ROLE OF REDUCED SUPPRESSION OF GLUCOSE PRODUCTION AND DIMINISHED EARLY INSULIN RELEASE IN IMPAIRED GLUCOSE TOLERANCE

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Abstract *Background.* Insulin resistance and impaired insulin secretion both occur in non-insulin-dependent diabetes (NIDDM), but their relative importance is unclear. Hyperglycemia itself has adverse effects on tissue insulin sensitivity and insulin secretion that make it difficult to distinguish between primary and secondary abnormalities. To avoid this problem we studied subjects with postprandial glucose intolerance but not sustained hyperglycemia.

Methods. We compared the rate of systemic appearance and disappearance of glucose, the output of endogenous hepatic glucose, splanchnic and muscle uptake of glucose, and plasma insulin and glucagon responses after the ingestion of 1 g of glucose per kilogram of body weight in 15 subjects with impaired glucose tolerance (8 of them nonobese and 7 obese) and in 16 normal subjects (9 nonobese and 7 obese) who were matched for age and weight.

Results. After glucose ingestion the mean (±SE) rate of total systemic appearance of glucose was significantly higher in both the nonobese subjects (455±12 mmol per five hours) and the obese subjects (486±17 mmol per five hours) with impaired glucose tolerance than in the respective normal subjects (411±11 and 436±7 mmol per five hours). This difference was fully accounted for by the reduced suppression of endogenous hepatic glucose in the subjects with impaired glucose tolerance (a reduction of about 28 percent, vs. 48 percent in the normal subjects; $P<0.01$). Despite late hyperinsulinemia, at 30 minutes the subjects with impaired glucose tolerance had smaller increases in plasma insulin and smaller reductions in plasma glucagon (both $P<0.01$). Molar ratios of plasma insulin to plasma glucagon levels correlated inversely ($r = -0.62$, $P<0.001$) with the rates of systemic glucose appearance; the latter correlated positively ($r = 0.72$, $P<0.0001$) with peak plasma glucose concentrations.

Conclusions. Impaired glucose tolerance, the precursor of NIDDM, results primarily from reduced suppression of hepatic glucose output due to abnormal pancreatic islet-cell function. The late hyperinsulinemia may be the consequence of an inadequate early beta-cell response rather than of insulin resistance. (N Engl J Med 1992;326: 22-9.)

IMPAIRED insulin secretion and insulin resistance are prominent features of non-insulin-dependent diabetes mellitus (NIDDM).[1,2] Despite extensive investigation, the role of these abnormalities in the pathogenesis of NIDDM remains controversial.[1-3] A major reason for this uncertainty is that hyperglycemia itself can impair insulin secretion[4-6] and cause insulin resistance.[5-7] Most, if not all, patients with NIDDM initially have impaired glucose tolerance for a period,[8] during which factors important for the development of NIDDM may already be present. To circumvent the confounding effects of glucose toxicity,[4-7] several investigators have recently undertaken studies of insulin secretion and action in patients with impaired glucose tolerance.[7,9-11]

Studies of the relation between the plasma glucose and insulin responses that occur two hours after oral glucose administration in patients with impaired glucose tolerance or mild NIDDM show that, up to a point, the plasma insulin level increases as plasma glucose increases, but it then decreases with further increases in plasma glucose.[10,12-18] This relation has been interpreted by some investigators to indicate that insulin resistance is already present in people with impaired glucose tolerance and that impaired insulin secretion is a later event.[1,10,12,15-18] However, since the plasma glucose concentration is the principal stimulus for insulin secretion, a higher two-hour plasma glucose concentration should be associated with a higher two-hour plasma insulin level. Indeed, there is evidence that the increased two-hour plasma insulin concentration in patients with impaired glucose tolerance may be inappropriately low for their hyperglycemia.[19]

The key question — the answer to which might provide insight into the pathogenesis of NIDDM — is, What causes the increased two-hour plasma glucose concentration in people with impaired glucose tolerance? The concentration at two hours is determined by the relative changes in the rates of appearance and disappearance of glucose soon after the ingestion of glucose.[20-22] These rates depend mainly on changes in insulin and glucagon secretion and the sensitivity of liver and muscle to these hormones. To address the question, we compared hepatic glucose output, splanchnic glucose sequestration, the uptake of glucose by muscle, and plasma insulin and glucagon concentrations after the administration of an oral glucose load in subjects with impaired glucose tolerance and in normal subjects of similar age, weight, and sex.

METHODS

Subjects

Informed written consent was obtained from 15 otherwise healthy subjects with impaired glucose tolerance according to World Health Organization criteria[23] (i.e., plasma glucose concentrations between 7.8 and 11.1 mmol per liter two hours after an oral glucose load of 75 g) and from 16 healthy subjects with normal glucose tolerance[23] with whom they were matched for age and weight. The clinical characteristics of the study subjects are shown in Table 1. The groups did not differ significantly in age, sex, muscle mass,[24] or degree of obesity. In subdividing the groups, we considered a body-

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Supported in part by grants from the National Institutes of Health (5M01 RR00056 and 5R37 DK20411) and the Department of Veterans Affairs. Drs. Mokan and Veneman were supported by a mentor-based fellowship from the American Diabetes Association.

mass index (defined as the weight in kilograms divided by the square of the height in meters) greater than 26 in women and 27 in men to indicate obesity. The study protocol was approved by the University of Pittsburgh Biomedical Institutional Review Board.

Protocol

The subjects were admitted to the General Clinical Research Center the evening before the study began, having eaten a weight-maintenance diet containing at least 200 g of carbohydrate for the preceding three days. After a standard dinner (10 kcal per kilogram of body weight, 50 percent carbohydrate, 35 percent fat, and 15 percent protein) between 5 and 7 p.m., the subjects received only water for the subsequent 12 to 14 hours before the study.

At about 5 o'clock the next morning, an 18-gauge catheter was inserted into a superficial forearm vein for initiation of a primed (28 μCi), continuous (0.40 μCi per minute) infusion of [6-^{3}H]glucose (New England Nuclear, Boston). The ipsilateral radial artery was cannulated with a 20-gauge arterial catheter (Arrow International, Greensboro, Pa.) for intermittent arterial sampling. In the contralateral arm, a large antecubital vein was cannulated in a retrograde direction for intermittent sampling of the forearm deep venous system. Saline without added heparin was infused slowly to maintain patency. After a four-hour isotope-equilibration period, each subject drank a 200-ml solution of glucose (1 g of Dextral per kilogram of body weight [maximum, 75 g], American Scientific Products, McGaw Park, Ill.) containing 100 μCi of [1-^{14}C]glucose (Research Products International, Gif sur Yvette, France) in five minutes. The subjects remained supine throughout the experiment. Simultaneous samples of arterial and venous blood were obtained at 30-minute intervals before and for 5 hours after the ingestion of glucose to determine plasma glucose concentrations, [1-^{14}C]glucose specific activity, and [6-^{3}H]glucose specific activity, as previously described.[25] Plasma insulin[26] and glucagon[27] concentrations were measured only in the arterial samples.

Calculations

We calculated the overall rate of appearance of glucose (endogenous plus exogenous) from the [6-^{3}H]glucose data, using the nonsteady-state equations of Hetenyi and Norwich.[28] The rate of appearance of the oral glucose in the systemic circulation was calculated from the [1-^{14}C]glucose data with the equation of Chiasson et al.[29] after correction for recycling,[30,31] as previously described.[25] The production of endogenous glucose was calculated as the difference between the overall rate of appearance of glucose and the rate of appearance of exogenous glucose.[25] Overall splanchnic uptake of glucose was calculated as the difference between the amount of oral glucose administered and the total systemic appearance of the oral glucose.[25] We assumed that absorption of the orally administered glucose was complete within the five-hour study period.[32]

The net forearm uptake of glucose was calculated at each sampling time as the product of the arteriovenous difference and the forearm blood flow, as determined by plethysmography.[25,33] We converted plasma concentrations to values for whole blood using the following equation: whole-blood value = plasma value × (1 − 0.0294 hematocrit).[34] The sum of each 30-minute measurement was used to determine the overall net balance during the 5-hour study period. We converted forearm data per 100 ml of tissue to values per kilogram of forearm muscle, assuming that 80 percent of the measured forearm blood flow perfused muscle[35] and that muscle made up 60 percent of the measured forearm volume.[36] These values were multiplied by total-body skeletal muscle mass, which we calculated from mid-arm circumference and triceps skin-fold thickness using the equation of Heymsfield et al.,[24] to obtain values for total-body skeletal muscle. The validity of these assumptions has been described in detail elsewhere.[24,35,36]

The results are presented as means ±SE. Statistical significance was determined by analysis of variance, paired t-tests, and least-squares linear regression. P values of <0.05 were considered to indicate statistical significance.

Table 1. Clinical Characteristics of the Study Subjects.*

Characteristic	Nonobese Subjects		Obese Subjects	
	Normal	Impaired Glucose Tolerance	Normal	Impaired Glucose Tolerance
Age (yr)	47±6	49±1	47±1	55±8
Sex (M/F)	5/4	7/1	3/4	2/5
Weight (kg)	74±12	73±6	84±8	84±8
Body-mass index	24.3±2.1	24.1±1.7	29.6±2.6	31.2±3.4
Muscle mass (kg)	27.8±6.0	29.3±5.1	31.5±4.7	29.8±3.4
Fasting plasma glucose (mmol/liter)†	5.2±0.6	6.2±0.6‡	5.2±0.5	6.1±0.5‡
Fasting plasma insulin (pmol/liter)†	49±9	75±23‡	71±18	126±31‡
Fasting plasma glucagon (pmol/liter)	42±9	43±14	47±11	44±11

*Plus–minus values are means ±SD.

†Values shown are means of the measurements made 60 and 30 minutes before the ingestion of glucose and at ingestion.

‡P<0.02 for the comparison with the subjects with normal glucose tolerance.

Results

Arterial Plasma Glucose, Insulin, and Glucagon Concentrations

The mean fasting arterial plasma glucose concentration was significantly higher in the subjects with impaired glucose tolerance than in the normal subjects (Table 1 and Fig. 1). After the ingestion of glucose, arterial glucose increased in the normal subjects

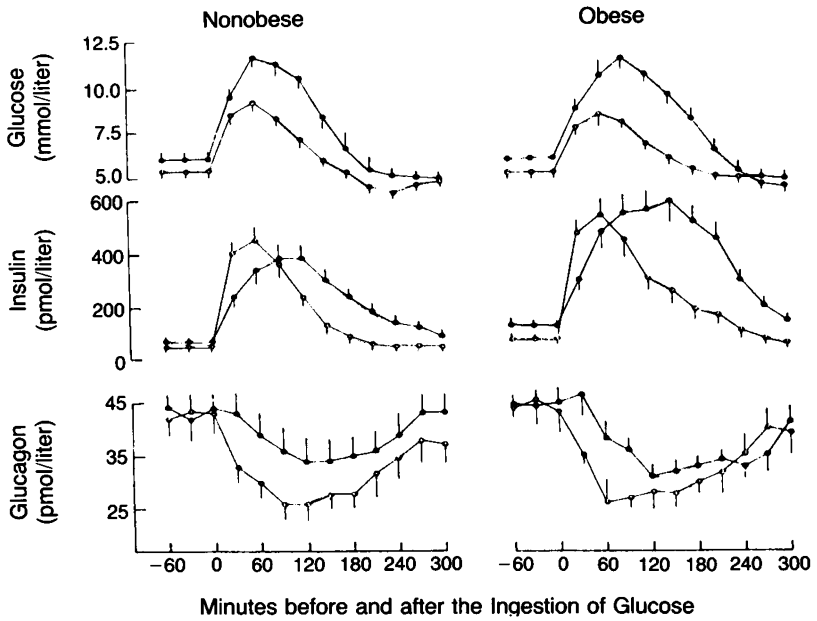

Figure 1. Mean (±SE) Arterial Plasma Glucose, Insulin, and Glucagon Concentrations before and after Glucose Ingestion in 16 Normal Subjects (○) and 15 Subjects with Impaired Glucose Tolerance (●).

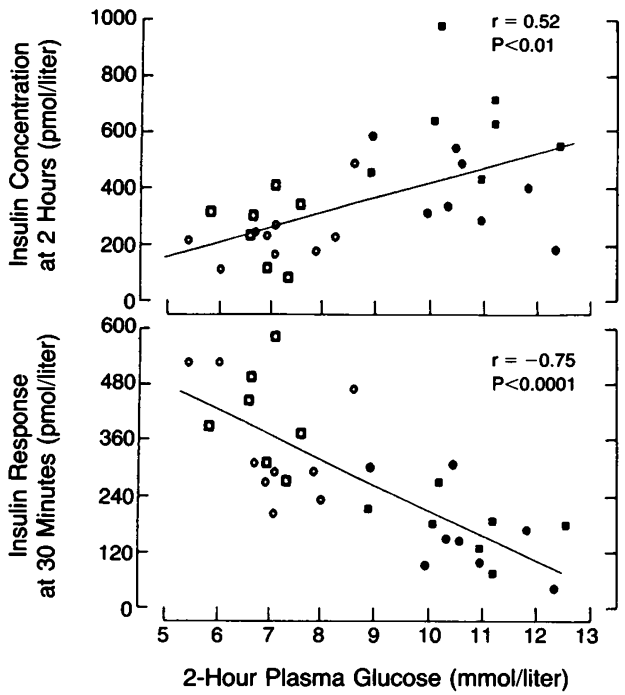

Figure 2. Correlations of 2-Hour Plasma Insulin Concentration and 30-Minute Plasma Insulin Response with 2-Hour Plasma Glucose Concentration in the Study Subjects.

Solid symbols represent subjects with impaired glucose tolerance, and open symbols normal subjects. Squares represent obese subjects, and circles nonobese subjects.

to peak concentrations of approximately 9 mmol per liter at 60 minutes and returned to basal values between 150 and 180 minutes. In the subjects with impaired glucose tolerance, arterial glucose increased to approximately 12 mmol per liter at 60 minutes and did not return to basal levels until 210 minutes. The mean arterial glucose concentrations during the five-hour study period in the nonobese and obese subjects with impaired glucose tolerance were 8.0±0.3 mmol per liter and 8.1±0.3 mmol per liter, respectively, as compared with 6.3±0.2 mmol per liter and 6.2±0.2 mmol per liter in the nonobese and obese normal subjects ($P<0.01$ for both comparisons).

The mean arterial insulin concentration while fasting was significantly higher in the subjects with impaired glucose tolerance than in the normal subjects (Table 1). Thirty minutes after the ingestion of glucose, although the plasma glucose concentrations were higher, the arterial insulin concentrations were significantly lower in both the nonobese subjects (244±35 pmol per liter) and the obese subjects (279±29 pmol per liter) with impaired glucose tolerance than in the respective normal subjects (396±42 pmol per liter and 474±36 pmol per liter, $P<0.01$ for both comparisons). Despite their reduced early insulin responses, the subjects with impaired glucose tolerance subsequently had greater increases in arterial insulin concentrations than the normal subjects. Consequently, the mean arterial plasma insulin concentration during the five-hour period was nearly 1½ times greater in both the nonobese subjects (244±28 pmol per liter) and the obese subjects (400±44 pmol per liter) with impaired glucose tolerance than in the respective normal subjects (187±16 pmol per liter and 259±31 pmol per liter, $P<0.02$ for both comparisons).

As shown in Figure 2, although 2-hour arterial insulin concentrations were positively correlated with 2-hour arterial glucose concentrations ($r = 0.52$, $P<0.01$), the arterial insulin concentrations at 30 minutes were negatively and more strongly correlated with 2-hour arterial glucose concentrations ($r = -0.75$, $P<0.0001$). Thus, the lower the initial insulin response, the greater the glucose intolerance.

The mean fasting arterial glucagon concentrations were similar in both groups of subjects (Table 1). Thirty minutes after the ingestion of glucose, the arterial glucagon level had decreased in the normal subjects ($P<0.01$). In the subjects with impaired glucose tolerance, the arterial glucagon level did not decrease until 60 minutes after ingestion, and the nadir values were significantly higher than in the normal subjects (Fig. 1). With a smaller initial decrease in arterial glucagon and a smaller initial increase in arterial insulin, at 30 minutes the subjects with impaired glucose tolerance had a molar ratio of arterial insulin to glucagon approximately half that of the normal subjects (6.0±0.7 vs. 11.7±1.0, $P<0.001$).

Rate of Appearance of Oral Glucose, Splanchnic Sequestration, and Rate of Appearance of Endogenous Glucose

In all the groups, the rate of systemic appearance of the ingested glucose was maximal 30 minutes after ingestion and then declined to values that were not significantly different from 0 between 270 and 300 minutes after ingestion (Fig. 3). There was no significant difference between the groups at any sampling time, and the overall rate of appearance of the oral glucose was thus comparable in all the groups (Table 2).

Splanchnic sequestration of the oral glucose did not differ significantly among the groups of subjects (Table 2) and amounted to approximately 27 percent of the oral glucose load.

Basal hepatic glucose output did not differ significantly among the groups (Table 2). After the ingestion of glucose, the output of endogenous glucose in the normal subjects decreased significantly within 30 minutes; during the entire 5-hour period it was suppressed by approximately 50 percent. In the subjects with impaired glucose tolerance, the output of endogenous glucose did not decrease significantly until 60 minutes after ingestion, and during the entire 5-hour period it was suppressed by less than 30 percent ($P<0.01$ for the comparison with the normal subjects). Consequently, total endogenous glucose output per five hours in both the nonobese (181±6 mmol) and the obese (201±9 mmol) subjects with impaired glucose tolerance was significantly higher than in the respec-

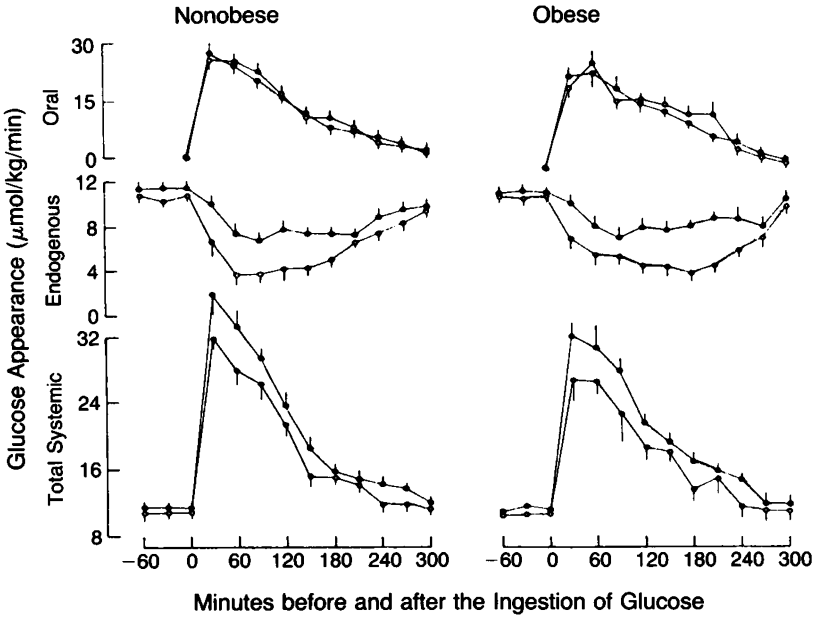

Figure 3. Mean (±SE) Rates of Appearance of Oral Glucose in the Systemic Circulation, Endogenous Glucose, and Total Systemic Glucose before and after Glucose Ingestion in 16 Normal Subjects (○) and 15 Subjects with Impaired Glucose Tolerance (●).

tive normal subjects (133±10 mmol and 136±8 mmol, $P<0.01$ for both comparisons).

Rates of Total Systemic Appearance and Disappearance of Glucose and Overall Tissue and Muscle Uptake of Glucose

As a consequence of the reduced suppression of the output of endogenous glucose, the rate of total systemic appearance of glucose was greater in the subjects with impaired glucose tolerance than in the normal subjects (Table 2). During the first 30 minutes, when the subjects with impaired glucose tolerance had a smaller increase in plasma insulin and a smaller decrease in plasma glucagon concentrations, their rate of overall glucose appearance was significantly higher than that of the normal subjects ($P<0.02$) (Fig. 3). As shown in Figure 4, the rates of overall glucose appearance were positively correlated with peak (1-hour) plasma glucose concentrations ($P<0.0001$) and negatively correlated with 30-minute molar ratios of plasma insulin to glucagon ($P<0.001$).

During the first 60 minutes after the ingestion of glucose, the rate of total systemic disappearance of glucose did not differ significantly among the groups (Fig. 5). The greater increase in the plasma glucose concentration during this period in the subjects with impaired glucose tolerance was thus due to greater rates of total glucose appearance rather than reduced rates of disappearance. Indeed, during the entire five-hour study period, the rate of total systemic disappearance of glucose was significantly higher in the subjects with impaired glucose tolerance (Table 3). Total tissue uptake of glucose, calculated by subtracting glucose that appeared in the urine from total glucose disappearance, was also significantly greater in the subjects with impaired glucose tolerance (Table 3).

Rates of forearm uptake of glucose at base line and after the ingestion of glucose were comparable in all the groups (Fig. 5). The extrapolated values for total-body uptake of glucose by muscle during the five-hour study period also did not differ significantly among the groups (Table 3).

Discussion

These studies demonstrate that reduced suppression of endogenous glucose output is primarily responsible for the excessive increases in plasma glucose concentrations that occur early after the ingestion of glucose in people with impaired glucose tolerance. A similar defect has also been found in people with NIDDM.[37-41]

The reduced suppression of endogenous glucose output could be due to hepatic insulin resistance,[42,43] abnormal insulin and glucagon secretion, or both.[20-22,44] The early (30 minute) plasma insulin concentrations were about 40 percent lower in our subjects with impaired glucose tolerance than in the normal subjects, and their plasma glucagon levels had not yet decreased. As a consequence, their molar ratio of plasma insulin to glucagon was reduced by about 50 percent. A lower ratio would be expected to result in less suppression of endogenous glucose output.[21,22,44] The significant inverse relation

Table 2. Splanchnic Glucose Metabolism during Assimilation of the Oral Glucose Load.*

Glucose Variable	Nonobese Subjects		Obese Subjects	
	Normal	Impaired Glucose Tolerance	Normal	Impaired Glucose Tolerance
Endogenous output before load (mmol/5 hr)	259±13	257±8	252±13	273±12
Endogenous output after load (mmol/5 hr)	133±10	181±6†	136±8	201±9†
Suppression of endogenous output (%)	49.2±3.1	29.4±2.2†	45.6±2.4	26.7±1.6†
Systemic appearance of oral load (mmol/5 hr)	279±8	273±8	301±11	287±11
Splanchnic sequestration of oral load (mmol/5 hr)	106±9	108±6	108±8	111±12
Percent of oral load sequestered	27.1±2.0	28.5±1.5	26.6±2.2	28.7±2.6
Total systemic appearance (mmol/5 hr)	411±11	455±12‡	436±7	486±17‡

*Plus–minus values are means ±SE. To convert millimoles of glucose to grams, multiply by 0.18.

†$P<0.01$ for the comparison with the subjects with normal glucose tolerance.

‡$P<0.02$ for the comparison with the subjects with normal glucose tolerance.

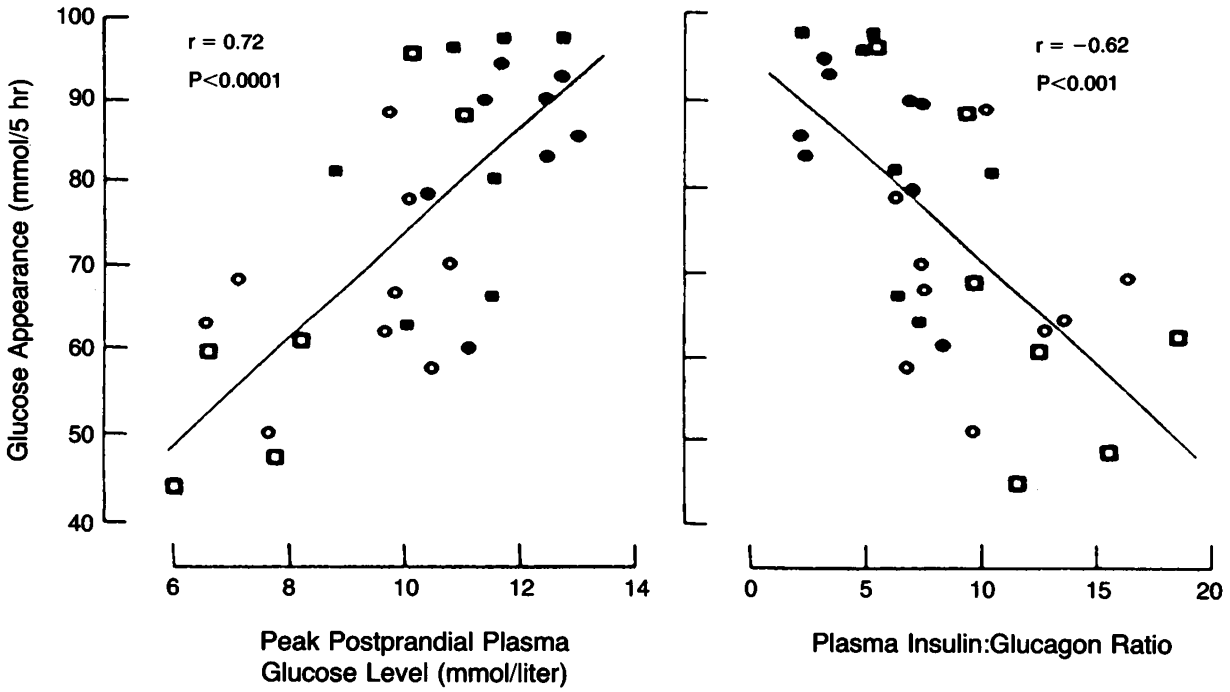

Figure 4. Correlations of Systemic Glucose Appearance with Peak Plasma Glucose Concentration and Molar Ratio of Plasma Insulin to Glucagon in the Study Subjects.

Solid symbols represent subjects with impaired glucose tolerance, and open symbols normal subjects. Squares represent obese subjects, and circles nonobese subjects.

between the plasma insulin:glucagon ratio and the rate of appearance of glucose 30 minutes after glucose ingestion provides evidence that impaired stimulation of early insulin secretion and impaired suppression of early glucagon secretion were the key pathogenetic factors.

It is important to point out that some of the plasma insulin in our subjects with impaired glucose tolerance was proinsulin. Several studies have found that in people with NIDDM or impaired glucose tolerance, an increased proportion of fasting and postprandial immunoreactive plasma insulin is accounted for by proinsulin immunoreactivity.[45-48] Thus, the reduction in early plasma insulin responses in our subjects with impaired glucose tolerance was probably underestimated.

Although insulin resistance was not formally measured, we found no evidence that it made a substantial contribution. The basal rates of glucose appearance, disappearance, and uptake by muscle were not significantly different in the subjects with impaired glucose tolerance and the normal subjects. The higher fasting and late postprandial plasma insulin concentrations in the subjects with impaired glucose tolerance could have reflected their higher plasma glucose levels, rather than insulin resistance. In normal subjects, the prolonged infusion of glucose results in the establishment of a new steady state, with increased plasma glucose and insulin concentrations.[49,50]

Tissue uptake after the ingestion of glucose was significantly greater in our subjects with impaired glucose tolerance; whether this was appropriate to their hyperglycemia and their late hyperinsulinemia is difficult to determine. It deserves emphasizing, however, that changes in plasma glucose concentrations are the result of absolute differences in the rates of appearance and disappearance of glucose, not differences between the rates of appearance and clearance of glucose. Glucose clearance is a measure of the efficiency of removal of glucose from the circulation. Initially, when plasma glucose levels increased more and plasma insulin levels increased less in the subjects with impaired glucose tolerance, their rates of overall glucose disappearance and muscle uptake were normal, but the rate of overall glucose appearance was increased. It would thus be difficult to ascribe an important role to peripheral insulin resistance. We cannot, however, definitely exclude the possibility that hepatic insulin resistance makes some contribution in addition to that expected from the reduced suppression of plasma glucagon.

Other investigators have found evidence of insulin resistance in subjects with impaired glucose tolerance.[7,9,10,51] In those studies, however, the subjects with impaired glucose tolerance were generally older or more obese than the normal subjects. Furthermore, in some of the studies[9,10,51] the subjects with impaired glucose tolerance had substantially higher postabsorptive plasma glucose and insulin concentrations than the normal subjects. Some of the insulin resistance

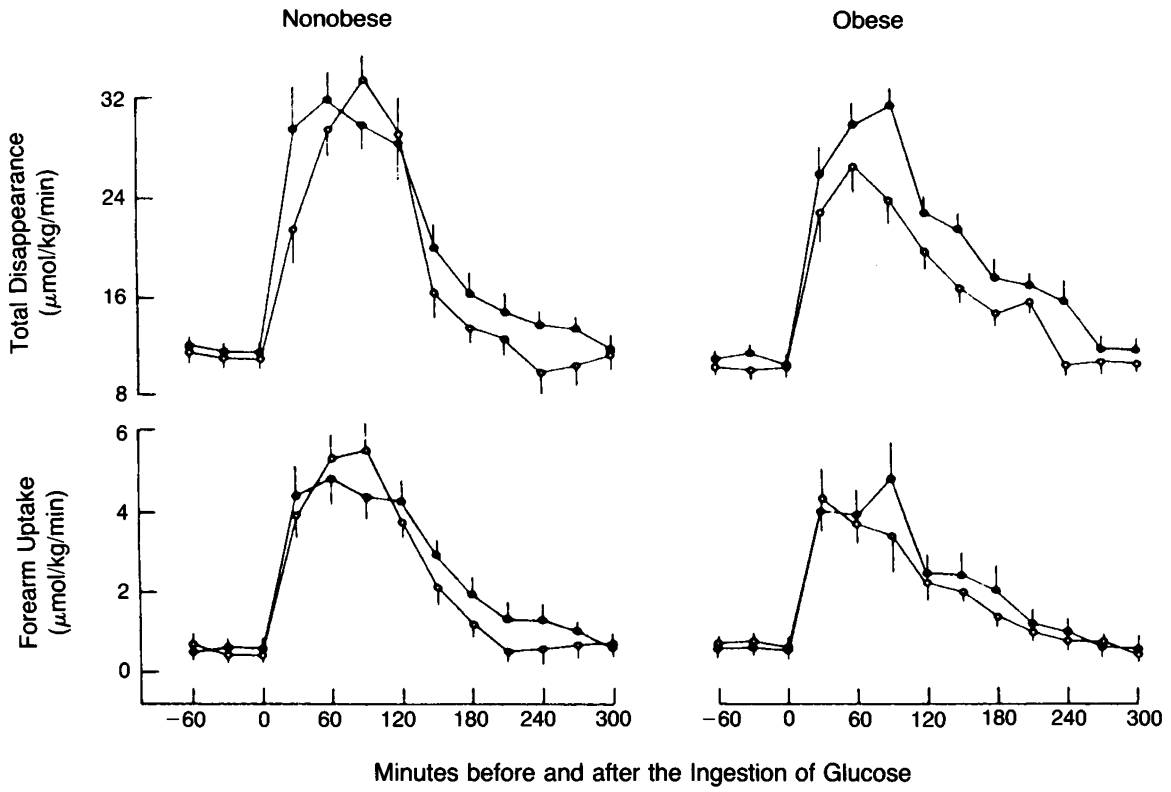

Figure 5. Mean (±SE) Rates of Systemic Glucose Disappearance and Forearm Glucose Uptake before and after Glucose Ingestion in 16 Normal Subjects (○) and 15 Subjects with Impaired Glucose Tolerance (●).

could therefore have been due to hyperglycemia[5,52] and hyperinsulinemia.[53]

Our findings have important implications for the interpretation of epidemiologic and other studies[1,10,11,18,20-22] in which conclusions regarding the pathogenesis of NIDDM have been based on the relation between two-hour plasma glucose and insulin concentrations. As in previous studies, the two-hour plasma insulin values in our study were higher in the subjects with impaired glucose tolerance (Fig. 2). This observation has led some investigators to conclude that hyperglycemia has occurred despite hyperinsulinemia and that insulin resistance was therefore the causative factor.[1,10,12,15-18] However, the abnormalities in plasma glucose and pancreatic islet-cell hormone kinetics demonstrated in this study suggest another interpretation — namely, that impaired early insulin secretion in conjunction with impaired suppression of glucagon secretion causes the excessive delivery of glucose into the systemic circulation. The larger increase in the plasma glucose concentration that results would then provide a greater stimulus for insulin secretion, so that higher plasma insulin concentrations would eventually occur. According to this interpretation, hyperglycemia, rather than insulin resistance, is primarily responsible for the delayed hyperinsulinemia characteristic of impaired glucose tolerance and mild NIDDM.

This interpretation, proposed by Perley and Kipnis 25 years ago,[19] is supported by several recent studies.[20-22] Luzi and DeFronzo[22] found that in normal subjects the inhibition of early-phase insulin release during the infusion of glucose reduced the suppression of hepatic glucose output by nearly 50 percent without reducing tissue uptake. These results are similar to those in our subjects with impaired glucose tolerance. In the study of Bruce et al.,[20] restoring nearly normal early plasma insulin responses to the ingestion of a meal in subjects with NIDDM by the intravenous administration of supplemental insulin reduced postprandial hyperglycemia and prevented delayed hyper-

Table 3. Peripheral-Tissue Glucose Metabolism during Assimilation of the Oral Glucose Load.*

Glucose Variable	Nonobese Subjects		Obese Subjects	
	Normal	Impaired Glucose Tolerance	Normal	Impaired Glucose Tolerance
Total disappearance (mmol/5 hr)	407±10	457±12†	432±10	498±20†
Urinary loss (mmol/5 hr)	0	1.1±0.5	0	0.5±0.5
Total tissue uptake (mmol/5 hr)	406±10	456±2†	432±9	482±21†
Muscle uptake (mmol/5 hr)	207±7	236±20	208±8	198±6

*Plus–minus values are means ±SE. To convert millimoles of glucose to grams, multiply by 0.18.

†$P<0.05$ for the comparison with the subjects with normal glucose tolerance.

insulinemia. These observations demonstrate the importance of reduced early increases in insulin secretion and the secondary nature of late hyperinsulinemia in NIDDM.

In conclusion, in persons with impaired glucose tolerance the excessive increase in plasma glucose concentrations after the ingestion of glucose results primarily from excessive entry of glucose into the circulation. This is due to the failure of the liver to reduce its glucose output appropriately and can largely be accounted for by diminished early insulin release and diminished suppression of glucagon secretion. We therefore suggest that insulin resistance in people with more severe glucose intolerance[7,9,10,51] may develop as a consequence of more prolonged hyperglycemia (glucose toxicity)[5-7,52] and compensatory hyperinsulinemia.[53] In susceptible people, a combination of these factors may ultimately cause further deterioration of beta-cell function and progression to NIDDM.

We are indebted to the staff of the General Clinical Research Center, Ms. Carol Korbanic, Ms. Dawn Purdy, and Ms. Lauri Henry, for their excellent technical help; to Ms. Cathy Butler and Ms. Laura Brinker for superb editorial assistance; and to Mr. E.W. Shirmer, Eli Lilly and Company, for the provision of ^{125}I-insulin.

References

1. DeFronzo RA, Ferrannini E, Koivisto V. New concepts in the pathogenesis and treatment of insulin-dependent diabetes mellitus. Am J Med 1983;74: Suppl 1A:52-81.
2. Gerich JE. Role of insulin resistance in the pathogenesis of type 2 (noninsulin-dependent) diabetes mellitus. Clin Endocrinol Metab 1988;2:307-26.
3. Greenstein BD. Improved insulin receptor assay: effects of an antidiabetic sulfonylurea on liver membrane insulin receptors from obese hyperglycaemic mice. Br J Pharmacol 1979;66:317-22.
4. Leahy JL, Cooper HE, Deal DA, Weir GC. Chronic hyperglycemia is associated with impaired glucose influence on insulin secretion: a study of normal rats using chronic in vivo glucose infusions. J Clin Invest 1986;77: 908-15.
5. Rossetti L, Giaccari A, DeFronzo RA. Glucose toxicity. Diabetes Care 1990;13:610-30.
6. Unger RH, Grundy S. Hyperglycaemia as an inducer as well as a consequence of impaired islet cell function and insulin resistance: implications for the management of diabetes. Diabetologia 1985;28:119-21.
7. Eriksson J, Franssila-Kallunki A, Ekstrand A, et al. Early metabolic defects in persons at increased risk for non-insulin-dependent diabetes mellitus. N Engl J Med 1989;321:337-43.
8. Ohlson LO, Larsson B, Bjorntorp P, et al. Risk factors for type 2 (non-insulin-dependent) diabetes mellitus: thirteen and one-half years of follow-up of the participants in a study of Swedish men born in 1913. Diabetologia 1988;31:798-805.
9. Kolterman OG, Gray RS, Griffin J, et al. Receptor and postreceptor defects contribute to the insulin resistance in noninsulin-dependent diabetes mellitus. J Clin Invest 1981;68:957-69.
10. Lillioja S, Mott DM, Howard BV, et al. Impaired glucose tolerance as a disorder of insulin action: longitudinal and cross-sectional studies in Pima Indians. N Engl J Med 1988;318:1217-25.
11. O'Rahilly SP, Nugent Z, Rudenski A, et al. Beta-cell dysfunction, rather than insulin insensitivity, is the primary defect in familial type 2 diabetes. Lancet 1986;2:360-4.
12. Reaven G, Miller R. Study of the relationship between glucose and insulin responses to an oral glucose load in man. Diabetes 1968;17:560-9.
13. Kipnis D. Insulin secretion in diabetes mellitus. Ann Intern Med 1968;67: 891-901.
14. Chiles R, Tzagournes M. Excessive insulin response to oral glucose in obesity and mild diabetes: study of 501 patients. Diabetes 1970;19:458-64.
15. Savage PJ, Dippe SE, Bennett PH, et al. Hyperinsulinemia and hypoinsulinemia: insulin responses to oral carbohydrate over a wide spectrum of glucose tolerance. Diabetes 1975;24:362-8.
16. Zimmet P, Whitchouse S, Kiss J. Ethnic variability in the plasma insulin response to oral glucose in Polynesian and Micronesian subjects. Diabetes 1979;28:624-8.
17. DeFronzo RA. The triumvirate: β-cell, muscle, and liver: a collusion responsible for NIDDM. Diabetes 1988;37:667-87.
18. Saad MF, Knowler WC, Pettitt DJ, Nelson RG, Mott DM, Bennett PH. Sequential changes in serum insulin concentration during development of non-insulin-dependent diabetes. Lancet 1989;1:1356-9.
19. Perley MJ, Kipnis DM. Plasma insulin responses to oral and intravenous glucose: studies in normal and diabetic subjects. J Clin Invest 1967;46:1954-62.
20. Bruce DG, Chisholm DJ, Storlien LH, Kraegen EW. Physiologic importance of deficiency in early prandial insulin secretion in non-insulin-dependent diabetes. Diabetes 1988;37:736-44.
21. Steiner KE, Mouton SM, Bowles CR, Williams PE, Cherrington AD. The relative importance of first- and second-phase insulin secretion in countering the action of glucagon on glucose turnover in the conscious dog. Diabetes 1982;31:964-72.
22. Luzi L, DeFronzo RA. Effect of loss of first-phase insulin secretion on hepatic glucose production and tissue glucose disposal in humans. Am J Physiol 1989;257:E241-E246.
23. WHO Expert Committee on Diabetes Mellitus: second report. WHO Tech Rep Ser 1980;646:1-80.
24. Heymsfield SB, McManus C, Stevens V, Smith J. Muscle mass: reliable indicator of protein-energy malnutrition seventy and outcome. Am J Clin Nutr 1982;35:1192-9.
25. Kelley D, Mitrakou A, Marsh H, et al. Skeletal muscle glycolysis, oxidation, and storage of an oral glucose load. J Clin Invest 1988;81:1563-71.
26. Herbert V, Lau KS, Gottlieb CW, Bleicher SJ. Coated charcoal immunoassay of insulin. J Clin Endocrinol Metab 1965;25:1375-84.
27. Faloona GR, Unger RH. Glucagon. In: Jaffe BM, Behrman HR, eds. Methods of hormone radioimmunoassay. New York: Academic Press, 1974:317-30.
28. Hetenyi G Jr, Norwich KH. Validity of rates of production and utilization of metabolites as determined by tracer methods in intact animals. Fed Proc 1974;33:1841-8.
29. Chiasson JL, Liljenquist JE, Lacy WW, Jennings AS, Cherrington AD. Gluconeogenesis: methodological approaches in vivo. Fed Proc 1977;36: 229-35.
30. Bloom B. The simultaneous determination of C^{14} and H^3 in the terminal groups of glucose. Anal Biochem 1962;3:85-7.
31. Reichard GA Jr, Moury FN Jr, Hochella NJ, Patterson AL, Weinhouse S. Quantitative estimation of the Cori cycle in the human. J Biol Chem 1963;238:495-501.
32. Radziuk J, McDonald TJ, Rubenstein D, Dupre J. Initial splanchnic extraction of ingested glucose in normal man. Metabolism 1978;27:657-69.
33. Greenfield ADM, Whitney RJ, Mowbray JF. Methods for the investigation of peripheral blood flow. Br Med Bull 1963;19:101-9.
34. Dillion RS. Importance of the hematocrit in interpretation of blood sugar. Diabetes 1965;14:672-4.
35. Cooper KE, Edholm OG, Moltram FR. The blood flow in skin and muscle of the human forearm. J Physiol 1955;128:258-67.
36. Andres R, Cader G, Zierler K. The quantitatively minor role of carbohydrate in oxidative metabolism by skeletal muscle in intact man in the basal state: measurements of oxygen and glucose uptake and carbon dioxide and lactate production in the forearm. J Clin Invest 1956;35:671-82.
37. Mitrakou M, Kelley D, Veneman T, et al. Contribution of abnormal muscle and liver glucose metabolism in postprandial hyperglycemia in NIDDM. Diabetes 1990;39:1381-90.
38. Firth RG, Bell PM, Marsh HM, Hansen I, Rizza RA. Postprandial hyperglycemia in patients with noninsulin-dependent diabetes mellitus: role of hepatic and extrahepatic tissues. J Clin Invest 1986;77:1525-32.
39. Ferrannini E, Simonson D, Katz L, et al. The disposal of an oral glucose load in patients with non-insulin-dependent diabetes. Metabolism 1988;37: 79-85.
40. Felig P, Wahren J, Hendler R. Influence of maturity-onset diabetes on splanchnic glucose balance after oral glucose ingestion. Diabetes 1978;27: 121-6.
41. Osei K. The role of splanchnic glucose output in determining glycemic responses after mixed meal in type II diabetic patients and normal subjects. Pancreas 1987;2:386-92.
42. Ferrannini E, Groop LC. Hepatic glucose production in insulin-resistant states. Diabetes Metab Rev 1989;5:711-26.
43. Campbell PJ, Mandarino LJ, Gerich JE. Quantification of the relative impairment in actions of insulin on hepatic glucose production and peripheral glucose uptake in non-insulin-dependent diabetes mellitus. Metabolism 1988;37:15-21.
44. Unger RH, Orci L. Physiology and pathophysiology of glucagon. Physiol Rev 1976;56:779-826.
45. Ward WK, LaCava EC, Paquette TL, Beard JC, Wallum BJ, Porte D Jr. Disproportionate elevation of immunoreactive proinsulin in type 2 (non-insulin-dependent) diabetes mellitus and in experimental insulin resistance. Diabetologia 1987;30:698-702.

46. Yoshioka N, Kuzuya T, Matsuda A, Tanigushi M, Iwamoto Y. Serum proinsulin levels at fasting and after oral glucose load in patients with type 2 (non-insulin-dependent) diabetes mellitus. Diabetologia 1988;31:355-60.
47. Saad MF, Kahn SE, Nelson RG, et al. Disproportionately elevated proinsulin in Pima Indians with noninsulin-dependent diabetes mellitus. J Clin Endocrinol Metab 1990;70:1247-53.
48. Temple RC, Clark PMS, Nagi DK, Schneider AE, Yudkin JS, Hales CN. Radioimmunoassay may overestimate insulin in non-insulin-dependent diabetics. Clin Endocrinol 1990;32:689-93.
49. Porte D Jr, Pupo AA. Insulin responses to glucose: evidence for a two pool system in man. J Clin Invest 1969;48:2309-19.
50. Pfeifer MA, Graf RJ, Halter JB, Porte D Jr. The regulation of glucose-induced insulin secretion by pre-stimulus glucose level and tolbutamide in normal man. Diabetologia 1981;21:198-205.
51. Sacca L, Orofino G, Petrone A, Vigorito C. Differential roles of splanchnic and peripheral tissues in the pathogenesis of impaired glucose tolerance. J Clin Invest 1984;73:1683-7.
52. Yki-Jarvinen H, Helve E, Koivisto VA. Hyperglycemia decreases glucose uptake in type I diabetes. Diabetes 1987;38:892-6.
53. Mandarino L, Baker B, Rizza R, Genest J, Gerich J. Infusion of insulin impairs human adipocyte glucose metabolism in vitro without decreasing adipocyte insulin receptor binding. Diabetologia 1984;27:358-63.