Effect of Priming of Amino Acids on Insulin and Growth Hormone Response in the Premature Infant

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SUMMARY

Glucose, α-amino nitrogen, serum insulin, and HGH were studied in preterm infants during the first 24 hours of life.

Glucose infusion (1.25 gm.) resulted in a slight elevation of serum insulin. When this amount of glucose was infused during the last 30 minutes of a two-and-a-half-hour infusion of a mixture of essential amino acids, there was a rapid and striking increase in serum insulin. However, this increase was not associated with a faster glucose disposal rate. The administration of this mixture of amino acids doubled the basal level of α-amino nitrogen, and during the first two hours, before glucose infusion, it caused a significant rise of serum insulin and HGH. In both cases glucose caused an increase of HGH secretion that was not significantly different in the two groups of infants. DIABETES 27:334-37, March, 1978.

Insulin is present in the fetal pancreas as early as the 10th to 12th gestational week and increases progressively up to birth.[1,2] Morphologic studies show that the beta cells possess typical storage granules and cell organelles,[3,4] but it has been demonstrated that the fetal[5] and neonatal[6] pancreas responds only slightly to an increase of serum glucose concentration. On the other hand, recent studies showed that in preterm neonates the unresponsiveness to glucose can be reversed.[7] When glucose is injected into them acutely it is a poor stimulant of insulin secretion, but if a small infusion of glucose precedes the acute injection of glucose, the latter becomes a potent stimulus of insulin secretion.

In line with this study, our paper demonstrates that in the preterm infant the insulin response to glucose infusion can also be enhanced by priming with a mixture of essential amino acids. The secretion of HGH was also evaluated.

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MATERIAL AND METHODS

Fifteen vaginally delivered preterm infants were included in our study. Prematurity was assessed on the basis of birth weight for gestational age and maternal dates as well as clinical appearance.[8,9] The mean gestational age was 33.2 ± 0.34 (± S.E.M.) weeks. Their birth weights ranged from 1,550 to 2,550 gm. At times varying from one to 24 hours after birth, two scalp vein needle sets were inserted into the peripheral vein to withdraw blood samples and administer infusions.

Two groups of infants received an intravenous infusion of 1.25 gm. of glucose over a 30-minute period. In the first group of seven infants a total of 1.20 gm. of a mixture of nine essential amino acids was administered over a two-and-a-half-hour period at the following rate: 10 mg. per minute for the first 90 minutes and 5 mg. per minute for the following 60 minutes. During the last 30 minutes of this infusion, 1.25 gm. of glucose was administered simultaneously with the amino acids.

The control infants, a group of eight, received a two-and-a-half-hour infusion of physiologic saline both before and during the 30-minute infusion of 1.25 gm. of glucose.

The mean age at testing was 14 ± 2.8 hours (range five to 24 hours) in the group preinfused with amino acids and 12.3 ± 3 hours (range, one to 20 hours) in the control group. The nature and the aim of the investigation were explained to the parents, and it was carried out with their consent.

The composition of the mixture of nine essential

amino acids is described elsewhere.[10] Glucose was measured by a glucose-oxidase method. Serum insulin and HGH were determined in triplicate by radioimmunoassay methods.[11,12] Concentrations of plasma α-amino nitrogen were determined by the technique of Matthews et al.[13]

RESULTS

Effect of Priming with a Mixture of Amino Acids on a 1.25-gm. Infusion of Glucose (Figure 1)

During the first two-hour part of priming with the mixture of amino acids in the seven infants with body weight of 2,157 ± 114 gm., the blood sugar level rose slightly from 50.5 ± 6.8 mg. to 61.5 ± 7.5 mg. per 100 ml., while the serum insulin, the α-amino nitrogen, which was measured in only five infants, and serum HGH rose significantly ($0.02>p>0.01$). Their respective values were: serum insulin rose from 13.2 ± 2.7 μU. per ml. to 25.7 ± 3.5 μU. per ml., the α-amino nitrogen increased from 3.9 ± 0.3 mg. to 6.9 ± 0.9 mg. per 100 ml., while the serum HGH increased from 19.5 ± 7 ng. per ml. to 46.5 ± 6.8 ng. per ml. When 1.25 gm. of glucose was added to the amino acid mixture during the last half hour of priming, the blood glucose went up acutely, to 200.8 ± 18 mg. per 100 ml. at 30 minutes and then fell to 116.1 ± 13.7 mg. and 65 ± 7.3 mg. per 100 ml. at 60 and 120 minutes, respectively. Serum insulin rose to 107.5 ± 21 μU. per ml. at 30 minutes and then decreased to 64.8 ± 17.6 and 21 ± 2 μU. per ml. at 60 and 120 minutes, respectively. α-Amino nitrogen was still 7.02 ± 1.4 mg. per 100 ml. at 30 minutes then decreased to 5.7 ± 1.3 mg. and 4.7 ± 1.1 mg. per 100 ml. Serum HGH increased gradually to 55.2 ± 16.4 ng. per ml. and 99 ± 20.1 ng. per ml. at 30 and 60 minutes, respectively, and then decreased to 45 ± 10.4 ng. per ml. at 120 minutes. Maximal increase of serum insulin occurred at 30 minutes in six subjects and at 60 minutes in one subject, while maximum increase of serum HGH occurred at 60 minutes in all subjects.

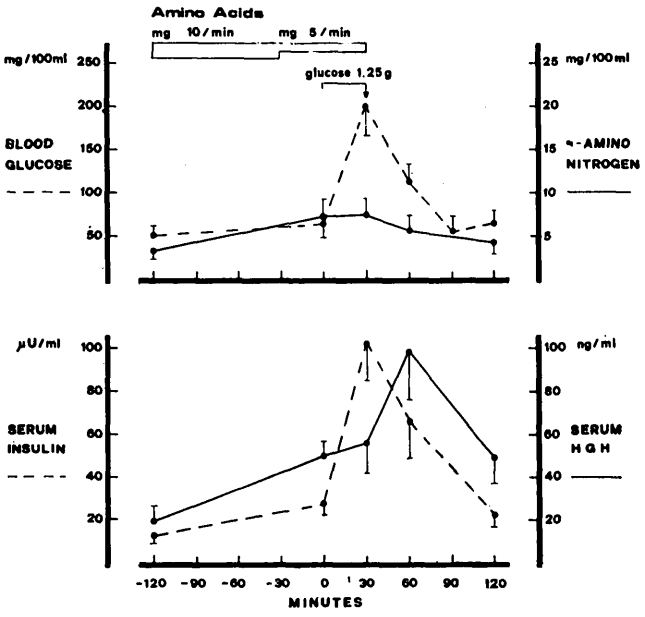

FIG. 1. Effect of priming with a mixture of amino acids on blood glucose, plasma α-amino nitrogen, serum insulin, and HGH in the preterm infant.

Effect of Priming with Saline on a 1.25-gm. Infusion of Glucose (Figure 2)

During the first two-hour part of priming with saline in eight infants with body weight of 2,197 ± 131 gm., the blood sugar, serum insulin, and HGH remained almost unchanged. When 1.25 gm. of glucose was added to the saline during the last half hour of priming, the blood glucose went up acutely from 62 ± 3.2 mg. at 0 minutes to 170.6 ± 8.7 mg. per 100 ml. at 30 minutes and then fell to 11.3 ± 6.5 mg., 83.8 ± 7.7 mg., and 82.6 ± 2.2 mg. per 100 ml. at 60, 90, and 120 minutes, respectively. However, the serum insulin went up from 10.8 ± 2.4 μU. per ml. at 0 minutes to only 18 ± 3.9 and 25.3 ± 6.5 μU. per ml. at 30 and 60 minutes, respectively. It was still 25.2 ± 5.8 μU. per ml. at 90 minutes but then decreased slightly, to 15.6 ± 2.4 μU. per ml. at 120 min. Serum HGH level rose from the basal level of 26.3 ± 5.2 at 0 minutes to 72.2 ± 18.2 ng. per ml. at 60 minutes. It remained 74.6 ± 15.4 ng. per ml. at 90 minutes, then decreased slightly, to 58.6 ± 12.5 ng. per ml. at 120 minutes.

DISCUSSION

Our data demonstrate that glucose primed with saline causes a small increase in serum insulin. However, if this same amount of glucose is infused during the last 30 minutes of a two-and-a-half-hour infusion of a mixture of essential amino acids it becomes a potent stimulus of insulin secretion. The administration of this mixture doubled the basal level of α-amino nitrogen, and during the first two hours, before the glucose infusion, it caused a significant rise of serum insulin and HGH. In both cases glucose provoked a marked rise in HGH secretion.

The biochemical explanation for the slight and delayed insulin response to glucose during the fetal and

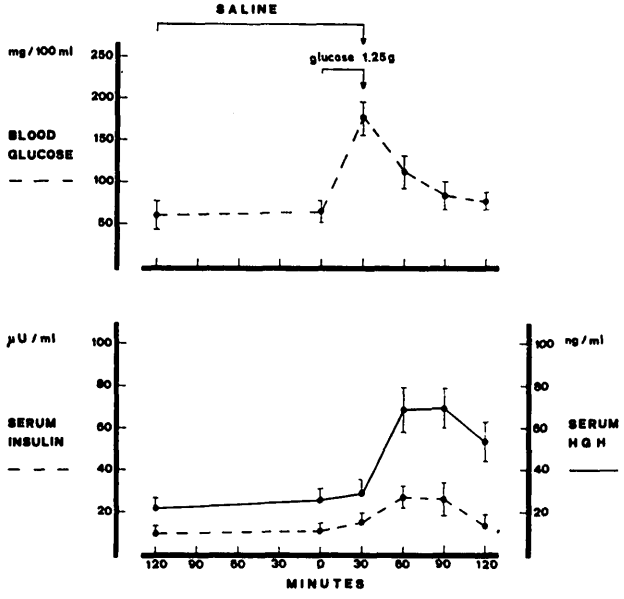

FIG. 2. Effect of priming with saline on blood glucose, serum insulin, and HGH in the preterm infant.

neonatal period is speculative, and several hypotheses have been put forward. It has been suggested that the low effectiveness of glucose in fetal and neonatal pancreas could be due to an inadequate intracellular accumulation of cyclic AMP within the beta cell.[14,15] In fact, in the preterm infant, theophylline and glucagon, agents known to increase cyclic AMP in the beta cells, stimulate insulin release.[16] In vitro, with fetal pancreatic explants, glucose becomes a potent stimulant in the presence of methylxanthines or of glucagon.[17]

On the other hand, it has been shown recently that D-glyceraldehyde stimulates insulin release in both the adult and newborn rat.[18] This effect is probably related to its metabolism in the beta cell. D-glyceraldehyde enters the glycolytic pathway at the triose phosphate step and would therefore stimulate the insulin secretion independently of the glucose phosphorylation of the beta cell. It has been suggested that the poor insulin response to glucose in the neonate would involve factors located above the triose phosphate step.

However, we have observed that a mixture of essential amino acids stimulates insulin release markedly during the neonatal period of the preterm infant.[6,10] The mechanism by which these substances stimulate the beta cell to release insulin remains unknown. Evidence has been presented that amino acids themselves, rather than their metabolites, are responsible for this action[19] and that different stimulatory mechanisms may exist for different amino acids.[20] In vivo in the dog[19] and in vitro with fetal rat pancreas,[21] nonmetabolized amino acid analogues stimulate insulin release.

In the present study we are also in doubt as to the mechanism(s) of the priming action of these substances on insulin secretion following glucose infusion. Several factors alone or in combination could contribute to these results. One possible factor may be that in the present study during the administration of the mixture of amino acids before the glucose infusion, there is a slight increase of blood glucose. We have previously shown that if an infusion of glucose administered for 120 minutes precedes the acute injection of glucose, the latter becomes a potent stimulus of insulin secretion.[7] However, in the same study the priming with 2.5 gm. of glucose and the successive rapid infusion of glucose (2.5 gm.) produced higher blood glucose levels, but the increase of serum insulin was identical. A second factor may be that glucose was infused during the last 30 minutes of amino acid priming, and the α-amino nitrogen remained significantly higher than the basal level during the glucose administration. Glucose and amino acids are synergistic in their insulin stimulatory action,[22] which could explain our findings. Contrary to the results obtained in this study, the blood glucose levels in the adult were lower during the infusion of the glucose-amino acid mixture than when glucose was infused alone.

In both full-term and preterm infants the disposal rate of glucose from the blood is much slower than that in older infants and seems to be influenced by other factors than merely changes in insulin levels. In our study the blood glucose curves in both groups of infants resemble each other closely in spite of the great difference in the insulin curves. Gentz et al.[23] showed that low-birth-weight infants cleared intravenously administered glucose at the same rate whether they showed an insulin response or not. Recently Zarif et al.,[24] in preterm infants given intravenous or oral glucose, showed that neither inadequate insulin nor elevated levels of HGH could be incriminated as significant etiologic factors in the glucose intolerance seen in such infants.

The human pituitary gland secretes HGH early in the fetal stage, which is immunologically and physicochemically similar to that in the child and adult.[25] Little is known about the secretion of this hormone in utero. It has been reported that, at term, maternal glucose infusion either does not influence fetal HGH secretion or, if so, causes only a slight decrease.[26,27] At birth the serum HGH is high, vary-

ing from subject to subject, and declines to lower levels after the first week of life.[28] In our study the intravenous injection of glucose in both groups of infants stimulated HGH secretion, in contrast to secretion in the fetus at term, older children, and adults. Similar data regarding full-term and preterm infants have been reported in the literature.[28,29] This paradoxic increase of HGH in response to hyperglycemia reverts to the adult mode on the sixth day of life[30] and appears to depend on hypothalamic neuroendocrine secretion as well as on an intact pituitary gland.[31] In fact, anencephalic infants without identifiable hypothalamic tissue do not respond at all.

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