Reprogramming of metabolic development by restriction of fetal growth

COLIN T. JONES
The Nuffield Institute for Medical Research, University of Oxford, Headley Way, Oxford OX3 9DS, U. K.

It has frequently been proposed that metabolic development occurs in predefined phases that are relatively invariant and critical in the sense that if they do not take place normally the end result is gross abnormality (Dobbing, 1968; Greengard, 1970). A good example of this is the delay of myelination caused by hypothyroidism (Balazs, 1979). Whether this is completely correct has been questioned (Jones et al., 1980b). A priori, if the onset of developmental processes result from local hormonal or cell contact stimuli, it could be expected that a precocious or remedial simulation of such normal changes should post- or post-maturely initiate the natural chain of development. There are plenty of such examples that equate the appearance of particular proteins or pathways with perinatal endocrine changes, such as the role of steroids in inducing lung phospholipid synthesis (Batenburg & Van Golde, 1979). However, the capacity to respond to hormones, as exemplified by the induction of liver enzymes (Greengard, 1970; Jones, 1982), is not constant throughout development. Moreover, the ability to initiate the precocious appearance of a protein by hormone administration does not prove a normal role for that agent, as exemplified by the complex function of cortisol in metabolism at relatively high doses to intact animals or cells in culture. Such techniques have not resolved the question of the flexibility of development and usually leave open the nature of the normal mechanisms of induction.

An alternative approach that has been relatively little used is to retard development by restricting nutrient supply and then to follow the subsequent timing of metabolic maturation and the factors associated with the natural reprogramming of growth patterns (Jones & Robinson, 1979; Lafeber et al., 1979; Jones et al., 1980b). The results from such experiments will be described here. A common experimental method of causing growth retardation is to restrict fetal nutrient supply by reducing maternal placental blood flow (Jones & Robinson, 1979; Lafeber et al., 1979; Jones et al., 1980a). This limits growth by as much as 75%, but is still consistent with survival. Organ development re-adjusts so that the growth of the visceral tissues is slowed considerably whilst that of the brain is largely maintained (Lafeber et al., 1979; Lafeber et al., 1984). The slowing of the growth rate of selected fetal organs is also associated with delayed matura tion. Thus in skeletal muscle, where maturation is relatively late by comparison with other tissues, the fusion of myoblasts to form myotubes and the appearance of an ordered myofibre structure may not be a prenatal event, as it is normal, but may occur many days later than expected. In the liver, enzymes such as those associated with gluconeogenesis or with fatty acid synthesis do not appear at the expected prenatal time and exhibit an abnormal pattern of relatively slow induction with peak activities occurring many days later than in well-nourished fetuses (Jones et al., 1980b). The heart shows a phase of delayed maturation, but because its development can occur relatively early in gestation, by term its biochemical characteristics can appear normal even though its size may be considerably reduced (Lafeber et al., 1979). Even the brain is subjected to a slowing of its normal maturation. Thus myelination, which in the guinea pig for instance is mostly prenatal, can not only be slowed by intra-uterine growth retardation but also caused to occur largely after birth (Jones et al., 1980b; Jones & Rolph, 1981).

The pattern that therefore emerges is of nutritional deprivation reprogramming normal developmental patterns so that they shift in time. Examples of this are shown in Fig. 1. In prenatal liver there are phases of DNA, RNA and protein synthesis that occur at distinct times (Miller, 1969). In liver of the growth-retarded fetal guinea pig such changes are blunted and the peak phases of deposition and synthesis are 5–10 days later than normal. A similar delay is seen in the appearance of the pathway for fatty acid synthesis (Jones et al., 1980b). Although slowing of development is a fairly common feature of growth retardation, enhanced maturation is seen in a number of specific instances; thus the onset of lung surfactant synthesis is accentuated.

A potential explanation for the general delay of selected gene expression, particularly that associated with maturation in the second half of gestation, may be found in the underlying hormonal changes (Jones & Robinson, 1979; Lafeber et al., 1979; Jones et al., 1980a,b; Jones et al., 1984). Hypoglycaemia is a common feature of growth retardation and thus it is not surprising that such fetuses are severely hypoinsulinaemic and have particularly high plasma glucagon concentrations (Jones & Robinson, 1979; Lafeber et al., 1979; Robinson et al., 1980; Jones et al., 1981; Jones et al., 1984). In contrast, although plasma cortisol concentrations can be somewhat lower, their developmental pattern is normal (Jones et al., 1980). The thyroid hormones show the greatest alteration although plasma prolactin may also be very low (Jones et al., 1980b; Robinson et al., 1980; Wrutniak & Cabello, 1984). Thus plasma thyroxine concentrations in the growth-retarded fetus are lower and change more slowly than normal (Fig. 2). It is of interest that in both normal and reduced growth states the capacity for hepatic fatty acid synthesis follows closely the plasma thyroxine level (Jones et al., 1980a,b). Plasma thyroid hormone changes may also be implicated in the inhibition of the
Fig. 1. Changes in the relative rates of DNA and RNA accumulation and of protein synthesis from leucine in liver of (a) normal and (b) growth-retarded fetal guinea pigs. Growth retardation was caused by uterine artery ligation at day 30 of pregnancy. (●) DNA accumulation; (▲) RNA accumulation; (○) protein synthesis. Vertical bars represent 2 S.D.

Fig. 2. Changes during gestation of plasma total thyroxine concentrations in normal and growth-retarded fetal guinea pigs. (●) Normal guinea pigs; (○) growth-retarded guinea pigs. Other details as given in the legend to Fig. 1.

Normal appearance of pyruvate carboxylase activity, which unlike the other gluconeogenic enzymes is not reversed immediately after birth, but only about 4–5 days later when plasma thyroxine is presumed to rise (Rolph & Jones, 1982). It is possible that low plasma thyroid hormone concentrations explain the slow rates of myofibrillar and hepatic maturation and of brain myelination, but many of the changes are not easily accounted for in this way.

Thus it is a paradox that a hepatic protein such as phosphoenolpyruvate carboxykinase, whose synthesis is regulated by glucagon and cortisol, should have much lower activity in the growth-retarded fetuses with high glucagon-to-insulin ratios (Jones et al., 1981). Sensitivity to these hormones could be regulated by thyroid hormone level, but alternative explanations are available. Precocious induction of gluconeogenic enzymes by cortisol and glucagon has been reported for fetal-rat liver (Hanson et al., 1973; Steele et al., 1980) and for isolated fetal-guinea-pig hepatocytes (Table 1). Thus there is a factor in vivo that limits the ability of fetal liver to respond. Previous studies have shown that plasma of growth-retarded fetuses contains anti-mitogenic factors inhibitory to cartilage growth (Jones & Robinson, 1979; Lefebre et al., 1979). This appears also to be true for hepatocyte growth which is inhibited by plasma from growth-retarded, but not from that of normal, fetal guinea pigs (Fig. 3). A similar action is observed on the induction of phosphoenolpyruvate carboxykinase (Table 1). Thus plasma from growth-retarded fetuses contains a heat- and trypsin-sensitive factor of about 40 K, able to suppress enzyme induction by glucagon and cortisol. There is some evidence to suggest that this factor could be of placental origin (Fig. 3), and there are reports of the placenta producing anti-mitogenic factors (Boden, 1975).

In summary there is considerable flexibility in the rate and extent of biochemical maturation. This is likely to ensure that vital developmental processes occur at the
Table 1. Induction of phosphoenolpyruvate carboxykinase in isolated hepatocytes from 40-day fetal guinea pigs

The hepatocytes were incubated for 48 h in medium containing insulin, thyroxine and epidermal growth factor and either 20% serum from normal 45-day or growth-retarded fetal guinea pigs with or without cortisol (1 µg/ml) and glucagon (5 µg/ml). The effects of a 40K protein fraction isolated by Biogel filtration from plasma of growth-retarded 50-day fetal guinea pigs were also investigated. Results are means ± S.D., with the number of observations in parentheses.

<table>
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<tr>
<th></th>
<th>Control</th>
<th>Glucagon + cortisol</th>
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<tr>
<td>Control</td>
<td>4.2 ± 1.9 (6)</td>
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<tr>
<td>Control + 48 h</td>
<td>7.6 ± 2.8 (5)</td>
<td>45.3 ± 11.6 (5)</td>
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<tr>
<td>Growth-retarded serum 48 h</td>
<td>3.5 ± 1.4 (6)</td>
<td>12.7 ± 4.9 (5)</td>
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<tr>
<td>+ 40K fraction 48 h</td>
<td>2.7 ± 1.1 (5)</td>
<td>9.3 ± 2.6 (5)</td>
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most favourable times. In addition to the role of plasma hormones it is likely that inhibitory factors, potentially derived in part from the placenta, communicate nutritional states to the fetal tissues.


Hanson, R. W., Fisher, L., Ballard, F. J. & Reshef, L. (1973) ENZYMES 15, 97-110