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The effect of intrauterine gonadectomy on fetal and neonatal gonadotrophin secretion in the lamb

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Summary. In Exp. 1, sheep fetuses (2 male and 3 female) were gonadectomized between Days 70 and 120 of gestation and their blood LH and FSH levels were compared to those of control animals (4 male, 6 female) at regular times during the first year after birth. In Exp. 2, fetuses (3 male and 5 female) were gonadectomized between Days 50 and 60 of gestation and gonadotrophin levels at Day 90 of gestation were compared to those of control fetuses of the same age. In neither study did the blood LH and FSH levels differ significantly between gonadectomized and intact lambs of either sex *in utero* or in the first week of life. However, by 2 weeks of age in the males and by 5 weeks of age in the females, gonadotrophin levels were higher in the gonadectomized than in the intact animals and this difference persisted throughout the first year of life. These results imply that fetal and neonatal gonadotrophin levels are suppressed by maternal and placental steroids in the sheep, and not by fetal gonadal hormones.

Introduction

It is well established that fetal thyroid and adrenal glands exert effects on the pituitary secretion of their respective trophic hormones (Hopkins, Wallace & Thorburn, 1975). By contrast, the effects of gonadal hormone production on pituitary gonadotrophin secretion within the fetus are not well known. Previous studies have investigated the patterns of serum gonadotrophins and gonadal steroids in normal lambs both *in utero* (Foster, Roach, Karsch, Norton & Nalbandov, 1972c) and from birth to sexual maturity (Lee *et al.*, 1976). The effects of neonatal gonadectomy on gonadotrophin secretion have also been reported (Foster, Cook & Nalbandov, 1972a).

In the present work, we performed intrauterine gonadectomy to determine whether this would lead to a change in serum gonadotrophin levels at birth and in the first year of life. In an additional study, we investigated whether intrauterine gonadectomy affected fetal gonadotrophin levels later in gestation.

Materials and Methods

Thirty-eight pregnant Merino–Corriedale crossbred ewes and their lambs were studied.

In Exp. 1, 6 fetuses (2 male, 4 female) were gonadectomized between Days 70 and 120 of

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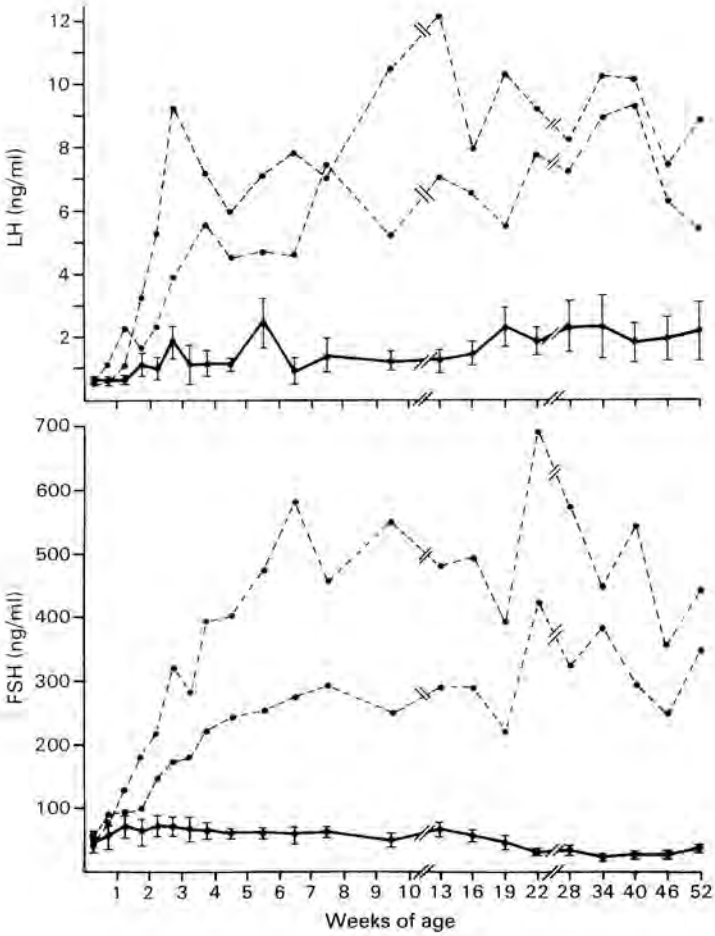
gestation, then allowed to go to term (145 days). Jugular venous blood samples (10 ml) were taken during the post-natal period, twice weekly during the first 4 weeks, once weekly during the second 4 weeks and at regular intervals thereafter during the first year of life. Blood samples were obtained at similar times from 10 normal control lambs (4 male and 6 female) born in the same flock within 1 week of the gonadectomized animals.

In Exp. 2, 8 fetuses (3 male and 5 female) were gonadectomized between Days 50 and 60 of gestation. Jugular venous blood samples (10 ml) from these animals and their mothers were obtained at Day 90 of gestation. Blood samples were also obtained from 14 intact fetuses (6 male and 8 female) and their mothers at Day 90 of gestation.

Surgery was performed under general anaesthesia (induction with thiopentone sodium and maintenance with halothane). The pregnant uterus was exposed through a midline lower abdominal incision and a second incision was made through the uterine wall to give access to the fetus. Gonadectomy was performed through bilateral lower abdominal or scrotal incisions, depending on the sex and age of the fetus. Survival to term of the gonadectomized fetuses was 35%. The cause(s) of fetal death was (were) not known. Plasma was frozen at −20°C until



Text-fig. 1. Plasma LH and FSH levels (mean ± s.e.m.) in normal male lambs (●——●, N = 4) and individual patterns for 2 male lambs (●– – –●) castrated at least 1 month before birth.

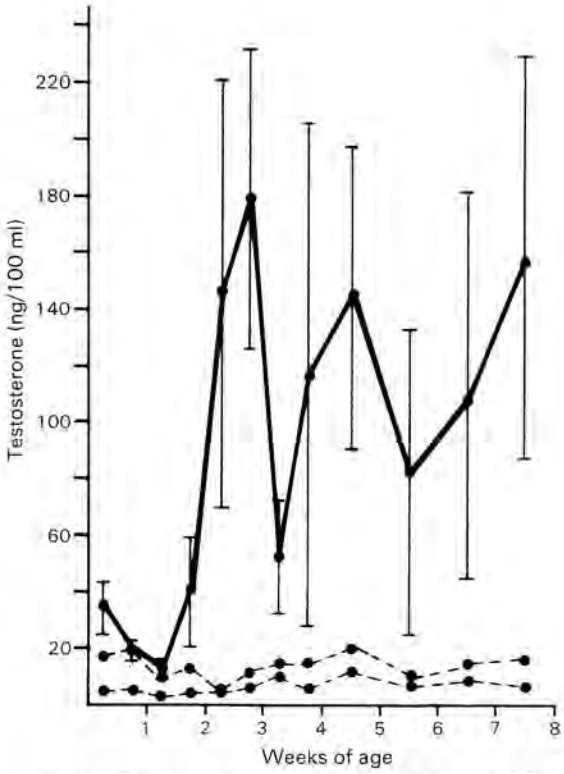
assay. The concentrations of luteinizing hormone (LH), follicle-stimulating hormone (FSH), testosterone and oestradiol were measured by previously described radioimmunoassays (Bremner, Findlay, Cumming, Hudson & de Kretser, 1976; Bremner, Findlay, Lee, de Kretser & Cumming, 1980).

Results and Discussion

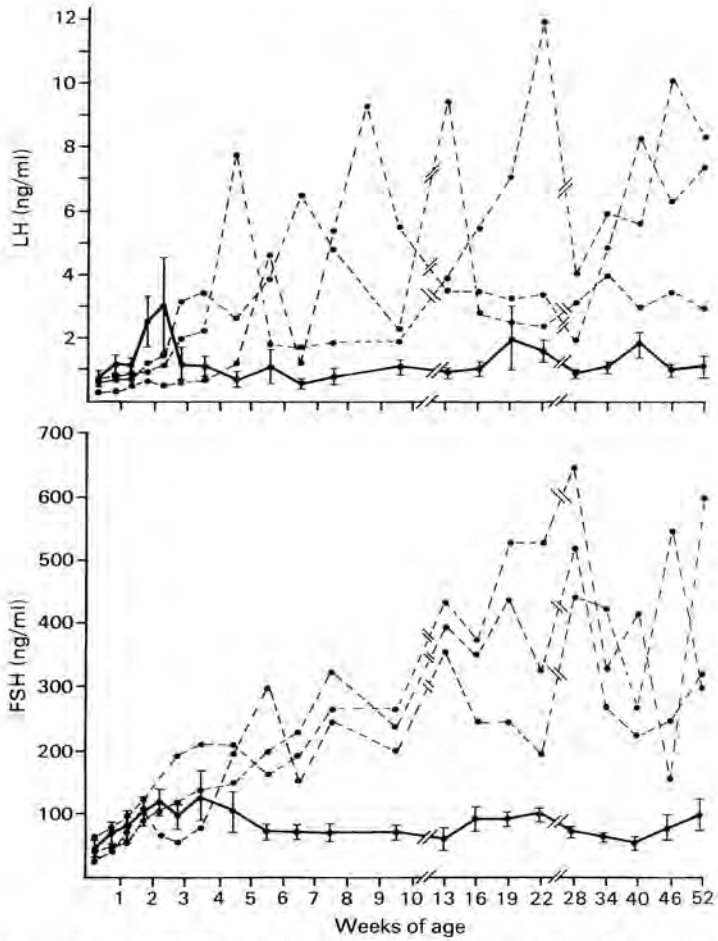
Effect of intrauterine gonadectomy on post-natal hormone levels

Among the male lambs castrated *in utero*, gonadotrophin levels during the first few days of life were indistinguishable from those of normal animals (Text-fig. 1). However, within 2 weeks after birth, both LH and FSH levels were elevated compared to controls and remained high for the first year of life. Increases of gonadotrophin levels, particularly LH, were also seen in the control animals during the first 3 weeks of life. This gonadotrophin rise stimulated an increase in testosterone levels in the normal animals beginning between 2 and 3 weeks of age (Text-fig. 2). Testosterone levels in the castrated males were consistently less than 20 ng/100 ml (Text-fig. 2).

In the females ovariectomized *in utero*, gonadotrophin levels for the first 3 weeks of life were indistinguishable from those of normal animals (Text-fig. 3). After 5 weeks of age, gonadotrophin levels were clearly higher in the ovariectomized than in the intact animals, although occasional single values overlapped between the two groups, particularly in the case of LH. As with the males, the intact females showed increased gonadotrophin levels in the first 3 weeks of life. This



Text-fig. 2. Plasma testosterone levels (mean ± s.e.m.) in normal male lambs (●——●, N = 4) and individual lambs (●– – –●) that had been castrated at least 1 month before birth.



Text-fig. 3. Plasma LH and FSH levels (mean ± s.e.m.) in normal female lambs (●——●, N = 6) and in 3 female lambs (●– – –●) that had been ovariectomized at least 1 month before birth.

increase is presumably important in stimulating the marked increase in ovarian size that occurs in the first 4 weeks of life in the lamb (Kennedy, Worthington & Cole, 1974). Oestradiol levels, in the ovariectomized and in the intact animals, were all below the sensitivity of the assay (12 pg/ml).

These results demonstrate clearly that gonadotrophin concentrations in the first week after birth do not differ between normal animals and those that have been gonadectomized at least 1 month before birth. Following this time, and occurring somewhat earlier in males than in females, LH and FSH levels in gonadectomized animals become consistently higher than those of intact lambs. The time course of the gonadotrophin increase in animals gonadectomized *in utero* is quite similar to that reported for animals gonadectomized immediately after birth (Foster *et al.*, 1972a).

The relatively low concentrations of gonadotrophins both in intact and gonadectomized animals in the first few days after birth could be due either to (a) immaturity of the hypothalamus and pituitary resulting in an inability to secrete more gonadotrophin or (b) suppression of the hypothalamus and pituitary by placental and maternal steroids. The second of these possibilities seems to be the more likely. Oestrogens and progesterone are produced in large amounts by the ewe and by the placenta in late pregnancy (Thompson & Wagner, 1974). Higher blood levels of

gonadotrophins may be found in fetal animals than in neonates (Foster *et al.*, 1972c) and fetal pituitaries are responsive to LH-releasing hormone (Foster, Cruz, Jackson, Cook & Nalbandov, 1972b). Gonadotrophin secretion in the ovine fetus in late gestation can be suppressed by gonadal steroid administration (P. D. Gluckman, C. Marti-Henneberg, S. L. Kaplan, A. M. Rudolph & M. M. Grumbach, personal communication). It is likely that, following clearance of maternal and placental steroids after birth, gonadotrophin levels rise in the neonate, stimulating gonadal hormone production. These gonadal hormones become the regulatory negative feedback signal controlling LH and FSH secretion by 2 weeks of age in the males and by 5 weeks of age in the females.

Effect of intrauterine castration on pre-natal levels of gonadotrophins

This study was undertaken to determine whether fetal gonadectomy relatively early in gestation (50–60 days) would lead to increased gonadotrophin levels later in pregnancy (90 days). The results in Table 1 indicate that there was no difference in LH and FSH levels in the gonadectomized animals of either sex when compared to values for the intact fetuses ($P > 0{\cdot}05$). Both LH and FSH were higher in female fetuses than in males. The fact that no differences in gonadotrophin levels were found in the gonadectomized fetuses suggests that the gonads are not important in controlling gonadotrophin secretion in fetuses of this age. This conclusion must be regarded as preliminary, however, because of the small number of animals involved in this study and particularly because of the fact that only one blood sample was taken from each fetus. It seems likely, however, that if a difference in gonadotrophin levels in gonadectomized and intact animals is present, it is small. In addition, the variability of the hormone levels found raises the possibility that pituitary gonadotrophins may be secreted episodically in the sheep fetus.

Table 1. Effect of fetal gonadectomy (Days 50–60) on mean ± s.e.m. concentrations of plasma LH and FSH at Day 90 of gestation

	LH (ng/ml)		FSH (ng/ml)	
	Fetal	Maternal	Fetal	Maternal
Intact males ($n = 6$)	2·0 ± 0·7	1·9 ± 0·2	37 ± 8	53 ± 4
Castrated males ($n = 3$)	3·2 ± 0·2	2·1 ± 0·4	45 ± 6	38 ± 7
Intact females ($n = 8$)	10·7 ± 2·1	2·2 ± 0·2	115 ± 13	74 ± 11
Ovariectomized females ($n = 5$)	10·3 ± 2·2	3·1 ± 0·7	109 ± 36	53 ± 5

The results of our studies suggest that the control of fetal pituitary gonadotrophin secretion may differ from that for thyrotrophin and adrenocorticotrophin. Removal of the target glands for TSH and ACTH, the thyroid and adrenal cortex, respectively, leads to increased blood levels of the pituitary hormones (Hopkins *et al.*, 1975). The present results suggest that removal of the gonads has little effect on gonadotrophin levels in ovine fetuses of either sex.

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