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TOPIC HIGHLIGHT

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Macro- or microencapsulation of pig islets to cure type 1 diabetes

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Abstract

Although allogeneic islet transplantation can successfully cure type 1 diabetes, it has limited applicability. For example, organs are in short supply; several human pancreas donors are often needed to treat one diabetic recipient; the intrahepatic site may not be the most appropriate site for islet implantation; and immunosuppressive regimens, which are associated with side effects, are often required to prolong survival of the islet graft. An alternative source of insulin-producing cells would therefore be of major interest. Pigs represent a possible alternative source of beta cells. Grafting of pig islets may appear difficult because of the immunologic species barrier, but pig islets have been shown to function in primates for at least 6 mo with clinically incompatible immunosuppression. Therefore, a bioartificial pancreas made of encapsulated pig islets may resolve issues associated with islet allotransplantation. Although several groups have shown that encapsulated pig islets are functional in small-animal models, less is known about the use of bioartificial pancreases in large-animal models. In this review, we summarize current knowledge of encapsulated pig islets, to determine obstacles to implantation in humans and possible solutions to overcome these obstacles.

Key words: Cell transplantation; Diabetes mellitus type 1; Islets of Langerhans; Porcine; Xenografts

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INTRODUCTION

Type 1 diabetes has been treated successfully by transplanting islets of Langerhans, the endocrine tissue that releases insulin[1]. Despite the clinical efficacy of islet transplantation, serious issues preclude its broad clinical application, including the side effects of chronic immunosuppressive regimens and a shortage of human donors. For example, it was recently estimated that only one pancreas is available per 333 patients with type 1 diabetes in the United States[2]. This situation is aggravated by the need of each recipient undergoing transplantation

for 2-4 pancreases[1,3]. This shortage of pancreas donors therefore justifies the search for alternative sources of insulin-producing cells. Swine appear to be the major candidates for islet procurement because: (1) humans have been treated with porcine insulin for > 40 years (pig and human insulin differ by only one amino acid); (2) pigs have large litters with offspring that rapidly attain adult size and are therefore amenable to genetic engineering; (3) pig pancreases contain large islets that respond to glucose stimulation; and (4) since pigs are widely bred and slaughtered for food, the use of their islets to restore human health may be an option that could satisfy sociocultural and ethical concerns[4-6].

Unlike primarily vascularized organs, pancreatic islets are implanted without direct connection to the host vascular network, with 7 to 14 d needed to re-establish blood flow[7-12]. Thus, it was thought that pig islet xenografts could escape typical hyperacute rejection and acute vascular rejection[13]. *In vivo*, however, pig islets in non-immunosuppressed nonhuman primates are rejected by both humoral and cellular immune reactions[14-16]. Diffuse, presumably nonspecific IgG deposits were observed within islet-associated accumulations of platelets 12 h after transplantation. Deposits of large amounts of IgM and moderate to large amounts of C3, C5, and C9 were present on islet surfaces 2 to 3 d after xenografting[15-17]. Anti-galactosyl (anti-Gal) and non-Gal[18,19] IgM antibodies bind to islet surfaces soon after transplantation and activate the classical complement pathway, as well as promoting neutrophil infiltration. These humoral immune responses to pig islets are consistent with early T-cell-independent immune system activation and are reminiscent of mechanisms that operate during the hyperacute rejection of solid-organ xenografts[20]. Humans and nonhuman primates have preformed anti-pig antibodies that rapidly recognize the Gal epitope on islet endothelial cells. During pig islet-to-primate xenotransplantation, however, the expression of Gal epitopes is influenced by the age of the pig. Gal residues are present on 20% of fetal, but on only 5.1% of adult, islet β cells[21-23]. Since Gal expression persists after islet isolation[24,25], Gal remains a target for humoral xenorejection.

Islet xenografts that survive immediate blood-mediated inflammatory reactions and additional humoral damage will be subject to acute cellular xenograft rejection. Following transplantation of fetal pig islets under the kidney capsule, the cellular infiltrate in primates has been found to consist mainly of CD8 T cells (implicating the indirect pathway), whereas the cellular infiltrate in rodents was dominated by macrophages. T-cell infiltration precedes macrophage influx, with small numbers of $CD3^+$ T cells observed 12 h after transplantation[14]. After 24 h, equal numbers of $CD3^+$ T cells and neutrophils were observed, and after 72 h, $CD3^+$ T cells dominated, representing 50% to 80% of all infiltrating cells. After 72 h, large numbers of macrophages were observed, with T cells localized at the periphery of and within transplanted islets. In addition, increased E-selectin expression on portal vein endothelial cells correlated with the infiltration of neutrophils, which caused tissue damage by releasing enzymes, active oxygen intermediates, and proinflammatory cytokines, and produced chemokines that attracted dendritic cells and T lymphocytes. Pig islet xenorejection seems theoretically easier to overcome, but because hyperacute and acute vascular rejections do not occur, the rapid destruction of pig islets within 72 h of transplantation into nonhuman primates demonstrates the strength of xenorejection.

Thus, an immunosuppressive regimen is mandatory for the long-term survival of pig islets in primates[26,27]. Although several immunosuppressive strategies have successfully suppressed alloimmune responses, T-cell-mediated xenoimmune responses have proven more resistant to immunosuppressive therapy[28,29]. This may be due to the greater molecular incompatibility between donor and recipient, which activates particularly the innate immune response[30].

Until recently, the maximum reported duration of pig islet survival (insulin-positive cells, no function) following transplantation under the kidney capsule in non-diabetic cynomolgus monkeys and immunosuppression with anti-thymocyte globulin, anti-interleukin-2R mAb, cyclosporine, and steroids was 53 d[31]. In March 2006, however, two studies reported that neonate or adult pig islets xenotransplanted into primates survived and functioned for > 180 d[26,27]. More recently, the transgenic expression of a human complement-regulatory protein (hCD46) on porcine islets was shown to enhance the survival of islets xenotransplanted into cynomolgus monkeys with streptozotocin (STZ)-induced diabetes for > 12 mo[32]. In addition, transplantation of galactosyl knock-out neonate pig islets was found to significantly enhance normoglycemia rates in diabetic primates, likely due to the decreased susceptibility of these xenografts to innate immunity mediated by complement and preformed xenoantibodies[33]. These results, however, required treatment with a heavy immunosuppressive regimen, in particular an anti-CD154-specific mAb, an antibody that induced thromboembolic events precluding its clinical use[34]. Despite the unacceptability of these immunosuppressive regimens in humans, these results are very encouraging since an alternative, nontoxic regimen combined with xenotransplantation of pig islets may induce normoglycemia in diabetic patients.

A bioartificial pancreas, in which islets of Langerhans are encapsulated within a semipermeable membrane, may be an alternative therapeutic device for patients with insulin-dependent (type 1) diabetes mellitus. It may constitute a safe and simple method of transplanting islets without the need for immunosuppressive therapy. Since the semipermeable membrane protects the islets from the host immune system, the islets are likely to survive and release insulin for a long period of time, thereby controlling glucose metabolism in the absence of immunosuppressive medication. Nevertheless, several important questions are associated with the transplanta-

Table 1 Bioartificial device configurations for encapsulation of pig islets

	Macroencapsulation	Microencapsulation	Conformal coating
Type of pig islets tested	Adult/neonate	Adult/neonate	Adult
Suitable material with biocompatibility	Alginate	Alginate with/ without PLL/PLO over-layer	PEG
Implantation site	Intraperitoneal	Intraperitoneal	Into the liver *via* the portal vein
	Subcutaneous	Kidney subcapsular space	
Proof of concept in preclinical studies in diabetic primates	+	+	–
Maximum survival of pig islets and diabetes correction	Mean 24 wk correction of glycated hemoglobin < 7% after transplantation of the MCD into subcutaneous tissues	Max 804 d but never reproduced; Biocompatibility confirmed under the kidney capsule up to 6 mo after transplantation	ND
Clinical study	+	+	ND
Clinical efficacy	No insulin-independence	No insulin independence	ND
	Glycated hemoglobin improvement; porcine insulin detected in recipient sera up to 4 yr after transplantation with insulin positive cells into hollow-fiber devices.	Survival up to 9.5 yr with insulin-positive cells after transplantation into the peritoneum and detection of urinary porcine C-peptide up to 11 mo after transplantation	
Advantages for large-scale clinical application	Easy procedure for transplantation into subcutaneous tissue	Easy procedure for transplantation by simple injection into the peritoneum	- Capability of transplantation into the liver
	Simple procedure to remove the graft from subcutaneous tissue	Large scale encapsulation of large number of islets	- Reduction of graft size
Limitations for large-scale clinical application	Limited islet oxygenation	Large volume of encapsulated islets limiting transplantation into the peritoneum	- No stability for long-term islet immunoprotection
	Difficulty to transplant into the peritoneal cavity	Islet survival limited by absence of biologic interaction with encapsulation material	
		No ability to remove the graft after transplantation	

PLL: Poly-lysine; PLO: Poly-ornithine; MCD: Monolayer cellular device; ND: Not determined; PEG: Polyethylene glycol.

tion of immunoisolated adult pig islets as a "bioartificial pancreas"[35] (Table 1).

VARIATIONS IN CAPSULE SIZE: MACROENCAPSULATION OR MICROENCAPSULATION

Macroencapsulation

In the first reports of encapsulation, a large number of islets were immunoisolated between flat-sheet double membranes[36,37]. This type of single macroencapsulation device could be implanted with minimal surgery at different sites, including the peritoneal cavity, subcutaneously, or under the renal capsule. Although several types of biomaterial have been used to produce microcapsules, including nitrocellulose, alginate, acrylonitrile, and agarose, these devices usually had some toxicity and activated nonspecific foreign body reactions, resulting in fibrotic overgrowth with subsequent necrosis of the encapsulated tissue[35]. A subcutaneously transplanted microdevice (TheraCyte device, Baxter Healthcare), 4 cm in length, shaped like a teabag, and made of a bilayered polytetrafluoroethylene membrane, was recently found to be biocompatible[38,39]. Neonatal pig cells inside the graft (i.e., cells immunohistochemically positive for insulin and glucagon) remained viable for up to 8 wk after xenotransplantation into nondiabetic cynomolgus monkeys, with no evidence of reaction with adjacent subcutaneous tissue[40]. Moreover, one of 12 non-immuno- suppressed adolescents became insulin independent and 5 children had reduced insulin requirement after transplantation of porcine islets encapsulated in hollow-fibers with porcine Sertoli cells, which likely have immunomodulating properties[41,42].

A "monolayer" configuration of macroencapsulated pig islets (monolayer cellular device) implanted subcutaneously (see below) has been found to significantly improve diabetes control (glycated hemoglobin < 7%) in primates for 6 mo without any immunosuppression[43]. In this encapsulation system, islets were seeded as a monolayer on an acellular collagen matrix, enhancing their interactions with a biologic membrane and increasing islet concentration per unit surface area. In addition, diabetes was controlled for up to 1 year in 2 diabetic primates after retransplantation with new monolayer cellular devices. Unfortunately, the lifespan of adult pig islets limited long-term graft function. Diabetic control was completely maintained for > 32 wk after the cotransplantation of adult pig islets and adipose mesenchymal stem cells[44]. A phase 1 clinical study is currently ongoing to assess the safety and efficacy of this device for allotransplantation of encapsulated islets into humans.

Microencapsulation

Another approach consists of the microencapsulation

of 1-3 islets per semipermeable immunoprotective capsule. The spherical configuration of these microcapsules resulted in a higher surface-to-volume ratio than did the tube or disk geometry of microcapsules, resulting in a higher diffusion rate[45]. Furthermore, microcapsules can be injected in large numbers, are durable and are difficult to disrupt mechanically[46,47].

Two recent studies describing transplantation of microencapsulated neonatal pig islets in an alginate matrix confirmed their biocompatibility in nondiabetic monkeys as well as their capacity to partially regulate diabetes[39,40]. Several protocols must be followed to increase the survival of alginate microencapsulated pig islets for up to 6 mo without immunosuppression in nondiabetic primates[25]: (1) Before transplantation, the islets should be cultured in medium containing 1.8 mmol $CaCl_2$; (2) Animal serum should be omitted from the culture medium; and (3) The graft should be composed of > 90% well-shaped capsules. Some of these islets survived for > 6 mo and were able to respond *in vitro* to glucose challenge 135 and 180 d after implantation. In addition, the implantation site (peritoneum, kidney capsule, or subcutaneous space) must be suitable[48,49], with the subcutaneous space considered a good choice for clinical applications[50].

Following transplantation of microencapsulated adult pig islets into spontaneously diabetic cynomolgus monkeys, blood glucose became normalized and the monkeys became insulin independent for periods ranging from 120 to 804 d[51]. Although these results were encouraging for the clinical application, they may have been dependent on the diabetic status of the recipient, the exact formulation of the capsules, and the immune response against pig islets (see below). To date, these results have been confirmed by only two casuistic manuscripts describing xenotransplantation in primates of microencapsulated neonatal pig islets[39,40]. One study confirmed the biocompatibility, for up to 8 wk, after transplantation of encapsulated pig islets in nondiabetic animals, and the second demonstrated that these microcapsules could regulate the diabetic state of diabetic recipients. Although the latter showed that daily exogenous insulin requirements were reduced by a mean of 43% compared with control animals transplanted with empty capsules, neither group showed changes in weekly blood glucose levels[39]. The absence of solid consistent data on glucose metabolism (e.g., changes in glycosylated hemoglobin concentration, glycosuria, intravenous glucose tolerance testing) renders this casuistic study difficult to interpret.

Living cell technologies (LCT) showed that porcine islet cells had survived and insulin production was maintained in a human patient 10 years after transplant of pig islet cells[52]. These findings demonstrated the long-term safety, viability, and functionality of encapsulated porcine islets in a human patient, without the use of immunosuppression. In 1996, a 41-year-old patient with diabetes was injected with pig islet cells to help regulate his blood glucose levels and control his diabetes. This patient's insulin requirement was reduced by 34% for over one year. Ten years later, the patient was still obtaining benefit from the transplant, and laparoscopic examination revealed living and functioning pig islet cells in his abdomen.

Two phase 1 trials have shown that intraperitoneally infused microencapsulated human islets can be considered safe for up to 3 years[53,54]. Although insulin independence was not achieved, glycemic control was improved, with a reduction in insulin daily requirement. In 2007, LCT launched a phase 1/2a study in Moscow of encapsulated neonatal insulin-producing porcine pancreatic islet cells (commercially called DIABECELL®). Seven patients with insulin-dependent diabetes have received between one and three implants of DIABECELL® (5000 and 10 000 IEQ/kg), with none showing marked adverse events 18 to 96 wk after transplantation. At last follow-up, the blood glucose concentrations in 5 patients were within the normal range (5.8-8.2 mmol/L). Two patients have shown excellent responses and do not require exogenously administered insulin. All recipients showed improvements in diabetes control, with lower glycated hemoglobin (% HbA1c) concentrations.

Following the successful completion of this phase 1/2a clinical trial in Russia, LCT launched phase 2b clinical trials, which are currently underway in New Zealand and Argentina. In contrast, although diabetes control was achieved by repeated injections of encapsulated islets (up to 4 infusions and up to 779 000 islets equivalent), a humoral response was induced, with cytotoxic antibodies found in the recipient sera 4 to 8 wk after transplantation, and necrosis of the islets at 16 mo[54].

Conformal coating of cell surfaces

A serious issue remains in using microcapsules as bioartificial pancreases, namely, the increase in total volume of the implant after microencapsulation. The average diameter of islets is roughly 150 μm, making the average diameter of capsules about three times as large and the total volume of microcapsules about 27 times as large. In clinical settings, the volume of islet suspensions is 10 mL, making it > 270 mL after microencapsulation. A site to implant such a large volume is difficult to find.

Much effort has been made to reduce the size of the capsules. For example, smaller microcapsules, about 300 μm in diameter, could result in a reasonable total volume for clinical application[55]. In clinical allotransplantation programs, islets are transplanted into the liver through the portal veins, and capsules of diameter larger than the islets themselves may plug larger blood vessels, resulting in severe thrombosis of the liver. The diameter of the encapsulated islets should therefore be much smaller than at present, to allow for their transplantation through the portal veins. A technique to coat islets with a very thin membrane or with conformal coating may reduce the diameter of these microcapsules, allowing their transplantation into the liver through the portal veins.

Several types of coating have been tested to im-

munoisolate islets from the host immune systems. For example, the surface of islets has been modified with thin membranes made of amphiphilic polymers, such as polyethylene glycol (PEG)-conjugated phospholipid (PEG-lipid) and polyvinyl alcohol carrying long alkyl chains[56-59]. The thickness of the PEG layer formed on the cell surface was several nanometers, but depended on the molecular weight of the PEG.

Surface modification did not change the morphology or viability of the islets[60]. Transplantation of 5000 porcine islets modified with PEG-N-hydroxysuccinimide (NHS) ester into the livers of NOD-severe combined immunodeficiency mice through the portal vein resulted in the transient normalization of blood glucose concentrations, but these concentrations later increased. The surface of islets covered with PEG reacted with the amino groups of the collagen layer remaining on the islet surface[61]. Although transplantation of islets covered with a PEG-NHS-modified surface into recipient rats treated with low-dose cyclosporine resulted in the maintenance of normoglycemia for 1 year, normoglycemia was maintained for only 11 d in the absence of cyclosporine despite the surface modification with PEG.

Although a conformal PEG layer may form on the cells or islet surfaces at the nanometer level and this method enables a drastic reduction in total graft volume compared with conventional microcapsules, the PEG layer on the islets was not stable and disappeared from the cell surface over 3 d.

Use of a layer-by-layer method may enhance the stability of PEG-lipid membranes on the cell surface. Various functional groups, such as maleimide and biotin, can be easily introduced to the end of the PEG chain of PEG-lipids[58] and can be used as reaction points to form multilayer membranes on the cell surface. A layer-by-layer membrane can also be formed by the reaction between biotin and streptavidin. Biotin-PEG-lipids are anchored to the cell membranes of islets and are further covered by streptavidin. The modified islets can be alternatively exposed to a biotin-bovine serum albumin conjugate solution and a streptavidin solution to form 20 layers. The thickness of the membrane is approximately 30 nm. A glucose stimulation test demonstrates the ability of the modified islets to control insulin release in response to changes in glucose concentration. After intraportal transplantation of modified islets with PEG-lipid into STZ-induced diabetic mice[59], most islets were not damaged and remained intact in the blood vessels of the liver for 1 h to 1 d after transplantation.

VARIATION OF MATERIALS

The choice of material to use for cellular encapsulation is a crucial parameter because failure of microencapsulated islet grafts is usually regarded as a consequence of insufficient biocompatibility, inducing a nonspecific foreign body immune reaction against the microcapsules and resulting in progressively fibrotic overgrowth of the capsules. This overgrowth interferes with adequate nutrition of the islets and consequently causes islet cell death. There are major distinctions between water-soluble polymers, such as alginate, and water-insoluble polymers, such as poly (hydroxyethyl methacrylate-methyl methacrylate)[35]. However, a major obstacle in using water-insoluble polymers for encapsulation of cells is the requirement for an organic solvent, which usually interferes with cellular function[62]. Despite their solubility in aqueous solutions, alginate-based capsules have been shown to remain stable for several years in small and large animals as well as humans[51,63-67]. The method of alginate capsule formation is based on the entrapment of islets in alginate droplets, which are transformed into rigid beads by gelification in a divalent cation solution, mostly Ca^{2+}. In most studies to date, alginate beads were coated with a second layer to reduce the porosity of the capsule membrane[35,68]. In the pig-to-primate model, alternating layers of poly-L-lysine and a polyornithine were used to surround the alginate core[40,51]. The latter type of layer, however, has been associated with polyamino acid cytotoxicity and mechanical instability of the microcapsules, limiting their application[35,69,70]. Several groups have recently reported that encapsulation in simple alginate microbeads can protect pig pancreatic cells against xenorejections in diabetic mice[46,47,71]. Although several chemical formulations of alginate (e.g., high-mannuronic/guluronic; high/low viscosity, with or without additional peptide sequences) have been proposed for islet immuno-isolation, we found that high-mannuronic alginate was the most suitable to obtain selective impermeability for molecules over 150 kDa (as an IgG) before and after transplantation, and optimal biocompatibility to avoid nonspecific inflammatory response associated with surrounding angiogenesis, resulting in sufficient oxygen tension (about 40 mmHg) for the survival and function of encapsulated islets[72]. This type of alginate was biocompatible not only in a small-animal model (Wistar rat recipients) but also in a pig-to-primate model of xenotransplantation under the kidney capsule and skin for up to 6 mo (see below)[25,43,48].

VARIATION OF IMPLANTATION SITES

The lack of revascularization of the encapsulated islets interferes with both the functional performance and the longevity of the grafts. Apparently, a site in which encapsulated islets are in close contact with the bloodstream is obligatory for clinical application. Unfortunately, it is difficult to find such a site because it must be of sufficient size to bear a large graft volume and be near blood vessels. Sites reported to allow successful nonencapsulated islet transplantation, such as the liver and spleen, do not meet these requirements because these sites are unable to tolerate the large volumes (> 16 mL) of capsules (of diameter > 600 μm) required for transplantation into primates. Therefore, most transplantations of encapsulated pig islets into primates were

intraperitoneal[39,39,51]. Although this technique seemed relatively easy, the peritoneal site was not optimal. Indeed, recent studies in mice found that macrophages and lymphocytes are involved in the rapid degradation of encapsulated pig islets after their transplantation into the peritoneum[71,73-76]. The peritoneum is, indeed, a preferential site for inflammation and immunologic reactions[77] and peritoneal mesothelial cells facilitate the action of powerful innate immune mechanisms[77]. Studies in mice showed that immunosuppression had beneficial effects, improving the biocompatibility and prolonging the survival of encapsulated pig islets transplanted into the peritoneum[73,74,78]. This method of combining encapsulation and immunosuppression, however, remains incompatible with clinical applications. The biocompatibility of alginate-encapsulated pig islets depends on the implantation site. Encapsulated pig islets transplanted under the kidney capsule and under the skin demonstrated better biocompatibility than capsules transplanted into the peritoneum[48]. Indeed, a cellular reaction essentially composed of macrophages was observed 7 d after transplantation into the peritoneum. This finding is in good agreement with results showing that macrophages are recruited 7 d after transplantation of encapsulated pig islets into the peritoneum of mice and rats[73,74]. In addition, severe fibrosis surrounding intraperitoneally implanted capsules was observed 30 d after transplantation and was correlated with the loss of porcine C-peptide 7 d after implantation. In contrast, subcutaneous and kidney capsule implantation resulted in very weak cellular immune reactions against encapsulated pig islets, along with improved porcine islet viability; porcine C-peptide was detected in the sera of rats for 30 d after transplantation of encapsulated pig islets at both sites. These findings indicate that implantation into the subcapsular kidney and subcutaneous spaces improves the biocompatibility and *in vivo* survival of encapsulated pig islets, as well as enhancing pig islet function during the first 7 d after transplantation. The loss of the *in vivo* function activity of encapsulated pig islets transplanted into the peritoneum correlated with significant alterations in islet viability, a loss of insulin content, and significant reductions in insulin secretion after glucose stimulation. These findings may be associated with macrophage overgrowth of the area surrounding the capsules, creating a microenvironment of stress, with low oxygen tension, for pig islets[75,76]. Indeed, macrophage activation, as shown by NO production and the release of the cytokines L-1β and tumor necrosis factor-alpha, had a deleterious effect on islet function and viability[73,75,76]. We found that implantation into kidney subcapsular and subcutaneous spaces improved the biocompatibility of encapsulated pig islets and significantly reduced macrophage recruitment. This reduction in pig islet stress and improved islet viability maintained insulin level per islet and insulin secretion after glucose stimulation[48]. Subcutaneous tissue was recently shown to provide oxygen tension (20-40 mmHg) compatible with the function and survival of encapsulated islets[72]. Among the sites being tested for islet transplantation, with or without encapsulation, to improve the survival, engraftment and function of islets, are the brachioradialis muscle[79], striated muscle[80], the greater omentum[81], and the anterior chamber of the eye[82].

CONCLUSION

Immunosuppression remains the major limitation of allotransplantation or xenotransplantation of islets for type 1 diabetes. Extended survival of transplanted pig islets has recently been observed in primate models, but several questions and problems associated with immunosuppression remain to be resolved in terms of adjustment before clinical trials. A bioartificial pancreas made of encapsulated pig islets may overcome the two major hurdles to islet transplantation: the shortage of human organ donors and the requirement for immunosuppressive regimens.

The development of a bioartificial pancreas for preclinical/clinical studies requires the conjunction of integrated parameters such as the choice of a biocompatible material for encapsulation to maintain selective permeability. The encapsulation device should be designed to maintain mechanical properties and stability at an implantation site compatible with the viability and physiology of the encapsulated islets to control glycemic homeostasis.

Of the three major types of bioartificial pancreases (macroencapsulation, microencapsulation, and conformal encapsulation), the macroencapsulation system is the only method that has demonstrated the capacity to control diabetes in large animals and in preliminary clinical studies.

Several improvements must be made to reduce the size of the implant (by increasing islet concentration relative to the surface or volume of the implant), to improve oxygenation of islets (to limit islet death), and to develop a simple clinical procedure for bioartificial transplantation and easy access to a device allowing "realimentation" of the islets.

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