EFFECT OF MATERNAL NUTRITION DURING EARLY AND MID-GESTATION ON FETAL GROWTH

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ABSTRACT

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It is generally assumed that, because the fetus has a small nutritional requirement relative to that of the dam in early gestation, differential maternal nutrition at this time is unlikely to influence fetal growth. Hence the dogma that females in early gestation need not be provided with nutrients additional to those required by comparable nonpregnant females. However, the effects of nutrition during early gestation on fetal and placental growth and development, and hence birth weight, have not been studied extensively. Nor does the current dogma take into account the fact that the placenta has a different pattern of growth from the fetus. Hence this study sought to further examine the effects of maternal nutrition during early and mid-gestation on placental and fetal growth.

The first study was conducted over two years, with crossbred heifers managed for High (H, 0.6 kg/day) or Low (L, 0.1 kg/day) liveweight gains from mating until day 140 of gestation. Treatments were then reversed so that effects of nutrition during early gestation were not confounded by differences in maternal live weight at calving. Averaged across years, maternal live weights (kg, Mean±SE, n=60) were (H vs L) 393.5 ± 4.3 vs 362.1 ± 4.3 ($P<0.001$) at day 140 of gestation and 417.6 ± 4.5 vs 408.7 ± 4.3 ($P>0.05$) at term. Calf birth weights were 31.1 ± 0.5 vs 31.4 ± 0.5kg and weaning weights (average calf age = 90 ± 14.4days) were 91.3 ± 2.0 vs 89.7 ± 2.0 (both $P>0.05$).

A second study using breeding ewes was designed to determine more directly whether early placental development and fetal growth may be modulated by nutritional manipulation of the female during early and mid-gestation, and so eliminated the potential for the dam to compensate for earlier treatments during late gestation.

Mixed-age Romney ewes (average live weight 54.5 ± 0.4 kg), pregnant to a synchronised oestrus, were allocated to three nutritional treatment groups (n = 20/group), Low (L = 0.5 maintenance (M)), Control (C = 1.0M) and High (H = 1.5M) from days 21 to 101 of gestation. Maintenance requirements for a 50 kg ewe were assumed to be approximately 0.9 kg DM/ewe/day (10 MJ ME/day) at a concentration of 11 MJ ME/kg DM. Ewes were weighed weekly, slaughtered at the end of treatment, and fetal and placental measurements recorded.

Live weights were significantly ($P<0.001$) different at slaughter (L, 45.8 ± 1.4 kg; C, 56.8 ± 1.4 kg; H, 69.1 ± 1.4 kg). At day 101 of gestation, measures of fetal and placental growth and development were (C vs H group): Uterus (minus fetus and fluids) (1290.7 ± 67.0 vs 1475.4 ± 64.8 g, $P<0.05$); fetal weight (1280.8 ± 38.0 vs 1379.8 ± 35.2 g, $P<0.05$); total placentome weight (631.0 ± 30.7 vs 702.9 ± 29.7 g, $P<0.01$) and total placentome number (102.6 ± 3.2 vs 93.4 ± 3.1 g, $P<0.05$). Low levels of maternal nutrition did not significantly influence these parameters (L vs C).

It is concluded that high levels of maternal nutrition in early and mid-gestation enhance fetal and placental growth and development in sheep, while low levels are without effect compared to ewes fed at maintenance. Thus there may be advantages to high levels of maternal nutrition in early and mid-gestation though the possible effects of compensation in later gestation, as may have occurred in the beef cow trial, are yet to be studied in sheep.

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°C	degrees celsius
%	percentage
µg	microgram
C	control (group)
CIDR	controlled internal drug releaser
cm	centimetre
CRL	crown rump length
d	day
DM	dry matter
DOP	dressing out percentage
g	gram
H	high (group)
kg	kilogram
L	low (group)
LWT	live weight
ME	metabolisable energy
MJ	megajoules
mm	millimetre
TGUW	total gravid uterus weight
vs	versus

Statistical:

n	number of experimental units
NS	non-significant (P>0.10)
SE	standard error of the mean

CHAPTER ONE

INTRODUCTION

PREAMBLE

This thesis addresses the extent to which placental development, fetal growth and birth weight may be manipulated in farm species by altering maternal nutritional levels during early to mid-gestation (days 0 to 100 in sheep, 0 to 140 in cattle).

The studies described herein were conducted because the literature examining the effects of nutrition during early to mid-gestation on placental development and birth weight in sheep and cattle is sparse and inconclusive. This study is potentially relevant to New Zealand farmers and the nutritional management of their ewes and/or beef cows throughout gestation.

Chapter One summarises the existing literature on the effects of maternal nutrition during gestation, and the ways in which placental and fetal growth, and hence birth weight, might be manipulated by way of maternal nutrition during early and mid-gestation.

PLACENTAL DEVELOPMENT

Introduction

The placenta plays a critical role in providing an environment by which the fetus receives metabolic substrates for growth throughout gestation. Therefore, growth and development of the functional ability of the placenta is vital to the fetus (McCrabb *et al.,* 1992; Anthony *et al.,* 1995). Placental weight alone is not an absolute determinant of placental function, as its surface area, vascularity and permeability continue to increase after maximal weight has been achieved (Kulhanek *et al.,* 1974). The placenta performs its role by providing the site of nutrient transfer from the dam to the fetus, and waste secretion from the fetus to the dam, as well as acting as a barrier against pathogens and having an endocrine function. These roles are determined by a placentas' size, functional capability, rate of blood flow and differences in concentrations of metabolites between the fetal and maternal circulation. The mechanism(s) by which each of these components are regulated is unknown, although it is understood that physical, hormonal and metabolic mechanisms are controlling factors (Kelly, 1992).

By definition, the placenta is the region of contact between chorion and maternal tissues (Ferrell and Reynolds, 1992), and is an active endocrine organ, capable of synthesising and secreting a broad range of protein, steroid and other hormones and growth factors (Anthony *et al.,* 1995). The placentae of sheep and cattle consist of a variable number of highly vascular structures called cotyledons. These are located on the chorionic membrane, and each is attached to a specific area of the uterine epithelium, normally referred to as cotyledonary burr, or caruncle (Robinson, 1982). It is known that there is substantial variation between individuals in the extent of development of placental mass and function, although factors controlling placental development, in biochemical or endocrine terms, are not fully known (Davis, 1983).

Stages of placental development

The cotyledonary placenta of the sheep is formed by the attachment of the chorionic membrane to specific endometrial thickenings, termed 'caruncles', on the uterine wall between days 20 to 30 of gestation. The non-pregnant sheep uterus contains between 60 and 150 caruncles (Alexander, 1964a) and, although numbers vary between species, breeds and strains, not all of these sites are occupied by the fetal cotyledons. The proportion of caruncle occupancy is widely variable, with an approximate 70% occupancy rate if the ewe carries a single lamb, and over 80% with twin lambs (Davis, 1983). The points of attachment develop into bulb- or button-shaped placentomes, which are essentially the fusion of the fetal cotyledons and the maternal caruncles, and which together constitute the placenta. When the maternal and fetal components of fresh placental tissue are separated manually, the cotyledonary portion, which probably contains fragments of caruncle tissue, contributes about 80% to the weight of each placentome (Mellor and Murray, 1981). As gestation proceeds, the bulb shape of the placentome changes to a flat button shape as the fetal tissue envelops the maternal tissue

(Kelly, 1992), and placental functions are largely executed at these highly vascularised sites. In all of the species for which data are available, placental growth proceeds rapidly and exceeds fetal growth rate until mid-gestation (e.g. day 90 to 100 of gestation in the sheep) (Robinson, 1982; Davis, 1983; Ferrell, 1989). In sheep, maximum cotyledonary size and weight is reached between days 70 to 80 of gestation after which it slowly declines by up to 20%, while fetal weight continues its exponential increase until term at days 145 to 150 (see Figure 1). While placental formation is similar between species, there is a distinct lack of data on the development of the bovine placenta.

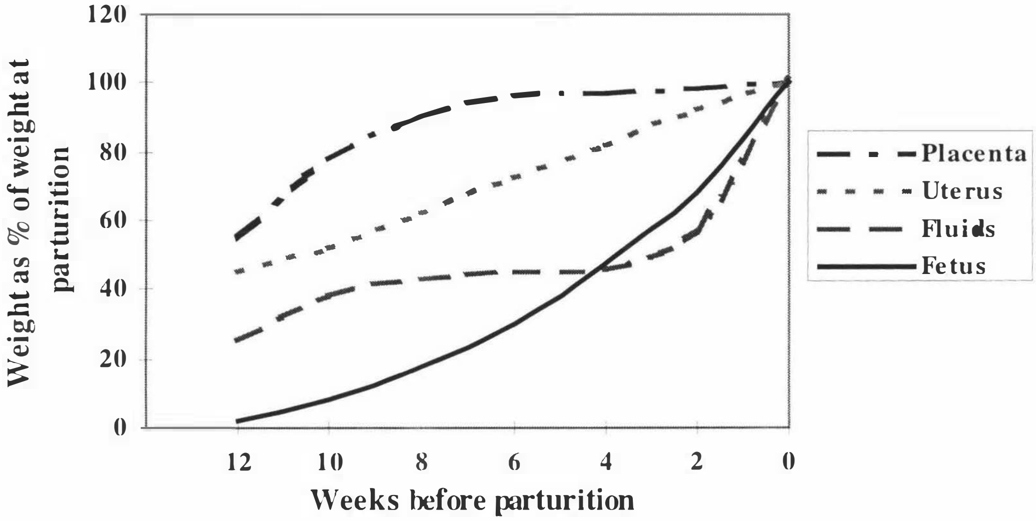

Figure 1: **Weight growth curves for the products of conception (Robinson *et al.*, 1977, p. 546).**

Individual cotyledon weights range from 0.1 to 45 g in the ewe during late gestation, with the total cotyledon weight from 'twin' pregnancies being 30-50% greater than the weight of those from a 'single' pregnancy. This indicates a considerable variation in cotyledon weight within pregnancy rank (Davis, 1983), although the placenta accounts for approximately 12 % of the total fetal weight at term across all birth ranks (Robinson, 1977). Uterine fluids continue to increase in weight after day 90 of gestation, with substantial increases occurring in the last 20 to 30 days prior to term. In contrast, the total fluid weight in relation to total fetal weight increases with increasing birth rank, being approximately 32, 36 and 40% for twins, triplets and quadruplets at parturition respectively (Robinson, 1977).

Placental exchange

The blood of the fetus and the dam never come into direct contact, yet the two circulations are close enough at the junction of the chorion and the endometrium for oxygen and nutrients to pass from maternal to fetal blood (Jainudeen and Hafez, 1980). The placenta permits the transport of sugars, amino acids, vitamins and minerals to the fetus as substrates for fetal growth (Jainudeen and Hafez, 1980), of which the major substrates for fetal growth and energy production by the pregnant uterus are oxygen, glucose and amino acids (Battaglia and Meschia, 1978). The placenta not only transfers oxygen and glucose to the fetus, but also converts glucose to lactate which is then released to the fetal and maternal circulations (Sparks *et al.,* 1983). During mid-gestation in sheep, fetal mass is less than half that of the placenta, and the utero-placental tissues consume more than 80% of the oxygen and glucose taken up by the pregnant uterus (Bell *et al.,* 1986). This demand by the utero-placental tissues reflects the growth curves of the placenta and fetus shown in Figure 1, where relative fetal growth is minimal during early gestation. However, during late gestation, fetal mass is four to six times greater than that of the placenta, but the placenta still accounts for 40 to 60% of uterine consumption of oxygen and glucose (Sparks *et al.,* 1983). Therefore, the placenta competes with the rapidly growing fetus during late gestation for substrates to support essential placental functions such as hormone production. However, while placental exchange mechanisms are subject to pathological alteration, they do not appear to be the mechanisms by which fetal growth is physiologically regulated (Gluckman and Liggins, 1984).

Factors affecting placental size

In view of the predominant influence of the placenta on fetal growth, factors affecting placental weight are of considerable interest, particularly as placental weight varies widely among uniformly treated animals (Alexander, 1964a; Alexander and Williams, 1971; Mellor and Murray, 1981; Mellor and Murray, 1982a), although the basis of this variation is poorly understood (Bell, 1984).

Endocrine factors

Mellor (1983) found very low levels of receptors for progesterone and oestradiol in maternal and fetal tissues between days 56 and 112 of gestation, relative to those in caruncular endometrium on day 0 of gestation. Steroid receptor levels were not related to the time-course of placental cellular growth or protein synthesis, suggesting that neither progesterone nor oestradiol has an important effect on these processes. Maternal corticosteroids may affect implantation and placental development; stressful husbandry practices or experimental treatment of the ewe with hydrocortisone or adrenocorticotrophin during the first 20 days of gestation increase embryo mortality (Doney *et al.,* 1976)

Nutritional and environmental factors

Maternal nutrition during gestation has been shown to have an effect on placental size, although the extent to which size is affected appears to depend on the stage of gestation and the level of nutrition. Some studies have suggested that the nutritional effect on placental growth is most prevalent in mid-gestation, while others examining nutrition levels during early (El-Sheikh *et al.,* 1955; Foote *et al.,* 1959; Parr *et al.,* 1986) and late (Mellor, 1983; Faichney and White, 1987; Holst *et al.,* 1992) gestation, reported no influence on placental size. However, other studies examining placental size during mid-gestation reported varying results, with maternal under-nutrition both increasing (Faichney and White, 1987), and retarding (Everitt, 1964; Mellor, 1983; McCrabb, *et al.,* 1986) placental growth. Under-feeding throughout the entire gestational term has been shown to significantly reduce placental weight at, or near, term (Wallace, 1948; Everitt, 1968; Alexander and Williams, 1971), and a similar effect was observed in pregnant ewes which were underfed prior to day 100 (Everitt, 1964) and day 119 (Mellor, 1983) of gestation.

Everitt (1964) managed single-bearing Merino ewes to lose 12% of their live weight by day 90 of gestation, a loss which resulted in a 30% reduction in functional cotyledon weight. Further data suggest that, following underfeeding from day 50 to 100 of gestation, a return to an adequate diet may prevent, or even reverse, the normal decline in placental weight during late gestation (Faichney and White, 1987). However, Mellor (1983) found no evidence of placental compensatory growth in ewes which were underfed between days 35 and 119 of gestation and well-fed from day 119 of gestation to term, but did observe that the response to change in maternal nutrition within groups depended on placental size. Placental growth was studied in two-year-old ewes maintained on a maintenance diet (900 g daily pelleted lucerne hay and oat grain) except when restricted to moderate dietary restriction (500 g daily) from day 50 to 100, day 100 to 135 or day 50 to 135. All restrictions were associated with increased placental size (Faichney and White, 1987). In cattle, moderate feeding levels to allow for liveweight gains of 0.1 and 0.6 kg/day resulted in higher total cotyledon weights than those in heifers fed to gain 1.1 kg live weight/day (Prior and Laster, 1979).

Placental growth is restricted by heat stress during both mid- and late gestation (Alexander and Williams, 1971; Bell *et al.,* 1989) although the relationship between placental weight and functional capacity is dependent on the time of gestation at which heat stress occurs (McCrabb *et al.,* 1993). Despite a relationship being observed, the mechanisms by which heat stress reduces placental growth are unknown. Ewes which were subjected to prolonged environmental heat, sufficient to elevate deep body temperature for several hours each day, were shown to have reduced placental size (Alexander and Williams, 1971). These effects were most marked when heat exposure was continued throughout mid- and late gestation, although significant reductions also occurred when heating was confined to either mid- or late gestation only. Whereas this study indicated that heating during late gestation may accelerate the normal rate of decline in placental weight, the results of Bell (1984) demonstrated that the placenta is capable of compensatory growth during this period. Thus, compensatory growth during late gestation may have masked any previous effect heat stress had on placental growth. McCrabb *et al.,* (1993) exposed ewes to 42°C heat for 9 hours per day (h/day) and 32°C for 15 h/day between days 30 and 80 of gestation with the sole objective of restricting placental weight. Results showed that ewes exposed to heat had lower placental weights than control ewes at days 80 and 140 of gestation, and the placentae exhibited a greater number of smaller placentomes, and a lower number of large placentomes.

Other factors

Fetal sex has been variably reported to have an effect on placental size. Everitt (1964) reported increased placental weights at day 90 of gestation in ewes bearing male, compared with female, fetuses, while McCrabb *et al.,* (1992) found no significant difference in placental growth associated with the sex of the fetus. Ewe age has been reported to have an effect on placental weight. Placental weight tends to increase, and the proportion of caruncle occupancy to decline, with increasing parity of the ewe (Ferrell, 1989).

Litter size is an influencing factor on placental weight and size, with the combined weight of twin placentas in sheep usually being greater than that of a single placenta, because greater proportions of available caruncles are occupied and individual placentomes are heavier (Alexander, 1964a). However, weights of placentas of individual twins are usually less than those of single fetuses, indicating that the increased placentome size does not completely compensate for the decrease in placentome number available for each twin (Ferrell, 1989).

FETAL GROWTH

Introduction

Birth weight of the neonate is determined by the mechanisms which promote and regulate fetal growth and development throughout gestation. Many studies have used birth weight as the index for fetal growth (Gluckman and Liggins, 1984) but, while birth weights are readily obtained, they provide no information on the modifying effects of nutrition on the preceding pattern of differential tissue and organ growth in the fetus during gestation (Robinson, 1982; Gluckman and Liggins, 1984). In particular, information concerning prenatal growth of the bovine has been primarily limited to birth weight, with few studies examining placental and fetal development at mid-gestation. Alterations in the prenatal growth and development process influence both size and viability at birth (Wallace *et al.,* 1996). Excessively high or low birth weights generally cause an increase in mortality rates, and thus, for example, birth weight has an important effect on the survival and growth of lambs (Schinckel and Short, 1961; McCutcheon *et al.,* 1981). Birth weights lower than optimum are associated with reduced fetal energy reserves of body lipid and glycogen, reduced levels of thermogenic brown adipose tissue, and lowered thermoregulatory capability due to the higher surface area/volume ratio of the neonate, as well as increased perinatal mortality (Ferrell, 1989; Rattray, 1992). It is estimated that some 15% of all lambs born in New Zealand die within the first month of life, and that death is often due to inappropriate size of the newborn (Dalton *et al.,* 1980). Conversely, cows which give birth to larger than normal calves are susceptible to dystocia, which can result in perinatal mortality and/or loss of the dam. A comprehensive Australian study concluded that the major cause of death in beef calves was slow or difficult birth (dystocia), which accounted for 63.5% of the total calf losses, and that 91.7% of these losses occurred within 24 hours after birth (Young and Blair, 1974). Therefore, although fetal growth accounts for only 5 to 8% of mature body size in ruminants, this developmental phase is of vital importance (Everitt, 1968).

Fetal growth is influenced to varying degrees by numerous factors, including sex (Meyer and Clarke, 1978), birth rank (Donald and Purser, 1956), breed or breed cross (Burris and Blunn, 1952; Bellows *et al.,* 1982), dam age (Burris and Blunn, 1952), heat or cold stress (Cartwright and Thwaites, 1976a; Thompson *et al.,* 1982; Symonds *et al.,* 1986; McCrabb *et al.,* 1993), placental size (Alexander, 1974) and maternal nutrition (Robinson, 1977; Mellor, 1983). The importance of these factors and the degree to which they affect fetal growth differ between species (Ferrell, 1989), although with regard to maternal nutrition, there are too few comparative slaughter studies involving undernourished ewes to draw any firm conclusions as to when and how nutrition affects fetal growth during gestation (Robinson, 1982).

Patterns of fetal growth

It is during intrauterine life that the initiation and differentiation of all cell types is completed, and maturation of most is well under way before birth (Everitt, 1968). The total extent of development that occurs prenatally, or the degree of maturity of the fetus at birth, varies enormously among species and remarkably little within species. For example, young rats acquire the ability to thermoregulate two weeks after birth while in sheep this particular homeostatic mechanism is in place about a week before normal term (Currie, 1988), reflecting a major developmental difference between the species.

There are many changes in the conceptus of sheep and cattle throughout gestation, particularly in the growth and development of the embryo. In sheep, the embryo develops from its initial weight of around 2 μg, to a full-term fetus approximately 20 to 21 weeks later, with a weight equivalent to 15 to 25% of the metabolic weight ($W^{0.75}$)

of its dam (Robinson, 1982). Figure 2 shows that, throughout gestation, fetal growth follows a distinct exponential pattern. The prenatal development of cattle is very similar to that of sheep (Eckstein and Kelly, 1964).

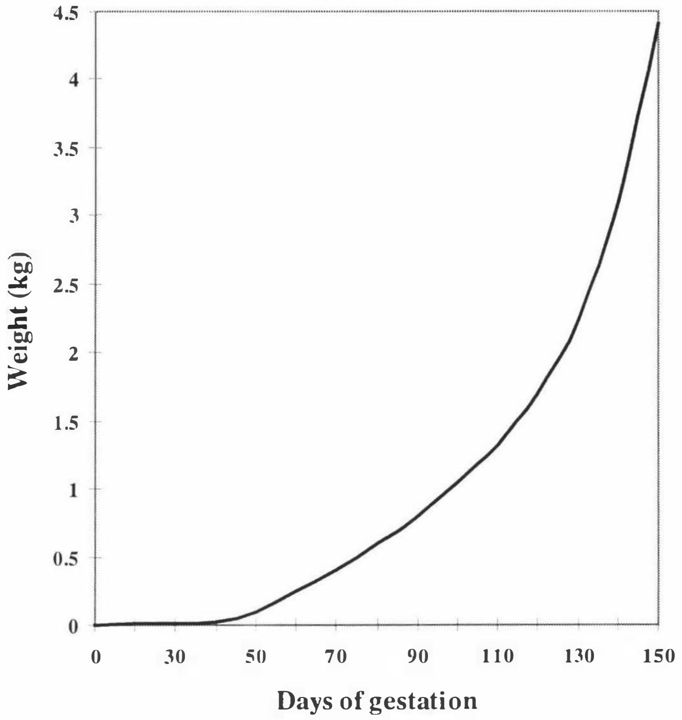

Figure 2: **The mean weights of twin fetuses at different stages of gestation in ewes of approximately 70 kg at mating (Robinson, 1982, p. 105).**

Fetal growth during early gestation

During early gestation (days 0 to 50 in sheep and 0 to 100 in cattle), the fetus grows to only approximately 15% of its size at term. For the first 100 days of gestation in sheep, fetal growth appears to be predominantly genetically motivated with external factors, such as the environment and nutrition level, appearing to have relatively small effects. However, studies have observed reductions in fetal growth up to days 90 to 100 of gestation in ewes which were severely undernourished (Everitt, 1968) or heat stressed (Alexander and Williams, 1971; McCrabb *et al.,* 1993).

The rate of growth in the fetus, i.e. the percentage increase in weight and dimensions per unit time (relative growth), is most rapid in early gestation, but decreases as gestation advances (Jainudeen and Hafez, 1980). Maximal rates of growth and development occur in a definite sequence, with the vital organs such as the heart and liver developing early, then the central nervous system, then bones and lastly adipose tissue and muscle. All fetal organs develop from primitive embryonic structures, a process which begins shortly after the time of implantation. During early fetal life, the cephalic region grows rapidly, and consequently the fetal head is disproportionately large. By day 110 of gestation, fetuses from well-fed ewes have achieved only about three quarters of their crown-rump length (CRL) and girth at term, and only about half of their weight at term, with changes in CRL and girth primarily reflecting skeletal growth (i.e. spinal column and rib growth) and changes in weight indicating soft tissue growth (Mellor and Murray, 1981).

Fetal growth during late gestation

The major characteristic of fetal growth in late gestation is that it is maternally constrained (Gluckman and Liggins, 1984). A classic example of this, is a study conducted by Walton and Hammond (1938) who examined the birth weights of pure-bred Shire and Shetland horses, and their respective crosses. The resulting birth weights of the crossbred foals were similar to those of the pure-bred of the same breed as their dam. This study demonstrated that maternal factors were the controlling feature in determining birth size.

During late gestation (days 100 to 150 in sheep and 190 to 280 in cattle), fetal mass increases in a curvilinear fashion such that (in the sheep) in the last 8, 4, and 2 weeks of gestation, the fetus is gaining equivalent to 85, 50 and 25%, respectively, of its final birth weight (Robinson, 1983). Consequently, the literature has concentrated on fetal growth and development during this period, which is considered the most important stage of gestation with regard to maternal nutrition levels. While the major fetal organs are already developed and functioning by mid-gestation, it is during late gestation that fetal hair follicles, horn pits, teeth, and finally wool or hair, are developed.

Factors affecting fetal growth

Maternal nutrition

Maternal nutrition, as well as influencing fetal growth, also affects fetal energy reserves, and colostrum production and composition, thus playing a key role in the survival of the neonate (Rattray, 1992). Nutritional manipulation of the intrauterine environment may affect embryo survival and the growth capacity of the fetus, through the development of the placenta and the transfer of nutrients between the maternal and fetal circulation (El-Sheikh *et al.,* 1955). However, the extent to which maternal nutrition can be used to manipulate fetal growth is debatable. It is often argued that the birth weight of offspring can be manipulated via varying levels of maternal nutrition, although the ability of the dam to buffer against under- or over-nutrition, by utilising her own body reserves, results in this technique being seldom successful. In ewes, liveweight differences of up to 10-12 kg have been observed to day 100 of gestation with no obvious effect on birth weight (Parr *et al.,* 1986; Rattray *et al.,* 1987). Alternatively, a 20% liveweight difference at mating, which was maintained throughout gestation, resulted in a pronounced influence on lamb size and survival which appeared to be linked to placental weight. Lambs born to light ewes (less than 55 kg) had lighter lambs and smaller placentas than those born to heavy (more than 60 kg) ewes (Clarke *et al.,* 1997). A study where ewes were managed to lose 7% of their live weight during the first 90 days of gestation demonstrated the ability of the gravid uterus to continue its growth, not only at the expense of the dam's accumulated reserves, but even when her vital structures were being depleted (Wallace, 1948). However, it has been demonstrated that the sensitivity of the ewe to feed restriction increases with the number of fetuses carried (Alexander and Williams, 1971; Alexander, 1974), thereby indicating that multiple-bearing ewes are more sensitive to under-feeding than single-bearing ewes.

The mechanism by which severe maternal under-nutrition leads to fetal growth retardation is complex, although the primary factor is clearly an inadequate supply of maternal glucose and amino acids to the fetoplacental unit (Gluckman and Liggins, 1984). While extreme maternal under- or over-nutrition during certain stages of gestation may result in a difference in birth weight, the later consequences on the dam can be detrimental. Severe under-nutrition during gestation may predispose the dam to metabolic diseases (such as hypoglycaemia), and may also reduce subsequent milk production, thus inhibiting lactational performance. Furthermore, wool production in sheep can be depressed with sudden changes in nutrition resulting in lower fleece weights, wool break (Hodge, 1966) and cotting (Coop and Clark, 1969). Controversially, it is also argued that fetal growth can only be altered by way of differing levels of maternal nutrition during late gestation. Likewise, fetal growth during late gestation may be influenced by placental development during early gestation. Altering placental development during this period may modify the placental supply of available nutrients to the fetus during its exponential period of growth throughout late gestation. However, answers to these arguments are currently unavailable and researchers still remain unable to adequately control birth weight via levels of maternal nutrition, while allowing the dam to maintain her maternal live weight. This illustrates our relative lack of understanding of the essential biological processes which initiate and regulate placental development and fetal growth, and the processes by which one develops in conjunction with the other.

Nutrition during early and mid-gestation

It has been stated that the level of maternal nutrition from embryo implantation, to day 100 of gestation, is relatively unimportant to fetal growth (Robinson, 1983; Coop, 1986; Rattray, 1992). Therefore, birth weight differences observed in the offspring of ewes which were underfed during early gestation, are unlikely to be the direct result of inadequate nutrition (Robinson, 1982). Thus, in view of the small size of the fetus and its low nutritional requirements during this period, few studies have been conducted examining the effects of maternal nutrition on fetal growth. It has also been stated that single fetuses are immediately vulnerable to large reductions in maternal feed intake only after day 110 of gestation, and that retarding effects on fetal growth are delayed if maternal feed intake is reduced prior to this stage of gestation (Mellor and Murray, 1981).

There have been few studies conducted where the effects of nutrition on fetal and placental growth and development in cattle have been examined, and consequently information concerning nutritional effects on prenatal growth of the bovine has been primarily limited to studying birth weight (Ferrell *et al.,* 1976). Fetal weight between days 90 and 255 of gestation was not affected when cows were fed to gain 0.1, 0.6 or 1.1 kg/day from day 38 of gestation to term (Prior and Laster, 1979). The results of this study were in agreement with other results (Rattray *et al.,* 1975; Ferrell *et al.,* 1976), which observed no effect on fetal weight when dams were fed near-maintenance levels of nutrition during gestation. Even when the level of maternal nutrition during early gestation was well below maintenance, fetal size (Koshy *et al.,* 1975) and birth weight (Hight, 1966) were unaffected.

Many studies examining the effects of maternal nutrition have implemented differential maternal feeding levels throughout early gestation, and split or reversed the treatments from mid-gestation onwards to term. Therefore, it cannot be determined at what stage of gestation maternal nutrition levels affected fetal and placental development. The studies of Wallace (1948) found no decrease in fetal weight in ewes which were managed to lose 7% of their maternal live weight through under-nutrition during the first 90 days of gestation. Since the publication of these results, it has been generally assumed that the level of maternal nutrition in the first 100 days of gestation in sheep does not affect early fetal growth. However, Everitt (1964) exposed the danger in this assumption by reporting a 10% reduction in fetal weight at day 90 of gestation in single-bearing Merino ewes which, as a result of under-nutrition, lost 12% of their body weight at mating in the first three months of gestation.

Holst *et al.,* (1992) found no significant difference in fetal weight from ewes fed differential levels of nutrition from days 79 to 107 of gestation. However, the changes in ewe live weight were minimal, with the ewes losing 0.8 kg, or gaining 2.75 kg live weight over the 28 day period. Where two groups of ewes were managed to maintain (1.0 times maintenance) and lose (0.8 times maintenance) live weight from days 30 to 100 of gestation, and then fed to provide for a small liveweight gain from day 100 to term, no significant difference in fetal weight occurred at days 96 or 142 of gestation. It was concluded that feeding the dam maintenance levels of nutrition from day 30 to 100 of gestation, would not result in reduced fetal growth providing that nutrition levels during the final 50 days of gestation were adequate (McCrabb *et al.,* 1986). Fetal growth was studied in two-year-old ewes fed a maintenance diet (900 g daily pelleted lucerne hay and oat grain), except when restricted to moderate dietary restriction (500 g daily) from day 50 to 100 (RM), day 100 to 135 (MR) or day 50 to 135 (MM). The nutritional restrictions resulted in smaller MM fetuses, larger RM fetuses, and MR fetuses were unaffected when compared to those from ewes fed a maintenance diet throughout gestation (Faichney and White, 1987).

Other experiments where ewes were managed to compensate during late gestation for under-feeding throughout early gestation, have reported varying results. Everitt (1967) observed almost complete compensation in birth weights of lambs from ewes which were severely underfed up to day 90 of gestation, then refed until term, whereas a study conducted by Taplin and Everitt (1964) attempted to reverse the effects of under-feeding during early gestation, which resulted in a number of ewes aborting. Mellor (1983) found no such effect in ewes moderately underfed from day 35 of gestation to 119 of gestation, then refed until day 142, although it has been suggested that at least partial compensation can occur (Everitt, 1966; 1967). However, the degree to which this effect can be reversed in the fetus by adequate feeding in late gestation may depend on the stage of gestation at which high levels of nutrition are introduced. It is also questionable whether the effects on fetal growth rates of under-nutrition during this period are directly due to an inadequate supply of nutrients *per se*, or are mediated through an effect of under-nutrition on the endocrine status of the dam (Robinson *et al.*, 1977).

Nutrition during late gestation

In line with the rapid development of fetal mass at the beginning of the fourth month of gestation in sheep, it is commonly recommended that an increase in maternal nutrition levels be available to the dam (Robinson, 1982).

A majority of studies examining the nutritional effects during late gestation on birth weight in beef cattle, have been motivated by the high incidence of dystocia which is related to calf size at birth (Laster, 1974; Hodge and Stokoe, 1974; Prior and Laster, 1979; Kroker and Cummins, 1979; Bellows *et al.*, 1982). Studies where cattle were subjected to differential feeding regimes in order to influence calf birth weight have exhibited varying results. No differences in birth weight were observed when pregnant cows with similar initial live weights (436 vs 443 kg) were fed to gain 0.6 or 0.3 kg live weight/day respectively, over the last 56 days of gestation (Nicoll, 1979). These findings are in agreement with studies where feeding treatments were commenced at different stages throughout late gestation, and resulted in no differences in calf birth weights (Hodge and Stokoe, 1974; Laster, 1974; Bellows and Short, 1978; Bellows *et al.,* 1982). These results demonstrate the remarkable ability of the dam to buffer against under-nutrition, and suggest that reductions in calf birth weight may occur only if the dam is fed levels of nutrition well below maintenance in the latter stages of gestation. However, the studies of Hight (1966; 1968) and Kroker and Cummins (1979) have successfully produced differences in calf birth weights from dams subjected to varying levels of nutrition during late gestation. Cows were managed to lose 34 or 46 kg maternal live weight over the last eight and three weeks of gestation respectively, and from then until term the animals were fed a high plane of nutrition. The liveweight losses which occurred over the last eight and three weeks of gestation, resulted in birth weight differences of 3.7 and 2.0 kg respectively, when compared to those animals fed a high plane of nutrition throughout gestation (Hight, 1968). In another study, two groups of cows were fed the same levels of nutrition up to day 160 of gestation, after which time they were fed high (H) and low (L) planes of nutrition. L cows lost 81.8 kg over the 120 day treatment period, which resulted in a 5.4 kg decrease in calf birth weight, when compared to birth weights of calves from the H group (Hight, 1966). Between-group differences in calf birth weights were observed when cows were fed to gain (1.12 kg/head/day), maintain or lose (-0.11 kg/ head/day), and lose (-0.57 kg/ head/day) live weight over the last 84 days of gestation (Kroker and Cummins, 1979). These three studies demonstrated that birth weight can be manipulated to some extent via levels of maternal nutrition during mid- to late gestation.

Similarly, studies examining the effects of maternal nutrition in ewes on fetal growth during late gestation, also report varying results. Chandler *et al.,* (1985) observed no decrease in fetal weight from ewes which lost 3 or 5 kg by day 143 of gestation, as a result of a treatment combination of exercise and under-feeding imposed between days 80 and 100 of gestation respectively. However, ewes fed a low plane of nutrition from days 95 to 123 of gestation, and a high plane of nutrition thereafter, exhibited increased birth weights, despite a 6.5 kg decrease in maternal live weight over the 28 day treatment period (Fogarty *et al.,* 1992). These results are in agreement with those of El-Sheikh *et al.,* (1955), where high levels of feeding were imposed on ewes from days 91 to 144 of gestation. West African Dwarf ewes fed to gain 0.67 kg live weight over the last 48 days of gestation, gave birth to lambs which were significantly lighter (1.1 vs 1.7 kg) than those from ewes which gained 4.7 kg over the same gestational period (Adu and Olaloku, 1979).

These studies indicate the relative flexibility of the fetus to gain weight in conjunction with increased levels of maternal nutrition past day 100 of gestation. Although results of other trials indicate that it is possible to detrimentally affect lamb birth weight via maternal undernutrition, this effect appears to only occur in situations of extreme underfeeding, such as situations in which ewe flocks normally weighing more than 53 kg at mating, achieve conceptus-free weights of less than 42 kg in late gestation (Smeaton *et al.,* 1985).

A 30 to 70 % decrease in fetal weight over a three day period has been observed, and in rare cases fetal growth almost ceased, when ewes were well fed prior to days 90 to 100 of gestation, and thereafter were severely underfed (Mellor and Murray, 1981). The incredible speed and extent of this fetal response indicated that the rate of mobilisation of growth substrates from maternal reserves is substantially less than fetal requirements and thus emphasises the importance of ensuring a continuous supply of high quality feed from day 100 of gestation onwards.

The placenta

Maternal nutrition and placental size are well recognised as major determinants of fetal growth rate. In practice they act simultaneously and their effects may be confounded because a high plane of maternal nutrition can partly offset the growth-retarding effects of a small placenta, and a large placenta can partly offset the retarding effects of maternal under-nutrition. However, the survival of the fetus is jeopardised more by a small placenta than by decreased maternal nutrition (Mellor, 1983), although given that reduced placental size could be a result of decreased maternal nutrition, survival of the neonate may still be indirectly affected. The well known correlation between placental mass and fetal weight (Dawes, 1968), and the growth-retarded lambs of ewes in which the placental mass has been surgically reduced (Alexander, 1964a; Owens *et al.,* 1995), demonstrate the long-term control of fetal growth by the placenta. This experimental manipulation can result in growth retarded fetuses whose viability after birth is reduced (Harding *et al.,* 1985). A relationship between birth weight and cotyledon weight exists in heat stressed and undernourished ewes, and becomes more marked as gestation proceeds (Alexander, 1974). This suggests that any restricting effect of the placenta on fetal growth increases as gestation advances.

Although placental size is closely correlated with the size of the fetus at birth in sheep (Alexander, 1974; Davis *et al.,* 1981; Bassett, 1991), and cattle (Prior and Laster 1979; Head *et al.,* 1981), the importance of placental size as a determinant of fetal growth rate, has in the past been greatly underestimated. This was demonstrated in a study conducted by Mellor (1983), who observed that, on average, approximately 75% of the within-group variance in lamb birth weight could be accounted for by variations in placental weight, compared to the within-group figure of 35%, observed by Alexander (1964a). The relationship between fetal growth and placental size is not entirely clear and it may not necessarily be one of cause and effect, because rather than limiting fetal growth, placental size may be regulated by, or together with, the fetus (Bell, 1984). The data of Dawes (1968) and Ferrell *et al.,* (1976) concluded that the development of the bovine fetus is dependent upon prior development of the uterus and fetal membranes, which are necessary to supply nutrients to the maturing fetus. This conclusion is supported by McNeill *et al.,* (1997) who speculated that an increase in ewe condition through improved maternal nutrition up to day 72 of gestation, could have resulted in a larger fetus because well conditioned ewes had larger placentas than those in poor condition.

Studies have shown that placental weight and placentome number per fetus are related to fetal weight (Owens and Hinch, 1984; Fennessy and Owens, 1985; Owens *et al.,* 1986). A significant correlation of fetal cotyledon weight with fetal weight was observed at day 137 of gestation (Davis, 1983), a relationship which has been described elsewhere for sheep (Alexander, 1974) and cattle (Head *et al.,* 1981). Data have shown that placental weight and fetal growth rate in sheep are positively correlated during periods of maternal underfeeding which start before 90 days, at 95 or at 112 days of gestation (Mellor and Murray, 1981, 1982a). However, placental size must be restricted by under-nutrition below a critical threshold level before subsequent fetal growth is affected, for abundant feeding in late gestation of ewes which were severely ill-fed during early gestation, resulted in lambs of average birth weight (Taplin and Everitt, 1964; Everitt, 1966; Everitt, 1967).

The provision of substrates to the fetus depends finally on the efficacy of the placenta (Gluckman and Liggins, 1984) which, if reduced, could have a negative effect on fetal growth. When placental growth is restrained, uterine blood flow is restricted, and hence transport of substrates to the fetus is reduced, thereby resulting in fetal growth retardation (Owens *et al.,* 1987a, 1987b). This was demonstrated by Alexander (1964b) who surgically removed most of the caruncles (i.e. placental attachment sites) from the non-pregnant uterus. This procedure resulted in restricted placental growth, and reduced fetal growth in the subsequent pregnancy. Therefore, restriction of fetal growth occurs when placental function is reduced, or an experimental restriction in placental growth is performed. As a consequence, the rate of uterine and umbilical blood flow, glucose transfer capacity, permeability to urea and the rate of delivery of substrates to the pregnant uterus and the fetus are reduced, and fetal hypoxaemia and hypoglycaemia can result (Owens *et al.,* 1987a, 1987b).

Both Alexander (1974) and Mellor (1983) interpreted the curvilinear relationship between fetal and placental weight to indicate decreased placental efficiency as placental weight increases. However, it is equally feasible that the placenta has a functional reserve, and that fetal growth is placentally limited only when this reserve is exceeded, because of either small placental size or large fetal growth potential (Bell, 1984). Even in ewes well-fed during late gestation, a decline in fetal growth rate during the last few weeks of gestation has been observed. Studies (Alexander, 1978; Mellor, 1983) have cited this observation as evidence for placental restriction of fetal growth, in agreement with the results of Davis (1981) which implied that the ability to alter lamb birth weight via maternal nutrition during late gestation is limited by the extent of placental growth. A study which conducted daily measurements of fetal CRL and thoracic girth in individual fetuses *in utero*, showed that differences in placental weight accounted for 69 to 91% of the variance in fetal weight, and was the greatest single source of variation in the weight of the fetus at term (Mellor and Murray, 1981, 1982b).

Observations by Alexander (1964a) concluded that the fresh weight of the placenta is correlated with fetal weight over the last 50 days of gestation, and these findings were interpreted by McCrabb *et al.,* (1986) to indicate placental restriction of fetal growth, due to substrate limitation. It appears that the rate of nutrient supply to the fetus *in utero* may be close to that of utilisation by the fetus, and therefore a reduction of placental mass, and presumably the maternal blood flow, causes a reduction of nutrient supply for fetal growth (Jones, 1976). Foote *et al.,* (1959) reported a significant association between fetal and membrane weight as early as day 25 of gestation, and by day 40 of gestation, when components of the placenta could be measured, the weight of the developing maternal components was significantly associated with fetal weight, independent of fetal membranes and fluids. Fetal cotyledon weight at day 140 of gestation also had a significant direct effect on fetal weight. McCrabb *et al.,* (1986) observed that maternal under-nutrition from days 30 to 100 reduced placental size at day 96 of gestation, but no difference was detected in weight or dimensions of the fetus. While studies have shown that placental size influences fetal growth, the exact mechanisms by which this process occurs remains unclear, although reduction in substrate supply to the fetus appears to be the most likely reason.

Environmental factors

Maternal heat stress during mid- and late gestation has been shown to affect fetal weight (Yeates, 1958; Alexander and Williams, 1971; Cartwright and Thwaites, 1976a; Brown, *et al.,* 1977). McCrabb *et al.,* (1993) reported that fetal weights were reduced not after the initial period of heat treatment from day 30 to 80 of gestation, but at day 140 of gestation. These findings confirm the theories of Alexander and Williams (1971) and Bell *et al.,* (1987) that fetal growth during late gestation is retarded in heat-stressed ewes as a result of a primary restriction in fetal growth.

Other factors

Following the early studies of Wallace (1948) which showed only small effects of litter sizes on fetal weights at day 90 of gestation, it has been widely assumed that variability in birth weight is due entirely to differences in fetal growth rates in late gestation (Dingwall *et al.,* 1987). Robinson *et al.,* (1977) questioned this assumption using a mathematical model for the description of fetal growth from day 55 to 145 of gestation in prolific ewes. The model implied that reductions in size at birth that accompany increases in litter size, originated in early gestation. While it has been shown that litter size influences fetal weight, it is generally accepted that the influence only becomes apparent after days 90 to 100 of gestation (Robinson, 1981). Studies detailing the associated placental and uterine structures, and their relationships to the weight of individual fetuses within the same litter, have shown that the presence of some very small lambs in large litters is not attributable to maternal undernutrition in late gestation (Rhind *et al.,* 1980; Dingwall *et al.,* 1987).

Fetal sex can have a significant effect on the weight of the fetus. In calves, males were found to be heavier at birth than their female counterparts (Gregory *et al.,* 1950; Echternkamp, 1993), while in sheep, male lambs tended to be on average 0.5 kg heavier than females (Alexander, 1974).

PURPOSE AND SCOPE OF THE INVESTIGATION

Fetal growth follows an exponential growth pattern and so has very low nutritional requirements in early gestation compared to those in later gestation (and compared to the requirements of the dam). Thus it has been generally assumed that females in early gestation need not be provided with nutrients additional to those required by comparable non-pregnant females. However, this ignores the fact that the placenta has a different growth pattern to the fetus and that nutritional manipulation of placental growth in early gestation could determine limits to fetal growth in later pregnancy.

Cattle with large calves are susceptible to dystocia which has potentially fatal results. Thus control of fetal growth and development in the beef cow is important to the well-being of both calf and dam. As previous attempts to control calf birth weight by nutritional manipulation in late gestation have not been consistently successful, the first objective of this study was to examine the effects of differential nutrition of the beef cow on fetal growth as measured by birth weight. This study did not, however, permit a direct assessment of effects on fetal growth because cows were taken to term and because the "switchback" of nutritional levels left open the possibility of compensatory effects in the second half of gestation.

The second study was therefore designed to examine the effects of nutrition on fetal growth at day 100, and so to eliminate the potential for the dam to compensate for earlier treatments during late gestation. This study investigates the hypothesis that, contrary to current dogma, high maternal nutrition during early gestation may indeed lead to increased placental and fetal growth. That is, while the early gestation fetus may not require high levels of maternal nutrition, the possibility exists that it may benefit from them.

CHAPTER TWO

THE EFFECTS OF MATERNAL NUTRITION DURING EARLY GESTATION ON CALF BIRTH WEIGHT IN BEEF CATTLE

ABSTRACT

It is generally assumed that, because the fetus has a small nutritional requirement relative to that of the dam in early pregnancy, differential maternal nutrition at this time is unlikely to influence fetal growth. However, this assumption has not been widely tested.

The study was conducted over two years, with the objective of examining the effects of differential nutrition of the beef cow on fetal growth as measured by birth weight. Crossbred heifers were managed for High (H, 0.6 kg/day) or Low (L, 0.1 kg/day) liveweight gains from mating until day 140 of gestation. Treatments were then reversed so that effects of nutrition during early gestation were not confounded by differences in maternal live weight at calving. Averaged across years, maternal live weights (kg, Mean±SE, n=60) were (H vs L) 393.5 ± 4.3 vs 362.1 ± 4.3 ($P<0.001$) at day 140 of gestation and 417.6 ± 4.5 vs 408.7 ± 4.3 ($P>0.05$) at term. Calf birth weights were 31.1±0.5 vs 31.4±0.5kg and weaning weights (average calf age = 90 days) were 91.3 ± 2.0 vs 89.7 ± 2.0 (both $P>0.05$).

These results indicate that birth weight in cattle is not altered by levels of maternal nutrition during the first 140 days of gestation if adequate nutrition is provided from day 140 of gestation to term. Any nutritional effects on fetal growth which could have occurred at mid-gestation may have been masked by the differential feeding after day 140 of gestation.

INTRODUCTION

Birth weight in beef cattle is an important factor in the well-being of both the newborn and dam. Cows carrying large calves are susceptible to dystocia, putting the life of both the calf and dam at risk. Dystocia, or calving difficulty, has been stated as the major cause of peri-natal calf losses (Anderson and Bellows, 1967) with high birth weights being the major cause of calving difficulty (Bellows *et al.,* 1971; Rice and Wiltbank, 1972; Laster and Gregory, 1973). A possible mechanism by which to control birth weight is by altering maternal nutrition during gestation, an attractive option for the New Zealand beef (and dairy) industry. The New Zealand industry would benefit greatly if birth weight could be manipulated by altering levels of maternal nutrition during gestation, without suppressing postnatal growth and development of the calf, and subsequent re-breeding performance of the dam.

Numerous experiments have been conducted examining the nutritional manipulation of birth weight in cows and have focused on nutrition during the latter third of gestation, the time when the majority of fetal growth occurs (Hight, 1968; Hodge and Stokoe, 1974; Anderson *et al.,* 1981; Bellows *et al.,* 1982). These studies concluded that reduced levels of maternal nutrition throughout late gestation decreased calf birth weights. However, other experiments have found that, while low levels of maternal nutrition prior to parturition altered birth weight of the calf, they had no beneficial effect (Bellows and Short, 1978) in reducing the incidence of dystocia (Tudor, 1972). As is the case with sheep, literature examining the effects of maternal nutrition during early gestation (i.e. the first three months) in cattle is sparse.

While the literature indicates that neither the calf nor the dam benefits consistently from varied levels of maternal nutrition throughout late gestation, there is a lack of information regarding maternal nutrition during early gestation in the beef cow, and its effect on calf birth weight.

The objective of this study was therefore to examine whether calf birth weight can be manipulated by varying levels of maternal nutrition throughout the first half of gestation in beef heifers, uncomplicated by a difference in dam live weight at calving.

MATERIALS AND METHODS

Animals and treatment

Beef × dairy heifers (15 months old) were assigned to the experiment, which involved two nutritional treatments during gestation. The experiment was repeated over two years and heifers in both years were managed at pasture to reach similar pre-calving live weights in each nutritional group.

In 1994, thirty-one Hereford × Friesian (H × F) and twenty-nine Hereford × Jersey (H × J) heifers were joined with two Angus bulls. Six did not conceive, and were subsequently sold as dries and excluded from the final analysis. Calving occurred during the period 27 August to 4 October 1994 (mean calving date 17 September). Three calves died due to dystocia-related problems at parturition.

In 1995, thirty Hereford × Friesian (H × F) and thirty Angus × Friesian (A × F) heifers were joined with two Angus bulls. Seven heifers did not conceive, and two heifers died. These heifers were discarded from the analysis. Calving occurred during the period 8 September to 11 October 1995 (mean calving date 25 September).

After the first cycle of mating, the heifers were randomly allocated by live weight and breed cross to the two nutritional treatments. For the first 140 days of gestation, half of the heifers were fed to gain live weight at 0.60 kg/day (High or H group), while those in the second group were fed to achieve a liveweight gain of 0.10 kg/day (Low or L group). From day 140 of gestation until term, the treatments were reversed so that the effects of early gestation nutrition were not confounded by differences in maternal live weight at calving. The heifers were weighed monthly, and at calving the dam's pre- and post-calving live weight was recorded along with calf birth weight. Calves were weighed and tagged at birth whether they were alive or dead. Average calf age was 90 days at weaning, when both calves and their dams were weighed.

From calving to weaning the heifers were grazed *ad libitum* on predominantly ryegrass (*Lolium perenne*) and white clover (*Trifolium repens*) pasture at Keeble farm, Massey University, Palmerston North.

Statistical analysis

Analysis of covariance was used to determine the effects of nutrition during early and mid-gestation on dam live weights from mating to term, and on calf birth and weaning weights. Calf birth and weaning weights were adjusted to a common maternal live weight and birth date. Data are expressed as least square means and standard errors for the treatment groups and their calves. Statistical analyses were conducted using the computer package 'SAS' (SAS, 1988).

RESULTS

Figures 3 and 4 show the live weight changes for the two treatment groups from day zero of gestation through to calving. The live weight differences between the H and L groups at day 140 of gestation were 43.7 and 14.5 kg for 1994 and 1995 respectively.

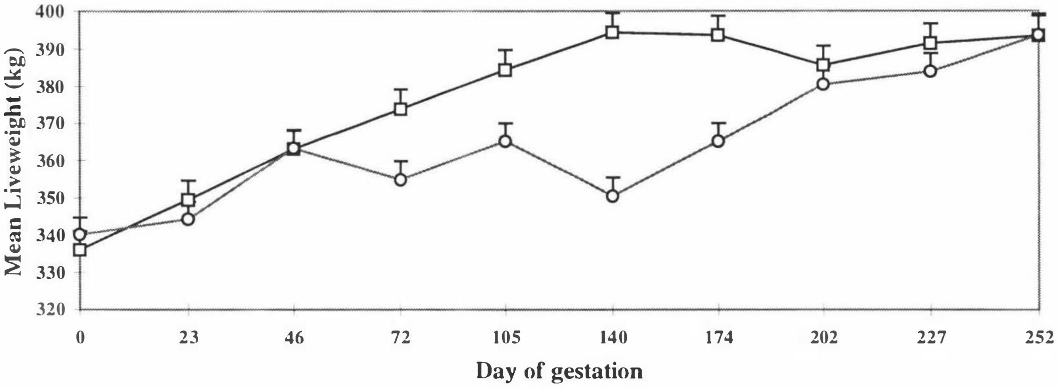

Figure 3: **Mean live weights of the 1994 High (-□-) and Low (-o-) treatment groups throughout gestation. Vertical bars represent the standard error of the mean.**

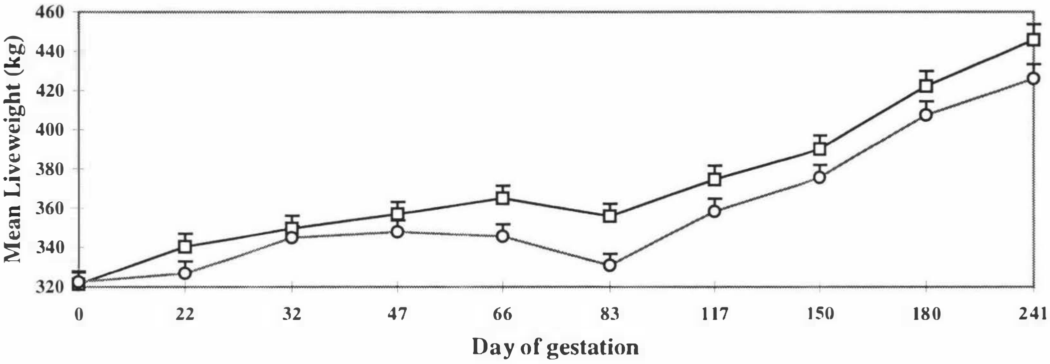

Figure 4: **Mean live weights of the 1995 High (-□ -) and Low (-o-) treatment groups throughout gestation. Vertical bars represent the standard error of the mean.**

Selected dam live weight parameters for the two treatment groups in 1994 and 1995 are shown in Table 1.

There was no interaction between dam breed and nutritional treatment in either year, and therefore results for the two main effects are treated separately.

In 1994, varied levels nutrition during early gestation increased live weight differences between the two treatment groups. Heifer live weights recorded on days 105, 140, 174 and 202 of gestation were significantly (P<0.05) different.

Differing levels of nutrition during early gestation in 1995 caused differences in heifer live weights between the two groups, although only weights recorded on days 66 and 83 of gestation were significantly (P<0.05) different.

Table 1: **Effects of early gestation nutrition on selected dam and calf live weights (Mean±SE).**

	1994		1995	
	Low	High	Low	High
n	27	27	26	25
Dam (kg)				
Gestation day 0 weight	340.3±4.5	336.2±4.9	322.3±5.6	321.3±6.0
Early gestation weight [1]	365.1±5.0^a	384.3±5.4^b	358.3±6.6	374.4±7.1
Mid-gestation weight [2]	350.6±4.9^a	394.3±5.3^b	375.3±6.7	389.8±7.2
Late gestation weight [3]	393.5±5.3	393.4±5.8	425.7±7.3	443.8±7.9
Weaning	452.2±6.0	453.3±6.5	368.8±6.1	374.9±6.6
Calf (kg)				
Birth weight	30.9±0.7	29.9±0.8	32.1±0.6	32.6±0.6
Weaning weight	137.7±3.7	130.2±4.3	116.3±3.2	121.3±3.5

^{ab} Means with different superscripts in the same row within year are significantly different ($P<0.05$)

[1] 1994 = day 105; 1995 = day 117

[2] 1994 = day 140; 1995 = day 150

[3] 1994 = day 252; 1995 = day 241

The effects of dam breed-cross on selected dam and calf weights in 1994 and 1995 are presented in Table 2. In 1994, H × F dams had significantly ($P<0.05$) greater live weights, when compared to their H × J counterparts. Calves from H × F dams exhibited greater birth and weaning weights ($P<0.05$) when compared to those from H × J dams. Dam breed-cross had no effect on the weights of either dams or calves in 1995.

Table 2: **Effect of dam breed-cross on selected dam and calf live weights (Mean±SE).**

	1994		1995	
	Hereford × Friesian	Hereford × Jersey	Hereford × Friesian	Angus × Friesian
n	26	28	26	25
Dam (kg)				
Gestation day 0 weight	351.8±4.7^{b}	324.7±4.8^{a}	318.6±5.7	325.5±5.7
Early gestation weight [1]	360.6±5.4	388.8±5.2	361.9±6.8	370.9±6.7
Mid-gestation weight [2]	386.3±5.2^{b}	358.5±5.3^{a}	377.3±6.9	387.8±6.8
Late gestation weight [3]	405.0±5.6^{b}	381.9±5.8^{a}	431.0±7.6	438.4±7.4
Weaning	434.9±6.0^{b}	400.2±6.1^{a}	365.9±6.3	377.8±6.2
Calf (kg)				
Birth weight	31.5±0.7^{b}	29.2±0.8^{a}	31.6±0.6	33.1±0.6
Weaning weight	141.4±4.1^{b}	126.4±4.0^{a}	120.9±3.3	116.8±3.2

[ab] Means with different superscripts in the same row within year are significantly different ($P<0.05$)

[1] 1994 = day 105; 1995 = day 117

[2] 1994 = day 140; 1995 = day 150

[3] 1994 = day 252; 1995 = day 241

DISCUSSION

The main objective of this study was to determine whether the birth weight of calves born to beef × dairy heifers could be influenced by varying levels of nutrition during early gestation (day 0 to 140), without the confounding effect of a difference in maternal live weight at calving.

Over the 140 day period, the 1994 L and H treatment groups achieved liveweight gains of 10.3 kg (0.07 kg/head/day) and 58.1 kg (0.41 kg/head/day) respectively. Over the same period, the 1995 heifers achieved liveweight gains of 53 kg (0.35 kg/head/day) and 68.5 kg (0.45 kg/head/day), for the L and H treatment groups respectively. The between-treatment live weight differences at day 140 in 1994 and 1995 were 47.8 kg and 15.5 kg respectively, although only in 1994 was the difference significant. Although between-treatment live weight differences were achieved by day 140 of gestation in both years, nutrition during early gestation did not have an effect on calf birth, and subsequent weaning, weights. The small and nonsignificant live weight difference at mid-gestation in 1995, a result of poor control of target daily liveweight gains, was attributed to the climatically good summer/autumn in that year. A 38% increase in pasture cover (kg dry matter (DM) per hectare) throughout March, April, May and June in 1995, compared with the same period in 1994, made control of heifer intake difficult, and this is reflected in the daily liveweight gain differences between years. The heifer live weights in 1995 ran almost parallel to one another (see Figure 4) and the target between-treatment live weight difference was not met (70 kg vs 15 kg).

Although the actual between-treatment live weight difference in 1994 did not meet the target (47.8 kg vs 70 kg), a distinctive live weight pattern developed over the treatment period (see Figure 3). A possible contributing factor for the nonsignificant difference between calf birth weights in 1995 could be the small between-treatment liveweight differences. However, the fact that the 1994 heifer live weights were significantly different at day 140 of gestation, and there was still no significant difference found in calf birth weight, indicates that the between-treatment live weight difference in 1994 was not large enough. It is possible that differences in birth weights would have been generated had the between-group live weight target been met. However, there may have been a difference in fetal weight at day 140 of gestation in 1994, but because the treatments were applied throughout the entire gestational period, the dams ability to buffer using her own reserves may have masked any potential difference in birth weight at term.

Therefore, birth weight in cattle is notoriously difficult to control, or alter, via levels of maternal nutrition during gestation. The ability of the dam (in both sheep and cattle) to buffer against under-nutrition during early gestation using her own body reserves, severely limits the extent to which birth weight can be altered via this period of early fetal development. The fact that the fetus places relatively small demands on the dam during early gestation, and that approximately 85% of fetal development occurs during late gestation (Robinson, 1983), is reason enough for researchers to concentrate on late gestation as the period in which levels of maternal nutrition are most likely to affect fetal growth. Therefore, literature examining the consequences of low levels of maternal nutrition during early gestation on calf birth weights is scarce.

Studies examining the potential manipulation of fetal development and birth weight via maternal nutrition during early and mid-gestation, have shown that near-maintenance levels of nutrition had no effect on fetal size or composition. However, near-maintenance levels of nutrition during late gestation showed to have an effect on calf birth weight (Laster, 1974; Anderson *et al.,* 1981; Pleasants and Barton, 1992) in cattle (Ferrell *et al.,* 1976), and lamb birth weight in sheep (Rattray *et al.,* 1975). These findings indicate that the level of nutrition throughout early gestation is required to be well below maintenance in order for an effect on birth weight to be significantly reduced.

Similarly, Prior and Laster (1979), fed heifers three nutritional levels to provide for liveweight gains of 0.1 (low), 0.6 (medium) and 1.1 (high) kg/day from day 42 of gestation, until the heifers were progressively slaughtered between days 90 and 245 of gestation. The study indicated that levels of maternal dietary energy did not influence fetal weight.

In conclusion, although levels of maternal nutrition during early gestation showed to have no effect on calf birth and subsequent weaning weights, the literature shows that it is possible to depress birth weight to some extent via the same mechanism during late gestation. It could be suggested that, had the proposed liveweight changes between the treatment groups been achieved, the ability of the dam to buffer against undernutrition may have been somewhat diminished, and a difference in birth weight may have been observed.

To generate an effect on birth weight via levels of maternal nutrition during early gestation, the necessary nutritional restrictions are required to be well below maintenance. Consequently, this would be impractical and unreasonable, considering the negative effects on the later reproductive and lactational performance of the dam. As the cattle study yielded no significant results, a more intensive second study was conducted, using sheep as a model, examining the effects of nutrition throughout the first 101 days of gestation on fetal and placental growth and development. Nutritional treatments were applied from days 21 to 101 of gestation, when the animals were slaughtered. This allowed an examination of the effects of nutrition on fetal growth at day 101, and also eliminated the potential for the dam to buffer during late gestation, and hence mask any difference in fetal weight that may have occurred during early gestation. Sheep were the chosen species due to their lesser area requirements, their manageability and the fact that their feed intakes can be manipulated to a greater extent under pastoral conditions. It was anticipated that this study would provide an insight into how levels of maternal nutrition during early gestation affect fetal and placental growth and development.

CHAPTER THREE

THE EFFECTS OF MATERNAL NUTRITION DURING EARLY AND MID-GESTATION ON FETAL AND PLACENTAL GROWTH IN BREEDING EWES

ABSTRACT

The effects of nutrition during early gestation on fetal and placental growth and development, and hence birth weight, have not been studied extensively. This study was conducted to determine whether placental development and fetal growth may be modulated by nutritional manipulation of the ewe during the first 101 days of gestation.

Mixed-aged Romney ewes (average live weight 54.5 ± 0.4 kg) pregnant to a synchronised oestrus, were allocated to three nutritional treatment groups (*n* = 20/group), Low (L = 0.5 maintenance (M)), Control (C = 1.0M) and High (H = 1.5M) from days 21 to 101 of gestation. Maintenance requirements for a 50 kg ewe were assumed to be 0.9 kg DM/ewe/day (10 MJ ME/day) at a concentration of 11 MJ ME/kg DM. Feed consumption rates were calculated as being 1.35, 0.9 and 0.45 kg DM/ewe/day for H, M and L treatment groups respectively. Ewes were weighed weekly and slaughtered at the end of treatment, and fetal and placental measurements recorded.

Live weights were significantly ($P<0.001$) different at slaughter (L, 45.8 ± 1.4 kg; C, 56.8 ± 1.4 kg; H, 69.1 ± 1.4 kg). At day 101 of gestation, measures of fetal and placental growth and development were (C vs H group): Uterus (minus fetus and fluids) (1290.7 ± 67.0 vs 1475.4 ± 64.8 g, $P<0.05$); fetal weight (1280.8 ± 38.0 vs 1379.8 ± 35.2 g, $P<0.05$); total placentome weight (631.0 ± 30.7 vs 702.9 ± 29.7 g, $P<0.01$) and total placentome number (102.6 ± 3.2 vs 93.4 ± 3.1 g, $P<0.05$). Low levels of maternal nutrition did not significantly influence these parameters (L vs C).

This study demonstrated that high levels of maternal nutrition during early and mid-gestation enhanced fetal and placental growth and development, while low levels were without effect compared to ewes fed at maintenance

INTRODUCTION

It is estimated that some 15% of all lambs born in New Zealand die within the first month of life and that, of these deaths, two-thirds are due to inappropriate size of the newborn (Dalton *et al.,* 1980). Birth weight, and hence fetal growth and development, is a major determinant of the survival and growth of young lambs. It is, however, notoriously difficult to control via maternal nutrition during gestation, because of the ability of the dam to buffer against external influences by using her own body reserves (Robinson, 1983). The extent to which fetal growth is affected by maternal nutrition depends greatly on the degree and duration of the nutritional treatment (Faichney and White, 1987).

The majority of experiments investigating the effects of nutrition during gestation in sheep have involved looking at one of three gestational stages (Robinson, 1983) - days 0 to 50, 50 to 100, and 100 to 150 of gestation. It is in the late gestation period (days 100 to 150), when growth of the fetus amounts to 85% of its birth weight (Robinson, 1977), that most studies have focused on and have indicated very clearly the large and important effects of nutrition on lamb birth weight (Wallace, 1948; Thomson and Aitken, 1959; Schinckel, 1963; Mellor and Matheson, 1979) and fetal growth (Bennett *et al.,* 1964; Adu and Olaloku, 1979; Rattray and Trigg, 1979; Mellor and Murray, 1982a).

While there have been numerous studies carried out on maternal nutrition during mid- to late gestation, there are few reports in the literature examining the effects of nutrition during the first 100 days of gestation on fetal growth. The extent to which birth weight can be manipulated via levels of maternal nutrition during this period is largely unknown, and studies carried out in this area report somewhat conflicting results. It has been shown that a 7% decrease in dam live weight during the first three months of gestation had no effect on fetal growth and development at day 90 of gestation (Wallace, 1948), suggesting that the ewes were using their own reserves to buffer against the lack of nutrition. However, there have been studies which showed that severe under-nutrition during early and mid-gestation (days 0 to 100) resulted in reduced placental weight (McCrabb *et al.,* 1992) and lamb birth weight (Everitt, 1967).

Some researchers observed seasonal differences in fetal growth between December- and March-mated ewes. These differences were thought to have occurred during early gestation, and were the result of reduced placental development in March-mated ewes, although the mechanism by which season influenced this process was unknown (Jenkinson *et al.,* 1995).

Manipulation of placental development is another mechanism by which to potentially alter fetal growth and development. Research examining the ability of the placenta to alter fetal growth has shown that birth weight is strongly correlated with placental weight in sheep (Everitt, 1967; Alexander, 1974; Mellor and Murray, 1981; Davis *et al.,* 1981; Hay Jr, 1991). As the placenta forms during the first three months of gestation, and is fully developed (at least in terms of size) by day 100, it is possible that placental development could be altered via maternal nutrition during early gestation. It has been suggested that poor nutrition during the first three months of gestation (Russel *et al.,* 1967, 1968) could retard development of the placenta in pregnant ewes, thereby aggravating the effects of maternal under-nutrition, and leading to small lambs with low viability (Purser and Young, 1959, 1964; Alexander, 1974; Khalaf *et al.,* 1979). Therefore, if varying degrees of maternal nutrition during early gestation can alter placental development, and taking into account the relationship between birth weight and placental weight, this process could potentially be used to manipulate fetal growth and development.

The objective of this study was to determine the extent to which placental, and hence fetal growth, may be manipulated by altering levels of nutrition during the first 100 days of gestation in mixed-age ewes.

MATERIALS AND METHODS

Animals and treatment

The trial was a 3 × 2 factorial design, incorporating three nutritional treatments and two pregnancy ranks (single vs twin).

One hundred and fifty mixed-age (three to seven years) Romney ewes were joined with Romney rams at a progesterone-synchronised oestrus (Eazi-breed CIDR Type G, Carter Holt Harvey Plastic Products, Hamilton, New Zealand) for two cycles. Only ewes pregnant to the first synchronised oestrus were used in the study. Average ewe live weight at mating was 53.6 ± 5.7 kg. Following stratification on the basis of ewe live weight, the ewes were randomly allocated to one of three nutritional treatments, with the average live weight of each treatment group being 54.5 ± 0.4 kg.

The three nutritional treatment groups (Low, L; Control, C and High, H) involved feeding levels of 0.5, 1.0 and 1.5 times maintenance, respectively, from days 21 to 101 of gestation. Maintenance requirements for a 50 kg ewe were assumed to be 0.9 kg DM/ewe/day (10 MJ ME/day) at an energy concentration of 11 MJ ME/kg DM (Robinson, 1983). The target levels of feed consumption were therefore, 0.90, 0.45 and 1.35 kg DM/ewe/day for the L, C and H treatment groups respectively.

The three treatment groups were set stocked on predominantly ryegrass (*Lolium perenne*) and white clover (*Trifolium repens*) pasture using electric fencing, at different stocking rates. Individual paddock size for each treatment group was set according to the herbage cover and feed requirements to ensure correct intakes were met. The treatment groups were rotated between three 1.5 hectare paddocks throughout the trial period. Animal intakes were controlled by varying paddock size according to herbage cover (measured by a rising-plate meter (Earle and McGowan, 1979)), ewe live weights and estimated feed requirements.

Using real-time ultrasound scanning (Carter, 1987), single or multiple pregnancy was confirmed at day 42 of gestation, and 20 ewes (10 single- and 10 twin-bearing) were selected from each treatment group to complete the factorial design. The ewes were weighed weekly (from day 0 of gestation) until slaughter, which began at day 98 of gestation (21 June). One ewe died and one other was, at slaughter, found to be carrying lambs conceived during the second cycle. These animals were subsequently excluded from the analysis

Slaughter procedure

Final ewe live weights were determined approximately 12 hours prior to the first day of slaughter and, from then on, each slaughter group was weighed the night prior to slaughter. Two single- and two twin-bearing ewes from each treatment group were randomly allocated to daily slaughter groups, thereby ensuring that the treatment groups were balanced across the slaughter dates. Each ewe was identified by a coloured number sprayed onto their back, which corresponded to the day and the order in which they were to be slaughtered (H Single, H twin, C single, C twin, L single, L twin etc). The ewes were slaughtered by captive bolt pistol and exsanguination. Slaughter was conducted between 0800 and 1600 hours, over a five day period. Following exsanguination, the abdominal cavity was opened and the gravid uterus removed. The cervix was trimmed off and the ovaries removed. The uterus was then weighed, giving total gravid uterus weight. The allantioc and amniotic fluids were removed through an incision made along the greater curvature of the pregnant horn and discarded. The fetus(es) were removed from the uterus and the umbilical cord ligated at the abdomen before being severed. Each fetus was gently squeezed by hand to remove amniotic fluid, and fetal number, weight and sex were then recorded. Any fetuses still alive were euthanased by intra-cardiac injection of sodium pentobarbitone (Pentobarb 500, Chemstock Animal Health, Christchurch, New Zealand). Fetal curved crown-rump length (with the fetus lying in a "relaxed" position) and chest circumference were then recorded.

Placentomes were dissected from the uterus using curved scissors, separated into their maternal (caruncle) and fetal (cotyledon) components, and their individual weights recorded. The myoendometrium and the fetal membranes were weighed. The total weight of the caruncle and cotyledon tissues was used in this study as an index of placental size, and is referred to as 'placental weight'.

The ewe carcasses were chilled overnight, weighed and a cut made between ribs 12 and 13. Measurements were made of soft tissue depth GR (110mm off the midline (Kirton and Johnson, 1979)) and fat depth c (over the maximum depth of the eye muscle (Palsson, 1939)).

Statistical analysis

Analysis of covariance was used to determine the effect of nutrition on weights of uterine components, numbers of placentomes, fetal weight and size (adjusted to a common pregnancy rank (single vs twin)). Data are expressed as least square means and standard errors for the three ewe treatment groups and their fetuses. Statistical analyses were conducted using the computer package 'SAS' (SAS, 1988).

RESULTS

Figure 5 shows the change in live weight for the three treatment groups from day 21 (end of mating) through to day 101 of gestation (average slaughter date). Average live weights for the three treatment groups were significantly different (P<0.001) for all live weights recorded from day 35 of gestation onwards. The final live weight difference between the H group and the L group was 23 kg, with ewe live weights increasing by -16.8%, 0.02% and 21.4% in the L, C and H groups respectively over the trial period.

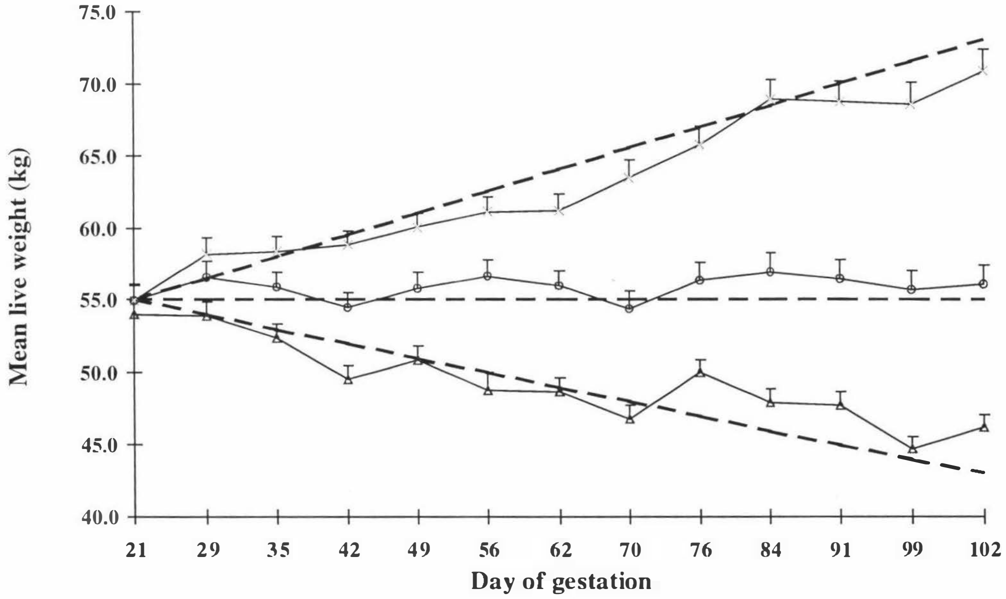

Figure 5: **Live weights of the three treatment groups, H (-x-), C (-o-), and L (-Δ-). The vertical bars represent standard errors, and the dashed lines are the target live weights for the three groups.**

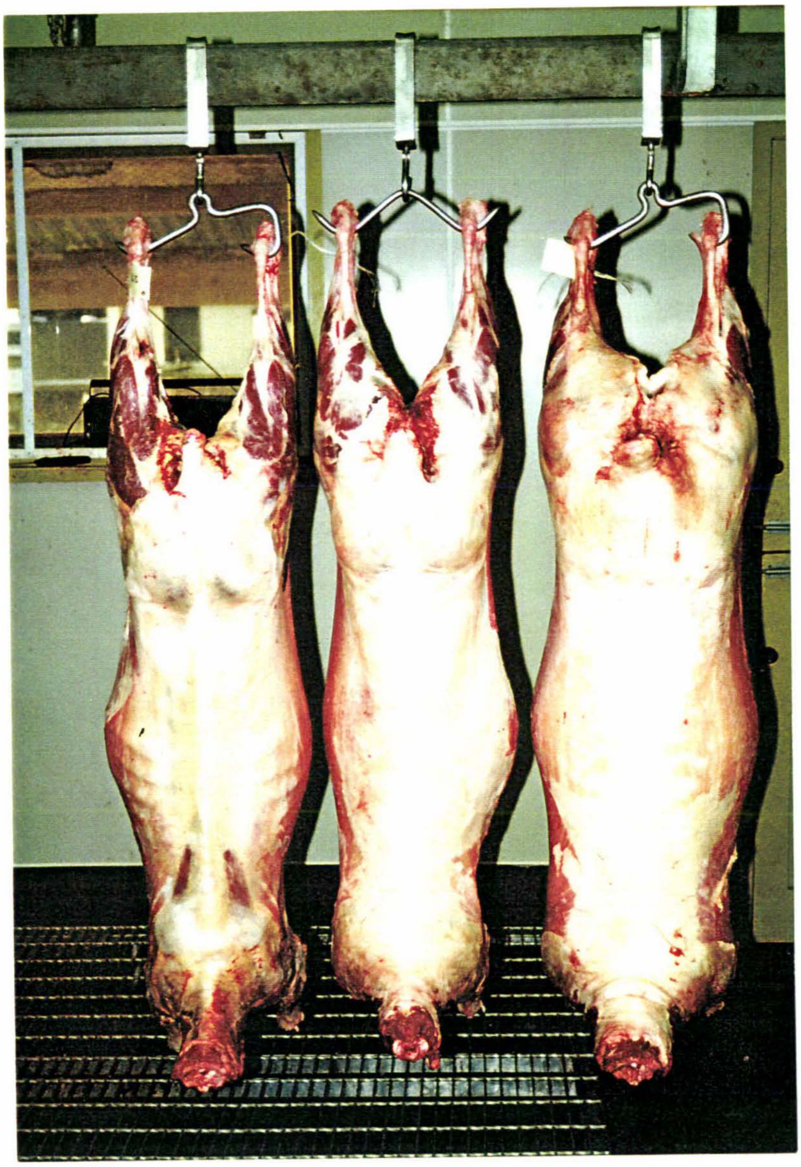

Figure 6: Carcasses from (left to right) L (16 kg), C (22 kg) and H (39 kg) groups.

Figure 6 graphically shows the difference in carcasses from the three treatments groups. Final maternal live weights, carcass weights, and carcass characteristics are presented in Table 3. High levels of nutrition increased and low levels of nutrition decreased, both final live weights and carcass weights compared to the C group ewes ($P<0.001$). However, while high nutrition led to increased dressing out percentages and fatness measurements (GR and c), these parameters were not significantly different in the L group ewes, compared to the C group ewes.

Table 3: Effects of nutritional treatment on final ewe live weight, carcass weight, and characteristics of the chilled carcass (Mean±SE).

	Group		
	Low	Control	High
n	19	20	19
Final live weight (kg)	46.1 ± 1.5^{a}	56.8 ± 1.4^{b}	69.1 ± 1.4^{c}
Carcass weight (kg)	17.0 ± 1.0^{a}	21.6 ± 1.0^{b}	30.1 ± 0.9^{c}
Dressing-out (%)	36.6 ± 0.9^{a}	37.8 ± 1.4^{a}	43.5 ± 0.8^{b}
Left GR (mm)	4.4 ± 1.8^{a}	7.9 ± 1.8^{a}	25.5 ± 1.7^{b}
Right GR (mm)	4.2 ± 1.9^{a}	7.7 ± 1.8^{a}	25.3 ± 1.7^{b}
Left c (mm)	3.0 ± 1.3^{a}	4.8 ± 1.0^{a}	13.1 ± 1.0^{b}
Right c (mm)	3.3 ± 1.3^{a}	4.8 ± 1.2^{a}	13.9 ± 1.2^{b}

^{abc} Means in the same row with different superscripts are significantly different (P<0.05)

Table 4 shows the effect of nutrition on placental components. H group ewes had similar weights of the gravid uterus, myoendometrium and fetal membranes (excluding cotyledons) to C group ewes, but a greater (P<0.05) weight of the uterus (minus fetus and fluids). A high plane of nutrition was (compared to C group) also associated with increased (P<0.01) total and average placentome weights, but a lower (P<0.05) number of placentomes and caruncle occupancy. The greater total placentome weight in the H group ewes reflected an increase (P<0.05) in total cotyledon weight, but no difference in the total weight of the caruncles.

Compared to the C group ewes, L group ewes exhibited lower (P<0.05) total gravid uterus weights and weights of the myoendometrium and fetal membranes. However, a low plane of nutrition did not reduce uterus weight (minus fetus and fluids), the total weight or number of placentomes, or total weights of the cotyledonary and caruncular components of the placentomes.

Table 4: **Effects of nutritional treatments on placental components at day 101 (±3 days) of gestation (Mean±SE).**

	Group		
	Low	Control	High
n	19	20	19
Weight (g)			
Gravid uterus	4010.9 ± 138.4^{a}	4376.8 ± 132.6^{b}	4422.9 ± 128.3^{b}
Uterus (minus fluids & fetus)	1242.8 ± 69.9^{a}	1290.7 ± 67.0^{a}	1475.4 ± 64.8^{b}
Myoendometrium (minus caruncles)	360.1 ± 13.2^{a}	376.4 ± 12.7^{b}	400.0 ± 12.2^{b}
Fetal membrane (minus cotyledons)	196.3 ± 10.8^{a}	219.4 ± 10.4^{b}	288.6 ± 10.0^{b}
Total placentome	584.9 ± 32.0^{a}	631.0 ± 30.7^{a}	702.9 ± 29.7^{b}
Average placentome	6.3 ± 0.3^{a}	6.1 ± 0.3^{a}	7.4 ± 0.3^{b}
Total caruncle	119.6 ± 8.4	126.9 ± 8.1	133.6 ± 7.8
Average caruncle	1.26 ± 0.08^{a}	1.25 ± 0.07^{a}	1.42 ± 0.07^{b}
Total cotyledon	470.1 ± 26.6^{a}	497.3 ± 25.5^{a}	571.8 ± 24.7^{b}
Average cotyledon	5.0 ± 0.3^{a}	4.9 ± 0.2^{a}	6.0 ± 0.2^{b}
Number			
Total placentomes	98.4 ± 3.4^{b}	102.6 ± 3.2^{b}	93.4 ± 3.1^{a}
Total caruncles	116.0 ± 4.9	125.7 ± 4.7	127.5 ± 4.5
Caruncle occupancy[1]	84.0 ± 2.6^{b}	82.1 ± 2.4^{b}	73.3 ± 2.4^{a}

[1] Number of placentomes/number of caruncles

[ab] Means in the same row with different superscripts are significantly different (P<0.05)

Table 5 compares the differences in fetal components between the three nutritional treatments. High levels of nutrition increased (P<0.05) fetal weight and chest circumference (P<0.01) compared to those from the C group, but had no effect on CRL or fetal:placental weight ratio.

Low nutrition resulted in reduced (but non-significant) fetal weight and chest circumference, when compared to those from the C group, although CRL was reduced (P<0.05). Nutritional levels had no effect on fetal:placental weight ratio.

Table 5: **Effects of nutritional treatments on fetal weight and other fetal components at day 101 (±3 days) of gestation (Mean±SE).**

	Group		
	Low	Control	High
n	28	30	29
Fetal weight (g)	1249.9 ± 40.6^a	1280.8 ± 38.0^a	1379.8 ± 35.2^b
Fetal weight/placental weight	2.7 ± 0.1	2.6 ± 0.1	2.6 ± 0.1
Crown rump length (mm)	37.6 ± 1.1^a	40.5 ± 1.0^b	40.6 ± 0.9^b
Chest circumference (mm)	23.6 ± 0.3^a	23.8 ± 0.2^a	24.7 ± 0.2^b

[ab] Means in the same row with different superscripts are significantly different ($P<0.05$)

No significant interactions were found between the effects of nutrition and birth rank in any of the parameters measured.

DISCUSSION AND CONCLUSIONS

This research studied the effect of maternal nutrition on fetal and placental growth and development in Romney ewes, throughout early and mid-gestation (days 21 to 101). The following hypotheses were developed and tested:

- Early placental growth and development can be influenced via maternal nutrition from day 21 to 101 of gestation.
- Maternal nutrition at 1.5 times maintenance from day 21 to 101 of gestation increases fetal weight.
- Maternal nutrition at 0.5 times maintenance from day 21 to 101 of gestation decreases fetal weight.

This study showed that an increase in maternal nutrition during the first 101 days of gestation increased fetal and placental weights, with maternal undernutrition decreasing fetal and placental weights to a much lesser (and generally non-significant) extent. Thus, the first and second hypotheses tested were accepted, because fetal growth was affected in conjunction with placental development via differing levels of maternal nutrition, and increased maternal nutrition significantly increased fetal weight at day 101 of gestation. However, the third hypothesis was rejected because decreased maternal nutrition did not significantly reduce fetal weight at day 101 of gestation.

Target ewe live weights were achieved throughout the trial period. All three treatment groups followed the planned pattern of liveweight change, with only minor deviations (see Figure 5). Over the 80 day trial period, a liveweight difference of 23.0 kg was achieved between the H and L treatment groups, a large difference to be generated in a pastoral based trial. Throughout the experiment, ewe live weights in the L, C and H groups changed by -16.8%, +0.02% and +21.4% respectively. These figures indicate a reliable level of control of all three groups. Managing ewes at pasture in order to achieve targeted liveweight differences at various stages during gestation can be difficult due to the remarkable buffering ability of the dam. For example, pregnant ewes managed at pasture specifically to lose live weight, may still manage to maintain their maternal live weight and even provide for fetal weight gain and development through changes in body composition. Indoor trials, where individual ewe feed allowances and intakes can be measured more precisely, are a more effective way of achieving the anticipated results, as external factors such as temperature, physical activity, feed levels, and dietary consumption can be more rigorously controlled. Nonetheless, the liveweight changes achieved allowed a good test of the hypotheses, in contrast to the second year of the study reported in Chapter Two.

Whereas uterine and total placentome weights were significantly heavier as a result of increased maternal nutrition, decreased levels of maternal nutrition did not significantly reduce uterine weight, or total and average placentome weights, when compared to the C group ewes. Although L group ewes exhibited (non-significantly) lighter placentomes, a greater number were formed (or fewer were lost), and therefore they achieved a higher caruncle occupancy than the H group ewes. Thus, the L group compared to the H group, utilised 84% versus 73.3% of potential fetal:maternal exchange points and this difference may have arisen during placental formation over the first 30 days of gestation. However, it is also possible that the L and H group ewes formed the same number of placentomes, but the L ewes maintained a higher number of fetal:maternal exchange points as compensation against undernutrition. The possibility that the weight and function of the fetal:maternal exchange points in the L group could be enhanced by an increase in maternal nutrition from day 100 to term, and therefore result in improved fetal growth and higher birth weights, is a factor to be further considered. Mellor (1983) showed that there is a strong association between fetal weight and placental weight in late gestation, with placental weight accounting for 69 to 91% of the variation in fetal growth. The relationship between the number of placentomes and their weight after day 90 of gestation has been examined in a number of studies (Alexander, 1964a; Rhind *et al.*, 1980; McDonald *et al.*, 1981) which observed decreased placentome number and total weight, but an increase in the weight of individual placentomes. This relationship was not observed in any of the treatment groups in the present study.

It has been suggested that impaired fetal growth in lambs is related to low placental weights (Jenkinson *et al.*, 1995), with the placenta being recognised as a major determinant of fetal growth (Mellor and Matheson, 1979). Thus, with the majority of placental development occurring during early gestation and peaking at day 90, altering placental development via maternal nutrition during this period could have a significant impact on fetal growth and development.

The differences in fetal weights (adjusted for birth rank) between groups are most likely to have occurred due to the large between-group live weight differences which were generated as a result of the differing levels of maternal nutrition. Increased maternal nutrition resulted in significantly increased fetal weights and chest circumference compared to those from C group ewes. Although decreased maternal nutrition resulted in lighter and physically smaller fetuses (CRL and chest circumference), these results were not significant (except for CRL). This result suggests that unless the maternal live weight is reduced by more than 16.8% during the first 101 days of gestation, fetal weight and chest circumference will not be significantly reduced when compared to fetuses from ewes fed to maintain maternal live weight. However, Everitt (1964) demonstrated that a 12% decrease in gross body weight of Merino ewes from mating to day 96 of gestation, led to significantly decreased fetal weights, while Wallace (1948) showed that a 7% decrease in dam live weight had no detrimental effect on fetal weight at day 90 of gestation. The fact that Everitt's (1964) results were not consistent with those of this study could be due to the fact that his study, and that of Wallace (1948), did not include a control group with which to compare results from the High and Low ewe treatment groups. The longer term implications of a 16% decrease in maternal live weight during the first 101 days of gestation are unknown. One possible implication of severe liveweight loss during early gestation is that the dam may be less able to nutritionally provide for the rapid development of the fetus during the latter stage of gestation, and therefore be less likely to produce a healthy viable lamb.

In the present study, the effects of varying levels of maternal nutrition after day 101 of gestation on fetal and placental development were not examined. This planned limitation leads to the following questions:

- What effect would reversing the H and L nutritional treatments from day 101 of gestation to term (i.e. differentially partitioning a given amount of feed between the two stages of gestation) have on fetal and placental components, and birth weights?
- What is the effect of such large nutritional differences during gestation on ewe wool growth and production?
- Would severe maternal undernutrition affect the ability of the fetus to deposit brown adipose tissue?
- Would lactational performance of the ewe, and therefore postnatal performance of her offspring, be affected by marked differences in nutritional levels during different stages of gestation?

Because there are as yet no clear answers to these questions, new areas of potential study are suggested. A further experiment following a similar design to the present one, but with the duration of the experiment extended to day 145 of gestation, with the treatments being split, reversed or both, at day 101, could help to further clarify these relationships. The proposed experimental design would allow for a number of ewes to be slaughtered at day 101 and at term, and the remainder carried through to parturition. A study designed in this way would allow the effects of nutrition on fetal and placental growth and development to be studied at both early to mid-gestation (day 101), and at late gestation (day 145). Leaving a number of ewes to carry their fetus(es) to term would allow the effects of maternal nutrition during gestation on birth weight and on lactational performance, to be studied.

While underfeeding cattle during the first half of gestation did not decrease birth weight (Chapter Two), it may have caused an unobserved difference in fetal weight at day 140 of gestation. The clear difference between the sheep and cattle studies was that the cattle did not lose any maternal live weight throughout the trial. For example, the L group ewes lost on average, 16.8 % (7.9 kg) of their maternal live weight during the first 101 days of gestation while, in comparison, the L group cattle (which were young females and therefore fed to allow for their own growth) gained an average of 23.8 % (32 kg) live weight during the first 140 days of gestation (averaged over two years). Therefore, any decrease in fetal weight which may have occurred in the cattle study at day 140 of gestation, could have been reversed by a combination of increased maternal nutrition from day 140 onwards, and the ability of the dam to compensate against under-nutrition which occurred during early gestation. It has also been suggested that the effects of a nutritionally retarded placenta on fetal development during early gestation can be reversed by way of increased maternal nutrition during late gestation. Had there been a difference in fetal weight at day 140 of gestation, the compensatory action by the placenta could have masked any difference in fetal weight that occurred at day 140 of gestation, either as a direct, or indirect result of maternal undernutrition. Thus, the combination of under-feeding during early, and over-feeding throughout late gestation, may have resulted in placental compensation during the latter half of gestation, thereby masking any difference in fetal growth at day 140 of gestation.

Contrary to the belief that early gestation is the least important period with regard to maternal nutrition, the sheep study in particular has effectively proven that maternal nutrition during early gestation is an important factor influencing fetal and placental growth. The study successfully demonstrated that increases in fetal weight and placental components are associated with, and likely to be the result of, increased levels of maternal nutrition throughout early to mid-gestation. The general understanding by farmers and researchers alike, is that the first 100 days of gestation is a period during which the fetus places little or no nutritional demand on the dam, and therefore maternal nutrition levels are relatively unimportant during this period. This misconception arises from the fact that only 20% of fetal growth and development is achieved in the first 100 and 190 days of gestation in sheep and cattle respectively.

Although the nutritional requirements of the fetus during early gestation are minimal, it is now clear that fetal growth is influenced either directly by levels of maternal nutrition, or by placental development which can in turn be influenced by maternal nutrition. Therefore, the results discussed here could potentially influence the way in which New Zealand farmers nutritionally manage their stock throughout gestation. Thus in relation to the findings of the present study, the following recommendations could be made to New Zealand farmers.

In New Zealand, the first 100 days of gestation in sheep fall over the months of March, April, May and June, a period where farmers could potentially utilise the late summer/autumn pasture, before pasture growth and nutritive value declines heading into colder winter months. Consequently, if farmers increase maternal nutrition levels significantly throughout the late summer and autumn season, fetal growth may be significantly increased by mid-gestation. High levels of feeding during the first half of gestation may not only increase fetal weight, but may also increase body reserves of the dam, and therefore enhance her ability to buffer against under-nutrition during late gestation. By utilising her own body reserves to maintain fetal growth throughout late gestation, the dam ensures that the nutritional demands for exponential growth by the fetus throughout this period, will be adequately provided for. Since the late gestation period occurs over the winter months when pasture growth is minimal and sometimes supplementary feed is utilised, it would be highly beneficial to the farmer if they could feed pregnant animals less over this period.

Another management method which could be implemented alone, or in conjunction with the one suggested above, is the differential feeding of ewes according to the number of fetuses carried. Pregnant ewes should be fed a high plane of nutrition until pregnancy diagnosis (after day 50 of gestation), after which single- and twin-bearing dams can be separated into two mobs, and fed two levels of nutrition. Multiple fetuses place higher nutritional demands on dams, resulting in them being more sensitive to low levels of feeding throughout gestation than those dams which bear a single fetus. Therefore, by feeding multiple-bearing dams higher levels of nutrition during the first

101 of gestation, the farmer will be ensuring that growth of both fetuses is maximised by mid-gestation, while the principle of compensation by the dam described above, will apply throughout late gestation. This method places emphasis on the economic value of two healthy neonates versus one, and also ensures that dams carrying single fetuses are not those which are receiving high levels of nutrition at the expense of those bearing multiple fetuses. The economic implications of managing pregnant stock in this manner would impact when a higher number of heavier lambs survive and are weaned or sold, as a result of maternal nutrition levels throughout gestation.

Therefore, sheep and cattle industries would benefit alike if fetal size at parturition could be controlled to some extent by maternal nutrition levels during stages of gestation, without suppressing neonatal growth and development, and without reducing subsequent rebreeding performance of the dam.

APPENDIX

Calendar of events for the sheep study

Number of Ewes H, M, L,	Day	Dates	Task
150	-14	4-Mar-96	CIDR'S Implanted, Weigh
150	0	18-Mar-96	CIDR removal = 2 days; Put rams out; Weigh
150	02	20-Mar-96	Assumed conception date
150	07	25-Mar-96	Weigh; Change raddle on rams
150	22	09-Apr-96	Remove all 2nd cycles; Allocate to treatments
90-120	35	22-Apr-96	Weigh
90-120	42	29-Apr-96	Weigh; Pregnancy Diagnosis; Select 30 twins and 30 singles
60	49	06-May-96	Weigh
60	56	13-May-96	Weigh
60	63	20-May-96	Weigh
60	70	27-May-96	Weigh
60	77	03-Jun-96	Weigh
60	84	10-Jun-96	Weigh
60	91	17-Jun-96	Weigh
60	100	26-Jun-96	Weigh; Slaughter

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