ISSN 0975-6299 Vol.1/Issue-4/Oct-Dec.2010

International Journal of Pharma and Bio Sciences

STEM CELL TREATMENT FOR DIABETES

SAKTHIVEL .K*, RAJESH .C AND SENTHAMARAI .R

Department of Pharmacy Practice, Periyar College of Pharmaceutical Sciences for Girls Tiruchirappalli – 620 021, TN State - INDIA

Corresponding Author tksakthivel@rediffmail.com

ABSTRACT

Diabetes is often called the "silent killer" because it attacks the body slowly and stealthily. Newly diagnosed adult diabetes patients are usually not overly concerned about it since their symptoms are often no more serious than frequent urination and increased thirst. Many other patients have no symptoms at all. It's actually a group of diseases characterized by abnormally high levels of the sugar glucose in the bloodstream. This excess glucose is responsible for most of the complications of diabetes, which include blindness, kidney failure, heart disease, stroke, neuropathy, and amputations. Diabetics using stem-cell therapy have been able to stop taking insulin injections for the first time, after their bodies started to produce the hormone naturally again. Diabetes patients are usually treated by injecting the stem cells into the pancreatic artery via catheter. Patients who cannot safely undergo the catheterization procedure may elect to receive the stem cells intravenously. Both methods are outpatient procedures that require patients to stay in hospital for 4 or 5 nights.

KEYWORDS

Diabetes, Stem cells, Insulin therapy, Pancreatic islet cells.

INTRODUCTION

Diabetes is often called the "silent killer" because it attacks the body slowly and stealthily. Newly diagnosed adult diabetes patients are usually not overly concerned about it since their symptoms are often no more serious than frequent urination and increased thirst. Many other patients have no symptoms at all. For decades, diabetes researchers have been searching for ways to replace the insulin-producing cells of the pancreas that are destroyed by a patient's own immune system. Now it appears that this may be possible. Each year, diabetes affects more people and causes more deaths than breast cancer and AIDS combined. Diabetes is the seventh leading cause of death in the United States today, with nearly 200,000 deaths reported each year. The American Diabetes Association estimates that nearly 16 million people, or 5.9 percent of the United States population, currently have diabetes.

Diabetes is actually a group of diseases characterized by abnormally high levels of the sugar glucose in the bloodstream. This excess glucose is responsible for most of the complications of diabetes, which include blindness, kidney failure, heart disease, stroke, neuropathy, and amputations. Type 1 diabetes, also known as juvenile-onset diabetes, typically affects children and young adults. Diabetes develops when the body's immune system sees its own cells as foreign and attacks and

destroys them. As a result, the islet cells of the pancreas, which normally produce insulin, are destroyed. In the absence of insulin, glucose cannot enter the cell and glucose accumulates in the blood. Type 2 diabetes, also called adult-onset diabetes, tends to affect older, sedentary, and overweight individuals with a family history of diabetes. Type 2 diabetes occurs when the body cannot use insulin effectively. This is called insulin resistance and the result is the same as with type 1 diabetes—a build up of glucose in the blood.

There is currently no cure for diabetes. People with type 1 diabetes must take insulin several times a day and test their blood glucose concentration three to four times a day throughout their entire lives. Frequent monitoring is important because patients who keep their blood glucose concentrations as close to normal as possible can significantly reduce many of the complications of diabetes, such as retinopathy (a disease of the small blood vessels of the eye which can lead to blindness) and heart disease that tend to develop over time. People with type 2 diabetes can often control their blood glucose concentrations through a combination of diet, exercise, and oral medication. Type 2 diabetes often progresses to the point where only insulin therapy will control blood glucose concentrations.

Each year, approximately 1,300 people with type 1 diabetes receive whole-organ pancreas transplants. After a year, 83 percent of these patients, on average, have no symptoms of diabetes and do not have to take insulin to maintain normal glucose concentrations in the blood. However, the demand for transplantable pancreases outweighs their availability. To prevent the body from rejecting the transplanted pancreas, patients must take powerful drugs that suppress the immune system for their entire lives, a regimen that makes them susceptible to a host of other diseases. Many hospitals will not perform a pancreas transplant unless the patient also needs a kidney transplant. That is because the risk of infection due to immunosuppressant therapy can be a greater health threat than the diabetes itself. But if a patient is also receiving a new kidney and will require immunosuppressant drugs anyway, many hospitals will perform the pancreas transplant.

Over the past several years, doctors have attempted to cure diabetes by injecting patients with pancreatic islet cells—the cells of the pancreas that secrete insulin and other hormones. However, the requirement for steroid immunosuppressant therapy to prevent rejection of the cells increases the metabolic demand on insulin-producing cells and eventually they may exhaust their capacity to produce insulin. The deleterious effect of steroids is greater for islet cell transplants than for whole-organ transplants. As a result, less than 8 percent of islet cell transplants performed before last year had been successful.

More recently, James Shapiro and his colleagues in Edmonton, Alberta, Canada, have developed an experimental protocol for transplanting islet cells that involves using a much larger amount of islet cells and a different type of immunosuppressant therapy[1].

The resulting pancreas is a combination of a lobulated, branched acinar gland that forms the exocrine pancreas, and, embedded in the acinar gland, the Islets of Langerhans, which constitute the endocrine pancreas.

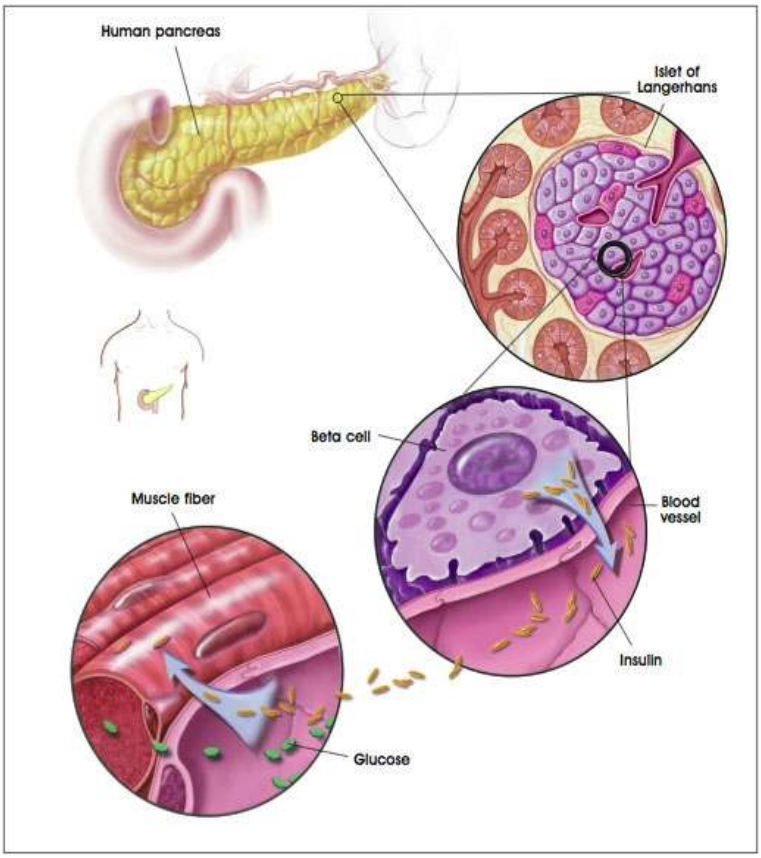


Figure1
(Insulin Production in the Human Pancreas)

The pancreas is located in the abdomen, adjacent to the duodenum (the first portion of the small intestine). A cross-section of the pancreas shows the islet of Langerhans which is the functional unit of the endocrine pancreas. Encircled is the beta cell that synthesizes and secretes insulin. Beta cells are located adjacent to blood vessels and can easily respond to changes in blood glucose concentration by adjusting insulin production. Insulin facilitates uptake of glucose, the main fuel source, into cells of tissues such as muscle.
(© 2001 Terese Winslow, Lydia Kibiuk)

Fetal Tissue as Source for Islet Cells

Therefore, many researchers believe that it will be preferable to develop a system in which stem or precursor cell types can be cultured to produce all the cells of the islet cluster in order to generate a population of cells that will be able to coordinate the release of the appropriate amount of insulin to the physiologically relevant concentrations of glucose in the blood.

Several groups of researchers are investigating the use of fetal tissue as a potential source of islet progenitor cells. For example, using mice, researchers have compared the insulin content of implants from several sources of stem cells—fresh human fetal pancreatic tissue, purified human islets, and cultured islet tissue[2]. They found that insulin content was initially higher in the fresh tissue and purified islets. However, with time, insulin concentration decreased in the whole tissue grafts, while it remained the same in the purified islet grafts. When cultured islets were implanted, however, their insulin content increased over the course of three months. The researchers concluded that precursor cells within the cultured islets were able to proliferate (continue to replicate) and differentiate (specialize) into functioning islet

tissue, but that the purified islet cells (already differentiated) could not further proliferate when grafted. Importantly, the researchers found, however, that it was also difficult to expand cultures of fetal islet progenitor cells in culture[3].

Adult Tissue as Source for Islet Cells

Many researchers have focused on culturing islet cells from human adult cadavers for use in developing transplantable material. Although differentiated beta cells are difficult to proliferate and culture, some researchers have had success in engineering such cells to do this. For example, Fred Levine and his colleagues at the University of California, San Diego, have engineered islet cells isolated from human cadavers by adding to the cells' **DNA** special genes that stimulate cell proliferation. However, because once such cell lines that can proliferate in culture are established, they no longer produce insulin. The cell lines are further engineered to express the beta islet cell gene, PDX-1, which stimulates the expression of the insulin gene. Such cell lines have been shown to propagate in culture and can be induced to differentiate to cells, which produce insulin. When transplanted into immune-deficient mice, the cells secrete insulin in response to glucose. The researchers are currently investigating whether these cells will reverse diabetes in an experimental diabetes model in mice[4, 5].

These investigators report that these cells do not produce as much insulin as normal islets, but it is within an order of magnitude. The major problem in dealing with these cells is maintaining the delicate balance between growth and differentiation. Cells that proliferate well do not produce insulin efficiently, and those that do produce insulin do not proliferate well. According to the researchers, the major issue is developing the technology to be able to grow large numbers of these cells that will reproducibly produce normal amounts of insulin[6].

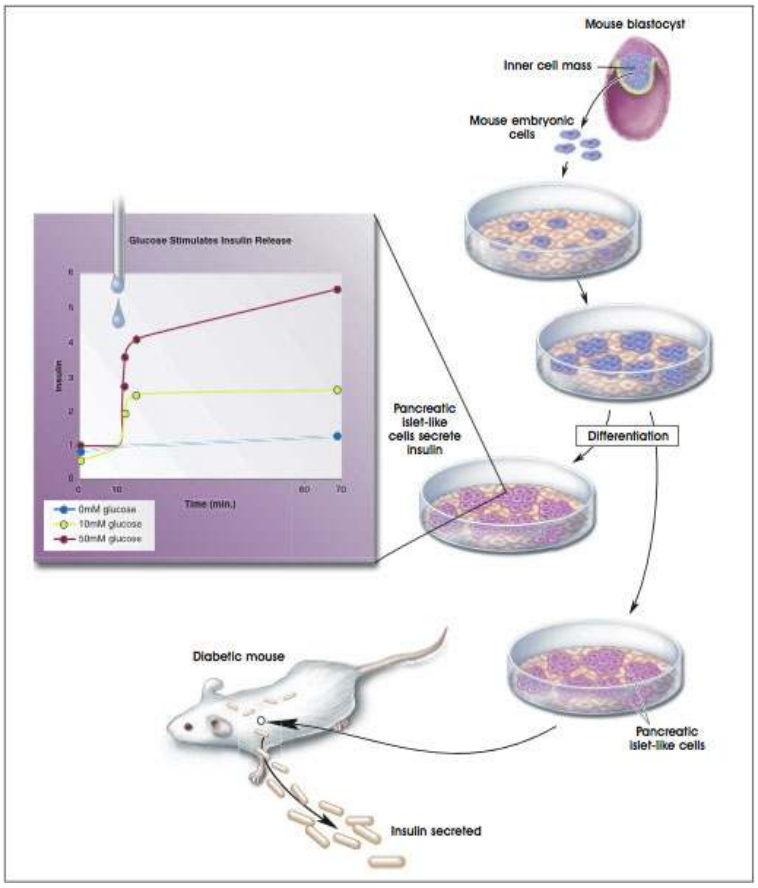
Another promising source of islet progenitor cells lies in the cells that line the pancreatic ducts. Some researchers believe that multipotent (capable of forming cells from more than one germ layer) stem cells are intermingled with mature, differentiated duct cells, while others believe that the duct cells themselves can undergo a differentiation, or a reversal to a less mature type of cell, which can then differentiate into an insulin-producing islet cell.

Susan Bonner-Weir and her colleagues reported last year that when ductal cells isolated from adult human pancreatic tissue were cultured, they could be induced to differentiate into clusters that contained both ductal and endocrine cells. Over the course of three to four weeks in culture, the cells secreted low amounts of insulin when exposed to low concentrations of glucose, and higher amounts of insulin when exposed to higher glucose concentrations. The researchers have determined by immunochemistry and ultrastructural analysis that these clusters contain all of the endocrine cells of the islet[7].

However, some researchers believe that it will be important to engineer systems in which all the components of a functioning pancreatic islet are allowed to develop.

Recently Ron McKay and his colleagues described a series of experiments in which they induced mouse embryonic cells to differentiate into insulin-secreting structures that resembled pancreatic islets[8]. McKay and his colleagues started with embryonic stem cells and let them form embryoid bodies—an aggregate of cells containing all three embryonic germ layers. They then selected a population of cells from the embryoid bodies that expressed the neural marker nestin. Using a sophisticated five-stage culturing technique, the researchers were able to induce the cells to form islet-like clusters that resembled those found in native pancreatic islets. The cells responded to normal glucose concentrations by secreting insulin, although insulin amounts were lower than those secreted by normal islet cells

(Figure2. Development of Insulin-Secreting Pancreatic-Like Cells from Mouse Embryonic Stem Cells). When the cells were injected into diabetic mice, they survived, although they did not reverse the symptoms of diabetes.



Mouse embryonic stem cells were derived from the inner cell mass of the early embryo (blastocyst) and cultured under specific conditions. The embryonic stem cells (in blue) were then expanded and differentiated. Cells with markers consistent with islet cells were selected for further differentiation and characterization. When these cells (in purple) were grown in culture, they spontaneously formed three-dimentional clusters similar in structure to normal pancreatic islets. The cells produced and secreted insulin. As depicted in the chart, the pancreatic islet-like cells showed an increase in release of insulin as the glucose concentration of the culture media was increased. When the pancreatic islet-like cells were implanted in the shoulder of diabetic mice, the cells became vascularized, synthesized insulin, and maintained physical characteristics similar to pancreatic islets.
(© 2001 Terese Winslow, Caitlin Duckwall)

According to McKay, this system is unique in that the embryonic cells form a functioning pancreatic islet, complete with all the major cell types. The cells assemble into islet-like structures that contain another layer, which contains neurons and is similar to intact islets from the pancreas[9].

METHOD

Bone Marrow Collection

On the first day, bone marrow is collected from the patient's iliac crest (hip bone) using thin-needle mini-puncture under local anesthesia. Although some pain is felt when the needle is inserted, most patients do not find the bone marrow collection procedure particularly painful. The entire procedure normally takes about 30 minutes. Once the bone marrow collection is complete, the patient

may return to their hotel and go about normal activities.

Laboratory Processing

The next day, the stem cells are processed from the bone marrow in a state-of-the-art, government approved (cGMP) laboratory. In the lab, both the quantity and quality of the stem cells are measured. These cells have the potential to transform into multiple types of cells and are capable of regenerating damaged cells such as pancreatic beta cells.

Stem Cell Implantation

On the third day, the stem cells are implanted into the pancreatic artery under local anesthesia using a fine wire (catheter) that is inserted into the patient's right femoral artery. X-ray scanning is used to guide the catheter into the pancreatic artery where the stem cells are injected through a small hole in the center of the wire. This procedure takes about 90 minutes. Afterwards, the patient will spend 2 or 3 hours in the recovery room to ensure that the entry site is not bleeding.

Patients who cannot be treated by catheter, such as those with kidney problems, are offered an alternative intravenous stem cell implantation. Patients who are suffering from diabetic peripheral neuropathy will receive a portion of their stem cells via intramuscular injections into the leg muscles.

Following Treatment

Patients are required to stay in town on the fourth day for general safety considerations. They may return home on the fifth day.

REFERENCES

1. Shapiro, J., Lakey, J.R.T., Ryan, E.A., Korbutt, G.S., Toth, E., Warnock, G.L., Kneteman, N.M., and Rajotte, R.V. (2000). Islet transplantation in seven patients with type 1 diabetes mellitus using a glucocorticoid-free immunosuppressive regimen. N. Engl. J. Med. *343,* 230–238.
2. Beattie, G.M., Otonkoski, T., Lopez, A.D., and Hayek, A. (1997). Functional beta-cell mass after transplantation of human fetal pancreatic cells: differentiation or proliferation? Diabetes. *46,* 244–248.
3. Hayek, A., personal communication.
4. Dufayet de la Tour, D., Halvorsen, T., Demeterco, C., Tyrberg, B., Itkin-Ansari, P., Loy, M., Yoo, S.J., Hao, S., Bossie, S., and Levine, F. (2001). B-cell differentiation from a human pancreatic cell line *in vitro* and *in vivo*. Mol. Endocrinol. *15,* 476–483.
5. Itkin-Ansari, P., Demeterco, C., Bossie, S., Dufayet de la Tour, D., Beattie, G.M., Movassat, J., Mally, M.I., Hayek, A., and Levine, F. (2001). PDX-1 and cell-cell contact act in synergy to promote d-cell development in a human pancreatic endocrine precursor cell line. Mol. Endocrinol. *14,* 814–822.
6. Levine, F., personal communication.
7. Bonner-Weir, S., Taneja, M., Weir, G.C., Tatarkiewicz, K., Song, K.H., Sharma, A., and O'Neil, J.J. (2000). *In vitro* cultivation of human islets from expanded ductal tissue. Proc. Natl. Acad. Sci. U. S. A. *97,* 7999–8004.
8. Lumelsky, N., Blondel, O., Laeng, P., Velasco, I., Ravin, R., and McKay, R. (2001). Differentiation of Embryonic Stem Cells to Insulin-Secreting Structures Similiar to Pancreatic Islets. Science. *292,* 1389–1394.
9. McKay, R., personal communication
10. Stem cell treatment for diabetes in type I & II