**Portal Vein Glucose Sensors Do Not Play a Major Role in Modulating Physiological Responses to Insulin-Induced Hypoglycemia in Humans**

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OBJECTIVE—Experimental data from animal studies indicate that portal vein glucose sensors play a key role in the responses to slow-fall hypoglycemia. However, their role in modulating these responses in humans is not well understood. The aim of the present study was to examine in humans the potential role of portal vein glucose sensors in physiological responses to insulin-induced hypoglycemia mimicking the slow fall of insulin-treated diabetic subjects.

RESEARCH DESIGN AND METHODS—Ten nondiabetic subjects were studied on two different occasions during intravenous injection of food (10) and stimulation of net hepatic glucose uptake (2,13). In addition, portal glucose sensors modulate the sympathetic responses to hypoglycemia (14,15). However, how portal glucose sensors may affect sympathetic responses to hypoglycemia and how their activity integrates with that of glucose-sensitive areas in the brain is not well understood (16). In fact, several studies in animals indicate that the brain is the prominent center for the sensing of hypoglycemia. In dogs in which insulin-induced hypoglycemia was allowed to occur peripherally while brain euglycemia was maintained by glucose infusions in carotid and vertebral arteries bilaterally, the responses of counterregulatory hormones decreased nearly completely compared with dogs with brain neuroglycopenia (17,18). In rats, the ventromedial hypothalamus (VMH) appears to be necessary to trigger counterregulation during hypoglycemia. In fact, bilateral lesions of the VMH in conscious rats suppresses glucagon and catecholamine responses during hypoglycemia (19,20), suggesting that the VMH is one of the most important sites acting as a glucose sensor (21,22). However, there is evidence that in rats, activation of portal glucose sensors by glucose may be the most important modulators of sympathetic response to hypoglycemia, resulting in a significant suppression of this response (23–25).

Recent studies in rats have established that portal vein glucose sensors, responsible for hypoglycemic detection, extend beyond the portal vein being placed also in the superior mesenteric vein and that their role is essential in detecting slow, but not fast, fall in blood glucose (26).

Limited knowledge is available about the potential role of portal glucose sensors in humans. Only three studies (5–7) have addressed the question, with conflicting results. In fact, counterregulatory hormone responses to hypoglycemia have been found potentiated (5), reduced (6), or reduced in early phase and potentiated in late phase (7) after ingestion of oral glucose (5,6) or orange juice (7). It is likely that methodological differences account, at least in part, for these divergent results.

It has been suggested that glucose sensors in the portal area are necessary to monitor glucose derived from the gut (1). In fact, when exogenous glucose is infused directly in the portal vein (2,3) or in the duodenum (4) or ingested orally as glucose load (5–7), a portal-arterial glucose gradient is generated with glucose concentrations higher in the portal vein than in arterial circulation. Such portal-arterial glucose gradient generates a portal signal that is probably dependent on glucose-sensitive nerves in the portal veins, the firing rate of which is inversely proportional to the portal glucose concentration (8). The signal then moves through the hepatic vagal afferences to modulate the function of different tissues (e.g., liver, pancreatic β-cells) involved in the control of glucose homeostasis (9). In addition, signals enter the central nervous system to regulate hypothalamic functions, such as feeding and satiety (10). Recent evidence indicates that GLUT2 transporter is essential for glucose sensing by the portal glucose sensor (11,12) and also that glucagon-like peptide 1 (GLP-1) receptor is required for the function of the portal glucose sensor in mice (9).

Earlier studies in animals have shown that portal-arterial glucose gradient is involved in the control of intake of food (10) and stimulation of net hepatic glucose uptake (2,13). In addition, portal glucose sensors modulate the sympathetic responses to hypoglycemia (14,15). However, how portal glucose sensors may affect sympathetic responses to hypoglycemia and how their activity integrates with that of glucose-sensitive areas in the brain is not well understood (16). In fact, several studies in animals indicate that the brain is the prominent center for the sensing of hypoglycemia. In dogs in which insulin-induced hypoglycemia was allowed to occur peripherally while brain euglycemia was maintained by glucose infusions in carotid and vertebral arteries bilaterally, the responses of counterregulatory hormones decreased nearly completely compared with dogs with brain neuroglycopenia (17,18). In rats, the ventromedial hypothalamus (VMH) appears to be necessary to trigger counterregulation during hypoglycemia. In fact, bilateral lesions of the VMH in conscious rats suppresses glucagon and catecholamine responses during hypoglycemia (19,20), suggesting that the VMH is one of the most important sites acting as a glucose sensor (21,22). However, there is evidence that in rats, activation of portal glucose sensors by glucose may be the most important modulators of sympathetic response to hypoglycemia, resulting in a significant suppression of this response (23–25).

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So far, no study has investigated in humans the role of portal glucose sensors on counterregulation, symptoms, and cognitive function during hypoglycemia induced slowly, to mimic the hypoglycemia of the clinical situation (27). It is worthy of note that in the postprandial state, a condition characterized by glucose arriving in the portal vein from the gut, sympathetic responses and some aspects of cognitive function are affected by the rate of fall of blood glucose (27). However, in the postprandial condition, it is not only glucose that enters in the portal system but also other substrates that may suppress sympathoadrenal responses to hypoglycemia (28).

The aim of the present study was to examine in humans the potential effects of portal glucose sensors on hormonal counterregulatory responses and responses of symptoms and cognitive function in a model of slow-fall insulin-induced hypoglycemia. For this purpose, healthy subjects were studied during hypoglycemia preceded by ingestion of either oral glucose to prevent portal hypoglycemia or placebo.

**RESULTS**

**Plasma glucose, C-peptide, and insulin concentrations and rates of glucose infusion.** In both study conditions, plasma glucose was maintained at euglycemia for 30 min by variable infusion of glucose. Thereafter, similar increments were produced from 30 to 60 min in both study conditions, and then the rate of glucose infusion was decreased to reduce plasma glucose concentration to the target hypoglycemic plateau of 47 ± 1.2 mg/dl at 120 min. Subsequently, plasma glucose concentration was maintained at the nominal plateau of 47 mg/dl for 40 min until the end of the studies with no differences between study conditions (P > 0.2). The calculated rate of fall of plasma glucose from euglycemia (60 min) to first hypoglycemic plateau point (120 min) was 0.76 ± 0.05 mg·dl⁻¹·min⁻¹ in subjects given glucose and 0.65 ± 0.05 in those given placebo (P = 0.106) (Fig. 1). Plasma C-peptide concentrations decreased at 30 min in both studies. However, at 60 min, plasma C-peptide concentration increased in the glucose study (1.3 ± 0.4 nmol/l), whereas it continued to decrease in the placebo study (0.5 ± 0.1 nmol/l, P = 0.035). Thereafter, plasma C-peptide concentrations decreased in both studies, although, on average, they were higher in subjects given glucose than in those given placebo (0.65 ± 0.2 and 0.34 ± 0.1 nmol/l, respectively, P < 0.04). Plasma insulin concentrations was similar in placebo and glucose studies (185 ± 4 and 181 ± 6 μU/ml, respectively, P > 0.2). The rates of glucose infusion were lower in the glucose and placebo compared with the placebo study (5.7 ± 0.4 vs. 2.8 ± 0.4 mg·kg⁻¹·min⁻¹, respectively, P < 0.001).

Plasma glucagon, norepinephrine, adrenaline, pancreatic polypeptide, cortisol, and growth hormone concentrations.

After an initial decrease at 60 min in both studies, plasma glucagon concentrations increased to
Levels were not different between glucose and placebo, respectively, $P = 0.018$, and from $252 \pm 16$ to $185 \pm 19\text{ pmol}/l$, $P = 0.034$, placebo and glucose studies, respectively) with no difference between study conditions ($P > 0.2$).

**Plasma nonglucose substrate.** Plasma FFA levels decreased with no difference between studies ($68 \pm 5$ and $84 \pm 12\text{ µmol}/l$ in placebo and glucose studies, respectively, $P > 0.2$). Similarly, plasma glycerol and $\beta$-OH-butyrate concentrations decreased from baseline with no difference between placebo and glucose studies (Fig. 3). Plasma lactate concentrations increased in both studies. The first similar increment was observed at 60 min, and then a further increase was detected at 140 min with no difference between studies ($P = 0.075$). Plasma alanine concentrations were similar during placebo and glucose studies ($366 \pm 45$ and $369 \pm 29\text{ µmol}/l$, respectively, $P > 0.2$).

**Symptoms.** Symptom scores increased during hypoglycemia during both placebo and glucose studies. However, mean and peak values for total, autonomic, neuroglycopenic, adrenergic, and cholinergic symptom scores were not different between studies ($P > 0.2$ for all comparisons) (Fig. 4).

**Cognitive function.** With the exception of digit vigilance test, all cognitive tests deteriorated significantly during hypoglycemia both in placebo and glucose studies (Table 1). The Stroop color and colored words subtest deteriorated less in the glucose study than in the placebo study ($P < 0.05$).

**DISCUSSION**

The present study was undertaken to examine the effects of oral glucose on the counterregulatory, symptomatic, and cognitive responses to slow-fall hypoglycemia in humans after concomitant ingestion of either glucose to prevent portal hypoglycemia while maintaining systemic hypoglycemia or placebo. The results indicate that prevention of portal hypoglycemia by oral glucose did not have any impact on counterregulatory and symptomatic response to hypoglycemia. Thus, this study supports the view that in nondiabetic subjects, it is systemic glucose sensing (i.e., the brain) that plays the key counterregulatory role when plasma glucose decreases (16). However, oral glucose affected responses of $\beta$-cells of pancreatic islets, as shown by the greater response of C-peptide. In addition, oral glucose preserved some aspects of cognitive function during hypoglycemia, such as mental flexibility and attention.

It has been suggested that glucose sensors localized in the portal vein modulate sympathoadrenal responses to hypoglycemia in rats (14) and dogs (15). In these animals, the increase in portal glucose levels, by infusing glucose directly in the portal vein, in the face of systemic hypoglycemia, causes a net suppression of the sympathoadrenal response, which is, instead, normally observed when hypoglycemia is allowed to occur in the portal vein (14,15).

Interestingly, recent studies in rats indicate that there are glucose sensors in the superior mesenteric vein (25), in addition to the portal vein, and that they play an essential role in sensing slow, but not fast, fall in blood glucose defined as rate of decrease of $1.6$ and $3.78\text{ mg} \cdot \text{dl}^{-1} \cdot \text{min}^{-1}$, respectively (26). In humans, it has been shown statistically significant ($P = 0.107$). Plasma ghrelin concentrations (data not shown) decreased over time from basal to nadir values during the hypoglycemic plateau (from $268 \pm 32$ to $172 \pm 19\text{ pmol}/l$, $P = 0.018$, and from $252 \pm 16$ to $185 \pm 19\text{ pmol}/l$, $P = 0.034$, placebo and glucose studies, respectively) with no difference between study conditions ($P > 0.2$).

Fig. 1. Plasma glucose, C-peptide, insulin concentrations, and rates of glucose infusion during clamped hypoglycemia both with placebo (●) and oral glucose (○).

A peak of $156 \pm 20\text{ pg}/ml$ (vs. baseline $68 \pm 3\text{ pg}/ml$, $P = 0.001$) in the placebo and to $143 \pm 17\text{ pg}/ml$ (vs. baseline $66 \pm 4.6\text{ pg}/ml$, $P = 0.001$) in the glucose study (Fig. 2). These differences were not statistically significant ($P > 0.02$). Plasma pancreatic polypeptide concentration increased both during placebo and glucose studies with no difference between studies (peak response $155 \pm 20$ and $166 \pm 18\text{ pmol}/l$, respectively, $P > 0.2$). Plasma adrenaline levels were not different between glucose and placebo studies (peak response $4 \pm 0.9$ and $3.7 \pm 1.0\text{ nmol}/l$, respectively, $P = 0.271$). Similarly, plasma norepinephrine concentrations increased in both placebo and glucose studies with no difference between studies. Responses of plasma cortisol were not different between studies (peak response $16.2 \pm 3.7$ and $15.5 \pm 2.6\text{ µg}/dl$, respectively, $P > 0.2$). Plasma growth hormone increased more in response to glucose than to placebo, although the difference was not
that in postprandial conditions (27), when portal glucose levels are higher than those in systemic circulation, responses to hypoglycemia are affected by the rate of fall of glucose. Specifically, the sympathoadrenergic response was greater in slow compared with fast fall of blood glucose, whereas cognitive function deteriorated more during fast fall of blood glucose. Notably in humans, slow and fast fall are usually at rates of 0.6 and 1.8 mg · dl⁻¹ · min⁻¹, respectively (27). However, under those conditions, the relative contribution of glucose and of other nutrients cannot be separated. Therefore, we carried out the present study by inducing hypoglycemia (47 mg/dl) slowly, over 60 min at the rate of 0.75 mg · dl⁻¹ · min⁻¹ (slow fall) and maintaining the hypoglycemic plateau for an additional 40 min. Because the rate of fall in our study was slower compared with that achieved in rats (~1.6 mg · dl⁻¹ · min⁻¹) (26) in which slow-fall but not fast-fall hypoglycemia had effects, it is concluded that in humans the portal glucose sensors do not play a major counter-regulatory role in the clinical situation in contrast to rats.

The oral load (28 g) was given before hypoglycemia, which was allowed to occur slowly. As a consequence, from the beginning of hypoglycemia the portal vein was exposed to high glucose levels arriving from the intestine. Most likely, elevation of portal plasma glucose prevented activation of portal sensors during systemic hypoglycemia. If portal glucose sensors had a prominent role in modulating the sympathoadrenal response, we should have observed a suppression of this response compared with placebo, as described in animals (14,15). However, that was not the case. Likely, species differences account for the different findings in the present study in humans compared with previous study in rats (26).

In the present study, sympathoadrenal responses, adrenaline and norepinephrine, were not different after oral glucose compared with placebo. In addition, we did not
observe any effects of oral glucose compared with placebo on the other counterregulatory hormones, such as glucagon, cortisol, and growth hormone. Plasma concentrations of ghrelin, a potent stimulator of growth hormone secretion (40,41), were similarly suppressed during insulin-induced hypoglycemia in both study conditions. Ghrelin decreases during insulin-induced hypoglycemia, being inhibited by hyperinsulinemia (42). Although oral glucose is a further mechanism of ghrelin inhibition (43), likely hyperinsulinemia was the main determinant of its suppression. Finally, it is interesting to note that, according to plasma ghrelin levels, the hunger symptom, which has been shown to increase after ghrelin administration (44), in our study was not different under both hypoglycemic conditions.

The greater plasma C-peptide levels observed after oral glucose might suggest that greater insulin levels have occurred in the portal area in the face of identical peripheral arterial plasma insulin concentrations. This is likely to be explained both by the slight increase in arterial plasma glucose after oral glucose and by incretin effects mediated mostly by GLP-1 release (45). Certainly GLP-1 favored insulin secretion in the portal vein. However, the relative contribution to portal hyperinsulinemia of GLP-1 and of the oral glucose per se cannot be inferred from our study. Although we did not measure GLP-1, its glucose-independent stimulation of insulin secretion after ingestion of glucose is well known (45).

GLP-1 has been found to suppress glucagon in healthy subjects under euglycemic (46) but not during hypoglycemic conditions (47). Because in our study glucagon levels were similar with oral glucose and placebo, it is likely that any inhibitory effect of GLP-1 on glucagon secretion was offset by the predominant stimulatory effects of systemic hypoglycemia achieved during the clamp procedure.

In line with counterregulatory hormone responses,
symptoms of hypoglycemia were not affected by oral glucose, in contrast to results previously reported by Smith et al. (6). However, we did find better preservation in the color and color-words subtests of the Stroop test. This finding is intriguing. Speculatively, it might be related to neuroprotective effects of GLP-1 (48), although peripheral infusion of GLP-1 during stepped hypoglycemic clamps does not alter cognitive function in healthy subjects (47). However, these results do not negate a possible effect related to portal increments of GLP-1 and its involvement in the generation of neuroprotective portal signals capable of modulating aspects of cognitive function.

One important limitation of our study is that we have no direct measurement of portal vein glucose and do not know with absolute certainty the degree of portal glucose elevation achieved after oral glucose. Therefore, the conclusions of the present study that portal vein glucose sensors do not play a role in hypoglycemia in humans rely entirely on the assumption that oral glucose prevented hypoglycemia in the portal vein in our experimental model during the slow fall of blood glucose (60–120 min) and during clamped hypoglycemia (120–160 min). We believe that this was the case for the following reasons. In fact, Smith et al. (6) have shown that the ingestion of 20 g glucose labeled with the tracer $[^{13}C_6]$glucose is fully absorbed in a parabolic manner over $\sim 2$ h in humans. Based on this finding, it is conceivable that in our study, in which a larger amount of oral glucose was given (28 g), portal hypoglycemia was prevented, both during the period of time of the slow fall of blood glucose (for 60 min) and during the stable hypoglycemic plateau (additional 40 min). In addition, and most importantly, the lower amount of glucose infused during hypoglycemia in the oral glucose study compared with placebo represents a compensation.
for the arrival of oral glucose absorbed from the intestine
in the portal vein and in the systemic circulation. Thus, it
is conceivable that the difference in glucose infusion rates
(GIRs) between the two studies represents a reliable
estimate of intestinal absorption of glucose, ultimately
elevating portal glucose. Based on the Smith et al. cal-
ulation of the rate of systemic absorption of the oral glucose
from the intestine, assuming absorption over 130 min and
a portal blood flow of 0.8 l/min, it is possible to estimate
that the administration of 28 g glucose in our study
increased the venous portal glucose by a mean of
200 DIABETES, VOL. 58, JANUARY 2009
Our results are at variance with previous studies in humans (5–7). However, only one study aimed at prevention of portal hypoglycemia after administration of 20 g oral glucose (6). In contrast to our study, Smith et al. (6) reported a slightly lower response of adrenaline early after oral glucose. However, the effect was modest and not confirmed over the entire hypoglycemic plateau (6). In the present study, greater oral glucose (28 g) given earlier than in Smith study and in a model of slow-fall hypoglycemia to prevent portal hypoglycemia had no effects.

The two other studies (5,7) do not explore the role of portal-hepatic glucose sensors during slow-fall hypoglycemia. In fact hypoglycemia was induced quickly in both studies (5,7); respectively ~2–4 and ~1.3 mg · dl−1 · min−1. In addition, in one study (5) oral glucose was given after, not before, induction of hypoglycemia; in the other study (7), a small amount of carbohydrates as orange juice (15 g), not glucose, was given, and calculations of estimated portal plasma glucose were not performed.

In conclusion, the results of the present study indicate that ingestion of oral glucose to prevent portal hypoglycemia from an early phase of slow-fall systemic hypoglycemia does not affect responses of counterregulatory hormones and symptoms to hypoglycemia. Thus, in contrast to rats (25), the putative portal glucose sensors in humans do not play an appreciable role in modulating these responses to hypoglycemia, at least in the clinically relevant condition of slow-fall hypoglycemia. In humans, portal glucose sensors likely have a role different from that in animals (14,15,23–25). Additional studies are required to explore the complex potential of portal glucose sensors in glucose homeostasis in humans.

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REFERENCES
1. Niijima A: Afferent impulse discharges from glucoreceptors in the liver of the guinea pig. Ann N Y Acad Sci 157:700–709, 1969
2. Pagliassotti MJ, Holste LC, Moore MC, Neal DW, Cherrington AD: Comparison of the time courses of insulin and the portal signal on hepatic glucose and glycogen metabolism in the conscious dog. J Clin Invest 97:81–91, 1996
3. Moore MC, Cardin S, Edgerton DS, Farmer B, Neal DW, Lautz M, Cherrington AD: Unlike mice, dogs exhibit effective gluco-regulation during insulin-induced hypoglycemia in dogs. Diabetes 43:1052–1060, 1994
4. Zangeneh F, Basu R, Shah P, Arora P, Camilleri M, Rizza RA: Enteral glucose homeostasis in humans. Diabetes 45:1052–1060, 1996
5. Hamilton-Wessler M, Bergman RN, Halter JB, Shulman GI: Local ventromedial hypothalamus glucopenia triggers counterregulatory hormone response. Diabetes 45:1929–1934, 1996
6. Smith D, Pernet A, Reid H, Bingham E, Rosenthal JM, Macdonald IA, Umpleby AM, Amiel SA: The role of hepatic portal glucose sensing in sympathoadrenal response to hypoglycemia. Diabetes 49:1643–1648, 2000
7. Cherrington AD: Central versus peripheral glucose sensing and the response to hypoglycemia. Diabetes 57:1158–1159, 2008
8. Niijima A: The effect of D-glucose on the firing rate of glucose-sensitive vagal afferents in the liver in comparison with the effect of 2-deoxy-D-glucose. J Auton Nerv Syst 10:255–260, 1984
9. Burcelin R, Da Costa A, Drucker D, Thorens B: Glucose competence of the hepatopetal vein sensor requires the presence of an activated glucagon-like peptide-1 receptor. Diabetes 50:1720–1728, 2001
10. Schmitt M: Inflences of hepatic portal receptors on hypothalamic feeding and satiety centers. Am J Physiol 225:1089–1095, 1973
11. Burcelin R, Dolci W, Thorens B: Glucose sensing by the hepatopetal sensor is GLUT2-dependent: in vivo analysis in GLUT2-null mice. Diabetes 49:1643–1648, 2000
12. Thorens B: GLUT2 in pancreatic and extra-pancreatic gluco-detection (Review). Mol Membr Biol 18:265–273, 2001
13. Galassetti P, Chu CA, Neal DW, Reed GW, Wasserman DH, Cherrington AD: A negative arterial-portal venous glucose gradient increases net hepatic glucose uptake in euglycemic dogs. Am J Physiol 277:E126–E134, 1999
14. Donovon CM, Halter JB, Bergman RN: Impotence of hepatic glucose receptors in sympathoadrenal response to hypoglycemia. Diabetes 40:155–158, 1991
15. Hamilton-Wessler M, Bergman RN, Halter JB, Watanabe RM, Donovon CM: The role of liver glucosesensors in the integrated sympathetic response induced by deep hypoglycemia in dogs. Diabetes 43:1052–1060, 1994
16. Cherrington AD: Reliability of glucose sensors in the central nervous system. J Auton Nerv Syst 54:127–134, 1995
17. Biggers DW, Myers SR, Neal D, Stinson R, Cooper NB, Jaspan JB, Williams PE, Cherrington AD, Frizzell RT: Role of brain in counterregulation of insulin-induced hypoglycemia in dogs. Diabetes 38:7–16, 1989
18. Prizzell RT, Jones EM, Davis SN, Biggers DW, Myers SR, Connolly CC, Neal DW, Jaspan JB, Cherrington AD: Counterregulation during hypoglycemia is directed by widespread brain regions. Diabetes 42:1253–1261, 1993
19. Borg WP, Sherwin RS, During MJ, Borg MA, Shulman GI: Local ventromedial hypothalamus glucopenia triggers counterregulatory hormone release. Diabetes 45:179–184, 1996
20. Borg MA, Sherwin RS, Borg WP, Tamborlane WV, Shulman GI: Local ventromedial hypothalamus glucose perfusion blocks counterregulation during systemic hypoglycemia in awake rats. J Clin Invest 99:351–356, 1997
21. Smith D, Amiel SA: The anatomy of the human hypoglycaemia sensor. Diabetes Nutr Metab 15:316–318, 2002
22. McCormron R: The mechanisms that underlie glucose sensing during hypoglycemia in diabetes. Diabet Med 25:513–522, 2008
23. Donovon CM, Hamilton-Wessler M, Halter JB, Bergman RN: Primacy of liver glucosesensors in the sympathetic response to progressive hypoglycemia. Proc Natl Acad Sci U S A 91:2963–2967, 1994
24. Hevener AL, Bergman RN, Donovon CM: Novel glucose sensor for hypoglycemic detection localized to the portal vein. Diabetes 46:1521–1525, 1997
25. Hevener AL, Bergman RN, Donovon CM: Hypoglycemic detection does not occur in the hepatic artery or liver: findings consistent with a portal vein glucosesensor locus. Diabetes 50:499–503, 2001
26. Saberi M, Bohland M, Donovon CM: The locus for hypoglycemic detection shifts with the rate of fall in glycemia: the role of portal-superior mesenteric vein glucose sensing. Diabetes 57:1380–1386, 2008
27. Fanelli CG, Pampanelli S, Porcellati F, Bartocci L, Scionti L, Rossetti P, Mitrakou A, Ryan C, Veneman T, Mokan M, Jenssen T, Kiss I, Durrant J, Cherrington AD: Afferent neural impulses from arterial and portal vein glucose sensors in conscious dogs. Diabetes 49:1643–1648, 2000
28. Matveyenko AV, Donovan CM: Metabolic sensors mediate hypoglycemic detection at the portal vein. Diabetes 55:1276–1282, 2006
29. McGuire E, Helderman J, Tobin J, Andres R, Berman M: Effects of arterial venous sampling on analysis of glucose kinetics in man. J Appl Physiol 41:565–573, 1976
30. Mitrakou A, Ryan C, Veneman T, Mokan M, Jenssen T, Kiss I, Durrant J, Cryer PE, G erich J: Hierarchical of glycemic thresholds for counterregulatory hormone secretion, symptoms, and cerebral dysfunction. Am J Physiol 260:E67–E74, 1991
31. Cryer PE: Symptoms of hypoglycemia, thresholds for their occurrence, and hypoglycemia unawareness. Endocrinology 98:454–500, 1999
32. Bol l AB, Barth J: Neuropsychology of brain damage. In Handbook of Clinical Neuropsychology. Flis skov S, Boll T, Eds. New York, Wiley, 1981, p. 418–452
33. Lezak MD, Howieson DB, Loring DW, Hannay HJ, Fischer JS: Neuropsychologica l Assessment. 4th ed. New York, Oxford University Press, 2004, p. 429–550
34. N euforia Boll: Manual of the Wechsler Adult Intelligence Scale-Revised. New York, The Psychological Corporation Limited, 1981
35. Golden C: Stroop Color and Word Test. Chicago, Stoelting, 1978
36. Gronwall DMA: Paced auditory serial-addition task: a measure of recovery from concussion. Percept Mot Skills 44:367–373, 1977
37. Fanelli CG, De Feo P, Porcellati F, F errilli G, Torlone E, Santusiano F, P. ROSETTI AND ASSOCIATES

DIABETES, VOL. 58, JANUARY 2009 201
GLUCOSE COUNTERREGULATION

Brunetti P, Bolli GB: Adrenergic mechanisms contribute to the late phase of hypoglycemic glucose counterregulation in humans by stimulating lipolysis. J Clin Invest 89:2005–2013, 1992
38. Winer BJ, Brown DR, Michels KM. Statistical Principles in Experimental Design. 3rd ed. New York, McGraw Hill, 1991, p. 497–582
39. Holland BS, Copenhagen M. Improved Bonferroni type multiple testing procedures. Psychol Bull 104:145–148, 1988
40. Takaya K, Ariyasu H, Kanamoto N, Iwakura H, Yoshimoto A, Harada M, Mori K, Komatsu Y, Usui T, Shimatsu A, Ogawa Y, Hosoda K, Akamizu T, Kojima M, Kangawa K, Nakao K: Ghrelin strongly stimulates growth hormone release in humans. J Clin Endocrinol Metab 85:4908–4911, 2000
41. Van der Lely AJ, Tschöp M, Heiman MI, Ghigo E: Biological, physiological, pathophysiological, and pharmacological aspects of ghrelin. Endocr Rev 25:426–457, 2004
42. Lucidi P, Murdolo G, Di Loreto C, De Cicco A, Parlanti N, Fanelli C, Santususiano F, Bolli GB, De Feo P: Ghrelin is not necessary for adequate hormonal counterregulation of insulin-induced hypoglycemia. Diabetes 51:2911–2914, 2002
43. Broglio F, Gottero C, Prodam F, Destefanis F, Gauna C, De Cicco A, Parlanti N, Fanelli C, Santususiano F, Bolli GB, De Feo P: Ghrelin is not necessary for adequate hormonal counterregulation of insulin-induced hypoglycemia. Diabetes 51:2911–2914, 2002
44. Wren AM, Seal LJ, Cohen MA, Brynes AE, Frost GS, Murphy KG, Dhillon WS, Ghafei MA, Bloom SR: Ghrelin enhances appetite and increases food intake in humans. J Clin Endocrinol Metab 86:5892, 2001
45. Nauck MA, Homberger E, Siegel EG, Allen RC, Eaton RP, Ebert R, Creutzfeldt W: Incretin effects of increasing glucose loads in man calculated from venous insulin and C-peptide responses. J Clin Endocrinol Metab 63:492–498, 1986
46. Qualmann C, Nauck MA, Holst JJ, Orskov C, Creutzfeldt W: Insulinoergic actions of intravenous glucagon-like peptide-1 (GLP-1) [7-36 amide] in the fasting state in healthy subjects. Acta Diabetol 32:13–16, 1995
47. Nauck MA, Heimesaat MM, Behle K, Holst JJ, Nauck MS, Ritzel R, Hüfner M, Schniegel W: Effects of glucagon-like peptide 1 on counterregulatory hormone responses, cognitive functions, and insulin secretion during hyperinsulinemic, stepped hypoglycemic clamp experiments in healthy volunteers. J Clin Endocrinol Metab 87:1239–1246, 2002
48. During MJ: 2003 Glucagon-like peptide-1 receptor is involved in learning and neuroprotection. Nat Med 9:1173–1179, 2003
49. Radziuk J, McDonald TJ, Rubenstein D, Dupre J: Initial splanchnic extraction of ingested glucose in normal man. Metabolism 27:657–669, 1978
50. Sacca L, Cicala M, Corso G, Ungaro B, Sherwin RS: Effect of counterregulatory hormones on kinetic response to ingested glucose in dogs. Am J Physiol 240:E465–E473, 1981
51. Petersen KP, Cline GW, Gerard DP, Magnusson I, Rothman DL, Shulman GI: Contribution of net hepatic glycogen synthesis to disposal of an oral glucose load in humans. Metabolism 50:598–601, 2001