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Diabetes Due to a Progressive Defect in β-Cell Mass in Rats Transgenic for Human Islet Amyloid Polypeptide (HIP Rat)

A New Model for Type 2 Diabetes

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The islet in type 2 diabetes is characterized by a deficit in β-cell mass, increased β-cell apoptosis, and impaired insulin secretion. Also, islets in type 2 diabetes often contain deposits of islet amyloid derived from islet amyloid polypeptide (IAPP), a 37–amino acid protein cosecreted with insulin by β-cells. Several lines of evidence suggest that proteins with a capacity to develop amyloid fibrils may also form small toxic oligomers that can initiate apoptosis. The amino acid sequence of IAPP in rats and mice is identical and differs from that in humans by substitution of proline residues in the amyloidogenic sequence so that the protein no longer forms amyloid fibrils or is cytotoxic. In the present study, we report a novel rat model for type 2 diabetes: rats transgenic for human IAPP (the HIP rat). HIP rats develop diabetes between 5 and 10 months of age, characterized by an ~60% deficit in β-cell mass that is due to an increased frequency of β-cell apoptosis. HIP rats develop islet amyloid, but the extent of amyloid was not related to the frequency of β-cell apoptosis (r = 0.10, P = 0.65), whereas the fasting blood glucose was (r = 0.77, P < 0.001). The frequency of β-cell apoptosis was related to the frequency of β-cell replication (r = 0.97, P < 0.001) in support of the hypothesis that replicating cells are more vulnerable to apoptosis than nondividing cells. The HIP rat provides additional evidence in support of the potential role of IAPP oligomer formation toward the increased frequency of apoptosis in type 2 diabetes, a process that appears to be compounded by glucose toxicity when hyperglycemia supervenes. *Diabetes* 53:1509–1516, 2004

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IAPP, islet amyloid polypeptide; TUNEL, TdT-mediated dUTP nick-end labeling.

In humans with type 2 diabetes, insulin secretion is defective (1,2), presumably at least in part because of an ~60% defect in β-cell mass when compared with BMI-matched nondiabetic humans (3,4). Several mechanisms likely contribute to the deficit in β-cell mass in type 2 diabetes. One potential mechanism is increased β-cell apoptosis induced by cytotoxic oligomers of islet amyloid polypeptide (IAPP) (5–7). IAPP is a 37–amino acid peptide that is coexpressed and cosecreted with insulin by pancreatic β-cells (8,9). The functional role of IAPP is unknown, although it is thought to be involved in negatively regulating insulin secretion in a paracrine manner within the islet (10). In most humans with type 2 diabetes, there are large extracellular deposits of islet amyloid derived from IAPP (11,12). It remains unknown why islet amyloid develops in patients with type 2 diabetes or if it plays any role in the development of islet failure. Recently, molecular-based studies of degenerative diseases characterized by amyloid reveal that the propensity of a protein to develop amyloid fibrils is predictive of the capacity of the same protein to develop toxic oligomers (13).

Recent evidence suggests that toxic oligomers of IAPP much smaller than amyloid fibrils are responsible for the documented cytoxicity of human IAPP (6,7,13,14). Human, monkey, and cat IAPP share close structural homology and spontaneously form amyloid fibrils in an aqueous solution (15–17). Application of human IAPP to β-cells in an aqueous solution induces β-cell apoptosis (5–7), the toxicity not being due to the amyloid fibrils (6,7,13). Rat and mouse IAPP share an identical sequence that differs from humans by the substitution of proline residues in the amyloidogenic region of the peptide, and neither peptide forms amyloid fibrils or is toxic when applied to cells (6,7,18). It is therefore of interest that neither mice nor rats spontaneously develop midlife diabetes characterized by islet amyloid (19). Monkeys and cats do so and have a comparable sequence of IAPP to humans. Taken together, these data support a potential role for formation of toxic oligomers in the increased frequency of apoptosis in type 2 diabetes and the resulting loss of β-cell mass.

Several mouse models transgenic for human IAPP develop diabetes (20–22) characterized by islet amyloid, one

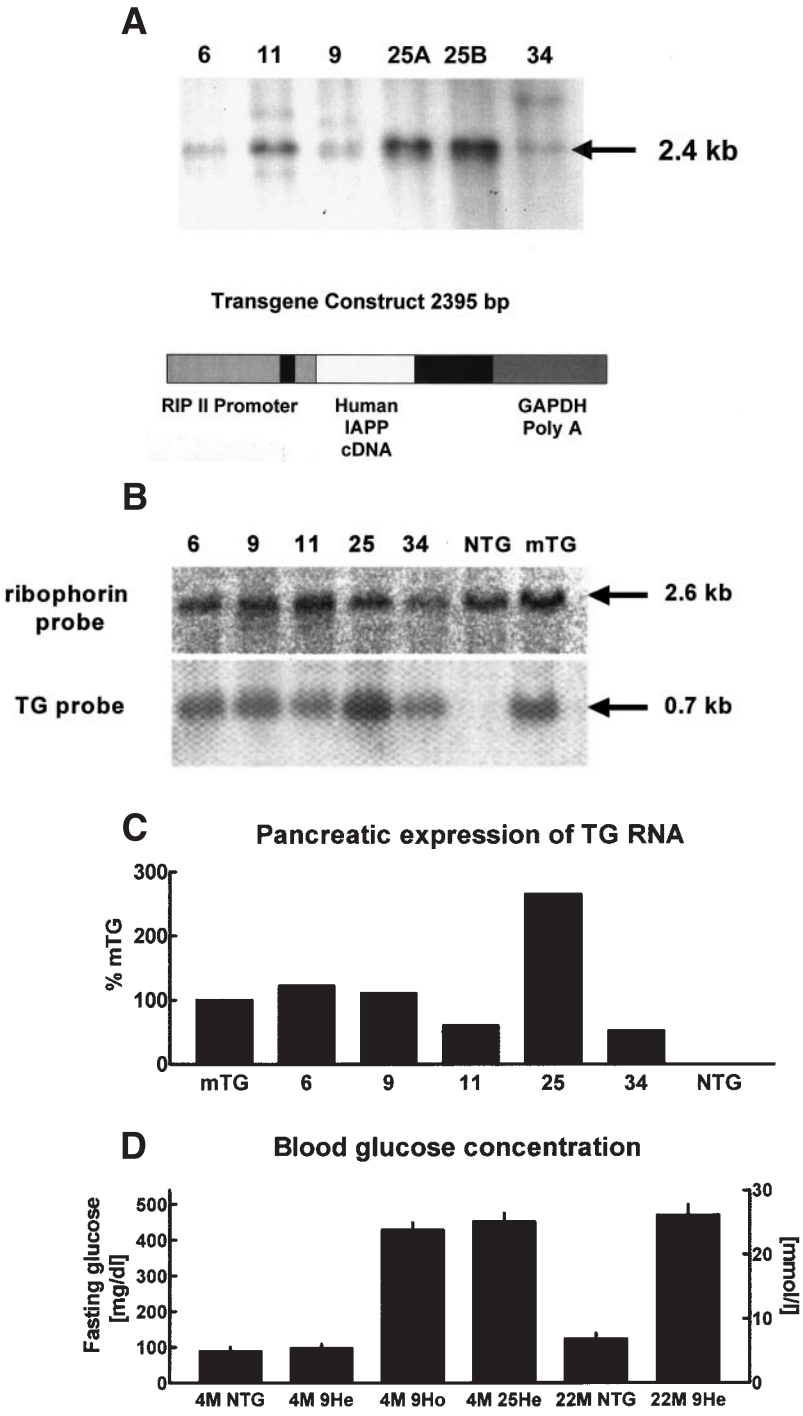

FIG. 1. ***A*****: Southern analysis for five independently derived lines of rats transgenic for hulAPP: 6, 11, 9, 25, and 34. For a time, breeding of line 25 was divided into two separate breeding isolators; these sublines were labeled 25A and 25B. Their genotypes and phenotypes proved to be indistinguishable. The transgene consisted of the rat insulin II promoter (light gray) ligated to the cDNA sequence encoding human IAPP (white); black boxes denote intron sequences. The RIP-II h-IAPP DNA fusion fragment was linked to the human serum albumin intron I sequence upstream of the human GAPDH polyadenylation signal (dark gray).** ***B*****: Northern analysis of hulAPP transgenic (TG) mRNA expression in pancreatic tissue of the TG lines. Pancreatic RNA from h-IAPP TG lines 6, 9, 11, 25, and 34 is shown. Pancreatic RNA from nontransgenic Sprague-Dawley rats (NTG) and pancreatic RNA from h-IAPP TG mice (mTG) were used as negative and positive controls.** ***C*****: Northern blots scanned to quantify RNA. mTG, transgenic mice; NTG, nontransgenic. 6, 9, 11, 25, 34 represent transgenic lines.** ***D*****: Fasting blood glucose concentrations for 4- and 22-month-old transgenic (TG) and nontransgenic rats. He, hemizygous; Ho, homozygous.**

of which has been shown to be characterized by increased β-cell apoptosis (23). Because of limitations of blood volume, relatively few physiological studies are possible in mice. We therefore elected to develop a rat model transgenic for human IAPP to address the following question: do rats transgenic for human IAPP develop diabetes, and if so is this diabetes characterized by a loss of β-cell mass and increased frequency of β-cell apoptosis?

RESEARCH DESIGN AND METHODS

Generation of transgenic rats. The transgene consisted of a recombinant DNA construct identical to that previously used to generate h-IAPP transgenic mice (20,21,23,24). This construct fused the rat insulin II promoter to the cDNA sequence encoding human IAPP. The RIP-II h-IAPP DNA fusion was linked to the human serum albumin intron I sequence upstream of the human GAPDH polyadenylation signal. DNX (Princeton, NJ) was contracted to perform microinjection of this DNA into fertilized Sprague-Dawley rat eggs and to generate and identify the resultant transgenic founder rats. Genomic DNA was extracted from tail snips and subjected to Southern analysis using the transgene DNA as a probe to confirm the PCR genotyping of transgenic founders by DNX. Five founders exhibited germline transmission. Of these five lines, lines 11 and 25 exhibited the highest copy number (Fig. 1). Total RNA extracted from the pancreas of the five lines was subjected to Northern analysis using the human GAPDH portion of the transgene as a probe. Ribophorin cDNA was used for normalizing total amounts of RNA loaded per lane. Pancreatic RNA from nontransgenic Sprague-Dawley rats and pancreatic RNA from previously reported h-IAPP transgenic mice (20) were used as negative and positive controls, respectively. Radioactive signals for RNA were scanned by phosphoimaging to quantify RNA levels.

Initially, fasting blood glucose concentrations were measured at only two

time points: 4 and 22 months of age in each of the five lines. Line 25, which had the highest transgene copy number and most intense RNA signal, developed diabetes spontaneously by 4 months of age (Fig. 1). In contrast, line 9 exhibited diabetes at 22 but not 4 months of age. However, when the transgene copy number of line 9 was doubled by breeding to homozygosity, diabetes was readily apparent at 4 months. These results strongly imply that there is a transgene threshold operating in this pathogenic process. This threshold effect was also observed in h-IAPP transgenic mice (20–24). Because line 9 hemizygotes exhibited a midlife onset of the diabetes reminiscent of type 2 diabetes in humans, we chose to focus our attention on this line of rats for subsequent studies. In keeping with other rodent models of diabetes, males manifest diabetes earlier than females; therefore, for the purposes of this study, we focused on male rats only.

Study design. Our overall goal was to establish a rat model with islet pathology that was reflective of humans with type 2 diabetes. To establish the time course of changes in blood glucose, insulin, and islet morphology, we then prospectively studied rats from line 9 and their nontransgenic counterparts from age 2 months to age 18 months. Blood glucose and insulin concentrations and pancreas morphology were quantified in 22 male h-IAPP transgenic rats (HIP rats) and 20 male control rats at 2, 5, 10, and 18 months of age.

Rat housing and experimental procedures. Rats were bred at Charles River Laboratories (Wilmington, MA), and males were shipped to the University of Southern California after weaning. The animals were housed in pairs (2–5 months) or singly (5–18 months) in controlled environmental conditions with a light cycle of 12 h per day. Animals were fed Rodent Diet 8604 (Harlan Teklad, Madison, WI) ad libitum. On the day of study, animals were anesthetized by inhalation of isoflurane (Abbott Laboratories, Chicago, IL) and intraperitoneal injection of pentobarbital sodium (Abbott Laboratories). Shortly after induction of anesthesia, blood was sampled for measurement of blood glucose and insulin concentrations after a 12-h fast.

Assays. Blood glucose concentrations were measured by the glucose oxidase method using a FreeStyle glucose meter (TheraSense, Alameda, CA). Plasma insulin concentration was measured using an in-house competitive colorimetric enzyme-linked immunosorbent assay as described previously (23). This assay has minimal cross-reactivity with proinsulin.

Morphological techniques. The complete pancreas was rapidly resected from killed rats, all fat and nonpancreas tissue was trimmed, and the pancreas was weighed. The mean weight of the pancreata did not differ between transgenic and nontransgenic rats at any age. A longitudinal section of the pancreas (tail through head in the flat plane of the pancreas) was fixed in formaldehyde and then embedded in paraffin. Sections of pancreas were then taken through the fixed tissue in the plane of embedding so that a near-complete section of pancreas (head, body, and tail) through its maximal width was obtained with each section. These sections were stained for hematoxylin/eosin and insulin as described before (20,21). In addition, adjacent sections were immunostained for the marker of replication Ki67 (rat anti-murine Ki67 monoclonal antibody TEC-3, 1/45; Dako, Carpinteria, CA) and stained by the TdT-mediated dUTP nick-end labeling (TUNEL) method for apoptosis using the TdT-Frag El Kit from Oncogene Research Products (Cambridge, MA) as previously described (4). The β-cell mass for each rat was measured by first obtaining the fraction of the cross-sectional area of pancreatic tissue (exocrine and endocrine) positive for insulin staining and then multiplying this by the pancreatic weight. The frequency of β-cell replication for each rat was calculated by averaging the number of $Ki67^+$ β-cells in ~30 islets from each rat by comparison of the insulin and Ki67 stains. The frequency of β-cell apoptosis for each rat was similarly computed by examination of the $TUNEL^+$ β-cells in the same islets as for the Ki67 stain. The frequency of β-cell replication and β-cell apoptosis was expressed as events per islet. To establish a relative frequency of new islet formation (islet neogenesis) between groups, we measured the percentage of exocrine duct cells positive for insulin as previously described (4). The frequency and extent of islet amyloid was quantified using Congo red–stained slides. Cases were scored for *1*) presence or absence, *2*) frequency of islet amyloid (using a scale from 0 to 3, where 0 indicated that no islets contained amyloid, 1 indicated a few islets in the sample had amyloid, 2 indicated that numerous islets in the sample had amyloid, and 3 indicated that most islets had amyloid), and *3*) extent of islet amyloid (μm^2 per islet).

Calculations and statistical calculations. Statistical comparisons were performed using ANOVA and regression analysis where stated. Data in graphs are presented as means ± SE. Findings were assumed to be statistically significant at the $P < 0.05$ level.

RESULTS

Body weight, blood glucose, and insulin concentrations. HIP and wild-type rats gained weight comparably until 5 months of age, and thereafter the mean weight of the HIP rats was ~20% less than that of the wild-type rats. This difference in weight most likely reflected glycosuria consequent on the onset of diabetes in the HIP rats between 5 and 10 months of age. Alternatively, the impaired weight gain may have been a consequence of relative insulin deficiency and impaired anabolic effects of insulin. Although the blood glucose progressively increased in HIP rats, the rats tolerated the hyperglycemia with no weight loss through to the end of the study at 18 months. The blood insulin concentrations showed variability in both wild-type and HIP rats with overlap between groups but a trend toward insulin deficiency after 10 months of age in HIP rats, particularly in light of their hyperglycemia. This impression was affirmed by examining the insulin-to-glucose ratio (insulinogenic index), which was significantly decreased in HIP versus wild-type rats at 10 and 18 months of age ($P < 0.05$) (Fig. 2).

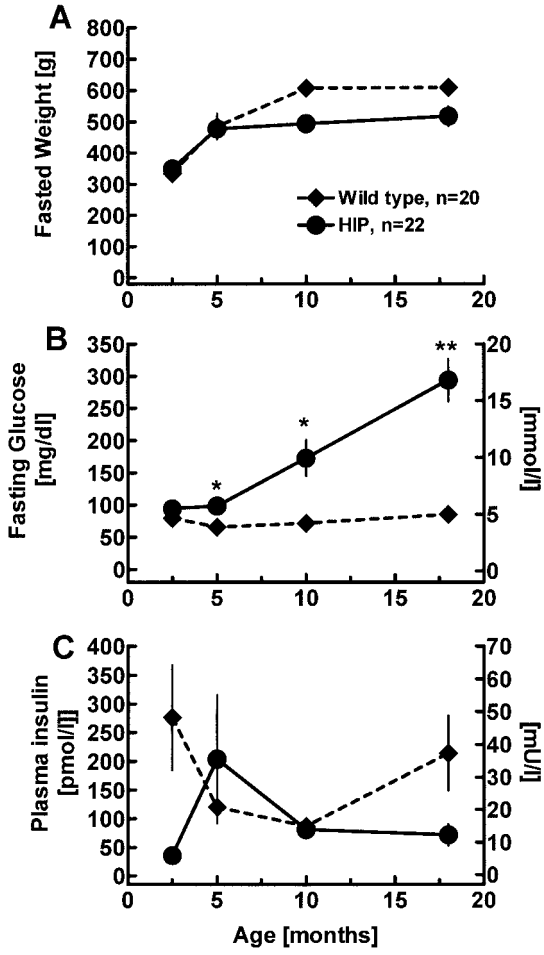

FIG. 2. The mean body weight and the mean fasting blood glucose and insulin concentrations in 22 HIP and 20 wild-type rats from 2–18 months of age.

β-Cell mass, β-cell replication, apoptosis, and islet neogenesis. There was no difference in the mean total pancreas weight between the HIP and wild-type rats at any age. β-Cell mass was comparable in HIP and wild-type rats until 5 months of age (Fig. 3). Thereafter, the β-cell mass continued to increase in wild-type rats but decreased in HIP rats, so that an ~60% deficit in β-cell mass coincided with the onset of diabetes. Morphological examination of

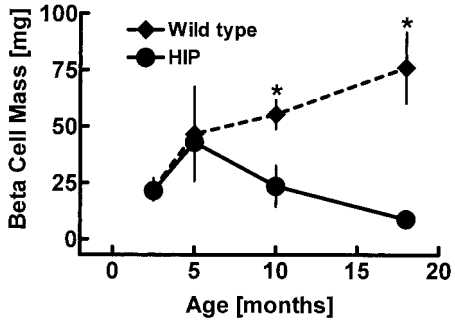

FIG. 3. The mean β-cell mass in 22 HIP and 20 wild-type rats from age 2–18 months.

the pancreata in HIP rats by 18 months of age (Fig. 4) revealed extensive islet amyloid and relatively frequent β-cell replication and apoptosis compared with wild-type rats. Although islet density was no different between HIP and wild-type rats (Fig. 5), the fraction of the islet occupied by β-cells (staining positively for insulin) progressively decreased in HIP rats compared with wild-type rats because amyloid occupied an increasing proportion of the islets. When the parameters of islet turnover were formally analyzed (Fig. 6), these impressions were affirmed with a greater frequency of apoptosis per islet present in HIP rats from 2 months of age, which progressively increased

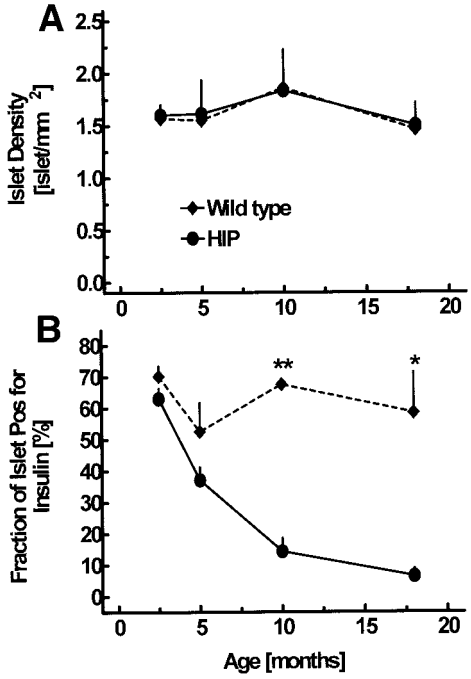

FIG. 5. The mean islet density and islet fraction positive for insulin in 20 wild-type and 22 HIP rats from age 2 to 18 months.

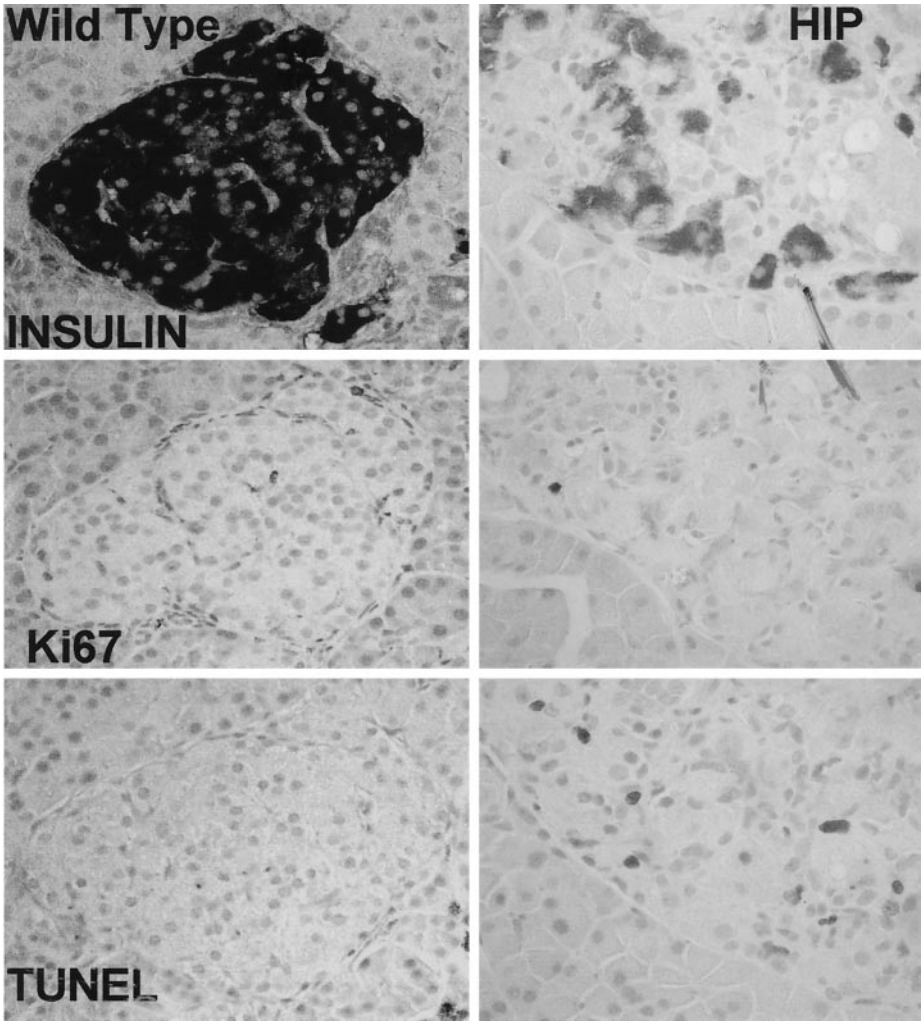

FIG. 4. Representative islets from 18-month-old HIP rat (*right three panels*) and wild-type rat (*left three panels*). Top panels stained for insulin show a relative loss of insulin-positive cells in HIP islets with amyloid deposits between cells. The middle panels show more KI67$^+$ cells in HIP islet. Lower panels show increased TUNEL$^+$ apoptotic cells in HIP islet.

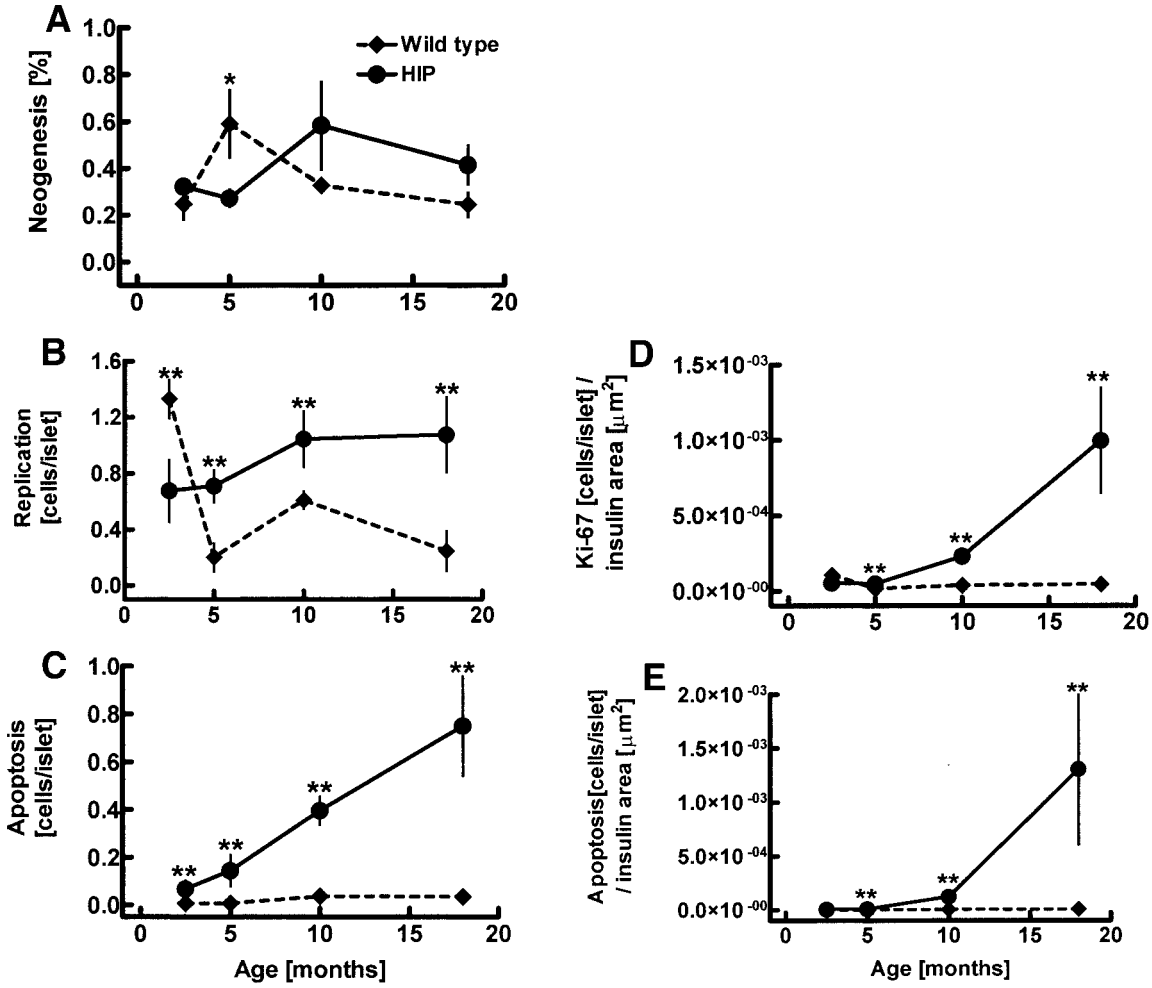

FIG. 6. Mean islet neogenesis (*A*) (estimated indirectly by percentage of duct cells positive for insulin), β-cell replication, and apoptosis expressed per islet (*B* and *C*) and per islet divided by fractional insulin area (*D* and *E*) in 22 HIP and 20 wild-type rats from 2 to 18 months of age.

thereafter. When this was normalized to insulin area per islet (to take into account the fewer β-cells per islet in HIP rats), the computed increased frequency of β-cell apoptosis was also increased, to an even greater extent after 5 months of age when β-cell mass was decreased in the HIP rats. β-Cell replication per islet was decreased in HIP versus wild-type rats at 2 months of age, but thereafter, coincident with the higher blood glucose concentration in HIP rats, β-cell replication was assumed increased. The percentage of duct cells positive for insulin (an indirect estimate of new islet formation) was not significantly different between groups but tended to be higher in the HIP rats than the wild-type rats once the former developed diabetes. Taken together, these data indicate that the mechanism subserving the deficit in β-cell mass in the HIP rats was an increased frequency of β-cell apoptosis and that this was not adequately compensated for by the coincident increased β-cell replication and possibly increased islet neogenesis observed in the HIP rats.

Islet amyloid frequency and extent. Islet amyloid (detected and quantified by Congo red staining) was present in small amounts by 2 months of age in occasional islets in the HIP rats. The percentage of islets with islet amyloid and the extent of amyloid increased to a plateau by 10 months of age. Amyloid was not present in the islets of wild-type rats. There was no obvious relationship between

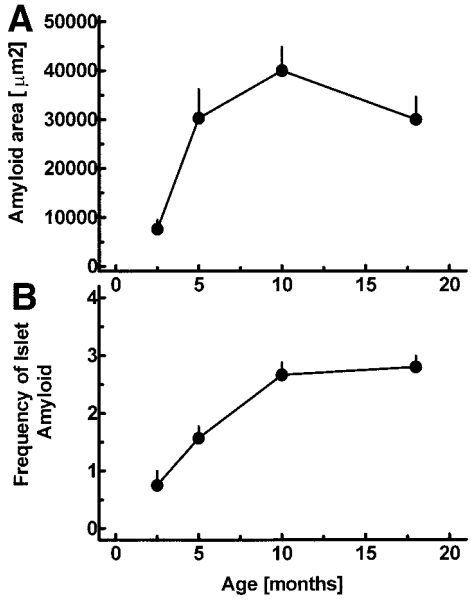

FIG. 7. Mean islet amyloid area (*A*) and frequency of islet amyloid (*B*) in 22 HIP rats. There was no islet amyloid in wild-type rats.

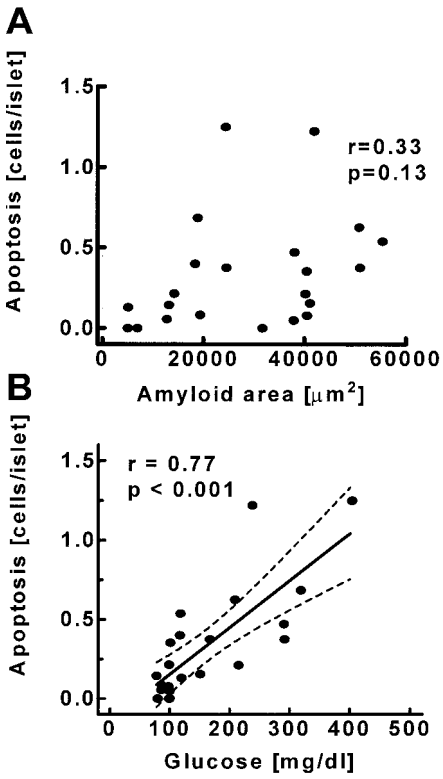

FIG. 8. Relationship between islet amyloid area and β-cell apoptosis (*A*) and fasting blood glucose concentration and apoptosis (*B*) in 22 HIP rats.

the location of amyloid in islets and the presence of β-cells undergoing apoptosis (Fig. 7).

There was no significant relationship between the extent of islet amyloid and the frequency of β-cell apoptosis in HIP rats whether expressed as cells/islet ($r = 0.33$, $P = 0.13$) or cells/islet corrected for insulin area ($r = 0.1$, $P = 0.65$) (Fig. 8). However, there was a positive relationship between the frequency of β-cell apoptosis and the blood glucose concentration (Fig. 8) in the HIP rats but not the wild-type rats ($r = 0.77$, $P < 0.001$). There was also a positive relationship between the frequency of β-cell replication and the frequency of β-cell apoptosis ($r = 0.97$, $P < 0.001$) in the HIP (but not wild-type) rats.

DISCUSSION

We report a novel rat model for type 2 diabetes. This human IAPP transgenic rat (the HIP rat) spontaneously develops diabetes characterized by islet amyloid and a deficit in β-cell mass due to increased β-cell apoptosis.

There are several mouse models transgenic for human IAPP that develop diabetes under conditions of insulin resistance, for example, because of obesity (22,24) or treatment with growth hormone and glucocorticoid treatment (21), or after breeding to homozygosity (20). Here, we report the first rat model transgenic for human IAPP. This model develops middle-age (~4–10 months) onset of diabetes, with islet pathology closely resembling that in humans with type 2 diabetes. In this model, diabetes onset coincides with the development of an ~60% deficit in β-cell mass, a defect that progresses to ~90% by 18 months of age. From age 5–18 months, β-cell mass increased by ~60% in wild-type rats but decreased by ~80% in HIP rats. β-Cell mass is regulated with input from new islet formation (islet neogenesis) as well as replication of β-cells within islets and output from β-cell apoptosis (25,26). In the present transgenic model, the mechanism subserving the defect in β-cell mass is an increased frequency in β-cell apoptosis. This increased frequency of β-cell apoptosis preceded the development of hyperglycemia. In common with a prior report in obese mice (23), the frequency of β-cell apoptosis did not correlate with the extent of islet amyloid as identified by Congo red staining. Furthermore, inspection of islets in HIP rats did not reveal a relationship between the location of apoptotic cells and the large extracellular amyloid deposits (Fig. 4). These data provide further support for the evolving concept that it is not the amyloid fibrils that are toxic in amyloidogenic diseases, but a distinct form of small toxic oligomers that appear to have a common structure, even when derived from separate amyloidogenic proteins, for example, IAPP, synuclein, and Alzheimer's β protein (5,6,13,23). Pathologically high glucose concentrations can also induce β-cell apoptosis (27). In the present model, there is a relationship between the blood glucose concentration and the frequency of β-cell apoptosis in HIP rats (Fig. 8), which is anticipated by the concordant increases in the frequency of β-cell apoptosis and blood glucose concentrations (~10–15 mmol/l) from 10 to 18 months of age. Interestingly, the threshold for glucose-induced apoptosis for rat islets lies within this range (27,28). In obese mice transgenic for human IAPP, we did not see a relationship between the frequency of β-cell apoptosis and the blood glucose concentration, but the mean blood glucose in these mice reached a plateau at ~10 mmol/l (23). These data suggest that the increased frequency of β-cell apoptosis in the HIP rat model reported here from age 2–10 months is due to toxic oligomers of human IAPP, but as the blood glucose concentration increases above 10 mmol/l after the age of 10 months, glucose toxicity may be superimposed on the toxicity of h-IAPP oligomers.

In the present studies, we also quantified β-cell input in the HIP rats versus control rats. The frequency of β-cell replication increased in transgenic rats in relation to the increased blood glucose, consistent with prior reports (25,26). This increased frequency of β-cell replication presumably to some extent offsets the increased frequency of apoptosis to reduce the relative loss of β-cell mass. However, because replicating β-cells are more susceptible to apoptosis than nonreplicating cells (7,23), this increased frequency of replication per se may have contributed to the observed progressive increased frequency of apoptosis from 10 to 18 months of age. Preferential apoptosis of replicating cells would be expected to result in a failure to appropriately expand β-cell mass, as observed in HIP rats. If this failed expansion of β-cell mass due to preferential apoptosis of replicating β-cells subject to toxic IAPP oligomers was further complicated by increased β-cell apoptosis caused by glucose toxicity once blood glucose concentrations had exceeded ~10 mmol/l, then a subsequent decrease in β-cell mass would be predicted, again as observed in the present studies. The other potential source of β-cell input is new islet formation (neogenesis) from ductal precursor cells (25). This is the most difficult component of islet turnover to measure and

is measured here indirectly by quantifying the percentage of ductal cells positive for insulin. Islets budding from exocrine ducts were present in both control and HIP rats at all ages, consistent with continued islet regeneration. When this was quantified, there was a modest but insignificant increased rate of islet regeneration in HIP rats from 10 months of age. Both short-term hyperglycemia and an abrupt decrease in β-cell mass by partial pancreatectomy induce a marked increase in islet neogenesis in rodents (25). As the HIP rats have both hyperglycemia and a deficit in β-cell mass, a more robust increased rate of islet neogenesis might have been expected. However, it is not clear that the increase in islet neogenesis observed in acute studies can be sustained for the many months of hyperglycemia and β-cell deficit present in these animals. We were unable to detect an increased rate of islet neogenesis in humans with type 2 diabetes when compared with BMI-matched nondiabetic control subjects (4).

How does this novel rat model for type 2 diabetes compare with models most commonly available at present? First, to compare the HIP rat with available h-IAPP transgenic mice, only one of these models has been examined longitudinally for the relationship between the development of hyperglycemia, changes in β-cell mass, and the balance of β-cell input and loss (23) as reported here in the HIP rat. This murine model is the first generation of a cross between a previously developed homozygous h-IAPP transgenic mouse (20) and the Avy/a mouse on the C57BL/6 background. The resulting male obese hemizygous h-IAPP transgenic mice develop diabetes. A limitation of this murine model is that two mouse colonies (Avy/agouti and homozygous h-IAPP transgenic mice) have to be maintained to generate the animals of interest, and then only ~13% of these are male obese h-IAPP transgenic and are therefore prone to diabetes. Another limitation of the murine model is the limited blood volume compared with the HIP rat, precluding physiological studies. There are also some interesting differences between these models. The murine model develops hyperglycemia up to ~200 mg/dl (~11 mmol/l) and then sustains this glucose value. In contrast, the HIP rat develops progressive hyperglycemia to ~300 mg/dl (~17 mmol/l). This difference likely explains that lack of a relationship between the blood glucose concentration and frequency of β-cell apoptosis in the murine model, whereas this relationship is present in the HIP model (Fig. 8). Thus, the HIP rat model should be useful in studies of glucose toxicity.

How does the HIP rat model compare with available rat models for type 2 diabetes? The diabetes-prone Zucker fatty rat model has been widely used for islet studies pertaining to type 2 diabetes (29–31). This rat develops extreme obesity because of a genetic defect in the leptin receptor (32,33). Whereas the original Zucker fatty rats compensate for the insulin resistance that develops as a consequence of obesity by increasing β-cell mass and insulin secretion, selective breeding has generated colonies of diabetes-prone Zucker fatty rats that develop diabetes. The mechanism subserving this propensity for development of diabetes is failure to adequately increase β-cell mass because of increased β-cell apoptosis (30,31). The mechanism underlying the increased frequency of β-cell apoptosis is not fully understood but has been attributed to lipotoxicity caused by lipid accumulation within islets as well as glucose toxicity (34–36). This model has the benefits that it mimics many aspects of the metabolic syndrome. Limitations of the model for studies of the evolution of the defect in islet turnover and function in relation to type 2 diabetes in humans is the extreme obesity required to provoke the diabetes phenotype and the fact that the resulting islet morphology does not resemble that in humans with type 2 diabetes. Another rodent model for type 2 diabetes is the gerbil *Psammomys obesus* (37,38). In captivity, if this rodent is fed a high-carbohydrate diet (versus its natural diet of a low-calorie salt brush), animals become obese and, similarly to the Zucker fatty rat, selective breeding has generated diabetes-prone animals that are characterized by a progressive loss of β-cell mass because of an increased frequency of β-cell apoptosis that has been attributed to glucose toxicity (38). Again, the islets in this model do not have the islet amyloid seen in humans with type 2 diabetes, but the animal does not require the extreme obesity of the diabetes-prone Zucker fatty rat to develop diabetes. Another well-characterized rat model for type 2 diabetes is the Goto-Kakizaki (GK) rat model developed by selective breeding of nondiabetic Wistar rats (39). In common with the HIP rat model, the GK rat is a nonobese model. In contrast to the HIP rat model, the GK model has a deficit in β-cell mass from birth that becomes progressively larger as a consequence of impaired new islet formation and β-cell replication rather than increased apoptosis (40–42). Interestingly, this deficit appears to be due to impaired IGF-II production (43). Therefore, the GK rat is a useful model of impaired β-cell replication leading to a deficit in β-cell mass, in contrast to the HIP rat, in which the deficit in β-cell mass is due to increased β-cell apoptosis rather than a decreased frequency of β-cell apoptosis.

In summary, we report a novel rodent model for type 2 diabetes: the HIP rat. This rat develops diabetes during midlife with a relatively gradual onset. It does not require extreme obesity to realize this phenotype and the islet pathology closely resembles that in humans with type 2 diabetes. This novel rodent model should be an important resource for studies seeking to elucidate the mechanisms leading to islet dysfunction in type 2 diabetes and therapies to treat and prevent it.

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