

# $\beta$ -Score

## An assessment of $\beta$ -cell function after islet transplantation

EDMOND A. RYAN, MD<sup>1</sup>  
BREAY W. PATY, MD<sup>1</sup>  
PETER A. SENIOR, MD, PHD<sup>1</sup>

JONATHAN R.T. LAKEY, PHD<sup>2</sup>  
DAVID BIGAM, MD<sup>2</sup>  
A.M. JAMES SHAPIRO, MD, PHD<sup>2</sup>

**OBJECTIVE** — Success after islet transplantation can be defined in terms of insulin independence, C-peptide secretion, or glycemic control. These measures are interdependent and all need to be considered in evaluating  $\beta$ -cell function after islet transplantation. For the current study, a composite  $\beta$ -score was developed that provides an integrated measure of  $\beta$ -cell function success after islet transplantation.

**RESEARCH DESIGN AND METHODS** — The proposed scoring system gave 2 points each for normal fasting glucose, HbA<sub>1c</sub>, stimulated C-peptide, and absence of insulin or oral hypoglycemic agent use. No points were awarded if the fasting glucose was in the diabetic range, the HbA<sub>1c</sub> was >6.9%, C-peptide secretion was absent on stimulation, or daily insulin use was in excess of 0.24 units/kg. One point was given for intermediate values. The score ranged from 0 to 8 and was correlated with the glucose value 90 min after a standard mixed meal challenge ( $n = 218$ ) in 57 subjects before and after islet transplantation. The score was also used to follow subjects for up to 5 years after islet transplantation.

**RESULTS** — The  $\beta$ -score correlated well with the plasma glucose level 90 min after a mixed meal challenge ( $r = -0.849$ ,  $P < 0.001$ ). On follow-up, the  $\beta$ -score rose after the first transplant and was maintained up to 5 years, demonstrating continuing function of the transplanted  $\beta$ -cells.

**CONCLUSIONS** — The  $\beta$ -score provides a simple clinical scoring system that encompasses glycemic control, diabetes therapy, and endogenous insulin secretion that correlates well with physiological measures of  $\beta$ -cell function. On this basis, it is suitable as an overall measure of  $\beta$ -cell transplant function. The  $\beta$ -score gives an integrated measure of  $\beta$ -cell function as a continuum that may be more useful than simply assessing the presence or absence of insulin independence.

*Diabetes Care* 28:343–347, 2005

Islet transplantation typically offers stabilization of blood glucose control, elimination of problematic hypoglycemia, and, frequently, insulin independence (1–4) and is being increasingly used worldwide (5–7). However, formal stimulation tests with intravenous glucose or arginine demonstrate that  $\beta$ -cell

reserve is often not completely normalized (1). Clinically, there is a spectrum of outcomes after islet transplantation, with some patients completely insulin independent with absolutely normal glucose profiles. Others have residual endogenous insulin secretion but may require supplementary insulin or oral hypoglycemic

mic agents (OHAs) to maintain appropriate glucose control, and some lose endogenous insulin secretion.

The simplest measure of success after islet transplantation is insulin independence. However, after islet transplant, a patient may be off insulin and yet have glucose values that are elevated with a raised HbA<sub>1c</sub>, clearly a suboptimal outcome. Equally, a patient may be taking insulin but, by virtue of some endogenous insulin secretion, have perfectly stable glucose values and excellent glucose control. In this setting, islet transplantation may be considered very successful even though the patient does not achieve insulin independence. C-peptide levels are useful for documentation of islet graft survival, but interpretation of the C-peptide values independent of simultaneous glucose levels is of limited value in deciding if  $\beta$ -cell function is adequate. Finally, simple measures of glucose control, such as fasting glucose, HbA<sub>1c</sub>, and glucose-to-insulin ratios, are useful but difficult to interpret if OHAs or insulin are being used.

Thus, there is need for a composite measure of clinical success after an islet transplant that incorporates insulin independence or the need for insulin/OHAs, glucose control, and graft survival. We have developed a  $\beta$ -score that provides an integrated measure of  $\beta$ -cell function after islet transplantation using these parameters.

### RESEARCH DESIGN AND METHODS

The proposed  $\beta$ -score is a composite scoring system based on fasting plasma glucose values, HbA<sub>1c</sub>, insulin independence or use of insulin/OHAs, and the determination of stimulated C-peptide levels. The scoring system is shown in Table 1. Normal values are given a score of 2, intermediate values merit a score of 1, and clearly abnormal values garner no points. Thus, a perfect score is 8, and a score of 0 indicates absolute absence of  $\beta$ -cell function. Normal fasting glucose was taken as <5.6 mmol/l (8), the upper limit of normal for HbA<sub>1c</sub> in our laboratory is 6.1%, and the lower limit of normal for C-peptide is 0.3

From the <sup>1</sup>Department of Medicine, Clinical Islet Transplant Program, University of Alberta and Capital Health Authority, Edmonton, Alberta, Canada; and the <sup>2</sup>Department of Surgery, Clinical Islet Transplant Program, University of Alberta and Capital Health Authority, Edmonton, Alberta, Canada.

Address correspondence and reprint requests to Edmond A. Ryan, 362 Heritage Medical Research Centre, Edmonton, Alberta, Canada T6G 2S2. E-mail: edmond.ryan@ualberta.ca.

Received for publication 11 August 2004 and accepted in revised form 11 November 2004.

**Abbreviations:** OHA, oral hypoglycemic agent.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

© 2005 by the American Diabetes Association.

The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked "advertisement" in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

**Table 1—Determination of components of the  $\beta$ -score**

Components	Score of 2	Score of 1	Score of 0
Fasting plasma glucose (mmol/l)	$\leq 5.5$	5.6–6.9	$\geq 7.0$
HbA <sub>1c</sub> (%)	$\leq 6.1$	6.2–6.9	$\geq 7.0$
Daily insulin (units/kg) or OHA use	—	0.01–0.24 and/or OHA use	$\geq 0.25$
Stimulated C-peptide (nmol/l)	$\geq 0.3^*$	0.1–0.29	$< 0.1^\dagger$

\*If fasting C-peptide was  $\geq 0.3$  nmol/l, then the stimulated C-peptide level was assumed to be  $\geq 0.3$  nmol/l.

†If stimulated C-peptide was  $< 0.1$  nmol/l, then an overall score of 0 was awarded.

nmol/l. Because it is possible, with good diabetes therapy, to have a normal HbA<sub>1c</sub> or fasting glucose level yet have no endogenous insulin secretion, an arbitrary value of 0 was allotted if the stimulated C-peptide was below the limit of detectability (0.1 nmol/l), as the primary goal of the score is to assess  $\beta$ -cell function. A stimulated C-peptide was defined as the C-peptide after a standard mixed meal challenge (see below). On five occasions we have found the C-peptide response to be present with mixed meal testing but absent with intravenous arginine testing. We have never had an instance of C-peptide being present with the intravenous arginine test but absent using the mixed meal test.

To validate the score, we examined the correlation between the 90-min glucose after a mixed meal stimulation test ( $n = 218$ ) and the  $\beta$ -score. We have previously indicated that a good simple measure of graft function in terms of insulin release is the glucose level determined 90 min after consuming a standard meal (Ensure HP) (1). The meal tolerance test was performed in the fasting state, with blood drawn for glucose and C-peptide at baseline and then at 90 min after drinking 360 ml of Ensure HP (providing 391 kcal with 8.5 g of fat, 44 g of carbohydrate, and 17 g of protein). For subjects on insulin or OHAs, no diabetes medications were taken until completion of the test. Included in this analysis were 57 subjects (mean age  $42.1 \pm 1.3$  years and mean duration of diabetes  $26.1 \pm 1.3$  years) who had a meal tolerance test either pretransplant ( $n = 35$ ) or posttransplant. In six further instances in which the subjects were on insulin and the fasting glucose was  $< 4.0$  mmol/l at the start of the mixed meal challenge, the 90-min values were not used because it was felt that there was a surfeit of exogenous insulin present, invalidating the stimulation test as a measure of  $\beta$ -cell reserve. The median

number of meal tolerance tests performed in each subject was three (interquartile range two to five). On 19 occasions for the meal tolerance test, the subjects were taking thiazolidinediones, and on 10 occasions, the subjects were on metformin, and these are included in the total 218 subjects. Results used were from subjects tested pretransplant and  $> 3$  months after transplant so that the HbA<sub>1c</sub> value could be interpreted appropriately.

We also determined the score obtained longitudinally in the 44 subjects who were given an adequate number of islets to become insulin independent after the islet transplant. We examined the scores obtained at 3 months after the first transplant and at 1, 2, 3, 4, and 5 years after the first transplant. Yearly ( $\pm 2$  months) intervals were taken in order to capture as many data points as possible. If a mixed meal test was not performed around the time of scoring but the basal C-peptide was  $\geq 0.3$  nmol/l, it was assumed that the stimulated C-peptide was likewise  $\geq 0.3$  nmol/l. If the C-peptide was absent upon stimulation at any time point, then it was assumed to remain negative and the zero value was carried out to the end of follow-up for that subject. In six instances where there was a repeat stimulation test after a negative one, it was always found to remain negative.

For comparison, we also determined the score in a group of patients ( $n = 12$ ) after whole-pancreas transplant. Eleven of the subjects were men. The mean age of this group was  $43.2 \pm 2.4$  years, and the median time from transplant was 33.4 months (interquartile range 20.3–41.7).

#### Assays

Plasma glucose concentrations were determined by the glucose oxidase method. C-peptide was measured using a commercial assay (Diagnostic Systems Laboratories, Webster, TX). The lower limit of sensitivity for C-peptide was 0.1 nmol/l in

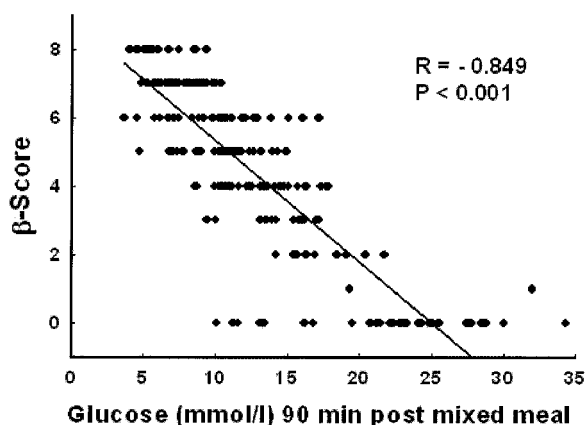
our laboratory, and the intra-assay and interassay coefficients of variations were  $< 9.5\%$  and the normal range was 0.3–1.32 nmol/l.

#### Statistics

All statistical analyses including linear regression were performed using SigmaStat for Windows (version 3.0; SPSS, Chicago, IL). For group comparisons, one-way repeated-measures ANOVA on ranks with a Holm-Sidak test for repeated comparisons of pre- versus posttransplant was used. Best subsets and stepwise forward regression analysis was performed to determine the appropriate contributors to the  $\beta$ -score. Descriptive statistics are given as the means  $\pm$  SE or median (25–75% interquartile range) as appropriate.  $P < 0.05$  was considered significant.

**RESULTS**— Using stepwise forward regression analysis, the  $\beta$ -score (dependent variable) could be predicted from a linear combination of the independent variables: fasting plasma glucose, stimulated C-peptide, daily insulin/OHA use, and HbA<sub>1c</sub> ( $P < 0.001$  for each). The addition of basal C-peptide to the model only improved the  $R^2$  marginally. The basal C-peptide was highly correlated with the stimulated C-peptide ( $r = 0.895$ ,  $P < 0.001$ ), and, thus, to prevent unnecessary overweighting of C-peptide, only the stimulated value was used in the scoring system.

The mixed meal tolerance tests were performed pretransplant, typically 3 months posttransplant, and then every 6–12 months for as long as 5 years' posttransplant. The relationship of the glucose level 90 min after consuming the mixed meal versus the  $\beta$ -scores is shown in Fig. 1, and a good correlation was evident ( $r = -0.849$ ,  $P < 0.001$ ). If the pretransplant results were removed from the analysis, the relationship was still present ( $r = -0.744$ ,  $P < 0.001$ ). Given the stringent criteria for a score of 8, a perfect score was achieved in only 16% of the subjects after islet transplant. Once adequate islet mass was provided and insulin independence achieved, the  $\beta$ -score was typically 6–7. The fasting glucose was often above the threshold necessary for a perfect score,  $\geq 5.6$  mmol/l, i.e., the patients would be insulin independent, have C-peptide present, and have an excellent HbA<sub>1c</sub>, but the mean fasting glucose remained slightly elevated. The  $\beta$ -score in



**Figure 1**— $\beta$ -Score in relationship to the plasma glucose level 90 min after consuming a standard mixed meal challenge ( $n = 218$ ) in 57 subjects before and after islet transplantation.

the patients who had a whole-pancreas transplant was  $7.3 \pm 0.3$ , with a median of 8 (interquartile range 7–8).

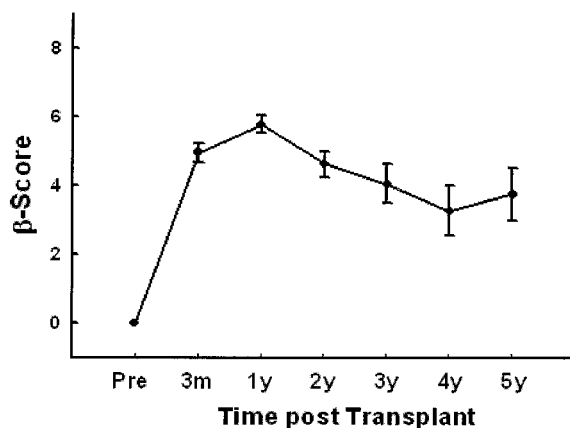
Examination of graft function longitudinally revealed a variety of patterns. Some patients lost graft function and showed a drop in their score, while others maintained good function. The score values over time are shown in Fig. 2. At 3 months, many of the patients were still between their first and second transplants, but once independence was achieved, the score was excellent.

**CONCLUSIONS**— Assessing the adequacy of  $\beta$ -cell function after an islet transplant is difficult. Single measures such as glycemic control, C-peptide secretion, or insulin independence do not suffice, as considering one in the absence of others is inadequate. For this reason, a

broader measurement that encompasses the major aspects of function is required. Such a measure would include the absence of or need for insulin/OHAs to control plasma glucose, measures of glycemia (fasting glucose or  $HbA_{1c}$ ), and an assessment of C-peptide secretion. The importance of maintaining some C-peptide secretion was evident in the Diabetes Control and Complications Trial (9), where the presence of some C-peptide was associated with less retinopathy and severe hypoglycemia. In addition, the benefit of being C-peptide positive after islet transplantation is being recognized in terms of improvement in long-term outcome for a concurrent kidney transplant (10) or diabetes complications (11,12). In terms of graft function, the currently proposed score is useful in that in the single value, an overall sense of how

well the  $\beta$ -cells are functioning can be gleaned. The advantages of the score are the ease of its calculation and its broad intuitive nature. It encompasses whether the patient is truly insulin independent or has some partial function in the setting of the glucose control and subsumes C-peptide secretion. In many ways, it is equivalent to an Apgar score in the neonate, a simple robust measure of outcome.

The  $\beta$ -score has an immediate application in the assessment of graft function after islet transplantation. As with pancreas transplantation, absolute success is easy to define in terms of normal glucose tolerance, insulin independence, and endogenous insulin release. Such a situation would merit a  $\beta$ -score of 8. However, frequently, the outcome is less clear cut. Insulin independence cannot be regarded as the only successful outcome measure, particularly if patients have unacceptable glycemic control or elevated  $HbA_{1c}$ . If a subject has an  $HbA_{1c}$  of 6.3% and a fasting glucose of 6.4 mmol/l and is insulin independent, the outcome is good but not perfect. The  $\beta$ -score in this situation would be 6. Another patient who is off insulin but has a fasting glucose of 7.2 mmol/l, an  $HbA_{1c}$  7.4%, and taking metformin to assist the endogenous insulin reserve that is present but blunted may be content to be off insulin, but the graft is clearly not functioning as well. This last case scenario would have a  $\beta$ -score of 2, indicative of the real situation in terms of graft function. A final scenario might be when the subject is placed on a small dose of insulin “to protect the cells.” In such a case, if all the other parameters were normal, the score would be 7. A score of 7 is clearly good, but if there is some doubt about the graft such that a small dose of insulin is being used, a perfect score should not apply. The  $\beta$ -score in the whole-pancreas transplant patients was excellent, confirming the perfect glucose control commonly found in these subjects. Thus, the score can provide an overall measure of the graft function, whether the setting is that of a whole-pancreas or islet transplant. A negative control group that could be considered are new-onset type 1 diabetic subjects who have some C-peptide secretion, but collecting such a group would be challenging given that only 35% of this population have demonstrable C-peptide secretion and half of



**Figure 2**— $\beta$ -Scores (means  $\pm$  SE) in 44 subjects before and after islet transplantation. Shown are pretransplant ( $n = 30$ ), 3-month ( $n = 26$ ), 1-year ( $n = 41$ ), 2-year ( $n = 31$ ), 3-year ( $n = 18$ ), 4-year ( $n = 11$ ), and 5-year ( $n = 4$ ) values. All the posttransplant values were significantly different from the pretransplant value ( $P < 0.05$ ).

these have lost their endogenous  $\beta$ -cell function by 1 year (9).

A potential drawback is the broad groupings that have been used. When other possibilities of varying degrees of glucose control, insulin use, or C-peptide secretion were tried, it became inordinately complex and difficult to use. We believe that the present scoring system achieves a balance that works. A second disadvantage is that in the presence of insulin resistance, a high C-peptide may be present, but yet the glucose may not be controlled. This may indicate that the cells are actually functioning quite well but reveals insufficient insulin for the degree of insulin resistance. In such a situation, the score will still be helpful in that it will not be normal by virtue of the hyperglycemia but still be in the upper tertile. By keeping to broad groupings rather than allotting extra points for higher C-peptide levels, a compromise has been achieved that is worthwhile. We have previously suggested that the 90-min glucose after a mixed meal challenge is a good measure of endogenous insulin release. While this remains the case, it does not incorporate the issue of ambient glycemic control as in the HbA<sub>1c</sub> and the use of insulin or OHAs, a deficit the  $\beta$ -score overcomes. We see both measures as complimentary: the  $\beta$ -score gives an overall measure of clinical outcome and the 90-min glucose an acute measure of  $\beta$ -cell reserve. Other measures of  $\beta$ -cell function are available, such as the C-peptide level after intravenous arginine (13) and more detailed metabolic tests (14,15), but none of these give a composite measure of the success of the transplant. The C-peptide response after arginine could replace the mixed meal challenge for the determination of the stimulated C-peptide (13), but this is in need of further study. For the  $\beta$ -score, what is required is a C-peptide after any adequate stimulus. A further issue is the weighting used in the score. Both glycemia and insulin secretion account for 75% of the values available, but this simply reflects the reality of the central nature of these components in determining  $\beta$ -cell function after an islet transplant. A point of note is that the score is assessing  $\beta$ -cell function and its impact on glycemia. Other important determinants of a successful islet transplant are measures of lability and hypoglycemia (4), the presence or absence of side effects (16), the

impact on the long-term complications of diabetes, and quality of life measures.

In addition to its obvious use for assessing postislet or postpancreas transplant  $\beta$ -cell function, it is possible that this score could also have a use in the evaluation of new studies proposed for the prevention or immunotherapy of new-onset diabetes. There are many proposed trials attempting to preserve  $\beta$ -cell function in new-onset type 1 diabetes or in subjects identified to be at high risk for type 1 diabetes. A problem that besets all of these studies is standardization of the outcomes. Simple measures of C-peptide without some concomitant determination of glucose are difficult to interpret. Likewise, insulin independence is often not achieved in these patients, so measures of glucose control in subjects who continue on insulin will be inadequate. Composite scores such as that currently proposed may be helpful in this setting. Finally, the  $\beta$ -score allows a comparison of outcomes after transplantation from different transplant centers. Using insulin independence alone is dependent on the investigator's decision as to whether or not to stop insulin, as there are no agreed criteria at what level of glucose should the insulin be discontinued.

In conclusion, we have described a simple scoring system that encompasses glycemic control, diabetes therapy, and  $\beta$ -cell survival suitable as an overall measure of  $\beta$ -cell transplant success. The  $\beta$ -score gives an integrated measure of  $\beta$ -cell function on a continuum that may be more useful than arbitrarily assessing the presence or absence of insulin independence.

**Acknowledgments**—The Clinical Islet Transplant Program receives funds from a Center Grant of Juvenile Diabetes Foundation International. A.M.J.S. is supported by a Canadian Institutes of Health Research (CIHR/Wyeth) Clinical Research Chair in Transplantation and through a Scholarship from the Alberta Heritage Foundation for Medical Research. J.R.T.L. is a recipient of scholarships from the Canadian Diabetes Association and the Alberta Heritage Foundation for Medical Research.

We thank the staff of the Clinical Investigation Unit, University of Alberta Hospitals; Sharleen Imes, Vijay Menon, and the nursing coordinators of the Clinical Islet Transplant Program at the University of Alberta/Capital Health in Edmonton; and the expert secretarial assistance of Louise Bohachyk.

## References

1. Ryan EA, Lakey JR, Paty BW, Imes S, Korbitt GS, Kneteman NM, Bigam D, Rajotte RV, Shapiro AM: Successful islet transplantation: continued insulin reserve provides long-term glycemic control. *Diabetes* 51:2148–2157, 2002
2. Markmann JF, Deng S, Huang X, Desai NM, Velidedeoglu EH, Lui C, Frank A, Markmann E, Palanjian M, Brayman K, Wolf B, Bell E, Vitamaniuk M, Doliba N, Matschinsky F, Barker CF, Naji A: Insulin independence following isolated islet transplantation and single islet infusions. *Ann Surg* 237:741–750, 2003
3. Hering BJ, Kandaswamy R, Harmon JV, Ansite JD, Clemmings SM, Sakai T, Paraskevas S, Eckman PM, Sageshima J, Nakano M, Sawada T, Matsumoto I, Zhang HJ, Sutherland DE, Bluestone JA: Transplantation of cultured islets from two-layer preserved pancreases in type 1 diabetes with anti-CD3 antibody. *Am J Transplant* 4:390–401, 2004
4. Ryan EA, Shandro T, Green K, Paty BW, Senior PA, Bigam D, Shapiro AM, Vantghem MC: Assessment of the severity of hypoglycemia and glycemic lability in type 1 diabetic subjects undergoing islet transplantation. *Diabetes* 53:955–962, 2004
5. Goss JA, Goodpastor SE, Brunnicardi FC, Barth MH, Soltes GD, Garber AJ, Hamilton DJ, Alejandro R, Ricordi C: Development of a human pancreatic islet-transplant program through a collaborative relationship with a remote islet-isolator center. *Transplantation* 77:462–466, 2004
6. Kessler L, Bucher P, Milliat-Guittard L, Benhamou PY, Berney T, Penformis A, Badet L, Thivolet C, Bayle F, Oberholzer J, Renoult E, Brun MJ, Riffle G, Atlan C, Colin C, Morel P, the GRAGIL Group: Influence of islet transportation on pancreatic islet allotransplantation in type 1 diabetic patients with the Swiss-French GRAGIL network. *Transplantation* 77:1301–1304, 2004
7. Sutherland DE, Gruessner A, Hering BJ:  $\beta$ -Cell replacement therapy (pancreas and islet transplantation) for treatment of diabetes mellitus: an integrated approach. *Endocrinol Metab Clin North Am* 33:135–148, 2004
8. American Diabetes Association: Diagnosis and classification of diabetes mellitus (Position Statement). *Diabetes Care* 27 (Suppl. 1):S5–S10, 2004
9. Diabetes Control and Complications Trial Research Group: Effect of intensive therapy on residual  $\beta$ -cell function in patients with type 1 diabetes in the Diabetes Control and Complications Trial. *Ann Int Med* 128:517–523, 1998
10. Fiorina P, Folli F, Zerbin G, Maffi P, Gremizzi C, Di Carlo V, Socci C, Bertuzzi F, Kashgarian M, Secchi A: Islet trans-

- plantation is associated with improvement of renal function among uremic patients with type 1 diabetes mellitus and kidney transplants. *J Am Soc Nephrol* 14: 2150–2158, 2003
11. Fiorina P, Folli F, Bertuzzi F, Maffi P, Finzi G, Venturini M, Socci C, Davalli A, Orsenigo E, Monti L, Falqui L, Uccella S, La Rosa S, Usellini L, Properzi G, Di Carlo V, Del Maschio A, Capella C, Secchi A: Long-term beneficial effect of islet transplantation on diabetic macro-/micro-angiopathy in type 1 diabetic kidney-transplanted patients. *Diabetes Care* 26: 1129–1136, 2003
  12. Fiorina P, Folli F, Maffi P, Placidi C, Venturini M, Finzi G, Bertuzzi F, Davalli A, D'Angelo A, Socci C, Gremizzi C, Orsenigo E, La Rosa S, Ponzoni M, Cardillo M, Scalamogna M, Del Maschio A, Capella C, Di Carlo V, Secchi A: Islet transplantation improves vascular diabetic complications in patients with diabetes who underwent kidney transplantation: a comparison between kidney-pancreas and kidney-alone transplantation. *Transplantation* 75:1296–1301, 2003
  13. Greenbaum C, Seidel K, Pihoker C: The case for intravenous arginine stimulation in lieu of mixed-meal tolerance tests as outcome measure for intervention studies in recent-onset type 1 diabetes. *Diabetes Care* 27:1202–1204, 2004
  14. Luzi L, Perseghin G, Brendel MD, Terruzzi I, Battezzati A, Eckhard M, Brandhorst D, Brandhorst H, Friemann S, Socci C, Di Carlo V, Piceni Sereni L, Benedini S, Secchi A, Pozza G, Bretzel RG: Metabolic effects of restoring partial  $\beta$ -cell function after islet allotransplantation in type 1 diabetic patients. *Diabetes* 50:277–282, 2001
  15. Teuscher AU, Kendall DM, Smets YF, Leone JP, Sutherland DE, Robertson RP: Successful islet autotransplantation in humans: functional insulin secretory reserve as an estimate of surviving islet cell mass. *Diabetes* 47:324–330, 1998
  16. Hirshberg B, Rother KI, Digion BJ 3rd, Lee J, Gaglia JL, Hines K, Read EJ, Chang R, Wood BJ, Harlan DM: Benefits and risks of solitary islet transplantation for type 1 diabetes using steroid-sparing immunosuppression. *Diabetes Care* 26:3288–3295, 2003