device fall into three categories: (1) Release of angiogenic factor(s) from drug delivery constructs or cells, (2) use of synthetic materials that induce neovascularization by virtue of their microarchitecture, and (3) minimization of the inflammatory response that leads to formation of a fibrotic layer adjacent to an implanted material.

4.2.1. Angiogenic protein

In order to induce vascularization, early studies perfused a solution containing vascular endothelial growth factor (VEGF) through the surface of the device [190] or incorporated VEGF together with islets in

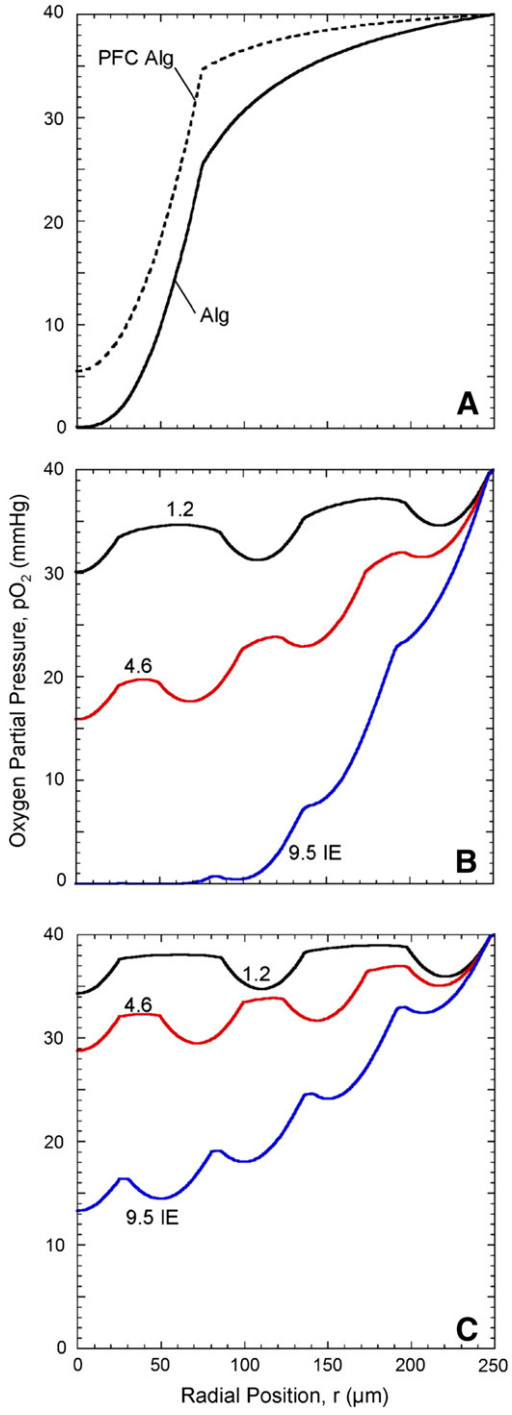

Fig. 3. Profile of pO_2 versus radial position in a 500-μm diameter spherical capsule. Each profile corresponds to the contour plot in specific panels in Fig. 2 as follows: (A) A, B; (B) C, E, and G; (C) D, F, and H.
Reproduced, with permission, from [149].

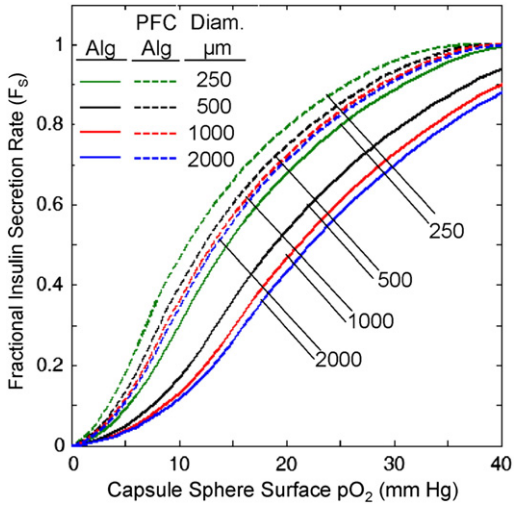

Fig. 4. Effect of capsule diameter on fraction of normal insulin secretion rate as a function of capsule surface pO_2. Solid lines — normal alginate, dashed lines — PFC alginate, corresponding to Fig. 2, panels A and B, respectively. Reprinted, with permission, from [149].

implanted hollow fibers in diabetic mice [189,190]. In the later study, VEGF increased the viability of encapsulated islets and the duration of normoglycemia following transplantation. Other studies incorporated endothelial cell growth factors into hollow fibers containing alginate [191,192], fibroblast growth factor-1 into alginate microcapsules for sustained release without cells [193–195], and fibroblast growth factor-2 in gelatin implanted in bag-like structures [196–198], all of which caused blood vessel density near the implant to increase for a period of time and then decline. Stable neovasculature formed adjacent to the devices only with encapsulated endothelial cells in these devices [199], which suggests that the vasculature was maintained over long periods of time by hypoxia-induced angiogenesis [200–202] attributable to the encapsulated endothelial cells. Continuous release of VEGF and/or other angiogenic factors from the device into the local environment is therefore likely to be necessary for formation of stable vasculature. This phenomenon can occur with other cell types including β-cells [203,204]. Such sustained release has also been achieved with coencapsulated mesenchymal stem cells [70,205] and encapsulated fibroblasts [206], vascular endothelial cells [199], bone marrow progenitor cells [207], and β cells and other cells engineered to increase secretion of vascular endothelial growth factor.

A far larger body of literature has recently developed around neovascularization for purposes other than support of microencapsulation devices, especially therapeutic vascularization and related tissue engineering applications. These areas have been extensively reviewed, and on-going research may provide new ideas and methods useful for encapsulated therapeutic cells [e.g., 208–219].

4.2.2. Vascularizing membranes

It was discovered at Baxter Healthcare in the 1990s that certain microporous membranes can induce neovascularization at the membrane surface by virtue of the membrane microarchitecture alone without the use of exogenous agents. Neovascularization at the membrane–tissue interface occurred with several membranes that had nominal pore sizes large enough to allow complete penetration by host cells (0.8–8 μm pore size) [72]. This discovery led to the development of a composite structure incorporating two membranes: (1) an exterior vascularizing membrane that has an optimal pore size (5 μm) so that cells can penetrate the layer and vascularization is induced, and (2) a cell-impermeable immunoisolating membrane for protection from the direct pathway, pore size of 0.45 μm, both made from PTFE [72]. Under some circumstances, a highly porous third outer layer was added that consisted of thicker fibers for mechanical support. Devices using vascularization membranes, including the Theracyte™ planar diffusion chamber (originally developed at Baxter Healthcare, Corp., later marketed by Theracyte, Inc.) has been effective in maintaining cell viability and protecting from allograft rejection in rodent models [220,221]. Because host tissue may integrate with the outer layers, device retrieval can be more difficult than with a device having a smooth surface. The device can be preimplanted in order to induce vascularization of the device prior to transplantation, which aids in islet survival post-transplantation [78–80]. In addition, infusion of the device with vascular endothelial growth factor (VEGF) improves the density of

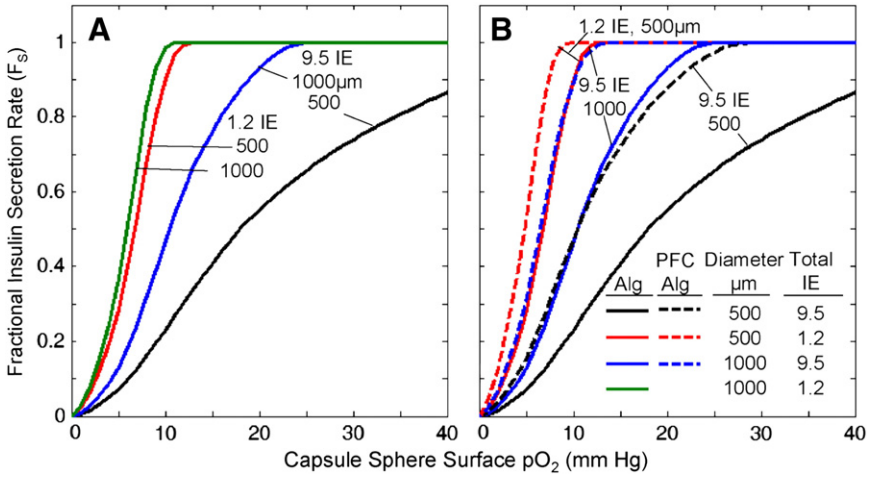

Fig. 5. Effect of capsule diameter and tissue loading on fraction of normal insulin secretion rate. These results correspond to contour plots designated by panels in Fig. 2 as follows: (A) C, G, I and K; (B) D, H, J and L. Note the improved performance of the larger capsules at high tissue loading as compared to smaller capsules at the same loading.
Reproduced, with permission, from [149].

blood vessels that form around the device [188]. Additional papers concerning Theracyte devices are discussed in the previous section on planar slab diffusion chambers. Other materials have been examined for use in promoting neovascularization, including a dual porosity electrospun nylon membrane [222] and a stainless steel mesh [223]. The timecourse of membrane microarchitecture-driven neovascularization involves migration of vessels towards the membrane surface over a 10-day period [224], after which the microvascular structure within 15 μm of the surface stabilizes for a long period of time [79,225].

As previously described, the observations with vascularizing membranes are consistent with the course of events seen in normal wound healing [226]. Neutrophils are the predominant cell type at the site of injury within the first 24–48 h, killing and phagocytosing any bacteria present. The macrophage becomes the predominant cell after this time, removing cellular and foreign debris from the area. Within 3–4 days, fibroblasts migrate out of the surrounding connective tissue into the wound area and begin to synthesize collagen, which quickly fills the wound space. New blood vessels begin to grow into the area

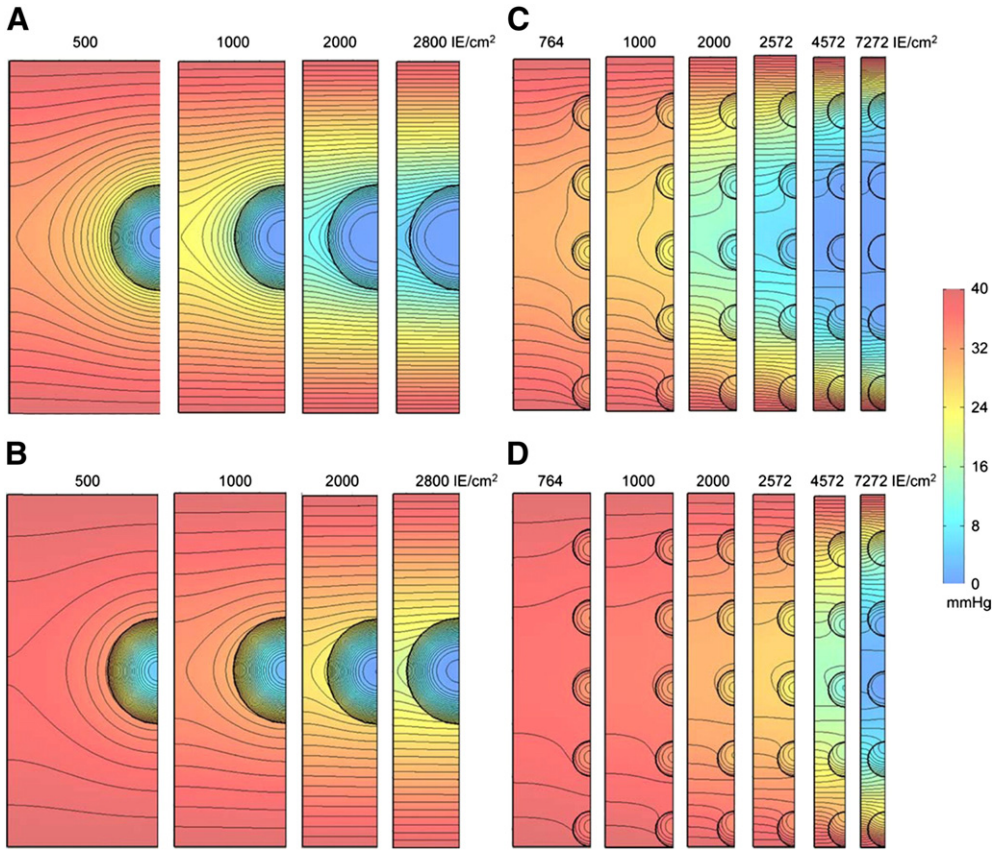

Fig. 6. Contours of constant pO_2 in a 500-μm thick slab containing centrally located islets (A, normal alginate; B, PFC alginate) or a multiplicity of 50-μm diameter spherical aggregates with 10 layers arranged in a body centered cubic array (C, normal alginate; D, PFC alginate) at a variety of surface densities.
Reprinted with permission from [149].

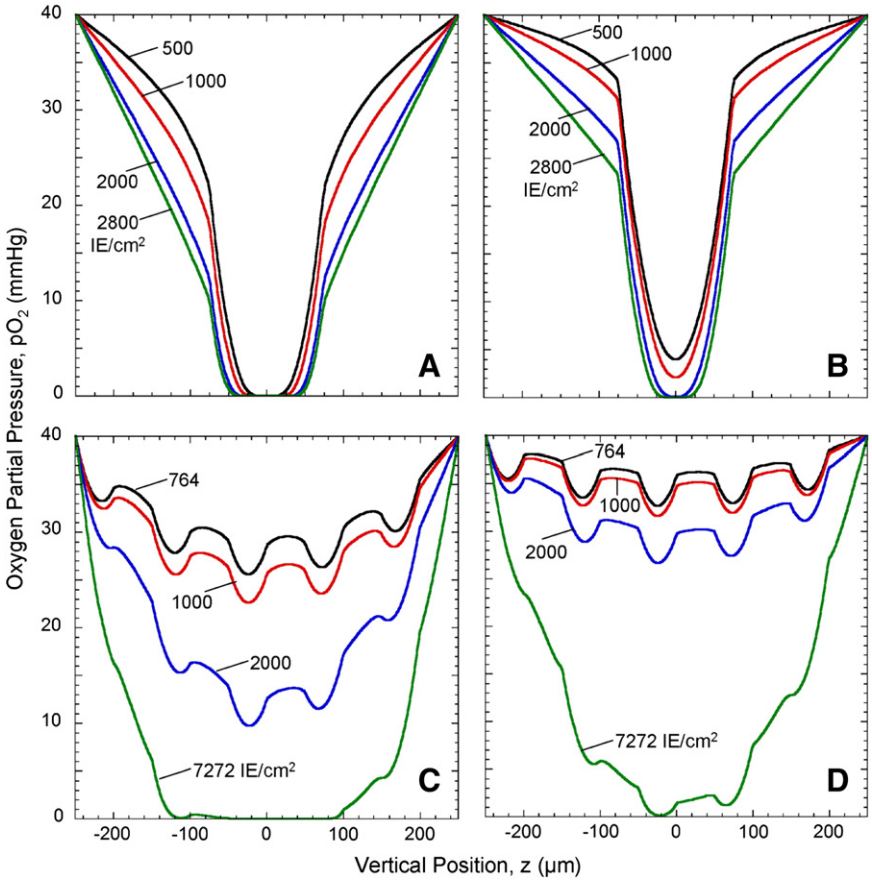

Fig. 7. Oxygen profiles plotted versus vertical position in a plane that cuts through the center of one column of tissue spheres in a 500-μm thick slab. Panels (A), (B), (C), and (D) correspond to equivalent panels in Fig. 5.
Reprinted, with permission, from [149].

at this time to supply oxygen and nutrients needed by the metabolically active fibroblasts and macrophages in the wound. The main difference between normal wound healing and membrane microarchitecture-driven neovascularization is that in normal wound healing the vessels begin to regress in the second week, while in membrane microarchitecture-driven neovascularization the vessels remain at the interface. The mature scar is avascular and acellular in a normal wound; in membrane microarchitecture-driven neovascularization, a multitude of vessels persist at the material–tissue interface, which is overlain by an otherwise largely acellular scar (akin to a foreign body response).

We have hypothesized [224] that host inflammatory macrophages invade these microporous materials and encounter locally reduced oxygen partial pressure caused by oxygen consumption of these cells. This hypoxia induces macrophages to secrete angiogenic factors provided that the macrophages have not flattened on the internal structural elements of the membrane. Macrophages are capable of producing a large number of molecules that are involved in the process of angiogenesis. Any flattened macrophages that adhere to the interior membrane elements would begin the process of "frustrated phagocytosis" that leads to the foreign body response and formation of a fibrotic capsule.

4.2.3. Minimization of inflammatory response

A different approach to improving oxygen supply falls in the general category of improved biocompatibility of materials in contact with host tissue [227,228]. This approach is based on the notion that elimination of the ubiquitous avascular foreign body response would result in improved oxygen supply in the vicinity of the implant, whether it be microcapsules that perform better when not attached to tissue surfaces or static macroencapsulation devices. Despite extensive efforts for decades, studies with new materials and coatings to reduce protein absorption and immune cell adhesion have achieved at best inconsistent results in vivo with respect to eliminating acute and chronic inflammation responses [229]. Active strategies for delivery of anti-inflammatory agents are currently in vogue, and it is too early to assess their effectiveness. Nonetheless, it is important to keep in mind that when neovascularization is successful, new vessels often develop immediately adjacent to the implanted material and can be overlain by a foreign body response, especially in the case of vascularizing membranes. In this context, elimination of the foreign body response may be neither sufficient nor necessary.

4.3. Elevated oxygen concentration adjacent to tissue

Induction of neovascularization adjacent to an implanted macroencapsulation device will improve viability and function of encapsulated tissue in comparison to no neovascularization at all. However, neovascularization should not be viewed as a panacea because the best it can do is to provide an oxygen microenvironment around the implant commensurate with the pO_2 in the capillaries and perivascular tissue within the microvasculature. The pO_2 measured in the microcirculation of the awake hamster skinfold reaches a minimum in the range of 25 to 35 mm Hg for tissue, capillaries, and the smallest arterioles and venules [230], which is comparable to values in the venous circulation of humans [231,232]. This is the same range for pO_2 values required at the surface of a single 150-μm diameter islet in order to keep cells alive all the way to the center (but not high enough to ensure full function by all cells). In contrast, pancreatic islets are perfused with arterial blood normally at a pO_2 in excess of 100 mm Hg. Because the oxygen level available from the microvasculature is actually insufficient to ensure that all of the islet tissue is maximally functional, it is desirable

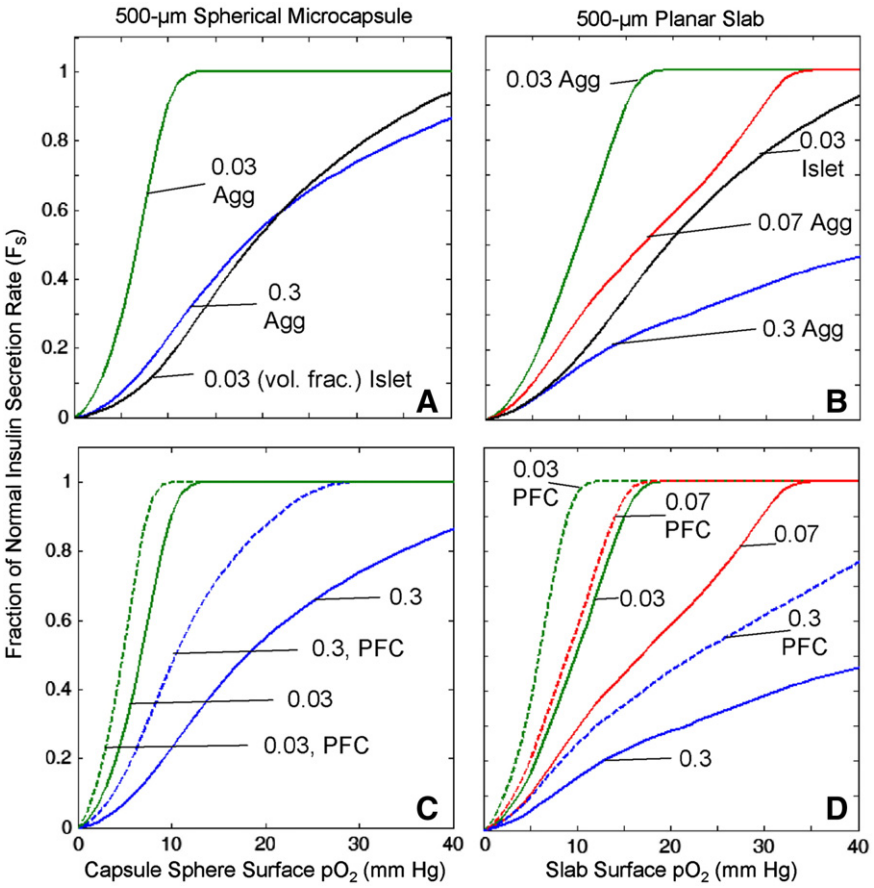

Fig. 8. Comparison of fraction of normal insulin secretion rate for a 500-μm microcapsule (A, C) and 500-μm planar slab (B, D) that have similar loading of 50-μm aggregates and 150-μm islets based on the encapsulated tissue volume fraction (indicated by numbers on the figures). The encapsulant is normal alginate (slid lines) or 70% (w/v) PFC alginate (dashed lines). For a microcapsule, cases examined are one centrally located 150-μm islet and 1.2 and 9.5 IE of 50-μm aggregates in a 500-μm microcapsule, which correspond to tissue volume fractions of 0.03, 0.07, and 0.3, respectively; for a planar slab, 150-μm islets arranged in 3 layers and 50-μm aggregates arranged in 10 layers at a tissue density of 764 IE/cm^2, which correspond to a tissue volume fraction of 0.03, and 10 layers of 50-μm aggregates at a density of 2000 IE/cm^2 for the 0.07 volume fraction and a density of 7272 IE/cm^2 for the 0.3 volume fraction.

to expose the encapsulated islet tissue to a level greater than that normally available locally in the tissue, even for supporting just a monolayer of islets. In order to increase islet density, which is extremely important in order to decrease required device dimensions, supply of exogenous oxygen at a higher pO_2 is needed.

One solution to this problem is to supply implanted tissue with additional oxygen at a higher pO_2 adjacent to one side of a tissue slab in a planar immunobarrier device, either by generation within the body or by injection from outside. On the other side, the exterior of the device is exposed to either culture medium for in vitro studies or to the host tissue for in vivo conditions. This concept of supplying exogenous oxygen at elevated pO_2 to one side of a tissue slab was first explored [84] using the electrolytic decomposition of water in an electrolyzer for in situ oxygen generation. The electrolyzer was in the form of a thin, multilayer sheet, within which electrolysis reactions took place on the anode and the cathode to form oxygen and hydrogen, respectively. In vitro studies with βTC3 cells contained in a chamber mated to the in situ oxygen generation device showed that the thickness of viable tissue increased with oxygen generation as compared to the situation of no oxygen generation [84]. Furthermore, glucose-stimulated insulin secretion rate from islets contained in such devices and placed in a hypoxic environment was higher with than without oxygen generation.

A detailed theoretical mathematical analysis [13] of oxygen diffusion and consumption in the tissue compartment of such a device containing islets encapsulated in a planner slab revealed that support of tissue at a higher surface density by virtue of either increased slab thickness at constant volumetric density or increased volumetric density at constant slab thickness would require some of the islets (those closer to the oxygen source) to be exposed to pO_2 substantially higher than physiological levels. This caused concern about the possibility of oxygen toxicity to encapsulated islets [13]. There is very limited prior work in the literature on the effect of hyperoxia on pancreatic islets, and even recent studies provide conflicting guidance [233,234]. Work on in situ electrochemical generation stopped in the early 2000s for non-technical reasons, and its ultimate utility remains to be further developed and tested.

Another method investigated for in situ oxygenation is light illumination of encapsulated algae to produce oxygen by photosynthesis [85,235]. However, this method is far less efficient than electrolysis of water. Both methods would require an implantable battery and transcutaneous energy transmission for recharging. Equipment and power requirements for water hydrolysis would be much smaller. Hydrolysis of calcium peroxide [236] and sodium percarbonate [237] has been proposed for oxygen generation on a temporary basis where a chronic increase in oxygen supply is not needed. The former study demonstrated improvement in viability and function of a β cell line and islets in vitro for a 3-week period.

Periodic supply of exogenous oxygen in gaseous form to an internal chamber in an implanted device is an alternative to in situ generation. One novel approach developed by Beta-O_2 Technologies in Israel, termed the βAir® device, contains islets immobilized within a flat alginate slab that is supplied with oxygen by diffusion through a gas permeable membrane from an adjacent inner gas chamber. The gas chamber is replenished daily through an external port. The encapsulated islets are protected from the host immune cells by a microporous membrane, the pores of which are filled with alginate. Initial studies with the βAir® device achieved normoglycemia and nearly-normal intravenous

tolerance tests in diabetic rats with isogenic and allogeneic islet implants [88,89] for up to 6 months without immunosuppression and demonstrated the benefit of supplying oxygen at increased pO_2 levels from one side of the alginate slab.

In a subsequent study [90], rat islets encapsulated in the device using a relatively high alginate concentration of 6% (w/v) and implanted in minipigs for up to 90 days demonstrated persistent graft functions, restoration of normoglycemia, and no evidence of any graft rejection without any immunosuppressive therapy. The success of this xenotransplantation study is consistent with the notion posed earlier in this review that islets maintained alive with good oxygenation behind a restrictive hydrogel immunobarrier do not release sufficient shed antigen to cause an immune response by the indirect pathway. The high alginate concentration may have also contributed by retarding the permeation of shed antigen across the immunobarrier.

In connection with the other studies just described, the effect of islet density on oxygen concentration profiles in the islet slab was measured with a microelectrode. These measurements were used to determine the pO_2 levels required in the gas chamber in order to ensure that the external surface of islets furthest away from the oxygen source were not exposed to a pO_2 less than 50 mm Hg so as to retain maximum function of all cells [92]. Islet densities as high as 4800 IEQ/cm^2 were studied. Devices containing various islet densities and sufficient gas chamber oxygen levels were implanted in STZ-induced diabetic rats for periods ranging from 2 to 6 months. The rats achieved normoglycemia for all time periods and displayed near-normal responses to intravenous glucose tolerance tests. Furthermore, the total oxygen consumption rate of islets in the explanted device was the same as when loaded into the device, thereby demonstrating that no viable islet tissue was lost. The data demonstrated the ability of the device to supply oxygen to implanted islets and to maintain islet viability and function at high islet immobilization densities, thereby reducing the required size of an implanted device suitable for humans. At the highest density studied, 250,000 human islets could be supported on a device having an active surface area on each side of 25 cm^2, which could be implanted in a human at a variety of sites.

Lastly, a 63-yer-old patient with type 1 diabetes was transplanted with 160,000 human islets in the device (2100 IE/kg, surface density of 4100 IE/cm^2) and followed for 10 months. The transplant demonstrated persistent graft function with regulated insulin secretion and preservation of islet morphology without any immunosuppressive therapy [91]. Hemoglobin A1C levels decreased, and the insulin requirement was reduced. Although only moderate improvements in clinical disease were observed, the results are nonetheless remarkable because they were achieved with an extremely low islet dose. After explantation, the complete device was submerged in an elevated glucose solution and demonstrated rapid insulin and C-peptide secretion in response to mild glucose challenge.

Taken together, these recently-published studies obtained a number of important findings, some for the first time: (1) At the highest density studied, islets nearest the gas chamber were exposed to pO_2 ranging from 570 to 350 mm Hg over each 24 hour period. No effects of hyperoxia were observed. (2) There was no significant change in total oxygen consumption rate of implanted islets from pre- to post-transplantation, and islet quality assessed as oxygen consumption rate per IE also did not change. (3) Almost all islets maintained their viability throughout the transplantation period, and few islets died. The encapsulated islets maintained normal glucose homeostasis during the period of implantation in diabetic animals. (4) Xenotransplantation was achieved in a large animal without immunosuppression. (5) An islet allograft functioned in a human for 10 months without immunosuppression.

These findings with the βAir® device suggest that a human clinical allograft trial and primate xenograft trial with therapeutically relevant islet doses are appropriate.

Although not aimed solely at improving oxygen supply, a different strategy has been to start from scratch and engineer an improved site and microenvironment for islet transplantation [70,101,238]. These efforts are generating novel ideas, some of which may be applicable to islet immunoisolation.

5. Perspective

In order for the promise of encapsulated cell therapy to be realized, many issues must be dealt with simultaneously. Foremost among these, at least for islet transplantation, is the problem of oxygen supply. Solving this problem alone is not sufficient, but research over four decades demonstrates that it is necessary. Furthermore, because loss of viability and function may be a consequence of different mechanisms, there is scant justification for extensive research in systems and configurations that suffer from substantial oxygen diffusion limitations and have no prospect of amelioration.

Adequate oxygenation of encapsulated tissue maintains its viability and function. It allows more efficient use of tissue, and it is likely to play a key role in eliminating the need for immunosuppression with xenogeneic tissue. Two approaches have emerged as potentially promising: (1) neovascularization to increase blood supply immediately adjacent to the device–host interface, and (2) use of exogenous oxygen to increase its concentration at the encapsulated tissue interface away from the device–host interface. Vascularizing membranes currently appear to be the simplest route to stable, long-term neovascularization. The Theracyte device has been used on a small scale by several academic and commercial laboratories for small volumes of tissue and for islet precursor cells that may be more resistant to hypoxia. However, vascularizing membranes have not yet been studied with large amounts of tissue nor successfully with adult islets. There is scant data in the literature to assess the maximum tissue density attainable. Mathematical model calculations indicate that the surface density will be modest even if complete, uniform neovascularization is attained, thereby necessitating a device with large surface area. Some of the other methods described, such as the use of an encapsulant with increased permeability or reduction in islet aggregate size, may be combined with vascularizing membranes to increase the supportable surface density.

Use of exogenous oxygen at higher concentrations than physiological has provided promising results recently, including demonstration that hypoxia is not toxic to islet tissue, virtually all of the implanted islets can be maintained viable and functional without loss of tissue, and xenogeneic rejection can be prevented by immunoisolation. As with any breakthrough, these findings need to be repeated and tested on a larger scale. The recently-reported device design currently requires frequent injection of oxygen-enriched gas through a port, which may be tolerable to only a fraction of the diabetic population. Transcutaneous power transmission to a rechargeable battery may be more palatable, in which case electrochemical generation of oxygen or related techniques may ultimately prove most attractive.

The recent developments for improving oxygen supply to encapsulated tissue are cause for optimism that clinical application may be possible in the foreseeable future.

Acknowledgment

Preparation of this manuscript was supported in part by the Juvenile Diabetes Research Foundation and the National Institutes of Health, USA.

References

[1] A. Omer, V. Duvivier-Kali, J. Fernandes, V. Tchipashvili, C.K. Colton, G.C. Weir, Long-term normoglycemia in rats receiving transplants with encapsulated islets, Transplantation 79 (1) (2005) 52–58.

[2] J. Brauker, G.H. Frost, V. Dwarki, T. Nijjar, R. Chin, V. Carr-Brendel, C. Jasunas, D. Hodgett, W. Stone, L.K. Cohen, et al., Sustained expression of high levels of human factor IX from human cells implanted within an immunoisolation device into athymic rodents, Hum. Gene Ther. 9 (1998) 879–888.

[3] F. Schwenter, B.L. Schneider, W.F. Pralong, N. Deglon, P. Aebischer, Survival of encapsulated human primary fibroblasts and erythropoietin expression under xenogeneic conditions, Hum. Gene Ther. 15 (2004) 669–689.
[4] C. Hasse, G. Klock, A. Schlosser, U. Zimmermann, M. Rothmund, Parathyroid allotransplantation without immunosuppression, Lancet North Am. Ed. 350 (1997) 1296–1297.
[5] E. Buchser, M. Goddard, B. Heyd, J.M. Joseph, J. Favre, N. de Tribolet, M. Lysaght, P. Aebischer, Immunoisolated xenogeneic chromaffin cell therapy for chronic pain: initial clinical experience, Anesthesiology 85 (5) (1996) 1005–1012.
[6] H. Kishima, T. Poyot, J. Bloch, J. Dauguet, F. Conde, F. Dolle, F. Hinnen, W. Pralong, S. Palfi, N. Deglon, et al., Encapsulated GDNF-producing C2C12 cells for Parkinson's disease: a preclinical study in chronic MPTP-treated baboons, Neurobiol. Dis. 16 (2004) 428–439.
[7] J. Bloch, A.C. Bachoud-Levi, N. Deglon, J.P. Lefaucheur, L. Winkel, S. Palfi, J.P. Nguyen, C. Bourdet, V. Gaura, P. Remy, P. Brugieres, M.-F. Boisse, S. Baudic, P. Cesaro, P. Hantraye, P. Aebischer, M. Peschanski, Neuroprotective gene therapy for Huntington's disease, using polymer-encapsulated cells engineered to secrete ciliary neurotrophic factor: results of a phase I study, Hum. Gene Ther. 15 (2004) 968–975.
[8] P. Aebischer, M. Schluep, N. Deglon, J.-M. Joseph, L. Hirt, B. Heyd, M. Goddard, J.P. Hammang, A.D. Zurn, A.C. Kato, et al., Intrathecal delivery of CNTF using encapsulated genetically modified xenogeneic cells in amyotrophic lateral sclerosis patients, Nat. Med. 2 (6) (1996) 696–699.
[9] C.K. Colton, E.S. Avgoustiniatos, Bioengineering in development of the hybrid artificial pancreas, J. Biomech. Eng. 113 (1991) 152–170.
[10] C.K. Colton, Implantable biohybrid artificial organs, Cell Transplant. 4 (4) (1995) 415–436.
[11] E.S. Avgoustiniatos, C.K. Colton, Effect of external oxygen mass transfer resistance on viability of immunoisolated tissue, Ann. N.Y. Acad. Sci. 831 (1997) 145–167.
[12] E.S. Avgoustiniatos, C.K. Colton, Design considerations in immunoisolation, in: R. Lanza, R. Langer, W. Chick (Eds.), Principles of Tissue Engineering, R.G. Landes, Austin, TX, 1997, pp. 333–346.
[13] E.S. Avgoustiniatos, H. Wu, C.K. Colton, Engineering challenges in immunoisolation device development, in: R.P. Lanza, R. Langer, J. Vacanti (Eds.), Principles of Tissue Engineering, Second ed., Academic Press, San Diego, 2000, pp. 331–350.
[14] A.S. Lewis, C.K. Colton, Tissue engineering for insulin replacement in diabetes, in: P.X. Ma, J. Elisseff (Eds.), Scaffolding in Tissue Engineering, Taylor & Francis, Boca Raton, FL, 2005, pp. 575–598.
[15] A.S. Lewis, C.K. Colton, Engineering challenges in immunobarrier device development, in: R.P. Lanza, R. Langer, J. Vacanti (Eds.), Principles of Tissue Engineering, Third ed., Academic Press, San Diego, 2007, pp. 405–418.
[16] R.P. Lanza, S.J. Sullivan, W.L. Chick, Islet transplantation with immunoisolation, Diabetes 41 (12) (1992) 1503–1510.
[17] R.P. Lanza, J.L. Hayes, W.L. Chick, Encapsulated cell technology, Nat. Biotechnol. 14 (1996) 1107–1111.
[18] A. Fort, N. Fort, C. Ricordi, C.L. Stabler, Biohybrid devices and encapsulation technologies for engineering a bioartificial pancreas, Cell Transplant. 17 (9) (2008) 997–1003.
[19] N. Kobayashi, Bioartificial pancreas for the treatment of diabetes, Cell Transplant. 17 (1–2) (2008) 11–17.
[20] J.T. Wilson, E.L. Chaikof, Challenges and emerging technologies in the immunoisolation of cells and tissues, Adv. Drug Deliv. Rev. 60 (2) (2008) 124–145.
[21] J.A. Giraldo, J.D. Weaver, C.L. Stabler, Enhancing clinical islet transplantation through tissue engineering strategies, J. Diabetes Soc. Tech. 4 (5) (2010) 1238–1247.
[22] N.V. Krishnamurthy, B. Gimi, Encapsulated cell grafts to treat cellular deficiencies and dysfunction, Crit. Rev. Biomed. Eng. 39 (6) (2011) 473–491.
[23] L. Robles, R. Storrs, M. Lamb, M. Alexander, J.R.T. Lakey, Current status of islet encapsulation, Cell Transplant. (2013), http://dx.doi.org/10.3727/096368913X670949 (in press).
[24] W.L. Chick, J.J. Perna, V. Lauris, P.M. Galletti, G. Panol, A.D. Whittemore, A.A. Like, C.K. Colton, M.J. Lysaght, An artificial pancreas utilizing living beta cells: effects on glucose hemostatis in diabetic rats, Science 197 (1977) 780–782.
[25] S.J. Sullivan, T. Maki, K.M. Borland, M.D. Mahoney, B.A. Solomon, T.E. Muller, A.P. Monaco, W.L. Chick, Science 252 (5006) (1991) 718–721.
[26] G.L. Warnock, R.V. Rajotte, Critical mass of purified islets that induce normoglycemia after implantation into dogs, Diabetes 37 (1988) 467–470.
[27] H. Ikeda, N. Kobayashi, Y. Tanaka, S. Nakaji, C. Yong, T. Okitsu, M. Oshita, S. Matsumoto, H. Noguchi, M. Narushima, K. Tanaka, A. Miki, J.D. Rivas-Carrillo, A. Soto-Gutierrez, N. Navarro-Alvarez, K. Tanaka, H.-S. Jun, N. Tanaka, J.-W. Yoon, A newly developed bioartificial pancreas successfully controls blood glucose in totally pancreatectomized diabetic pigs, Tissue Eng. 12 (7) (2006) 1799–1809.
[28] J. Park, Y.V. Kalinin, S. Kadam, C.L. Randall, D.H. Gracias, Design for a lithographically patterned bioartificial endocrine pancreas, Artif. Organs 37 (12) (2013) 1059–1067.
[29] F. Lim, A.M. Sun, Microencapsulated islets as bioartificial endocrine pancreas, Science 210 (1980) 908–910.
[30] N. Trivedi, M. Keegan, G.M. Steil, J. Hollister-Lock, W.M. Hasenkamp, C.K. Colton, S. Bonner-Weir, G.C. Weir, Islets in alginate macrobeads reverse diabetes despite minimal acute insulin secretory responses, Transplantation 71 (2) (2001) 203–211.
[31] L.S. Gazda, H.V. Vinerean, M.A. Laramore, C.H. Diehl, R.D. Hall, A.L. Rubin, B.H. Smith, Encapsulation of porcine islets permits extended culture time and insulin independence in spontaneously diabetic BB rats, Cell Transplant. 16 (6) (2007) 609–620.
[32] R.W. Holdcraft, L.S. Gazda, L. Circle, H. Adkins, S.G. Harbeck, E.B. Meyer, M.A. Bautista, P.C. Martis, M.A. Laramore, H.V. Vinerean, R.D. Hall, B.H. Smith, Enhancement of in vitro and in vivo function of agarose encapsulated porcine islets by changes in the islet microenvironment, Cell Transplant. (2013), http://dx.doi.org/10.3727/096368913X667033 (In Press).
[33] R.P. Lanza, D.M. Ecker, W.M. Kühtreiber, J.P. Marsh, J. Ringeling, W.L. Chick, Transplantation of islets using microencapsulation: studies in diabetic rodents and dogs, J. Mol. Med. 77 (1999) 206–210.
[34] C. Toso, Z. Mathe, P. Morel, J. Oberholzer, D. Bosco, D. Sainz-Vidal, D. Hunkeler, L.H. Buhler, C. Wandrey, T. Berney, Effect of microcapsule composition and short-term immunosuppression on intraportal biocompatibility, Cell Transplant. 14 (2005) 159–167.
[35] S. Schneider, P.J. Feilen, F. Brunnenmeier, T. Minnemann, H. Zimmermann, U. Zimmermann, M.M. Weber, Long-term graft function of adult rat and human islets encapsulated in novel alginate-based microcapsules after transplantation in immunocompetent diabetic mice, Diabetes 54 (2005) 687–693.
[36] R.P. Lanza, W.K. Kühtreiber, D. Eckler, J.E. Staruk, W.L. Chick, Xenotransplantation of porcine and bovine islets without immunosuppression using uncoated alginate microspheres, Transplantation 59 (10) (1995) 1377–1384.
[37] P. de Vos, B.J. De Haan, A. De Haan, J. van Zanten, M.M. Faas, Factors influencing functional survival of microencapsulated islet grafts, Cell Transplant. 13 (2004) 515–524.
[38] K.K. Papas, R.C. Long Jr., I. Constantinides, A. Smabanis, Effects of short-term hypoxia on a transformed cell-based bioartificial construct, Cell Transplant. 9 (3) (2000) 415–422.
[39] R. Calafiore, G. Basta, G. Luca, M. Calvitti, G. Calabrese, L. Racanicchi, G. Macchiarulo, F. Mancusso, L. Guido, P. Brunetti, Grafts of microencapsulated pancreatic islet cells for the therapy of diabetes melliltus in nonimmunosuppressed animals, Biotechnol. Appl. Biochem. 39 (2004) 159–164.
[40] M.D. Darrabie, W.F. Kendall, E.C. Opara, Characteristics of poly-L-ornithine-coated alginate microcapsules, Biomaterials 26 (2005) 6846–6852.
[41] B. Amsden, N. Turner, Diffusion characteristics of calcium alginate gels, Biotech. Bioeng. 65 (5) (1999) 605–610.
[42] F. Cellesi, W. Weber, M. Fussenegger, J.A. Hubbell, N. Tirelli, Towards a fully synthetic substitute of alginate: optimization of a thermal gelation/chemical cross-linking scheme ("tandem" gelation) for the production of beads and liquid-core capsules, Biotechnol. Bioeng. 88 (6) (2004) 740–749.
[43] G. Orive, R.M. Hernandez, A.R. Gascon, R. Calafiore, T.M.S. Chang, P. de Vos, G. Hortelano, D. Hunkeler, I. Lacik, J.L. Pedraz, History, challenges and perspectives of cell microencapsulation, Trends Biotechnol. 22 (2) (2004) 87–92.
[44] V. Vijayaganapathy, B.E. Tuch, Islet transplantation and encapsulation: an update on recent developments, Rev. Diabet. Stud. 8 (1) (2011) 51–67.
[45] E.S. O'Sullivan, A. Vegas, D.G. Anderson, G.C. Weir, Islets transplanted in immunoisolation devices: a review of the progress and the challenges that remain, Endocr. Rev. 32 (6) (2011) 827–844.
[46] P. de Vos, H.A. Lazarjani, D. Poncelet, M.M. Faas, Polymers in cell encapsulation from an enveloped cell perspective, Adv. Drug Deliv. Rev. 67–68 (2014) 15–34.
[47] Y. Sun, X. Ma, D. Zhou, I. Vacek, A.M. Sun, Normalization of diabetes in spontaneously diabetic cynomologus monkeys by xenografts of microencapsulated porcine islets without immunosuppression, J. Clin. Invest. 98 (6) (1996) 1417–1422.
[48] R. Calafiore, G. Basta, G. Luca, A. Lemmi, M.P. Montanucci, G. Calabrese, L. Racanicchi, F. Mancuso, P. Brunetti, Microencapsulated pancreatic islet allografts into nonimmunosuppressed patients with type 1 diabetes: first two cases, Diabetes Care 29 (2006) 137–138.
[49] B.J. Hering, M. Wijkstrom, M.L. Graham, M. Hardstedt, T.C. Aasheim, T. Jie, J.D. Ansite, M. Nakana, J. Cheng, W. Li, K. Moran, U. Christians, C. Finnegan, C.D. Mills, D.E. Sutherland, P. Bansal-Pakala, M.P. Murtaugh, N. Kirchof, H.J. Schuurman, Prolonged diabetes reversal after intraportal xenotransplantation of wild-type porcine islets in immunosuppressed nonhuman primates, Nat. Med. 12 (2006) 301–303.
[50] J.L. Foster, G. Williams, L.J. Williams, B.E. Tuch, Differentiation of transplanted microencapsulated fetal pancreatic cells, Transplantation 83 (2007) 1440–1448.
[51] R.B. Elliott, L. Escobar, P.L. Tan, M. Muzina, S. Zwain, C. Buchanan, Live encapsulated porcine islets from a type 1 diabetic patient 9.5 yr after xenotransplantation, Xenotransplantation 14 (2007) 157–161.
[52] B.E. Tuch, G.W. Keogh, L.J. Williams, W. Wu, J.L. Foster, V. Vaithilingam, R. Philips, Safety and viability of microencapsulated human islets transplanted into diabetic humans, Diabetes Care 32 (2009) 1887–1889.
[53] T.A. Desai, T. West, M. Cohen, T. Boiarski, A. Rampersaud, Nanoporous microsystems for islet cell replacement, Adv. Drug Deliv. Rev. 56 (2004) 1661–1673.
[54] O. Hauser, E. Prieschl-Grassauer, B. Salmonss, Encapsulated, genetically modified cell producing in vivo therapeutics, Curr. Opin. Mol. Ther. 6 (4) (2004) 412–420.
[55] R.P. Lanza, D.H. Butler, K.M. Borland, J.E. Staruk, D.L. Faustman, B.A. Solomon, T.E. Muller, R.G. Rupp, T. Maki, A.P. Monaco, et al., Xenotransplantation of canine, bovine, and porcine islets in diabetic rats without immunosuppression, Proc. Natl. Acad. Sci. U. S. A. 88 (24) (1991) 11100–11104.
[56] P.E. Lacy, O.D. Hegre, A. Gerasimidi-Vazeou, F.T. Gentile, K.E. Dionne, Maintenance of normoglycemia in diabetic mice by subcutaneous xenografts of encapsulated islets, Science 254 (1991) 1782–1784.
[57] R.P. Lanza, A.M. Beyer, J.E. Staruk, W.L. Chick, Biohybrid artificial pancreas, Transplantation 56 (5) (1993) 1067–1072.
[58] D.W. Scharp, C.J. Swanson, B.J. Olack, P.P. Latta, O.D. Hegre, E.J. Doherty, F.T. Gentile, K.S. Flavin, M.F. Ansara, P.E. Lacy, Protection of encapsulated human islets implanted without immunosuppression in patients with type I or type II diabetes and in nondiabetic control subjects, Diabetes 43 (1994) 1167–1170.
[59] R.P. Lanza, A.M. Beyer, W.L. Chick, Xenogeneic humoral responses to islets transplanted in biohybrid diffusion chambers, Transplantation 57 (9) (1994) 1371–1375.
[60] P. Aebischer, M. Schluep, N. Déglon, J.-M. Joseph, L. Hirt, B. Heyd, M. Goddard, J.P. Hammang, A.D. Zurn, A.C. Kato, F. Regli, E.E. Baetge, Intrathecal delivery of CNTF

using encapsulated genetically modified xenogeneic cells in amyotrophic lateral sclerosis patients, Nat. Med. 2 (6) (1996) 696–699.
[61] D.W. Scharp, P. Marchetti, Encapsulated islets for diabetes therapy: history, current progress, and critical issues requiring solution, Adv. Drug Deliv. Rev. 67–68 (2014) 35–73.
[62] J.-L. Dulong, C. Legallais, A theoretical study of oxygen transfer including cell necrosis for the design of a bioartificial pancreas, Biotech. Bioeng. 96 (2007) 990–998.
[63] G. Erdodi, J. Kang, B. Yalcin, M. Cakmak, K.S. Rosenthal, S. Grundfest-Broniatowski, J.P. Kennedy, A novel macroencapsulating immunoisolatory device: the preparation and properties of nanomat-reinforced amphiphilic co-networks deposited on perforated metal scaffold, Biomed. Microdevices 11 (2009) 297–312.
[64] R. Storrs, R. Dorian, S.R. King, J. Lakey, H. Rilo, Preclinical development of the islet sheet, Ann. N. Y. Acad. Sci. 944 (2001) 252–266.
[65] S.R. King, R. Dorian, R.W. Storrs, Requirements for encapsulation technology and the challenges for transplantation of islets of Langerhans, Graft 4 (2001) 491–499.
[66] P. Gianello, D. Dufrane, Correction of a diabetes mellitus type 1 on primate with encapsulated islet of pig pancreatic transplant, Bull. Mem. Acad. R. Med. Belg. 162 (2007) 439–449.
[67] D. Dufrane, R.-M. Goebbels, P. Gianello, Alginate macroencapsulation of pig islets allows correction of Streptozotocin-induced diabetes in primates up to 6 months without immunosuppression, Transplantation 90 (10) (2010) 1054–1062.
[68] D. Dufrane, P.R.R. Gianello, J.E. Melvik, Alginate coated, collagen matrix, cellular device, preparative methods and uses thereof (2008), US Patent 8168215 B2
[69] S. Véríter, N. Aouassar, P.Y. Adnet, M.S. Paridaens, C. Stuckman, B. Jordan, O. Karroum, B. Gallez, P. Gianello, D. Dufrane, The impact of hyperglycemia and the presence of encapsulated islets on oxygenation within a bioartificial pancreas in the presence of mesenchymal stem cells in a diabetic Wistar rat model, Biomaterials 32 (26) (2011) 5945–5956.
[70] S. Véríter, P. Gianello, Y. Igarashi, G. Beaurin, A. Ghyselinck, N. Aouassar, B. Jordan, B. Gallez, D. Dufrane, Improvement of subcutaneous bioartificial pancreas vascularization and function by co-encapsulation of pig islets and mesenchymal stem cells in primates, Cell Transplant. (2013), http://dx.doi.org/10.3727/096368913X663550 (In Press).
[71] J.I. Lee, R. Nishimura, H. Saka, N. Sasaki, T. Kenmochi, A newly developed immunoisolated bioartificial pancreas with cell sheet engineering, Cell Transplant. 17 (2008) 51–59.
[72] J. Brauker, V. Carr-Brendel, L. Martinson, J. Crudele, W. Johnston, R.C. Johnson, Neovascularization of synthetic membranes directed by membrane microarchitecture, J. Biomed. Mater. Res. 29 (12) (1995) 1517–1524.
[73] S.F. Josephs, T. Loudovaris, A. Dixit, S.K. Young, R.C. Johnson, In vivo delivery of recombinant human growth hormone from genetically engineered human fibroblasts implanted with Baxter immunoisolation devices, J. Mol. Med. 77 (1999) 211–214.
[74] T. Loudovaris, S. Jacobs, S. Young, D. Maryanov, J. Brauker, R.C. Johnson, Correction of diabetic nod mice with insulinomas implanted within Baxter immunoisolation devices, J. Mol. Med. 77 (1999) 219–222.
[75] A. Tibell, E. Rafael, L. Wennberg, J. Nordenstrom, M. Bergstrom, R.L. Geller, T. Loudovaris, R.C. Johnson, J.H. Brauker, S. Neuenfeldt, A. Wernerson, Survival of macroencapsulated allogeneic parathyroid tissue one year after transplantation in nonimmunosuppressed humans, Cell Transplant. 10 (2001) 591–599.
[76] S.-H. Chen, S.-C. Huang, C.-C. Lui, T.-P. Lin, F.-F. Chou, J.-Y. Ko, Effect of theracyte-encapsulated parathyroid cells on lumbar fusion in a rat model, Eur. Spine J. 21 (2012) 1734–1739.
[77] X. Jia, A. Sharma, M. Kumagai-Braesch, A.M. Wernerson, A.K. Sörenby, S. Yamamoto, F. Wang, A.B. Tibell, Extendin-4 increases the expression of hypoxia-inducible factor 1α in rat islets and preserves the endocrine cell volume of both free and macroencapsulated islet grafts, Cell Transplant. 21 (2012) 1269–1283.
[78] M. Kumagai-Braesch, S. Jacobson, H. Mori, X. Jia, T. Takahashi, A. Wernerson, M. Flodström-Tullberg, A. Tibell, The TheraCyte™ device protects against islet allograft rejection in immunized hosts, Cell Transplant. 22 (7) (2013) 1137–1146.
[79] E. Rafael, G.S. Wu, K. Hultenby, A. Tibell, A. Wernerson, Improved survival of macroencapsulated islets of Langerhans by pre-implantation of the immunoisolating device: a morphometric study, Cell Transplant. 12 (2003) 407–412.
[80] A.K. Sorenby, M. Kumagai-Braesch, A. Sharma, K.R. Hultenby, A.M. Wernerson, A.B. Tibell, Pre-implantation of an immunoprotective device can lower the curative dose of islets to that of free islet transplantation: studies in a rodent model, Transplantation 86 (2008) 364–366.
[81] K.E. La Flamme, G. Mor, D. Gong, T. La Tempa, V.A. Fusaro, C.A. Grimes, T.A. Desai, Nanoporous alumina capsules for cellular macroencapsulation: transport and biocompatibility, Diabetes Technol. Ther. 7 (5) (2005) 684–694.
[82] S.F. Grundfest-Broniatowski, G. Tellioglu, K.S. Rosenthal, J. Kang, G. Erdodi, B. Yalcin, M. Cakmak, J. Drazba, A. Bennett, L. Lu, J.P. Kennedy, A new bioartificial pancreas utilizing amphiphilic membranes for the immunoisolation of porcine islets, ASAIO J. 55 (4) (2009) 400–405.
[83] J. Kang, G. Erdodi, J.P. Kennedy, H. Chou, L. Lu, S. Grundfest-Broniatowski, Toward a bioartificial pancreas: diffusion of insulin and IgG across immunoprotective membranes with controlled hydrophilic channel diameters, Macromol. Biosci. 10 (2010) 369–377.
[84] H. Wu, E.S. Avgoustiniatos, L. Swette, S. Bonner-Weir, G.C. Weir, C.K. Colton, In situ electrochemical oxygen generation with an immunoisolated device, Ann. N. Y. Acad. Sci. 875 (1999) 105–125.
[85] K. Bloch, E. Papismedov, K. Yaviryants, M. Vorobeychik, S. Beer, P. Vardi, Immobilized microalgal cells as an oxygen supply system for encapsulated pancreatic islets: a feasibility study, Artif. Organs 30 (9) (2006) 715–718.
[86] K. Bloch, E. Papismedov, K. Yavriyants, M. Vorobeychik, S. Beer, P. Vardi, Photosynthetic oxygen generator for bioartificial pancreas, Tissue Eng. 12 (2) (2006) 337–344.
[87] B. Ludwig, B. Zimerman, A. Steffen, K. Yavriants, D. Azarov, A. Reichel, P. Vardi, T. German, N. Shabtay, A. Rotem, Y. Evron, T. Neufeld, S. Mimon, S. Ludwig, S.R. Bornstein, U. Barkai, A novel device for islet transplantation providing immune protection and oxygen supply, Horm. Metab. Res. 42 (13) (2010) 918–922.
[88] B. Ludwig, A. Rotem, J. Schmid, G.C. Weir, C.K. Colton, M.D. Brendel, T. Neufeld, N.L. Block, K. Yavriyants, A. Steffen, S. Ludwig, T. Chavikis, A. Reichel, D. Azarov, B. Zimermann, S. Maimon, M. Balyura, T. Rozenschtein, N. Shabtay, P. Vardi, K. Bloch, P. de Vos, A.V. Schally, S.R. Bornstein, U. Barkai, Improvement of islet function in a bioartificial pancreas by enhanced oxygen supply and growth hormone release hormone agonist, Proc. Natl. Acad. Sci. U. S. A. 109 (13) (2012) 5022–5027.
[89] U. Barkai, G.C. Weir, C.K. Colton, B. Ludwig, S.R. Bornstein, M.D. Brendel, T. Neufeld, C. Bremer, A. Leon, Y. Evron, K. Yavriyants, D. Azarov, B. Zimermann, S. Maimon, N. Shabtay, M. Balyura, T. Rozenschtein, P. Vardi, K. Bloch, P. de Vos, A. Rotem, Enhanced oxygen supply improves islet viability in a new bioartificial pancreas, Cell Transplant. 22 (8) (2013) 1463–1476.
[90] T. Neufeld, B. Ludwig, U. Barkai, G.C. Weir, C.K. Colton, Y. Evron, M. Balyura, K. Yavriyants, B. Zimermann, D. Azarov, S. Maimon, N. Shabtay, T. Rozenshtein, D. Lorber, A. Steffen, U. Willenz, K. Bloch, P. Vardi, R. Taube, P. de Vos, E.C. Lewis, S.R. Bornstein, A. Rotem, The efficacy of an immunoisolation membrane system for islet xenotransplantation in minipigs, PLOS One 8 (8) (2013) e70150.
[91] B. Ludwig, A. Reichel, A. Steffen, B. Zimerman, A.V. Schally, N.L. Block, C.K. Colton, S. Ludwig, S. Kersting, E. Bonifacio, M. Solimena, Z. Gendler, A. Rotem, U. Barkai, S.R. Bornstein, Transplantation of human islets without immunosuppression, Proc. Natl. Acad. Sci. U. S. A. 110 (47) (2013) 19054–19058.
[92] Y. Evron, C.K. Colton, B. Ludwig, U. Barkai, G.C. Weir, B. Zimermann, S. Maimon, T. Neufeld, N. Shalev, T. Goldman, A. Leon, K. Yavriyants, N. Shabtay, T. Rozenshtein, A. Stefen, P. Vardi, K. Bloch, P. de Vos, S.R. Bornstein, A. Rotem, Viability and function of transplanted encapsulated islets at high density is maintained by enhanced oxygen supply, 2014. (submitted for publication).
[93] J.A. Emamaullee, A.M. Shapiro, Factors influencing the loss of beta-cell mass in islet transplantation, Cell Transplant. 16 (2007) 1–8.
[94] L.A. Slezak, D.K. Andersen, Pancreatic resection: effects on glucose metabolism, World J. Surg. 25 (4) (2001) 452–460.
[95] G.C. Weir, S. Bonner-Weir, Five stages of evolving beta-cell dysfunction during progression to diabetes, Diabetes 53 (Suppl. 3) (2004) S16–S21.
[96] B. Hellman, The frequency distribution of the number and volume of the islets of Langerhans in man. I. Studies on non-diabetic adults, Acta Soc. Med. Ups. 64 (1959) 432–460.
[97] A. Pisania, Development of Quantitative Methods for Quality Assessment of Islets of Langerhans, (Ph.D. Thesis) Mass. Institute of Technology, Cambridge, MA, May 2007.
[98] A.M. Davalli, Y. Ogawa, C. Ricordi, D.W. Scharp, S. Bonner-Weir, G.C. Weir, A selective decrease in the beta cell mass of human islets transplanted into diabetic nude mice, Transplantation 59 (6) (1995) 817–820.
[99] E.A. Ryan, J.R. Lakey, B.W. Paty, S. Imes, G.S. Korbutt, N.M. Kneteman, D. Bigam, R.V. Rajotte, A.M. Shapiro, Successful islet transplantation: continued insulin reserve provides long-term glycemic control, Diabetes 51 (7) (2002) 2148–2157.
[100] M.R. Rickels, M.H. Schutta, J.F. Markmann, C.F. Barker, A. Naji, K.L. Teff, β-Cell function following human islet transplantation for type 1 diabetes, Diabetes 54 (1) (2005) 100–106.
[101] A.M. Smink, M.M. Faas, P. de Vos, Toward engineering a novel transplantation site for human pancreatic islets, Prospect. Diabetes 62 (5) (2013) 1357–1364.
[102] T. Aoki, H. Hui, Y. Umehara, S. LiCalzi, A.A. Demetriou, J. Rozga, R. Perfetti, Intrasplenic transplantation of encapsulated genetically engineered mouse insulinoma cells reverses streptozotocin-induced diabetes in rats, Cell Transplant. 14 (2005) 411–421.
[103] N.E. Simpson, N. Khokhlova, J.A. Oca-Cossio, S.S. McMarlane, C.P. Simpson, I. Constantinidis, Effects of growth regulation on conditionally transformed alginate-entrapped insulin-secreting cell lines in vitro, Biomaterials 26 (2005) 4633–4641.
[104] E. Kroon, L.A. Martinson, K. Kadoya, A.G. Bang, O.G. Kelly, S. Eliazer, H. Young, M. Richardson, N.G. Smart, J. Cunningham, A.D. Agulnick, K.A. D'Amour, M.K. Carpenter, E.E. Baetge, Pancreatic endoderm derived from human embryonic stem cells generates glucose-responsive insulin-secreting cells in vivo, Nat. Biotechnol. 26 (2008) 443–452.
[105] L. Martinson, C. Green, E. Kroon, A. Agulnick, O. Kelly, E.E. Baetge, Encapsulation of pancreatic cells derived from human pluripotent stem cells, US Patent Publication (2013) US 8,425,928 B2.
[106] S.-H. Lee, E. Hao, A.Y. Savinov, I. Geron, A.Y. Strongin, P. Itkin-Ansari, Human β-cell precursors mature into functional insulin-producing cells in an immunoisolation device: implications for diabetes cell therapies, Transplantation 87((7) (2009) 983–991.
[107] O. Korsgren, J. Jansson, Porcine islet-like cell clusters cure diabetic nude rats when transplanted under the kidney capsule, but not when implanted into the liver or spleen, Cell Transplant. 3 (1994) 49–54.
[108] M.R. Clatworthy, B cell response to allograft — more common than we thought? Am. J. Transplant. 13 (2013) 1629–1630.
[109] J. Arribas, A. Borroto, Protein ectodomain shedding, Chem. Rev. 102 (2002) 4627–4638.
[110] S.P. Dello, E. Rovida, Transmodulation of cell surface regulatory molecules via ectodomain shedding, Biol. Chem. 383 (2002) 69–83.
[111] M.L. Albert, B. Sauter, N. Bhardwaj, Dendritic cells acquire antigen from apoptotic cells and induce class I-restricted CTLs, Nature 392 (1998) 86–89.
[112] S. Gallucci, P. Matzinger, Danger signals: SOS to the immune system, Curr. Opin. Immunol. 13 (2001) 114–119.

[113] Y. Shi, J.E. Evans, K.L. Rock, Molecular identification of a danger signal that alerts the immune system to dying cells, Nature 425 (2003) 516–521.
[114] B.A. O'Brien, B.V. Harmon, D.P. Cameron, D.J. Allan, Apoptosis is the mode of α-cell death responsible for the development of IDDM in the nonobese diabetic (NOD) mouse, Diabetes 46 (1997) 750–757.
[115] J.D. Trudeau, J.P. Dutz, E. Arany, D.J. Hill, W.E. Fieldus, D.T. Finegood, Neonatal β-cell apoptosis: a trigger for autoimmune diabetes? Diabetes 49 (2000) 1–7.
[116] S. Turley, L. Poirot, M. Hattori, C. Benoist, D. Mathis, Physiological β-cell death triggers priming of self-reactive T cells by dendritic cells in a type-1 diabetes model, J. Exp. Med. 198 (2003) 1527–1537.
[117] E. Sun, Cell death recognition model for the immune system, Med. Hypothesis 70 (3) (2008) 585–596.
[118] J. Brauker, L.A. Martinson, S.K. Young, R.C. Johnson, Local inflammatory response around diffusion chambers containing xenografts: nonspecific destruction of tissues and decreased local vascularization, Exp. Transplant. 61 (12) (1996) 1671–1677.
[119] I.R. Sweet, O. Yanay, L. Waldron, M. Gilbert, J.M. Fuller, T. Tupling, A. Lernmark, W.R. Osborne, Treatment of diabetic rats with encapsulated islets, J. Cell. Mol. Med. 12 (2008) 2644–2650.
[120] T. Loudovaris, B. Charlton, R.J. Hodgson, T.E. Mandel, Destruction of xenografts but not allografts within cell impermeable, membranes, Transplant. Proc. 24 (1992) 2291–2292.
[121] T. Loudovaris, T.E. Mandel, B. Charlton, $CD4^+$ T-cell mediated destruction of xenografts within cell-impermeable membranes in the absence of $CD8^+$ T-cells and B-cells, Transplantation 61 (1996) 1678–1684.
[122] A.W. McKenzie, H.M. Georgiou, Y. Zhan, J.L. Brady, A.M. Lew, Protection of xenografts by a combination of immunoisolation and a single dose of anti-CD4 antibody, Cell Transplant. 10 (2001) 183–193.
[123] K.S. Jones, M.V. Sefton, R.M. Gorcynski, In vivo recognition by the host adaptive immune system of microencapsulated xenogeneic cells, Transplantation 78 (10) (2004) 1454–1462.
[124] A. Sharma, A. Sorenby, A. Wernerson, S. Efendic, M. Kumagai-Braesch, A. Tibell, Extendin-4 treatment improves metabolic control after rat islet transplantation to athymic mice with streptozotocin-induced diabetes, Diabetologia 49 (2006) 1247–1253.
[125] H. Iwata, K. Kobayashi, T. Takagi, T. Oka, H. Yang, H. Amemiya, T. Tsuji, F. Ito, Feasibility of agarose microbeads with xenogeneic islets as a bioartificial pancreas, J. Biomed. Mater. Res. Part A 28 (1994) 1003–1011.
[126] R.P. Lanza, R. Jackson, A. Sullivan, J. Ringeling, C. McGrath, W. Kühtreiber, W.L. Chick, Xenotransplantation of cells using biodegradable microcapsules, Transplantation 67 (8) (1999) 1105–1111.
[127] S.A. Safley, J.A. Kapp, C.J. Weber, Proliferative and cytokine responses in CTLA4-Ig-treated diabetic NOD mice transplanted with microencapsulated neonatal porcine ICCs, Cell Transplant. 11 (2002) 695–705.
[128] D. Dufrane, R.M. Goebbels, A. Saliez, Y. Guiot, P. Gianello, Six month survival of microencapsulated pig islets and alginate biocompatibility in primates: proof of concept, Transplantation 81 (2006) 1345–1353.
[129] D. Dufrane, P. Gianello, Pig islet xenotransplantation into non-human primate model, Transplantation 86 (6) (2008) 753–760.
[130] D. Dufrane, P. Gianello, Macro- or microencapsulation of pig islets to cure type 1 diabetes, World J. Gastroenterol. 18 (47) (2012) 6885–6893.
[131] S. Vériter, P. Gianello, D. Dufrane, Bioengineered sites for islet cell transplantation, Curr. Diab. Rep. 13 (2013) 745–755.
[132] T. Maki, I. Otsu, J.J. O'Neil, K. Dunleavy, C.J.P. Mullon, B.A. Solomon, A.P. Monaco, Treatment of diabetes by xenogeneic islets without immunosuppression, Diabetes 45 (1996) 342–347.
[133] I.F. Tannock, Oxygen diffusion and the distribution of cellular radiosensitivity in tumors, Br. J. Radiol. 45 (1972) 515–524.
[134] K.E. Dionne, C.K. Colton, M.L. Yarmush, Effect of hypoxia on insulin secretion by isolated rat and canine islets of Langerhans, Diabetes 42 (1993) 12–21.
[135] N.A. Deters, R.A. Stokes, J.E. Gunton, Islet transplantation: factors in short-term islet survival, Arch. Immunol. Ther. Exp. (Warsz) 59 (6) (2011) 421–429.
[136] P.O. Carlsson, F. Palm, A. Andersson, P. Liss, Markedly decreased oxygen tension in transplanted rat pancreatic islets irrespective of the implantation site, Diabetes 50 (3) (2001) 489–495.
[137] R. Olsson, J. Olerud, U. Pettersson, P.-O. Carlsson, Increased numbers of low-oxygenated pancreatic islets after intraportal islet transplantation, Diabetes 60 (2011) 2350–2353.
[138] J. Lau, J. Henriksnas, J. Svensson, P.-O. Carlsson, Oxygenation of islets and its role in transplantation, Curr. Opin. Organ Transplant. 14 (6) (2009) 688–693.
[139] A.M. Davalli, Y. Ogawa, C. Ricordi, D.W. Scharp, S. Bonner-Weir, G.C. Weir, A selective decrease in the beta cell mass of human islets transplanted into diabetic nude mice, Transplantation 59 (6) (1995) 817–820.
[140] K.K. Papas, A. Pisania, H. Wu, G.C. Weir, C.K. Colton, A stirred microchamber for oxygen consumption rate measurements with pancreatic islets, Biotechnol. Bioeng. 87 (2007) 1071–1082.
[141] P. De Vos, J.F.M. Van Straaten, A.G. Nieuwenhuizen, M. de Groot, R.J. Ploeg, B.J. De Haan, R. Van Schilfgaarde, Why do microencapsulated islet grafts fail in the absence of fibrotic overgrowth? Diabetes 48 (7) (1999) 1381–1388.
[142] J. Schrezenmeir, J. Kirchgessner, L. Gero, L.A. Kunz, J. Beyer, W. Mueller-Kliesser, Effect of microencapsulation on oxygen distribution in islets organs, Transplantation 57 (1994) 1308–1314.
[143] M. de Groot, T.A. Schuurs, R. van Schilfgaarde, Causes of limited survival of microencapsulated pancreatic islet grafts, J. Surg. Res. 121 (2004) 141–150.
[144] M. De Groot, T.A. Schuurs, P.P.M. Keizer, S. Fekken, H.G.D. Leuvenink, R. Van Schilfgaarde, Response of encapsulated rat pancreatic islets to hypoxia, Cell Transplant. 12 (2003) 867–875.
[145] J.L. Dulong, C. Legallais, Contributions of a finite element model for the geometric optimization of an implantable bioartificial pancreas, Artif. Organs 26 (7) (2002) 583–589.
[146] J.L. Dulong, C. Legallais, A novel model of solute transport in a hollow fiber bioartificial pancreas based on a finite element method, Biotechnol. Bioeng. 78 (5) (2002) 576–582.
[147] J.L. Dulong, C. Legallais, What are the relevant parameters for the geometrical optimization of an implantable bioartificial pancreas, J. Biomed. Eng. 127 (7) (2005) 1054–1061.
[148] A.S. Lewis, R.J. Fisher, G.C. Weir, C.K. Colton, Improving oxygen supply to encapsulated cells and islets, in: J.-P. Halle, P. De Vos, L. Rosenberg (Eds.), Transworld Research Network, Kerala India, 2007, pp. 205–241.
[149] A.S. Johnson, R.J. Fisher, G.C. Weir, C.K. Colto, Oxygen consumption and diffusion in assemblages of respiring spheres: performance enhancement of a bioartificial pancreas, Chem. Eng. Sci. 64 (2009) 4470–4487.
[150] P. Buchwald, A local glucose-and oxygen concentration-based insulin secretion model for pancreatic islets, Theor. Biol. Med. Model. 8 (2011) 20–45.
[151] E.S. Avgoustiniatos, Oxygen Diffusion Limitations in Pancreatic Islet Culture and Immunoisolation, (Ph.D. Thesis) Mass. Institute of Technology, Cambridge, MA, 2002.
[152] A. Pisana, G.C. Weir, J.J. O'Neil, A. Omer, V. Tchipashvili, J. Lei, C.K. Colton, S. Bonner-Weir, Quantitative analysis of cell composition and purity of human pancreatic islet preparations, Lab. Invest. 90 (2010) 1661–1675.
[153] D. Lazard, P. Vardi, K. Bloch, Induction of beta-cell resistance to hypoxia and technologies for oxygen delivery to transplanted pancreatic islets, Diabetes Metab. Res. Rev. 28 (6) (2012) 475–484.
[154] D. Klein, M.M. Ribeiro, V. Mendoza, S. Jayaraman, N.S. Kenyon, A. Pileggi, R.D. Molano, L. Inverardi, C. Ricordi, R.L. Pastori, Delivery of Bcl-XL or its BH4 domain by protein transduction inhibits apoptosis in human islets, Biochem. Biophys. Res. Commun. 323 (2004) 473–478.
[155] R.I. Mahato, Gene expression and silencing for improved islet transplantation, J. Control. Release 140 (2009) 262–267.
[156] X. Wang, M. Meloche, C.B. Verchere, D. Ou, A. Mui, G.L. Warnock, Improving islet engraftment by gene therapy, J. Transplant. (2011), http://dx.doi.org/10.1155/2011/594851 ((2011), Article ID 594851, 7 pp).
[157] A. Hughes, C. Jessup, C. Drogemuller, D. Mohanasundaram, C. Milner, D. Rojas, G.R. Russ, P.T.H. Coates, Gene therapy to improve pancreatic islet transplantation for type 1 diabetes mellitus, Current. Diabetes Rev. 6 (5) (2010) 274–284.
[158] R.A. Stokes, K. Cheng, N. Deters, S.M. Lau, W.J. Hawthorne, P.J. O'Connell, J. Stolp, S. Grey, T. Loudovaris, T.W. Kay, H.E. Thomas, F.J. Gonzalez, J.E. Gunton, Hypoxia-inducible factor-1α (HIF-1α) potentiates β-cell survival after islet transplantation of human and mouse islets, Cell Transplant. 22 (2013) 253–266.
[159] S. Duncanson, A. Sambanis, Dual factor delivery of CXCL12 and exendin-4 for improved survival and function of encapsulated beta cells under hypoxic conditions, Biotech. Bioeng. 110 (2013) 2292–2300.
[160] J.F. Lo, Y. Wang, A. Blake, G. Yu, T.A. Harvat, H. Jeon, J. Oberholzer, D.T. Eddington, Islet preconditioning via multimodal microfluidic modulation of intermittent hypoxia, Analyt. Chem. 84 (2012) 1987–1993.
[161] K. Bloch, O. Bloch, I. Tarasenko, D. Lazard, M. Rapoport, P. Vardi, A strategy for the engineering of insulin producing cells with a broad spectrum of defense properties, Biomaterials 32 (7) (2011) 1816–1825.
[162] F. Wiegand, K.-D. Kroncke, V. Kolb-Bachofen, Macrophage-generated nitric oxide as cytotoxic factor in destruction of alginate-encapsulated islets, Transplantation 56 (1993) 1206–1212.
[163] S.Y. Chae, M. Lee, S.W. Kim, Y.H. Bae, Protection of insulin-secreting cells from nitric oxide-induced cellular damage by cross-linked hemoglobin, Biomaterials 25 (2004) 843–850.
[164] S.Y. Chae, Y.Y. Kim, S.W. Kim, Y.H. Bae, Prolonged glucose normalization of streptozotocin-induced diabetic mice by transplantation of rat islets coencapsulated with crosslinked hemoglobin, Transplantation 78 (3) (2004) 392–397.
[165] C.K. Colton, P. Stroeve, J. Zahka, Mechansim of hemogloblin facilitation of oxygen transport, J. Appl. Physiol. 35 (1973) 307–309.
[166] V. Nadithe, D. Mishra, Y.H. Bae, Poly(ethylene glycol) cross-linked hemoglobin with antioxidant enzymes protects pancreatic islets from hypoxic and free radical stress and extends islet functionality, Biotech. Bioeng. 109 (9) (2012) 2392–2401.
[167] J.I. Stagner, R.S. Seelan, R.N. Parthasarathy, K. White, Reduction of ischemic cell death in cultured islets of Langerhans by the induction of cytoglobin, Islets 1 (2009) 50–54.
[168] J.I. Stagner, R.S. Seelan, R.N. Parthasarathy, Maintenance of aerobic metabolism increases immunoisolated islet survival, Islets 3 (2011) 89–92.
[169] A. Leung, Y. Ramaswamy, P. Munro, G. Lawrie, L. Nielsen, M. Trau, Emulsion strategies in the microencapsulation of cells: pathways to thin coherent membranes, Biotechnol. Bioeng. 92 (1) (2005) 45–53.
[170] T. Zekorn, U. Siebers, A. Horcher, R. Schnettler, U. Zimmermann, R.G. Bretzel, K. Federlin, Alginate coating of islets of Langerhans: in vitro studies on a new method for microencapsulation for immuno-isolated transplantation, Acta Diabetol. 29 (1992) 41–45.
[171] S. Kizilel, A. Scavone, X. Liu, J.M. Nothias, D. Ostaega, P. Witkowski, M. Millis, Encapsulation of pancreatic islets within nano-thin functional polyethylene glycol coatings for enhanced insulin secretion, Tissue Eng. A 16 (7) (2010) 2217–2228.
[172] G.M. Cruise, O.D. Hegre, F.V. Lamberti, S.R. Hager, R. Hill, D.S. Scharp, J.A. Hubbell, In vitro and in vivo performance of porcine islets encapsulated in interfacially photopolymerized poly(ethylene glycol) diacrylate membranes, Cell Transplant. 8 (3) (1999) 293–306.

[173] H. Dong, T.M. Fahmy, S.M. Metcalfe, S.L. Morton, X. Dong, L. Inverardi, D.B. Adams, W. Gao, H. Wang, Immuno-isolation of pancreatic islet allografts using pegylated nanotherapy leads to long-term normoglycemia in full MHC mismatch recipient mice, PLOS One 7 (12) (2012) e50265.
[174] Y. Teramura, H. Iwata, Bioartificial pancreas: microencapsulation and conformal coating of islet of Langerhans, Adv. Drug Deliv. Rev. 62 (2010) 827–840.
[175] J.D. McMillan, D.I.C. Wang, Mechanisms of oxygen transfer enhancement during submerged cultivation in perfluorochemical-in-water dispersions, Ann. N. Y. Acad. Sci. 589 (1990) 283–300.
[176] D. Poncelet, R. Leung, L. Centomo, R.J. Neufeld, Microencapsulation of silicone oils within polyacrylamide–polyethylene membranes as oxygen carriers for bioreactor oxygenation, J. Chem. Technol. Biotechnol. 57 (1993) 253–263.
[177] T. Zekorn, U. Siebers, R.G. Bretzel, S. Heller, U. Meder, H. Ruttkay, U. Zimmermann, K. Federlin, Impact of the perfluorochemical FC43 on function of isolated islets, Horm. Metab. Res. 23 (1991) 302–303.
[178] E. Maillard, M.T. Juszcak, A. Clark, S.J. Hughes, D.R.W. Gray, Perfluorodecalin-enriched fibrin matrix for human islet culture, Biomaterials 32 (2011) 9282–9289.
[179] S. Matsumoto, Y. Kuroda, Perfluorocarbon for organ preservation before transplantation, Transplantation 74 (12) (2002) 1804–1809.
[180] S.F. Khattak, K.-S. Chin, S.R. Bhatia, S.C. Roberts, Enhancing oxygen tension and cellular function in alginate cell encapsulation devices through the use of perfluorocarbons, Biotechnol. Bioeng. 96 (2007) 156.
[181] K. Chin, S.F. Khattak, S.R. Bhatia, S.C. Roberts, Hydrogel–perfluorocarbon composite scaffold promotes oxygen transport to immobilized cells, Biotechnol. Prog. 24 (2008) 358.
[182] K.M. Gattás-Asfura, C.A. Fraker, C.L. Stabler, Perfluorinated alginate for cellular encapsulation, J. Biomed. Mater. Res. Part A 100 (8) (2012) 1963–1971.
[183] F. Goh, R. Long Jr., N. Simpson, A. Sambanis, Dual perfluorocarbon method to non-invasively monitor dissolved oxygen concentration in tissue engineered constructs in vitro and in vivo, Biotechnol. Prog. 27 (4) (2011) 1115–1125.
[184] C.A. Fraker, A.J. Mendez, C.L. Sabler, Complementary methods for the determination of dissolved oxygen content in perfluorocarbon emulsions and other solutions, J. Phys. Chem. B 115 (2011) 10547–10552.
[185] C.A. Fraker, A.J. Mendez, L. Inverardi, C. Ricordi, C.L. Stabler, Optimization of perfluoro nano-scale emulsions: the importance of particle size for enhanced oxygen transfer in biomedical applications, Colloids Surf. B: Biointerfaces 98 (2012) 26–35.
[186] A.S. Johnson, E. O'Sullivan, L.N. D'Aoust, A. Omer, S. Bonner-Weir, R.J. Fisher, G.C. Weir, C.K. Colton, Quantitative assessment of islets of Langerhans encapsulated in alginate, Tissue Eng. Part C 17 (4) (2011) 435–449.
[187] E.S. O'Sullivan, A.S. Johnson, A. Omer, J. Hollister-Lock, S. Bonner-Weir, C.K. Colton, G.C. Weir, Rat islet cell aggregates are superior to islets for transplantation in microcapsules, Diabetologia 53 (5) (2010) 937–945.
[188] N. Trivedi, G.M. Steil, C.K. Colton, S. Bonner-Weir, G.C. Weir, Improved vascularization of planar membrane diffusion devices following continuous infusion of vascular endothelial growth factor, Cell Transplant. 9 (2000) 115–124.
[189] S. Sigrist, A. Mechine-Neuville, K. Mandes, V. Calenda, S. Braun, G. Legeay, J.-P. Bellocq, M. Pinget, L. Kellser, Influence of VEGF on the viability of encapsulated pancreatic rat islets after transplantation in diabetic mice, Cell Transplant. 12 (2003) 627–635.
[190] S. Sigrist, A. Mechine-Neuville, K. Mandes, V. Calenda, G. Legaey, J.-P. Bellocq, M. Pinget, L. Kessler, Induction of angiogenesis in omentum with vascular endothelial growth factor: influence on the viability of encapsulated rat pancreatic islets during transplantation, J. Vasc. Res. 40 (2003) 359–367.
[191] S.K. Hunter, J.M. Kao, Y.E. Wang, J.A. Benda, V.G.J. Rodgers, Promotion of neovascularization around hollow fiber bioartificial organs using biologically active substances, ASAIO J. 45 (1) (1999) 37–40.
[192] H.K. Tilakaratne, S.K. Hunter, M.E. Andracki, J.A. Benda, V.G. Rodgers, Characterizing short-term release and neovascularization potential of multi-protein growth supplement delivered via alginate hollow fiber devices, Biomaterials 28 (1) (2007) 89–98.
[193] O. Khanna, M.L. Moya, E.C. Opara, E.M. Brey, Synthesis of multilayered alginate microcapsules for the sustained release of fibroblast growth factor-1, J. Biomed. Mater. Res. Part A 95A (2010) 632–640.
[194] O. Khanna, J.-J. Huang, M.L. Moya, C.-W. Wu, M.-H. Cheng, E.C. Opara, E.M. Brey, FGF-1 delivery from multilayer alginate microbeads stimulates a rapid and persistent increase in vascular density, Microvasc. Res. 90 (2013) 23–29.
[195] J.P. McQuilling, J. Arenas-Herrera, C. Childers, R.A. Pareta, O. Khanna, B. Jiang, E.M. Brey, A.C. Farney, E.C. Opara, New alginate microcapsule system for angiogenic protein delivery and immunoisolation of islets for transplantation in the rat omentum pouch, Transplant. Proc. 43 (2011) 3262–3264.
[196] T. Sakurai, A. Satake, N. Nagata, Y. Gu, A. Hiura, K. Doo-Hoon, H. Hori, Y. Tabata, S. Sumi, K. Inoue, The development of new immunoisolatory devices possessing the ability to induce neovascularization, Cell Transplant. 12 (2003) 527–535.
[197] T. Sakurai, A. Satake, S. Sumi, K. Inoue, N. Nagata, Y. Tabata, M. Junji, The efficient prevascularization induced by fibroblast growth factor 2 with a collagen-coated device improves the cell survival of a bioartificial pancreas, Pancreas 28 (3) (2004) e70–e79.
[198] T. Yuasa, J.D. Rivas-Carrillo, N. Navarro-Alvarez, A. Soto-Gutierrez, Y. Kubota, Y. Tabata, T. Okitsu, H. Noguchi, S. Matsumoto, S. Nakaji, N. Tanaka, N. Kobayashi, Neovascularization induced around an artificial device implanted in the abdomen by the use of a gelatinized fibroblast growth factor 2, Cell Transplant. 18 (2009) 683–688.
[199] A. Miki, J.D. Rivas-Carrillo, N. Navarro-Alvarez, A. Soto-Gutierrez, Y. Chen, K. Tanaka, M. Narushima, Y. Tabata, T. Okitsu, H. Noguchi, S. Matsumoto, N. Tanaka, N. Kobayashi, Maintenance of neovascularization at the implantation site of an artificial device by bFGF and endothelial cell transplant, Cell Transplant. 15 (2006) 893–901.
[200] H.J.H. Marti, M. Bernaudin, A. Bellail, H. Schoch, M. Euler, E. Petit, W. Risau, Hypoxia-induced vascular endothelial growth factor expression precedes neovascularization after cerebral ischemia, Am. J. Pathol. 156 (3) (2000) 965–976.
[201] B.L. Krock, N. Skuli, M.C. Simon, Hypoxia-induced angiogenesis: good and evil, Genes Cancer 2 (12) (2011) 1117–1133.
[202] S. Oladipupo, S. Hu, J. Kovalski, J. Yao, A. Santeford, R.E. Sohn, R. Shohet, K. Maslov, L.V. Wang, J.M. Arbeit, VEGF is essential for hypoxia-inducible factor-mediated neovascularization but dispensable for endothelial sprouting, PNAS 108 (32) (2011) 13264–13269.
[203] M. Bensellam, B. Duvillíe, G. Rybachuk, D.R. Laybutt, C. Magnan, Y. Guiot, J. Pouysségur, J.-C. Jonas, Glucose-induced O_2 consumption activates hypoxia inducible factors 1 and 2 in rat insulin-secreting pancreatic beta-cells, PLOS One 7 (2012) e29807.
[204] J. Cantley, S.T. Grey, P.H. Maxwell, D.J. Withers, The hypoxia response pathway and β-cell function, Diabetes Obes. Metab. 12 (2) (2010) 159–167.
[205] S. Vériter, N. Aouassar, P.-Y. Adnet, M.-S. Paridaens, C. Stuckman, B. Jordan, O. Karroum, B. Gallez, P. Gianello, D. Dufrane, The impact of hyperglycemia and the presence of encapsulated islets on oxygenation within a bioartificial pancreas in the presence of mesenchymal stem cells in a diabetic Wistar rat model, Biomaterials 32 (2011) 5945–5956.
[206] N.C. Hunt, R.M. Shelton, D.J. Henderson, L.M. Grover, Calcium-alginate hydrogel-encapsulated fibroblasts provide sustained release of vascular endothelial growth factor, Tissue Eng. Part A 19 (2013) 905–914.
[207] J.J. Vallbacka, M.V. Sefton, Vascularization and improved in vivo survival of VEGF-secreting cells microencapsulated in HEMA-MMA, Tissue Eng. 13 (2007) 2259–2269.
[208] J. Lee, K.Y. Lee, Local and sustained vascular endothelial growth factor delivery for angiogenesis using an injectable system, Pharm. Res. 26 (7) (2009) 1739–1744.
[209] J.M. Chan, I.K. Zervantonakis, T. Rimchala, W.J. Polacheck, J. Whisler, R.D. Kamm, Engineering of in vitro 3D capillary beds by self-directed angiogenic sprouting, PLOS One 7 (12) (2012) e50582.
[210] I. Sukmana, Bioactive polymer scaffold for fabrication of vascularized engineering tissue, J. Artif. Organs 15 (2012) 215–224.
[211] A.-C. Brady, M.M. Martino, E. Pedraza, S. Sukert, A. Pileggi, C. Ricordi, J.A. Hubbell, C.L. Stabler, Proangiogenic hydrogels within macroporous scaffolds enhance islet engraftment in an extrahepatic site, Tissue Eng. A 19 (23–24) (2013) 2544–2552.
[212] E.A. Phelps, D.M. Headen, W.R. Taylor, P.M. Thulé, A.J. García, Vasculogenic bio-synthetic hydrogel for enhancement of pancreatic islet engraftment and function in type 1 diabetes, Biomaterials 34 (2013) 4602–4611.
[213] Y. Cao, A. Hong, H. Schulten, M.J. Post, Update on therapeutic neovascularization, Cardiovasc. Res. 65 (2005) 639–648.
[214] M. Lovett, K. Lee, A. Edwards, D.L. Kaplan, Vascularization strategies for tissue engineering, Tissue Eng. Part B 15 (3) (2009) 353–370.
[215] E.A. Phelps, A.J. Garcia, Engineering more than a cell: vascularization strategies in tissue engineering, Curr. Opin. Biotechnol. 21 (5) (2010) 704–709.
[216] J.C.-Y. Chung, D. Shum-Tim, Neovascularization in tissue engineering, Cells 1 (2012) 1246–1260.
[217] J.G. Nemeno-Guanzon, S. Lee, J.R. Berg, J.H. Jo, J.E. Yeo, B.M. Nam, Y.-G. Koh, J.I. Lee, Trends in tissue engineering for blood vessels, J. Biomed. Biotechnol. 2012 (2012) 1–14.
[218] I. Sukmana, Microvascular guidance: a challenge to support the development of vascularized tissue engineering construct, Sci. World J. 2012 (2012) 1–10.
[219] F.A. Auger, L. Gibot, D. Lacroix, The pivotal role of vascularization in tissue engineering, Annu. Rev. Biomed. Eng. 15 (2013) 177–200.
[220] I. Yakhneco, W.K. Wong, I.I. Katkov, P. Itkin-Ansari, Cryopreservation of human insulin expressing cells macro-encapsulated in a durable therapeutic immunoisolating device Theracyte, Cryo Letters 33 (6) (2012) 518–531.
[221] M. Kumagai-Braesch, S. Jacobson, H. Mori, X. Jai, T. Takahashi, A. Wernerson, M. Flodstr m-Tullberg, A. Tibel, The TheraCyte™ device protects against islet allograft rejection in immunized hosts, Cell Transplant. 22(7) 1137–1146
[222] L. Krishnan, L.R. Clayton, E.D. Boland, R.M. Reed, J.B. Hoying, S.K. Williams, Cellular immunoisolation for islet transplantation by a novel dual porosity electrospun membrane, Transplant. Proc. 43 (9) (2001) 3256–3261.
[223] A. Pileggi, R.D. Molano, C. Ricordi, E. Zahr, J. Collins, R. Valdes, L. Inverardi, Reversal of diabetes by pancreatic islet transplantation into a subcutaneous, neovascularized devise, Transplantation 15;81 (9) (2006) 1318–1324.
[224] R.F. Padera, C.K. Colton, Time course of membrane microarchitecture-driven neovascularization, Biomaterials 17 (3) (1996) 277–284.
[225] E. Rafael, B. Gazelius, G.S. Wu, A. Tibell, Longitudinal studies on the microcirculation around the TheraCyte immunoisolation device, using the laser Doppler technique, Cell Transplant. 9 (1) (2000) 107–113.
[226] J.M. Anderson, Inflammatory response to implants, Trans. Am. Soc. Artif. Inter. Organs 34 (1988) 101–107.
[227] T.D. Zekorn, A. Horcher, J. Mellert, U. Siebers, T. Altug, A. Emre, H.J. Hahn, K. Federlin, Biocompatibility and immunology in the encapsulation of islets of Langerhans (bioartificial pancreas), Int. J. Artif. Organs 19 (4) (1996) 251–257.
[228] U. Siebers, A. Horcher, R.G. Bretzel, K. Federlin, T. Zekorn, Alginate-based microcapsules for immunoprotected islet transplantation, Ann. N. Y. Acad. Sci. 831 (1997) 304–312.
[229] A.W. Bridges, A.J. García, Anti-inflammatory polymeric coatings for implantable biomaterials and devices, J. Diabetes Sci. Technol. 2 (6) (2008) 984–994.
[230] M. Intaglietta, P.C. Johnson, R.W. Winslow, Microvascular and tissue oxygen distribution, Cardiovasc. Res. 32 (1996) 632–643.

[231] W. Wang, P. Vadgama, O_2 microsensors for minimally invasive tissue monitoring, J. R. Soc. Interface 1 (2004) 109–117.
[232] F.L. Huber, T.D. Latshang, J.S. Goede, K.E. Bloch, Does venous blood gas analysis provide accurate estimates of hemoglobin oxygen affinity? Ann. Hematol. 92 (4) (2013) 517–521.
[233] S.J. Hughes, S.E. Davies, S.H. Powis, M. Press, Hyperoxia improves the survival of intraportally transplanted syngeneic pancreatic islets, Transplantation 75 (12) (2003) 1954–1959.
[234] Z. Ma, N. Moruzzi, S.-B. Catrina, V. Grill, A. Björklund, Hyperoxia inhibits glucose-induced insulin secretion and mitochondrial metabolism in rat pancreatic islets, Biochem. Biophys. Res. Commun. 443 (1) (2014) 223–228.
[235] E. Pedraza, M.M. Coronel, C.A. Fraker, C. Ricordi, C.L. Stabler, Preventing hypoxia-induced cell death in beta cells and islets via hydrolytically activated, oxygen-generating biomaterials, Proc. Natl. Acad. Sci. U. S. A. 109 (11) (2012) 4245–4250.
[236] E. Pedraza, A.C. Brady, C.A. Fraker, C.L. Stabler, Synthesis of macroporous poly(dimethylsiloxane) scaffolds for tissue engineering applications, J. Biomater. Sci. Polym. Ed. 24 (9) (2013) 1041–1056.
[237] C.L. Ward, B.T. Corona, J.J. Yoo, B.S. Harrison, G.J. Christ, Oxygen generating biomaterials preserve skeletal muscle homeostasis under hypoxic and ischemic conditions, PLOS One 8 (8) (2013) e72485.
[238] M.M. Coronel, C.L. Stabler, Engineering a local microenvironment for pancreatic islet replacement, Curr. Opinion Biotechnol. 24 (50) (2013) 900–908.
[239] C.K. Colton, K.K. Papas, A. Pisania, M.J. Rappel, D.E. Powers, A. Omer, G. Weir, S. Bonner-Weir, Characterization of islet preparation, in: C. Halberstadt, D.F. Emerich (Eds.), Cellular Transplantation from Laboratory to Clinic, Elsevier, Inc., New York, 2007, pp. 85–134.