dependent upon NEFA provided by hydrolysis of milk during the neonatal period, since the milk of most of the mammals has a high fat and low carbohydrate content. Therefore the newborn has to conserve the limited supply of glucose from the diet for glucose-dependent tissues while providing alternative substrates such as NEFA* and ketone bodies to other tissues. A marked hyperketonaemia has been observed in newborn infants (Melichar et al., 1965; Persson & Gentz, 1966), guinea pigs (P. H. Dûe & J. R. Girard, unpublished work), rabbits (Callikan et al., 1979) and rats (Page et al., 1971; Dahlquist et al., 1972). In contrast, blood ketone-body concentrations remain very low in newborn sheep (Varnam et al., 1978), pigs (Bengtson et al., 1969) and dogs (Spitzer & Weng, 1972). In these species ketone bodies are unlikely to play an important role as energy substrates. In most physiological situations an increased rate of hepatic ketone-body production leads to hyperketonaemia, which in turn enhances ketone-body utilization in peripheral tissues. As the regulation of ketone-body utilization in the adult and the newborn has been reviewed recently (Robinson & Williamson, 1980), the present paper will be focused on the development of ketogenesis in rat and rabbit during the neonatal period.

Regulation of ketogenesis by NEFA availability

The major precursors of ketone bodies are NEFA, and changes in the rate of their delivery to the liver can directly influence ketogenesis. During the neonatal period the NEFA supplied to the liver have two different origins: (1) the triacylglycerols of the milk. The relative importance of dietary or endogenous triacylglycerols can be easily studied by starving the newborns. The newborn guinea pigs and rabbits, which have very little white adipose tissue but large fat stores in the liver, develop a marked hyperketonaemia after 12–24 h even if they are unfed from birth (Callikan et al., 1979; P. H. Dûe & J. R. Girard, unpublished work). In these species NEFA provided by hepatic triacylglycerol breakdown are the major precursors of ketone bodies during starvation. In keeping with this, it has been shown that glycodizane, an inhibitor of hepatic triacylglycerol lipase, completely suppressed the endogenous production of ketone bodies by isolated hepatocytes from 24–48 h old starved newborn rabbits (L. El Manoubi & J. R. Girard, unpublished work). In contrast, newborn rats, which have no fat stores at birth, remain hypoketonaemic if they are starved from birth (Girard et al., 1973), and they are entirely dependent upon NEFA provided by hydrolysis of milk triacylglycerol to perform efficient ketogenesis (Ferré et al., 1978). It is noteworthy that in rat and rabbit the milk contains a high proportion of medium-chain fatty acids, respectively 40 and 70% (Dils et al., 1977), whose transport into the mitochondria is not dependent upon carnitine acyltransferase (see McGarry & Foster, 1980).

In suckling rats blood ketone-body concentrations increase very slowly during the first 12 h after delivery despite the large increase in plasma NEFA which occurs 2 h after birth (Ferré et al., 1978). Then blood ketone bodies increase sharply between 12 and 16 h, without any change in the plasma NEFA concentrations (Ferré et al., 1978). A similar pattern in the ketogenic capacity of the newborn rats is observed when starved newborns are fed with a triacylglycerol emulsion containing long-chain fatty acids (Ferré et al., 1978) or medium-chain fatty acids (J.-P. Pégrier & J. R. Girard, unpublished work), at different times after birth. Recent experiments using hepatocytes isolated from newborn rats at different times after birth and incubated in the presence of constant concentrations of oleate (1 mM) or octanoate (2 mM) also suggest that factors other than NEFA availability are responsible for the onset of ketogenesis at birth. In hepatocytes of newly born rats, octanoate is more ketogenic than oleate (Table 1). The ketogenic capacity of isolated hepatocytes increased markedly during the 16 h after birth with both oleate and octanoate as substrates (Table 1). Data obtained with octanoate suggest that carnitine acyltransferase is not the only limiting step in the development of ketogenesis in rat liver.

Intra-hepatic regulation of ketogenesis in the newborn

Most of the information at present available on the development of hepatic ketogenesis has been obtained in the rat. The rates of palmitate oxidation and ketone-body production are very low in foetal rat liver, and they markedly increase in newborns which are 1 day old or older (Drahota et al., 1964; Augenfeld & Fritz, 1970; Lockwood & Bailey, 1970; Foster & Bailey, 1976a). These changes are accompanied by an increase in the activity of carnitine acyltransferases (Augenfeld & Fritz, 1970; Lockwood & Bailey, 1970; Warshaw, 1972; Foster & Bailey, 1976b; Yeh & Zee, 1979) and of the enzymes of the hydroxy methylglutaryl-CoA pathway (Lockwood & Bailey, 1971; Lee & Fritz, 1971; Hipolito-Reis et al., 1974; Shah & Bailey, 1977). Unfortunately these studies do not provide any

<table>
<thead>
<tr>
<th>Substrates added</th>
<th>Age of newborns</th>
<th>Rate of ketogenesis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0h</td>
<td>6h</td>
</tr>
<tr>
<td>Oleate (1 mM)</td>
<td>16±2</td>
<td>22±4</td>
</tr>
<tr>
<td>Octanoate (2 mM)</td>
<td>94±13</td>
<td>166±20</td>
</tr>
<tr>
<td></td>
<td>173±22</td>
<td>229±25</td>
</tr>
</tbody>
</table>

*Abbreviation: NEFA, non-esterified fatty acids.

Table 1. Ketone-body synthesis in isolated hepatocytes from starved newborn rats

Rates of ketogenesis are expressed as µmol of ketone bodies formed/ h per 10^6 hepatocytes. The values are means ± S.E.M. of six to ten determinations.
Table 2. Developmental changes in the activity of liver mitochondrial carnitine palmitoyl transferase in unfed newborn rats

<table>
<thead>
<tr>
<th>Age of newborns</th>
<th>Activity of liver carnitine palmitoyl transferase</th>
</tr>
</thead>
<tbody>
<tr>
<td>0h</td>
<td>1.5 ± 0.1</td>
</tr>
<tr>
<td>4h, unfed</td>
<td>2.0 ± 0.2</td>
</tr>
<tr>
<td>12h, unfed</td>
<td>3.7 ± 0.6</td>
</tr>
<tr>
<td>16h, unfed</td>
<td>5.2 ± 0.3</td>
</tr>
<tr>
<td>16h, suckled</td>
<td>4.4 ± 0.3</td>
</tr>
</tbody>
</table>

Enzyme activity was measured as described by Solberg (1974) and is expressed as nmol/min per mg of protein. Values are means ± S.E.M. for 6 to 16 determinations.

Information on the time course or on the possible factors that may regulate hepatic ketogenesis during the first 24h of life. The regulation of ketogenesis in the adult rat has been reviewed recently (McGarry & Foster, 1980). The mechanisms proposed to explain an increase in hepatic ketogenesis in various conditions include an increase in plasma glucagon/insulin ratio, which in turn serves to activate the liver carnitine acyltransferase reaction via an increase in carnitine and a fall in malonyl-CoA concentration in the liver. To what extent can these conclusions be applied to the foetal–neonatal transition?

(1) Changes in plasma insulin and glucagon concentrations

Immediately after birth there is a sharp decrease in plasma insulin coupled with a marked increase in plasma glucagon in both rat (Girard et al., 1978) and rabbit (Callikan et al., 1979). These hormonal changes are thus appropriate to explain the rise in the rate of hepatic ketogenesis. Moreover, experimental situations in which a rise in plasma glucagon/insulin ratio is observed in utero (prolonged pregnancy (Portha et al., 1978) or injection of phlorizin to the mother (Freund et al., 1980)) are associated with a premature appearance of ketogenesis in the newborn rat (J.-P. Pégourier & J. R. Girard, unpublished work).

(2) Changes in liver carnitine acyltransferase activity

As shown in Table 2, carnitine acyltransferase increases progressively in the liver of newborn rats during the first 24h of life. An increased activity of carnitine acyltransferase has been found in the liver of postmature foetuses (J.-P. Pégourier & J. R. Girard, unpublished work).

(3) Liver carnitine

It has been suggested that the hepatic carnitine is a key factor in the regulation of ketogenesis in the newborn rat, since liver content of carnitine is increased in sucking rats (Robles-Valdes et al., 1976). However, the difference between the time course of increase in liver carnitine and blood ketone-body concentrations suggests that carnitine is not a key factor determining the onset of hepatic ketogenesis after birth (Ferré et al., 1978). Moreover, 16h-old newborn rats starved from birth show a marked rise in blood ketone-body concentrations in response to a feed of triacylglycerol emulsion despite a low and unchanged hepatic carnitine concentration (Ferré et al., 1978).

(4) Liver carbohydrate metabolism

In newborn rats (Ferré et al., 1978), as in adult rats (review in McGarry & Foster, 1980), there is an inverse relationship between liver glycogen concentration and the capacity for ketogenesis. The postnatal onset of ketogenesis may be linked to a metabolic event accompanying the decreased availability of hepatic carbohydrates after birth, such as decreased lipogenesis (Ballard & Hanson, 1967; Taylor et al., 1967) or increased gluconeogenesis (Ballard, 1971; Girard et al., 1975). In vivo, the rate of lipogenesis from \(^3\)H\(_2\)O is markedly decreased 3h after birth in the liver of starved or suckled rats and remains very low until 24h (M. Escriva, J.-P. Pégourier & J. R. Girard, unpublished work). It is reasonable to assume that [malonyl-CoA is very low a long time before ketogenesis becomes active. Therefore the neonatal period is ideal to test whether factors other than [malonyl-CoA regulate hepatic ketogenesis. Among these factors the large increase in gluconeogenesis after birth (Ballard, 1971; Girard et al., 1975) could influence the rate of intramitochondrial acetyl-CoA by channeling oxaloacetate toward glucose synthesis, thus decreasing citrate synthesis and favouring ketogenesis.


