Effect of Insulin and Glucose Infusions on Sympathetic Nervous System Activity in Normal Man

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SUMMARY

Recent studies indicate a link between carbohydrate intake and the functional state of the sympathetic nervous system. Fasting or carbohydrate restriction decreases sympathetic activity, while glucose ingestion or dietary supplementation with sucrose increases sympathetic nerve activity. To examine the potential contributions of hyperglycemia and hyperinsulinemia to sympathetic stimulation, sympathetic activity was assessed by measurement of plasma norepinephrine (NE) levels and concomitant cardiovascular indices in nonobese young men during glucose and insulin infusions using glucose clamp techniques. In the insulin infusion studies (euglycemic glucose clamp), insulin was administered at 2 mU/kg/min and 5 mU/kg/min for 2 h while blood glucose was maintained at basal levels by a variable rate of glucose infusion. In the hyperglycemic studies, blood glucose was raised 125 mg/dl above basal and maintained at that level for 2 h.

In response to both insulin infusions, plasma NE rose progressively over the course of the study, increasing 50% with the 2-mU infusion (from mean basal value of 240 ± 34 pg/ml to 360 ± 41 at 150 min, $P < 0.001$ for changes over time by analysis of variance) and 117% with the 5-mU infusion (from 254 ± 20 pg/ml to 551 ± 88 at 150 min, $P < 0.001$). The plasma NE response was greater with the 5-mU than with the 2-mU insulin infusion ($P < 0.001$), and similarly, was greater during the 2-mU insulin infusion than during a control test in which neither insulin nor glucose was infused ($P < 0.001$). Associated with the elevations in plasma NE in the 2-mU insulin infusion were increases in pulse rate ($P < 0.05$), pulse pressure ($P < 0.005$), and pulse rate – systolic blood pressure product ($P < 0.01$), and during the 5-mU insulin infusions there were increases in pulse pressure ($P < 0.001$), mean arterial blood pressure ($P < 0.001$), and pulse rate – systolic blood pressure product ($P < 0.001$). Plasma NE did not change during the hyperglycemic glucose clamp nor during control tests, and pulse pressure in the hyperglycemic studies ($P < 0.005$) was the only cardiovascular measurement increased by these two infusion protocols. The clearance of NE in three subjects was unaffected by the 2-mU insulin infusion. Thus, insulin infusion increases sympathetic nervous system activity in the absence of changes in blood glucose.

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Studies in experimental animals and man indicate that the activity of the sympathetic nervous system is influenced by the level of caloric intake.[1] Fasting decreases and sucrose overfeeding increases norepinephrine (NE) turnover, a direct, in vivo measure of neuronal activity in sympathetically innervated organs of rats and mice.[2–4] In humans, a reduction in carbohydrate intake lowers plasma NE concentrations in obese subjects,[5,6] while the oral administration of glucose raises plasma NE levels to a greater extent than that seen following ingestion of a noncaloric control drink.[7] These experiments in human subjects suggest that the pattern of sympathetic nervous system responses to dietary manipulation is similar in laboratory animals and man. The nature of the link between dietary intake and sympathetic nervous system function is unknown, but the apparent importance of dietary carbohydrate raises the possibility that either glucose or insulin may play a central role in this regulatory process. The present study was designed to examine the potential contributions of hyperglycemia and hyperinsulinemia to the sympathetic activation observed after carbohydrate administration.

The glucose clamp technique was developed to produce a stable blood glucose concentration at predetermined levels by means of servo-correction of the rate of a glucose infusion based on repeated glucose measurements and em-

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pirically derived formulas.[8] In the hyperglycemic variant of this technique, blood glucose is raised to the desired level and maintained constant despite increased insulin secretion by periodic increments in the rate of glucose infusion. Another form of the glucose clamp (the euglycemic glucose clamp) combines steady-state hyperinsulinemia, produced by a constant insulin infusion, with maintenance of basal glucose levels by means of a variable rate of glucose infusion. Thus, the influence of increased levels of circulating insulin can be assessed independently from that of alterations in blood glucose concentration and, moreover, the dose-response relationship between insulin level and a particular physiologic variable can be studied.

The results of these experiments demonstrate that insulin increases plasma NE concentrations in a dose-dependent fashion during maintenance of euglycemia over a 2-h period of observation. In contrast, plasma NE levels are not affected by 2 h of sustained hyperglycemia (125 mg/dl above basal) nor by the testing procedure itself in the absence of insulin or glucose infusions. The insulin-induced elevations in plasma NE imply activation of the sympathetic nervous system, as the clearance of circulating NE is not altered by insulin infusion and the pattern of cardiovascular stimulation produced by insulin is consistent with primary sympathetic stimulation.

METHODS

Subjects. Twelve male volunteers (18–36 yr of age) participated in these studies. All were within 20% of desirable body weight (obesity index 1.05 ± 0.04; mean ± SE). On screening, none showed evidence of chronic or intercurrent illness and none were taking any medication. All subjects had normal oral glucose tolerance and no family history of diabetes. Before each study, subjects supplemented their individual diets with additional carbohydrate for 3 days to insure a daily intake of 150–200 g. Subjects were hospitalized on the evening before the test and were studied in the following morning. An interval of at least 1 wk separated any two tests on the same subject. The protocols were approved by the Committee on Clinical Investigations, New Procedures and New Forms of Therapy at the Beth Israel Hospital, and informed consent was obtained from all subjects before study.

Glucose clamp studies. Glucose clamp techniques were employed as recently described.[8] Glucose in water (20%) was infused by calibrated infusion pump (Harvard Apparatus, Millis, Massachusetts) into an antecubital vein. A second catheter for collection of arteriolized venous blood[9] was inserted into a forearm vein and passed retrograde to the dorsum of the hand. The hand was placed in a heated chamber (68°C) for the duration of the study. In order to assess the influence of the warming chamber on plasma NE, simultaneous samples for plasma NE were drawn from the warmed hand and from a contralateral antecubital vein in three studies. The mean difference between plasma NE levels drawn from the two sites was 6 ± 8 pg/ml (mean ± SE). Insulin was infused into an antecubital vein in the arm opposite to the glucose infusion.

Subjects were supine and awake throughout all studies. An equilibration period of 45 min followed insertion of the intravenous catheters before blood collections began for glucose, insulin, and NE. Two basal samples were obtained before the start of the clamp study and at 5-min intervals for glucose and at 30-min intervals for insulin and NE thereafter. Pulse rate and blood pressure were measured by arm cuff technique at regular intervals in all studies. Pulse pressure was calculated as the difference between systolic and diastolic BP, and mean arterial BP as diastolic BP plus one-third of the pulse pressure. The double product, a noninvasive correlate of myocardial oxygen consumption,[10] was calculated by multiplying the pulse rate and the systolic BP. Seven hyperglycemic glucose clamp (G + 125) studies were performed. Blood glucose concentration was raised 125 mg/dl at the start of the experiment and maintained at that level for 120 min with a coefficient of variation for steady-state glucose concentrations (20–120 min) of 3.8 ± 1.4% (mean ± SE). In the euglycemic glucose clamp studies, crystalline pork insulin was infused in seven tests at an infusion rate of 2 mU/kg/min (2 mU), and in seven tests at 5 mU/kg/min (5 mU). To reduce insulin adherence to syringe and tubing, 2 ml of the subject's blood was added to each 50 ml of insulin infusate. Four minutes after the start of the insulin infusion, glucose was administered at an initial rate of 2 mg/kg/min with subsequent adjustments in the rate to maintain basal glucose levels. Coefficients of variation for blood glucose during the steady-state period of the insulin infusion studies were 6.3 ± 0.9% and 5.3 ± 0.4% for the 2- and 5-mU infusions, respectively. In the insulin infusion experiments, insulin was infused for 120 min. Glucose was maintained at euglycemic levels for an additional 30 min after the termination of insulin infusions.

Five control studies were performed in which 0.45% NaCl replaced the 20% glucose infusion and 0.9% NaCl the insulin infusion. Three of the studies followed the pattern of the hyperglycemic glucose clamps, and two followed the pattern of the euglycemic glucose clamps. The testing procedure, including phlebotomy, was otherwise identical to that in the glucose clamp experiments. In all studies, approximately 1000 ml, including 540 ± 60 ml of 0.9% NaCl and 400 ml of 20% glucose (or 0.45% NaCl in control tests), was infused.

Norepinephrine infusion studies. The clearance of NE from the circulation was evaluated in three subjects in the presence and absence of an insulin infusion. The two studies in each subject were separated by an interval of at least 1 wk. The subjects were prepared as described previously for a control or a 2-mU euglycemic glucose clamp. NE (Winthrop), in 0.9% NaCl to which ascorbic acid (0.5 mg/ml) had been added, was administered intravenously at the rate of 2 μg/min for 15 min (60–75 min after the start of the control or 2-mU clamp). Samples for NE were drawn from the arm opposite the NE infusion at 10 and 5 min before the start of the NE infusion, at 10, 12.5, and 15 min after the start of the NE infusion, and at 1, 2, 4, and 10 min following cessation of the NE infusion. Metabolic clearance rates and rates of NE disappearance were calculated for each individual in both tests, as previously described.[7]

Analytical methods. The glucose content of whole blood was determined throughout these studies with the glucose-oxidase technique using a glucose analyzer (Yellow Spring Instruments, Yellow Springs, Ohio). Insulin was measured in serum by the method of Soeldner and Slone.[11]

Samples for plasma NE were collected in iced, heparinized tubes and the plasma separated within 10 min. Sam-

ples were stored at −70°C as perchloric acid extracts of plasma until assayed (usually within 2–3 wk). Plasma NE samples in different experiments from the same subject were determined in the same assay. NE was measured by radioenzymatic assay[12] which, in our laboratory, has an intra-assay coefficient of variation of 8–11% and an interassay coefficient of variation of 15–18%. The limit of sensitivity of the assay, defined as the amount of NE required to give a reading twice blank, is 25–40 pg. In all studies, 20–120 min was considered the steady-state period for calculation of glucose and insulin results. Glucose metabolized (M) is calculated as the amount of glucose infused during the steady-state period minus glucose space corrections and urinary losses. M is a measure of tissue uptake of glucose minus hepatic glucose production, which has been shown to be suppressed under conditions of the glucose clamp.[8]

Data are presented as mean ± SE. Statistical analyses employed analysis of variance (ANOVA) for comparisons within a given test and between the 2- and 5-mU euglycemic glucose clamp experiments. Comparisons between changes during control and glucose and insulin infusions used a modification of two-way ANOVA for unequal but proportionate sample sizes.[13] Coefficient of correlation was determined for changes in NE and changes in blood glucose.[14]

RESULTS

Glucose and insulin. Insulin levels and glucose metabolism rates (M) during control, hyperglycemic and euglycemic glucose clamps are shown in Figure 1 and Table 1. In control studies neither blood glucose nor serum insulin changed over the 2 h of observation. In G + 125 studies, blood glucose levels were 81.4 ± 1.0 mg/dl before the test began and were raised to 208 ± 1.6 mg/dl during the steady-state period. A biphasic pattern of insulin response was observed with an early rise followed by a sustained increase over the period of glucose infusion. Serum insulin increased from basal values of 5.8 ± 0.7 μU/ml to mean steady-state levels of 44 ± 7.6 μU/ml. M increased progressively over the course of the infusion, averaging 8.15 ± 1.00 mg/kg/min, which represented approximately 71 g of glucose administered over the course of the study.

In the 2-mU infusion studies, serum insulin increased from a basal value of 7.7 ± 1.3 μU/ml to a mean level of 154 ± 32 μU/ml during steady state. Blood glucose concentrations were 81.6 ± 0.7 mg/dl before the test started and averaged 78.6 ± 0.5 mg/dl during the 20–120-min interval. M rose during the study and remained elevated in the 30-min period following termination of the insulin infusion. During steady state, M was 8.82 ± 0.75 mg/kg/min, which was not significantly different from that obtained during G + 125 studies.

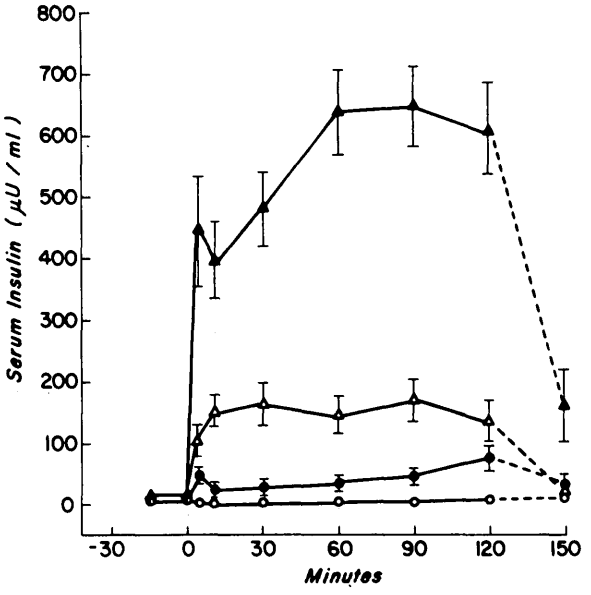


FIGURE 1. Serum concentrations of insulin during control (○), G + 125 hyperglycemic (●), and 2-mU (△) and 5-mU (▲) euglycemic studies. Points are mean ± SE for five to seven subjects per test.

In the 5-mU infusion, serum insulin increased from 9.2 ± 0.9 μU/ml to a mean level of 601 ± 64 μU/ml during steady state. Blood glucose concentrations were unchanged from a basal value of 82.2 ± 1.0 mg/dl to a mean steady-state level of 80.2 ± 1.0 mg/dl. M, during the 5-mU infusion, appeared to approach a maximum value within the first hour of insulin infusion, and over the course of the steady-state period was greater during the 2-mU than during the 5-mU infusion ($F_{1,60} = 6.43$, $P < 0.025$).

Plasma NE responses. The effects of control, hyperglycemic, and euglycemic glucose clamps on plasma NE concentration are shown in Table 2 and changes from basal are shown in Figure 2. In the control and G + 125 studies, plasma NE levels did not vary during the 2 h of observation. In contrast, in both 2- and 5-mU infusions, the rise in plasma NE levels throughout the course of the study was highly significant ($P < 0.001$ for both the 2- and the 5-mU infusions). With the 2-mU infusion, plasma NE increased 50% above the mean basal value of 240 ± 34 pg/ml to a peak of 360 ± 41 pg/ml at 150 min, 30 min after the end of the insulin in-

TABLE 1
Effect of hyperglycemic and euglycemic glucose clamps on glucose metabolism*

	Time intervals (min)						
	20–40	40–60	60–80	80–100	100–120	120–150	20–120
G + 125 (7)†	6.06 ± 0.44	6.30 ± 0.61	8.14 ± 1.42	9.45 ± 1.26	10.77 ± 1.41	—	8.15 ± 1.00
2 mU (7)‡	6.66 ± 0.51	8.69 ± 0.87	9.11 ± 0.85	9.60 ± 0.73	10.06 ± 0.90	10.02 ± 0.86	8.82 ± 0.74
5 mU (7)§	8.14 ± 0.36	10.23 ± 0.84	10.35 ± 0.61	10.67 ± 0.76	10.67 ± 0.79	11.10 ± 0.77	10.01 ± 0.66

* All data represent mean ± SE, mg/kg/min.
† The numbers in parentheses represent the number of subjects in each study.
‡ F = 1.25, not statistically different from G + 125 (ANOVA).
§ F = 5.76, P < 0.025, compared with G + 125 and 2 mU (ANOVA).

TABLE 2
Effect of control, hyperglycemic, and euglycemic glucose clamps on plasma norepinephrine*

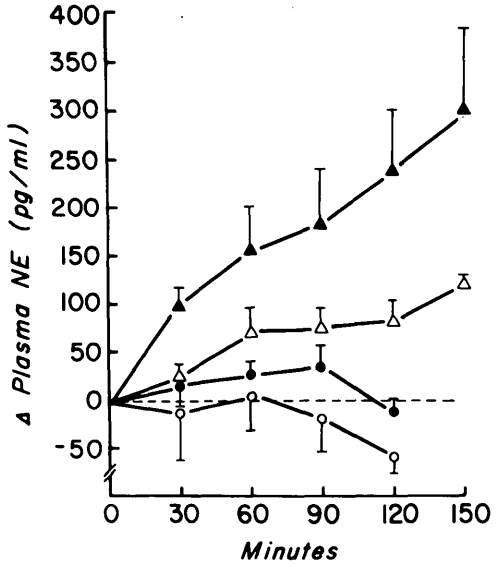
	Time (min)								
	−15	0	+30	+60	+90	+120	+150	F ratio†	P
Control (5)‡	241 ± 42	296 ± 29	257 ± 46	273 ± 35	254 ± 18	230 ± 21	—	0.84	NS
G + 125 (7)	204 ± 16	230 ± 23	239 ± 24	245 ± 14	252 ± 22	206 ± 19	—	1.97	NS
2 mU (7)	232 ± 41	246 ± 29	260 ± 35	310 ± 56	314 ± 44	321 ± 49	360 ± 41	9.44	<0.001
5 mU (7)§	248 ± 27	260 ± 22	353 ± 33	410 ± 57	438 ± 62	491 ± 67	551 ± 88	10.02	<0.001

* All values represent mean ± SE, pg/ml.
† Comparisons made over time within a given test (ANOVA).
‡ The numbers in parentheses represent the number of subjects in each group.
§ F = 15.65, P < 0.001 for comparison between 2 and 5 mU (ANOVA).

fusion. In response to the 5-mU infusion, plasma NE rose 117% above a mean basal value of 254 ± 25 pg/ml to a peak level of 551 ± 88 pg/ml at 150 min. Plasma NE levels and changes in plasma NE from basal were greater in the 2-mU glucose clamp than in control tests ($F_{1,60}$ = 53.32, P < 0.001 and $F_{1,40}$ = 13.05, P < 0.001, respectively). Plasma NE levels and the changes in plasma NE from basal values during the insulin infusion were both significantly greater in the 5-mU than in the 2-mU glucose clamp ($F_{1,84}$ = 15.65, P < 0.001 and $F_{1,60}$ = 21.67, P < 0.001 for differences between 2 and 5 mU in plasma NE levels and in change in plasma NE, respectively).

To exclude the possibility that elevations in plasma NE during the insulin infusions might be secondary to transient decrements in blood glucose, changes in plasma NE from basal were tested for correlation with the average change in blood glucose and with the maximum decrease in blood glucose below basal observed in the 30 min preceding the 60-, 90-, 120-, and 150-min plasma NE values. In the 2-mU insulin infusion, neither correlation was statistically significant (r = 0.01 for mean blood glucose versus ΔNE and r = 0.003 for the largest decrement in blood glucose versus ΔNE). Similarly, in the 5-mU infusion, no correlation was observed between the average change (r = 0.05) or the maximum fall below basal in blood glucose (r = −0.14) and the changes measured in plasma NE concentration. Furthermore, the mean difference from the basal glucose value for the period of 60–150 min, the time when the plasma NE levels were greatest, was −0.8 ± 0.3 mg/dl for the 2-mU infusion and −0.3 ± 0.1 for the 5-mU study.

FIGURE 2. Change from basal in plasma NE during control, G + 125 hyperglycemic, and 2- and 5-mU euglycemic clamps. Points are mean ± SE for five to seven subjects per test. Symbols are as indicated for Figure 1.



Cardiovascular responses. The effects of control, hyperglycemic, and euglycemic glucose clamps on cardiovascular measurements are depicted in Figure 3 as changes from basal over the course of the studies. Control infusions produced no significant changes in cardiovascular measurements during the 120 min of observation. In the G + 125 experiments, only pulse pressure increased during the infusion ($F_{9,54}$ = 3.09, P < 0.005). With the 2-mU infusion, mean arterial BP did not change, but pulse rate ($F_{11,66}$ = 2.13, P < 0.05), pulse pressure ($F_{11,66}$ = 2.88, P < 0.005), and double product ($F_{11,66}$ = 2.66, P < 0.01) all increased over the course of the study, changes that were all significantly greater than seen during control ($F_{1,70}$ = 8.27, P < 0.01; $F_{1,70}$ = 35.99, P < 0.001; and $F_{1,70}$ = 14.28, P < 0.001, respectively). Cardiovascular activity was more markedly stimulated by the 5-mU infusion. Although pulse rate did not increase, pulse pressure, mean arterial blood pressure, and double product all rose ($F_{11,66}$ = 9.83, P < 0.001; $F_{10,60}$ = 3.81, P < 0.001; $F_{11,66}$ = 7.23, P < 0.001, respectively) during the 5-mU test. Mean arterial BP, the increase above basal in mean arterial BP, and double product were greater in the 5-mU than in the 2-mU studies ($F_{1,132}$ = 18.77, P < 0.001; $F_{1,108}$ = 8.20, P < 0.005; $F_{1,144}$ = 12.50, P < 0.001, respectively).

NE clearance. The effect of insulin infusion on NE clearance is shown in Table 3. Neither the metabolic clearance rate nor the disappearance rate of circulating NE was affected by the 2-mU insulin infusion.

DISCUSSION

The effects of control, hyperglycemic, and euglycemic glucose clamps on plasma NE levels and cardiovascular measurements are summarized in Table 4. In general, increments in the level of circulating insulin are associated with progressive elevations in plasma NE concentrations and with increasing evidence of cardiovascular stimulation. The rise in plasma NE during the two insulin infusions is statistically significant as compared with the control infusion; the low and the high dose insulin infusions are also significantly different, thus indicating a dose-response relationship be-

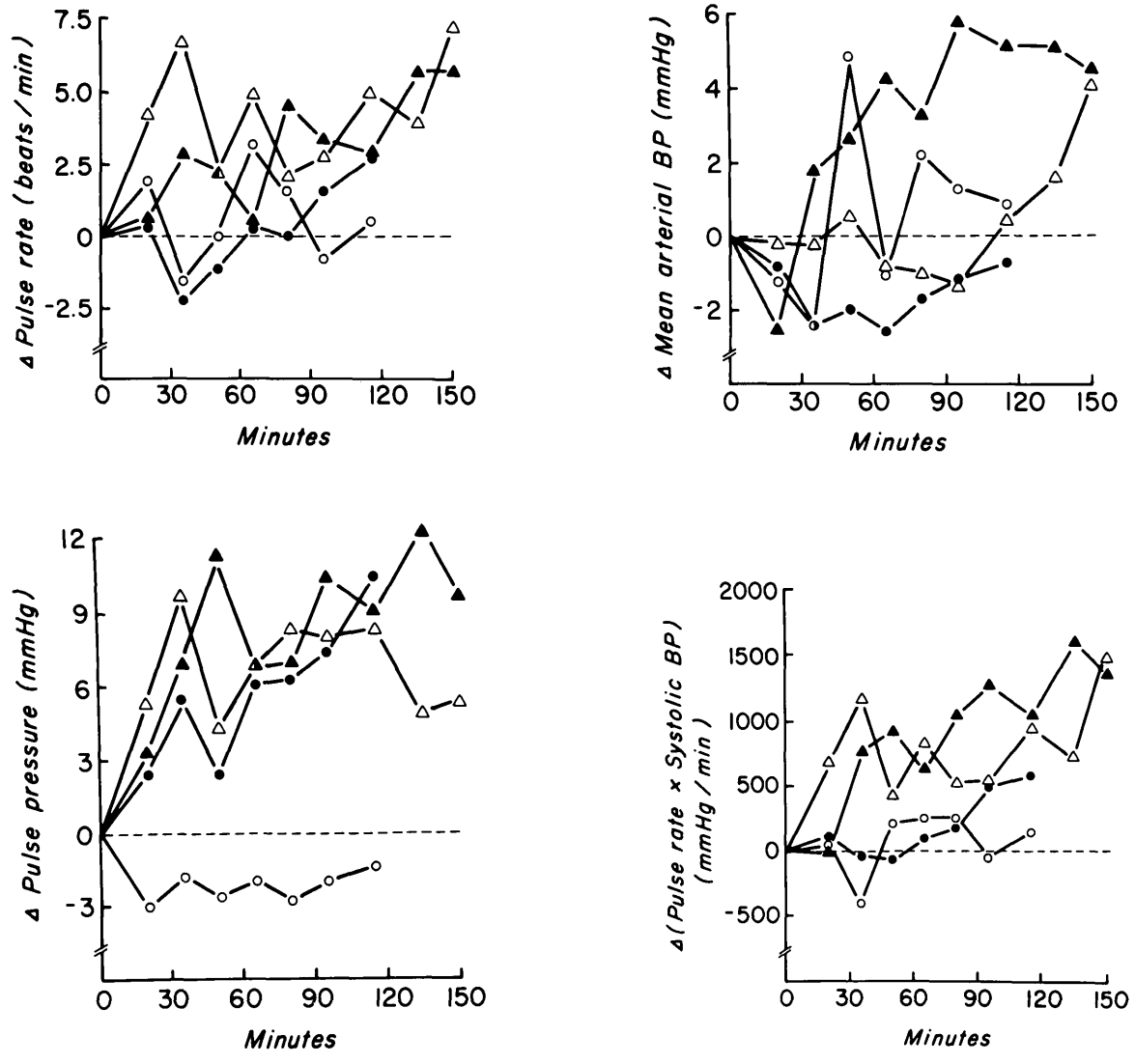


FIGURE 3. Changes from basal in pulse rate, mean arterial blood pressure, pulse pressure, and double product (pulse rate × systolic BP) during control, G + 125 hyperglycemic, and 2- and 5-mU euglycemic clamps. Points represent mean change in variable for five to seven subjects per test. Symbols are as indicated for Figure 1.

tween plasma norepinephrine and infused insulin. Moreover, the magnitude of the rise in plasma NE is not inconsiderable; in the 5-mU study, the NE level rose 117% above mean basal at 150 min, a rise similar to that observed in comparable subjects after upright posture.[7] The assessment of cardiovascular changes, although by methods that are inherently less precise than invasive techniques, yields data entirely consistent with activation of the sympathetic nervous system. The cardiovascular measurements, furthermore, are internally consistent; in the 2-mU study in the absence of a change in mean arterial blood pressure, pulse

TABLE 3
Effect of insulin infusion on NE clearance

Metabolic clearance rate for NE (ml/min)			NE disappearance rate (%/min)	
Subject	Control	2 mU	Control	2 mU
1	1160	1120	35	44
2	1800	1960	65	83
3	2610	1740	56	32
Mean	1860	1610	52	53
SE	420	250	9	15

TABLE 4
Summary of effects of control, hyperglycemic, and euglycemic glucose clamps on insulin, NE, and cardiovascular measurements

	Insulin	NE	Pulse rate	Pulse pressure	Double product	Mean arterial BP
Control	0	0	0	0	0	0
G + 125	+	0	0	+	0	0
2 mU	++	+	+	+	+	0
5 mU	+++	++	0	+	+	+

0 indicates no statistically significant change in a particular variable during the study.
+ indicates a statistically significant increase in a particular variable during the study.
++ and +++ indicate quantitatively greater changes in a particular variable in one test compared with another noncontrol test.

rate increased, while during the 5-mU infusion, mean arterial blood pressure increased without a rise in pulse rate.

Plasma NE elevations in response to insulin administration have been reported previously[15–18] and have usually been attributed to the insulin-induced fall in glucose concentration. The drop in glucose may be either to profoundly hypoglycemic levels of less than 50 mg/dl[15,16] or to relatively hypoglycemic levels of 65 mg/dl after a basal period at 95 mg/dl.[18] Such changes in blood glucose were not observed during the present studies. In the final 90 min of the 2- and 5-mU euglycemic glucose clamps, when plasma NE elevations were greatest, the blood glucose measurements obtained at 5-min intervals clustered around the desired basal values. Furthermore, blood glucose levels were similar in the two insulin infusion protocols while the rise in plasma NE was greater during the high dose 5-mU infusion. Thus, the rise in plasma NE with insulin administration during euglycemic glucose clamps is not secondary to an effect of insulin on blood glucose concentration. While an adrenal medullary origin for the plasma NE response to insulin cannot be excluded, the lack of correlation between measurements of plasma NE and decrements in blood glucose, the stimulus for adrenal medullary secretion, strongly points to the sympathetic nervous system as the source of the increase in plasma NE.

Another potential mechanism to account for an insulin-induced increase in plasma NE has been proposed by Gundersen and Christensen. They reported that glucose lowering by insulin to nonhypoglycemic levels in diabetic subjects was associated with a decrease in intravascular volume and a rise in plasma NE, which they assumed was secondary to baroreceptor-mediated stimulation of the sympathetic nervous system.[17] Subsequent workers have failed to confirm this contraction of plasma volume in diabetics following insulin administration;[19] moreover, no evidence exists to indicate that such a postulated effect of insulin on plasma volume occurs in nondiabetics when blood glucose is maintained constant, as in the current studies. In addition, the cardiovascular data obtained during the two insulin infusions are physiologically incompatible with a diminution in intravascular volume; with volume contraction, pulse pressure narrowed,[20] whatever the underlying cause, while in the euglycemic clamps, pulse pressure widened markedly. Thus, reflex activation of the sympathetic nervous system consequent to an insulin-induced reduction in plasma volume is not an adequate explanation for the rise in plasma NE with euglycemic insulin infusions.

An alternate possibility is that insulin or insulin-mediated glucose metabolism directly stimulates the sympathetic nervous system. Such a hypothesis has been raised before[21] and is consistent with the developing body of information suggesting that dietary carbohydrate has an important influence on sympathetic nervous system activity.[1–7] The continuing rise in plasma NE during the 30 min beyond cessation of the insulin infusions suggests that either insulin in some nonvascular compartment (perhaps the "third compartment" insulin described previously[22]) and/or a persistent action of insulin (as on intracellular glucose metabolism) may be the proximate stimulus to sympathetic activation. Comparable rates of peripheral glucose utilization (M) achieved in the G + 125 studies and in the 2-mU infusions suggest that the rate of glucose uptake by peripheral tissues is not the principal determinant of sympathetic activation. M, however, may not reflect glucose utilization in individual tissues and, thus, these results do not exclude a possible connection between sympathetic nervous system activity and glucose metabolism in a specific tissue compartment. The recent observation that 2-deoxyglucose, a nonmetabolizable glucose analogue, reduces sympathetic activity in rats[23] supports a role for glucose metabolism in mediating the effect of insulin on the sympathetic nervous system. Since changes in sympathetic outflow originate within the central nervous system, alterations in glucose metabolism within a small region of the brain, perhaps in the hypothalamus,[4] may coordinate sympathetic responses to changes in total body glucose metabolism and may serve as the link between administered insulin and sympathetic activation.

The high physiologic and supraphysiologic levels of circulating insulin achieved in the two infusion protocols, respectively, raise the possibility that insulin stimulation of the sympathetic nervous system may be of pharmacologic interest only. While this may be true, it must be recognized that peripheral infusions of insulin and glucose are in and of themselves nonphysiologic. Since metabolic responses to various substances (e.g., glucose and potassium) are influenced by the route of administration,[24] the effect of insulin on sympathetic activity in these studies may be diminished by the means chosen to deliver the insulin and glucose. Thus, the artificial nature of experimental design can only indicate potential physiologic effects of insulin, no matter what insulin dose is used.

While, as indicated above, extracellular volume considerations cannot account for the plasma NE elevations observed during the insulin infusions, they may explain the lack of plasma NE response in the G + 125 hyperglycemic glucose clamp. The sustained presence of the extracellular osmotic load in the G + 125 protocol creates a situation analogous to that produced by mannitol administration. While data to support a proposed sympatholytic effect of mannitol are not available, the decrease in renal vascular resistance following administration of hypertonic mannitol is consistent with such a hypothesis.[25] Thus, the altered volume status in the subjects during the G + 125 glucose clamp may have precluded the demonstration of a rise in plasma NE during glucose infusion, despite the endogenous insulin response.

A stimulatory action of insulin on the cardiovascular system, apart from hypoglycemic activation of the adrenal medulla, has been reported by numerous investigators. Insulin increases cardiac contractility whether measured in vitro with isolated papillary muscles or perfused whole hearts, or in vivo in anesthetized, open-chest preparations.[26–29] In intact, lightly anesthetized dogs, insulin administration leads to a transient rise in blood pressure of 70–80% before hypoglycemia occurs, a response which is actually enhanced by concomitant glucose infusion.[21] Associated with the brief elevation in mean arterial pressure is an increase in right atrial pressure, an observation which strongly argues against an effect of insulin to decrease intravascular volume in normal dogs. Furthermore, the transient hypertension is attenuated by ganglionic and adrenergic blockade and also follows intracarotid injections of insulin at doses which do not affect blood pressure when given systemically, thus providing evidence of central stimulation of peripheral sympa-

thetic activity by insulin. To what extent the cardiotonic effects of insulin in intact animals and man are directly produced or indirectly mediated through sympathetic stimulation remains to be determined. Nonetheless, the pattern of cardiovascular response observed during the insulin infusions is similar to that seen with exposure to environmental cold, a known sympathetic stimulus.[30]

The effects of insulin described here may have pharmacologic implications as well. The cardiotonic effect of glucose-insulin-potassium infusions may depend in part on sympathetic stimulation by insulin. The potential role of euglycemic insulin infusions in situations such as acute, severe injury,[31] in which sympathetic stimulation may be desirable, warrants further study.

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