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Metabolic changes in ewes shorn during mid-pregnancy

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ABSTRACT

Previous work has shown that, under appropriate circumstances, mid-pregnancy shearing results in an increase of both the birth and weaning weights of multiple-born lambs. However, the physiological mechanisms for this response are poorly understood. This paper attempts to clarify the link between mid-pregnancy shearing and heavier lamb weights by investigating the metabolic changes that occur in ewes that are shorn in mid-pregnancy.

Twenty-four twin-bearing ewes were assigned to one of two treatment groups; mid-pregnancy shorn (70 days after the mid-point of mating: P70) or unshorn. Groups were balanced for ewe age and weight. Blood samples were collected from all animals on days P53, P69, P81, P84, P90, P109 and P130, for measurement of concentrations of triiodothyronine (T3), thyroxine (T4), cortisol, glucose, non-esterified fatty acids (NEFA) and ,-hydroxybutyrate. Lamb birth weight, girth and crown-rump length (CRL) measurements were collected within 24 hrs of birth. Lambs were weighed at weaning. Mid-pregnancy shearing increased lamb birth weights and girths by 0.43 kg ($P<0.05$) and 2.18 cm ($P<0.05$) respectively although these differences were not significant once adjusted for date of birth. There was no effect on CRL. Plasma concentrations of T3 and T4 were significantly higher in mid-pregnancy-shorn ewes relative to unshorn ewes at both P81 ($P<0.01$) and P84 ($P<0.01$ and $P<0.05$ respectively) and T4 concentrations tended to be higher at P90 ($P<0.10$). There was a significant effect of shearing treatment at P81 on plasma NEFA concentrations ($P<0.05$). There were no significant post-shearing differences in mean cortisol, glucose or β-hydroxybutyrate concentrations. The study proposes that the most likely mechanism by which mid-pregnancy results in higher lamb birth weights is by increasing maternal thyroid hormone concentrations, resulting in an increase in fat mobilisation causing increased NEFA concentrations in the maternal blood supply, which in turn improves placental nutrition and, thus, lamb birth weight.

Keywords: mid-pregnancy shearing; birth weight; thyroxine; cortisol; NEFA; metabolism.

INTRODUCTION

Shearing pasture-fed ewes during the mid-pregnancy period has been found to increase lamb birth weights (Morris & McCutcheon, 1997; Morris et al., 2000; Kenyon et al., 2002ab), however shearing ewes during winter exposes ewes to conditions that may cause hypothermia and death. It would be desirable to obtain the birth-weight response without exposing ewes to this risk. In order to mimic the birth-weight response due to mid-shearing an understanding of the mechanism responsible is required.

Previous studies have tried to link the birth-weight response to increases in dam intake and gestation length with little success (Kenyon et al., 2002ab). Dabiri et al. (1996) and Jopson & Davis (2000) both reported that back fat depths were lower in shorn ewes during late pregnancy than in their unshorn counterparts, which may indicate that the dam's body reserves were being mobilised, resulting in enhanced fetal growth. Similarly, Kenyon et al. (2002b) suggested that the dam needs to have adequate body condition, prior to pregnancy shearing, if a birth-weight effect is to be observed. If the dam's body reserves are a source of extra energy for increased fetal growth, a change in the dam's metabolism must occur.

Glucose is the central fuel of energy metabolism and plasma concentrations are known to rise in response to cold stress (Thompson et al., 1982). As reserves of glucose are depleted, the animal begins to mobilise fat reserves to meet energy requirements. Increased plasma concentrations of non-esterified fatty acids (NEFA) and β-hydroxybutyrate are indicators of fat mobilisation. Shearing is known to increase cortisol concentrations (Pierzchala et al., 1983). An increase in cortisol concentration can result in increased blood glucose concentrations and increased rates of lipolysis in adipose tissue. Thyroid hormones increase oxygen consumption of tissues, affect carbohydrate metabolism and lipid metabolism (Greco and Stabenfeldt, 1997)

The majority of the research on the effect of pregnancy shearing on ewe metabolites and hormones has been undertaken under housed conditions (Aulie et al., 1971; Symonds et al., 1986; 1989; 1992; Revell et al., 2000; Black & Chestnutt, 1990; Vipond et al., 1987) which are likely to be different from that experienced under pastoral conditions where the pregnant ewe can be exposed to cold, wet and windy conditions. Furthermore, the results of these indoor studies have been inconsistent in the responses observed (for example Aulie et al. (1971) reported a rise in NEFA post-shearing in contrast to Symonds et al. (1986; 1992). Similarly, Black & Chestnutt (1990) reported a rise in glucose concentrations post shearing, but Symonds et al. (1986; 1992) failed to observe a change in glucose concentration.). In addition, individual studies have often only examined one or two hormones or metabolites.

Under pastoral conditions, Morris et al. (2000) observed both higher maternal glucose concentrations and elevated maternal triiodothyronine (T3) concentrations in shorn ewes over a 20-day period post mid-pregnancy shearing. Elvidge & Coop (1974) reported that shorn ewes had elevated plasma NEFA concentrations in comparison to their unshorn counterparts.

There have been no trials, to these authors' knowledge, in which either β-hydroxybutyrate or cortisol concentrations have been measured in mid-pregnancy-shorn ewes under pastoral conditions or where glucose, NEFA, T3 and T4 have been measured within one study.

Therefore, the aim of this study was to examine the effects of mid-pregnancy shearing on plasma glucose, NEFA, β-hydroxybutyrate, cortisol and thyroid hormone concentrations in ewes under pastoral conditions to try to identify a possible mechanism for the lamb birth-weight effect.

MATERIALS AND METHODS

Experimental design and animals

Thirty twin-bearing ewes that conceived over a 20-day period were randomly selected post-scanning (49 days after the mid-point of mating, P49) from a commercial flock of Coopworth ewes mated to Coopworth rams. On P52, ewes were randomly allocated (balanced for live weight and age) into one of two treatment groups (n = 15 in each group); shorn at P70 or left unshorn (with approximately six months of wool growth at P70).

From P50 until P131, all ewes were grazed under commercial conditions as one group. At P131, ewes were randomly allocated to one of three paddocks for the lambing period.

Animal measurements

Ewes

Ewes were weighed (unfasted) at P49, P84, P109, P130 and at weaning (P249). Blood samples (10ml) were taken via jugular venepuncture (Lithium Heparin vacutainer, Becton Dickinson Vacutainer Systems, USA) from all ewes on P53, P69, P81, P84, P90, P109 and at P130. Blood samples were placed immediately on ice and centrifuged at 3500 rpm for 15 minutes within an hour of collection. The plasma was harvested and stored at below -18°C until analysis. Plasma concentrations of T3, T4 and cortisol were analysed by radioimmunoassay using diagnostic kits (DiaSorin, Minnesota, USA). Plasma concentration of ,-hydroxybutyrate and NEFA were analysed using enzymatic assays (Sigma, Illinois, USA and Wako Pure Chemical Industries Ltd, Osaka, Japan respectively). Glucose plasma concentrations were analysed using a hexokinase assay (Roche Diagnostics Ltd, Switzerland).

Lambs

Lambs were identified to their dams, tagged, weighed and had crown rump length (CRL) and girth measurements taken within 24 hours of birth. Live weights of lambs were recorded again at weaning.

The trial was undertaken with the approval of the Massey University Animal Ethics committee.

Data analysis

Comparative least-squares means between groups were estimated for parameters measured on the lambs using the Generalised Linear Model procedure of the statistical package 'MINITAB' (Minitab, 2000). Sex of the lamb was used as a fixed effect in the model used to partition variation in birth weight and date of birth (paddock, and age of dam were tested and removed from the model because they were non-significant). Data are presented as least-squares means and standard errors.

Dam plasma parameters and ewe live weight were analysed using repeated measures using the Mixed procedure of 'SAS' (SAS, 2001**)**. Data are presented as least-squares means and standard errors. For all parameters, except ewe live weight, the first measurement on P53 was used to adjust for pre-experimental variation between the ewe treatment groups.

RESULTS

Lamb measurements

Shearing dams at mid-pregnancy increased lamb birth weights by 0.5 kg compared to unshorn ewes (Table 1). Lambs born to shorn ewes were born 4.5 days later on average than those born to unshorn ewes. Similarly there was a 2.2 cm increase in lamb girth ($P<0.05$), due to mid-pregnancy shearing dams, until date-of-birth effects were taken into account. Mid-pregnancy shearing had no effect on either CRL (Table 1) or weaning weight (Table 2).

Ewe live weights increased by approximately 5 kg (from 59 to 64 kg) between P49 and P130. After accounting for the weight of fleece removed from the shorn ewes, mid-pregnancy shearing had no effect on ewe live weight at any stage during pregnancy.

At day P69 (the day before shearing), sheep in the shorn group tended to have lower plasma concentrations of glucose ($P=0.08$) than their counterparts in the unshorn group (Figure 1A). Eleven days after shearing, plasma glucose concentrations tended to be higher in the shorn group ($P=0.11$). No treatment differences were noted thereafter.

Plasma NEFA concentrations were increased by mid-pregnancy shearing at P81 ($P<0.05$), but by 20 days post-shearing there was no difference between treatment groups (Figure 1B). There were no differences between treatment groups in plasma concentrations of β-hydroxybutyrate in response to mid-pregnancy shearing (Figure 1C).

Plasma cortisol concentrations of the unshorn ewes were higher ($P<0.05$) than those of their shorn

TABLE 1: The effect of mid-pregnancy shearing on the birth weight (BWt), crown rump length (CRL) and girth of twin lambs (least-squares means ±SEM).

Dam Treatment	(n)	BWt(kg)	DoB[1](days)	Girth(cm)	Girth Adj[1](cm)	CRL(cm)
Unshorn	20	4.7 ±0.14	9.5 ±1.1	40.5 ±0.81	41.4 ±0.73	53.9 ±0.74
Shorn	28	5.2 ±0.11	14.1 ±1.3	42.7 ±0.66	42.1 ±0.59	54.0 ±0.61
Signif.	*	*	*	*	NS	NS

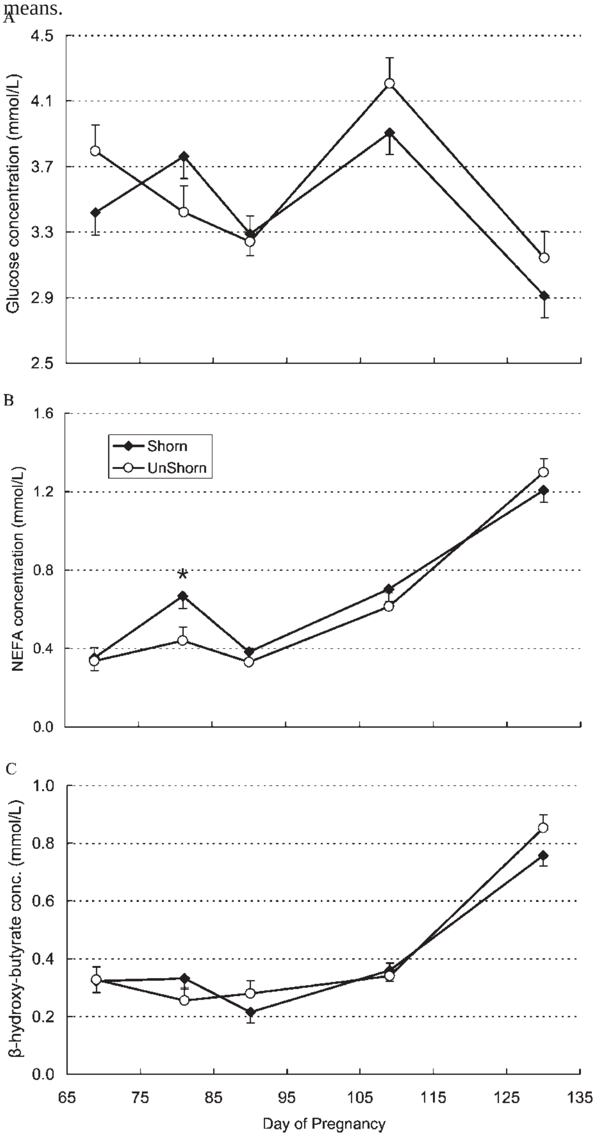
[1]Calculated as the number of days from when the first lamb was born.

TABLE 2: The effect of mid-pregnancy shearing on the weaning weight (WWt) of twin lambs (least-squares means ±SEM).

Dam Treatment	(n)	WWt(kg)	WWt Adj[1](kg)
Unshorn	19	26.2 ±0.85	25.6 ±0.85
Shorn	22	24.6 ±0.77	25.1 ±0.76
Signif.		NS	NS

[1]Adjusted for the effect of birth date.

FIGURE 1: Effect of dam treatment (mid-pregnancy shorn, solid diamonds; unshorn, open circles) on maternal plasma concentrations of glucose (A), non-esterified fatty acids (NEFA) (B) and β-hydroxybutyrate (C). The vertical bars indicate standard errors of the means.



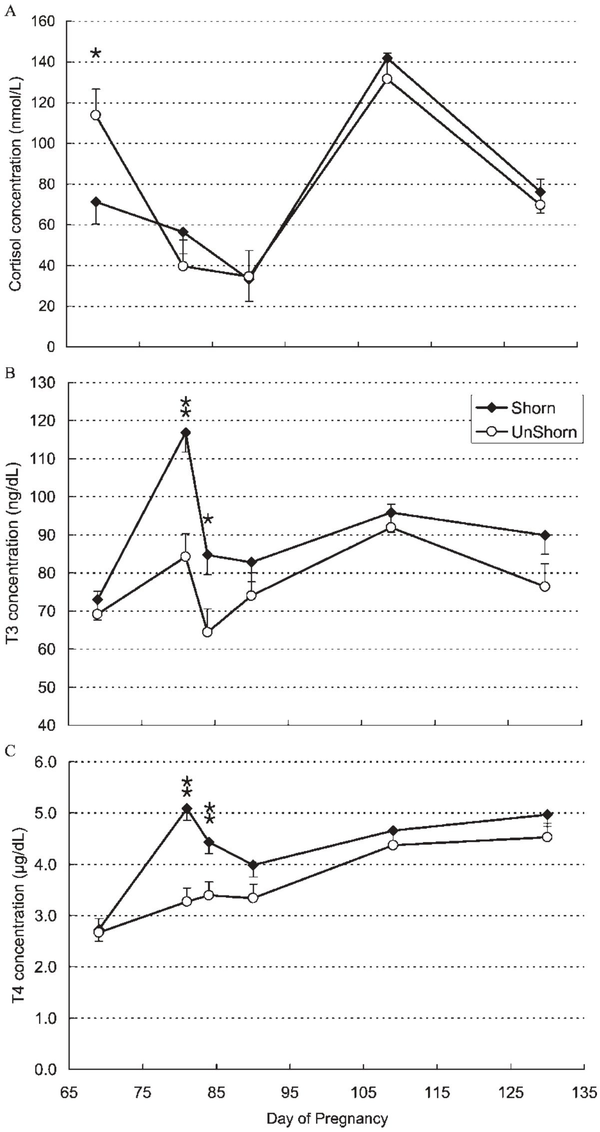
counterparts the day before shearing at P69 (Figure 2A). There were no significant treatment differences in cortisol concentration post shearing.

Plasma concentrations of both T3 and T4 were elevated by mid-pregnancy shearing at days P81 ($P<0.01$) and P84 ($P<0.05$ and $P<0.01$ respectively) and T4 concentrations tended to be higher at P90 ($P=0.07$) (Figures 2B and 2C).

DISCUSSION

Lambs born to mid-pregnancy shorn dams were 0.5kg heavier at birth. Because individual tup marks were not recorded in the present study, it was not possible to

FIGURE 2: Effect of dam treatment (mid-pregnancy shorn, solid diamonds; unshorn, open circles) on maternal plasma concentrations of cortisol (A), triiodothyronine (T3) (B) and thyroxine (T4) (C). The vertical bars indicate standard errors of the means.



determine whether there was a difference in gestation length due to shearing treatment. Therefore, whether the observed difference in birth weight was due to increased gestation length, increased growth rates from conception to birth, or a combination of both is unclear. However, the aim of this paper was to identify metabolic differences between shorn and unshorn ewes and not to re-establish that mid-pregnancy shearing can increase lamb birth weight per-se.

Previous studies have found no increase in intake following mid-pregnancy shearing (Husain et al., 1997; Revell et al., 2002) perhaps due to rumen restriction in later stages of pregnancy (Forbes, 1968). However, improved foetal growth is unlikely to occur without improved foetal nutrition. Given the pivotal role of glucose in energy metabolism and that it is readily transferred from the dam, across the placenta, to the foetus, glucose is a strong candidate for explaining increased foetal growth. However, in the current study, plasma glucose concentrations did not differ between shorn and unshorn ewes. Symonds et al. (1986; 1992) also reported no effect of pregnancy shearing on maternal glucose concentrations, despite the fact that they observed a birth-weight response. This indicates that a birth-weight response to mid-pregnancy shearing is not necessarily dependent on an increase in glucose concentrations.

Symonds et al. (1989) reported increased rates of lipolysis and NEFA oxidation in pregnancy-shorn ewes and Jopson et al. (2002) found that mid-pregnancy-shorn ewes lost more body fat than their unshorn contemporaries and subsequently gave birth to heavier lambs. In the present study NEFA concentrations were elevated in shorn ewes at P81, but by P91 had returned to the levels observed in unshorn ewes. This suggests that shorn ewes could have been mobilising fat reserves for a considerable proportion of the 20 day period from shearing until P90. Given that this period is also characterised by rapid placental growth (Ehrhardt & Bell., 1995) and that placental size is proportional to birth weight (Dingwall et al., 1987), an increase in available nutrients may help explain any increase in birth weight of lambs born to mid-pregnancy-shorn ewes.

Both NEFA and β-hydroxybutyrate levels increased substantially in late pregnancy, indicating that the ewes were in energy deficit. There were, however, no significant treatment differences in either NEFA or β-hydroxybutyrate concentrations, indicating that any effect of mid-pregnancy shearing on these metabolites was not prolonged through late pregnancy.

Cortisol and thyroid hormones are both known to increase fat breakdown that would lead to an increase in NEFA concentrations. In the current study, there were no differences between the post-shearing plasma concentrations of cortisol, which implies that cortisol was not responsible for the post-shearing increase in NEFA concentrations. However, the first blood sample analysed post shearing was after 11 days and may have been outside the critical window to observe a difference in cortisol due to mid-pregnancy shearing. In future studies, it would be prudent to measure concentrations of cortisol closer to mid-pregnancy shearing. Plasma concentrations of cortisol (and glucose) were significantly higher in the unshorn group pre-shearing at P69. It is difficult to explain the difference between the groups at this time, given that they were managed together.

Of all the hormones and metabolites measured in the current study, levels of T3 and T4 showed the largest and most prolonged response to mid-pregnancy shearing. A direct response in foetal growth due to maternal T3 and T4 is unlikely because they do not cross the placenta (Fisher et al., 1977). An increase in the circulating concentration of maternal thyroid hormones may however affect lamb growth by changing nutrient availability. Symonds et al. (1989) proposed that higher plasma concentrations of T3 and T4 could be responsible for increased NEFA oxidation & lipolysis. Pierzchala et al. (1983) found a rise in T4 concentrations in non-pregnant shorn animals that seemed to be dependent on the level of cooling. Given that the thyroid hormones increase oxygen consumption in the tissues and, hence, increase heat production, the increased levels of T3 and T4 observed in mid-pregnancy shorn ewes may well be a response to cold stress. Increased T3 and T4 concentrations have been previously found in mid-pregnancy-shorn ewes, both indoors (Symonds et al., 1989) and under pastoral conditions (Morris et al., 2000). However a birth-weight response is not always observed (Symonds et al., 1989; Morris et al., 2000). Morris et al. (2000) explained the absence of a birth-weight response in twin lambs by suggesting that the lambs born to unshorn ewes in that study were already close to their potential birth weight under the nutritional environment experienced. The lack of a consistent relationship between elevated maternal T3 and T4 and increased lamb birth weights, suggests that other factors must also be present for a birth-weight response to occur.

CONCLUSION

It is possible that the higher maternal thyroid hormone concentrations observed resulted in an increase in fat mobilisation causing increased NEFA concentrations in the maternal blood supply, which in turn could improve placental nutrition and, thus, lamb birth weight.

If this is indeed the mechanism by which mid-pregnancy shearing increases lamb birth weights, then further work should focus on why maternal T4 and T3 concentrations increase in response to mid-pregnancy shearing. More work is also required to examine the other prerequisites, in addition to elevated thyroid hormones, necessary to obtain an increase in the birth weight of lambs born to mid-pregnancy shorn ewes.

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