

PANCREATIC ISLET CELLS TRANSPLANTATION IN EXPERIMENTAL RATS AND INSULIN DEPENDENT DIABETIC PATIENTS

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INTRODUCTION

AND AIM OF THE WORK

INTRODUCTION AND AIM OF THE WORK

It has been more than 100 years ago that Paul Langerhans presented his inaugural thesis on the "Microscopic Anatomy of The Pancreas". In this dissertation, he described the specific cells of the pancreas that were later shown to secrete insulin. These specific cells, of course, later bore his name and became the "Islets of Langerhans" (Langerhans, 1869).

Langerhans' thesis stimulated further studies on the pancreas by many other investigators. Several of these came close to unraveling the relationship between the islet of Langerhans and diabetes mellitus. None came closer than Moses Barron, who wrote in (1920) that the islets secrete a hormone directly into lymph or blood stream (internal secretion), which has a controlling power over carbohydrate metabolism (Barron 1920).

The observation of Barron spurred the investigation of Banting and Best and in (1922), they published their epochal paper in the Canadian Medical Association Journal, entitled "Pancreatic Extracts in the Treatment of Diabetes Mellitus" (Banting et al 1922). Indeed, it appeared that the increasing availability of insulin heralded the beginning of the end of diabetes mellitus as a scourge of mankind. Yet

the administration of exogenous insulin is inadequate to restore metabolic homeostasis and to prevent the devastating secondary complications of diabetes. Hyperglycemia, persistent or intermittent, appears to be a major cause of abnormal capillary basement membrane formation leading to microangiopathy (Spiro, 1973). Glucose sensing devices implanted into patients for 24 hours monitoring of blood glucose levels have demonstrated many periods of hyperglycemia, even in patients considered well controlled with insulin (Service et al., 1970). A more physiologic supply of insulin such as transplantation of normal pancreatic beta cells, would seem to be a logical solution for those Juvenile diabetic patients with a beta cell deficiency.

When islet transplantation therapy becomes a clinical reality, the need for islet tissue will increase enormously, with the demand for human islets presumably exceeding current availability. The pig has been proposed as a donor source of islets of Langerhans that might be applicable to human transplantation in the future, mainly because of the similar structure and biological activity of porcine and human insulin, and the reasonably unlimited availability of pig pancreata. Further support is based on the evidence that the distribution of islets and endocrine cells in the islets is similar in the human and pig pancreas (Marchetti et al., 1990).

The aim of our work is to introduce a method for mass isolation of purified porcine islets, and to assess the in vitro and in vivo function of isolated islets. In vitro study by detection of glucose induced insulin release into the culture media from isolated pancreatic islets. In vivo study by intraperitoneal transplantation of isolated islets into streptozotocin induced diabetic rats.

REVIEW OF LITERATURE

IDIOPATHIC DIABETES MELLITUS

Diabetes is not a disease in the classic sense because of its heterogeneous aetiologies, rather it should be viewed as a syndrome characterized by a state of chronic hyperglycaemia. However, this definition alone implies a limited approach to a complex metabolic disorder with various long term structural tissue damage.

So, a broader definition of diabetes is that it is a syndrome characterized by a state of chronic hyperglycaemia caused by diminished insulin action (insulin deficiency or/and insulin resistance), metabolic defects and structural damage triad:

1) Metabolic defects beside hyperglycaemia there is accelerated fat and protein catabolism.

2) Structural damage triad (long term sequelae):

a) Large vessel disease including accelerated atherosclerosis and medial calcification.

b) Microvascular disease characterized by thickening and abnormality of function of capillary basement membrane resulting in nephropathy and retinopathy.

c) Neuropathy: there are peripheral sensory and motor defects, autonomic nervous system dysfunction, segmental demyelination and abnormalities of Schwann cell (Porte and Halter, 1981).

Since labelling one patient as "diabetic" carries both social, psychological and clinical implications, diagnostic criteria for diabetes and other categories of impaired glucose tolerance have been periodically and thoroughly revised in the last years.

It should be emphasized firstly that in patients presenting with acute symptoms namely thirst, polyuria and unexplained weight loss progressing sometimes to life threatening ketoacidosis or hyperosmolar coma, or those presenting with subacute symptoms as fatigue, pruritis vulvae, skin infection or visual impairment, all that is required is an elevated random blood glucose estimation to make the diagnosis.

On the other hand, chronic hyperglycemia may be asymptomatic or in some instances glucosuria is detected accidentally, e.g., during routine medical examination. It is here that more clear cut guidelines for diagnosis are required.

Revised criteria for the diagnosis of diabetes mellitus and other categories of glucose intolerance have been developed recently by the National Diabetes Data Group (1979) and these have been endorsed by the World Health

Organization (1980) and supported by other national groups as the American Diabetes Association:

1- Single blood glucose measurement: with acute symptoms, gross and unequivocal elevation of blood glucose confirms the diagnosis, namely fasting venous plasma glucose level of 7.8 mmol/L or more (140 mg/dl or more) on post absorptive (or random) venous plasma glucose level of 11.1 mmol/L or more (200 mg/dl or more).

2- Two blood glucose measurement: in the absence of symptoms of diabetes at least two abnormal values fasting and post absorptive as defined above, are required to establish diagnosis.

3- Oral glucose tolerance test (OGTT): this should be reserved for specific indication since in florid diabetes the test is unnecessary, wasteful, and can sometimes, precipitate hyperosmolar coma. The indications are:

- a) equivocal fasting value in asymptomatic patient, e.g., between 120-140 mg/dl or equivocal casual blood glucose values.
- b) suspicion of gestational diabetes.
- c) for clinical or epidemiological diabetes.
- d) suspicion of impaired glucose tolerance category to exclude diabetes mellitus.

The standard OGTT should normally be conducted with specific prerequisites that include ambulatory status, unrestricted diet and cognizance of diabetogenic drugs

(e.g., diuretics). The test must be conducted in the morning after an unrestricted diet (more than 150 g carbohydrate for 3 days) and a 10-16 hours over night fast. At zero time a fasting blood sample should be obtained and a 300 ml solution of 75 g glucose administered orally over 5 minutes. The 75 g glucose load is a compromise, while the North American 100 g load often produces nausea and the 50 g European load may not be a sufficiently vigorous stimulus. In children the load is 1.75 g/kg with a maximum of 75 g. It is recommended that samples are collected at 30 minutes intervals for 2 hours.

The generally ignored differences between plasma and whole blood glucose level, and between capillary and venous levels should be stressed. Whole blood values are some 10-15% lower than plasma values. Similarly capillary values are 7% higher than venous. This has become more important since laboratories may use venous plasma while bedside monitoring techniques use whole blood or plasma capillary blood (National Diabetic Data group, 1979).

Next table demonstrates the diagnostic glucose values for oral glucose tolerance tests using a specific enzymatic glucose assay.

Table (A): Diagnostic glucose values for oral glucose tolerance test (WHO Expert committee On Diabetes Mellitus 1980):

Fasting (mM/L) and/or 2 hour post glucose		
Diabetes Mellitus		
Venous whole blood	≥7	>10
Capillary whole blood	≥7	>11
Venous plasma	≥8	>11

Impaired glucose tolerance		
Venous whole blood	≤7	>7- <10
Capillary whole blood	≤7	>8- <11
Venous plasma	≤8	>8- <11

Normal		
Venous whole blood	≤7	<7
Capillary whole blood	≤7	<8
Venous plasma	≤8	<8

N.B.: mM/L X18 = mg/dl.

The new category of "impaired glucose tolerance" created by these recent criteria is not regarded as diabetes, however, they have an increased risk of progression to diabetes (2-4% per year) which can not be ignored and may be amenable to intervention, as with dietary advice this may be reduced to about 1.3%. Sartor et al. (1980) have claimed that with the further addition of Tolbutamide, the rate may be cut to zero. It has also been suggested that mild diabetes in children may be reversed by chlorpropamide (Mutch and Stowers, 1980).

Patients with impaired glucose tolerance have increased risk to atherosclerotic disease but this is due to association with known risk factors for arterial disease