Insulin Counterregulatory Hormones Are Ineffective in Neonatal Hyperinsulinemic Hypoglycemia

Richard M. Cowett, Robert E. Rapoza, and Nancy L. Gelardi

Insulin counterregulatory hormones play a major role in the maintenance of glucose homeostasis. To evaluate the hypothesis that the reported imprecise control of glucose production by insulin is mirrored by a corresponding lack of response to the various insulin counterregulatory hormones, 30 spontaneously delivered mixed-breed term lambs weighing 4.9 ± 0.5 kg (mean ± SD) were studied at 5.0 ± 0.7 days after birth following administration of 100 μCi D-[6-$^3H_2$]glucose in 0.9% NaCl by the primed-constant infusion technique to measure glucose kinetics. Infusion of 2.0 mU · kg^{-1} · min^{-1} insulin produced hyperinsulinemic hypoglycemia and was combined with 1.0 mg · kg^{-1} · min^{-1} somatostatin (SRIF) to block insulin, glucagon, and growth hormone release. Infusion of 2 ng · mg^{-1} · min^{-1} glucagon or 10 μg · kg^{-1} · h^{-1} growth hormone with SRIF and insulin isolated the glucagon or growth hormone effect, respectively. The addition of metyrapone blocked cortisol release. Controls received only the isotope. In toto, the data can be interpreted to suggest that insulin has a greater effect on glucose uptake than on glucose production, and that neither glucagon, growth hormone, nor cortisol appreciably influenced the endogenous glucose production rate (Rp) during hyperinsulinemic hypoglycemia. The imprecise effect of these insulin counterregulatory hormones on neonatal glucose production mirrors the previously documented imprecise control by insulin.

AS DISCUSSED ELSEWHERE, the neonate is in a transitional state of glucose homeostasis. While the fetus is completely dependent on the mother for glucose (nutrient) transfer across the placenta, the nonpregnant nondiabetic adult is independent. The neonate must obtain substrate, especially carbohydrate, for energy and growth to maintain a balance between glucose deficiency and excess.[1]

As the major glucoregulatory hormone, insulin suppresses glucose production and accelerates glucose utilization. Insulin appears to be important in the hormonal control of neonatal glucose homeostasis, although adult-like control of glucose homeostasis is not evident in the neonate in most published series.[2-5]

It is apparent that maturation of neonatal glucose homeostasis results from a balance between substrate availability, insulin action, and the developing insulin counterregulatory hormones. Most investigations in this area to date have focused on either insulin action or the insulin counterregulatory hormones rather than substrate availability.[6-9]

There is a paucity of data about the effect of insulin counterregulatory hormones during the neonatal period. Under conditions of euglycemia, Cowett[7] suggested that there may be a decreased epinephrine response influencing glucose homeostasis (kinetics) in the neonatal period as compared with the adult. In parallel studies in the neonatal puppy, Hetenyi et al[10] concluded that the pancreatic and gastric α cells are unresponsive to stimuli normally effective in grown dogs.

To evaluate the importance of insulin counterregulatory hormones in the ontogeny of glucose homeostasis in the neonatal period, the present studies have been performed in a lamb model of hyperinsulinemic hypoglycemia. We hypothesized that the reported imprecise control of glucose production by insulin is mirrored by a corresponding lack of response to the various insulin counterregulatory hormones.

MATERIALS AND METHODS

Thirty mixed-breed neonatal lambs weighing 4.9 ± 0.5 kg (mean ± SD) were studied at 5.0 ± 0.7 days after birth. The ewes were allowed to deliver spontaneously, and the lambs were fed ad libitum by their mother until approximately 12 hours prior to the study. The lamb was considered a nonruminant, since it had not been weaned. The catheterization protocol has previously been described in detail.[6-9,11] At the time of catheterization, the animal was lightly restrained and blindfolded. Under local anesthesia of 1% lidocaine (Elkins-Sinn, Cherry Hill, NJ), the internal carotid artery and external jugular vein were catheterized for blood sampling and infusion, respectively.

After 1 hour of stabilization following the catheterization procedure, the lamb received 100 μCi · kg D-[6-$^3H_2$]glucose in 0.9% NaCl at 0.06 mL · kg^{-1} · min^{-1} by the primed-constant infusion technique of Steele for 150 minutes. During the next 30 minutes, a total of four baseline samples were drawn every 10 minutes for determination of plasma concentrations of glucose, insulin, glucagon, growth hormone, cortisol, and glucose specific activity. Corresponding recordings of the heart rate and systolic and diastolic blood pressure were made as well. Subsequently, blood samples were taken and measurements were recorded every 20 minutes during the experimental period when one of the following six combinations of infusates were administered: (1) five lambs received 1.0 μg · kg^{-1} · min^{-1} somatostatin ([SRIF] Sigma, St Louis, MO) to block insulin, glucagon, and growth hormone release (SRIF group); (2) five lambs received 1.0 μg · kg^{-1} · min^{-1} SRIF and 2 mU · kg^{-1} · min^{-1} insulin (Eli Lilly, Indianapolis, IN) to produce hyperinsulinemic hypoglycemia (hyperinsulinemia group); (3) five lambs received 1.0 μg · kg^{-1} · min^{-1} SRIF, 2 mU · kg^{-1} · min^{-1} insulin, and 0.3 mg · kg^{-1} · min^{-1} glucagon (Eli Lilly) to isolate the glucagon effect (glucagon group); (4) five lambs received 1.0 μg · kg^{-1} · min^{-1} SRIF, 2 mU · kg^{-1} · min^{-1} insulin, and 10 μg · min^{-1} · h^{-1} growth hormone (Sigma) to isolate the growth hormone effect (growth hormone group); (5) five lambs received 1.0 μg · kg^{-1} · min^{-1} SRIF, 2 mU · kg^{-1} · min^{-1} insulin, 0.3 mg · kg^{-1} · min^{-1} glucagon, 10 μg · kg^{-1} · h^{-1} growth hormone, and 0.3 mg · kg^{-1} · min^{-1} metyrapone (Sigma) to isolate the cortisol effect (metyrapone group); and (6) five lambs continued to receive 0.9% NaCl alone as the diluent for the isotopic tracer throughout the study (control). The administered doses

From the Department of Pediatrics, Brown University School of Medicine, Providence; and Department of Pediatrics, Women & Infants' Hospital of Rhode Island, Providence, RI.

Submitted April 10, 1998; accepted November 23, 1998.

Supported in part by National Institutes of Health Grant No. R-01-27287.

Address reprint requests to Richard M. Cowett, MD, Department of Neonatology, The Cleveland Clinic Foundation Children's Hospital-M 62, 9500 Euclid Ave, Cleveland, OH 44195-0001.

0026-0495/99/4805-0006$10.00/0

of the infusates were similar to those previously used in studies in this lamb model or other animal models,[10] or modified from those used in studies in adult males on a per-kilogram basis.[12,13] In a parallel series of studies to be reported elsewhere, α- and β-adrenergic blockade was used separately and in concert to isolate the adrenergic effects using the same model. In this series of studies, adrenergics were not blocked and remain an underlying component of the physiological response throughout.

The plasma glucose concentration was measured on a YSI 2700 Select Biochemistry Analyzer (YSI, Yellow Springs, OH). The insulin level was measured by a single-antibody radioimmunoassay (Coat-A-Count Insulin; Diagnostic Products, Los Angeles, CA). The glucagon concentration was measured by a double-antibody radioimmunoassay (ICN Pharmaceuticals, Costa Mesa, CA). The cortisol level was measured by a single-antibody radioimmunoassay (Clinical Assays GammaCoat Cortisol ^{125}I RIA Kit; Incstar, Stillwater, MN). The growth hormone level was measured by a single-antibody radioimmunoassay (National Hormone and Pituitary Program, NIH, NIDDK, Bethesda, MD). Blood gases were measured on a Ciba-Corning 238 pH/blood gas analyzer (Ciba-Corning, Norwood, MA). Heart rate and systolic and diastolic blood pressure were recorded on a Corometrics 556 Monitor (Corometrics Medical Systems, Wallingford, CT).

The procedure for glucose specific activity has been previously published.[6-9,11] From the determination of glucose specific activity, the glucose production rate was calculated according to the equations of Steele et al [14]

Statistical analysis was performed by SAS Proc Mixed Design in a two-stage model (SAS Institute, Cary, NC). The data obtained during the baseline period were averaged to determine the mean value for each concentration (ie, blood glucose) or measurement (ie, heart rate per minute) for each group. Cluster analysis of the specific concentration or measurement for each neonate provided a regression for the specific parameter being evaluated over the 2-hour period of the perturbation (ie, plasma glucose concentration or heart rate). Subsequently, the specific individual neonatal regression coefficients were combined to calculate a group regression coefficient for each specific parameter. The six groups were subsequently compared, and a Bonferroni correction was calculated for sample size. Statistical significance was set at a P level of .0083 or less.

RESULTS

Figure 1 depicts the blood glucose concentration over time through the 2-hour period of administration by group. There were no significant differences between the mean blood glucose concentration of the groups during the basal period. Over time, there was a significant increase in blood glucose in the SRIF group compared with the control group ($P < .0001$) and a significant decrease in blood glucose in the hyperinsulinemia, glucagon, growth hormone, and metyrapone groups versus the control group ($P < .0001$). Blood glucose concentrations of the hyperinsulinemia, glucagon, growth hormone, and metyrapone groups were not significantly different.

Figure 1 depicts the specific activity for Rp over time through the 2-hour period of administration by group. There were no significant differences between the mean specific activity for Rp of the groups during the basal period or throughout the period of the various infusions.

Figure 1 depicts the mean Rp over time through the 2-hour period of administration by group. There were no significant differences between the Rp of the groups during the basal period. Over time, there was a significant increase of the Rp of the SRIF group compared with the control group ($P < .0033$) and a significant decrease of the Rp of the hyperinsulinemia and growth hormone groups compared with the control group ($P < .0002$). The Rps of hyperinsulinemia, glucagon, growth

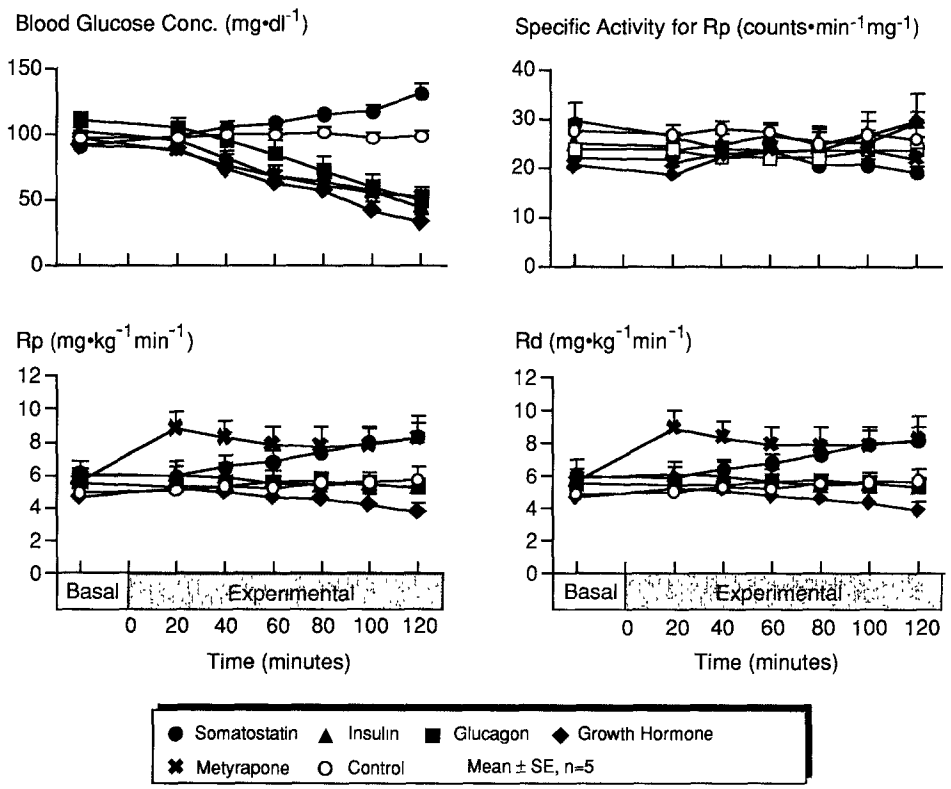

Fig 1. Blood glucose concentration, specific activity for glucose production (Rp), glucose production rate (Rp), and glucose utilization rate (Rd) over time from the means of the basal period through the 2-hour period of administration of the various infusions by group (experimental).

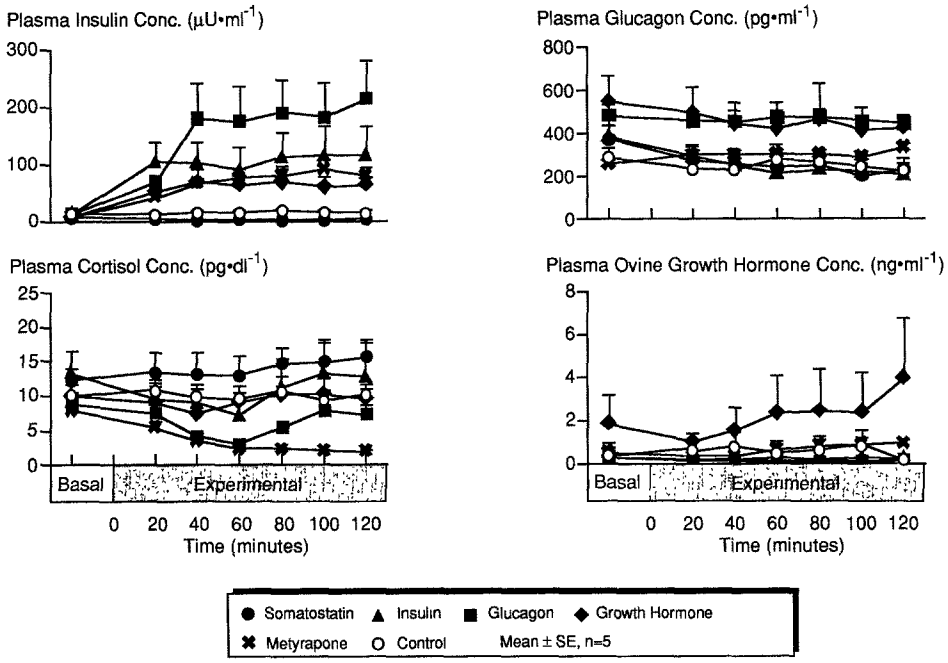

Fig 2. Plasma insulin, glucagon, cortisol, and growth hormone concentrations over time from the means of the basal period through the 2-hour period of administration of the various infusions by group (experimental).

hormone, and metyrapone groups were not significantly different.

Figure 1 also depicts the mean glucose utilization rate (Rd) over time through the 2-hour period of administration by group. There were no significant differences between the Rd of the groups during the basal period. Over time, there was a significant increase of the Rd of the SRIF group compared with the control group ($P < .0045$) and a significant decrease of the Rd of the hyperinsulinemia ($P < .0002$) and growth hormone ($P < .0004$) groups compared with the control group. The Rds of hyperinsulinemia, glucagon, growth hormone, and metyrapone groups were not significantly different.

Figure 2 depicts the plasma insulin concentration over time through the 2-hour period of administration by group. There were no physiological differences between the mean plasma insulin concentrations of the groups, which had a range of 8 ± 1 to 13 ± 4 μU/mL during the basal period. Over time, there was a significant decrease of plasma insulin in the SRIF group compared with the control group ($P < .0001$). There was a significant increase in plasma insulin in the glucagon, growth hormone, and metyrapone groups compared with the control group ($P < .0011$ or less). Plasma insulin concentrations of the hyperinsulinemia, growth hormone, and metyrapone groups were not significantly different.

Figure 2 depicts the plasma glucagon concentration over time through the 2-hour period of administration by group. There were no physiological differences between the mean plasma glucagon concentrations of the groups, with a range of 262 ± 19 to 550 ± 119 pg/mL during the basal period. Over time, there was a significant increase in plasma glucagon in the hyperinsulinemia, growth hormone, and metyrapone groups compared with the control group ($P < .0023$ or less). The plasma glucagon concentrations of SRIF, glucagon, and control groups were not significantly different.

Figure 2 depicts the plasma cortisol concentration over time through the 2-hour period of administration by group. There were no significant differences between the mean plasma cortisol concentrations of the groups during the basal period. Over time, there was a significant decrease in plasma cortisol in the metyrapone group compared with the SRIF, hyperinsulinemia, glucagon, growth hormone, and control groups ($P < .0029$ or less). There were no differences in plasma cortisol concentrations of the SRIF, hyperinsulinemia, glucagon, growth hormone, and control groups.

Figure 2 depicts the plasma growth hormone concentration over time through the 2-hour period of administration by group. There were no significant differences between the mean plasma growth hormone concentrations of the groups during the basal period or throughout the period of the various infusions.

Figure 3 depicts the heart rate over time through the 2-hour period of administration by group. There were no significant differences between the mean heart rate of the groups during the basal period. Over time, the heart rate of the SRIF and hyperinsulinemia groups was significantly decreased compared with the control group ($P < .0048$ or less). The mean heart rates of the hyperinsulinemia, glucagon, growth hormone, and metyrapone groups were not significantly different.

Figure 3 depicts the systolic blood pressure over time through the 2-hour period of administration by group. There were no significant differences between the systolic blood pressure of the groups during the basal period. Over time, the systolic blood pressure of the glucagon group increased compared with the SRIF, hyperinsulinemia, metyrapone, and control groups ($P < .0058$ or less).

Figure 3 depicts the diastolic blood pressure over time through the 2-hour period of administration by group. There were no significant differences between the mean diastolic blood pressure of the groups during the basal period or throughout the period of the various infusions.

Figure 4 depicts the pH over time through the 2-hour period

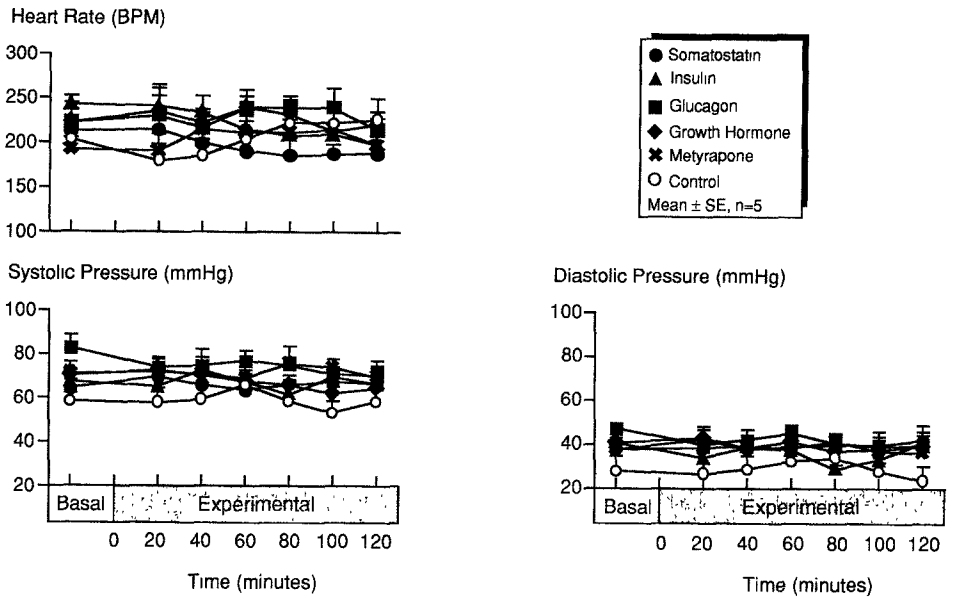

Fig 3. Heart rate (per minute), systolic blood pressure, and diastolic blood pressure over time from the means of the basal period through the 2-hour period of administration of the various infusions by group (experimental).

of administration by group. There were no significant differences between the mean pH of the groups during the basal period or throughout the period of the various infusions.

Figure 4 depicts the pO_2 over time through the 2-hour period of administration by group. There were no significant differences between the mean pO_2 of the groups during the basal period or throughout the period of the various infusions.

Figure 4 depicts the pCO_2 over time through the 2-hour period of administration by group. There were no significant differences between the mean pCO_2 of the groups during the basal period. Over time, the pCO_2 of the cortisol group was significantly decreased in comparison to the control group ($P < .0018$).

DISCUSSION

Insulin counterregulatory hormones play a major role in the maintenance of glucose homeostasis.[12,13] Our hypothesis was that there would be a parallel imprecise response to the various insulin counterregulatory hormones to a degree comparable to that previously reported for insulin itself. We chose to isolate the effect(s) of specific primary and secondary insulin counterregulatory hormones during hyperinsulinemic hypoglycemia, which has not been studied in the neonatal period heretofore. We used the following: SRIF, a known inhibitor of insulin, glucagon, and growth hormone; and metyrapone, a known inhibitor of cortisol. This series of inhibitors were infused collectively with and without simultaneous administration of specific hormone replacement to isolate their response(s) as a component of the ontogeny of neonatal glucose homeostasis.

The lamb has been used to study glucose kinetics in the neonatal period.[6-9,11] It was fasted for a period of 12 hours. This length of time resulted in a fasted state because the lamb does not become a ruminant until it is weaned.

The general stability of this model is confirmed by the

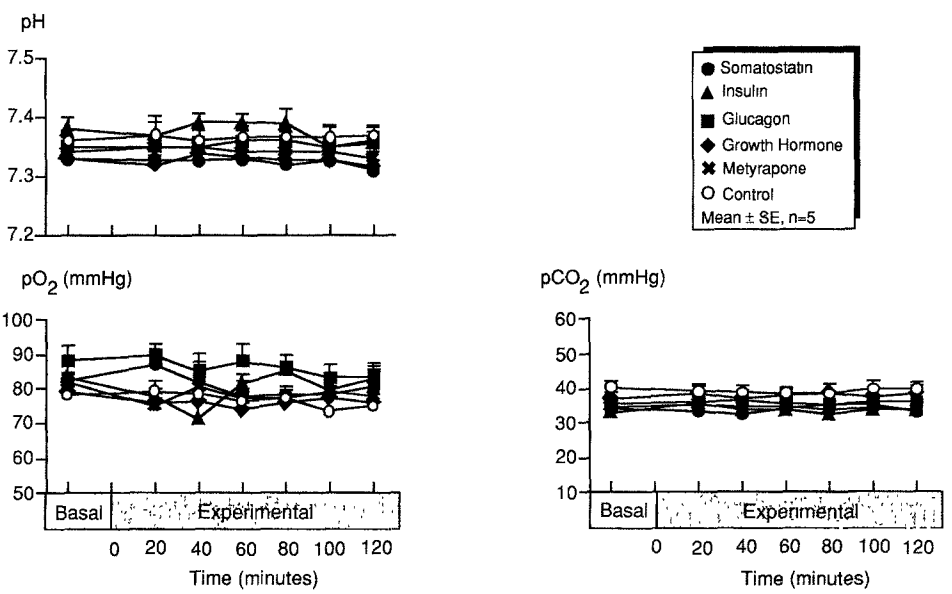

Fig 4. pH, pO_2, and pCO_2 over time from the means of the basal period through the 2-hour period of administration of the various infusions by group (experimental).

general uniformity of the data relative to the plasma glucose concentration, concentration of the specific hormones, heart rate, systolic and diastolic blood pressure, and blood gases throughout of the control group when only the isotope was infused in 0.9% saline. We recognize that our peripherally placed catheters (the internal carotid artery was used for sampling) did not allow for direct measurement of substrate or hormone concentrations secreted from the pancreas and/or liver. However, since the hormones are well known to exert specific predictable effects on the rate of glucose turnover (production), the measurement of Rp has been used as the marker for the differential effect of these hormones on neonatal glucose kinetics, as studied here.[6-9,11]

Four previous studies have used SRIF administration to evaluate glucose homeostasis in the newborn. Hetenyi et al[10] studied the newborn puppy, and Sperling et al,[15] Cowett and Tenenbaum,[9] and Cowett et al[11] studied the newborn lamb. Hetenyi et al[10] failed to show a decrease in plasma glucagon but noted a decrease in plasma insulin comparable to when the puppy was infused with saline alone. Sperling et al[15] noted a decrease in plasma glucagon but a variable response in plasma insulin. Cowett and Tenenbaum[9] and Cowett et al[11] noted a decrease in plasma insulin but variable changes in plasma glucagon. The reasons for these differences are not apparent. Hetenyi et al[10] suggested that in the newborn puppy, α and β cells are less responsive to SRIF compared with the adult. The degree to which SRIF depressed plasma insulin concentrations in the studies reported in this data set was similar to the degree shown by our laboratory previously, as well as by others, in the adult.[16]

During kinetic studies using stable and/or radioactive isotope methodology, glycogenolysis and gluconeogenesis may collectively contribute to the rate of glucose appearance (production). Hormones and substrates that are known to play a role in suppressing endogenous glucose production include insulin and glucose, respectively.[2,17-20]

In the newborn, the apparent inefficiency of glucose homeostasis from a hormonal standpoint may be due to (1) decreased hepatic or peripheral response to insulin or to the insulin counterregulatory hormones, glucagon, catecholamines, steroids, and/or growth hormone, or (2) decreased insulin secretion in response to glucose infusion. Originally, an indirect technique of stepwise incremental glucose infusion was used in infants to infer the rate of basal glucose output compared with adults.[21] The inference was predicated on the assumption that the newborn was as responsive as the adult. However, Varma et al[22] initially showed that a 2- to 3-hour infusion of glucose did not produce a steady-state plasma glucose concentration in the newborn dog as it did in the adult dog. They suggested that the neonatal puppy lacked effective regulation to handle excess glucose, resulting in an ineffective decrease in hepatic glucose turnover. They postulated that there was a decreased hepatic and peripheral response to insulin. These conclusions have largely been substantiated by studies in the human newborn and in the newborn lamb from this laboratory.[2,8,9,11]

The utilization of the hyperinsulinemic-euglycemic clamp technique in the human neonate has recently been reported.[23] Persistent glucose production was apparent during a wide range of insulin infusion rates. Endogenous glucose production was sensitive to a low insulin concentration, reached a plateau quickly, and then became nonresponsive to higher insulin concentrations. Endogenous glucose production was reduced by about 50% at a plasma insulin concentration up to 89 mU/mL. In another series, this parallel response existed in the term and preterm neonate early in the neonatal period and in the preterm neonate at the end of the neonatal period.[24] Thus, we have concluded that insulin only partially suppresses the endogenous glucose production rate.

In this lamb model, infusion of SRIF resulted in an elevation of blood glucose and an increase in the rate of glucose production. As we have noted in prior studies using SRIF, there was a concomitant decrease in plasma insulin and no significant decrease in plasma glucagon compared with controls. It is logical to interpret these data as indicating that glucagon continued to have a stimulatory effect on the glucose production rate without the inhibitory presence of insulin. In fact, these data are again compatible with data reported by Felig et al[25] and Sherwin et al,[26] who infused SRIF both in adult humans and in an adult dog model for a relatively prolonged period of 5 to 8 hours. They performed their studies to determine if the hypoglycemia observed with short infusions of SRIF would continue to be present following a prolonged infusion. While they initially noted the presence of hypoglycemia following a relatively short SRIF infusion, hyperglycemia occurred during a more prolonged infusion despite persistent hypoglucagonemia. They suggested that insulin deficiency rather than glucagon availability was of primary importance in the control of glucose homeostasis, since glucagon appeared to have an evanescent effect on hepatic glucose production and insulin predominated in the long-term (relative) control of glucose homeostasis.

In contrast to the conclusions of Felig et al[25] and Sherwin et al,[16,26] Cherrington et al[27] have suggested that even after a significant fast, glucagon is responsible for at least one third of basal glucose production and basal insulin acts to prevent the increase in the rate of glucose production that would follow from the unrestrained effect of glucagon. They noted, using an adult dog model, that administration of SRIF resulted in diminution of both insulin and glucagon. This resulted in a rapid and significant decline in plasma glucose and the rate of glucose production. They suggested that glucagon played a significant role in maintaining hepatic glucose production in the overnight-fasted adult dog and that insulin acts as a potent antagonist of glucagon. They concluded that when glucagon is deficient in the adult, hepatic glucose production is responsive to the presence of insulin, which would lead to the decreased glucose production rate and plasma glucose concentration noted.

The relatively elevated and variable baseline levels of plasma glucagon have also been noted in our previous studies in the newborn.[9,11] However, the concentrations are in the same range noted by others studying different species of the newborn.[10,28] Hetenyi et al[10] have postulated that the relatively elevated plasma glucagon concentration in the newborn may partially be secondary to a large and readily metabolizable store of biologically active glucagon. It has not been determined whether elevated levels represent "true" glucagon, glucagon precursor

molecules, and/or glucagon-like immunoactivity in the newborn with this methodology.

In this series, hyperinsulinemic hypoglycemia was produced following administration of 2 $mU \cdot kg^{-1} \cdot min^{-1}$ insulin. As would be expected, plasma glucose declined, as did the endogenous glucose production rate. Both of these data are in keeping with the known metabolic effects of insulin.[2,9,18,20,23,24,28,29] Following administration of insulin, there were significant increases of the plasma insulin concentration in the glucagon, growth hormone, and metyrapone groups compared with the control group. Plasma insulin concentrations of the hyperinsulinemia, growth hormone, and metyrapone groups were not significantly different. This suggests that hyperinsulinemia generally resulted from the 2-$mU \cdot kg^{-1} \cdot min^{-1}$ insulin infusion during concomitant infusion of SRIF.

Following administration of glucagon during hyperinsulinemia, a persistent decline of plasma glucose was apparent without a significant difference in the endogenous glucose production rate compared with the hyperinsulinemia group. This suggests that glucagon was not able to functionally counteract the effects of hyperinsulinemia. Likewise, during growth hormone administration, which did not evidence a significant increase in the concentration due to the wide standard error within the group, there was a significant decrease in plasma glucose and the endogenous glucose production rate. This is interpreted to indicate that growth hormone, as well, was not able to functionally counteract the effects of hyperinsulinemia.

Another significant set of data included the effect of administration of metyrapone, which is known to block the release of cortisol. The plasma cortisol concentration of the metyrapone group was decreased in comparison to all other groups, and the plasma glucose concentration and endogenous glucose production rate were not different from those of the hyperinsulinemia group. The absence of cortisol following metyrapone administration did not accentuate the changes observed in the hyperinsulinemic hypoglycemic group in which cortisol was present. This is interpreted to indicate that cortisol is probably a secondary counterregulatory hormone that plays a minor role in the neonatal response to hyperinsulinemic hypoglycemia. Parallel conclusions could be inferred for growth hormone as well.

The decrease in plasma glucose and the lack of change in endogenous glucose production in the glucagon-infused hyperinsulinemic group is interpreted to indicate that insulin has a greater effect on glucose uptake than on glucose production and/or glucagon does not appreciably influence glucose production during hyperinsulinemic hypoglycemia in the neonatal period.

In summary, SRIF and other known inhibitors of insulin counterregulatory hormones have been infused in combination with specific hormonal replacement to isolate the various insulin counterregulatory hormones during hyperinsulinemic hypoglycemia. It is apparent from the data generated in this neonatal lamb model that there is an imprecise response of insulin counterregulatory hormones during hypoglycemic hyperinsulinemia. These data contribute, for the first time, the concept that this imprecise response mirrors the generally accepted imprecise control by insulin of glucose production as one of the components of the ontogeny of neonatal glucose homeostasis.

ACKNOWLEDGMENT

We appreciate the expert statistical support provided by Joseph Hogan, ScD, Assistant Professor of Statistics, Department of Community Medicine, Brown University. Patricia Knight provided expert secretarial assistance.

REFERENCES

1. Cowett RM: Neonatal glucose metabolism, in Cowett RM (ed): Principles of Perinatal-Neonatal Metabolism (ed 2). New York, NY, Springer-Verlag, 1998, pp 683-722

2. Cowett RM, Oh W, Schwartz R: Persistent glucose production during glucose infusion in the neonates. J Clin Invest 71:467-475, 1983

3. Hertz DE, Karn CA, Liu YM, et al: Intravenous glucose suppresses glucose production but not proteolysis in extremely premature newborns. J Clin Invest 92:1752-1758, 1993

4. Kalhan SC, Oliven A, King KC, et al: Role of glucose in the regulation of endogenous glucose production in the human newborn Pediatr Res 20:49-52, 1986

5. van Goudoever JB, Sulkers EJ, Chapman TE, et al: Glucose kinetics and glucoregulatory hormone levels in ventilated preterm infants on the first day of life. Pediatr Res 33:583-589, 1993

6. Cowett RM: α-Adrenergic agonists stimulate neonatal glucose production less than β-adrenergic agonists in the lamb. Metabolism 37:831-836, 1988

7. Cowett RM: Decreased response to catecholamines in the newborn. Effect on glucose kinetics in the lamb. Metabolism 37 736-740, 1988

8. Cowett RM, Susa JB, Oh W, et al: Endogenous glucose production during constant glucose infusion in the newborn lamb. Pediatr Res 12:853-857, 1978

9. Cowett RM, Tenenbaum D: Hepatic response to insulin in control of glucose kinetics in the neonatal lamb. Metabolism 36:1021-1026, 1987

10. Hetenyi G Jr, Kovacevic N, Hall SEH, et al: Plasma glucagon in pups, decreased by fasting, unaffected by somatostatin or hypoglycemia. Am J Physiol 231:1377-1382, 1976

11. Cowett RM, Rapoza RE, Gelardi NL: The contribution of glucose to neonatal glucose homeostasis in the lamb. Metabolism 47:1239-1244, 1998

12. Cryer PE: Glucose counterregulation: The prevention and correction of hypoglycemia in humans. Am J Physiol 264:E149-E155, 1993

13. Gerich JE, Cryer PE: Counterregulatory hormones: Biochemical and physiological aspects, in Cowett RM (ed): Principles of Perinatal-Neonatal Metabolism (ed 2). New York, NY, Springer-Verlag, 1998, pp 155-182

14 Steele RS, Wall RC, DeBodo RC, et al: Measurement of size and turnover rate of body glucose pool by the isotope dilution method. Am J Physiol 187:215-224, 1956

15. Sperling MA, Grajwer L, Leake RD, et al: Effects of somatostatin (SRIF) infusion on glucose homeostasis in newborn lambs: Evidence for a significant role of glucagon. Pediatr Res 11:962-967, 1977

16. Sherwin RS, Hendler R, DeFronzo R, et al: Glucose homeostasis during prolonged suppression of glucagon and insulin secretion by somatostatin. Proc Natl Acad Sci USA 74:348-352, 1977

17. Bolli G, De Feo G, Perriello S, et al: Role of hepatic autoregulation in defense against hypoglycemia in humans. J Clin Invest 75:1623-1631, 1985

18. Hulman SE, Kliegman RM: Assessment of insulin resistance in newborn beagles with the euglycemic hyperinsulinemic clamp. Pediatr Res 24:219-223, 1989

19. Rizza RA, Mandarino LJ, Gerich JE: Dose response characteristics for effects of insulin on production and utilization of glucose in man. Am J Physiol 240:E630-E639, 1981

20. Wolfe R, Allsop JR, Burke JF: Glucose metabolism in man: Responses to intravenous glucose infusion. Metabolism 28:210-220, 1979

21. Adam PAJ, King KC, Schwartz R: Model for the investigation of intractable hypoglycemia: Insulin-glucose interrelationships during steady state infusions. Pediatrics 41:91-105, 1968

22. Varma S, Nickerson H, Cowan JS, et al: Homeostatic responses to glucose loading in newborn and young dogs. Metabolism 22:1367-1375, 1973

23. Farrag HM, Nawrath LM, Healey JE, et al: Persistent glucose production and greater peripheral sensitivity to insulin in the neonate vs the adult. Am J Physiol 272:E86-E93, 1997

24. Farrag HM, Dorcus EJ, Cowett RM: Maturation of the glucose utilization response to insulin occurs before that of glucose production in the preterm neonate. Pediatr Res 39:308A, 1996 (abstr)

25. Felig P, Wahren J, Sherwin R, et al: Insulin, glucagon, and somatostatin in normal physiology and diabetes mellitus. Diabetes 25:1091-1099, 1976

26. Sherwin RS, Fisher M, Hendler R, et al: Hyperglucagonemia and blood glucose regulation in normal and obese diabetic subjects. N Engl J Med 294:455-461, 1976

27. Cherrington AD, Chaisson JL, Liljenquist JE, et al: The role of insulin and glucagon in the regulation of basal glucose production in the post absorptive dog. J Clin Invest 58:1407-1418, 1976

28. Girard JR, Ceundet GS, Marliss EB, et al: Fuels, hormones and liver metabolism at term and during the early postnatal period in the rat. J Clin Invest 52:3190-3200, 1973

29. Gruppuso P: Insulin: Biochemical and physiological aspects, in Cowett RM (ed): Principles of Perinatal-Neonatal Metabolism (ed 2). New York, NY, Springer-Verlag, 1998, pp 135-154