EFFECTS OF ELECTROCOAGULATION OF THE FOETAL LAMB HYPOPHYSIS ON GROWTH AND DEVELOPMENT

G. C. LIGGINS* AND P. C. KENNEDY

Department of Physiological Sciences, University of California
School of Veterinary Medicine, Davis, California, U.S.A.

(Revised manuscript received 23 October 1967)

SUMMARY

Electrocoagulation lesions involving 70–100% of the hypophysis of 17 foetal lambs were made at various times between 93 and 136 days of gestation. Foetal growth and development after the operation was compared with that in a group of seven control lambs comprising three intact littermates of operated foetuses and four sham-operated foetuses. Hypophysial damage was found to be associated with retardation of somatic development which was most marked when the operation was performed early in gestation. Retardation was particularly evident in epiphysial development. Hypoplasia of the adrenal cortices, the interstitial cells of the testis and the thyroid glands confirmed the assumption made by previous workers based on decapitation experiments, that the foetal hypophysis exerts a trophic influence on the development of these endocrine organs. Reduced liver glycogen content and plasma glucose levels indicated a disturbed carbohydrate metabolism. Abnormal subcutaneous fat deposition suggested disorders in fat metabolism. Delayed involution of haemopoietic tissues, hypoplasia of peripheral lymphoid tissue and reduction in the number of mast cells in the skin were also noted. Structures of ectodermal origin were relatively unaffected by hypophysial lesions. It is concluded that the hypophysis of the foetal lamb subserves many of the functions attributed to it during extrauterine life.

INTRODUCTION

The foetal hypophysis is usually considered to make no significant contribution to foetal growth. Decapitation of rabbit foetuses (Jost, 1947) and of rat foetuses (Wells, 1947; Jost, 1951a) was not followed by retardation of somatic growth. Raynaud & Frilley (1947) reported similar findings after destruction of the hypophysis in mice by X-rays. In man also the available evidence does not support an important role of the hypophysis in foetal growth: the anencephalic foetus may attain a normal birth weight at term. Similarly, newborn infants who show signs of hypopituitary

* Address for reprint requests: Postgraduate School of Obstetrics and Gynaecology, University of Auckland, Auckland, New Zealand.
dwarfism after 6 months are usually a normal birth weight (Brasel, Wright, Wilkins & Blizzard, 1965) as are children with pituitary aplasia who die soon after birth (Blizzard & Alberts, 1956; Brewer, 1957; Reid, 1960). Nevertheless there are some indications that foetal growth may depend to some extent on hypophysial secretion of foetal origin. Heggstad & Wells (1965), who decapitated foetal rats, concluded that 20% of foetal growth in late pregnancy depended on the foetal hypophysis. Guernsey calves with a genetically determined anomaly involving the central nervous system and hypophysis usually fail to attain normal maturity even when the pregnancy is prolonged for several months beyond term (Kennedy, Kendrick & Stormont, 1957). Moreover, the human anencephalic foetus, although cited as evidence against a role of the foetal hypophysis in somatic growth, may remain within the range of normal birth weights when delivered 2 months or more beyond term (Frandsen & Stakeman, 1961; Comerford, 1965).

Apart from growth, other trophic activities of the foetal hypophysis can be demonstrated readily. Adrenal hypoplasia after foetal decapitation has been observed in the rat by Wells (1947), Jost (1951a), Christianson & Chester Jones (1957) and Kitchell & Wells (1952), who also showed that hypoplasia was prevented by giving adrenocorticotropic hormone (ACTH). Thyroid hypoplasia in the foetus has been demonstrated after decapitation in the hamster (Foote & Foote, 1949), the rat (Jost, 1966), the rabbit (Jost, 1953) and the mouse (Raynaud, 1950) and has been associated with reduced secretion of thyroxine (Geloso, 1958). The hypoplasia may be remedied by administration of thyrotrropic hormone (Geloso, 1964). The interstitial cells of the foetal testis show similar hypoplastic changes after decapitation in the rat (Wells, 1950) and the rabbit (Jost, 1951b), although Raynaud (1950) failed to confirm this finding in the mouse. The human anencephalic foetus may have marked hypoplasia of the adrenal cortex (Lanman, 1957), the thyroid (Saxén, 1958) and the testis (Zondek & Zondek, 1965).

The purpose of this report is to describe observations on foetal hypophysial function in the sheep, a species not previously investigated.

**Materials and Methods**

A flock of 25 mature ewes of mixed breeding were run in groups with a ram carrying a marking harness; the ewes were inspected daily and the marked ones separated from the ram. The date of marking was taken as the first day of gestation and term was calculated by adding 150 days. Foetal maturity was estimated by X-rays at 60-100 days of gestation and at laparotomy by estimation of skull dimensions.

Ablation of the hypophysis was performed between the 93rd and 136th day of gestation by electrocoagulation with an insulated probe passed into the sella turcica by an approach through the intact uterine wall into the frontal bone in the midline of the skull. Details of the surgical techniques are given elsewhere (Liggins, Kennedy & Holm, 1967). Lambs with lesions involving less than 70% of the whole gland were excluded. In three cases of multiple pregnancy, one foetus was left unoperated. The probe failed to reach the vicinity of the hypophysis or hypothalamus in four which have been classified as sham operations. The remaining 11 ewes carried foetuses with hypophysial lesions involving 70-100% of the gland. In some ewes parturition...
occurred spontaneously at term. In others, elective caesarean section was performed 8–37 days beyond term.

Lambs delivered by caesarean section were killed by exsanguination within 15 min. Those born spontaneously were killed when found, the oldest being 12 hr. Heparinized foetal and maternal blood was immediately refrigerated, centrifuged and the plasma was separated. Lambs were weighed after emptying the stomach. A detailed macroscopic examination was made of all tissues. Thyroids, adrenals and testes were weighed and then fixed in formol-Zenker solution; other tissues were fixed in 10% formalin. A block containing the sella turcica was cut from the base of the skull in such a way that a portion of hypothalamus remained attached to the intact hypophysial stalk. A lateral radiograph was taken of a hind limb from each lamb and the degree of osseus maturity was assessed by counting the number of ossification centres (Lascelles, 1959).

Examination of hypophysis. After decalcification in 15% formic acid, sagittal sections were cut through the hypophysial block. A macroscopic estimate of the extent of the lesion was made. This was expressed as a percentage, to the nearest 10%, of the expected volume of the whole gland.

Examination of adrenals. In addition to routine staining, histochemical examination was made by the method of Rubin, Deane & Hamilton (1963) for demonstrating the location and degree of activity of the enzyme Δ4-3β-hydroxysteroid dehydrogenase. Fresh foetal adrenals were set in small blocks of foetal liver and were frozen in liquid nitrogen. Sections were cut at 12 μ in a cryostat at −20°C. Control media were identical except for the absence of the substrate dehydroepiandrosterone.

Biochemical methods. Plasma total-reducing substances were estimated by the method of Folin & Malmros (1929), fructose by the method of Roe (1934) and glucose by the glucose oxidase method (Marks, 1959). Plasma protein-bound iodine was estimated by the method of Farrell & Richmond (1961).

RESULTS

Birth weight

The effects of hypophysial lesions were best observed in a group of three ewes with multiple pregnancies in which an intact foetus served as a control (Table 1). In each instance, the lamb with a hypophysial lesion weighed less than the control twin or triplet. The difference in weight was greatest the earlier the operation was performed in pregnancy.

In the prolonged pregnancy group (Table 2) the mean birth weight of lambs delivered 7–37 days beyond term did not differ from that of term controls (Table 3). The two largest lambs in the former group, weighing 5·55 and 5·9 kg respectively, were exceptional in having extensive cerebral degeneration as well as total destruction of the hypophysis; each had marked subcutaneous oedema which contributed an unknown increment to the body weight.

Bone age

Examples of term epiphysial development were provided by four sham-operated lambs, two unoperated twins and one unoperated triplet. Bone ages of 13 lambs with
hypophysial lesions involving more than 70% of the gland ranged from 110 days to term (Table 2). The most marked retardation was observed after ablation of the hypophysis at 93 days; the bone age of this lamb at 173 days of gestation was 110 days. The least marked retardation occurred in a lamb which attained a bone age of

Table 1. Birth weight, organ weight and bone age of lamb foetuses with hypophysial lesions, of their intact littermates and of a group of sham-operated foetuses

<table>
<thead>
<tr>
<th>Ewe no.</th>
<th>Maturity at operation (days)</th>
<th>Maturity at delivery (days)</th>
<th>Birth weight (kg.)</th>
<th>Adrenals (mg.)</th>
<th>Thyroid (mg.)</th>
<th>Testes (mg.)</th>
<th>Bone age (days)</th>
<th>Extent of hypophysial lesion (% of whole gland)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A. Multiple pregnancies</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>2358</td>
<td>95</td>
<td>147</td>
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<tr>
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<td>874</td>
<td>576</td>
<td>1795</td>
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<td></td>
</tr>
<tr>
<td>191</td>
<td>122</td>
<td>149</td>
<td>3.50</td>
<td>352</td>
<td>1050</td>
<td>880</td>
<td>135</td>
<td>90</td>
</tr>
<tr>
<td></td>
<td>5-10</td>
<td>888</td>
<td>1660</td>
<td>1500</td>
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<td></td>
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<td>146</td>
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<td>398</td>
<td>460</td>
<td>—</td>
<td>130</td>
<td>&gt; 90</td>
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<tr>
<td></td>
<td>3-60</td>
<td>300</td>
<td>500</td>
<td>1090</td>
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</tr>
<tr>
<td></td>
<td>3-90</td>
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<td>775</td>
<td>1650</td>
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<td>B. Sham-operated foetuses</td>
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<td>740</td>
<td>—</td>
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</tr>
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<td>142</td>
<td>147</td>
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<td>907</td>
<td>—</td>
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<tr>
<td>2285</td>
<td>142</td>
<td>145</td>
<td>4-35</td>
<td>760</td>
<td>890</td>
<td>—</td>
<td>Term</td>
<td>0</td>
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</tbody>
</table>

Table 2. Birth weight, organ weight and bone age of 13 lamb foetuses with hypophysial lesions in single pregnancies or twin pregnancies with both foetuses operated upon

<table>
<thead>
<tr>
<th>Ewe no.</th>
<th>Maturity at operation (days)</th>
<th>Maturity at delivery (days)</th>
<th>Birth weight (kg.)</th>
<th>Adrenals (mg.)</th>
<th>Thyroid (mg.)</th>
<th>Testes (mg.)</th>
<th>Bone age (days)</th>
<th>Extent of hypophysial lesion (% of whole gland)</th>
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<tr>
<td>2355</td>
<td>93</td>
<td>165</td>
<td>4.35</td>
<td>251</td>
<td>315</td>
<td>970</td>
<td>124</td>
<td>90</td>
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<td>2360</td>
<td>93</td>
<td>173</td>
<td>4.00</td>
<td>209</td>
<td>273</td>
<td>1020</td>
<td>110</td>
<td>80</td>
</tr>
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<td>101</td>
<td>164</td>
<td>5.55</td>
<td>380</td>
<td>494</td>
<td>1135</td>
<td>120</td>
<td>&gt; 90</td>
</tr>
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<td>2383</td>
<td>102</td>
<td>187</td>
<td>5.90</td>
<td>205</td>
<td>410</td>
<td>1200</td>
<td>135</td>
<td>&gt; 90</td>
</tr>
<tr>
<td>263</td>
<td>103</td>
<td>166</td>
<td>3.60</td>
<td>267</td>
<td>680</td>
<td>850</td>
<td>115</td>
<td>&gt; 90</td>
</tr>
<tr>
<td>4048</td>
<td>120</td>
<td>175</td>
<td>5.35</td>
<td>232</td>
<td>822</td>
<td>1038</td>
<td>145</td>
<td>&gt; 90</td>
</tr>
<tr>
<td>254</td>
<td>120*</td>
<td>166</td>
<td>3.50</td>
<td>740†</td>
<td>585</td>
<td>1023</td>
<td>130</td>
<td>&gt; 90</td>
</tr>
<tr>
<td></td>
<td>3-80</td>
<td>293</td>
<td>455</td>
<td>—</td>
<td>130</td>
<td>&gt; 90</td>
<td></td>
<td></td>
</tr>
<tr>
<td>265</td>
<td>126</td>
<td>161</td>
<td>4.20</td>
<td>374</td>
<td>657</td>
<td>1105</td>
<td>Term</td>
<td>80</td>
</tr>
<tr>
<td>4032</td>
<td>132</td>
<td>158</td>
<td>4.40</td>
<td>272</td>
<td>953</td>
<td>—</td>
<td>Term</td>
<td>&gt; 90</td>
</tr>
<tr>
<td>4031</td>
<td>133</td>
<td>182</td>
<td>4.10</td>
<td>360</td>
<td>553</td>
<td>—</td>
<td>Term</td>
<td>70</td>
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<tr>
<td>4012</td>
<td>136*</td>
<td>157</td>
<td>4.02</td>
<td>383</td>
<td>725</td>
<td>—</td>
<td>Term</td>
<td>&gt; 90</td>
</tr>
<tr>
<td></td>
<td>136*</td>
<td>157</td>
<td>3-50</td>
<td>207</td>
<td>497</td>
<td>1182</td>
<td>Term</td>
<td>&gt; 90</td>
</tr>
</tbody>
</table>

Mean ± s.d. 4.33 ± 0.75 316 ± 70 594 ± 273 1040 ± 107 — —

* Twins.  † Foetus infused with synthetic ACTH for 7 days before delivery.

145 days at 175 days of gestation after total destruction of the hypophysis at 120 days. With this exception, destruction of the gland before 125 days limited bone age to 135 days at gestational ages ranging from 145 to 187 days. Operation after 125 days was consistent with the bone age at term when the pregnancy continued.
Hypophysectomy in the foetal lamb

at least a week beyond term. In general, the earlier the hypophysial lesion the greater the discrepancy between bone age and gestational age (Pl. 1, figs. 1, 2).

Gross X-ray changes were represented histologically by inactivity of the osteoblastic cells, both periostial ones and those lining the trabeculae (Pl. 5, figs. 15–18). In foetuses with hypophysial lesions the trabeculae were much less closely spaced.

Table 3. Comparison of mean birthweight and organ weights of lamb foetuses with hypophysial lesions and of control foetuses (means ± S.D.)

<table>
<thead>
<tr>
<th>Experimental group*</th>
<th>Birth weight (kg.)</th>
<th>Adrenals (mg.)</th>
<th>Thyroid (mg.)</th>
<th>Testes (mg.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Multiple pregnancies, operated foetuses (4)</td>
<td>3·26 ± 0·33</td>
<td>316 ± 70</td>
<td>566 ± 295</td>
<td>868 ± 131</td>
</tr>
<tr>
<td>P</td>
<td>&lt; 0·02</td>
<td>&lt; 0·01</td>
<td>N.S.†</td>
<td>&lt; 0·01</td>
</tr>
<tr>
<td>Multiple pregnancies, intact litter-mates (3)</td>
<td>4·60 ± 0·51</td>
<td>770 ± 158</td>
<td>1004 ± 470</td>
<td>1648 ± 125</td>
</tr>
<tr>
<td>Single pregnancies and twin pregnantuses with both foetuses operated (13)</td>
<td>4·33 ± 0·76</td>
<td>316 ± 70</td>
<td>594 ± 273</td>
<td>1040 ± 107</td>
</tr>
<tr>
<td>P</td>
<td>N.S.</td>
<td>&lt; 0·001</td>
<td>&lt; 0·03</td>
<td>&lt; 0·001</td>
</tr>
<tr>
<td>Control foetuses (7)</td>
<td>4·32 ± 0·44</td>
<td>680 ± 158</td>
<td>907 ± 324</td>
<td>1533 ± 217</td>
</tr>
</tbody>
</table>

* Number of observations in parentheses.
† N.S., not statistically significant (Student’s ‘t’ test).

Ectodermal structures

Whereas the bone age and body weight of lambs with hypophysial lesions were abnormal at gestation, the degree of development of ectodermal structures was more or less normal. This led to an unusual appearance in some of the post-term foetuses, the wool and hair being unduly long and straight, attaining lengths of 4 cm. or more. Teeth had erupted in both jaws, a finding not usual in normal lambs until several weeks after birth.

Adrenals

The mean adrenal weight in lambs with hypophysial lesions was approximately half that of normal controls (Table 3). The earlier the operations the greater the differences.

The failure of growth of the zona fasciculata in the lambs with pituitary lesions appeared to account for these weight differences. No changes were detected in the adrenal medulla and the changes in the zona glomerulosa were minor in operated foetuses compared with the controls. In the zona fasciculata, however, the normal cellular hypertrophy and hyperplasia failed to occur (Pl. 2, figs. 3–6). When the hypophysis was completely destroyed early in the last trimester of pregnancy the zona fasciculata was severely hypoplastic, averaging 0·12 mm. in thickness and being only slightly wider than the glomerulosa. In the normal lamb at term only a single inner zona could be clearly recognized; it was approximately 0·30 mm. wide. This increase in width of the zona fasciculata in the normal foetal lamb consisted of hyperplasia, enlargement of individual cells and loosening of the entire zone. It contrasted sharply with the appearance of this zone in the foetus with hypophysial damage.
Histochemical examination showed no characteristic differences in the distribution of $\Delta^3$-3$\beta$-hydroxysteroid dehydrogenase between adrenal cortices of control lambs and lambs with hypophysial lesions. In both groups, diformazan deposition was sparse in the zona glomerulosa but was heavy and evenly distributed in the inner zone of the cortex (Pl. 3, figs. 7, 8).

**Thyroids**

The mean thyroid weight of lambs with hypophysial lesions was reduced by more than one-third compared with control lambs (Table 3). There was greater variation in thyroid weights among the former group, the extremes being 273 and 1050 mg. The lowest thyroid weights were associated with operation before 120 days of gestation. In both the experimental and the control groups, follicles had formed, and colloid had accumulated; in many individual microscopic fields it was impossible to distinguish between experimental groups. In general, the thyroids of the foetuses with hypophysial lesions had a flatter follicular epithelium which contained less cytoplasm than the epithelium of the thyroids of the controls (Pl. 3, figs. 9, 10). The glands of the experimental group were less vascular than those of the controls.

Plasma protein-bound iodine levels were low in the lambs with marked thyroid hypoplasia but the levels were normal despite ablation of the hypophysis when the thyroid weight was within normal limits.

**Testes**

The mean weight of testes from lambs with hypophysial lesions was about two-thirds that of control lambs (Table 3). On histological examination, the testes of lambs whose hypophysis had been damaged were hypoplastic (Pl. 4, figs. 11, 12). This hypoplasia involved both the seminiferous tubules and the interstitium but it was most noticeable with respect to growth and differentiation of Leydig cells. In lambs operated on near the beginning of the last trimester the interstitial tissue was loose and only a small number of Leydig cells had formed. The tubules were also reduced in diameter. These changes were much less obvious when the operation had been performed near term.

**Ovaries**

Only four female foetuses in the series were suitable for histological examination. No changes in ovarian structure were found.

**Placenta**

In placentae of foetuses with pituitary lesions the chorionic epithelium of the tips of the villi was markedly vacuolated in many cases. The severity of these changes did not correlate closely with either the completeness of the operation or the length of time the foetus was retained in utero beyond term. Since normal chorionic epithelium may contain cells with vacuolated cytoplasm this feature is difficult to interpret. The cytoplasmic vacuoles did not contain stainable glycogen and their significance is not known.
Haemopoietic tissue

In the normal foetal lamb at term there are only small infrequent foci of haemopoietic tissue in the liver; this contrasts with the large amount of haemopoietic tissue found in the livers of the foetuses with hypophysial lesions (Pl. 4, figs. 13, 14). Extramedullary haemopoiesis was also seen in other sites; these included spleen, adrenal cortex and lymph nodes but in these organs the change was less obvious than in the liver. Differences between experimental and control foetuses were seen even when the operation was performed within 10 days of term, although the increased amounts of haemopoietic tissue in these cases was slight. The amounts of haemopoietic tissue present were not increased proportionately in foetuses operated on earlier than 20 days before term.

Table 4. Foetal (F) and maternal (M) plasma levels of protein-bound iodine, glucose and fructose in a sham-operated foetus and a group of foetuses with lesions of the hypophysis

<table>
<thead>
<tr>
<th>Ewe no.</th>
<th>Maturity at operation (days)</th>
<th>Maturity at delivery (days)</th>
<th>Extent of hypophysial lesion (% of whole gland)</th>
<th>Protein-bound iodine (µg./100 ml.)</th>
<th>Plasma glucose (mg./100 ml.)</th>
<th>Plasma fructose (mg./100 ml.)</th>
<th>Total reducing substances in plasma (mg./100 ml.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2396</td>
<td>147</td>
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<td>892</td>
<td>5-6</td>
<td>55</td>
<td>114</td>
<td>3</td>
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<tr>
<td>2355</td>
<td>93</td>
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<td>37</td>
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<td>157</td>
<td>725</td>
<td>4-7</td>
<td>5</td>
<td>32</td>
<td>29</td>
</tr>
</tbody>
</table>

* Less than 1 mg./100 ml.  † N.M., not measured.

Carbohydrate metabolism

Periodic acid-Schiff staining of histological sections of liver demonstrated marked reduction in glycogen content in lambs with hypophysial lesions (Pl. 4, figs. 13, 14). Plasma glucose levels were markedly reduced in most of the lambs with hypophysial lesions (Table 4). Fructose levels, on the other hand, were within the limits found by Barklay, Haas, Huggett, King & Rowley (1949) in normal lambs. Histological examination of the pancreas showed no characteristic changes.

Miscellaneous observations

Normal lambs were devoid of subcutaneous fat but lambs with hypophysial lesions of more than a few days standing were invariably covered by a layer of subcutaneous fat which reached a thickness of up to 1.5 cm. over the sternum. Histological examination showed that this was normal 'white' fat; 'brown' fat from the perinephric region was inconspicuous.

Lymph nodes were reduced in number and size in lambs with hypophysial lesions.
made before 120 days of gestation. Inspection of the thymus did not suggest a substantial alteration in weight and no histological changes were detected.

The accumulation of granules within mast cells was slowed or stopped by destruction of the hypophysis. When comparing the twin foetuses delivered spontaneously at term, measured sections of skin were examined for recognizable mast cells. On the average, twenty times as many mast cells were counted in the controls as in the foetuses with hypophysial damage. One or two granulated mast cells could be seen per high power field in the dermis of a normal newborn lamb. Granulated mast cells did not appear in normal numbers even where pregnancy was prolonged for 13 days beyond normal term by hypophysial damage.

**DISCUSSION**

Previous studies of foetal hypophysial function have been made either by observing the effects of decapitation or by examination of the newborn with congenital anomalies which include aplasia of the hypophysis. In either case the assumption was made that the absence of the hypophysis was the factor responsible for the observed changes. This assumption has been adequately tested in some instances by reversal of the effects of decapitation by injection of adenohypophysial hormones into the foetus. The present results which depend upon discrete hypophysial lesions add unequivocal support to the established framework of foetal endocrinology.

The wide range of disturbances of growth and development which follow interference with hypophysial function in the foetal lamb indicate that the foetal hypophysis may fulfil many of the functions attributed to it in postnatal life. Indeed, it subserves one vital function never again required of it—the initiation of parturition; for parturition in the sheep occurs in the absence of the maternal hypophysis (Denamur & Martinet, 1961) but not of the foetal hypophysis (Liggins et al. 1967).

With respect to somatic growth as judged by body weight, our results in three multiple pregnancies agree with the findings of Heggestad & Wells (1965) in the rat, that removal of the foetal hypophysis is followed by retardation of growth. Comparison of the birth weights of the single foetuses with those of the sham-operated ones is difficult, since the former group were delivered 7–37 days beyond term; absence of significant differences in birth weights despite the extended duration of intra-uterine life of the foetuses with lesions of the hypophysis does not indicate necessarily that a difference was present at term, since retardation of weight-gain might have occurred during the post-term period only. It is not possible from our experiments to determine the stage of gestation at which growth becomes dependent on endocrine influences or even to determine its dependence on adenohypophysial hormones at various stages of gestation after the 100th day. The observation that hypophysial lesions at 93–103 days lead to more marked retardation than lesions made later in pregnancy points to an effect at the earlier time. On the other hand, lambs which had prolonged gestation periods due to hypophysial lesions made after 130 days were not of excessive size, suggesting a growth-limiting effect of such lesions in late pregnancy. It seems that the hypophysis of the foetal lamb has an influence on growth throughout the last third of pregnancy at least.

Tibial length and volume were measured by Heggestad & Wells (1965) in the foetal
rat after decapitation but estimates of bone age have not been reported. Our results agree with those of Lascelles & Setchell (1959), who showed that in the foetal lamb osseous maturity is the most sensitive indicator of growth retardation. This was well illustrated in a foetus which was operated on at 93 days of gestation; when delivered 80 days later bone age had advanced only 17 days. The trophic hormones responsible for maintaining epiphysial development in the foetus are unknown and the present results offer no evidence in this direction. A rough correlation was present between the degree of retardation of epiphysial development, the reduction in weight of the thyroid and the plasma protein-bound iodine levels but no causal relationship could be established. Lascelles & Setchell (1959) observed delayed bone maturation in foetal lambs whose mothers were hypothyroid, suggesting that lack of thyroxine may have contributed to the retarded bone maturation in foetuses in our series. Growth hormone deficiency could also be involved, since Heggestad & Wells (1965) noted that injection of this hormone into decapitated foetal rats prevented the reduction in tibial length and volume which occurred in decapitated controls.

Our observations in the ruminant on the relationship between the foetal hypophysis and the development of the adrenals, thyroid and testes and its effect on carbohydrate metabolism agree with previous work in rodents and man as reviewed by Jost (1966) and Dawkins (1966). Likewise, disturbed lipid metabolism as demonstrated in the foetal lamb by the abnormal deposition of subcutaneous fat has been demonstrated previously in decapitated rat and rabbit foetuses by Picon & Jost (1963), who measured total body fat, and by Bearn, Antonis & Pilkington (1967), who determined plasma triglyceride and cholesterol levels in decapitated rabbit foetuses. Persistence of haemopoietic tissue in the liver following hypophysectomy in rabbit foetuses was suggested by unpublished observations of Jacquot (see Jacquot, 1965); in the foetal lamb this effect of hypophysectomy was clearly evident. Lack of mast cells in the skin has not been reported.

Dr Liggins carried out this work during the tenure of a Lalor Foundation Fellowship. The research was aided by grants GMS-05784 and HD-02299 from the National Institutes of Health and by a grant from the Association for the Aid of Crippled Children.

REFERENCES


G. C. Liggins and P. C. Kennedy


**DESCRIPTION OF PLATES**

**PLATE 1**

Fig. 1. X-ray of hind legs of twins delivered spontaneously at term. Leg A, from unoperated lamb, showing normal term epiphysial development. Leg B, from lamb with total destruction of the hypophysis at 95 days of gestation, had a bone age of 115 days.

Fig. 2. X-ray of hind legs of triplets delivered spontaneously at term. Leg A, from an intact lamb, showing normal epiphysial development. Legs B and C, from lambs with near-total hypophysial destruction at 124 days, had a bone age of 130 days.

**PLATE 2**

Figs. 3, 4. Adrenal of normal lamb at term (fig. 3) compared with that of twin with destruction of the hypophysis at 124 days of pregnancy (fig. 4). Medullary veins are in the same relative position with respect to medulla. (× 35.)

Figs. 5, 6. Detailed appearance of adrenal cortices of fig. 3. Note increased amount of cytoplasm in cells of normal zona fasciculata on left and hypoplastic fasciculata on right. (× 150.)

**PLATE 3**

Figs. 7, 8. Adjacent frozen sections of adrenal from lamb with destruction of the hypophysis. Sections in fig. 7 stained with haematoxylin-eosin, sections in fig. 8 incubated for 1 hr. to show 3β-hydroxysteroid dehydrogenase activity. The hypoplastic fasciculata shows strong, evenly distributed enzymic reaction. The glomerulosa shows a weak reaction. (× 150.)

Figs. 9, 10. Thyroid glands of twins at term: fig. 9 from an intact normal lamb; fig. 10 from a lamb with destruction of the hypophysis at 122 days of pregnancy. Note flattened, apparently inactive epithelium. (× 150.)

**PLATE 4**

Figs. 11, 12. Testes from twins at term: fig. 11 from an intact normal lamb; fig. 12 from lamb with destruction of the hypophysis at 93 days of pregnancy. Note loose, poorly differentiated interstitial tissue and small tubules. (× 150.)

Figs. 13, 14. Liver from twins at term: fig. 13 from intact normal lamb; fig. 14 from lamb with destruction of the hypophysis at 124 days of pregnancy. The small cells are haemopoietic elements. Note also foamy cytoplasm in normal liver cells with deposition of glycogen. (× 150.)

**PLATE 5**

Metatarsal epiphyses from twins at term

Figs. 15, 17. Unoperated normal lamb. (× 35 and × 150.)

Figs. 16, 18. Lamb with destruction of the hypophysis at 122 days of pregnancy. Growth of cartilage appears normal. Formation of bone trabeculae is greatly reduced. Comparable areas show inactivity of osteoblasts after destruction of the hypophysis. (× 35 and × 150.)
G. C. LIGGINS and P. C. KENNEDY