INTRODUCTION

After the classical demonstration of the role of the pancreas in diabetes mellitus (1,2), and since the identification of the cellular components of the pancreatic islets and their hormonal products (3-6), the development of the islets of Langerhans of the pancreas has attracted the attention of researchers (7-10). Major progress in the understanding of the cell lineages of the endocrine pancreas has come, among others, from ablation studies (11-13), from analyses of the coexpression of endocrine hormones (14-16) and from transgenic approaches (17,18). In the last five years, however, the primordium of the endocrine pancreas has received special and renewed attention, mainly because advances in the field of molecular biology have brought about new insights into the intricacies of islet differentiation (19-25). Perhaps the most important recent contributions to the field have been a consequence of the study of the pancreatic phenotypes of mouse lines carrying induced mutations of developmental control genes - HNF1-α (26); Isll (27); Beta2/NeuroD (28); Nkx2.2 (29); P48 (30); Pax4 (31); Pax6 (32,33); pdx-1 (34-36). A large part of the excitement that surrounds the study of islet development is due to the therapeutic promise harboured by every advance in the understanding of the development and function of the endocrine pancreas.

Cell differentiation, the process by which embryonic cells become different from one another and acquire specialized functions, has been intensively studied in a variety of systems. In every one of the models, one particular differentiation strategy has been found – muscle differentiation is achieved with the help of master genes (37); in the hematopoietic system, pluripotent stem cells and growth factors have a fundamental role (38); in the neural crest, environmental factors found during extensive migrations are probably responsible for the final fate of these multipotent cells (39). Except for the general mechanisms of regulation of gene expression, the details of the differentiation process do not show much similarity between systems – it would seem that, perhaps not surprisingly, cells achieve their differentiated state by different mechanisms – no “model” can possibly be found that applies to the differentiation of every kind of organ or tissue.

Complex combinations of transcription factors control cell differentiation, and although some of them have a very restricted pattern of expression, others are common to many cell types. An emerging concept is that groups of transcription factors perform same or similar work in the differentiation of a variety of tissues or organs (epigenetic code). Perhaps we do not have cellular or organic models of differentiation, but molecular models of co-operation between transcription factors to
bring about similar effects in many kinds of cellular systems. One family of transcription factors that is currently under scrutiny from precisely this point of view, and some of whose members have major roles in islet differentiation is formed by the Pax genes.

The Pax gene family

The Pax genes form a family of developmental control genes coding for transcriptional regulator proteins that share a highly conserved 128 amino acid domain or "paired domain" (reviewed in reference 40). Nine mammalian Pax genes have been cloned. Mutations in three of the human Pax genes cause different human developmental phenotypes – a Pax2 mutation causes the renal-coloboma syndrome (41-43), Pax3 mutations cause Waardenburg type I syndrome (44) and mutations of Pax6 have been identified as the cause of aniridia (45). In addition, several human tumors show chromosomal translocations involving Pax genes, or alterations of the regulation of some members of the family (46-53). The implication of Pax genes in carcinogenesis (54-57) and in the development of organs (58,59) has been recently reviewed. Apart from the pancreas, Pax genes have a major involvement in the differentiation of other two organs, the kidney (60-62) and the thyroid gland (63-66).

Vertebrate Pax genes have been classified into four paralogous (related, non-allelic genes within a single species) groups (I-IV) by structural similarities (reviewed in references 58 and 67). The members of each group share a specific assembly of three motifs: the paired box, the homeobox and the octapeptide. Group I is formed by Pax1 and Pax9, Group II by Pax2, 5 and 8, Group III by Pax3 and 7 and Group IV by Pax 4 and 6. Members of a group have very high sequence similarity within the paired domain and show resembling expression patterns during development. In this context, it is also interesting that Pax transcription factors appear in groups of two in a variety of differentiating systems: Pax2 and Pax6 in the eye (68-70), Pax2 and Pax5 in the midbrain and cerebellum (71, 72), Pax2 and Pax8 in the kidney (62,73), Pax3 and Pax7 in the spinal cord (74-76) and of course Pax4 and Pax6 in the pancreas (31,32). In the model of axial mesoderm differentiation, and with respect to the classical homeobox genes, it has been proposed that the identity of a vertebral segment could be determined by the particular combination of Hox genes expressed at a certain time in that particular segment; these significant combinations of transcription factors would form a "Hox code" (77). The paired combinations of Pax transcription factors that cooperate in differentiation processes make it tempting to speculate about the existence of a "Pax code" to cellular identity at work in many tissues or organs.

Pax4 and Pax6

The role of Pax genes in islet development was instantly emphasized in 1997, with the phenotypes of two targeted mutant mouse lines which show major pancreatic differentiation alterations (31,32): Pax4 and Pax6 are essential for the differentiation of the β- and α-cell-lineages, respectively. Analysis of a natural mutant of Pax6 confirmed and extended the data on the involvement of this gene in the differentiation of the α-cells (33). Together, Pax4 and Pax6 form Group IV of the structural classification of vertebrate paired box genes – they have paired box and homeobox, but not the octapeptide (reviewed in reference 58).

Pax6 (78) is widely expressed in the developing nervous system and eye (79) and in the endocrine pancreas (80). The brain phenotype of mice homozygous for the Pax6 mutation Small eye (Sey) (81,82) shows great complexity and is currently the object of intensive studies (69,83-88). There are two known human pathological conditions