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Metabolic effects of septicaemia in newborn and young infants with particular reference to the plasma free amino acids

by

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Blood samples were obtained from 13 infants with clinically and microbiologically confirmed septicaemia. Two subgroups of the septic infants were formed according to postnatal age, an early onset group consisting of 6 infants younger than 7 days, and a late onset group comprising 7 infants older than 7 days. The controls, divided into similar age groups, consisted of 20 infants. The blood samples were analysed for 18 plasma amino acids and glucose, lactate and FFA. Except the significantly lowered blood glucose in the early onset group of infants, no appreciable change occurred in the glucose, lactate and FFA levels. The plasma aminogram was different in the two groups, as hypoaminoacidaemia was more pronounced in the early onset group. The branched chain amino acids decreased significantly in both groups. The three important glucogenic amino acids alanine, glycine and proline showed a decrease only in the early onset group. Among the calculated plasma amino acid ratios the phenylalanine: tyrosine quotient was markedly increased. This, in addition to the response of the branched chain amino acids, has been found characteristic of a variety of clinical and experimental infections.

Septicaemia in the neonatal and postnatal period is a major cause of morbidity and mortality. It is therefore of importance to investigate the metabolic responses to severe infection in relation to the characteristics of energy and nutrient metabolism in this period of extrauterine life. The present study was undertaken to examine the plasma nutrient concentrations in neonatal septicaemia with particular emphasis on the changes of the plasma aminogram. In view of the observations made in infected adults [4, 5, 19, 22, 23], it is of interest whether similar quantitative changes occur in the plasma amino acid pattern in response to severe bacterial infection in the newborn infant. Increased flux of amino acids from the muscle, stimulated hepatic gluconeogenesis, increased anabolic and catabolic utilization of amino acids associated with infection [13, 19, 20, 21, 23] are, in fact, expected to influence the plasma total and individual amino acid level.

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Material and Methods

The data presented in this paper were collected from 13 formula fed septicaemic infants admitted to the intensive care unit. Two subgroups were formed according to postnatal age: an early onset group comprising 6 infants younger than 7 days and a late onset group comprising 7 infants older than 7 days when they had fallen ill. The controls, divided into similar age groups, consisted of 20 infants admitted for examination because of minor congenital anomalies or various non-infectious and non-metabolic disturbances, in whom blood sample was taken for their routine management. The means and ranges of gestational age, postnatal age, birth weight as well as the age at which the blood samples were drawn are shown in Table I.

The relevant clinical, microbiological and autopsy findings of the two groups of septic infants are demonstrated in Table II. The diagnosis of septicaemia was based on well-known alarming symptoms, like abdominal distension, unexplained unconjugated hyperbilirubinaemia, fever and hepatomegaly together with positive blood cultures. Two infants showed signs of peripheral circulatory failure at the time of blood sampling. A positive blood culture in association with clinical symptoms was regarded as an important direct proof of septicaemia. Four infants of the early onset, and three of the late onset group died indicating that severely ill infants were studied. It should be emphasized that the early onset group was additionally burdened with the occurrence of prematurity which, in all probability, contributed to the high mortality.

Blood was taken from a peripheral vein in the postabsorptive state soon after the clinical symptoms of septicaemia had become manifest and before infusion was started in the septic infants, and before the first morning feeding in the control infants.

The blood was placed in tubes containing heparin. Glucose and lactate concentration was determined by the method described by Price [17] and Huckabee [11], respectively. The remainder of the blood sample was centrifuged for analysis of FFA [12]. The protein-free supernatant corresponding to 0.5 ml of plasma was immediately frozen and stored at −20 °C until assayed. Amino acid determination was performed by an automatic Beckman Multichrome Liquid Column Chromatograph, and norleucine was used as the internal reference standard.

For statistical analysis, the means and standard errors were calculated. Significance was estimated by Student's t test.

Table I

Means and ranges of gestational age, postnatal age, birth weight as well as the age at which the blood samples were drawn

	Early onset group (postnatal age: 1—6 days)		Late onset group (postnatal age: 7—42 days)	
	control	septicaemia	control	septicaemia
Gestational age (wk)	32.6 (29—38)	33.6 (30—37)	36.3 (30—41)	36.2 (32—41)
Birth weight (g)	1835 (1550—2780)	1876 (1070—2600)	2557 (1570—3610)	2810 (1470—3600)
Postnatal age at the clinical manifestation of septicaemia and withdrawal of blood (day)	2.1 (1—5)	2.3 (1—5)	18.9 (7—30)	19.7 (7—42)
Number of infants	10	6	10	7

TABLE II

Relevant clinical and microbiological findings in the two groups of septic infants

No	Gestational age/wk	Birthweight (g)	Onset of symptoms (day)	Clinical symtoms	Blood culture	Clinical diagnosis	Outcome
1	37	2600	2	jaundice, hepatomegaly	E. coli	Septicaemia. DIC.*	D +
2	—	1750	1	jaundice, abdominal distension	Clostridium p.	Necrotizing enterocolitis Peritonitis.	D
3	33	1500	2	jaundice, abdominal distension	Staphylococcus aur. haem.	Septicaemia. Pneumonia.	D
4	30	1070	2	cyanosis, abdominal distension	Pseudomonas p.	Septicaemia. Peritonitis.	D
5	33	2460	5	hepato-splenomegaly, abdominal distension	E. coli	Septicaemia. Pyelonephritis.	S ++
6	35	1900	2	jaundice, hepatosplenomegaly	Enterococcus	Septicaemia. Pyelonephritis.	S
7	37	3600	8	abdominal distension, fever, hepatomegaly	Pseudomonas p.	Septicaemia.	D
8	32	1470	32	jaundice, abdominal distension	Pseudomonas p.	Septicaemia. Pyelonephritis.	D
9	38	3450	42	convulsions, petechiae	Pseudomonas p.	Purulent meningitis.	D
10	33	2240	17	jaundice, pyuria	E. coli	Septicaemia. Pyelonephritis.	S
11	41	3160	21	jaundice, hepatomegaly	E. coli	Septicaemia. Pyelonephritis.	S
12	32	2150	11	abdominal distension, hepatomegaly	Staphylococcus aur. haem.	Septicaemia. Pneumonia. Endotoxic shock.	S
13	41	3600	7	jaundice, fever, hepatomegaly	Proteus	Septicaemia. Endotoxic shock.	S

* Disseminated intravascular coagulopathy. + D = died, ++ S = survived

TABLE III

Means±SE of blood glucose, lactate and plasma free fatty acids in the control and septic infants

	Early onset group		Late onset group	
	control (n = 10)	septicaemia (n = 6)	control (n = 10)	septicaemia (n = 7)
Glucose, mmol/l	3.4±0.3	1.8±0.4*	3.6±0.2	3.4±0.4
Lactate, mmol/l	2.4±0.2	2.8±0.8	1.4±0.1	2.3±0.5
FFA, μE/l	751±156	573±193	562±182	715±148

TABLE IV

Plasma concentrations (μmol/l, M±SE) of individual amino acids in control and septic infants

	Early onset group control	Early onset group septic infants	Late onset group control	Late onset group septic infants
	(n = 10)	(n = 6)	(n = 10)	(n = 7)
Taurine	248±25	139±37+	77± 5	169±21**
Aspartate	51± 5	39± 6	50± 4	53± 6
Glutamate	92±12	52±10+	71± 7	163±32++
Citrulline	27± 5	18± 3	14± 2	—
Proline	222±33	178±14	211±24	258±33
Glycine	321±18	266±43	214±21	218±40
Alanine	344±16	248±26*	308±21	311±60
Cystine	150±29	82±11+	81±14	66± 6
Valine	182±13	128±18+	200±19	142±21
Methionine	30± 5	17± 3+	29± 4	20± 5
Isoleucine	46± 2	26± 7**	56± 6	26± 3**
Leucine	104± 9	63±13+	115±14	70± 7++
Tyrosine	131±18	70±13++	133±18	36± 4**
Phenylalanine	132±12	126±33	195±28	231±82
Lysine	207±24	151±16	277±32	105±12**
Histidine	73±20	77±16	90±16	88±19
Arginine	62±10	44± 9	60± 7	35± 4*
Ornithine	78± 6	56±11	101±10	54± 6*
Total	2497±87	1783±104**	2267±99	2045±70

+ $p < 0.05$
++ $p < 0.02$
* $p < 0.01$
** $p < 0.001$

Results

The mean values for blood glucose, lactate and plasma FFA concentrations are shown in Table III. It can be seen that the mean glucose level in the early onset group was significantly lower than in the respective controls. Apart from this, no additional significant differences were observed in the glucose, FFA and lactate levels of the control and septicaemic infants in both age groups.

The mean concentrations of individual amino acids are shown in Table IV. It is seen that the qualitative and quantitative responses of the plasma aminogram to severe bacterial infections were somewhat different in the early onset and the late onset group. While in the former the total amount of the 18 amino acids was significantly decreased in relation to the control value, in the latter group, although a definitely lower concentration was observed, the difference was not significant statistically.

As regards the individual plasma concentrations, 9 amino acids (taurine, glutamate, alanine, cystine, valine, leucine, isoleucine, methionine and tyrosine) in the early onset group, and 6 amino acids (leucine, isoleucine, tyrosine, lysine, arginine and ornithine) in the late onset group were found to be significantly lower than the control levels. The unchanged alanine concentration, the non-significantly lowered cystine, valine and methionine, as well as the significantly lowered lysine, arginine and ornithine concentrations obtained in the older septicaemic infants were the main differences between the two septicaemic amino acid profiles. Besides, cystine, valine and methionine in the older group, and lysine, arginine and ornithinine in the younger group showed a definite fall in their plasma concentration, which, however, was not significant statistically. The concentration of alanine remained unchanged in the older infants, whereas it decreased significantly in the early onset group.

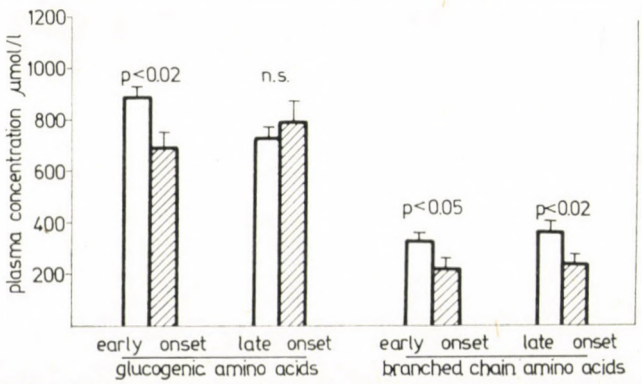

Fig. 1. Sum of the plasma concentrations of the three prominent glucogenic amino acids (ALA + PRO + GLY) and of the branched chain amino acids (VAL + ILEU + LEU) in neonatal and infant septicaemia in relation to the respective controls (▨ septic infants, ▭ control infants)

The sum of the three prominent glucogenic amino acids alanine, proline and glycine and that of the branched chain amino acids valine, leucine and isoleucine are shown in Fig. 1. It can be seen that while the glucogenic group showed a significant decrease in the early onset group only, the branched chain amino acids were significantly lowered in both groups of septic infants.

Deficient calorie and/or protein intake and hence impaired nutritional status are often associated with infection. Since an increased glycine: valine ratio due to increased level of glycine and decreased level of valine has been suggested to be characteristic of experimental and clinical protein-calorie malnutrition [1, 3, 8], this quotient has also been calculated (see Table V). In both groups of septic infants a somewhat increased glycine: valine ratio was obtained, but it did not reach the level of significance.

It has been reported that a number of infections are characterized by an increase in the plasma phenylalanine: tyrosine ratio [19, 20]. In order to see whether this applied to infected neonates and older infants, the ratio of these two aromatic amino acids has also been calculated. As it is seen in Table V, the ratio was significantly increased in both groups of septic infants.

Discussion

The quantitative and qualitative responses of plasma amino acids to severe bacterial infections in the neonatal and postnatal period were similar to those observed in adults suffering from sandfly or typhoid fever [18, 22, 23]. This similarity applied particularly to the changes in the total and individual amino acids obtained in early onset septicaemia. First of all, the marked hypoaminoacidaemia should be interpreted as a characteristic phenomenon and a global indicator of the profound changes induced in the circulating amino acid pool. The decreased plasma amino acid content resulted from the decreased concentration of several individual amino acids.

In full agreement with observations in infected adult patients [18], both the individual and the combined plasma levels of the branched chain amino acids were significantly depressed in the two groups of septic infants.

Table V

Plasma amino acid ratios in neonatal and postneonatal septicaemia

	Early onset group		Late onset group	
	control (n = 10)	septicaemia (n = 6)	control (n = 10)	septicaemia (n = 7)
PHE/TYR ratio	1.15±0.16	2.22±0.46*	1.77±0.28	6.12±1.86*
GLY/VAL ratio	1.81±0.11	2.28±0.41	1.19±0.17	1.69±0.37

* $p < 0.05$

This response represents a common and typical alteration in the plasma amino acid pattern of septic neonates, infants and adults. As regards the mechanism of the decreased concentrations of branched chain amino acids, experimental observations [7, 15, 16, 18, 24] suggest a decreased release into the circulation, and an increased utilization in the muscles as a source of energy. It is postulated that the branched chain amino acids liberated in the muscles by the increased protein catabolism induced by severe infection are metabolized in increased amounts for the production of glucogenic amino acids, such as alanine and glutamine.

According to experimental observations in animals and humans, it appears that the increased gluconeogenesis plays a central role in the adaptation to infection [1, 18, 21]. The limited availability of fuels reflected by the decreased glucose and FFA utilization induces increased proteolysis and amino acid oxidation to supply energy needs. This postulated chain of events seems to be applicable to the present findings, particularly to those obtained in early neonatal septicaemia where hypoglycaemia was a frequent occurrence. Since most of the infants included in this group were born before 37 weeks of gestation, prematurity, in addition to sepsis, could have also been responsible, for the glucose deficiency. One infant showed somatic features of intrauterine malnutrition, a well-known and frequent cause of neonatal hypoglycaemia. The plasma aminogram of the septic neonate, however, was not indicative of a decreased gluconeogenic capacity [10, 14], as instead of an increased total plasma amino acid level, hypoaminoacidaemia was observed.

The metabolic derangements different from that reported for hypoglycaemic malnourished neonates [10, 14] was also reflected by the behaviour of the glucogenic amino acids. Alanine concentration and the combined level of the three prominent glucose precursors, alanine, proline and glycine was appreciably decreased in early onset septicaemia and somewhat but not significantly lowered in late onset septicaemia. This suggests a stimulated hepatic gluconeogenesis in contrast with the hypoglycaemic intrauterine malnourished infants in whom a marked accumulation of glucogenic amino acids was observed due to a decreased capacity of endogenous glucose production.

When interpreting the alterations of the plasma aminogram associated with infections, the effect of prolonged starvation and/or malnutrition should be considered [2, 6, 8, 9]. They are often concurrent conditions, when it is difficult to clarify which is mostly responsible for the plasma free amino acid deviations.

The present findings in septic infants showed features similar to the plasma aminogram in prolonged fasting and protein deficiency. Since metabolic studies in septic patients are often complicated by starvation or malnutrition, it would be important to separate the effects of infection

from those of calorie and/or protein deficiency. Besides the efforts made by Ghisolfi et al. [9] in infected and malnourished children, additional studies in different age groups would be necessary to find reliable discriminating criteria of the amino acid pattern related to infection and malnutrition.

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