Comparison of the Glucose Dependency of Glucagon-Like Peptide-1(7-37) and Glyburide In Vitro and In Vivo

Diane M. Hargrove, Nancy A. Nardone, Lorna M. Persson, Kim M. Andrews, Kandace L. Shepherd, Ralph W. Stevenson, and Janice C. Parker

The purpose of the present study was to compare the glucose dependency of the insulin secretagogue activity of the sulfonylurea, glyburide, versus that of glucagon-like peptide-1(7-37) [GLP-1(7-37)] in vitro and in vivo. In freshly isolated rat islets, maximally effective concentrations of glyburide (10 μmol/L) and GLP-1(7-37) (10 nmol/L) were equally effective in stimulating insulin secretion in the presence of 15 mmol/L glucose (2.4-fold increase relative to 15 mmol/L glucose alone). At 5 mmol/L glucose, both agents increased insulin secretion, but the effect for glyburide was threefold greater than for GLP-1(7-37) (122% and 41% increase in insulin secretion, respectively). In conscious catheterized rats infused with glucose at a variable rate to clamp plasma glucose concentration at 11 mmol/L, glyburide (1 mg/kg orally) and GLP-1(7-37) (infused intravenously [IV] at 5 pmol/min/kg) produced similar increases in insulin levels (1.8-fold relative to the respective vehicle controls) that were sustained through 60 minutes of measurement. These doses of GLP-1(7-37) and glyburide were then administered to fasted and fed rats (basal plasma glucose concentration, 5.8 and 7.3 mmol/L, respectively). Relative to the vehicle control group, GLP-1(7-37) infusion produced a transitory increase (30%) in plasma insulin concentration and a modest sustained decrease (10% to 20%) in glucose in both fasted and fed rats, whereas glyburide induced a sustained 2.4- and 1.7-fold increase in plasma insulin concentration in fasted and fed rats, respectively, and a 50% decrease in plasma glucose in both fasted and fed rats. Results of these studies demonstrate the higher glucose threshold for the insulin secretagogue activity of GLP-1(7-37) relative to glyburide in vitro and in vivo.

SECOND-GENERATION sulfonylureas such as glyburide and glipizide and first-generation sulfonylureas such as tolbutamide and chlorpropamide are insulin secretagogues that have been used extensively for treatment of non-insulin-dependent diabetes mellitus (NIDDM). Sulfonylureas stimulate insulin secretion through interaction with pancreatic adenosine triphosphate–sensitive potassium channels. Although they are effective in many patients, there is a high rate of primary and secondary failure, and because they stimulate insulin secretion at low glucose concentrations, there is a risk of hypoglycemia in those patients who respond well to sulfonylureas. Therefore, there is a need for new agents that stimulate insulin secretion by a mechanism distinct from that of sulfonylureas.

Truncated glucagon-like peptides-1 (GLP-1(7-37) and GLP-1(7-36)-amide) are naturally occurring peptides that are synthesized primarily in the intestine and secreted following a meal or glucose challenge.[1-3] Several lines of evidence suggest that endogenous GLP-1(7-37)/(7-36)-amide act physiologically as insulin secretagogues and may function as incretins.[4-6] GLP-1(7-37) and GLP-1(7-36)-amide have potent insulin secretagogue activity in isolated islets, perfused pancreas, and cultured β-cell lines.[7-12] Specific receptors for the GLPs have been demonstrated on rat insulinoma cell lines[13,14] and rat islets of Langerhans,[15] where their activity is linked to the stimulation of adenyl cyclase.[16] Exogenous administration of GLP-1(7-37) or GLP-1(7-36)-amide has been shown to increase circulating insulin levels and decrease blood glucose in animal models and humans.[3,17-26]

Because of their potent insulin secretagogue activity and different mechanism of action versus sulfonylureas, truncated GLPs represent potential therapeutic agents for treatment of NIDDM. There are no reports that directly compare GLPs and sulfonylureas for effects on insulin secretion.

The present studies were conducted to compare the glucose dependency of the insulin secretagogue activity of GLP-1(7-37) and glyburide both in vitro and in vivo to examine whether GLP-1(7-37) is likely to have advantages over the sulfonylureas.

MATERIALS AND METHODS

In Vitro Studies

Rat islets. Rat islets were isolated by a modification of the method of Lacy and Kostianovsky,[27] in which the collagenase digest of pancreatic tissue was separated on a Ficoll gradient (27%, 23%, 20.5%, and 11% in Hanks balanced salt solution, pH 7.4). Islets were collected from the 20.5%/11% interface, washed, and hand-picked free of exocrine and other tissue under a stereomicroscope. Groups of eight isolated islets were preincubated in glucose-free buffer (Krebs-Ringer bicarbonate buffer, pH 7.4 at ambient CO_2, containing 0.25% bovine serum albumin) for 1 hour at 37°C. They were then transferred to 500 μL buffer containing 5 or 15 mmol/L glucose with or without drug and incubated for 30 minutes at 37°C. An aliquot of islet-free medium was collected for determination of insulin concentration.

In Vivo Studies

The effects of glyburide and GLP-1(7-37) on insulin concentrations were studied in conscious catheterized male Sprague-Dawley rats at three levels of glycemia: 11 (clamped), 5.8 (fasted), and 7.3 mmol/L (fed).

Animals and surgical preparation. Rats (body weight, 300 to 350 g; Charles River, Wilmington, MA) were housed in an environmentally controlled room (21°C with a 12-hour light/dark cycle) with food (standard rodent chow) and water available ad

From the Department of Metabolic Diseases, Pfizer Central Research, Groton, CT.

Submitted August 1, 1995; accepted August 27, 1995.

Address reprint requests to Diane M. Hargrove, PhD, Pfizer Inc, Central Research Division, Eastern Point Road, Groton, CT 06340.

0026-0495/96/4503-0019$03.00/0

libitum. For surgical implantation of catheters, rats were anesthetized (70 mg/kg ketamine and 8.5 mg/kg xylazine by intramuscular injection), and PE50 tubing was implanted in the left carotid artery and right jugular vein. The catheters were exteriorized at the back of the neck, filled with a polyvinylpyrrolidone solution in heparinized saline, flame-sealed, and secured with tape. Rats were housed individually and given 7 days' recovery from the surgery. On the day of the experiment, the catheters were untaped and flushed with saline and the animals were allowed to settle for at least 30 minutes before initiation of the experiment. During the experiment, rats were allowed to move about freely.

Hyperglycemic clamp. A 2-hour hyperglycemic clamp was performed in fed rats. Glucose (50% solution) was infused at a variable rate to maintain plasma glucose concentration at 11 mmol/L. Blood samples (30 μL) for glucose determination were taken every 5 minutes and immediately centrifuged. Plasma was analyzed for glucose concentration, and the exogenous rate of glucose infusion was adjusted accordingly. Samples (250 μL) for insulin determination were taken every 15 to 30 minutes from the carotid artery catheter. Replacement blood (6 mL whole rat blood obtained by decapitation and collected through gauze into tubes containing 4 mL saline and 1 mL heparin) was administered (500 μL) via the carotid artery catheter every 15 minutes. GLP-1(7-37) (5 pmol/kg/min) or vehicle (2% heat-inactivated serum + 1% sodium acetate in normal saline) were infused IV from 60 to 120 minutes. Glyburide (1 mg/kg by gavage) or vehicle (0.05N NaOH) were administered at 60 minutes of the 2-hour clamp. At the end of the clamp, a blood sample was taken from rats infused with GLP-1(7-37), into tubes containing EDTA and aprotinin for analysis of GLP-1(7-37) concentration.

Fasted and fed rats. Fasted rats had food removed at 4 to 5 PM the evening before the experiment. On the day of the experiment, the catheters were exteriorized and the animals were allowed to settle. Basal samples for glucose and insulin determination were taken, and treatment with GLP-1(7-37) (5 pmol/kg/min infused IV for 60 minutes), glyburide (1 mg/kg orally), or vehicle (2% heat-inactivated serum and 1% sodium acetate in normal saline infused IV at 1 mL/h plus 0.5 mL 0.05N NaOH orally) was initiated. To control for the effects of handling and vehicle, GLP-1(7-37)–infused rats were administered the glyburide vehicle by gavage (0.05N NaOH 1 mL), glyburide-treated rats were infused IV with GLP-1(7-37) vehicle (2% heat-inactivated serum + 1% sodium acetate in normal saline IV), and vehicle control rats were given both vehicles. Blood samples were taken from the carotid artery catheter at −1, 15, 30, and 60 minutes for plasma insulin and every 5 minutes for plasma glucose determinations. Replacement blood (0.5 mL prepared as described above) was given every 15 minutes. After the 60-minute insulin sample was collected, a blood sample was taken from rats infused with GLP-1(7-37), into tubes containing EDTA and aprotinin for analysis of GLP-1(7-37) concentration.

Analytical Methods

Plasma glucose was determined either with the glucose oxidase method using a Beckman Glucose Analyzer 2 (Beckman Instruments, Brea, CA) or spectrophotometrically (hexokinase and glucose-6-phosphate dehydrogenase) on an Abbott VP Supersystem (Abbott Laboratories, North Chicago, IL). Plasma and islet-free media insulin concentrations were determined by a radioimmunoassay kit (Binax, Portland, ME). Plasma GLP-1(7-37) concentration was determined on unextracted plasma by radioimmunoassay (kit from Peninsula Laboratories, Belmont, CA). GLP-1(7-37) was obtained from Bachem (Torrance, CA).

Statistical Analysis

Statistical evaluation of the data was performed with one-way ANOVA using StatView 512+ computer software (Brain Power, Agoura Hills, CA). When ANOVA indicated significant differences ($P < .05$), a Fisher's protected least-significant difference test was performed to determine which means were statistically different.

RESULTS

In Vitro Studies

In pilot studies with freshly isolated islets, 10 nmol/L GLP-1(7-37) and 10 μmol/L glyburide were maximally effective concentrations for stimulation of insulin secretion, and these concentrations were used for the comparison of glucose dependency. At 15 mmol/L glucose, GLP-1(7-37) and glyburide induced a similar stimulation of insulin secretion (2.4-fold relative to 15 mmol/L glucose without drug). At 5 mmol/L glucose, both agents increased insulin secretion; however, the effect of glyburide was threefold greater ($P < .05$) than the effect of GLP-1(7-37) (Fig 1).

In Vivo Studies

Effects of glyburide and GLP-1(7-37) during hyperglycemia. In previous studies in rats,[20] we have shown that GLP-1(7-37) at 5 pmol/kg/min (IV) is a submaximal dose for increasing insulin concentration during an 11 mmol/L hyperglycemic clamp. In pilot hyperglycemic clamp studies in which glyburide doses of 0.05, 1, and 20 mg/kg were tested, a dose of 1 mg/kg produced insulin secretagogue activity comparable to GLP-1(7-37) at 5 pmol/min/kg (data not shown). Thus, a side-by-side comparison was performed using 5 pmol/kg/min GLP-1(7-37) and 1 mg/kg (orally) glyburide. During the first 60 minutes of the clamp (glucose infusion only), insulin increased from 252 to 858 pmol/L, with a glucose infusion rate of 183 μmol/min/kg (mean of all the treatment groups). There were no differences between the groups at the basal or 60-minute time points (Table 1). Glyburide (given orally at 60 minutes of the clamp) and GLP-1(7-37) (infused IV from 60 to 120 minutes of the clamp) each enhanced the glucose-induced increase in plasma insulin concentration by 1.8-fold ($P < .05$) relative to values in the respective vehicle-treated groups during the last 30 minutes of the clamp. There was no difference between glyburide and GLP-1(7-37) in the increase in insulin concentration (Table 1), and the effect of the two agents was sustained through 60 minutes (Fig 2). Relative to the respective vehicle groups, the glucose infusion rate required to maintain glucose at 11 mmol/L was increased 54% in both the GLP-1(7-37) and the glyburide groups ($P < .05$ for drug *v* respective vehicle; Table 1).

Comparison of GLP-1(7-37) and glyburide in fed and fasted Sprague-Dawley rats. Glyburide and GLP-1(7-37) were tested in fed and fasted Sprague-Dawley rats for effects on glucose and insulin concentrations at the doses that caused similar increases in insulin levels during the hyperglycemic clamp.

A 1-hour infusion of GLP-1(7-37) produced transitory

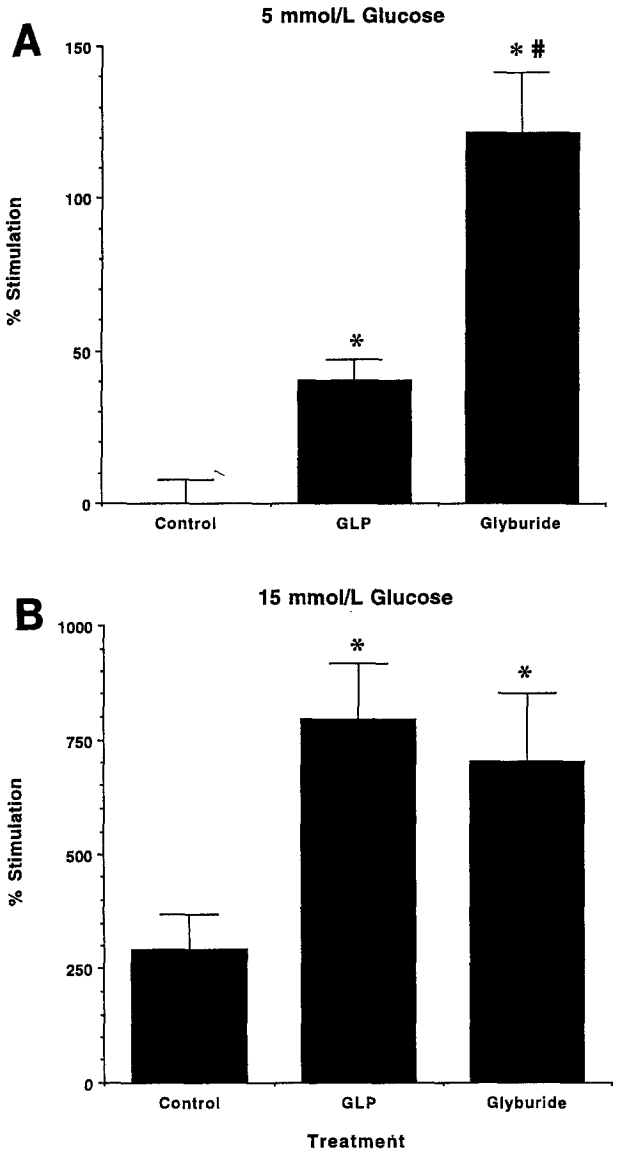

Fig 1. Insulin secretion from freshly isolated rat islets in static incubation at 5 mmol/L glucose (A) or 15 mmol/L glucose (B) in the absence of drug (control) or in the presence of GLP-1(7-37) (10 nmol/L) or glyburide (10 μmol/L) as indicated. Data are expressed as % stimulation relative to the mean control value at 5 mmol/L glucose (which was 221 ± 36 pmol/L, n = 14). Values are the mean ± SEM (n = 12 to 16). *Significant difference between drug and control ($P < .05$). #Significant difference between glyburide and GLP-1(7-37).

increases (~30%) in insulin relative to the vehicle-treated control groups; however, under these conditions, the increase was not statistically significant ($P > .05$; Fig 3). GLP-1(7-37) infusion induced sustained significant decreases in glucose concentrations (10% and 14% to 20% in fasted and fed groups, respectively, relative to the time-matched values in vehicle-treated control groups, $P < .05$) beginning at 45 minutes in fasted rats and at 15 minutes in fed rats (Fig 3). It is noteworthy that there were small increases in glucose and insulin from baseline in the vehicle-treated group, probably as a result of handling. GLP-1(7-37) attenuated vehicle-induced increases in glucose, but glucose values did not decrease to less than baseline. GLP-1(7-37) concentrations in fasted and fed rats infused with GLP-1(7-37) were 227 ± 14 and 187 ± 21 pmol/L, respectively.

In both fasted and fed rats, glyburide induced an increase in insulin and a decrease in glucose ($P < .05$) at 15 minutes postdose, and the effects were sustained through 60 minutes of measurement (Fig 3). In comparison to vehicle-treated controls, glyburide induced a 2.3- and 1.8-fold increase in insulin in fasted and fed rats, respectively, and an approximately 50% decrease in glucose in both groups. At most time points tested, glyburide had a greater ($P < .05$) effect on glucose and insulin concentrations than GLP-1(7-37) in both fasted and fed rats (Fig 3).

DISCUSSION

These results demonstrate the lower glucose threshold for insulin secretagogue activity of the second-generation sulfonylurea, glyburide, relative to GLP-1(7-37) in vitro and in vivo.

In rat pancreatic islets, glyburide stimulated insulin secretion in the presence of 5 mmol/L glucose (Fig 1), a concentration corresponding to fasting glycemia in rats and humans. GLP-1(7-37) had a significantly smaller effect on insulin secretion from islets at 5 mmol/L glucose, although at 15 mmol/L glucose, its maximal effect was comparable to that of a maximally effective concentration of glyburide (Fig 1). Thus, GLP-1(7-37) stimulated insulin secretion most effectively at concentrations that simulated hyperglycemia. These data are consistent with the observations reported by Holz et al,[28] who found that in rat pancreatic β cells the inhibition by GLP-1(7-37) of adenosine triphosphate–sensitive K^+ channels exhibited an absolute requirement for the presence of glucose, whereas the inhibition of these channels by glyburide was more direct and glucose-independent.

The higher glucose threshold for insulin secretagogue activity of GLP-1(7-37) relative to glyburide was also demonstrated in vivo. At appropriate doses, the two drugs induced identical changes in circulating insulin under the conditions of a hyperglycemic clamp (Table 1 and Fig 2). However, when these same doses were administered to fed and fasted rats, differences in glycemic responses to the two drugs became apparent. In fasted animals, GLP-1(7-37) was essentially without effect on plasma glucose and insulin levels (Fig 3). In contrast, glyburide administration to fasted animals stimulated plasma insulin to levels comparable to those observed in the fed animals, causing a corresponding decrease in plasma glucose to less than 3.4 mmol/L. In fed animals, effects of the two drugs were less strikingly different (Fig 3). Both caused an increase in circulating insulin, although the effect of GLP-1(7-37) did not achieve statistical significance relative to the corresponding vehicle control. Both drugs caused a decrease in circulating glucose. The glucose level achieved in glyburide-treated animals was significantly less than that in GLP-1(7-37)–treated animals. Higher doses of GLP-1(7-37) pro-

Table 1. Plasma Glucose and Insulin Concentrations and Glucose Infusion Rate During the 11-mmol/L Hyperglycemic Clamp Before (0-60 min) and After (90-120 min) Administration of Glyburide (1 mg/kg, orally) or GLP-1(7-37) (5 pmol/min/kg IV) Starting at 60 Minutes

	Glucose Concentration (mmol/L)			Insulin Concentration (pmol/L)			Glucose Infusion Rate (μmol/min/kg)	
Treatment	0 min	60 min	90-120 min	0 min	60 min	90-120 min	60 min	90-120 min
Vehicle (orally)	6.7 ± 0.3	11 ± 0.3	11.5 ± 0.2	258 ± 18	900 ± 72	936 ± 156	178 ± 11	167 ± 17
Glyburide (1 mg/kg orally)	6.9 ± 0.3	11.1 ± 0.5	11.1 ± 0.2	264 ± 30	948 ± 48	1,660 ± 174*	189 ± 17	255 ± 22*
Vehicle (IV)	6.8 ± 0.3	10.7 ± 0.2	11.1 ± 0.2	228 ± 30	900 ± 102	980 ± 120	194 ± 17	205 ± 17
GLP-1(7-37) (5 pmol/min/kg IV)	6.9 ± 0.3	10.6 ± 0.3	11.2 ± 0.1	264 ± 24	804 ± 138	1,760 ± 240*	178 ± 17	316 ± 6*†

NOTE. Values are the mean ± SE (n = 5 to 7). For 90- to 120-minute values, measurements of glucose infusion rate, and glucose and insulin concentrations obtained every 5 to 30 minutes during the last 30 minutes of the clamp were averaged for each rat and mean group values are presented.

*Significantly different from corresponding vehicle-treated group ($P < .05$).

†Significant difference between GLP-1(7-37)– and glyburide-treated groups ($P < .05$).

duced no additional effect on insulin secretion or glucose levels in fed or fasted rats (data not shown). The transitory insulin response to GLP-1(7-37) under fasted conditions was probably a consequence of the glucose concentration decreasing to less than the threshold for stimulation of insulin secretion by GLP-1(7-37), since GLP-1(7-37) induced a sustained stimulation of insulin secretion when glucose was maintained at a hyperglycemic level (Fig 2). Consistent with this interpretation, in a previous report, GLP-1(7-37) infusion induced a transitory increase in insulin and a sustained decrease in glucose (to 5.8 mmol/L) in fed Zucker diabetic fatty rats (an animal model with hyperglycemia and hyperinsulinemia) and their lean littermates. However, in fed and fasted Sprague-Dawley rats, GLP-1(7-37) induced a transitory effect on both glucose and insulin, with a threshold for insulin secretagogue activity of 5.3 mmol/L in fasted rats and 5.8 mmol/L in fed rats.[20] The apparently higher glucose threshold and the more sustained glucose-lowering effect in the present report may be the result of differences in handling. In the previous report, rats received an IV infusion only; in the present report, they also received a vehicle gavage to compare the data with those obtained in glyburide-treated rats, and the gavage produced a modest increase in glucose.

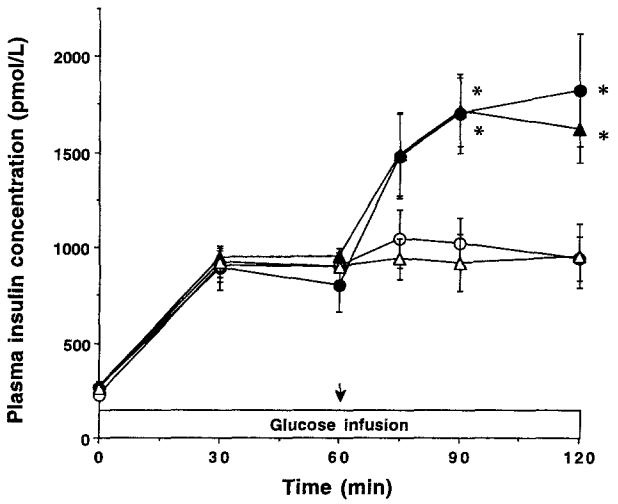

Fig 2. Time course of insulin concentration during the 11-mmol/L hyperglycemic clamp. Catheterized Sprague-Dawley rats were infused with glucose at a variable rate to maintain plasma glucose at 11 mmol/L for 120 minutes. At 60 minutes of glucose infusion, rats received either GLP-1(7-37) infusion (5 pmol/min/kg IV, ●), IV vehicle infusion (○), glyburide (1 mg/kg orally, ▲), or vehicle (orally, △). Values are the mean ± SEM (n = 6 to 7). *Significant difference between drug and corresponding vehicle ($P < .05$).

In nondiabetic human subjects, GLP-1(7-37)/GLP-1(7-36)-amide appears to have a lower glucose threshold of action than in the rat, as demonstrated by a small decrease in plasma glucose in subjects with fasting plasma glucose levels of 5 mmol/L[24] and approximately 4.7 mmol/L.[26] However, this action was transitory, since plasma insulin levels started to decline when plasma glucose decreased to 4.1 to 4.4 mmol/L, suggesting a similar glucose dependency of action in the rat, but with a slightly lower glucose threshold. In patients with NIDDM, GLP-1(7-37)/GLP-1(7-36)-amide increased insulin secretion for longer periods[24,25] and in the latter study, plasma glucose was reduced to a nadir of 4.9 mmol/L after 4 hours' administration. It is noteworthy that this activity of the incretin, GLP-1(7-37), represents an amplification of the peptide's normal role to stimulate insulin secretion, that leads to a reduction in hyperglycemia in patients with NIDDM, but that another putative incretin, GIP, is ineffective in NIDDM, in contrast to its effects in nondiabetics.[17,25]

We conclude that although glyburide and GLP-1(7-37) are both effective insulin secretagogues and hence have glucose-lowering activity, they differ in the glucose threshold for stimulation of insulin secretion. Glyburide stimulates insulin secretion even at glucose concentrations that correspond to hypoglycemia. However, GLP-1(7-37) is relatively inert at low glucose concentrations and becomes an effective secretagogue only at glucose concentrations that correspond to hyperglycemia. As glucose decreases toward euglycemic levels, the effectiveness of GLP-1(7-37) as an insulin secretagogue diminishes and once euglycemia is achieved, apparently ceases, despite continued infusion of the drug. Thus, the effects of GLP-1(7-37) are, in effect, self-limiting. It follows that GLP-1(7-37) is less likely than glyburide to cause hypoglycemia in vivo. This property has important implications for its therapeutic potential in the treatment of NIDDM.

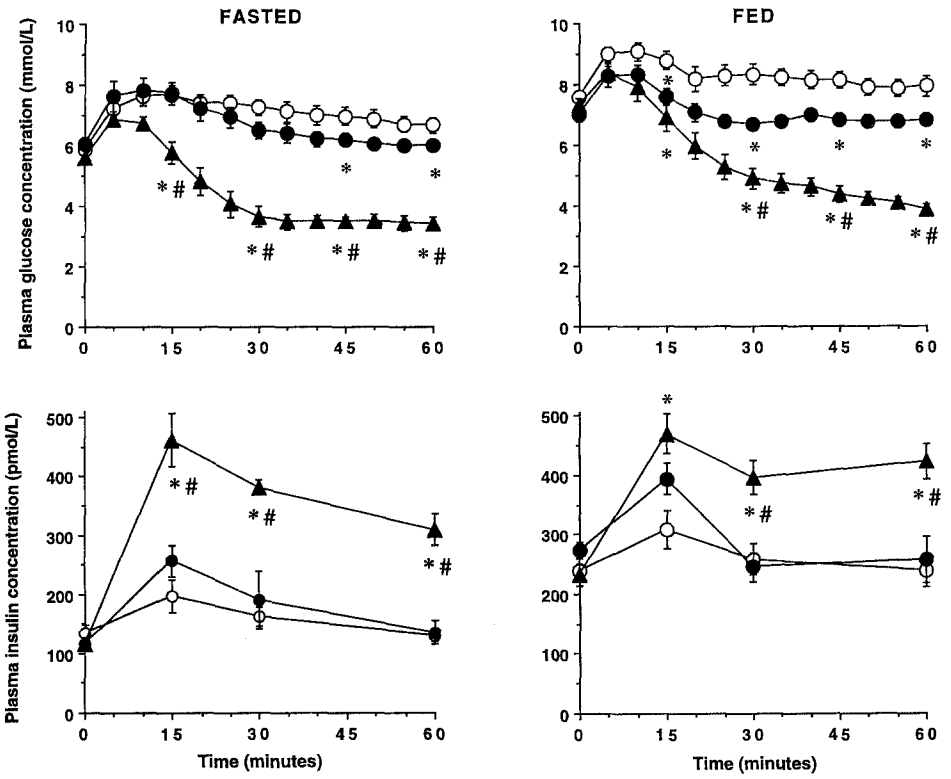

Fig 3. Time course of insulin and glucose concentrations in fasted (left) and fed (right) catheterized Sprague-Dawley rats treated with vehicle (2% heat-inactivated serum + 1% sodium acetate in normal saline IV and 0.05N NaOH orally, ○), GLP-1(7-37) (5 pmol/min/kg IV, ●), or glyburide (1 mg/kg orally, ▲). Values are the mean ± SEM (n = 7 to 10). *Significant difference between drug and corresponding vehicle ($P < .05$). #Significant difference between glyburide and time-matched GLP-1(7-37). For clarity, significance symbols for glucose concentration time course graphs are shown at 15-minute intervals.

REFERENCES

1. Mojsov S, Kopczynski MG, Habener JF: Both amidated and nonamidated forms of glucagon-like peptide I are synthesized in the rat intestine and the pancreas. J Biol Chem 265:8001-8008, 1990
2. Ørskov C, Rabenhøj L, Wettergren A, et al: Tissue and plasma concentrations of amidated and glycine-extended glucagon-like peptide I in humans. Diabetes 43:535-539, 1994
3. Fridolf T, Bottcher G, Sundler F, et al: GLP-1 and GLP-1(7-36)amide: Influences on basal and stimulated insulin and glucagon secretion in the mouse. Pancreas 6:208-215, 1991
4. Habener JF: The incretin notion and its relevance to diabetes. Endocrinol Metab Clin North Am 22:775-794, 1993
5. Weir GC: Glucagon-like peptide-1 (GLP-1): A piece of the incretin puzzle. J Clin Invest 95:1, 1995
6. Wang Z, Wang RM, Owji AA, et al: Glucagon-like peptide-1 is a physiological incretin in rat. J Clin Invest 95:417-421, 1995
7. Drucker DJ, Philippe J, Mojsov S, et al: Glucagon-like peptide I stimulates insulin gene expression and increases cyclic AMP levels in a rat islet cell line. Proc Natl Acad Sci USA 84:3434-3438, 1987
8. Goke R, Wagner B, Fehmann H-C, et al: Glucose-dependency of the insulin stimulatory effect of glucagon-like peptide-1(7-36) amide on the rat pancreas. Res Exp Med 193:97-103, 1993
9. Schmid R, Schusdziarra V, Aulehner R, et al: Comparison of GLP-1(7-36 amide) and GIP on release of somatostatin-like immunoreactivity and insulin from the isolated rat pancreas. Z Gastroenterol 28:280-284, 1990
10. Siegel EG, Schulze A, Schmidt WE, et al: Comparison of the effect of GIP and GLP-1(7-36 amide) on insulin release from rat pancreatic islets. Eur J Clin Invest 22:154-157, 1992
11. Weir GC, Mojsov S, Hendrick GK, et al: Glucagonlike peptide 1(7-37) actions on endocrine pancreas. Diabetes 38:338-342, 1989
12. Zawalich WS, Zawalich KC, Rasmussen H: Influence of glucagon-like peptide-1 on β cell responsiveness. Regul Pept 44:277-283, 1993
13. Goke R, Conlon JM: Receptors for glucagon-like peptide-1(7-36) amide on rat insulinoma-derived cells. J Endocrinol 116:357-362, 1988
14. Ørskov C, Neilsen JH: Truncated glucagon-like peptide-1 (proglucagon 78-107 amide), an intestinal insulin-releasing peptide, has specific receptors on rat insulinoma cells (RIN 5AH). FEBS Lett 229:175-178, 1988
15. Ørskov C, Poulsen SS: Glucagon-like peptide-1-(7-36)-amide receptors only in islets of Langerhans: Autoradiographic survey of extracerebral tissues in rats. Diabetes 40:1292-1296, 1991
16. Thorens B: Expression cloning of the pancreatic β cell receptor for the gluco-incretin hormone glucagon-like peptide 1. Proc Natl Acad Sci USA 89:8641-8645, 1992
17. Elahi D, McAloon-Dyke M, Fukagawa NK, et al: The insulinotropic actions of glucose-dependent insulinotropic polypeptide (GIP) and glucagon-like peptide-1(7-37) in normal and diabetic subjects. Regul Pept 51:63-74, 1994
18. Faulkner A, Pollock HT: Effects of truncated glucagon-like peptide-1 on the responses of starved sheep to glucose. J Endocrinol 129:55-58, 1991
19. Gutniak M, Ørskov C, Holst JJ, et al: Antidiabetogenic effect of glucagon-like peptide-1(7-36) amide in normal subjects and patients with diabetes mellitus. N Engl J Med 326:1316-1322, 1992
20. Hargrove DM, Nardone NA, Persson LM, et al: Glucose dependent action of glucagon-like peptide-1(7-37) in vivo during acute or chronic administration. Metabolism 44:1231-1237, 1995
21. Hendrick GK, Gjinovci A, Baxter LA, et al: Glucagon-like peptide-1-(7-37) suppresses hyperglycemia in rats. Metabolism 42:1-6, 1993

22. Kawai K, Suzuki S, Ohashi S, et al: Effects of truncated glucagon-like peptide-1 on pancreatic hormone release in normal conscious dogs. Acta Endocrinol (Copenh) 123:661-667, 1990

23. Kreymann B, Williams G, Ghatei MA, et al: Glucagon-like peptide-1 7-36: A physiological incretin in man. Lancet 2:1300-1304, 1987

24. Nathan DM, Schreiber E, Fogel H, et al: Insulinotropic action of glucagonlike peptide-1-(7-37) in diabetic and nondiabetic subjects. Diabetes Care 15:270-276, 1992

25. Nauck MA, Heimesaat MM, Ørskov C, et al: Preserved incretin activity of glucagon-like peptide 1[7-36 amide] but not of synthetic human gastric inhibitory polypeptide in patients with type-2 diabetes mellitus. J Clin Invest 91:301-307, 1993

26. Ørskov C, Wettergren A, Holst JJ: Biological effects and metabolic rates of glucagonlike peptide-1 7-36 amide and glucagon-like peptide-1 7-37 in healthy subjects are indistinguishable. Diabetes 42:658-661, 1993

27. Lacy PE, Kostianovsky M: Method for the isolation of intact islets of Langerhans from the rat pancreas. Diabetes 16:35-39, 1967

28. Holz GG IV, Kuhtreiber WM, Habener JF: Pancreatic beta-cells are rendered glucose-competent by the insulinotropic hormone glucagon-like peptide-1(7-37). Nature 361:362-365, 1993