**The Effects of Feed Composition and Level on
Lactational Performance in Rats and Dairy Cows:
A Basic Approach to Feed Description**

by

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To the rats, Mum and Baby.

They represent the good and the bad in
our relationship with animals

ABSTRACT

An investigation into the effects of feed composition on lactational performance was carried out using rats and cows. A graphical representation of the feed as a triangle was used to aid the interpretation of results. The first rat experiment showed that, on high protein feeds, the lactational performance of rats is not depressed when offered feeds of very low carbohydrate content. This was substantiated by the other rat experiments. When carbohydrate in the feed was replaced by fat at low protein content (rat experiment 2) there was a large depression in lactational performance, effectively a cessation of milk production. The interaction between the three feed components protein, carbohydrate, and fat was highly significant. The hypothesis that maternal heat production was limiting food intake was advanced. The third rat experiment used feeds whose composition was marginal in relation to lactational success. The feeds also allowed comparison between feeds of constant nutrient:energy ratio. The results of this experiment indicated that there is an extremely abrupt threshold in feed composition for adequate lactation. This effect could not be attributed to any one nutrient:energy ratio. This experiment also showed the importance of maternal body reserves in support of lactation. A model was developed to explore the hypothesis that maternal heat production was limiting performance, however this model failed. An experiment using sheep was conducted in order to permit prediction of the volatile fatty acid proportions arising from a range of feeds. This experiment was designed to allow application of the rat work to dairy cows. A dairy cow trial was conducted, to compare different feed types and feeding levels. The results of this trial showed no effect of feed type on lactational performance. A linear relationship between food intake and level of milk production was found. This included an effect of feeding level on rate of decline in milk yield. All these findings are discussed in detail.

DECLARATION

I declare that this thesis has been written by myself. The experimental work and analyses were carried out by myself, with the assistance of members of the Edinburgh School of Agriculture which is duly acknowledged. The results and the analyses have not previously been submitted for any other degree or qualifications.

Nicolas Friggens

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Lang Tran did all the drawings in this thesis.

To all the people mentioned above I am deeply grateful.

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<u>List of abbreviations:</u>

The abbreviations used in the lactation model (chapter 7) are listed on pp. 88-90. The abbreviations used in appendices are listed in each appendix.

ADF	acid detergent fibre
ADL	acid detergent lignin
AEE	acid hydrolysed ether extract
C	carbohydrate (in the triangles)
c.o.	adjusted for carry over effect
calc.	calculated
CF	crude fiber
CHO	carbohydrate
coeff.	coefficient
covar.	covariate
CP	crude protein
d14LBP	litter body protein gain on day 14
d14MBP	maternal body protein gain on day 14
d2mCP	maternal crude protein gain on day 2
d2mEE	maternal ether extract gain on day 2
d2Mlwt	maternal liveweight on day 2
DE	digestible energy
DM	dry matter
DMI	dry matter intake
DMRQ	dry matter requirement to meet predicted metabolisable energy needs for the end of the experiment (chapter 9)
E	energy
EE	ether extract
evap.	evaporative
F	fat (in the triangles)
g	grams
GE	gross energy
iu	international units
kg	kilograms
kJ	kilo Joule
LBF	litter body fat gain
LBP	litter body protein gain

LWT	liveweight
main.	maintenance
MBF	maternal body fat gain
MBP	maternal body protein gain
mCPg	maternal crude protein gain
ME	metabolisable energy
mEEg	maternal ether extract gain
MFC	milk fat content
MJ	mega Joule
MLC	milk lactose content
MLW	avarage maternal liveweight
MPC	milk protein content
MY	milk yield
N	nitrogen
n.s.	non-significant
NDF	neutral detergent fibre
NDFIP	neutral detergent fibre insoluble protein
NPNL	non-pregnant non-lactating
OM	organic matter
P	protein (in the triangles)
prot.	protein
R1	the first rat experiment
R^2	percentage of variation in Y accounted for
R2	the second rat experiment
R^2adj.	R^2 adjusted for deviant points
R3	the third rat experiment
R4	the fourth rat experiment
s	residual standard error
S.D.	standard deviation
se	standard error
sed	standard error of the difference
SPF	specified pathogen free
t	students t ratio
temp.	temperature
VFA	volatile fatty acid
vit.	vitamins

CHAPTER 1

General Introduction

Note: All tables and figures are numbered according to the page on which they are found. As such table and figure numbers are not consecutive (see list of tables and figures, p. ix).

Overview

This thesis describes four feeding experiments with lactating rats, one with lactating dairy cows, and an investigation of rumen fermentation in sheep. The choice of species used and of experimental design were based *in part* on the available facilities, the cost, and the academic environment. I make no apology for these realities, indeed they play their part in shaping this study. Experiments were chosen to develop a set of ideas, the aim of which was to explore the effect on milk production of differences in feed composition.

In this work, the feed components studied were the main nutrients; protein, carbohydrate, and fat. The effects of variation in the mineral and vitamin content of feeds for lactation were not examined.

Lactational performance was measured as input to and output from the lactating mother, using (where possible) the same description as for the composition of the feed; protein, carbohydrate, and fat. More complex physiological and biochemical measures, such as hormonal and enzymic changes, were not made. It was decided that the number of measures of this kind necessary to augment the knowledge gained from the more simple, holistic measures, was beyond the scope of available resources, mental and physical.

The Limitation of Applying Conventional Feed Descriptions to Lactating Animals

Lactation is the only physiological state in which mammals produce large amounts of carbohydrate, usually in form of milk lactose. Despite this, feeds for lactating animals are usually described in terms of protein and energy content alone. Implicit in this two component description of feeds is the assumption that carbohydrate and fat are of equal value for lactation in energy terms. Since fat cannot be converted to carbohydrate in the body, this assumption is only valid if two conditions apply:

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1) That carbohydrate supply is not limiting for milk lactose production.

2) That feed carbohydrate content does not influence lactational performance other than by lactose production.

These two conditions are discussed below, mainly in relation to rats and dairy cows, the species studied subsequently.

Sufficiency of Carbohydrate Supply for Lactose Production

Estimates of the carbohydrate requirement for maintenance and for milk production are given for several species in table 3. These values incorporate estimates of the inefficiency of carbohydrate metabolism in meeting these requirements. Since the major component (of the digestible portion) of most plants and their products is carbohydrate, the majority of animal feeds are not limiting in carbohydrate content. There are however two exceptions to this. Feeds containing a high proportion of animal products or of added fat may be limiting in carbohydrate content, as may feeds for lactating ruminants.

Table 3: Estimated glucose requirements for maintenance and milk production in rat, pig, sheep, and cow.

	Maintenance carbohydrate (g/d)	Milk carbohydrate (g/d)	Total yield (kg/d)
Rat	0.25	0.59	0.03
Pig	200	302	7
Sheep	110	150	3.9
Cow	242	1738	35

Estimates derived from:
Rats: Donaldson, 1915; Brody and Nisbet, 1938; Luckey et al, 1955; Canas, 1974; Dahlquist and Persson, 1976.
Pigs: Elsley, 1970; Mullan et al, 1989; Emmans, 1990.
Sheep: Jenness and Sloan, 1970; Wilson et al, 1983.
Cows: Young, 1977; Girdler et al, 1986.

The advantage of being able to digest cellulose conferred on ruminants by the microbes in the rumen, carries the penalty that the major end-products of rumen fermentation are lipogenic. As a consequence of this there is some evidence to suggest that particularly in high yielding dairy cows, the supply of glucogenic carbohydrate may limit milk production (Frobish and Davies, 1977; Vik-Mo et al, 1974). Thus in some situations condition 1; that carbohydrate supply is not limiting for lactose production, may not apply.

Interaction between Feed Carbohydrate Content and Lactational Performance

In non-ruminant mammals, there are few experiments which offer direct evidence of interactions between feed components affecting milk production. There are however experiments which indicate that feeds of high carbohydrate content may be detrimental to lactational performance (Maynard and Rasmussen, 1942; Naismith et al, 1982). These effects, not attributable to influences on lactose production, are described in greater detail in chapter 2.

In ruminants, interaction between the type of feed carbohydrate and lactational performance has been observed frequently. Increasing the proportion of starch in the feed may result in a depression in milk fat concentration, particularly in feeds which have a high cereal content and are rapidly fermented (Sutton et al, 1987). Feeds which are rapidly fermented generally yield more glucogenic end-products than do slowly fermented feeds. Additionally, rapid fermentation tends to alter the rumen environment such that lipogenic end-products are reduced (Jorgensen and Schultz, 1963). Milk fat depression may therefore be explained by changes in the rumen. However some studies in which glucose has been infused post-ruminally have also found a depression in milk fat production (Storry and Rook, 1965; Frobish and Davies, 1977). Thus condition 2; that feed carbohydrate content does not influence lactation beyond its effect on lactose production, may not always apply.

Both of the conditions necessary for the assumption that feed carbohydrate and feed fat can be equated in energy terms do not apply in a number of circumstances; this is

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particularly so for ruminants. For this reason, the work described in this thesis was directed towards the role of feed carbohydrate in lactation. In order to investigate this adequately in ruminants, it is important to be able to measure or predict accurately the proportions of glucogenic and lipogenic end-products arising from the fermentation of different feeds. Measurement of these end-products requires complex techniques with fistulated animals and prediction at present incorporates too many uncertainties (Sutton, 1985). Because of these difficulties I chose to work initially with a non-ruminant species, the rat. Subsequently the approach was extended to a study with lactating cattle. Additional work was designed to approach the problem of estimating the proportions of glucogenic and lipogenic end-products arising from feedstuffs offered to ruminant animals. The first six chapters of this thesis are concerned with the effects of feed composition on lactational performance in rats. The remainder of the thesis addresses this subject in ruminants.

The Effect of Differences in Task Complexity on
Variation of Performance in the Rat Tested at...

*Things are not, as soon as I took my place at the table, would start up my leg and
on the table and, if not vigilantly watched, would carry off the bread, or a piece of
cheese, which it would nibble a little, and leave the remainder...*
(Moor, 18..)

Note: All tables and figures are numbered according to the page on which they are
found. As such table and figure numbers are not consecutive (see list of tables and
figures, p. ix).

The Effect of Differences in Feed Composition on
Lactational Performance in the Rat: The Approach

"Scugg, the rat, as soon as I took my place at the table, would run up my leg, get on the table, and, if not vigilantly watched, would carry off the sugar, pastry, or cheese, of which it would nibble a little, and leave the rest to Flora..."

(Moss, 1836)

Note: All tables and figures are numbered according to the page on which they are found. As such table and figure numbers are not consecutive (see list of tables and figures, p. ix).

Introduction

When considering the role of feed carbohydrate content on the lactational performance of rats, two important points are raised.

Firstly, there are relatively few experiments designed specifically to investigate the effects of feed carbohydrate in rats. Additional information from other non-ruminant species is lacking, the two main species which have been studied are humans and pigs. Results from human lactational studies are generally incomplete because of the obvious experimental limitations; the reliability of indirect measurements has been questioned (Singh et al, 1989). Whilst there are a number of trials investigating the lactational requirements of sows (Mullan et al, 1989), these are almost exclusively in terms of energy and protein alone, and thus not suitable within the present context.

Nearly all of the experiments discussed subsequently were not designed with the objective of investigating the role of feed content on lactational performance. As such, they may have been described in different terms to those of the authors. In particular, some re-calculation of feed compositions has been made using the values given in table 7.

Table 7: Values used for re-calculation of feed compositions (g/kg OM unless otherwise stated), derived from the ingredient composition in chapter 3.

	Casein	Fats, and Oils[a]	Starch, and Sugar	Wheat flour	Skimmed milk powder[b]
Crude protein	964	0	0	129	382
Ether extract	5	1000	0	20	10
Carbohydrate	32	0	1000	851	607
Ash (g/kg DM)	19	0	0	20	86
GE (kJ/g OM)	23.7	39.6	16.5	18.2	19.5

a: Groundnut -, vegetable -, corn - and cod liver oil, butterfat and lard. Source: MAFF (1986). b: Source: McDonald et al. (1981).

Secondly, it is clear that when comparing feeds of different carbohydrate content the compensating difference in the other feed components must be considered. For instance, "successful" lactation has been observed in rats on very low carbohydrate feeds (140 g/kg OM; Steingrimsdottir et al, 1980) and even on a carbohydrate free diet (Follis and Straight, 1943). In these experiments, the low carbohydrate content was achieved by high inclusion of protein (270 and 450 g/kg OM respectively), providing an alternative source of glucose. Experiments which offered lactating rats feeds of low glucogenic potential, i.e. low carbohydrate *and* low protein content, do not exist. Therefore, subsequent discussion in this chapter does not consider the effect of feed carbohydrate as a limitor of lactose production.

In order to clarify the interactions between feed components a graphical description of feeds was used in the design of the rat experiments. This description and the discussion of the relevant literature are presented under the following headings:

- A Triangular Description of Feeds.

- The Effects of Substituting Feed Carbohydrate for Feed Protein on Lactational Performance.

- The Effects of Substituting Feed Carbohydrate for Feed Fat on Lactational Performance.

- Interactions Between Feed Components on Lactational Performance.

A Triangular Description of Feeds

The results presented above emphasize the problem of attempting to consider one feed component alone. Effectively there is no such thing as a "carbohydrate experiment" since variation in feed carbohydrate content forces a change in the content of at least one other component. There are however "carbohydrate/protein experiments" and "carbohydrate/fat experiments". A useful description of feed mixtures which helps to clarify this has long been used by geologists (Rankin and Wright, 1915). It has been comprehensively described for nutrient mixtures in mathematical terms by Parks (1982), and has been applied by Krishmanachar and Canolty (1985).

For a mixture of three components whose sum is unity, such as carbohydrate, protein, and fat in the digestible organic matter of a feed, the graphical representation is an equilateral triangle (figure 10). Any point on the triangle represents a feed consisting of some combination of the three components. The apices of the triangle represent feeds consisting only of the component named at that apex. Any point on the edge of the triangle represents feeds consisting of only two components, those named at the apices which that edge connects (figure 10). Thus the edge connecting the carbohydrate apex to the fat apex represents feeds of the same protein content; zero protein. Feeds of equal protein content lie along lines parallel to the carbohydrate-fat edge, with increasing protein content the nearer to the protein apex they are (figure 10). Similar lines drawn parallel to the protein-fat edge are isocarbohydrate lines, and lines parallel to the protein-carbohydrate edge are isofat lines. The visual representation of feeds in the triangle, emphasises the fact that changes in feed composition always involve at least *two* feed components.

Interactions between feed components on lactational performance have frequently been described in the literature. There is however, a need to distinguish between "real" and "apparent" interactions. Apparent interactions arise from inappropriate comparisons between feeds. These interactions are defined below, in terms of a mixture of three components A, B, and C, using the simplest substitutions:

An <u>apparent interaction</u> occurs when the effect of a change in the level of component A, substituting for component B, is different from the effect of the same change in component A, substituting for component C.

A <u>real interaction</u> occurs when the effect of a change in the level of component A, substituting for component B, is different from the effect of the same change in component A, substituting for component B at a different level of component C.

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Fig 10: A triangular representation of feeds consisting of three components.
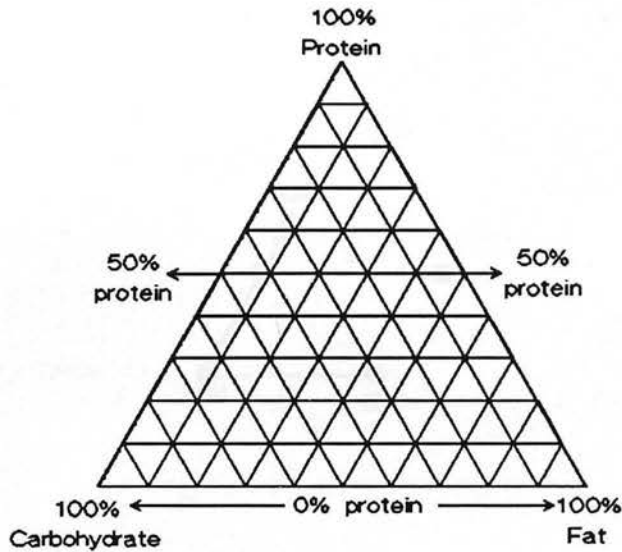
An experiment by Nelson and Evans (1947b) provides a simple example of an apparent interaction (figure 11). The feeds that they used are represented by the filled circles in figure 11 with the average pup liveweight gains (in grams from days 1 to 21 of lactation) presented in brackets. From figure 11 it can be seen that the decrease in feed carbohydrate content labelled 'x' caused a depression in pup weight gain, at constant feed casein (250 g/kg OM). When feed carbohydrate content is decreased to the same extent by comparison 'y' there is no significant depression in pup weight gain. This is an apparent interaction between carbohydrate and protein since the changes in feed composition, x and y are clearly of a different nature and thus not comparable.

To test for a real interaction between carbohydrate and protein the effect of a decrease in carbohydrate content x should be compared with the same decrease in carbohydrate content at a different protein content, for example 'z'. Both x and z being the same *change* in feed composition. The advantage of this graphical representation is that the difference between these comparisons becomes immediately apparent.

Fig 11: A triangular representation of feeds consisting of three components showing the treatments from Nelson and Evans (1947b).
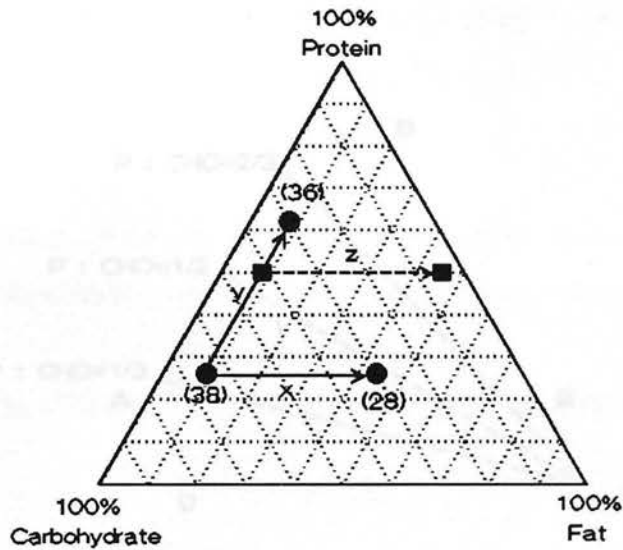
The feeds from this experiment have been expressed in proportions of digestible organic matter. This was done so as to exclude minerals and indigestible matter from the feed description, maintaining a three component description. The number of dimensions necessary to represent a mixture is equal to the number of components in that mixture less one. Whilst it is perfectly possible to describe a ten component mixture mathematically, it does create a problem of visual representation.

As well as clarifying the interrelationships between nutrients, this representation allows the interrelationships between nutrients and energy in the feed to be more easily seen. Examples of lines of constant nutrient:nutrient ratio and of constant nutrient:energy ratio are shown in figures 12a and 12b. In these figures, line AB represents substitution of feed carbohydrate for fat at constant protein content and line CD represents substitution of feed carbohydrate for protein at constant fat content. There are three main types of alteration in the feed composition of lactating rats which are described in the literature. The first two are represented by lines AB and CD; these are discussed in

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Fig 12a: A triangular representation of feeds
showing substitutions at constant fat (CD) and
at constant protein (AB). The broken lines are
lines of constant protein : carbohydrate.

Knowledge of the effects of single substitutions of one feed component for another is a prerequisite for understanding the effects of mixtures of components.

The Effects of Substitutions of Carbohydrate or Fat for Protein on Lactational Performance

Protein cannot be created from carbohydrate or fat alone, and the stores of protein in the body are small relative to the stores of fat. For lactating rats (exercise 8, feed protein 182 g/kg fresh DM), Naismith et al (1982) reported maternal protein losses of 0.13-0.3 from an initial body protein of 37 and fat losses of 21.6 g/1d from an initial 33.5g. The body stores of protein are depleted after reductions in dietary protein supply below requirements, as demonstrated by Mueller and Cox (1946, figure 14a). The manner in which lactating rats respond to a reduction in feed protein content, may provide information about the factors controlling their food intake.

Fig 12b: A triangular representation of feeds
showing substitutions at constant fat (CD) and
at constant protein (AB). The broken lines are
lines of constant protein : energy (mg/kJ).

In an experiment substituting casein for carbohydrate in the range 120 to 450 g casein/kg DM, Nelson and Evans (1958) replicated the results of Mueller and Cox (1946) as shown in figure 14a. In addition they measured maternal food intake and weight change (figure 14b). Below a certain protein concentration maternal weight gain declined and maternal liveweight loss increased. The lactating rat appeared to be trying to compensate for inadequate feed protein by eating more of the diet (figure 14b). However, the maternal food intake also declined, increasing the inadequacy of the protein supply (figure 14b). This is interpreted if the large claims that lactating rats are able to compensate for dilution of their protein supply by increasing their intake (Munson et al, 1974). Further, lactating rats are able to compensate for the increased demand of

subsequent sections of this chapter. The third is dilution of stock feed with a single feed component. In this type of alteration, all three feed component proportions are changed, making it difficult to attribute any observed effects to specific aspects of the total change in feed composition. For this reason, trials of this nature have not been discussed.

Knowledge of the effects of simple substitutions of one feed component for another is a prerequisite for understanding interactions between feed components.

<u>The Effects of Substituting Feed Carbohydrate for Feed Protein on Lactational Performance.</u>

Protein cannot be created from carbohydrate or fat alone, and the stores of protein in the body are small relative to the stores of fat. For lactating rats (littersize 8; feed protein 104 g/kg fresh OM), Naismith et al. (1982) reported maternal protein losses of 6g in 14 days from an initial body protein of 46g and fat losses of 21.6 g/14d from an initial 35.3g. The lactating rat is therefore vulnerable to depression in dietary protein supply below required levels as shown by Mueller and Cox (1946; figure 14a). The manner in which lactating rats respond to changes in feed protein content, may provide information about the factors controlling their food intake.

In an experiment substituting casein for sucrose over the range 120 to 480 g casein/kg DM, Nelson and Evans (1958) replicated the results of Mueller and Cox (1946) as shown in figure 14a. In addition they reported maternal food intake and weight change (figure 14b). Below a casein content of 250 g/kg DM pup liveweight gain declined and maternal liveweight loss increased. The dams were evidently trying to compensate for inadequate feed protein intake by mobilising body reserves. However, the maternal feed intake also declined exacerbating the inadequacy of the protein supply (figure 14b). This is unexpected since it has been shown that non-lactating rats are able to compensate for dilution of their protein supply by increasing their intake (Musten et al, 1974). Further, lactating rats are able to compensate for the increased demand of

Fig 14a: The effect of changing feed protein content on litter growth.

Av. pup weight on day 21 (g)

Feed casein content (%)

—+— Mueller and Cox (1946)   —△— Nelson and Evans (1958)

Fig 14b: The effect of changing feed protein content on maternal intake and weight change (Nelson and Evans, 1958)

Av. daily food intake (gDM)

Maternal weight change (g/21d)

Feed casein content (%)

—+— Food intake   —△— Weight change

is rather by choice than necessity, in view of the subsequent feeding experiments (Hansen and Heggset 1969). These results suggest that the lactating rats on low .... also but were constrained from so doing.

.... the nature of this study and .... on a key experiment by Nasmith and co-workers (1958). They compared two feeds, a high protein (239 g/kg fresh DM) and a low protein feed (104 g/kg fresh DM), substituting casein for starch. Feeds were offered ad libitum to dams nursing litters of 8 pups. The low protein group ate 29 % less than the high protein group, despite being clearly in protein deficit (table 15). As in the above work (Nelson and Evans, 1958), the low protein group appear to have had their intake constrained .... food. Presumably, intake became constrained as a result of the change in feed composition. The excess intake per unit protein on the low protein feed was relatively high. If the energy content of the feed was constraining intake, it would be expected that the low protein group would sustain an excess load .... equal to that of the high protein group. .... low protein group .... only increased their .... If .... low protein group sustained a large amount of .... amount of dietary energy absent .... the energy derived from body lipid mobilization was equivalent to the (non-protein) energy coming from 50g of feed. Had that energy been derived from feed rather than body lipid, it would have supplied .... 50g of extra needed protein. It would ....

Table 15: Feed intake (g fresh), litter gains and maternal weight changes (g DM) over 19 days of lactation (day 2 to 19), from Nasmith et al. (1982).

Feed	HP	LP
Protein content (g/kg fresh DM)	239	104
Feed intake	405	240
Protein intake	105.3	53.1
Litter protein gain		13.0
Litter fat gain		10.2
Maternal protein gain		-2.6
Maternal fat gain		-21.0

....

lactation by choosing a higher proportion of casein in choice feeding experiments (Richter and Barelare, 1938). These results suggest, that the lactating rats on low protein feeds wanted to increase their intake but were constrained from so doing.

Evidence as to the nature of this constraint comes from a key experiment by Naismith and co-workers (1982). They compared two feeds, a high protein (238 g/kg fresh OM) and a low protein feed (104 g/kg fresh OM), substituting casein for starch. Feeds were offered ad libitum to dams nursing litters of 8 pups. The low protein group ate 29 % less than the high protein group, despite being clearly in protein deficit (table 15). As in the above work (Nelson and Evans, 1958), the low protein group appear to have had their intake constrained. Presumably, intake became constrained as a result of the change in feed composition. The energy intake per unit protein on the low protein feed was relatively high. If the energy content of the feed was constraining intake, it would be expected that the low protein group could achieve an energy intake equal to that of the high protein group. Had they done so, they would have concurrently increased their protein intake by 48 %. Further, the low protein group mobilised a large amount of body fat (21.6g, table 15). This would have exacerbated a situation of dietary energy excess. The energy derived from body lipid mobilisation was equivalent to the (non-protein) energy arising from 56g of feed. Had that energy been derived from feed rather than body lipid, it would have supplied 4.8g of much needed protein. It would

Table 15: Food intake (g fresh), litter gains and maternal weight changes (g DM) over 14 days of lactation (day 2 to 16). From Naismith et al. (1982).

Feed	HP	LP
Protein content (g/kg fresh OM)	238	104
Food intake	485	346
Protein intake	105.1	33.1
Litter protein gain	31.6	13.8
Litter fat gain	27.9	10.2
Maternal protein gain	2.3	-5.8
Maternal fat gain	-19.7	-21.6

therefore appear that the energy content of the low protein feed was not constraining intake. In addition to the changes in casein content, these two feeds differed in their contents of starch and methionine. The high protein was methionine supplemented (2.5 g/kg feed) and the low protein feed was not.

Work by Drori and Folman (1973) with methionine supplementation, and by Grigor et al. (1987) without methionine, produced results similar to those of Naismith et al. (1982). These results suggest that the differences in methionine supplementation in the experiment by Naismith et al. (1982) did not cause the effect seen on the low protein feed. Grimble (1981) showed that lactating rats were sensitive to large changes in protein quality, and it is possible, that in the experiment of Naismith et al. (1982) differences in the absolute amino acid requirements of the dams arose from different levels of milk production on the high and low protein feeds. However, there is relatively little difference between the efficiency of casein use for maintenance (66%) and for milk production (69%; both calculated from Luckey et al, 1955; Kuzdzal-Savoie et al, 1980; Fuller et al, 1989), implying that protein quality was not affected by level of performance. Thus in the experiment of Naismith et al. (1982) it would appear that the changes in the carbohydrate content of the feed were important.

There are indications that carbohydrate in excess may have adverse effects on performance, and that there is an optimal ratio of carbohydrate to the other feed components. These indications come from two sources; from the work of Sainz et al. (1986) and from choice feeding experiments.

Sainz compared three levels of protein (148, 296, and 446 g/kg OM) in the feed of lactating rats at a *constant* fat:carbohydrate ratio. Rats were fed at different levels of intake, including *ad libitum*. As the protein content declined to 148 g/kg OM there was a small non-significant increase in food intake, In contrast to the decrease observed by Naismith et al. (1982). The increase in intake was not sufficient to compensate for the fall in protein content and consequently, the dams on the low protein feed lost the most body protein. However, the losses of body fat (by the ad libitum fed rats) were least on the low protein feed. Whilst there were only three rats fed ad libitum per feed, these

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results support the proposition that feed carbohydrate content may be important in the control of feed intake on low protein feeds. The role of the feed carbohydrate:fat ratio on lactational performance is explored in greater detail in the next section (p. 19) of this introduction.

It has been shown that when animals are allowed to choose from a range of feeds the mixture which they desire, it usually results in optimal performance (Richter, 1943; Kyriazakis, 1989). In an elegant experiment, Richter and Barelare (1938) offered a choice of casein, sucrose, and oil plus a range of mineral and vitamin solutions to dams through one reproductive cycle. At the onset of lactation there was a huge increase in protein intake in the form of casein and also in energy intake. These dams selected their extra energy intake largely in the form of oil, resulting in lactational performance equal to that of stock fed dams. Rolls et al. (1984) also found, that dams selected a food mixture with a decreased carbohydrate content when offered a choice of salami, crackers, cookies, and stock feed (table 18). Clearly, when allowed to, lactating rats seek a relatively low carbohydrate intake. As a consequence of choosing a diet of low carbohydrate content, the choice fed dams in the experiment of Rolls et al. (1984) were eating a feed of lower protein content than the stock fed dams. Despite this, their litters grew better than those of the stock fed dams. This was achieved in part by an average increase in intake of 28% (DM) of the choice fed, low protein group over the stock fed, high protein group. In this experiment, the low protein feed was of a lower carbohydrate content than the high protein feed. This higher intake by rats on a feed of lower protein content was contrary to the findings of experiments, in which low protein content was accompanied by a high carbohydrate content (Nelson and Evans, 1958; Drori and Folman, 1973; Naismith et al, 1982; Grigor et al, 1987). This indicates, that the carbohydrate content of the feed influenced the food intake of the rats on low protein feeds. The choice feeding experiments indicate, that lactating rats prefer oil or fat as an energy source over carbohydrate. Choice fed dams, eating a high fat/low carbohydrate mixture, had a lactational performance equal to that of the stock fed dams, but achieved with only 74% of the energy intake of the stock fed dams (Richter and Barelare, 1938). It would appear that the choice fed dams used feed energy with a

Table 18: The feed composition (g/kg OM) selected by lactating rats when offered a choice in relation to stock fed rats.

| | Richter and Barelare (1938) | | Rolls et al. (1984) | |
	stock[a]	choice	stock	choice[b]
Protein	235	337	232	151
Fat	67	397	29	338
Carbohydrate	698	266	739	511

a: stock composition from Wang (1925).
b: derived from Rolls et al. (1986); the choice was more restricted than in the experiment of Richter and Barelare.

higher efficiency, and therefore produced less heat than the stock fed dams. There is strong evidence from work with *growing* rats, that on feeds of low protein/high carbohydrate content (less than 100 g protein/kg OM) intake was restricted by the capacity of those rats to loose heat (Meyer and Hargus, 1959; Andik et al, 1963). It is therefore reasonable to suggest, that the same constraint applies to *lactating* rats on low protein/high carbohydrate feeds. If this is the case, then a feed which is more efficiently utilised for milk production would be advantageous, as it results in less heat production. In the experiment of Richter and Barelare (1938), the high fat feed appeared to be such a feed. Unfortunately, the data of this experiment do not include information on maternal body mobilisation, and so the reason for the apparent differences in efficiency cannot by defined. However, the experiments described in this section collectively suggest, that the carbohydrate content of the feed is important in determining the food intake of lactating rats.

In summary, the following effects on lactational performance of substituting feed protein for carbohydrate have been identified:

i) Decreasing feed protein content (as casein) below approximately 250 g/kg OM and concurrently increasing carbohydrate content results in a depression in litter growth.

ii) Lactating rats on low protein/high carbohydrate feeds could not compensate for their protein deficit by increasing their intake, indeed intake on these feeds was depressed.

iii) This constraint on food intake was not due to the energy content of the low protein/high carbohydrate feeds.

iv) When offered a choice, lactating rats avoided excess carbohydrate intake. A reduced carbohydrate content allowed dams on low protein feeds to maintain or increase their level of feed intake.

v) It is likely that the carbohydrate content of low protein feeds is an important determinant of intake, probably because of its contribution to maternal heat production.

The Effects of Substituting Feed Carbohydrate for Feed Fat on Lactational Performance

It has been suggested in the preceding section, that carbohydrate and fat are not of equal value as sources of energy (on an equal energy basis). This is examined further in this section by considering experiments, where the daily protein intake was constant and feed carbohydrate has been substituted for fat.

In an experiment carried out by Loosli and co-workers (1944) lactating dams, each nursing 6 pups, were rationed one of four feeds containing 608, 712, 777, or 874 g/kg OM of carbohydrate. The four groups were intended to have equal energy and protein intakes; however this was only the case for the groups fed 712 and 777 g/kg OM carbohydrate (table 20). Despite equal intakes of energy and protein, the group receiving 712 g/kg OM carbohydrate achieved greater litter weight gains accompanied by greater maternal weight losses. Results similar to these have been observed by Maynard and Rasmussen (1942), Nelson and Evans (1947a,b) and by Steingrimsdottir et al. (1980).

Maynard and Rasmussen (1942) compared two feeds differing in carbohydrate content, rationed so as to give equal energy and protein intakes. This difference in carbohydrate

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Table 20: The effect of altering the carbohydrate content of the feed on lactational performance at (approx.) equal energy and protein intakes (Loosli et al, 1944). All measures are g/17d, except energy (MJ/17d).

Feed carbohydrate content (g/kg OM)	608	712	777	874
Energy intake	7.08	7.37	7.33	7.14
Protein intake	89.3	94.0	93.6	83.0
Litter weight gain	150.1	161.2	152.1	121.9
Maternal weight gain	-17	-8	3	-6

content was achieved by substitution of corn meal for corn oil, with an adjustment in the protein content (casein) to maintain the protein:energy ratio. The feeds were pair fed at the level of the lowest ad libitum energy intake of the pair. This was always the intake of the dam on the higher carbohydrate feed. Because the energy intake differed between pairs of dams, the difference in carbohydrate intake cannot be expressed in absolute terms. It was equivalent to a difference in carbohydrate content of 120 g/kg OM within each pair (after correction of an error in the reported feed compositions, recalculated from the ingredient compositions (MAFF, 1986 and table 7)). The dams on the low carbohydrate feed actually received slightly less protein than those on the high carbohydrate feed. Despite this, the low carbohydrate feed resulted in an increase in litter growth and maternal weight loss (figures 21a and 21b). From the data it was possible to relate litter liveweight gain and maternal weight change to feed energy intake and feed type. This was done by regression using the following equation:

$$Y = k + aN + bX + cNX$$

Where

Y is litter *or* maternal weight change (g/17d)
X is energy intake (kJ/17d)
N codes for feed type; 0 = high carbohydrate
 1 = low carbohydrate

k is the regression constant
a, b, and c are the regression coefficients

Fig 21a: Litter growth vs energy intake in relation to carbohydrate content (Maynard and Rasmussen, 1942)
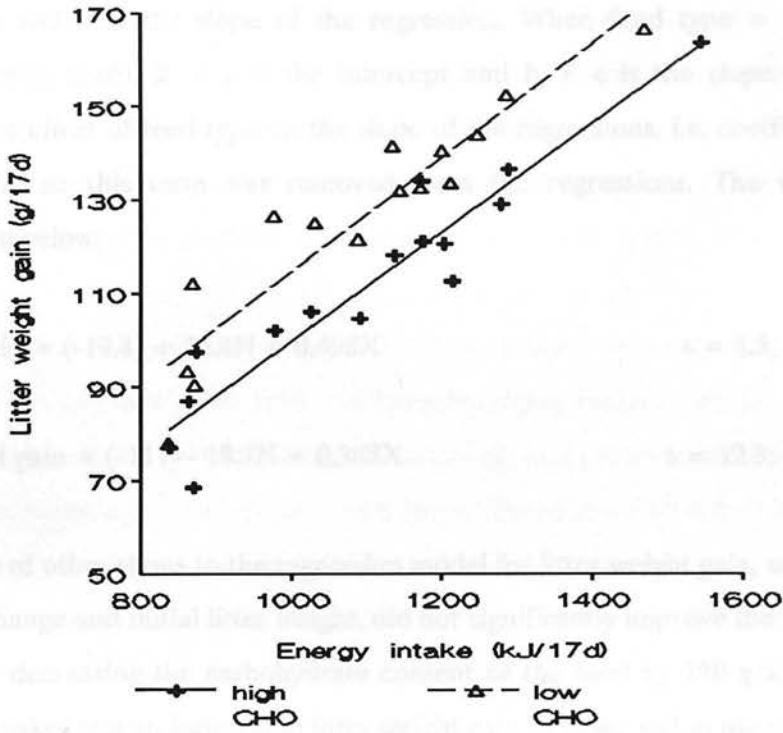
Fig 21b: Maternal weight change vs energy intake at different feed carbohydrate contents (Maynard and Rasmussen, 1942). One missing value.
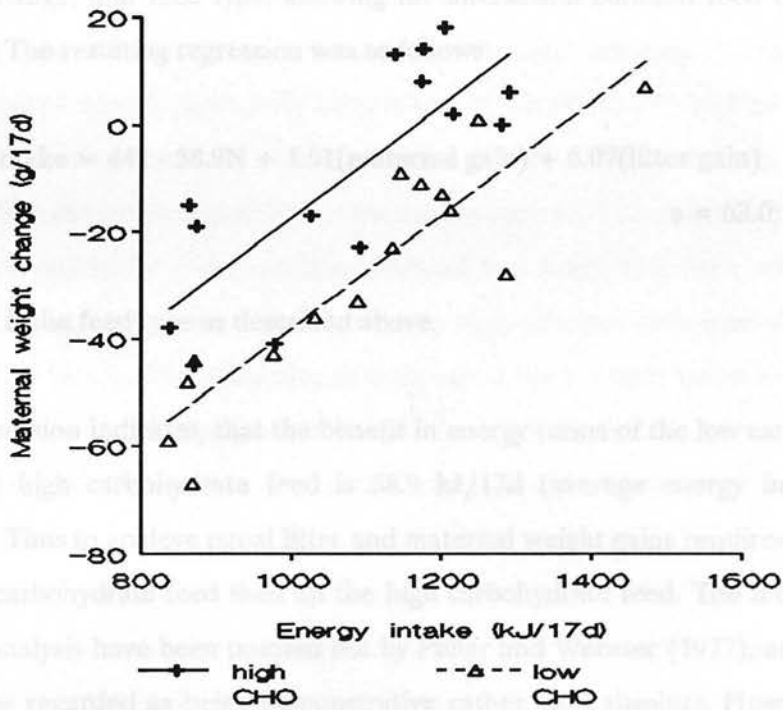
In this regression, when feed type = 0 (i.e. the high carbohydrate feed), **k** is the intercept and **b** is the slope of the regression. When feed type = 1 (i.e. the low carbohydrate feed), **k** + **a** is the intercept and **b** + **c** is the slope. There was no significant effect of feed type on the slope of the regressions, i.e. coefficient **c** was not significant, so this term was removed from the regressions. The regressions are presented below:

Litter gain = (-19.4) + 15.4N + 0.498X $\quad\quad$ s = 8.5; R^2adj = 88.4%

Maternal gain = (-111) - 18.7N + 0.388X $\quad\quad$ s = 12.3; R^2adj = 72.3%

Addition of other terms to the regression model for litter weight gain, such as maternal weight change and initial litter weight, did not significantly improve the regression. The effect of decreasing the carbohydrate content of the feed by 120 g/kg OM at equal energy intakes was an increase in litter weight gain of 15.4g and in maternal weight loss of 18.7g. By using energy intake as the dependent variable, estimates of the partition of feed energy can be made. Energy intake was regressed on litter weight gain, maternal weight change, and feed type, allowing for interaction between feed type and weight changes. The resulting regression was as follows:

Energy Intake = 441 - 58.9N + 1.91(maternal gain) + 6.07(litter gain)

$$s = 63.0; R^2adj = 88.8\%$$

where N is the feed type as described above.

This regression indicates, that the benefit in energy terms of the low carbohydrate feed over the high carbohydrate feed is 58.9 kJ/17d (average energy intake was 1098 kJ/17d). Thus to achieve equal litter and maternal weight gains requires 58.9 kJ less on the low carbohydrate feed than on the high carbohydrate feed. The limitations of this type of analysis have been pointed out by Pullar and Webster (1977), and these results should be regarded as being demonstrative rather than absolute. However, it is clear that substituting fat for carbohydrate improves the efficiency of food use in lactation.

Similar, but more rigourous comparisons than this one, have been made with non-lactating rats in an excellent series of experiments (Forbes et al, 1946a,b,c; French et al, 1948) summarised by Swift and Black (1949). They pair fed high and low carbohydrate feeds at equal protein and energy intakes. These experiments measured carcass composition, nitrogen and energy retention, and respiratory gases. Their results show that feed fat is used more efficiently than carbohydrate for growth.

In addition to an improvement in the efficiency of feed use, there was an increase in maternal weight loss when feed carbohydrate was replaced by fat (Maynard and Rasmussen, 1942; Loosli et al, 1944). Since energy and protein intakes on the different treatments were equal, this effect cannot be attributed to a deficit in food intake on the low carbohydrate/high fat feeds. As a result of this increased maternal body mobilisation, these litters gained more weight, thus the extra weight loss by these dams was beneficial. It would therefore be reasonable to assume, that the body mobilisation of the dams on the high carbohydrate/low fat feeds was constrained.

The intake of *growing* rats on low protein feeds has been shown to be constrained by the ability of these rats to dispose of heat (Meyer and Hargus, 1959; Andik et al, 1963). It was proposed in the previous section that the same constraint applied to lactating rats on low protein/high carbohydrate feeds. If this constraint applied to the dams in the experiment of Maynard and Rasmussen (1942), the difference in maternal body mobilisation can be explained. Dams on the low carbohydrate/high fat feed utilised the feed more efficiently and therefore produced less heat, than the dams on the other feed. This permitted them to mobilise more body reserves before reaching a maximal rate of heat production. Assuming that the extra body weight loss was all fat and was used to make milk fat then, using a value for the heat of milk fat production of 3.96 kJ/g (Chudy and Schiemann, 1969), the amount of extra body weight loss possible can be calculated.

Dams on the low carbohydrate feed achieved the same performance as dams on the high carbohydrate feed, but with 58.9 kJ less energy intake (regression 3) Therefore their heat production derived from feed energy was 58.9 kJ lower. This difference in

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heat production is equivalent to a body fat loss of 15g; the average measured difference in weight loss between the two feeds was 19.6g. In this context, the large maternal lipid losses occurring on low protein/high carbohydrate feeds described in the previous section, may be seen as a means by which to maintain milk fat production on a restricted food intake whilst incurring a minimal penalty in terms of heat production.

To explain differences in maternal weight loss it was assumed that heat production was maximal for the feeds in the above experiment (Maynard and Rasmussen, 1942). This assumption is derived from work with feeds of low protein content (Meyer and Hargus, 1959; Andik et al, 1963); the feeds in the above experiment were not of low protein content. However, the assumption was still considered to be valid on the basis of the findings described below.

The food intake of lactating rats is greatly elevated, being up to three times that of non-lactating rats (Slonaker, 1925). The measured heat production of lactating rats was found to be 713 kJ/kg$^{0.73}$/d at an environmental temperature of 28°C (Brody et al, 1938). In non-lactating rats, this rate of heat loss is only achieved at an environmental temperature of 42°C, close to the lethal temperature for rats (Kirmiz, 1962;). Thus the assumption, that lactating rats are limited by their heat production even when feed protein content is not low, seems tenable.

An attempt to substantiate this hypothesis is described in chapter 7. There is a paucity of reliable data on the efficiencies of milk production from different feed components and for this reason, this aspect of the hypothesis has not been discussed here.

The effects of substituting feed carbohydrate for feed fat on lactational performance may be summarised as follows:

i) There is an improvement in the efficiency of feed conversion to milk when fat replaces carbohydrate in the feed.

ii) At equal energy intakes, low carbohydrate/high fat feeds result in greater maternal weight loss than high carbohydrate/low fat feeds.

Further, the discussion has proposed that:

iii) The above effect can be explained by the assumption that the heat production of lactating rats is maximal over a wide range of feed compositions and hence constraining.

iv) Indirect evidence supports the assumption that the heat production of lactating rats is maximal, but no direct evidence exists to substantiate this.

Interactions Between Feed Components on Lactational Performance

Experiments which have substituted one feed component for another have been discussed above. It appears that an important effect of these substitutions is a change in maternal heat production. If this is the case, interactions between feed components would seem likely. There are however few experiments which have investigated interactions between feed components on lactational performance.

Nelson and Evans (1947a,b) compared three feeds; one high in protein, one high in carbohydrate, and one high in fat (figure 11). Dams with a standard littersize of six pups were offered these feeds ad libitum. They showed a depression in litter growth on the high fat feed (figure 11). There was no difference in litter growth between the high protein and high carbohydrate feeds. However, this experiment as described on page 10, was not adequate to investigate real as opposed to apparent interactions.

A comparison of three feeds differing in protein content from 148 to 446 g/kg OM was made by Sainz et al. (1986). Because the fat:carbohydrate ratio was constant, these feeds varied in their contents of fat and carbohydrate, as well as protein. There were no significant effects of feed composition on litter growth.

Another experiment from the same group of workers (Taylor et al, 1986) compared three feed protein contents (147, 311, and 565 g/kg OM) and two feed fat contents (26 and 197 g/kg OM) allowing a test for real interactions. Again, no significant effects of feed composition on lactational performance were found. This was surprising, given the results of other experiments (Naismith et al, 1982; Maynard and Rasmussen, 1942) which have found significant effects on lactational performance within the range of feed compositions tested by Taylor and co-workers. This experiment also studied the effect of feeding level on lactational performance, so that within each feed type, rats were allocated to different levels of feeding. The effects of feed composition on lactational performance were presented as adjusted marginal means pooled across levels of intake. This form of presentation prevents comparison of their results with other work, in absolute terms. In addition, it is not clear whether this analysis made any adjustment for interactions between feed composition and level of feeding effects. This is important, given that there were only two replicates per treatment cell. The original data for this experiment (Taylor, 1985) are not available in a usable form.

To my knowledge, there are no other experiments with lactating rats on interactions between feed components. There are a number of experiments with non-lactating rats (Schreiber and Elvehejm, 1955; Siedler et al, 1962; Krishmanachar and Canolty, 1986). The often cited experiment by Hartsook et al. (1973) has been omitted from this list, because the central composite design which they used, was inadequate to provide the "description of a large outcome surface", which they extrapolated from their data. However, given the differences between non-lactating and lactating animals in terms of productive outputs, food intake, and nutrient utilisation the relevance of these studies is questionable.

The lack of information about interactions between feed components on lactational performance, and the doubts about the existing experiments do not allow any valid conclusions to be drawn. It was therefore decided to investigate this subject. Four experiments and an attempt to model the effects of feed composition on lactational performance in rats, are presented in the following chapters.

The Rat Experiments - General Methodology.

I am grateful to Jack McGowan and his staff for their efforts in chemically analysing the carcasses and feeds

Note: All tables and figures are numbered according to the page on which they are found. As such table and figure numbers are not consecutive (see list of tables and figures, p. ix).

Introduction

The four experiments (R1, R2, R3, and R4) with lactating rats share a common methodology, which evolved with time, but was developed largely on the basis of experience gained in the first experiment (R1). The chronology of the experiments is R1, R4, R2 and R3. In this chapter, descriptive statements which are specific to an experiment are followed by the relevant experiment number in parentheses. Absence of experiment numbers indicates that the description applies to all the rat experiments.

Synopsis

Individually housed Sprague-Dawley rats nursing standardized litters were offered experimental feeds and water ad libitum from day 2 until day 14 (day 16, R1; day 12, R4) of lactation. During the experimental period intakes of food and water, maternal and litter liveweight, and room temperature were measured daily. Changes in carcass composition during the experimental period were measured by comparative slaughter. Carcasses and feeds were analysed for water, nitrogen, ether extracted fat, ash, and gross energy.

Animals

Sprague-Dawley rats were used in all the experiments. They were supplied by Harlan Olac UK Ltd as specified pathogen free (SPF) rats, that had previously produced two litters, except in the first experiment (R1) where virgins were used (suppliers B, S & S; Edinburgh). Similarly experienced males were used for mating in all the experiments except R1. The rats were allowed to adjust to their new environment for one week before mating. The number of females per male, conception rates and spread of births are given for the different experiments in table 29a. Fecundity of these rats was consistently underestimated, which created difficulties at cross-fostering. Work of this nature would be considerably easier if the experimental rats were drawn from a larger breeding population. This would allow variation in *natural* littersize and in spread of births to be reduced.

Table 29a: General reproductive data, pup mortality, and spread of births in the four experiments.

Experiments in chronological order:	R1	R4	R2	R3
Number of females	25	58	73	24
Number of males	5	13	30	24
Mating duration (days)	4	4[a]	5	7
Females/male	5	3	2 or 3	1
Conception rates	0.4	0.69	0.67	0.88
Total number of pups born	96	442	598	232
Stillborn (%)	5.2	1.4	2.8	0.4
Mortality (%)[b]	4.4	2.5	0.3	0
Spread of births[c] (days)	2.0	3.8	5.0	6.0

a: Each male spent 24 hours with each of three females in a cycle for up to 4 times per female.
b: Mortality does not include stillborn.
c: 3rd quartile minus 1st quartile.

Environmental Conditions

Females were individually housed in solid floor plastic cages (W=21.0 cm; L=34.5 cm; H=18.5 cm). Initially they had sawdust for bedding and shredded paper for nesting (R1). These were replaced by cat litter and shredded plastic in subsequent experiments (R4, R2, R3) to remove potential dietary fibre sources. Room temperature for the different experiments are detailed in table 29b.

Table 29b: Room temperature data (°C) in the four experiments.

	R1	R4	R2	R3
Mean temperature	19.7	22.8	24.6	23.9
S.D.	1.6	0.6	0.6	0.5
Top-bottom*	2.7	3.0	0.3	0.5
Extreme temp. measured:				
- max.	23.0	25.5	26.4	25.1
- min.	15.0	21.0	22.5	22.1

* The difference between the top cages and the bottom cages. In the last two experiments, a different ventilation system resulted in a more even room temperature.

Cross-Fostering and Handling

The day of birth was designated day 0 of lactation on which females were left alone. On day 1 of lactation pups were cross-fostered to give standard littersizes, these varied between experiments depending upon the average littersize of that experiment. In the first experiment (R1) it was necessary to delay some fostering until day 3 because of pup availability. Pups, which were fostered into a litter, were gently rubbed with the bedding of the recipient dam and were then placed in the middle of the natural litter whilst the dam was temporarily in another cage. The maximum number of pups successfully fostered into a litter was 5. Female rats with newborn litters are notoriously nervous and aggressive, and sudden unexpected disturbances may cause them to eat their litters. Several steps were taken to reduce pup mortality in the last three experiments, as a result of experience gained in R1. Dams were handled daily at a routine time throughout pregnancy. From the time of the first birth until the last litter was 5 days old, the only person who entered the room was myself. A radio-timer came on half an hour before I entered the room in the later experiments (R4, R2, R3). On opening any cage, the dam was always approached and allowed time for recognition before the litter was handled.

Daily Measures

The experimental period lasted from day 2 until day 14 (R2, R3, R4) or day 16 (R1) of lactation. During the experimental period all rats were offered the experimental feeds and water ad libitum, with the following measures being made daily:

> Feed intake
> Water intake (except R4)
> Maternal liveweight
> Litter liveweight
> Room temperature

These measures were made at the same time each day (starting at 08:45 am) and in the same sequence.

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Feed Intake and Composition

The rats were introduced to mashed feeds during pregnancy and no digestive problems of feeding were encountered. Fresh feed was weighed out daily, the weight of feed offered was designed to result in a 10% refusal. Refusals were dried in an oven at 60°C for 48 hours before weighing (R1, R4). In experiments R2 and R3 daily fresh refusals were weighed and stored under trichloroethane for subsequent chemical analysis. Unexpectedly this did not prevent microbial degradation of the feeds, resulting in the loss of these refusal samples. Beyond day 15 of lactation measurement of maternal food intake became less reliable because the pups opened their eyes and were able to eat the food as well (R1). For this reason the experimental period was ended on day 14 of lactation (R4, R2, R3). Feed samples were taken every third day throughout the experiment and dried. Samples were analysed for protein, ash, ether extract and gross energy using the same techniques as described for analysis of the carcass samples. The feed offered during pregnancy contained 25% protein and 40% (OM) fat. The compositions of the experimental feeds are given in the relevant chapters.

Feed Ingredients and Feed Manufacture

The components of all the feeds used throughout were casein, groundnut oil, maize starch, sucrose, mineral and vitamin mixes. The casein was supplemented with DL-methionine, the starch and sucrose were always fed in the ratio 2:1. Vitamins and minerals were initially incorporated into the feeds to supply 20 g/kg DM and 50g ash/kg DM respectively (R1, R4); subsequently levels were increased to 40 g/kg DM and 100g ash/kg DM respectively (R2, R3). The composition of the individual feed components is given in tables 32a, 32b and 32c; the composition of the feed mixtures for each experiment is given in the relevant chapters. The consistency of the final feed was important for accurate measurement of food intake. It was necessary to create a feed which was too soft for the rats to be able to remove in lumps from the feed container, but firm enough so as to remain in the feed container if knocked over. Water was added to the feeds to achieve the correct consistency, necessitating storage of the feeds in a freezer. Feeds of a high oil content tended to sediment out and so in the later experiments (R2, R3) an emulsifier combination (Montane 80 plus Montanox 80,

Table 32a: Composition of the individual feed ingredients (g/kg DM; except DM (g/kg fresh) and GE (MJ/kg DM)).

	Casein (1% met)	Starch	Sugar	Groundnut oil[a]	Vit.
DM	943.3	911.6	990.0	999.9	893.0
CP	945.2[b]	4.9	0.9	1.2	112.8
EE	4.7	3.5	2.7	998.8[c]	57.0
CF	-	-	-	-	31.4
Ash	19.0	0.0	0.0	0.0	30.5
CHO[d]	31.1	991.6	996.4	0.0	768.3
GE	23.5	16.5	16.3	39.6	18.3

a: Groundnut oil supplemented with antioxidant (1.5 g/kg Rendox; Kemin Europa Ltd.).
b: Crude protein = N x 6.407; where 1/6.407 is the calculated N-content of [0.99 casein + 0.01 methionine]; casein composition from Kuzdzal-Savoie et al. (1980).
c: Calculated; assuming CHO content = 0.
d: CHO = 1000 - (CP + EE + CF + Ash).

Table 32b: Composition of the vitamin mix; the filler was maize meal.

Vitamin	mg/kg
Biotin	18.8
Pantothenate	500
B$_1$	600
B$_2$	1500
Niacin	9894
B$_6$	980
B$_{12}$	2.4
K$_3$	800
Folate	1980
Choline	50000
	iu/g
A	1600
D$_3$	200
E	12

Inclusion at 2% DM in the feed meets the mineral requirements for lactating rats (NRC, 1978).

Table 32c: Composition of the mineral mix.

Mineral	g/kg DM
Calcium	126.1
Phosphorous	101.6
Sodium	76.5
Potassium	70.9
Magnesium	50.4
Sulphur	7.5
Chloride	118.0
Iron	1.5
Manganese	1.3
	mg/kg DM
Zinc	300.6
Selenium	2.5
Copper	101.6
Cobalt	20.0
Iodine	71.6

Inclusion at 2% DM in the feed meets the mineral requirements for lactating rats (NRC, 1978).

50:50, 5% inclusion, Honeywell and Stein, Leatherhead) was used with all feeds. This combination was robust enough to create a homogeneous mousse from feeds containing up to 55% oil, which survived being frozen and thawed. Unfortunately it resulted in problems with the chemical analysis of the feeds. The fat determinations by ether extract for those feeds which contained emulsifiers were clearly in error. Alternative fat determinations (acidified ether extract and chloroform-methanol extraction) were carried out on these samples but proved unsatisfactory, and feed fat content was therefore calculated from the gross energy of the feed. This assumed that there were no errors in the measured protein and ash contents of the feed. The feeds for each experiment were made up in one batch (R2, R3, R4) using a commercial dough mixer. Since the feeds contained no fibre, a plastic "chew" ring was given to each rat to allow natural trimming of teeth.

Feed Containers

Feed were weighed into pre-weigh jars (120 ml Beatsen wide necked). To prevent these being tipped over, a holder was developed which consisted of a cross section of plastic pipe glued to a plastic base, into which the jars just fitted (figure 34).

Measurement of Milk Production

No attempt was made to directly measure milk production or milk composition. Milk production was assessed indirectly from litter liveweight gain and gain in litter carcass constituents. Litters were weighed daily from day 2 to day 14 of lactation. The decision to index, rather than measure, milk production was a conscious one, based on the large variation in reported values and on the difficulties of the technique involved. Values range from 1.6g (Cox and Mueller, 1937) up to 51g (Brody and Nisbet, 1938) of milk produced daily from rats suckling an average of 8 pups in days 15-21 of lactation. The variety of techniques used have been discussed by Linzell (1972). They are all either accompanied by serious reservations concerning their validity and effect on litter growth, or demand special equipment and techniques. In the experiment by Brody and

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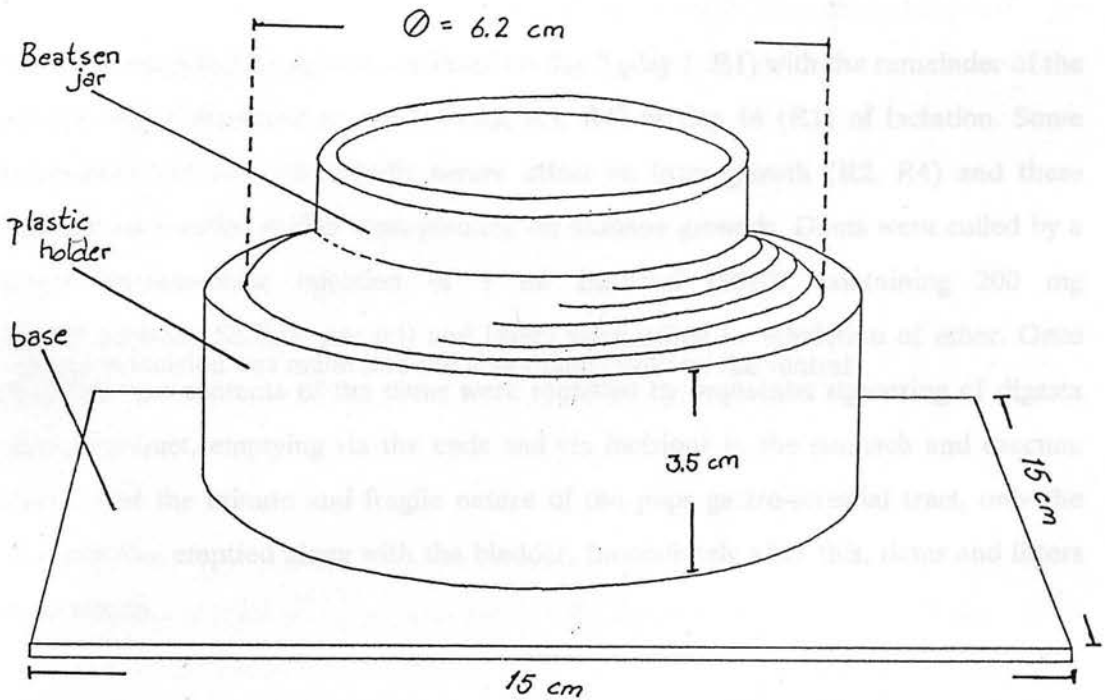
Figure 34. Diagram of feed container and jar.

Nisbet (1938), milk yield was measured by pup weight increase on suckling and also calculated from comparative slaughter data with good agreement between the two. This approach of using carcass data and assumed efficiencies has been used by others (Canas, 1974) and appears to give as good results as direct attempts at measurement. For these reasons it was decided not to measure milk production directly.

The only non-ruminant mammal, in which milk production can be measured accurately without concern for the physiological relevance of so doing, is the rabbit. The nursing behaviour of the doe is such that she only visits the litter once every 24 hours. The rabbit kittens receive their total daily milk intake in one suckling bout lasting only a few minutes (Hudson and Distell, 1986). The rabbit is therefore ideally suited to

measurement of milk production by pup weight increase on suckling.

Carcass Analysis

An initial slaughter group was sacrificed on day 2 (day 1; R1) with the remainder of the animals being sacrificed on day 14 (R2, R3, R4) or day 16 (R1) of lactation. Some treatments had an unexpectedly severe effect on litter growth (R2, R4) and these animals were culled earlier than planned on humane grounds. Dams were culled by a single intramuscular injection of 1 ml Euthatal (RMB; containing 200 mg Pentobaribitone Sodium per ml) and litters were culled by inhalation of ether. Once dead, an incision was made into the abdominal cavity on the ventral side. The gut contents of the dams were removed by sequential squeezing of digesta along the tract, emptying via the ends and via incisions in the stomach and caecum. Because of the minute and fragile nature of the pups gastro-intestial tract, only the stomach was emptied along with the bladder. Immediately after this, dams and litters were frozen.

In the subsequent analysis dams and litters carcasses were treated in the same fashion. The frozen whole carcasses were chopped up, minced and freeze dried (R2, R3) or freeze dried without mincing (R1, R4). In the latter case the whole carcass was dried, whereas a small portion of the fresh mince was retained from the fresh mince samples (R2, R3). After 48 hours in the freeze dryer (FTS Systems Inc.), all sample types had reached constant weight. They were then milled in a Retsch ultra-centrifugal mill designed to prevent overheating of the samples and consequent loss of fat. The milling of dried chopped unminced carcass created heat due to difficulty of milling skin. This meant, that a considerable amount of time had to be spent cooling the mill between samples to avoid overheating. For this reason, the procedure was modified to include the mincing step. The dried ground samples was then split and analysed in duplicate for:

Ash	(500°C for a minimum of 5 hours)
Nitrogen	(Automated Kjeldahl method; using Technicon block digestor and Tecator 1030 analyzer)
Ether extract	(40°-60°C pet. ether extraction for 5 hours; residue dried at 100°C)
Gross energy	(Gallenkamp adiabatic bomb calorimeter)

Discrepancies between replicates of more than 1% of the absolute % in DM (0.5% for ether extract and gross energy) resulted in further replicates being analyzed. The results arising from carcass analysis were further checked by the following calculations:

Sum = 6.25N + EE + Ash

Calc. GE = 23.8(6.25N) + 39.6EE

Protein:fat free DM = 6.25N/(100 - EE)

Protein/ash = 6.25N/ash

Any samples which showed a significant deviation in any one of these measures was re-analysed. It was found for all samples (R2, R3, R4) that the average sum was low (92%) but that there was good agreement between the calculated and measured GE. The discrepancy in the sum was due to incomplete drying of samples in the freeze dryer. This was checked by further drying samples at 60°C and 100°C. Freeze dried samples were shown to have contained some moisture, which was not fully expelled by drying at 60°C. However, at this temperature, sample weight does not stabilise but decreases by a small constant amount each day. By extrapolating back to time zero a corrected dry weight at 100°C can be calculated. Given the assumptions involved in this extrapolation, carcass composition data were not corrected to the 100°C dry weight since the error due to incomplete drying cancels out in the calculation of carcass gains (g/12d).

To consolidate the relationship between liveweight and initial carcass composition, data from three of the experiments (R2, R3, R4) were combined (appendix 3). This was justified, since there were no significant differences between experiments for this relationship. The regressions relating initial liveweight and littersize to carcass composition were of the form

Initial component weight (g DM) = a(inLlwt) + b(inMlwt) + c(natlit)

where   inLlwt is the initial litter liveweight.
              inMlwt is the initial maternal liveweight.
              natlit is the natural littersize.
              a, b, and c are the coefficients, which are given in table 37.

Table 37: Regressions relating initial liveweight and littersize to initial carcass component weights (g DM) or energy (kJ).

	a	b	c	s*
Maternal carcass composition:				
Crude protein	+0.0725	+0.160	0	2.548
Ether extract	-0.323	+0.262	0	7.378
Ash	+0.0159	+0.0321	0	1.010
Gross energy	-10.6	+13.8	0	271.9
Litter carcass composition:				
Crude protein	+0.0965	0	+0.0796	0.4884
Ether extract	+0.132	0	-0.756	0.5479
Ash	+0.0176	0	+0.0109	0.1139
Gross energy	+7.6	0	-29.2	24.80

*: s = residual standard deviation

In the first experiment (R1) the average values of carcass composition were applied to all animals. There was no justification from the data on the initial culls in this experiment to use a more sophisticated approach.

Statistical Analysis

Initial data processing and simple descriptive statistics were carried out using the Minitab software. Subsequent analysis of variance was carried out using Genstat software. A standard analysis of variance structure was used in conjunction with Levenes test for homogeneity of variance (Snedecor and Cochrane, 1980). In experiments R2 and R4 more complex structures were necessary to accommodate missing values and the structure of the design respectively. All Genstat analysis of variance programs are given in appendix 1. All statistical analyses were validated by a statistician.

CHAPTER 4

The First Rat Experiment (R1)

The Effect of Substituting Carbohydrate for Fat in the Feed of Lactating Rats

Note: All tables and figures are numbered according to the page on which they are found. As such table and figure numbers are not consecutive (see list of tables and figures, p. ix).

<u>Introduction</u>

It has been shown that lactating rats offered a low protein feed have a greatly impaired performance when compared to rats offered a high protein feed (Naismith et al, 1982). They are unable to compensate for the decreased protein content of the feed by increasing their intake, which appears to be constrained. Work by Maynard and Rasmussen (1942) showed, that at constant protein and energy intake lactating rats receiving feeds of a lower carbohydrate content grew larger litters and would, had they been fed ad libitum, have had a higher energy and protein intake than those rats on the high carbohydrate feed. It thus seems likely, that the effects of different feed protein contents might be modified by the carbohydrate content of the feed. It was decided to investigate this using extremes of feed composition.

The two objectives of this, the first, rat experiment were:

- To gain experience of working with lactating rats and to develop a methodology for subsequent experiments.

- To compare feeds differing in fat and carbohydrate content at two different protein levels.

Because of the lack of expertise and of a proven methodology it was decided to work with a small number of female rats (n=25). This limited the extent to which the second objective could be fulfilled.

<u>Method</u>

The original plan of the experiment was to use four feeds (figure 40). The design had to be reduced due to difficulties in achieving a satisfactory number of pregnancies in an acceptable time span. Groups of five dams were caged with one male for a week and inspected daily for vaginal plugs. Relatively few plugs were seen, but because the cages had solid floors it was assumed that the plugs had been eaten and that by the end of

the week all females would have been inseminated. Abdominal palpation on day 14 of "pregnancy" revealed, that there were insufficient lactating dams to allow four treatments. Thus only two feeds were offered; their compositions are presented in table 41a. These feeds were offered to both lactating rats and to those rats which did not conceive, the non-pregnant non-lactating (NPNL) rats. The numbers of rats per treatment are given in table 41b. Between days 1 and 3 of lactation pups were cross-fostered to achieve a standard littersize of 10 pups. On day 1 of lactation a group of rats were culled to provide initial body composition estimates. The experimental feeds were offered ad libitum from day 2 until day 16 of lactation, when the remaining animals were culled for carcass analysis. Food intake, water intake, maternal liveweight and litter liveweight were measured daily during this period. Full details of the methods are given in the preceding chapter. The full data are presented in appendix 2.

Fig. 40: The original design
for the first rat experiment.
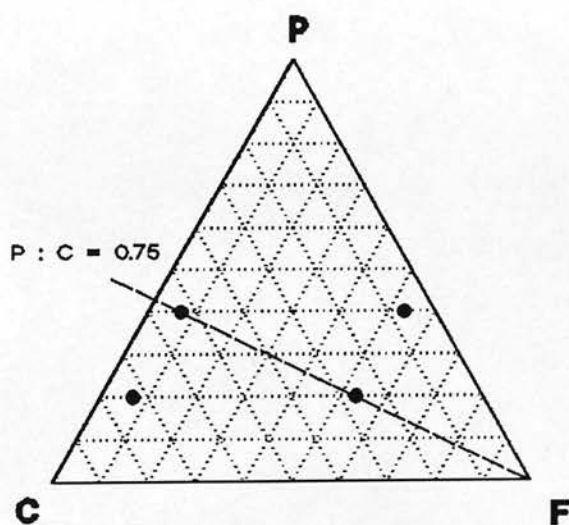
Table 41a: Composition of the feeds used in the first rat experiment (g/kg OM unless otherwise stated).

	Low carbohydrate (L)	High carbohydrate (H)
DM (g/kg fresh)	965.5	399.4
CP	400.0	400.0
EE	533.3	6.7
CHO	6.7	533.3
GE (MJ/kg OM)	31.87	21.02

DM is oven dry matter at 60°C; CP is crude protein = N x 6.407 (see p. 32); EE is ether extract; CHO is carbohydrate calculated as (1000-CP-EE); GE is gross energy. Mineral content is 100 g/kg DM.

Table 41b: Number of rats per treatment

Treatment	L	H	Initial cull
NPNL	5	6	3
Lactating	4	3	3

Results

General Health: It became apparent during the experiment that these rats were suffering from a respiratory infection from which one rat died. The infection was probably responsible in part for the low conception rate achieved; improvements in the mating scheme and in other aspects of the methodology have been reported in the preceding chapter (3), including a change in the supplier of rats. These changes, in subsequent experiments, have resulted in decreased pup mortality, improved conception rates and increased precision of measurement.

Lactating Rats: Despite reservations due to the health of these rats, the results (summarized in table 42) are in general agreement with subsequent experimental results. It can be seen from table 42, that litter growth was not significantly affected by feed composition except for an increase in lipid gain (p<0.01) on the low

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carbohydrate/high fat feed. There were no significant effects of changing carbohydrate/fat content on maternal liveweight or body reserves. Protein intake was significantly decreased (p < 0.01) on the low carbohydrate/high fat feed when compared to the high carbohydrate/low fat feed. This resulted in a non-significant depression in litter protein content and almost equal maternal protein gains. The significant increase in litter lipid gain was the result of a non-significant increase in energy intake on the low carbohydrate/high fat feed. It should be noted, that the variation within groups was relatively high, presumably due in part to the health problems of these animals. The variation was not equal between feeds, such that the low carbohydrate group had higher standard deviations for most variables. Homogeneity of variance was tested for, using Levenes' test of homogeneity (Snedecor and Cochran, 1980). Those measures, which did not show homogeneity of variance, were transformed to log values for analysis of variance, which did not alter the significance of the results. The standard errors of the difference shown in the tables are from the analysis of untransformed data.

Table 42: The effect of altering the carbohydrate/fat content of the feed on lactational performance of dams fed ad libitum (All values in g/14d, except the energy values in MJ/14d).

| | Carbohydrate content | | sed | p< | coefficient of variance |
	Low (n=4)	High (n=3)			
Intake of					
DM	390	546	30	0.01	0.09
Protein	149.8	209.6	11.4	0.01	0.09
Carbohydrate	25.0	279.5	4.6		0.05
Fat	199.7	35.0	14.7		0.15
Gross energy	11.9	11.0	0.9	n.s.	0.10
Water	961	1366	51	0.001	0.06
Maternal gain of					
Liveweight	8.5	1.4	8.5	n.s.	1.99
Protein	7.6	7.7	2.7	n.s.	0.45
Lipid	-20.1	-27.6	5.4	n.s.	0.30
Litter gain of					
Liveweight	285	247	25	n.s.	0.11
Protein	44.8	45.7	4.2	n.s.	0.11
Lipid	47.4	17.1	6.4	0.01	0.21

Non-Pregnant Non-Lactating Rats (NPNL): Average intakes and changes in body stores for the NPNL rats are given in table 43. The effects of altering the carbohydrate/fat content of the feed were quantitatively the same as the effects on intake and body changes in the lactating rats. Because there was not the additional demand of lactation, the non-lactating rats did not lose body lipid. However, in both states the rats on the low carbohydrate/high fat feed had a tendency to have more body lipid, than the rats on the high carbohydrate/low fat feed.

Discussion

Altering the carbohydrate/fat content of the feed of lactating rats from 533 to 67 g/kg OM had no effect on litter liveweight gain. The extremely low content of carbohydrate (67 g/kg OM) in the low carbohydrate feed did not appear to compromise the milk production of those dams. This is in agreement with other published work (Steingrimsdottir et al, 1980), although the current low carbohydrate content was chosen to be lower than that in the work of Steingrimsdottir and co-workers (at a similar fat content). Follis and Straight (1943) reported on a dam who was able to raise

Table 43: The effect of altering the carbohydrate/fat content of the feed on 3 months old female rats (All values in g/15d except energy values in MJ/15d).

| | Carbohydrate content | | | | |
	Low (n=5)	High (n=6)	sed	p<	coefficient of variance
Intake of					
DM	154.7	210.6	15.8	0.01	0.13
Protein	59.4	80.9	5.8	0.01	0.13
Carbohydrate	9.9	107.8	6.2		0.16
Fat	79.2	13.5	4.7		0.18
Gross energy	4.7	4.3	0.4	n.s.	0.14
Water	353	517	53	0.01	0.20
Body gains of					
Liveweight	25.5	19.6	3.8	n.s.	0.28
Protein	9.2	8.3	1.7	n.s	0.32
Lipid	3.9	3.7	6.5	n.s.	2.77

a litter on a feed which consisted only of protein and fat, but they did not quantify the litter growth. On extremely low carbohydrate feeds, provided that there is an ample protein supply, it would appear that lactating rats are not only able to maintain their litters but to grow them at a rate equal to that of dams on high carbohydrate feeds.

Litter gains of protein were not significantly affected by feed. This indicates, that there was an adequate supply of milk protein on both feeds, despite a significantly lower maternal protein intake on the low carbohydrate/high fat feed. Dams on both feeds gained similar amounts of body protein, indicating that there was an adequate supply of feed protein, surplus to protein requirements for milk.

The only significant effect on milk production was that litters on the low carbohydrate /high fat feed gained more lipid than litters on the high carbohydrate/low fat feed. This effect was due to the non-significant increase in the energy intake of the low carbohydrate/high fat feed. The greater energy intake on the low carbohydrate/high fat feed was not the consequence of an intake driven by a protein demand, since the dams had a protein intake surplus to their requirements for milk production. Further, the dams lost body lipid, suggesting that they were in energy deficit. It would appear the low carbohydrate/high fat feed, despite being of extremely low carbohydrate content, improved lactational performance as compared to the high carbohydrate/low fat feed.

As described in the results, these rats were suffering from a respiratory infection, and the variation associated with these results was high (tables 42 and 43). This was probably due to the impaired health and consequent low animal numbers per treatment. To attempt to develop the discussion any further on the basis of these results would be unproductive. Having achieved a measure of competence in working with rats, and a greatly improved methodology, these results indicated that the effect of altering the carbohydrate/fat content of the feed was worthy of further exploration.

CHAPTER 5

The Second Rat Experiment (R2)

The Effect of Substituting Feed Carbohydrate for Fat at Two Different Levels of Protein Inclusion on the Lactational Performance of Rats.

Note: All tables and figures are numbered according to the page on which they are found. As such table and figure numbers are not consecutive (see list of tables and figures, p. ix).

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Introduction

The first rat experiment (chapter 4) was a preliminary comparison of the influence on lactational performance of two feeds which differed in their carbohydrate/fat content. Replacement of carbohydrate by fat in high protein feeds (400 g/kg OM in R1) did not depress lactational performance. This has been found by others but not at such a low level of carbohydrate inclusion (Maynard and Rasmussen, 1942; Nelson and Evans, 1948; Canas, 1974). It is likely, that those rats which were offered diets of low carbohydrate/high fat and relatively high protein content, were augmenting their glucose supply by conversion of surplus dietary protein to glucose via gluconeogenesis in order to maintain their milk production. If a feed of equally low carbohydrate content, but of limiting protein content, were offered to lactating rats, gluconeogenesis would result in a protein deficit. In this situation, the lactating dam could not maintain production of both milk lactose and milk protein. The effect of substituting feed carbohydrate for feed fat on lactational performance may, for this reason, be influenced by the protein content of that feed. As discussed in chapter 2 (p. 25), there are no adequate tests reported in the literature of this potential interaction. In order to explore this potential interaction, the present experiment was designed with the following objectives:

- To measure the effect of substituting feed carbohydrate for feed fat over a wide range, at fixed feed protein content.

- To measure the effect of varying the carbohydrate/fat content of the feed at two different feed protein contents; one expected to be in excess of protein requirements, the other expected to be below the protein requirement for adequate lactation.

- To satisfy the above objectives with feeds which permit the interactions between protein, fat and carbohydrate to be quantified.

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The resultant design is shown in figure 48a. Given that these objectives were met, the following expectations were held:

1) That at the super-adequate protein content, feeds of decreasing carbohydrate content would support adequate lactation and allow litters to have increased body lipid gains, as in the first experiment (R1). This assumes, that the energy content of these feeds does not prevent minimum desired protein intake from being achieved. By design, the protein:energy ratios of the feeds in this experiment were less favourable than in the previous experiment (R1; figure 48b).

2) That dams on the low protein feeds would have lower protein and energy intakes than those on the high protein feeds (Naismith et al, 1982).

3) That the lactational performance of dams on the low protein feeds would be depressed in relation to the high protein feeds, resulting in smaller growth rates in the litters and in increased maternal protein mobilization.

4) That on the low protein feeds, substitution of feed fat for carbohydrate would affect lactational performance, but the effect would depend upon the total glucogenic supply (carbohydrate + protein catabolism) in relation to glucogenic requirements for milk production.

5) In those low protein feeds where total glucogenic supply *was* limiting (figure 48b), decreasing carbohydrate content would result in a depression in lactose availability and thus milk production and litter growth.

6) In those low protein feeds where total glucogenic supply was *not* limiting (figure 48b), decreasing carbohydrate content would not depress litter growth. The decrease in carbohydrate content might even result in increased litter lipid gain.

Fig. 48a: Feeding treatments in the second rat experiment.
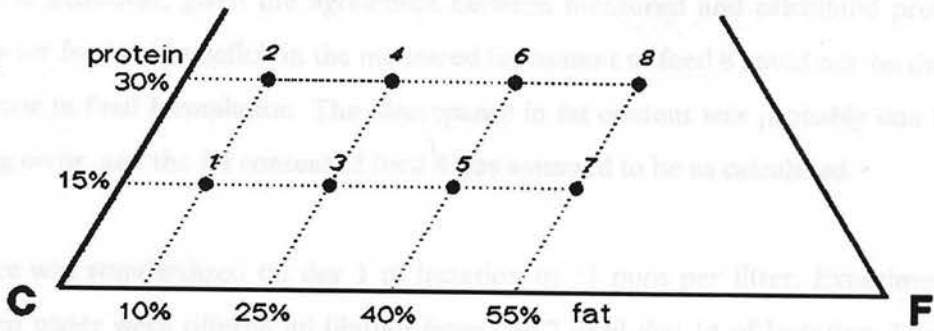
Fig. 48b. Feeding treatments for the first (▼) and second (●) rat experiments; the dotted lines are of constant protein : energy ratio.

Table 49: The composition of the feeds used in the second rat experiment (g/kg OM, unless otherwise stated).

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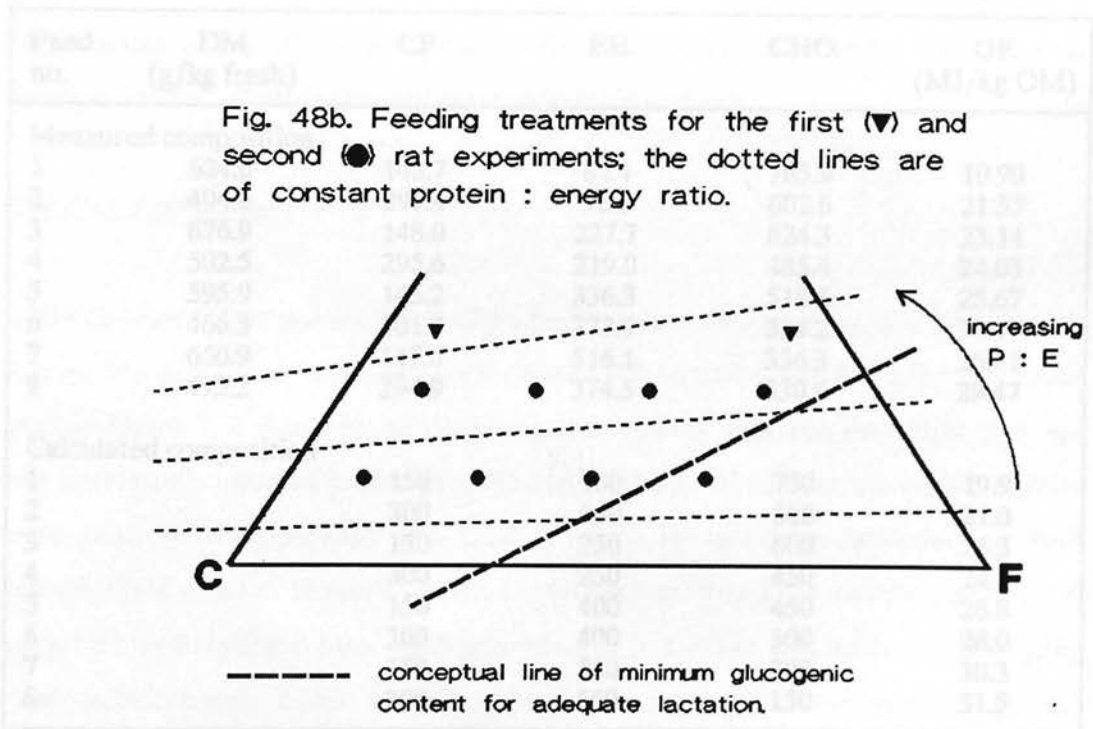
Method

A total of 47 lactating dams were allocated to one of 9 treatments; 8 feeding treatments and an initial cull group. The composition of the feeds are given in table 49. The measured feed composition was in agreement with the calculated composition, except for feed 8. However, given the agreement between measured and calculated protein contents for feed 8, the deficit in the measured fat content of feed 8 could not be the result of an error in feed formulation. The discrepancy in fat content was probably due to a sampling error, and the fat content of feed 8 was assumed to be as calculated.

Littersize was standardized on day 1 of lactation to 13 pups per litter. Experimental feeds and water were offered ad libitum from day 2 until day 14 of lactation. During

Table 49: The composition of the feeds used in the second rat experiment (g/kg OM, unless otherwise stated).

Feed no.	DM (g/kg fresh)	CP	EE	CHO	GE (MJ/kg OM)
Measured composition*					
1	624.6	148.7	85.4	765.9	19.90
2	494.0	299.4	98.0	602.6	21.57
3	676.9	148.0	227.7	624.3	23.14
4	502.5	295.6	219.0	485.4	24.03
5	595.9	145.2	336.3	518.5	25.67
6	466.5	301.9	373.9	324.2	27.57
7	660.9	147.6	516.1	336.3	26.61
8	475.2	294.9	374.5	330.6	29.47
Calculated composition					
1		150	100	750	19.9
2		300	100	600	21.0
3		150	250	600	23.3
4		300	250	450	24.5
5		150	400	450	26.8
6		300	400	300	28.0
7		150	550	300	30.3
8		300	550	150	31.5

* DM is oven dry matter at 60°C; CP is crude protein = N x 6.407 (see p. 32); EE is ether extract calculated from gross energy; CHO is carbohydrate calculated as (1000-CP-EE); GE is gross energy. Mineral content is 100 g/kg DM.

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this period, the following daily measures were made:

> Food intake
> Water intake
> Maternal liveweight
> Litter liveweight
> Room temperature

The initial cull was made on day 2 with the remaining rats being culled on day 14 for carcass analysis. Carcasses were analysed for water, nitrogen, ether extract, ash, and gross energy. Further details of the method are described in chapter 3. The full data are presented in appendix 3.

Results

The results are presented in three sections; the first two sections describe the effects of substituting carbohydrate for fat in the feeds of high protein content, and in the feeds of low protein content. These two sections have been sub-divided into effects on litter growth, maternal body reserves, and food intake. The third section makes comparisons between the two protein contents. Statistical significances quoted in the first two sections refer to comparisons between feeds of constant protein content. Statistical significances quoted in the third section refer to comparisons made between all feeds. F-values for the different comparisons are given in table 55.

The High Protein Feeds

Litter Growth: Litter gains of liveweight, body protein, and body fat, are presented in figures 51a and 51b. When dams were offered a feed containing 300 g/kg OM crude protein (feeds 2, 4, 6 and 8), all the litters grew quickly and litter liveweight gain was not significantly affected by changing the carbohydrate/fat content. As expected, litter lipid gains were significantly enhanced ($p < 0.05$) by successive reductions in feed carbohydrate content. However, at the lowest feed carbohydrate content (feed 8; 150 g/kg OM carbohydrate) litter lipid gain was less (n.s.) than that on feed 6 (300 g/kg OM carbohydrate). Litter protein gains were not significantly affected by the carbohydrate/fat content of the high protein feeds, but there was a trend for litter protein gain to decline with decreasing carbohydrate content.

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Fig. 51a: Maternal and litter liveweight gains (g/12days);
(sed = 12.6 and 26.5 g/12 days for maternal and litter gains).
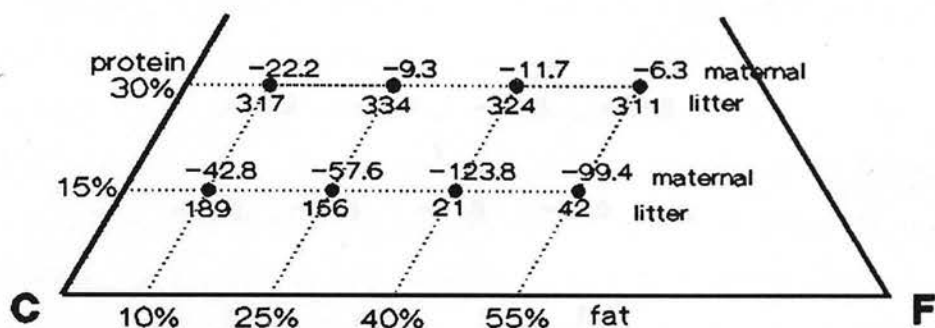
Fig. 51b: Litter gains of body protein and fat (g/12 days);
(sed = 3.5 and 5.7 g/12 days for body protein and fat).
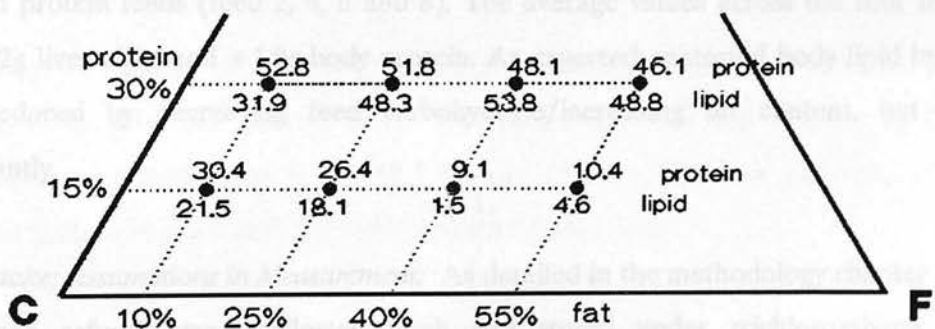
Fig. 52: Maternal gains of body protein and fat (g/12 days):
(sed = 2.4 and 5.9 g/12 days for body protein and lipid).
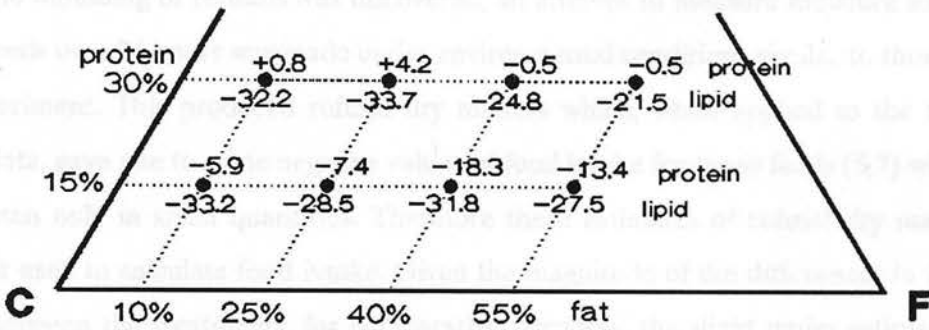
Maternal Body Reserves: On all feeds dams lost weight, protein, lipid and energy between day 2 and 14 of lactation. Changes in maternal liveweight and body protein (figures 51a and 52) were not significantly affected by the carbohydrate/fat content of the high protein feeds (feed 2, 4, 6 and 8). The average values across the four feeds were -12g liveweight and +1.0g body protein. As expected, maternal body lipid losses were reduced by decreasing feed carbohydrate/increasing fat content, but not significantly.

Food Intake; Assumptions in Measurement: As detailed in the methodology chapter (3), daily feed refusals were collected fresh and stored under trichloroethane for subsequent chemical analysis. Unfortunately, the solvent did not, as expected, prevent the refusals fermenting and growing mould. Composition data for the refusals were therefore lost. This had two effects:

- That refusal compositions has been assumed to the same as that of the feed offered. This is highly likely since the feeds were emulsified and no visible separation or sedimentation occurred.

- That refusal dry matter has been assumed to the same as that of the feed offered. When the moulding of refusals was discovered, an attempt to measure moisture losses of the feeds over 24 hours was made under environmental conditions similar to those in the experiment. This produced refusal dry matters which, when applied to the feed intake data, gave rise to some negative values of food intake for those feeds (5,7) which were eaten only in small quantities. Therefore these estimates of refusal dry matter were not used to calculate food intake. Given the magnitude of the differences in feed intake between the treatments, for comparative purposes, the slight under-estimation of feed intake resulting from this assumption is not important.

Food Intake; Treatment Effects: As the feed carbohydrate content of the high protein feeds decreased, food intake (figure 54a) and consequently protein intake also decreased ($p < 0.01$). Despite this decline in food intake, there was a non-significant increase in energy intake (figure 54b) as fat replaced carbohydrate, because of the increasing energy content of the feeds. At the lowest feed carbohydrate content (feed 8; 150 g/kg OM) this increased energy intake was curbed to a level similar to feed 4 (600 g/kg OM carbohydrate).

The energy intake data complement the results on litter lipid gain and maternal lipid loss. As the carbohydrate content of the feed decreased from 600 to 300 g/kg OM, energy intake increased from 9.3 to 10.2 MJ per 12 days. This was accompanied by increased litter lipid gains and decreased maternal lipid losses.

The Low Protein Feeds

Litter Growth: In general, performance on the low protein feeds for both dams and litters were much inferior to that on high protein feeds (see p. 51). Decreasing the

Fig. 54a: Maternal dry matter intake (g/12 days; sed = 39).
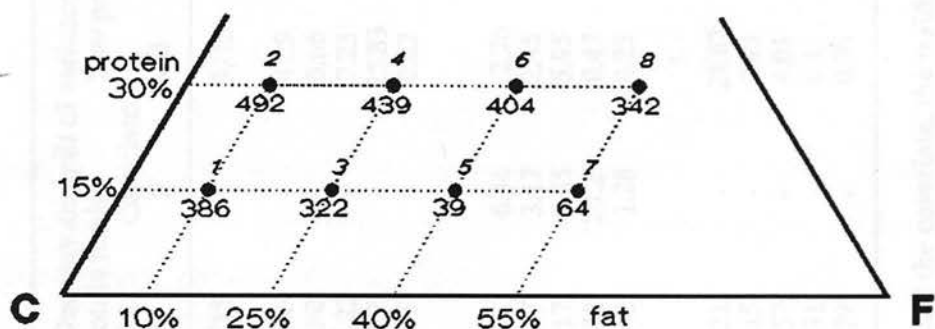
Fig. 54b: Maternal gross energy intake (MJ/12 days; sed = 0.8)
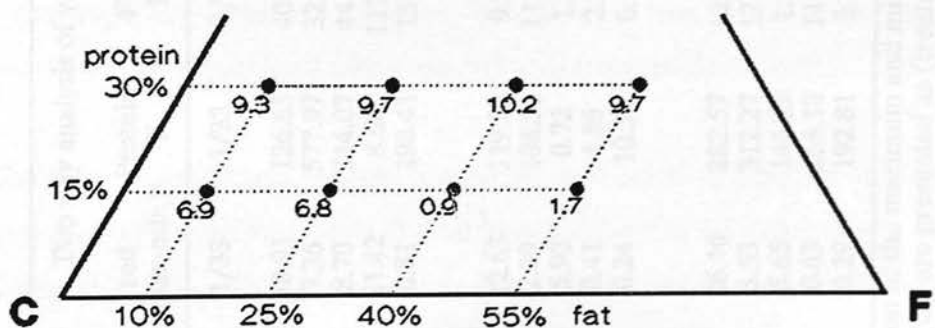
Table 55: The standard error of the difference between means (sed) and the variance ratios (F) from the two-way analysis of variance comparing all feeds, and from the one-way analyses of variance for feeds of the same protein content. Maternal gains have been analysed using maternal liveweight on day 2 of lactation as a covariate.

	Two way analysis of variance; all feeds					One way analysis of variance					
						High protein feeds			Low protein feeds		
	sed (max-min)[a]	Protein F	Fat F	Interaction F	Covariate F	sed	Fat F	Covariate F	sed	Fat F	Covariate F
Degrees of freedom[b]	1/33	3/33	3/33	1/33	1/32	3/16	1/15	1/15	3/16	1/16	1/16
Maternal intake of											
DM	38.91	126.65	40.73	11.68	-	31.6	8.80	-	41.9	36.73	-
Protein	7.36	577.97	32.48	3.16	-	8.54	8.80	-	5.66	36.73	-
Fat	8.70	314.07	44.75	65.32	-	9.65	75.22	-	7.23	22.48	-
Carbohydrate	21.42	8.68	112.03	4.27	-	11.87	142.71	-	25.83	45.84	-
Gross energy	0.81	198.41	15.07	20.14	-	0.74	0.55	-	0.82	31.21	-
Maternal gains of											
Liveweight	12.63	119.93	9.20	12.48	12.95	11.95	0.71	6.46	12.76	18.54	6.59
Protein	2.39	108.50	11.11	4.37	9.78	2.09	2.17	3.17	2.45	11.97	7.78
Fat	5.90	0.72	1.50	0.88	0.79	5.67	2.17	0.65	5.95	0.37	0.19
Ash	0.41	1.86	2.96	1.94	21.16	0.30	2.32	20.29	0.47	2.59	7.17
Gross energy	0.24	10.38	0.78	1.51	2.31	0.23	1.42	1.28	0.25	0.91	1.05
Litter gains of											
Liveweight	26.46	282.57	12.26	10.63	-	29.6	0.21	-	21.67	30.69	-
Protein	3.53	312.27	17.48	5.64	-	4.31	1.15	-	2.43	41.32	-
Fat	5.65	146.83	1.28	12.35	-	6.81	4.57	-	4.01	12.54	-
Ash	0.63	268.18	14.98	9.85	-	0.75	0.31	-	0.45	44.13	-
Gross energy	0.29	192.81	2.79	10.41	-	0.36	1.79	-	0.20	19.66	-

a: sed for the comparison of the maximum and minimum number of replicates.

b: degrees of freedom (df) are presented as (treatment df)/(residual df). For analyses which used the covariate, the residual df for all comparisons is as given for the covariate.

carbohydrate content of the low protein feeds (feeds 1, 3, 5 and 7) had dramatic effects on litter gains. The effects of successive declining steps in carbohydrate content of 150 g/kg OM can be seen from figures 51a and 51b. The effect of decreasing carbohydrate/increasing fat was not uniform across the range of feed carbohydrate contents offered.

At the high carbohydrate/low fat end of the range, defined as feeds 1 and 3, the average litter liveweight gain was 173 g/12 days. The drop of 150 g/kg OM in carbohydrate content between feeds 1 and 3 resulted in decreased litter liveweight, protein, and lipid gains of -33g, -4.0g, and -3.4g per 12 days respectively. This effect, though non-significant, was contrary to expectations.

The subsequent step down in feed carbohydrate content and hence step up in fat content from feed 3 to 5 had a catastrophic effect on lactation. The differences between feeds 3 and 5 values for litter liveweight, protein and lipid gains were -135g, -17.2g, and -16.6g per 12 days respectively ($p < 0.001$).

At the low carbohydrate end of the range, defined as feeds 5 and 7, the average litter liveweight gain was 31 g/12 days. The effect of the final step down in carbohydrate content from feed 5 to 7 was a small non-significant increase in litter performance; a decrease had been expected. The mean values of litter performance, maternal weight change, and food intake of the group on feed 7 were heavily influenced by one individual rat (rat 21, appendix 3) which was excluded from the data.

Dams and litters on feed 5 and 7 were culled earlier than day 14 of lactation on account of the unexpectedly severe depression in litter growth. The average cull dates for feeds 5 and 7 were days 11.9 and 10.5 of lactation respectively; by this time the litters had ceased gaining weight. For comparison with the other groups, it was assumed that the weight of these dams and litters on day 14 of lactation would have been the same as their weight when culled.

Maternal Body Reserves: Changes in the maternal body stores (figure 52) were in accordance with the effects seen on litter growth. All dams on the low protein feeds (feeds 1, 3, 5 and 7) lost liveweight, body lipid, and body protein. At the high carbohydrate end of the range (feeds 1,3), average maternal liveweight and body protein losses were 50 g/12 days and 6.6 g/12 days. At the low carbohydrate end of the range (feeds 5,7) average maternal liveweight and body protein losses were 112 g/12 days and 15.9 g/12 days, respectively. The difference between the high and low carbohydrate feeds for both liveweight and body protein losses was highly significant ($p < 0.001$). There was no significant effect of changing feed carbohydrate/fat content on maternal body lipid losses. The average body lipid loss was 30.3 g/12 days.

Food Intake: Food intake data for the low protein feeds (feeds 1, 3, 5 and 7) are presented in figures 54a and 54b. Dams and litters on feeds 5 and 7 were culled earlier than day 14 of lactation. To allow comparison between these dams and those on other feeds, 12 day cumulative intakes (up to day 14 of lactation) were calculated by extrapolation from existing daily intakes (appendix 3).

Feeds 1 and 3 were of high carbohydrate content, not limiting glucogenic supply. Contrary to the expectation, that food intake would increase as fat replaced carbohydrate in these feeds, there was a non-significant decrease in food intake and consequently protein intake.

There was a massive decline ($p < 0.001$) in food intake when the carbohydrate content of the feed decreased from 450 to 300 g/kg OM (and fat content increased). Average intakes for the high and low feed carbohydrate contents (feed 1, 3 and 5, 7) were 353.7 and 60.7 g/12 days, respectively. Despite the increasing energy density of the feed as feed carbohydrate was replaced by fat, energy intake fell ($p < 0.001$) as the carbohydrate content decreased (figure 54b).

There was no significant difference in food intake between feeds 5 and 7. This was contrary to expectation for feeds which severely limited lactational performance.

Comparison Between Protein Levels

The effect of a decrease in feed protein content from 300 g/kg OM to 150 g/kg OM was, as expected, a depression in energy and protein intakes which resulted in poorer litter growth, litter carcass gains, and greater mobilisation of maternal body protein ($p < 0.001$). Maternal body lipid mobilisation was not significantly affected by feed composition.

There was a massive and highly significant interaction between the effect of feed protein content and the effect of feed carbohydrate/fat content on lactational performance (figure 59) except for maternal lipid loss. The interaction was such that the difference between the two protein levels in lactational performance was amplified as carbohydrate was replaced by fat in the feed.

Discussion

For the high protein feeds (feeds 2, 4, 6, and 8) the results of this experiment substantiated the preliminary results from the first experiment (R1). At a high fixed level of feed protein content, the only significant effect on lactational performance of replacing feed carbohydrate by feed fat was to increase litter lipid gain. This resulted in part from a non-significant increase in energy intake as feed carbohydrate was replaced by fat, except on the lowest carbohydrate feed (8). Since the energy density and energy intake of the feeds increased as feed fat replaced feed carbohydrate, food intake and consequently protein intake declined significantly. This did not significantly affect litter or maternal protein gains, though there was a trend for litter protein gains to decline as the feed carbohydrate content decreased, and protein intake fell. The dams on these feeds did not lose body protein.

These results suggest that even on the low carbohydrate/high fat feed protein intake was not being limited. However, litter lipid gains were sensitive to differences in feed carbohydrate/fat content and their resultant effect on energy intake. This indicates that

Fig. 59: The interaction between feed protein and carbohydrate/fat content on litter liveweight gain.
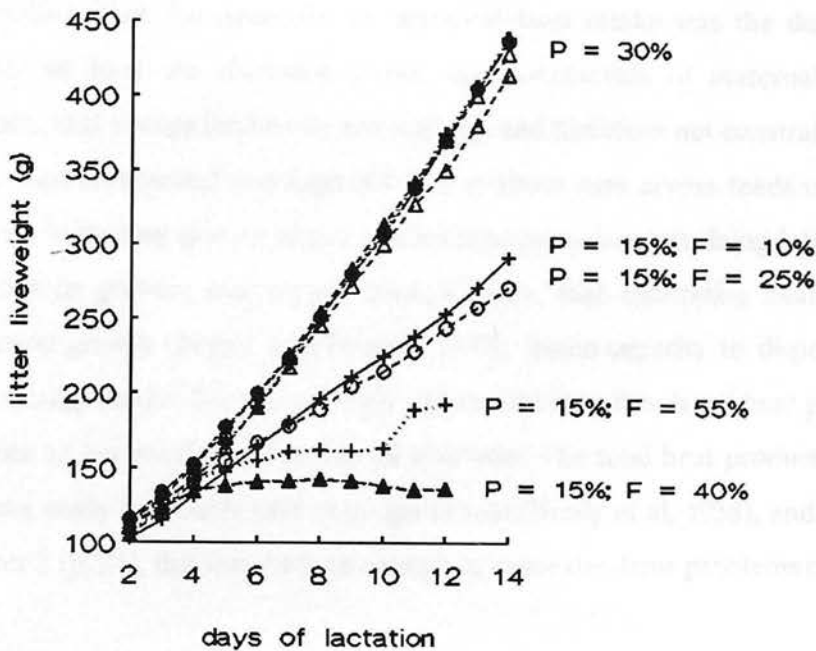
the intake of energy was either excessive on the low carbohydrate/high fat feeds or being constrained on the high carbohydrate/low fat feeds. These two contradictory conclusions are discussed below

If the energy intake of the low carbohydrate/high fat feeds was excessive, then it is difficult to reconcile the considerable losses of maternal body lipid which occurred on these feeds. Mobilisation of large amounts of lipid, is usually an indication that dams are short of energy. Thus in this situation, a reason for body lipid mobilisation other than supplementation of an energy deficit is required. One explanation would be, that under non-limiting conditions the size of the body lipid store is related to the animals physiological state. If this were the case and if maximum lipid store size decreased through lactation, then dams which had previously attained their desired fatness would be obliged to lose body lipid. Even if there were an obligatory body lipid loss, this does not adequately explain why energy intake increased as feed carbohydrate content decreased. No adequate explanation for this can be advanced given the assumption that energy intake was excessive.

If the energy intake on the high protein feeds was constrained, a more satisfying explanation for the observed effects can be constructed. This explanation relies on the assumption, that the constraint on maternal food intake was the dams' capacity to dispose of heat. As discussed above, the mobilisation of maternal lipid reserves suggests, that energy intake was not maximal and therefore not constraining intake. No other feed component was ingested at a constant rate across feeds of equal protein content, indicating that no single feed component was constraining intake. It has been shown with growing rats on low protein feeds, that facilitating heat loss results in improved growth (Meyer and Hargus, 1959), hence capacity to dispose of heat was constraining intake. For this to apply to lactating rats, the dams' heat production must be close to her maximum capacity to lose heat. The total heat production of lactating rats can easily be double that of pregnant rats (Brody et al, 1938), and as described in chapter 2 (p. 24), this may be high enough to cause the dams problems of heat disposal.

If heat production is the constraining factor on food intake, then energy intake is by definition limited and so there is a requirement for energy from body lipid. Conversion of body reserves into milk is more efficient than conversion of food into milk; estimates of 89% and 72% respectively have been cited (Noblet and Etienne, 1987). Body lipid mobilisation would therefore be a means to supplement the limited energy intake, with the minimum possible increment in heat production.

This explanation also accounts for the observed increase in energy intake as feed fat replaced feed carbohydrate. Forbes and his co-workers (1946a,b) have shown in non-lactating rats, that the heat increment per unit feed energy decreases as fat replaces carbohydrate in that feed, at equal protein intakes (summarized by Swift and Black, 1949). Therefore, in order to reach the same feeding derived heat production, more feed energy could be ingested as feed carbohydrate content decreased. This would be further accentuated by the decline in the protein:energy ratio, as the feed carbohydrate content declined and consequently the energy content of the feed increased.

These arguments suppose, that the heat production of the dam, other than the heat

increment, is constant. Whilst adequacy of protein and carbohydrate supply exists, such an assumption is justified. Subadequate protein or carbohydrate supply may result in a drop in milk production with consequent changes in the dams' metabolism and heat production, rendering the assumption false.

The final step down in feed carbohydrate content on the high protein feeds, from feed 6 to feed 8, resulted in a drop in energy intake and in litter lipid gain. This was contrary to the trend seen with decreasing carbohydrate content of the other high protein feeds. Within the limitations of the experimental numbers, the effect of decreasing feed carbohydrate from 300 g/kg OM to 150 g/kg OM (feed 6 to 8) was not significant and therefore could be ascribed to unaccounted variation. The initial litter liveweight on feed 8 was on average lower than on feed 6, 105 and 115 g respectively (sed=3.5). However, it would be reasonable to suggest, that feed 8 represents a situation of protein or carbohydrate deficiency, or both, and that consequently the assumption of comparability with the other high protein treatments no longer holds. For instance, a shortage of carbohydrate supply would either result in a fall in milk production or in an increased conversion of protein to glucose. The former would decrease the total heat of milk production, and would divert nutrients from milk production to body reserves or to catabolism. The latter would increase the heat production due to de-amination and gluconeogenesis. Both would alter heat production, and this is only one scenario in a fairly complex set of possibilities.

The hypothesis, that food intake and hence lactational performance was being constrained by a maximal heat production, has been developed from considerations of the results for the high protein feeds. This is now discussed in relation to the low protein feeds.

It is obvious, that the dams on the low protein feed had a shortage of protein; maternal protein losses were several times greater than the losses on the equivalent high protein feed. Consequently, litter growth was severely depressed. Food intake of the dams on· the low protein feeds was also depressed. Within the context of a maximal heat production this is not surprising, since the protein:energy ratios of all the low protein

feeds are lower than those of all the high protein feeds. Thus achieving an adequate protein intake on the low protein feeds requires the use or disposal of a far greater amount of energy than on the high protein feeds. If feed energy content *per se* is put forwards as the constraint on intake then, as with the high protein feeds, there exists the apparent contradiction of a constrained feed intake and a concomitant maternal lipid loss.

Accepting that the heat increment of the feed was the main factor limiting the intake of the low protein feeds, then the decrease in energy intake as feed carbohydrate was replaced by feed fat can be explained. This effect is the opposite of that seen with the high protein feeds, however these feeds were protein limited and resulted in a decreased milk production. A lower milk production also represents a lower capacity for energy disposal into milk. Thus dams would be forced to dispose of a larger amount of energy by other means. If the carbohydrate content of the feed was limiting milk production, then decreasing the carbohydrate content of the feed would reduce milk production and consequently increase the energy surplus which required disposal. Even if the carbohydrate content of the feed was not limiting (feeds 1 and 3), replacing feed carbohydrate with feed fat would increase the energy content of the feed, exacerbating the problem of disposal. Especially for feeds in which carbohydrate content was limiting milk production, it is possible to envisage a threshold in feed composition which, once exceeded, results in a large drop in feed intake.

Using heat production as the constraining factor, a simple scheme for regulation of food intake can be constructed, as shown in figure 63. Of central importance to this scheme is the milk production possible from the carbohydrate and protein intake. It is reasonable to suppose, that the dam would regulate her conversion of available protein to glucose so that milk volume at an acceptable protein:lactose ratio was maximal. If the carbohydrate and protein content of a feed were very low, the resultant drop in milk energy yield would lead to an increased heat of disposal. If that increased heat of disposal was greater than the concomitant fall in heat of milk production, total heat·

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Fig. 63: A scheme for regulation of food intake by lactating dams.
Solid lines are mass transfers; stippled lines are heat productions.

Food intake

Protein intake   Carbohydrate intake   Fat intake

Body protein

Possible protein limited milk production

Possible carbohydrate limited milk production

Body fat

Actual milk production is the smaller of the two possible productions

milk fat production

Heat of protein catabolism   Heat of milk production   Maintenance heat   Surplus   Heat of fat synthesis

Total heat production (THP)   Capacity to lose heat (HLC)

HLC>THP?

Y

must decrease food intake   N   no need to decrease food intake

production would rise. In a situation where heat production was already maximal, food intake would be forced down. In such a scheme, a spiralling down of feed intake could be an outcome, if the reduction in intake led to a further drop in milk production, leading to an increase in total heat production, forcing a further drop in intake and so on. Thus, the threshold feed composition would be that composition, where the drop in heat increment was equal to the increase in [heat of disposal - heat of milk production] resulting from a decrease in feed intake. To calculate this would require detailed knowledge of the biochemical and energetic efficiencies of conversion of feed carbohydrate and protein to milk lactose and milk protein, and their interconversion; the possible range of milk compositions; the energetic efficiencies of conversion of feed to maternal body stores and vice versa; and the capacity for heat loss of the dam.

Such a spiralling down of feed intake would, however, reach a point where the total heat production no longer exceeded the dams' capacity for heat loss. Yet, as can be seen in figure 54a, the dams on feeds 5 and 7 reduced their daily feed intake to very low, submaintenance levels, so this model alone cannot fully explain these results.

When considering the high fat, low protein feeds (5 and 7), it is difficult to explain the catastrophic effect that these feeds had on lactational performance. Alternative explanations for this dramatic interaction between feed components on lactational performance have been classified and discussed as follows:

- Nutrient deficiencies
- Toxicity and unpalatability
- Metabolic disorders

Nutrient Deficiencies: By the nature of the experimental design, this result cannot be explained as the effect of low feed protein content or of low carbohydrate content alone. There were high protein feeds of lower carbohydrate content than feeds 7 and 8, and other feeds of equal protein content on which dams and litters performed very · much better (figure 51a). Using [protein + carbohydrate] as a crude measure of glucogenic material, feeds 6 and 8 were of equal or lower [protein + carbohydrate]

content than feeds 5 and 7. The high protein feeds were of similar "glucogenic" content to the low protein feeds per unit energy content of the feed. Yet they resulted in vastly superior lactational performance.

One cause of concern in the design of the experiment was that despite meeting NRC requirements (1978), the level of mineral and vitamin inclusion in preceding experiments (R1, R4) may have been too low, in particular for the high fat feeds, where it was possible that insoluble mineral soaps might form. In this experiment (and R3) the mineral and vitamin contents were doubled to counter this possibility. In relation to NRC recommendations these contents were adequate, provided that the intake was at least 84 g/12d. Except for feeds 5 and 7, all rats had intakes greater than this (more than 4 times this level). If vitamin and mineral contents of the feeds were imbalanced or inadequate, the expectation would have been, that difficulties and reduced intakes would have been found with all feeds. A very short time elapsed between the start of the treatments and the virtual cessation of food intake on feeds 5 and 7 (figure 66). Further, no such disruption to intake was found with the other feeds. The depression in intake was far too rapid to be attributable to a mineral or vitamin deficiency.

Toxicity or Unpalatability: Intakes as low as those on feeds 5 and 7 suggest, that these feeds were either toxic to the rats or that they resulted in illness causing loss of appetite. Given the design of the experiment, there is no convincing evidence for these feeds being toxic. All eight feeds were made using the same batch of ingredients and were mixed and stored (frozen) in the same manner at the same time. Unpalatability cannot be ascribed to any one component of these feeds (5 and 7) on its own, since there were always other feeds which had an equal or higher content of that component. Similarly, there were always other feeds (than 5 and 7) which contained equal or lower amounts of any one component. Thus, to propose a viable explanation on the grounds of unpalatability requires the effect to be the result of an interaction between two or more components of the feed, which only occurs within a limited range of the composition of the total feed mixture. Most examples of feed preference, ascribed to · palatability have been shown to arise from poor interpretation or poor design of the

Fig. 66: The effect of feed composition
on average daily maternal food intake.

feed number —△— 1   --▲-- 2   —▽— 3   --▼-- 4
             —◆— 5   ···○··· 6   ···+··· 7   —□— 8
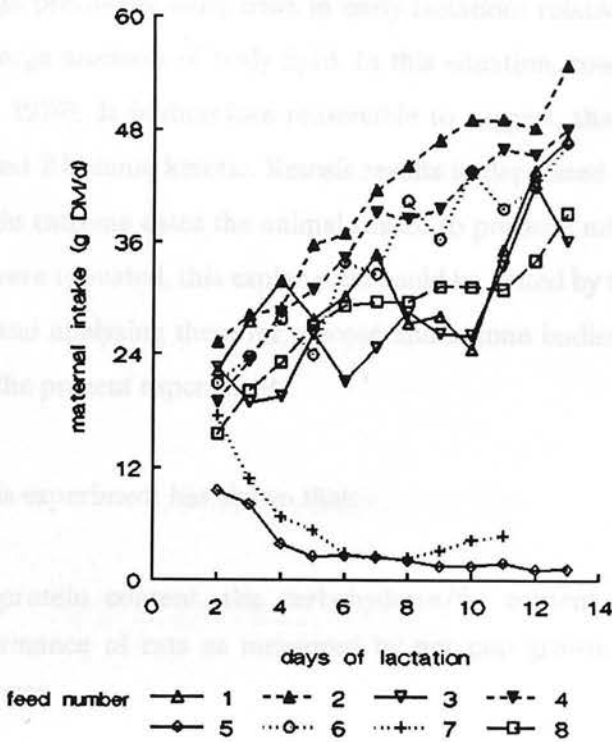
experiment in question (Kyriazakis, 1989). Palatability cannot be measured in absolute terms, but must be expressed as the preference for a given food *relative* to another food or to no food. The usefulness of such a concept is therefore questionable. When offered only one food, the preference for that food over no food is influenced by physiological state and environment. The ingredients used in feeds 5 and 7 were palatable for lactating rats when mixed in other proportions (all other feeds). It is extremely difficult to envisage a combination of these ingredients, which in other proportions are palatable, being so unpalatable as to make maternal weight losses of 30% and negative litter weight gain preferable to eating the food.

Metabolic Disorders: A far more plausible hypothesis is, that these feeds resulted in a metabolic disorder associated with appetite loss by the dams. The circumstances suggest, that a ketotic state may have been induced. These two feeds were of low glucogenic content and with the highest energy content per unit protein. Assuming that the food intake of these feeds was forced down to a low level, as suggested in the scheme on page 63, then these dams attempted to maintain milk production by massive

use of body reserves, greater than on all the other feeds. This is a situation similar to that found in high producing dairy cows in early lactation; relatively short of glucose, and mobilizing large amounts of body lipid. In this situation, cows are most prone to ketosis (Hibbitt, 1979). It is therefore reasonable to suggest, that the lactating dams offered diets 5 and 7 became ketotic. Ketosis results in depressed milk production and loss of appetite, in extreme cases the animal ceases to produce milk (Schultz, 1979). If the experiment were repeated, this explanation could be tested by taking blood samples from the dams, and analyzing them for glucose and ketone bodies. Unfortunately this was not done in the present experiment.

In conclusion, this experiment has shown that:

i) At constant protein content, the carbohydrate/fat content of the feed affects lactational performance of rats as measured by net pup growth and maternal body composition changes.

ii) At constant carbohydrate or fat content, the feed protein content affects lactational performance.

iii) There is an interaction between feed protein, carbohydrate and fat contents, which results in massive depression of lactational performance at low protein, low carbohydrate contents.

Further, the discussion has proposed that:

iv) The collapse of lactation at low protein, low carbohydrate content may be the result of ketosis in the dams.

v) The effects observed can be explained by the hypothesis, that heat production is maximal in these lactating dams and constrains food intake.

vi) That to adequately explore this hypothesis requires the construction of a more complex model allowing it to be tested both quantitatively and qualitatively.

CHAPTER 6

The Third Rat Experiment (R3)

Critical Nutrient Proportions for Lactation

Note: All tables and figures are numbered according to the page on which they are found. As such table and figure numbers are not consecutive (see list of tables and figures, p. ix).

Introduction

The results of the preceding experiment (R2) showed that there was a huge interaction between feed protein, carbohydrate and fat content, such that relatively small changes in feed composition could have a catastrophic effect on lactational performance (figure 59). As elaborated in the discussion of that experiment (p. 60), a possible explanation of the phenomenon is based on the assumption that maternal heat production is maximal and that this limits the intake of certain feeds. The lactational failure seen on these feeds may have been the result of a metabolic disorder such as ketosis. These feeds were of low glucose content and the dams were mobilising large amounts of body reserves; conditions predisposed towards ketosis. A model, described in the next chapter (7), explores this argument quantitatively.

A complication in the interpretation of the results of experiment R2 was that proportions of nutrients, particularly protein in relation to energy, changed in the different feeds. The effects of changes in feed composition could therefore be ascribed to changes in nutrient proportions or to changes in the nutrient:energy ratios. In this experiment (R3), this was investigated by comparing feeds of equal nutrient:energy ratio. Mapping the feeds from R2 and R3 onto two triangles, one of *mass* proportions (g/kg OM) and one of *energy* proportions (kJ/MJ OM) clarifies this difference (figures 70a and 70b).

In the triangle of energy proportions (figure 70b), lines of constant protein mass:total energy ratio run parallel to the base of the triangle. These lines are directly related to lines of constant protein:energy in the triangle of mass proportions as follows:

Define: Protein content (g/kg) = P
Carbohydrate content (g/kg) = C
Fat content (g/kg) = F

$$\text{Protein energy/Total energy} = (23.9P)/(23.9P + 16.5C + 39.6F)$$

Therefore $P/(\text{Total energy}) = (1/23.9)(\text{Protein energy/Total energy})$

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The objectives of this experiment were as follows:

- To further define the interaction between lactational performance and ... from the interaction between trait components.

- To account for the effect of differences in nutrient:energy ratio on this interaction.

- To obtain further information for constructing a lactation model ... the effect on lactation of decreasing the protein:energy ...

A limited number of rats was available for this experiment (feed R3) and the experiment designed with this constraint is shown in figure 70a. Three different feeds were offered, one of which was a replicate of a feed from the previous experiment. The following expectations were held:

1) That the performance of rats in this experiment on the repeat feed (feed III) from the last experiment would ... the performance of those rats in the previous experiment (R2, feed ...).

2) Decreasing the protein content of the feeds would cause a depression in litter growth and intake, accompanied by an increase in maternal body catabolism.

3) Provided that feeds I and II were of non-limiting carbohydrate content it was expected there would be no difference between their lactational performance and that the protein:energy ratio would be the main determinant of total intake (because of the heat production associated with disposal of the net dietary ...

4) Since feed I from the previous experiment (R2), was of a similar protein:energy ratio it was expected that lactational performance on this feed would be similar to that of feeds I and II (R3).

Fig. 70a: Feeding treatments for the second (●) and third (■) rat experiment, mapped onto a triangle of mass proportions. The solid line shows constant protein : energy ratio.
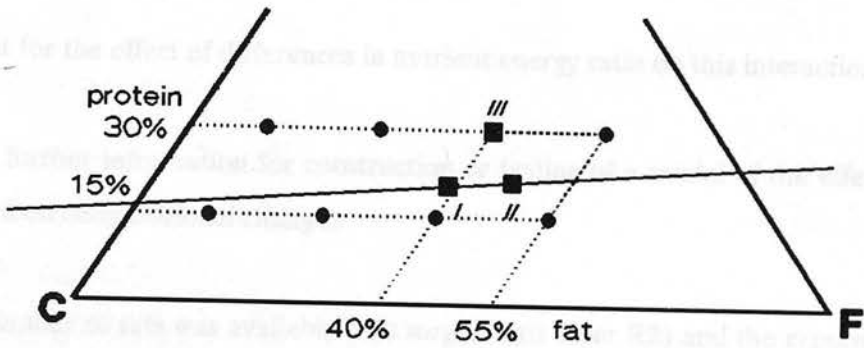
Fig. 70b: Feeding treatments for the second (●) and third (■) rat experiment, mapped onto a triangle of energy proportions. The lines show constant nutrient : energy ratio.
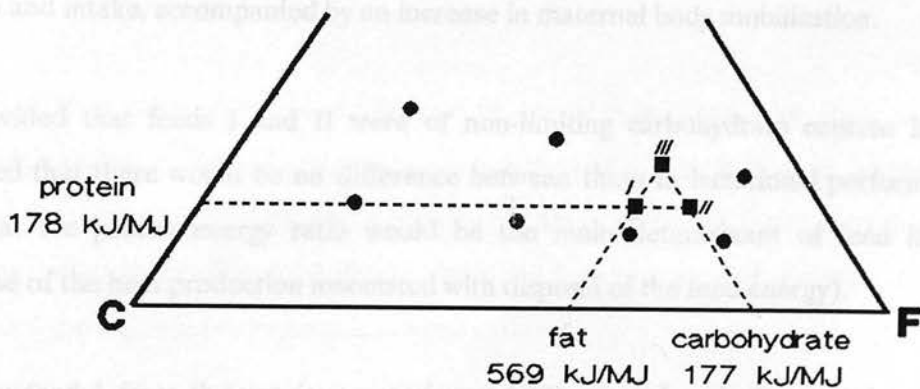
The objectives of this experiment were as follows:

- To further define the threshold found in R2 between adequate lactational performance and catastrophic lactational performance resulting from the interaction between feed components.

- To account for the effect of differences in nutrient:energy ratio on this interaction.

- To obtain further information for construction or testing of a model of the effect on lactation of feed compositional changes.

A limited number of rats was available (the surplus rats from R2) and the experiment designed with this constraint is shown in figure 70a. Three different feeds were offered, one of which was a replicate of a feed from the previous experiment, The following expectations were held:

1) That the performance of rats in this experiment on the repeat feed (feed III) from the last experiment would be the same as that of the rats on that feed in the previous experiment (R2, feed 6).

2) Decreasing the protein content of the feeds would cause a depression in litter growth and intake, accompanied by an increase in maternal body mobilisation.

3) Provided that feeds I and II were of non-limiting carbohydrate content it was expected that there would be no difference between them in lactational performance and that the protein:energy ratio would be the main determinant of feed intake (because of the heat production associated with disposal of the feed energy).

4) Since feed 1 from the previous experiment (R2), was of a similar protein:energy ratio it was expected that lactational performance on this feed would be similar to that of feeds I and II (R3).

Table 72: Composition of the feeds used in the third rat experiment (g/kg OM unless otherwise stated).

Feed	I	II	III
Measured composition*			
DM (g/kg fresh)	492.4	483.6	463.6
CP	199.8	214.9	301.3
EE	354.9	442.6	374.2
CHO	445.3	342.2	324.5
GE (MJ/kg OM)	26.41	28.58	27.53
Calculated composition (mass)			
CP	204	212	300
EE	389	470	400
CHO	407	312	300
GE (MJ/kg OM)	27.0	28.8	28.0
Calculated composition (energy)			
CP (kJ/MJ OM)	178	178	254
EE (kJ/MJ OM)	569	644	569
CHO (kJ/MJ OM)	253	177	177

* DM is oven dry matter at 60°C; CP is crude protein = N x 6.407 (see p. 32); EE is ether extract calculated from gross energy; CHO is carbohydrate calculated as (1000-CP-EE). Mineral content is 100 g/kg DM.

5) If the proviso in expectation 3) was not met, it would be expected that performance on feeds I and II would be different. Since feed II contained less carbohydrate than feed I, it was expected that lactational performance would be poorer on feed II than on feed I

Method

A total of 20 lactating dams were allocated to one of four treatments; three feeding treatments and an initial cull group. The composition of the feeds is given in table 72. Littersize was standardized on day 1 of lactation to 12 pups per litter. Experimental feeds and water were offered ad libitum from day 2 until day 14 of lactation. During this period the following daily measures were made:

        Food intake
        Water intake
        Maternal liveweight
        Litter liveweight
        Room temperature

The initial cull was carried out on day 2 of lactation with the remaining rats being culled on day 14 of lactation for carcass analysis. Carcasses were analysed for water, nitrogen, ether extract, ash, and gross energy. Further details of the method are described in chapter 3. The full data are presented in appendix 4.

Results

Treatment means for litter growth and maternal body mobilisation are presented in figures 74a, 74b and 75; feed intake results are presented in figures 76a and 76b. The differences between treatments were all highly significant ($p < 0.01$) except maternal lipid loss which was significant at the 5% level. It should be noted, that the mean values for feed II hide a large variation between individuals. Levene's test for homogeneity of variance (Snedecor and Cochrane, 1980) was applied to the data. Those measures which did not show homogeneity of variance were transformed to $\log_{10}$ values for analysis of variance. This transformation did not alter the statistical significance of the results; the standard errors (of the difference) presented are from the untransformed analyses. The large variation between individuals on feed II is considered separately in the discussion and shown in figure 77 and table 77.

Comparison between Experiments of the Same Feed: Lactational performance on feed III was not significantly different from the lactational performance achieved on the same feed in the previous experiment (feed 6; R2). This is an important result for further comparison between the present and preceding experiments.

Comparison between Protein:Energy Levels (feed III vs feeds I, II): Feed intake results are presented in figures 76a and 76b. As expected, intake of feed III was significantly greater than the intakes of feeds I and II ($p < 0.05$). Consequently, dams on these feeds had decreased intakes of protein and energy, resulting in significantly poorer litter growth and greater loss of maternal body stores (figures 74a and 75).

Fig. 74a: Maternal and litter liveweight gains (g/12 days);
sed = 12.2 and 44.8 g/12 days for maternal and litter
gains. Feed 1 from R2 is shown for comparison.

protein
30%

−5.4 maternal
345   litter

15%

−44.3 −83.3 maternal
273   117   litter

−42.7
190

C

40%   55% fat   F
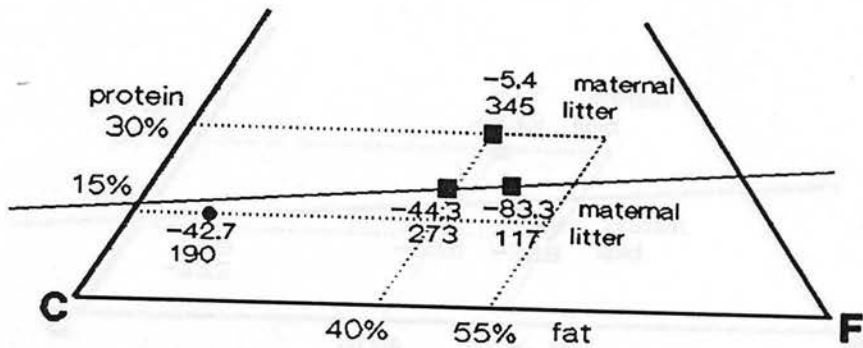
Fig. 74b: Litter gains of body protein and fat (g/12 days);
sed = 5.2 and 8.1 for body protein and fat. Feed 1
from R2 is shown for comparison.

protein
30%

51.6   protein
48.6   lipid

15%

39.2   21.5 protein
44.2   15.9 lipid

30.4
21.5

C

40%   55% fat   F

74
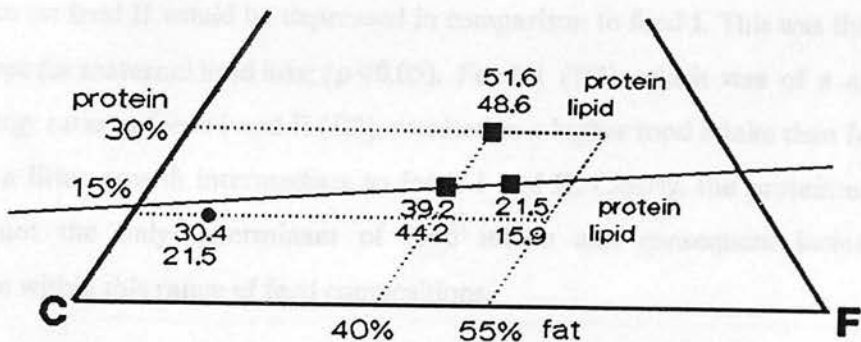
Fig. 75: Maternal gains of body protein and fat (g/12 days); sed = 2.4 and 4.3 for body protein and lipid. Feed 1 from R2 is shown for comparison.
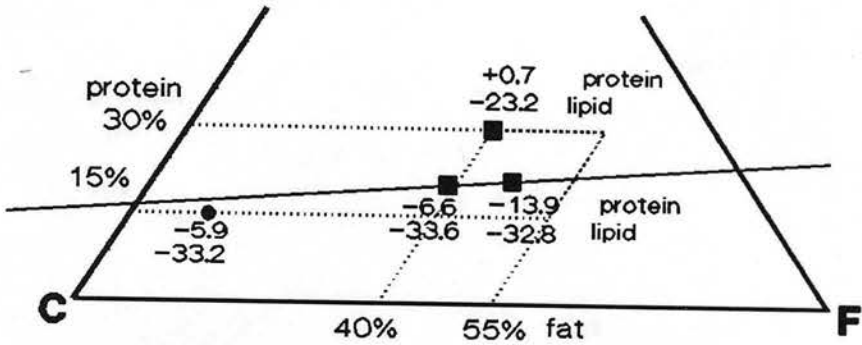
Comparison between Feeds at Constant Protein:Energy ratio: Feeds I and II were of equal protein:energy ratio. Despite this, intakes of protein and energy on feed I were greater ($p < 0.01$) than on feed II (figure 76b). This was contrary to expectation 3 and resulted in significantly ($p < 0.01$) better litter growth on feed I (figure 74a). Given that expectation 3 was not met, it was predicted (expectation 4) that intake and lactational performance on feed II would be depressed in comparison to feed I. This was the case ($p < 0.01$ except for maternal lipid loss; ($p < 0.05$). Feed 1 (R2), which was of a similar protein:energy ratio as feeds I and II (R3), resulted in a higher food intake than feeds I and II and a litter growth intermediate to feeds I and II. Clearly, the protein:energy ratio was not the only determinant of food intake and consequent lactational performance within this range of feed compositions.

Individual Variation within Treatments: Daily litter liveweights are presented in figure 77. For feeds I and III, the average treatment values have been plotted, for feed II individual rat data are shown. Individual differences in litter and maternal gains for

75

Fig. 76a: Maternal organic matter intake (g/ 12 days; sed = 59). Feed 1 from R2 is shown for comparison.
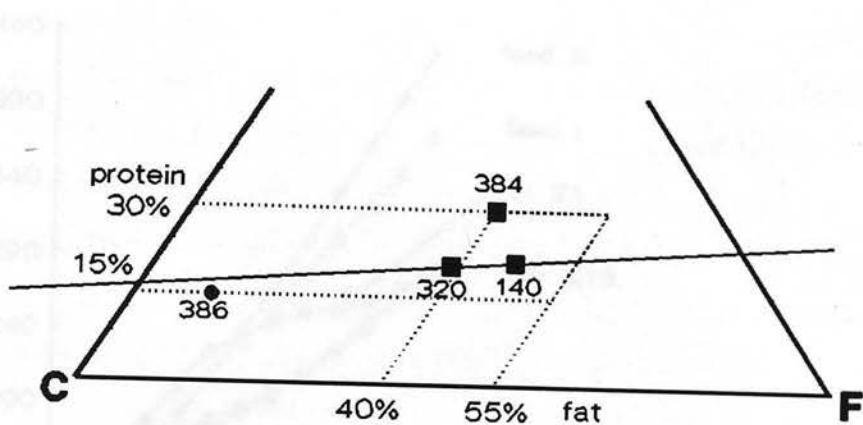
Fig. 76b: Maternal intake of gross energy (MJ/12 days) and protein (g/12 days); sed = 12.0 g/12 days for protein and 1.5 MJ/12 days for energy intake. Feed 1 from R2 is shown for comparison.

three of the rats on feed II, ... on this feed are presented in table 77.
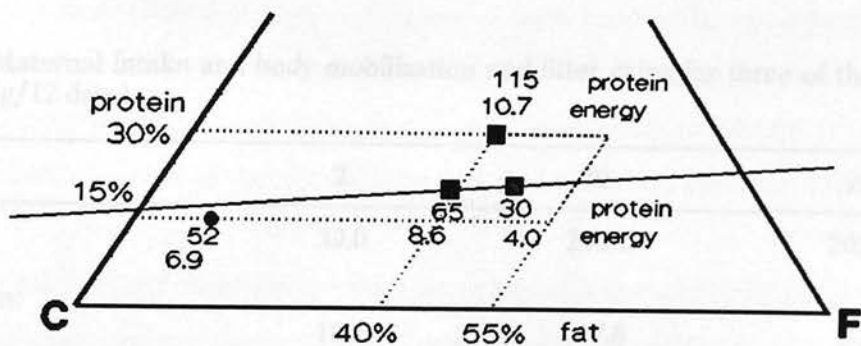
Fig. 77: The variation in litter growth supported by individual dams on the same feed (II).

It is clear from these results that the depression in reproductive performance observed in the preceding experiment ... ... relatively on a change in feed composition resulted ... variation in performance (figure). In experiment, a ... similar flows rat 21 (142 g/kg OM) transferring largely by ... of reproductive efficiency corded litter growth and maternal ...

Unlike this ... ... was associated with changes in the protein energy ... of the feed. The previous experiment ... that the protein concentration ... effect of the feed. The ... in feed intake or feed protein content ... not only be confined to altering in the current results changes in protein energy content. This hypothesis has been developed in greater detail in the discussion to the preceding experiment.

However, the current results comparing feeds of constant protein/energy ratio clearly show that this was not the only determinant of lactational performance. Had it been, then feeds I and II (R3) and feed 3 (R3) would have resulted in equal lactational ...

Table 77: Maternal intake and body mobilisation and litter gains for three of the rats on feed II (g/12 days).

Rat no.	2	19	21
Intake	39.0	244.1	201.7
Litter gains:			
Protein	15.1	27.6	27.6
Fat	17.4	20.8	19.9
Maternal gains			
Protein	-21.2	-3.8	-16.0
Fat	-25.6	-35.3	-30.8

three of the rats on feed II, representing the extremes of performance on this feed, are presented in table 77.

Discussion

It is clear from these results that the threshold in feed composition for lactational adequacy, observed in the preceding experiment, is very sharp indeed. Relatively small changes in feed composition resulted in massive differences in lactational performance (figure 74a). In particular, a decrease in carbohydrate content from 445 to 342 g/kg OM (substituting largely for fat; 88 g/kg OM) at constant protein:energy caused litter growth and maternal intake to halve and maternal body losses to double.

Unlike the previous experiment, this effect was not confounded with changes in the protein:energy ratio of the feed. The previous experiment suggests, that the protein:energy ratio is an important characteristic of the feed. The depression in feed intake as feed protein content fell can only be explained by alluding to the concomitant changes in protein:energy content. This hypothesis has been developed in greater detail in the discussion to the preceding experiment.

However, the current results comparing feeds of constant protein:energy ratio clearly show, that this was not the only determinant of lactational performance. Had it been, then feeds I and II (R3) and feed 1 (R2) would have resulted in equal lactational performance. The difference between feeds 1 (R2) and I (R3) can be explained in terms of a constraining heat production, feed II requires an additional factor to explain the performance achieved on that feed. Feed I and feed 1 (R2) are considered first.

Feed I resulted in significantly greater litter growth ($p < 0.05$) than feed 1 (R2). Since the protein:energy ratios of these two feeds were the same, this cannot be attributed to the higher protein content of feed I. Dams on feed I achieved a greater energy intake than those on feed 1 (R2), 8.6 and 6.9 MJ/12d respectively. Had they eaten equal amounts of energy, their protein intake would have been identical. The major difference between these two feeds was in the carbohydrate content (feed I, 407 g/kg

OM; feed 1 (R2), 750 g/kg OM), the balance being largely feed fat. That a decline in feed carbohydrate content (at constant protein) allows an increase in energy intake has been found in previous experiments (R1, R2) and is in agreement with Maynard and Rasmussen (1942). It can be explained by the heat increment of feed carbohydrate being greater than that of feed fat (Forbes et al, 1946a,b,c). Hence the heat increment per unit feed declines as carbohydrate content declines, permitting a greater energy intake to achieve the same heat production. The lower feed carbohydrate content of feed I therefore allowed the dams on this feed to have a higher energy and protein intake than the dams on feed 1 (R2) and consequently to raise heavier litters.

However, decreasing the carbohydrate content further from 413 g/kg OM in feed I to 312 g/kg OM in feed II at a constant protein:energy ratio, did not result in a further amelioration of lactational performance. The observed decline in lactational performance clearly does not conform to the hypothesis, that decreasing carbohydrate content will allow an increase in energy intake. The reason is, that the hypothesis has an important accompanying proviso which does not hold on feed II. It only holds *provided* that total heat production is not markedly altered by the change in carbohydrate content. A change in total heat production would occur if milk production was impaired. There was probably an inadequate glucose supply from feed II to maintain the milk production possible at that protein:energy ratio. In this situation, described in greater detail in chapter 5, (pp. 60-64), food intake is forced down as milk production declines, because heat disposal into milk decreases faster than heat production from food intake. On extreme feeds this probably leads to metabolic disorders such as ketosis.

Alternative potential causes of such a depression in milk production, for instance a mineral deficiency or a toxic effect, have been discussed and discounted in the discussion to the preceding experiment (R2) and are no more plausible there than in the current experiment.

On those feeds in the preceding experiment (feeds 5 and 7) where the threshold of nutritional support for lactational adequacy was exceeded, the effect was an almost

complete cessation of litter growth and of feed intake, probably due to the onset of a ketosis or similar metabolic disorder. In the current experiment, feeds were chosen to be as close to that threshold as possible. Consequently. the average effect of feed II was not as severe as the effects of feeds 5 and 7 (R2). The individuals on feed II fell into two distinct categories, those who could maintain a lactation, albeit of limited capacity, and those which failed to maintain their lactation (figure 77 and table 77). On a marginal feed such as this one, the importance of maternal body reserves for lactational support is amplified. The point at which daily litter liveweight gains cease for rats 2 and 12 (figure 77) is probably the point at which their reserves of protein are depleted. The characteristics of the individual dam that govern the extent to which she will mobilise reserves are clearly important.

Litter protein gain was related to the food intake of the dams. An attempt to relate dry matter intake, maternal protein and maternal lipid losses to estimated initial body composition (g) was made (table 81). There was no relationship between these factors and the ability to maintain (or not) intake on feed II. Considerably more data and more detailed measures of initial state would be required to investigate this further. This assumes, that it is known which measures best describe these maternal characteristics.

In conclusion, this experiment has shown that:

i) An extremely abrupt threshold in feed composition for lactational adequacy is encountered as feed carbohydrate content decreases in low protein feeds of constant protein:energy.

ii) The effects of changing feed composition cannot be explained by changes in any single nutrient:energy ratio alone.

iii) The exact threshold feed composition is to some extent dependant on poorly defined characteristics of the individual animal.

Table 81: The relationship between intake, body mobilisation, and initial body reserves for dams on feed II. (Initial body reserves derived from regression between initial liveweight and body composition of the initial cull group; see p. 37).

Y variate =	constant +	coeff.(X variate*)	s	R^2adj.(%)
Litter CP gain	8.58	+0.082(DMI)	3.365	86.3
Dry matter intake	292	+10.9(mCPg)	85.96	31.3
Dry matter intake	-11.5	-8.06(mEEg)	108.5	0.0
Dry matter intake	324	-2.43(d2mCP)	117.1	0.0
Dry matter intake	283	-1.94(d2mEE)	116.6	0.0
Litter CP gain	29.2	+0.62(mCPg)	9.356	0.0
Maternal CP gain	22.6	-0.508(d2mCP)	5.564	28.8
Maternal EE gain	-55.1	+0.325(d2mEE)	4.361	35.9

*: DMI is dry matter intake; mCPg is maternal crude protein gain; mEEg is maternal ether extract gain; d2mCP is maternal crude protein on day 2, and d2mEE is maternal ether extract on day 2.

Further the results of this experiment can be explained by the same hypothesis as was proposed in the preceding chapter, namely that the dams capacity to dispose of heat is constrained.

CHAPTER 7

The Lactation Model and the Fourth Rat Experiment (R4)

Prediction of Lactational Performance in Rats Using
Heat Production as the Controlling Factor.

Note: All tables and figures are numbered according to the page on which they are found. As such table and figure numbers are not consecutive (see list of tables and figures, p. ix).

Introduction

The last two rat experiments (chapters 5 and 6) have shown that feed composition affects lactational performance. There was an interaction between feed protein, carbohydrate, and fat which caused a large depression in the lactational performance of dams on feeds with low contents of both protein and carbohydrate. As eloquently discussed by Kronfeld (1976), the demands of lactation decrease the mothers ability to accommodate extremes of food composition because she is functioning at a level close to her capacity. This concept was used to discuss the above results according to the following hypothesis:

Food intake, and therefore lactational performance, is being constrained by the capacity of the dams to dissipate heat.

There is no direct experimental evidence to support this hypothesis. However, Brody et al. (1938) reported the heat production of lactating rats to be 4.6 kJ/weight(g)$^{0.73}$/day at 28°C. For a 250g rat this is a heat production of 259 kJ/day. By comparison, the basal heat production of fasted, non-lactating 250g rats in a thermoneutral environment is 104 kJ/day ((2.9 MJ/m²/d)x(0.09*weight(kg)$^{2/3}$); Herrington, 1940 and Meeh, 1879, respectively). Non-lactating (250g) rats which are trying to lose heat only reach that level of heat production at an environmental temperature of approximately 42°C. Rats kept at a temperature of 45°C for two hours died (Kirmiz, 1962). These observations indicate that the above hypothesis is tenable.

Given the above hypothesis, it was proposed that the interaction between feed components on lactational performance was due to changes in the resultant heat production by the lactating animal. The manner in which changing feed composition would affect maternal heat production is difficult to predict intuitively. There are a number of possible consequences of changing feed composition on energy balance and heat production which have been categorised as follows:

1) The heat increments associated with protein, carbohydrate, and fat are different (Forbes et al, 1946a,b,c). Hence changes in feed composition would result in changes in the overall heat increment of the feed.

2) Disposal of feed energy into milk is an important part of the dams' energy balance. The volume of milk produced determines the maximum amount of energy which the dam can dispose of as milk fat (Mueller and Cox, 1946). The volume of milk produced is proportional to the amount of lactose produced. Further, there appears to be an association between production of milk lactose and milk protein, as suggested by the small variation in milk protein content (Mueller and Cox, 1946). Thus the level of milk production is largely dependent on the intakes of carbohydrate and protein. Given that food intake is affected by the composition of the feed, it follows that milk fat production and therefore the energy surplus (to milk production) per unit feed is affected by feed composition.

3) Variations in feed composition result in differences in maternal body mobilisation (chapters 5 and 6). It is possible that this may be a response to maximal heat production rather than a direct effect of feed composition. However, it still represents a changing component of maternal energy balance and as such has an effect on heat production.

To test the hypothesis that food intake, and hence lactational performance, is being constrained by the capacity of the dams to dissipate heat, a model was constructed. The model was designed as a tool to aid in the discussion of observed results and not to provide absolute outputs. It was constructed in two parts, one based on back calculation from litter growth, and the other based on calculation from maternal inputs. The two model parts have been presented as flow diagrams in figures 86 and 87. Both parts were derived from balance equations for protein, carbohydrate, and fat mass, and from an equation balancing energy inputs and outputs. The final equations were computed using Minitab software. The development of the model, the assumptions and definitions which are built into it are described under the following headings:

- Objectives
- General Assumptions
- Notation of Variables and Constants
- Prediction of Milk Production from Litter Growth Data.
- Prediction of Lactational Performance from Maternal Data.
- Derivation of Constants and Known Variables.

Objectives

1) To calculate maternal milk output from litter growth data.

2) Given feed composition, maternal body losses, milk production, and maternal food intake; to calculate maternal heat production.

3) Given feed composition, maternal body losses, a capacity for heat disposal, and maternal food intake; to calculate milk production.

4) Given feed composition, maternal body losses, and a capacity for heat disposal; to calculate maximum maternal food intake and milk production.

If any one objective is not satisfactorily met then subsequent objectives cannot be achieved.

General Assumptions

A1) That the ratio of protein to carbohydrate in milk is constant; without this assumption milk carbohydrate cannot be calculated. Davies et al. (1983) showed a negative relationship between protein and lactose concentrations in the milk of 130 different species with a slope of approximately 2.3 (protein/lactose). Within the species *Rattus Norvegicus*, there is variation between experiments in reported protein:carbohydrate ratios (Mueller and Cox, 1946; Luckey et al, 1955; Rolls et al,

Fig. 86: A model of lactational performance. I. The scheme by which milk production was calculated from litter growth. Solid lines are mass transfers; stippled lines are heat production. Lines which have the same number are mutually exclusive.

1963; Fleckbeck and Rivenbark, 1965). However, within experiments the protein:carbohydrate ratio is reasonably constant across a wide range of dietary treatments (Mueller and Cox, 1946; Leibet al., 1963; Rolls et al, 1969; Fleckbeck and ...hausen, 1967). This divergence may reflect the inability of obtaining representative samples of milk from rat (as discussed in section). A protein:carbohydrate ratio in rat milk of 3:4 was used in the model. Three assumptions underlie the model:

A.I. That the growth of the litter is limited by the protein supply in the milk; the litter holds no surplus protein. This assumption is necessary to calculate the carbohydrate supply from milk, as opposed to supply from maternal metabolism. Since the growth of newborn animals is rapid, this is a justifiable assumption.

A.II. That energy requirements of maintenance and costs of producing milk (measured as maternal) are met primarily by oxidation of carbohydrate and maternal body energy, apart from obligatory protein catabolism. The primary substrate oxidised; the energy available from carbohydrate that is oxidised to meet the remaining energy requirement of the dam, and the surplus requirement is converted to carbohydrate and oxidised.

A.III. That the heat production of resting rats is equal to their thermally absorbed heat. The arguments in support of this assumption have been discussed in the two preceding chapters. The calculation of maternal heat production (without this assumption) returns in values similar to those calculated for maternal oxygen) to a certain limit. Only this assumption can be used to calculate maternal heat production independently of the maternal energy balance. This would allow milk production to be calculated from ... milk data only.

Definition of Variables and Constants

All the variables are in g DM per unit time, except for those variables which are in energy terms (EMA, HEAT, and HCAP) expressed as kJ per unit time.

Fig. 87: A model of lactational performance. Solid lines are mass transfers; stippled lines are heat productions. Lines which have the same number are mutually exclusive. The scheme by which maternal production of heat and milk was calculated.

Figure labels: Feed fat intake; Feed carbohydrate (CHO) intake; Feed protein intake; Body fat; Milk fat; Combusted fat; CHO converted to fat; Surplus CHO; Combusted CHO; Maint. CHO; Milk CHO; Maint. energy; Combustion to meet energy requirement to balance heat production; Catabolised protein; Urinary N; Milk prot.; Maint. prot.; Surplus prot.; Body prot.; (1-biol. value)

1986; Fischbeck and Rasmussen, 1987). However, within experiments the protein:carbohydrate ratio is remarkably consistent across a wide range of dietary treatments (Mueller and Cox, 1946; Luckey et al, 1955; Rolls et al, 1986; Fischbeck and Rasmussen, 1987). This discrepancy may reflect the difficulty of obtaining representative samples of milk from rats (as discussed in chapter 3, p. 33). A protein:carbohydrate ratio in rat milk of 2.4 was used in the model (Luckey et al, 1955).

A2) That the growth of the litters is limited by the protein supply in the milk, i.e. the litters have no surplus protein. This assumption is necessary to calculate litter carbohydrate supply from milk, as opposed to supply from protein catabolism. Since the growth of newborn animals is rapid, this is a justifiable assumption.

A3) That energy requirements of maintenance and work of producing mass (expressed as heat) are met primarily by oxidation of carbohydrate and non-carbohydrate energy yielded from obligatory protein catabolism. If the energy requirements exceed the energy available from carbohydrate then fat is oxidised to meet the remaining energy requirements. In the dams, protein surplus to requirement is converted to carbohydrate and urea.

A4) That the heat production of lactating rats is equal to their capacity to dissipate heat. The arguments in support of this assumption have been discussed in the two preceding chapters. If calculation of maternal heat production (without this assumption) results in values similar to those calculated for maternal capacity to dissipate heat, then this assumption can be used to calculate maternal heat production independently of the maternal energy balance. This would allow milk production to be calculated from maternal data only.

Notation of Variables and Constants

All the variables are in gDM per unit time, except for those variables which are in energy terms (EMA, HEAT, and HCAP) expressed as kJ per unit time.

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Variables to be determined:

I   Food intake

PMK   Milk protein

CMK   Milk carbohydrate

FMK   Milk Fat

PCAT   Catabolised protein

CCAT   Catabolised carbohydrate

CCON Carbohydrate converted to fat

FCAT   Catabolised fat

HEAT   Heat production

Variables directly measured or calculated:

BP   Body protein gain

BF   Body fat gain

PMA   Maintenance protein

EMA   Maintenance energy

BPCOR Correction for body protein (defined on p. 90)

BFCOR Correction for body fat (defined on p. 91)

HCAP   Capacity to dispose of heat

Constants:

a	heat of body protein loss\gain	$(29.9\text{kJ/g})*\text{BPCOR/BP}$
b	heat of body fat loss\gain	$(4.4\text{kJ/g})*\text{BFCOR/BF}$
c	proportion of carbohydrate in the feed	g/g
d	heat of milk protein formation	16.7kJ/g PMK
e	heat of milk carbohydrate formation	0.88kJ/g CMK
f	proportion of fat in the feed	g/g
g	heat of milk fat formation	4.4kJ/g FMK
h	heat of protein catabolism	4.9kJ/g PCAT
i	digestibility of protein	0.95
j	digestibility of carbohydrate	0.95
k	digestibility of fat	0.97

m	carbohydrate from protein catabolism	0.6g/g PCAT
n	heat correction for carbohydrate conversion to fat	3.14kJ/g CCON
p	proportion of protein in the feed	g/g
q	heat of combustion of protein	23.7kJ/g
r	heat of combustion of carbohydrate	16.5kJ/g
s	heat of combustion of fat	39.6kJ/g
t	efficiency of milk protein use for maintenance and body protein gain	0.85g/g
u	energy loss as urine	5.63kJ/g PCAT
v	ratio of protein to carbohydrate in milk	2.4
w	fat yield from carbohydrate conversion	0.3g/gCCON
x	non-carbohydrate energy yield from protein catabolism	8.17kJ/gPCAT

Correcting factors:

These corrections apply to the pairs of pathways in the flow-charts (figures 86 and 87) which carry the same numbers. The effect of the corrections is to create a switch between the two alternatives. The corrections for body protein (BPCOR) and body fat (BFCOR) alter the heat of changing body reserves, depending upon whether or not reserves are being gained, as follows:

$$BP = day14BP - day2BP$$

ie. BP is body protein gain

$$BPCOR = (((BP^2)^{0.5}) + BP)/2$$

BPCOR = 0 if BP is -ve

BPCOR = BP if BP is +ve

$$a = 29.9BPCOR/BP$$

a = 0 if BPCOR = 0

a = 29.9 if BPCOR = BP

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BF = day14BF - day2BF   ie. BF is body fat gain

BFCOR = $(((BF^2)^{0.5}) + BF)/2$ BFCOR = 0 if BF is -ve

                                         BFCOR = BF if BF is +ve

b = 4.4BFCOR/BF   b = 0 if BFCOR = 0

                                         b = 4.4 if BFCOR = BF

The correction for carbohydrate conversion to fat (CCON) prevents fat catabolism (FCAT) when carbohydrate is being converted to fat, and adjusts the heat production for the relative inefficiency of fat formation from carbohydrate, as follows:

rCCAT - (HEAT - xPCAT) = the energy available from carbohydrate minus the heat production which needs to be met. If this is +ve, carbohydrate is surplus to requirements for milk and energy, and this is converted to fat. If (rCCAT - (HEAT - xPCAT)) is -ve, then fat catabolism is necessary to meet the energy requirement for heat production.

CCON = $(((rCCAT+xPCAT-HEAT)^2)^{0.5})+rCCAT+xPCAT-HEAT)/2r$

If (rCCAT+xPCAT-HEAT) is -ve, CCON = 0

If (rCCAT+xPCAT-HEAT) is +ve, CCON = (rCCAT+xPCAT-HEAT)

Prediction of Milk Production from Litter Growth

The equations describing lactational performance are of two kinds; equations balancing inputs and outputs, and equations decribing the assumptions discussed above. For the litters, the equations are as follows:

Accounting for protein, carbohydrate, and fat as:

Supply from milk plus breakdown of the nutrients = growth + catabolism.

$$iPMK = BP + PCAT \tag{1}$$

$$jCMK + mPCAT = CCAT \tag{2}$$

$$kFMK + wCCON = BF + FCAT \tag{3}$$

Balancing milk energy intake with energy output:

$$iqPMK + jrCMK + ksFMK = qBP + sBF + uPCAT + HEAT \tag{4}$$

where $HEAT = aBP + bBF + hPCAT + EMA + nCCON \tag{5}$

From assumptions A1) A2), and A3):

$$CMK = PMK/v \tag{6}$$

$$PCAT = iPMK(1-t) + PMA \tag{7}$$

$$FCAT = ((HEAT - xPCAT) - rCCAT + CCON)/s \tag{8}$$

where CCON acts as a switch which causes FCAT to be equal to zero when there is conversion of carbohydrate to fat (defined on p. 91).

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Substituting (7) in (1):

iPMK = BP + PMK(i - it) + PMA

PMK(i - (i - it)) = BP + PMA

PMK = (BP + PMA)/ti

$$(9)$$

Substituting (8) in (3):

kFMK + wCCON = BF + ((HEAT - (rCCAT + xPCAT) + rCCON)/s)

ksFMK + swCCON = sBF + HEAT - rCCAT - xPCAT + rCCON

CCON(r - sw) = ksFMK + rCCAT + xPCAT - sBF - HEAT

$$CCON = \frac{ksFMK + rCCAT + xPCAT - sBF - HEAT}{r - sw}$$

$$(10)$$

Substituting (2) in (10):

$$CCON = \frac{ksFMK + r(iCMK + mPCAT) + xPCAT - sBF - HEAT}{r - sw}$$

$$= \frac{ksFMK + riCMK + PCAT(rm + x) - sBF - HEAT}{r - sw}$$

$$(11)$$

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Substituting (11) in (5):

$$HEAT = aBP + b B\bar{F} + hPCAT + EMA + n(ksFMK + rjCMK + PCAT(rm + x) - sBF - HEAT)$$

$$HEAT(r - sw) = (r - sw)(aBP + bBF + hPCAT + EMA) + n(ksFMK + rjCMK + PCAT(rm + x) - sBF - HEAT)$$

$$HEAT(r - sw + n) = (r - sw)(aBP + bBF + hPCAT + EMA) + nksFMK + nrjCMK + nPCAT(rm + x) - nsBF$$

$$HEAT = \frac{BPa(r-sw) + BF(b(r-sw)-ns) + PCAT(h(r-sw) + n(rm+x)) + EMA(r-sw) + nksFMK + nrjCMK}{r - sw + n} \quad (12)$$

Substituting (12) in (4):

$$iqPMK+jrCMK+ksFMK = qBP+sBF+uPCAT+BPa(r-sw)+BF(b(r-sw)-ns)+PCAT(h(r-sw)+n(rm+x))+EMA(r-sw)+nksFMK+nrjCMK$$
$$\overline{\qquad\qquad\qquad\qquad\qquad\qquad r - sw + n \qquad\qquad\qquad\qquad\qquad\qquad}$$

$$(iqPMK+jrCMK+ksFMK)(r-sw+n) = (qBP+sBF+uPCAT)(r-sw+n) + BPa(r-sw) + BF(b(r-sw)-ns)$$
$$+ PCAT(h(r-sw)+n(rm+x)) + EMA(r-sw) + nksFMK + nrjCMK$$

$$FMK(ks(r-sw)) = PCAT((r-sw)(h+u)+n(rm+x+u)) + BP((r-sw)(q+a)+qn) + BF((r-sw)(s+b))-PMKiq(r-sw+n) + CMKjr(sw-r) + EMA(r-sw) \quad (13)$$

Substituting (7) in (13):

$$FMK(ks(r-sw)) = (PMK(i-it)+PMA)((r-sw)(h+u)+n(rm+x+u)) + BP((r-sw)(q+a)+qn) + BF((r-sw)(s+b))-PMKiq(r-sw+n)$$
$$+ CMKjr(sw-r) + EMA(r-sw)$$

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$$FMK = \frac{PMK((i\text{-}it)((r\text{-}sw)(h+u)+n(rm+x+u))\text{-}iq(r\text{-}sw+n))+BP((r\text{-}sw)(q+a)+qn)+BF(r\text{-}sw)(s+b)+CMKir(sw\text{-}r)+EMA(r\text{-}sw)}{ks(r\text{-}sw)}$$

$$+ \frac{PMA((r\text{-}sw)(h+u)+n(rm+x+u))}{ks(r\text{-}sw)} \qquad (14)$$

Given litter protein gain (BP) and litter body fat gain (BF) and using the values reported subsequently for the constants, then milk production (PMK, CMK, and FMK) can be calculated as follows:

From (9):

PMK = (BP + PMA)/(0.85*0.95)

From (6):

CMK = PMK/2.4

From (14):

r - sw = 16.5 - 39.6*0.3 = 4.62

n(rm + x+ u) = 3.14(16.5*0.6 + 8.17 + 5.63) = 74.418

$$FMK = \frac{PMK((0.95-0.95*0.85)(4.62(4.9+5.63)+74.418)-0.95*23.7(4.62+3.14))+BP(4.62(29.9(BPCOR/BP)+23.7)+23.7*3.14)}{39.6*0.97(16.5-\mathbf{39.6*0.3})}$$

$$+ \frac{BF*4.62(39.6+4.4(BFCOR/BF))+CMK*0.95*16.5(-4.62)+PMA(4.62(4.9+5.63)+74.418)+4.62*EMA}{39.6*0.97(16.5-\mathbf{39.6*0.3})}$$

(15)

$$FMK = \frac{-157.179PMK+183.912BP+138.138BPCOR+182.952BF+20.328BFCOR-72.419CMK+123.067PMA+4.62EMA}{177.463}$$

$$FMK = -0.886PMK + 1.036BP + 0.778BPCOR + 1.031BF + 0.115BFCOR - 0.408CMK + 0.693PMA + 0.026EMA$$

Prediction of Lactational Performance from Maternal Data

Accounting for protein, carbohydrate, and fat as:

intake = output + catabolism.

$$piI = PMK + PCAT + BP \qquad (16)$$

$$cjI + mPCAT = CMK + CCAT \qquad (17)$$

$$fkI + wCCON = FMK + FCAT + BF \qquad (18)$$

Balancing energy intake with energy output:

$$I(piq + crj + fks) = qPMK + rCMK + sFMK + qBP + sBF + uPCAT + HEAT \qquad (19)$$

$$\text{where } HEAT = aBP + bBF + dPMK + eCMK + gFMK + hPCAT + EMA + nCCON \qquad (20)$$

Assumptions A1), A4), and A3) which have been described above, result in the following equations:

$$CMK = PMK/v \qquad (21)$$

$$HEAT = HCAP \qquad (22)$$

$$FCAT = ((HEAT - xPCAT) - rCCAT + rCCON)/s \qquad (23)$$

where CCON acts as a switch which causes FCAT to be equal to zero when there is conversion of carbohydrate to fat (defined on p. 91).

From (16):

$$PCAT = piI - BP - PMK \qquad (24)$$

From (17):

$$CCAT = cjI + mPCAT - CMK \qquad (25)$$

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Substituting (23) in (18):

$fkI + wCCON = FMK + BF + ((HEAT - (rCCAT + xPCAT) + rCCON)/s)$

$rCCON - swCCON = sfkI - sFMK - sBF - HEAT + rCCAT + xPCAT$ (26)

Substituting (25) in (26):

$CCON(r - sw) = sfkI - sBF - sFMK - HEAT + r(cjI + mPCAT - CMK) + xPCAT$

$$CCON = \frac{I(rcj + sfk) - sBF - sFMK - HEAT + PCAT(rm + x) - rCMK}{r - sw}$$ (27)

Substituting (27) in (20):

$HEAT = aBP + bBF + dPMK + eCMK + gFMK + hPCAT + EMA + \frac{n(I(rcj + sfk) - sBF - sFMK - HEAT + PCAT(rm + x) - rCMK)}{r - sw}$

$HEAT(r-sw) = (r-sw)(aBP+bBF+dPMK+eCMK+gFMK+EMA)+nI(rcj+sfk)-nsBF-nsFMK-nHEAT+PCAT(h(r-sw)+n(rm+x))-nrCMK$ (28)

Substituting (24) in (28):

$HEAT(r-sw+n) = (r-sw)(aBP+bBF+dPMK+eCMK+gFMK+EMA)+(piI-BP-PMK)(h(r-sw)+n(rm+x))$

$+ nI(rcj+sfk)-nsBF-nsFMK-nrCMK$

$$HEAT = \frac{BP((r-sw)(a-h)-n(rm+x)) + BF(b(r-sw)-ns) + PMK((r-sw)(d-h)-n(rm+x)) + CMK(e(r-sw)-nr' + FMK(g(r-sw)-ns)}{r-sw+n}$$

$$+ \frac{EMA(r-sw) + I(pi(h(r-sw) + n(rm+x)) + n(rcj+sfk))}{r-sw+n} \tag{29}$$

Given maternal body weight change (BP and BF), feed composition (p,f, and c), maternal intake (I), and milk production (PMK, CMK, and FMK) derived from litter growth and using the values reported subsequently for the constants, heat can be calculated from equation (29):

$r - sw = 16.5 - 39.6*0.3 = 4.62$

$rm + x = 16.5*0.6 + 8.17 = 18.07$

$$HEAT = \frac{BP(4.62(29.9(BPCOR/BP)-4.9)-3.14*18.07) + BF(4.4(BFCOR/BF)*4.62)-3.14*18.07) + PMK(4.62(16.7-4.9)-3.14*18.07)}{4.62+3.14}$$

$$+ \frac{CMK(0.88*4.62-3.14*16.5) + FMK(4.4*4.62-3.14*39.6) + 4.62EMA + I(p*0.95(4.9*4.62+3.14*18.07) + 3.14(c*16.5*0.95+f*39.6*0.97)}{4.62+3.14}$$

$$= \frac{-79.378BP + 138.138BPCOR - 124.344BF + 20.328BFCOR - 2.224PMK - 47.744CMK - 104.016FMK + 4.62EMA + I(75.409p + 49.220c + 120.614f)}{7.76}$$

$$= -10.229BP + 17.801BPCOR + 16.024BF + 2.620BFCOR - 0.287PMK - 6.153CMK - 13.404FMK + 0.595EMA + I(9.718p + 6.343c + 15.543f) \tag{30}$$

From (19):

$qPMK = I(piq + crj + fks) - qBP - sBF - rCMK - sFMK - uPCAT - HEAT \tag{31}$

Substituting (21) and (24) in (31):

qPMK = I(piq + crj + fks) - qBP - sBF - sFMK - (rPMK/v) - u(piI - BP - PMK) - HEAT

PMK(q + (r/v) - u) = I(pi(q - u) + crj + fks) - BP(q - u) -sBF - sFMK - HEAT

$$PMK = \frac{v(I(pi(q-u)+crj+fks)+BP(u-q)-sBF-sFMK-HEAT)}{qv+r-uv}$$ (32)

Substituting (21) in (29):

$$HEAT = \frac{I(pi(h(r-sw)+n(rm+x)+n(sfk+rcj))+BP((r-sw)(a-h)-n(rm+x))+BF(b(r-sw)-ns)+PMK((r-sw)(d-h)-n(rm+x)+((e(r-sw)-nr)/v))}{r-sw+n}$$

$$+ \frac{FMK(g(r-sw)-ns)+EMA(r-sw)}{r-sw+n}$$ (33)

HEAT(r-sw+n) = I(pi(h(r-sw)+n(rm+x)+n(sfk+rcj)) + BP((r-sw)(a-h)-n(rm+x)) + BF(b(r-sw)-ns) + PMK(1/v)((r-sw)(dv-hv+e)-nv(rm+x)-nr)

+ FMK(g(r-sw)-ns) + EMA(r-sw)

Substituting (32) in (33):

HEAT(r-sw+n) = I(pi(h(r-sw)+n(rm+x)+n(sfk+rcj)) + BP((r-sw)(a-h)-n(rm+x)) + BF(b(r-sw)-ns)

$$+ \frac{v(I(pi(q-u)+crj+fks)+BP(u-q)-sBF-sFMK-HEAT)(1/v)((r-sw)(dv-hv+e)-nv(rm+x)-nr)}{qv+r-uv}$$

+ FMK(g(r-sw)-ns) + EMA(r-sw)

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Abbreviating:

$(qv+r-uv)$ to z

$((r-sw)(dv-hv+e)-nv(rm+x)-nr)$ to y

$$HEAT(r-sw+n) = I(pi(h(r-sw)+n(rm+x)+n(sfk+rcj))+BP((r-sw)(a-h)-n(rm+x))+BF(b(r-sw)-ns)$$
$$+ \frac{v(I(pi(q-u)+crj+fks)+BP(u-q)-sBF-sFMK-HEAT)(y/v)+FMK(g(r-sw)-ns)+EMA(r-sw)}{z}$$

$$HEAT(r-sw+n) = I(pi(h(r-sw)+n(rm+x)+n(sfk+rcj))+BP((r-sw)(a-h)-n(rm+x))+BF(b(r-sw)-ns)$$
$$+ I(y/z)(pi(q-u)+crj+fks)+BP(y/z)(u-q)-(sy/z)BF-(sy/z)FMK-(y/z)HEAT)$$
$$+ FMK(g(r-sw)-ns)+EMA(r-sw)$$

$$FMK(g(r-sw)-ns-(sy/z)) = HEAT(r-sw+n+(y/z))-BP((r-sw)(a-h)-n(rm+x)-(y/z)(q-u))-EMA(r-sw)$$
$$- BF(b(r-sw)-ns-(sy/z)-I(pi(h(r-sw)+n(rm+x)+(y/z)(q-u))+crj(n+(y/z))+fks(n+(y/z))$$

$$FMK = \frac{HEAT(r-sw+n+(y/z))-BP((r-sw)(a-h)-n(rm+x)-(y/z)(q-u))-EMA(r-sw)-BF(b(r-sw)-ns-(sy/z)}{g(r-sw)-ns-(sy/z)}$$
$$- \frac{I(pi(h(r-sw)+n(rm+x)+(y/z)(q-u))+(crj+fks)(n+(y/z))}{g(r-sw)-ns-(sy/z)} \qquad (34)$$

Given maternal body weight changes (BP and BF), feed composition (p,c, and f), maternal intake (I), and assuming that heat production is maximal ($=$HCAP), milk production (PMK, CMK, and FMK) can be calculated as follows:

Derivation of Equation 1 from Known Variables

The protein intake of the chicks (denoted PI) per unit proportional protein density per day was (from 21):

When a protein density is maintained, feed was held to be scaled according to a metabolic energy requirement function per bird (from Emmans and Fisher, 1978). Using a metaprotein density function for an average current protein requirement from measured protein values obtained on days 2 and 14 to estimate the approximate amount required to maintain liveweight was:

$$n_r = 42*10^4(0.6+8.17)+e(23.7-5.63+8BW)/(2*10^{18}) \qquad (i)$$

The amino acid requirement is proportional to a mean metabolic order of liveweight in the form of liveweight loss: $BW^{0.75}$ an as completely efficient liveweight gain, with liveweight gain of liveweight standing in same proportion as the absolute metabolic order of liveness (Hepburn et al., 1996). When expressed conventionally as a measure in rats of different ages it has been found to be scaled similarly across poultry over a large range of liveweight/age (Hepburn, 1996). Denoting expression (i) and the protein requirement above scaled by protein requirement function to maintenance per 12 hours was calculated as:

r - sw = 16.5 - 39.6*0.3 = 4.62

n(rm + x) = 3.14(16.5*0.6 + 8.17) = 56.740

$$\frac{y}{z} = \frac{(r-sw)(dv-hv+e)-nv(rm+x)-nr}{qv+r-uv} = \frac{4.62(16.7*2.4-4.9*2.4+0.88)-2.4(16.7*2.4+0.88)-2.4*56.740-3.14*16.5}{23.7*2.4 + 16.5-5.63*2.4} = \frac{-53.082}{59.868} = -0.887$$

From (34) and (22):

$$FMK = \frac{HCAP(4.62+3.14-0.887)-BP(4.62(29.9(BPCOR/BP)-4.9)-56.740+0.887(23.7-5.63))-BF(4.4(BFCOR/BF)*4.62-3.14*39.6*0.887)}{4.4*4.62-3.14*39.6+39.6*0.887}$$

$$- \frac{4.62EMA-I(p*0.95(4.9*4.62+56.740-0.887(23.7-5.63))+(c*0.95*16.5+f*0.97*39.6)(3.14-0.887)}{4.4*4.62-3.14*39.6+39.6*0.887}$$

$$FMK = \frac{6.873HCAP+63.350BP-138.138BPCOR+89.219BF-20.328BFCOR-4.62EMA-I(60.182p+35.316c+86.542f)}{-68.891}$$

FMK = -0.100HCAP-0.920BP+3.489BPCOR-1.295BF+0.514BFCOR+0.117EMA+I(0.874p+0.513c+1.256f)

From (32) and (22):

$$PMK = \frac{2.4(I(0.95p(23.7-5.63)+0.95*16.5*c+0.97*39.6*f)-BP(23.7-5.63)-39.6BF-39.6FMK-HCAP)}{23.7*2.4+16.5-5.63*2.4}$$

$$= \frac{I(41.200p + 37.62c + 92.189f) - 43.368BP - 95.04FMK - 2.4HCAP}{59.868}$$

= I(0.688p + 0.628c + 1.540f) - 0.724BP - 1.587BF - 1.587FMK - 0.040HCAP

From (21):   CMK = PMK/2.4

Derivation of Constants and Known Variables

PMA: **The protein required for maintenance** is 10g per unit proportional protein maturity per day (Emmans, and Oldham, 1988).

Where possible, functions related to body size have been scaled according to protein mass as a proportion of mature protein mass (Emmans and Fischer, 1986). Using a mature protein mass of 0.07kg and an average current protein mass derived from measures of protein mass (BP) in grams on days 2 and 14 of lactation, the following expression for scaling to metabolic liveweight was used:

$$(0.07^{-0.27})*(day2BP + day14BP)/(2*1000) \qquad (i)$$

The above scalar was used in preference to the more usual scalar of (liveweight in kg)$^{0.75}$. This was because liveweight$^{0.75}$ did not completely eliminate differences in body size due to age as opposed to weight, resulting in underestimation of the metabolic liveweight of litters (Kleiber et al, 1956). Whilst expression (i) has not been tested in rats of different ages, it has been found to be satisfactory for growing poultry over a large range of liveweight:age (Emmans, 1990). Combining expression (i) and the protein requirement given above, the protein requirement for maintenance per 12 days was calculated as:

$$PMA = 12*10*(0.07^{-0.27})*(day2BP + day14BP)/(2*1000)$$

EMA: **The energy requirement for maintenance** was scaled in the same way as described for protein maintenance using 1630 kJ/unit protein mass (Emmans and Oldham, 1988). Given this the energy requirement for maintenance per 12 days was calculated as:

$$EMA = 12*1630*(0.07^{-0.27})*(day2BP + day14BP)/(2*1000)$$

HCAP: The maximum maternal heat which the dam can dispose of, is derived from data on rats in hot environments (Kirmiz, 1962). Because of the physiological mechanisms by which animals can regulate their heat loss, it was necessary to use data from rats who would have been trying to lose rather than to conserve heat. The work by Kirmiz was the only source of such data found.

An attempt to calculate capacity to lose heat from theoretical considerations of radiation, conduction, convection, and evaporation (Blaxter, 1989; Monteith and Unsworth, 1990) was made. However, this approach required too many assumptions and approximations, rendering it worthless.

Kirmiz (1962) measured total and evaporative heat losses in a sealed calorimeter, by calculation from the amounts of oxygen consumed, CO_2 excreted, and liveweight loss (rats which produced faeces or urine during the measurement period were excluded from the results). Between 0°C and 25°C the total heat loss and evaporative heat loss are described by the following regressions (figure 105):

Total heat = 1143.0 - 30.4(temp. °C) $s = 42.5$; R^2adj. = 97.6%
Evaporative heat = 104.5 - 2.4(temp. °C) $s = 3.7$; R^2adj. = 97.2%

By subtraction, non-evaporative heat loss is described by:

Non-evap. heat = 1038.5 - 28(temp. °C)

Thus, at 37.1°C non-evaporative heat loss has declined to zero, and this is in good agreement with the measured body temperature of these rats (37.5°C). As the environmental temperature increases from 30°C to 35°C there was a linear increase in evaporative heat loss. Above 35°C there was a large increase in both the evaporative heat loss and in body temperature. The large increase in body temperature indicates, that these rats were storing heat and that they

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Fig 105: The relationship between room temperature and heat loss in mature rats (Kirmiz, 1962)
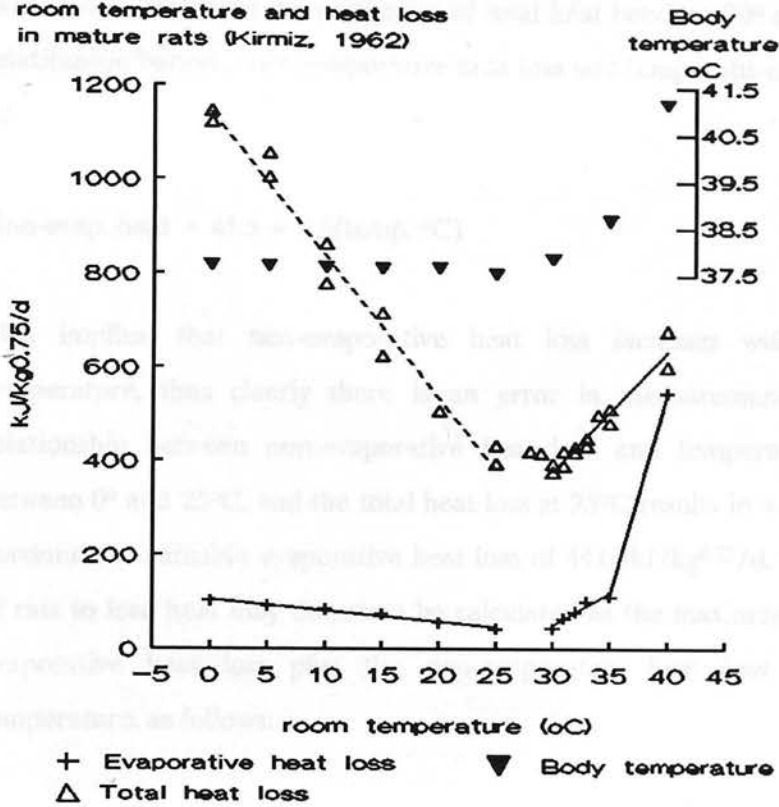
were not able to tolerate temperatures above 35°C for longer periods such as 12 days. From the heat loss data between 30°C and 35°C the following regressions were derived (figure 105):

Total heat = -318.9 + 23.4(temp. °C) $s = 15.6$; R^2adj. $= 87.9\%$

Evap. heat = -364.4 + 13.8(temp. °C) $s = 8.0$; R^2 adj. $= 91.6\%$

Assuming that the maximum sustainable evaporative heat loss occurs at 35°C, then from the regression of evaporative heat loss this is $118.6 kJ/kg^{0.75}/d$. However, this implies a non-evaporative heat loss of $381.5 kJ/kg^{0.75}/d$, which is 6.52 times the value predicted from the relationship between non-evaporative heat loss and temperature given above. Either the relationship does not hold at temperatures in excess of 25°C, or there is an error in the measurement of evaporative heat loss above 25°C. By subtraction of the regression for

evaporative heat from the regression of total heat between 30° and 35°C, the relationship between non-evaporative heat loss and temperature in this range is:

Non-evap. heat = 45.5 + 4.6(temp. °C)

This implies, that non-evaporative heat loss *increases* with increasing temperature, thus clearly there is an error in measurement. Using the relationship between non-evaporative heat loss and temperature derived between 0° and 25°C, and the total heat loss at 35°C results in a value for the maximum sustainable evaporative heat loss of $441.6kJ/kg^{0.75}/d$. The capacity of rats to lose heat may therefore be calculated as the maximum sustainable evaporative heat loss plus the non-evaporative heat loss at a given temperature, as follows:

HCAP = 441.6 + (1038.5 - 28(temp. °C))

$= (1480.1 - 28(temp. °C)) kJ/kg^{0.75}/d$

Brody and co-workers (1938) measured the heat production of lactating rats at a temperature of 28°C to be $1.1 kcal/g^{0.73}/d$. The two relationships are compared in figure 107. The agreement between these two relationships suggests, that the expression calculating capacity to lose heat is adequate.

These data did not allow the expression to be based on proportional protein maturity. However, since it is only applied to the dams, the drawback of scaling to liveweight$^{0.75}$ is of little consequence.

a: **The heat of body protein gain** is derived from energy balance data on growing rats using dietary casein as a protein supply (29.9kJ/gBP; Pullar and Webster, 1977). In the absence of evidence to the contrary from calorimetric studies, the heat of body protein mobilisation was taken to be zero (biochemical suggestions that this may not be so, have been disregarded).

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Fig. 107: A comparison at 28°C between the heat
capacity equation (HCAP) and an equation for the heat
production of lactating rats (Brody et al, 1938).
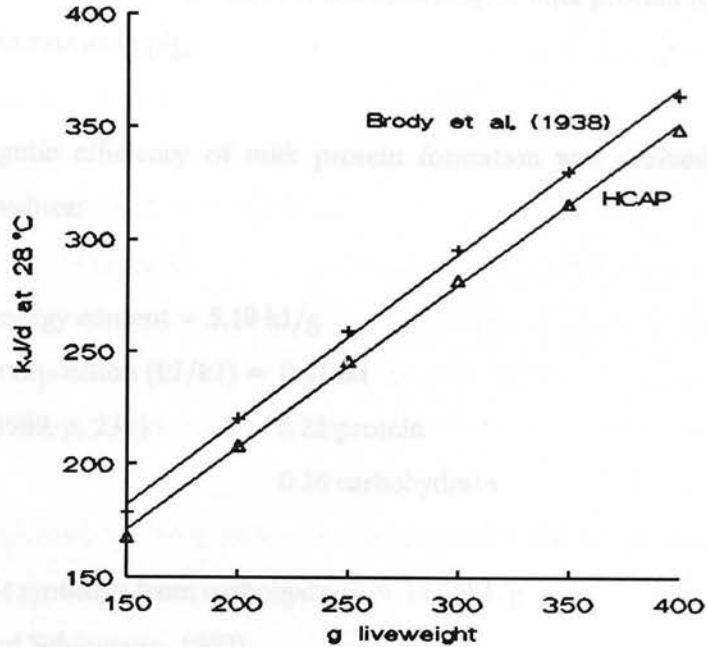
HCAP = (1480.1 − 28×28) $kJ/kg^{0.75}/d$
Brody et al. (1938): heat = 4.60 $kJ/g^{0.73}/d$

b: **The heat of body fat formation** is derived from the work of Chudy and Schiemann (1969), who measured the efficiency of rat body lipid gain from dietary fat as 0.9. Therefore 0.1*39.6 kJ of heat are produced per 0.9g of body fat synthesised.

b = 3.96/0.9 = 4.4kJ/g body fat formed.

d: No direct measurement of **the energy requirement for production of milk protein** has been made. Theoretical estimates of the energetic efficiency of milk protein formation (0.82; Baldwin, 1968) are markedly different from measures of the energetic efficiency of body protein formation (0.44, Pullar and Webster, 1977). The difference between these values is probably related to the extent of protein turnover incorporated into the estimate. An alternative approach to estimating the energetic efficiency of milk protein formation is to account for the energy cost of synthesising milk lactose and milk fat and attribute the remaining energy expenditure to the cost of synthesising milk protein. This was done using data from pigs, because no

acceptable measure of the energetic efficiency of milk production in rats was found. It is therefore assumed, that the efficiency of milk protein formation is the same in rats as in pigs.

The energetic efficiency of milk protein formation was derived from the following values:

Pig milk energy content = 5.19 kJ/g

Pig milk composition (kJ/kJ) = 0.61 fat

(Blaxter, 1989, p. 235)   0.22 protein

                               0.16 carbohydrate

Heat of fat synthesis from carbohydrate = 14.65kJ/g

(Chudy and Schiemann, 1969)

Heat of lactose synthesis = 0.88 kJ/g

(Baldwin, 1968)

Using an energetic efficiency, measured in pigs, for milk synthesis from feed of 0.72 (Noblet and Etienne, 1987), the heat of milk protein synthesis was calculated as follows:

Energy to make 1g of milk = 5.19/0.72 = 7.208 kJ

Heat production = 7.208 - 5.19 = 2.018 kJ

Heat of milk fat = 14.65(5.19*0.61/39.6) = 1.171 kJ

Heat of milk lactose = 0.88(5.19*0.16/16.5) = 0.044 kJ

Remaining heat = 2.018 - (1.171 + 0.044) = 0.803 kJ

Milk protein content = (5.19*0.22)/23.7 = 0.0482 g/g

Heat of milk protein synthesis (d) = 0.803/0.0482 kJ/g

d = 16.7 kJ/g milk protein.

e: **The heat of milk carbohydrate formation** was derived from the following stoichiometry (Baldwin, 1968):

2 liver glucose + 4 ATP ----> 1 lactose + 4 ADP

(2 ATP for transport from liver to mammary gland)

energy yield from ATP = 75.3 kJ/mol

mw lactose = 342 g/mol

e = 4 * 75.3/342

approx. = 0.9 kJ/g milk carbohydrate

g: **The heat of milk fat formation** was assumed to be the same as the heat of body fat formation (**b**); 4.4kJ/g milk fat formed.

h: **The heat of protein catabolism**, i.e. the energy cost of synthesising and excreting urinary N, was taken to be 4.9kJ/g protein catabolised (Whittemore and Fawcett. 1976).

i: **The digestibility of milk protein and of feed casein** was derived from the measured casein digestibilities of feeds ranging in casein content from 100 to 700 g/kg (Radcliffe and Webster, 1976, 1978). **i = 0.95**

j: **The digestibility of carbohydrate from feed and milk** was taken to be 0.95. In the work of Radcliffe and Webster (1976,1978) the digestibility of energy was 0.95. Given a casein digestibility of 0.95 and a constant low fat content of 50g/kg the digestibility of the remaining energy, that is carbohydrate, must be very close to 0.95.

k: **The digestibility of feed fat**, in this case groundnut oil, was taken to be the same as the digestibility of corn oil; 0.97 (Chudy and Schiemann, 1969). **The digestibility of milk fat**, in the absence of other data, was assumed to be the same as the digestibility of feed fat.

m: **The carbohydrate yield from protein catabolism** was calculated from the stoichiometry of individual amino acid catabolism to urea and glucose (Schulz, 1978) and the amino acid composition of casein (Kuzdzal-Savoie et al, 1980).

m = 0.6 g carbohydrate per g protein catabolised

n: This constant adjusts for **the extra heat produced by fat synthesis from carbohydrate as opposed to fat synthesis from fat.** The heats of body fat and milk fat synthesis (**b** and **g**) are calculated as if dietary fat is the substrate for body fat and milk fat synthesis; 4.4 kJ per g fat synthesised (Chudy and Schiemann, 1969). Using an energetic efficiency of 0.73 for fat synthesis from carbohydrate (Chudy and Schiemann, 1969), the heat of fat synthesis from carbohydrate is 14.65 kJ per g fat synthesised. Hence the extra heat of fat synthesis from carbohydrate is (14.65 - 4.4)kJ per g fat synthesised. The feed carbohydrate necessary to make 1 gram of fat is:

$((39.6/0.73)/16.5) = 3.29$ g CHO

n $= (14.65 - 4.4)/3.29 = 3.12$ kJ/g CHO

t: **The efficiency with which milk protein is used by the litters for body protein gain** was taken to be 0.85; the biological value (measured in rats) of milk protein (McDonald et al, 1981). The efficiency of milk protein use for maintenance was assumed to be the same.

Using the measured amino acid requirements for maintenance in pigs (Fuller et al, 1989) and the amino acid composition of rat milk (Luckey et al, 1955), the first limiting amino acid in rat milk, when used for maintenance, was found to be threonine. The ratio between the threonine contents of "ideal" protein for maintenance and rat milk, was 0.79. Given that this calculation was based on maintenance amino acid requirements for pigs, and given the considerable variation in reported maintenance amino acid requirements for rats and pigs (Fuller et al, 1989) the above assumption was justified.

u: **The energy loss in urine,** from protein catabolism was taken to be 5.63 kJ/g protein catabolised (Emmans, 1990)

v: **The ratio of protein to carbohydrate in rat milk** was assumed to be 2.4 (Luckey et al, 1955), this has been described in more detail in assumption A1).

w: **The yield of fat per gram of carbohydrate converted to fat** was calculated, using an energetic efficiency of conversion of 0.73 (Chudy and Schiemann, 1969) as:

$$w = (16.5*0.73)/39.6 = 0.30$$

Stoichiometric calculations, assuming that the fat produced is tripalmitylglycerol, result in a value of 0.36 (Schulz, 1978).

x: **The non-carbohydrate energy yield from protein catabolism** is calculated as the heat of combustion of protein minus the energy recovered as carbohydrate and lost as urine:

$$x = 23.7 - 5.63 - 0.6*16.5 = 8.17 \text{ kJ/gPCAT}$$

Model results

The average values from the treatments used in experiment R2 were used as inputs to the model (table 112a). The model was then used to calculate milk production from litter growth data. Predicted milk productions from litter growth data are shown in table 112b. The ratio of milk protein to milk fat is high when compared to the range of published values (Jenness and Sloan, 1970), but is in agreement with more recent measures of milk composition (Fischbeck and Rasmussen, 1987; Grigor et al, 1987).

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Table 112a: Data inputs for the model; average values from feeds in the second rat experiment (R2).

Feed	Protein (g/g feed)	Fat (g/g feed)	Carbohydrate (g/g feed)	Food intake (kg)	Temperature (°)C
1	0.1487	0.0854	0.7659	347.4	24.6
2	0.2994	0.0980	0.6026	442.8	24.6
3	0.1480	0.2277	0.6243	289.8	24.6
4	0.2956	0.2190	0.4854	395.1	24.6
5	0.1452	0.3363	0.5185	35.1	24.6
6	0.3019	0.3739	0.3242	363.6	24.6
7	0.1476	0.5161	0.3363	57.5	24.6
8	0.2949	0.5500	0.1551	307.8	24.6

Feed	MBP (g/12d)	MBF (g/12d)	LBP (g/12d)	LBF (g/12d)	MLW (kg)	d14MBP (g)	d14LBP (g)
1	-5.9	-33.2	30.4	21.5	0.3428	60.2	41.4
2	0.8	-32.2	52.8	31.9	0.3586	68.4	64.8
3	-7.4	-28.5	26.4	18.1	0.3489	61.3	38.5
4	4.2	-33.7	51.8	48.3	0.3717	72.0	63.0
5	-18.3	-31.8	9.1	1.5	0.3048	48.8	21.1
6	-0.5	-24.8	48.1	53.8	0.3826	69.5	60.1
7	-13.4	-27.5	10.4	4.6	0.3289	54.8	21.8
8	-0.5	-21.5	46.1	48.8	0.3775	67.8	57.4

MBP is maternal body protein gain; MBF is maternal body fat gain; LBP is litter body protein gain; LBF is litter body fat gain; MLW is average maternal liveweight; d14MBP is day 14 maternal body protein; d14LBP is day 14 litter body protein.

Table 112b: Predicted milk productions, heat productions and heat capacities from model using litter growth data from the second rat experiment (R2).

Feed	Milk protein (g/12d)	Milk carbohydrate (g/12d)	Milk fat (g/12d)	HCAP (kJ/12d)	HEAT (kJ/12d)
1	45.6	19.0	63.4	4251.9	3770.3
2	77.1	32.1	97.5	4398.1	4273.0
3	40.4	16.8	56.7	4308.5	3798.7
4	75.5	31.4	114.6	4518.0	4184.1
5	16.3	6.8	21.1	3893.2	2098.8
6	70.5	29.4	118.1	4617.0	4209.6
7	17.9	7.5	25.3	4121.9	2321.9
8	67.6	28.1	109.9	4570.8	4124.0

Assuming a milk carbohydrate content of 37g/kg (Luckey et al, 1955), the calculated milk yields of the dams on the high protein feeds are in agreement with the values measured by Fischbeck and Rasmussen (1987) who used the tritiated water technique (Rath and Thenen, 1979) corrected for measured milk composition. Within the range of milk yields reported, these values, together with the estimates of Brody and Nisbet (1938), are high. However these two sources represent the most reliable estimates of rat milk production although, as discussed by Linzell (1972), there is a degree of uncertainty associated with all the reported values.

Maternal heat production was calculated from the litter derived milk production, maternal intake and body mobilisation. The capacity of the dams to lose heat was calculated from maternal liveweight and environmental temperature (table 112b). The heat production on all the feeds was lower than the calculated heat loss capacity (figure 114). However, for those feeds which supported a successful lactation (1,2,3,4,6,8) the ratio between the heat production and the independantly calculated heat loss capacity was constant as follows (The regression constant was not significant):

HEAT = 0.914HCAP   s = 143.5

Given the assumptions and simplifications used, particularly in the derivation of the equation for the calculation of the heat loss capacity, this discrepancy is acceptable. The importance of these results is that they show the rats which lactated successfully to be producing heat at a rate close to our best estimates of the maximum possible rate of heat production. Those rats which failed to lactate (feeds 5,7), had heat productions well below their capacity for heat disposal.

Given the agreement between heat production and heat capacity for most feeds, the assumption that the heat capacity was equal to the heat production could be used in the model. This permitted prediction of the maternal milk production when heat production was maximal, without using litter growth data. Initially these results were calculated using the value of heat production derived from the litter growth estimated

Fig. 114: Predicted heat production in relation to predicted capacity for heat disposal.

milk production ... that this was a back calculation to ... the slopes of the equations. Clearly the equations to predict milk production from maternal data alone are not satisfactory.

The algebra in the equations is correct, and the logic from which the equations were derived is sound. ... one of coefficients in the final equations for prediction of milk production (eqns. (34),(32), and (21)) is high, and there is an error of measurement associated with each coefficient. In some cases the data from which coefficients were estimated are poor and simplifying assumptions were made. In this model it is easily appreciated that the cumulative error in the equations is large relative to the values of the controlling heat capacity. One effect of using heat capacity as the controlling factor in the model is to make the partition of protein between catabolism and milk sensitive to relatively small changes in the heat capacity, particularly for milk protein formation ... protein accretion. The sensitivity of the model to the partition in protein supply ... heat of milk protein synthesis (constant d, ordinarily 16.7) from 20.7 to 12.7 as shown in table 114. These values of d are well within the acceptable range of values arising from theoretical and measured estimates for the work of protein synthesis (see definition of constant d).

It is clear from these results that the model cannot predict milk production from maternal data alone. Thus the main objective of the model, namely to test the ...

Table 114: Predicted milk productions (g/12d) from maternal data only, using different values for the heat of milk protein synthesis (**d**). The value of **d** ordinarily used in the model was 16.7 kJ/g milk protein synthesised. Data were from the second rat experiment (R2).

if	Fat $d=12.7$	Protein $d=12.7$	Fat $d=16.7$	Protein $d=16.7$	Fat $d=20.7$	Protein $d=20.7$
Feed						
1	51.3	77.4	186.6	-141.6	238.7	-228.6
2	118.2	25.0	242.8	-180.0	289.2	-261.1
3	40.2	83.4	182.0	-145.4	236.9	-236.4
4	149.6	-13.1	273.1	-216.3	319.3	-296.8
5	58.2	-77.9	148.6	-223.0	184.1	-280.9
6	140.8	13.6	266.0	-191.9	313.1	-273.4
7	59.2	-68.1	159.0	-228.1	198.1	-291.9
8	124.1	32.1	251.6	-176.7	299.8	-259.6

milk production as the capacity for heat loss (table 114; d = 16.7). In effect this was a back calculation to check the algebra of the equations. Clearly, the equations to predict milk production from maternal data alone are not satisfactory.

The algebra in the equations is correct, and the logic from which the equations were derived is sound. The number of coefficients in the final equations for prediction of milk production (eqns. (34),(32), and (21)) is high, and there is an error of measurement associated with each coefficient. In some cases the data from which coefficients were estimated are poor and simplifying assumptions were made. In this model it would appear that the cumulative error in the equations is large relative to the effect of the controlling heat capacity. One effect of using heat capacity as the controlling factor in the model is to make the partition of protein between catabolism and milk sensitive to relatively small changes in the heat coefficients, particularly for milk protein formation and protein catabolism. The sensitivity of the model to the partition in protein supply was tested by varying the value for the heat of milk protein synthesis (constant d, ordinarily 16.7) from 20.7 to 12.7 as shown in table 114. These values of d are well within the acceptable range of values arising from theoretical and measured estimates for the work of protein synthesis (see definition of constant d).

It is clear from these results that the model cannot predict milk production from maternal data alone. Thus the main objective of the model, namely to test the hypothesis that heat capacity was constraining milk production, cannot be realised beyond the initial comparison.

Model Modification

There are insufficient data available at the present time to provide coefficients of a precision high enough to allow modifications to the prediction of rat milk production from maternal data. Information of particular value would be a better understanding of the relationship between the components within the milk; this would allow a simplification of the model and the discarding of one assumption (A1). However, to see if the litter growth based predictions of heat production were valid between

experiments, the calculation of the dams capacity to lose heat was scaled by the discrepancy between the heat capacity and the heat production discussed above (figure 114):

$$HCAP = 0.914(1480.1 - 28(temp. \,^{\circ}C)) \; kJ/kg^{0.75}/d$$

This modification was tested against data from the fourth rat experiment.

The Fourth Rat Experiment: A Comparison between Feeds Differing in Protein:Carbohydrate or in Protein:Energy Ratio on Lactational Performance

Introduction

The fourth rat experiment (R4) was in chronological order the second experiment to be done. It did not fit conveniently into the development of a hypothesis for the effects of feed composition on lactational performance. The results from this experiment (R4) were not used in the development of the model and have therefore been used as a test of the modification to the heat capacity equation.

The experiment was designed to investigate the effect of protein:carbohydrate ratio and protein:energy ratio on lactational performance, using two feeds per ratio (figure 117). In addition, a comparison between feeds of equal energy content (feeds 1,5 vs 2,6) and equal protein content (feeds 1,2 vs 5,6) was included in the design (figure 117).

Method

A total of 35 lactating dams were allocated to one of six feeding treatments and an initial cull group. The composition of the feeds is given in table 117. The difference between the measured and calculated composition of particularly the high fat feeds was due to the sampling difficulties associated with high fat mixtures. The measured

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Fig. 117: Feeding treatments in the fourth rat experiment. Horizontal dotted lines are between feeds of equal protein content; vertical dotted lines are between feeds of equal energy content.
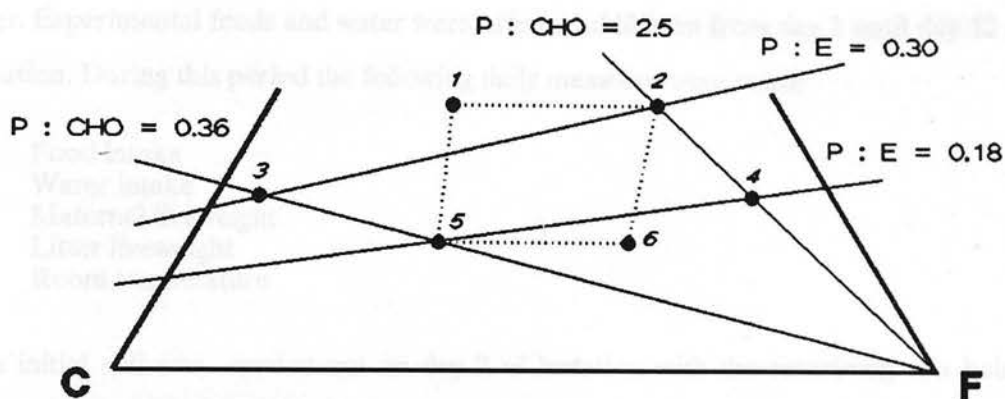
Table 117: The composition of the feeds used in the fourth rat experiment (measured values in % DM, calculated values in % OM, unless otherwise stated.)

Feed no.	DM (g/kg fresh)	CP	EE	CHO	GE (MJ/kg OM)
Measured composition*					
1	339.9	36.22	22.22	41.56	25.74
2	998.3	34.78	40.41	24.81	26.83
3	483.0	22.90	3.18	73.92	18.45
4	998.0	24.81	59.26	15.93	30.16
5	997.7	19.08	18.73	62.19	21.75
6	798.1	19.85	33.56	46.59	27.06
Calculated composition					
1		38.00	23.32	38.69	24.69
2		38.00	47.00	15.00	30.16
3		25.00	5.00	70.00	19.49
4		25.00	65.00	10.00	33.35
5		18.52	29.62	51.86	24.69
6		18.52	53.28	28.20	30.16

* DM is oven dry matter at 60°C; CP is crude protein = N x 6.407 (see p. 32); EE is ether extract; CHO is carbohydrate calculated as (1000-CP-EE); GE is gross energy. Mineral content is 100 g/kg DM.

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composition does not agree with the recorded weights of ingredients in the feeds, the feed compositions calculated from the ingredient proportions were in good agreement with the calculated composition. The feeds were therefore assumed to have their intended compositions.Littersize was standardized on day 1 of lactation to 12 pups per litter. Experimental feeds and water were offered ad libitum from day 2 until day 12 of lactation. During this period the following daily measures were made:

    Food intake
    Water intake
    Maternal liveweight
    Litter liveweight
    Room temperature

The initial cull was carried out on day 2 of lactation with the remaining rats being culled on day 12 of lactation for carcass analysis. Carcasses were analysed for water, nitrogen, ether extract, ash, and gross energy. Further details of the method are described in chapter 3. The full data are in appendix 5.

Results

The treatment averages are given in table 118 and the F-values arising from these comparisons are given in table 119.

Table 118: The treatment averages for the lactational performance of the rats in the fourth rat experiment (gDM/10d except energy values kJ/10d).

Feed	1	2	3	4	5	6	sed.
Intake	289	126	381	96	332	127	45
Maternal gains of[*]:							
liveweight	-18.8	-80.0	4.4	-76.8	-1.6	-60.6	15.1
Protein	-0.6	-0.5	4.6	-11.5	1.3	11.9	6.0
Fat	-24.9	-44.6	-20.0	-31.4	-33.2	-31.5	7.1
Litter gains of:							
liveweight	244	105	236	60	265	95	25
Protein	59.3	30.0	53.2	17.1	58.9	20.7	12.3
Fat	50.4	25.9	37.4	13.5	79.2	22.3	12.52

* Maternal gains are covariate adjusted by initial maternal liveweight

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In order to evaluate the effects on lactational performance of energy content, protein content, protein:energy ratio, and protein:carbohydrate ratio in the feed the analysis of variance (given in appendix 1) made the following comparisons between feeds:

	Feeds
Energy contents at equal protein content	1, 5 vs 2, 6
Protein contents at equal energy content	1, 2 vs 5, 6
P:CHO at equal P:energy	2, 4 vs 3, 5
P:energy at equal P:CHO	2, 3 vs 4, 5

and the following interactions between:

Protein and energy contents	1, 6 vs 2, 5
P:CHO and P:energy	2, 5 vs 3, 4

In this experiment feeds 2 and 5 are of central importance, since they form part of every comparison made. The performance resulting from feed 2 is clearly atypical when compared to similar feeds in experiments R1 and R2 (figures 120 and 121); this was

Table 119: The F-values arising from the analysis of variance of the fourth rat experiment.

Comparison	1,5 vs 2,6	1,2 vs 5,6	1,6 vs 2,5	2,3 vs 4,5	2,4 vs 3,5	2,5 vs 3,4	covariate d2Mlwt*
Intake	37.64	0.57	0.50	0.78	4.25	0.07	
Maternal gains of:							
Liveweight	35.72	3.25	0.01	0.06	61.25	0.20	9.24
Protein	2.76	1.42	2.92	2.45	4.95	1.00	2.25
Fat	3.69	0.26	5.24	0.02	0.00	6.33	0.02
Litter gains of:							
Liveweight	87.69	0.12	0.94	0.05	95.18	4.73	
Protein	17.03	0.36	0.30	0.03	14.42	1.23	
Fat	23.85	2.27	3.77	4.49	18.77	10.03	

*: d2Mlwt is maternal liveweight on day 2.

Fig. 120: The effect of feed composition on litter liveweight gain (g/12d). The original litter gains from R1 and R4 have been adjusted to 12 day gains.
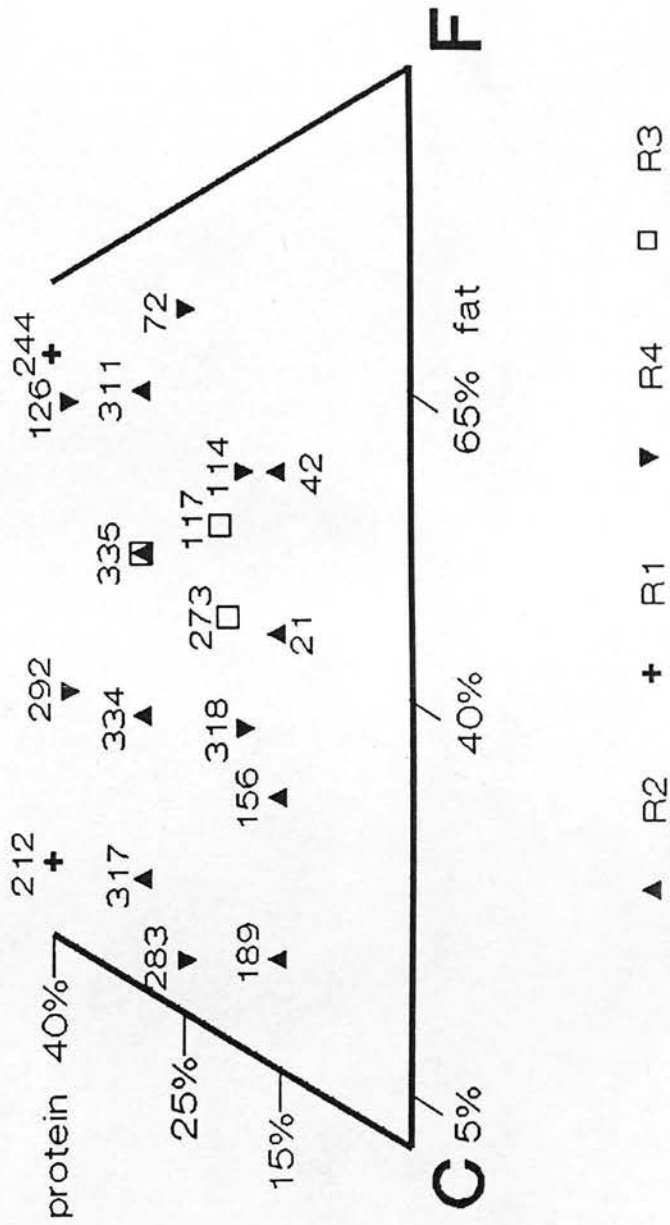
Fig. 121: The effect of feed composition on the food intake of lactating rats (gOM/12d). The original intakes from R1 and R4 have been adjusted to 12 day intakes.

use discussion... some similarity... line from here. Given that the feed line resulted in similar... maternal performance, no useful comparison can be made within the experiment.

The feed intake data were used to calculate milk production. Using these values and approximate..., heat production was calculated (figure 122). The calculated heat production... heat production... the relationship between heat production and capacity to lose heat is modified.

Discussion

The Basis of Performance on feed 2

The feed level performance resulting from feed 2 in this experiment (fed) in terms of litter growth and feed intake was 13g/12d and 145gOM/12d respectively (figures 120...

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Fig. 122: Predicted heat production in relation to modified capacity for heat disposal.
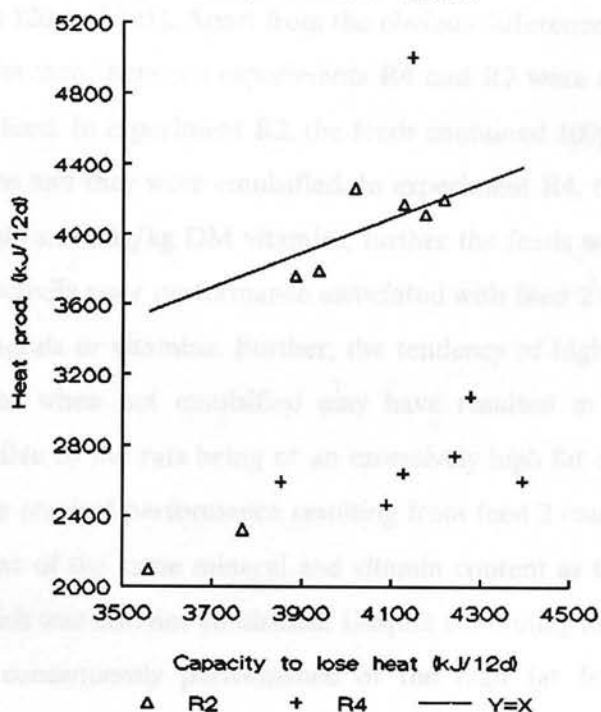
not due to any one individual within the group. Given that this feed has resulted in abnormal lactational performance, no meaningful comparisons can be made within the experiment.

The litter growth data were used to calculate milk production. Using these values and maternal data, heat production was calculated (figure 122). The calculated heat productions do not conform to the relationship between heat production and capacity to lose heat as modified.

Discussion

The Atypical Performance on Feed 2

The lactational performance resulting from feed 2 in this experiment (R4) in terms of litter growth and food intake was 126g/12d and 145gOM/12d respectively (figures 120

and 121). This was markedly less than the performance on similar feeds in experiments R1 and R2 (figures 120 and 121). Apart from the obvious differences in animals and in time, the main differences between experiments R4 and R2 were associated with the formulation of the feed. In experiment R2, the feeds contained 100g/kg DM minerals, 40g/kg DM vitamins and they were emulsified. In experiment R4, the feeds contained 50g/kg DM minerals and 20g/kg DM vitamins, further the feeds were not emulsified. As such the unexpectedly poor performance associated with feed 2 could be related to a deficiency of minerals or vitamins. Further, the tendency of high fat feeds, such as feed 2, to sediment when not emulsified may have resulted in the layer of feed immediately accessible to the rats being of an excessively high fat content. The above explanations for the atypical performance resulting from feed 2 cannot be discounted. However, feed 2 was of the same mineral and vitamin content as the high fat feed in experiment R1, which was also not emulsified. Despite containing more fat than feed 2 (R4), intake and consequently performance of the high fat feed from the first experiment was more than twice that of feed 2 (R4). Since all the feeds in this experiment were made using the same batch of oil (including an anti-oxidant) and were stored in the same manner, any possible rancidity would have affected all the feeds and not just feed 2. No satisfactory explanation for the unexpectedly poor performance on feed 2 (R4) was found.

The Test of the Heat Prediction Equation

The modified heat productions and the heat capacities calculated using the treatment averages are plotted in figure 122. Values from both the current experiment (R4) and the second experiment (R2) are shown. The relationship between the heat capacity and heat production for values from the current experiment is poor. Further, there appears to be a discrepancy between values derived from the two experiments. The modification made to the heat capacity equation (p. 115) does not improve the relationship between heat capacity and heat production. Indeed, the derivation of both these equations is brought into question. It is more likely, given the assumptions made, that the derivation of the heat capacity equation is in error. However, no better data are available at present than those already used (Kirmiz, 1962) with which to re-evaluate this equation.

Fig. 124: Characterisation of the feed composition triangle in terms of lactational success or failure (up to 40% protein). The values from which the threshold of lactational adequacy was derived are shown as triangles (interpolated values) or squares (extrapolated values). The dots are feeds from all rat experiments.
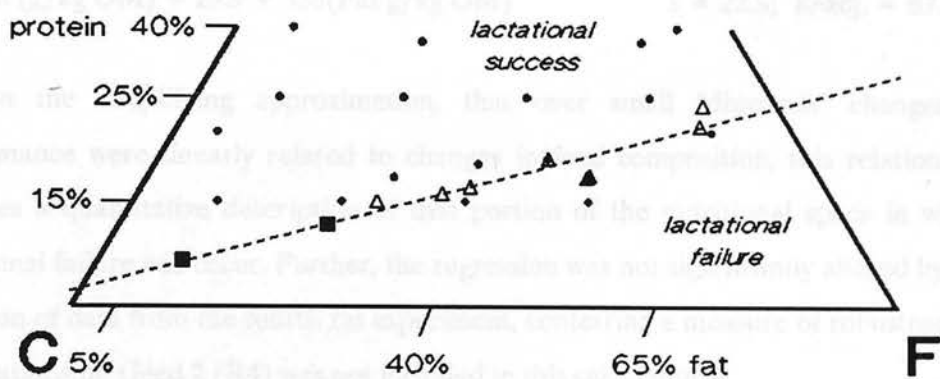
Characterisation of the Feed Composition Triangle

The four rat experiments were designed to collectively allow characterisation of an area of the feed composition triangle in terms of lactational performance. The feed compositions used in this experiment are particularly important to the description of the lower boundary in feed composition for lactational adequacy (figures 120 and 121). Using the data from all four experiments this threshold was characterised as follows.

The average level of performance attained on feed II in the third experiment (117g litter growth per 12 days) was defined as the "threshold performance". Some of the dams on this feed achieved an adequate lactation whilst other suffered lactational failure (p. 77). Clearly this feed was on, or very close, to a threshold in feed composition for lactational adequacy. Given the threshold level of performance, other threshold feed compositions were calculated by interpolation between pairs of feeds one of which supported adequate lactation and the other which did not (figure 124). Two additional points were derived by extrapolation from the relationship between

litter growth and protein content, to provide data for the low protein/high carbohydrate area of the triangle (figure 124). The following relationship between feed protein content and feed fat content describing the threshold in feed composition was derived by regression:

Protein (g/kg OM) = 29.9 + 358(Fat g/kg OM)   s = 22.8; R^2adj. = 87.6%

Despite the simplifying approximation, that over small "distances" changes in performance were linearly related to changes in feed composition, this relationship provides a quantitative description of that portion of the nutritional space in which lactational failure will occur. Further, the regression was not significantly altered by the omission of data from the fourth rat experiment, conferring a measure of robustness to the relationship (feed 2 (R4) was not included in this calculation).

Summary

This model was constructed to test the hypothesis that intake of food and consequently lactational performance was being constrained by the dams capacity to dissipate heat. The predictions of milk production from litter growth data were within the range of published values. Using litter predicted milk production, the calculated heat production resulting from those feeds which supported adequate lactation was of a similar magnitude to the predicted capacity for disposal of heat. However the relationship between heat production and heat capacity was poor, with differences between experiments. The model failed to predict milk production from maternal data only, precluding any exploration of the relationship between heat production and food intake.

The results from the model did not discredit the hypothesis that lactational performance was constrained by heat capacity, neither do they substantiate it. This was not evident at the outset. To properly test this hypothesis, these experiments would need to be repeated at different environmental temperatures. This would allow dams in low temperatures the opportunity to dispose of more heat, eat more, and consequently have an improved lactational performance.
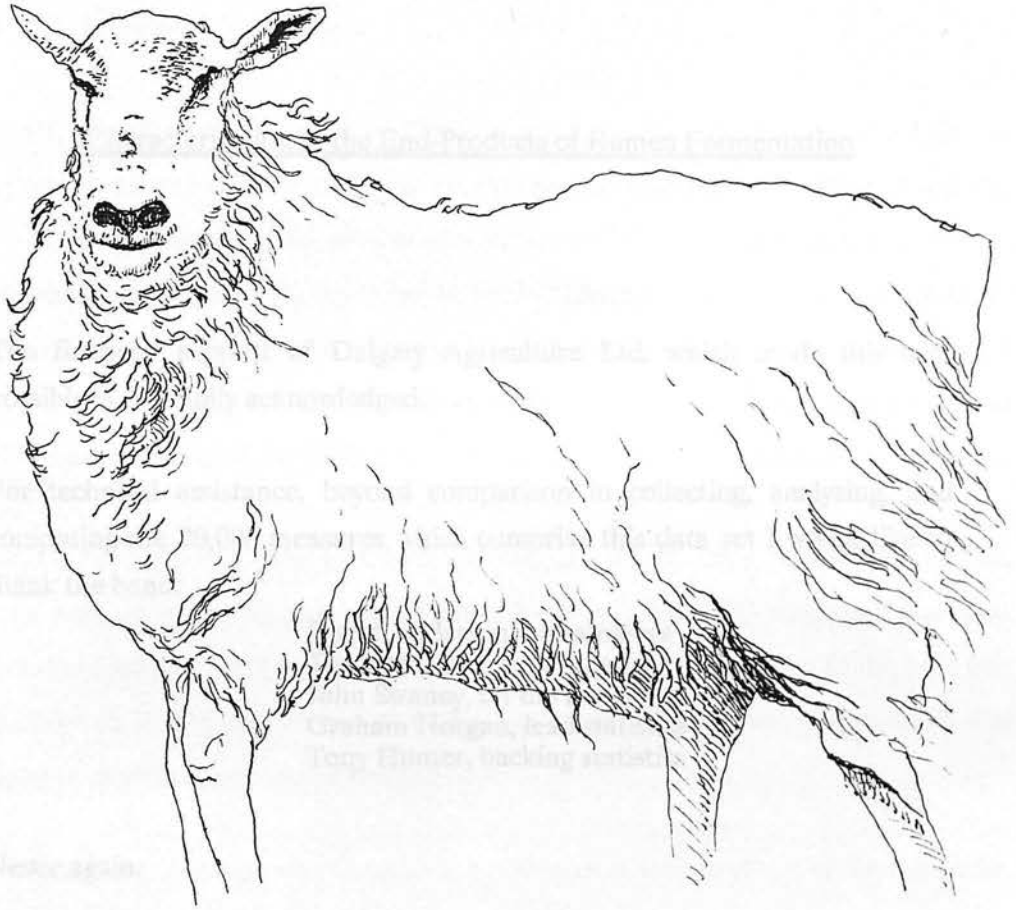
Estimation of ... the End Products of Rumen Fermentation

This ... work ... of Dalgety Agriculture Ltd, whose work and full publication ... is fully acknowledged.

Note: again.

Note: All tables and figures are numbered according to the page on which they are found. As such table and figure numbers are not consecutive (see list of tables and figures, p. ix).

CHAPTER 8

Characterisation of the End-Products of Rumen Fermentation

The financial support of Dalgety Agriculture Ltd, which made this trial possible is gratefully acknowledged.

For technical assistance, beyond comparison in collecting, analysing, and computing the 20,000 measures which comprise this data set I would like to thank the band:

David Anderson, on samplers
Terry McHale, on hard graft
John Swaney, on the integrator
Graham Horgan, lead statistics
Tony Hunter, backing statistics

Never again.

Note: All tables and figures are numbered according to the page on which they are found. As such table and figure numbers are not consecutive (see list of tables and figures, p. ix).

Introduction

The effect of feed composition on the lactational performance of rats has been described in chapters 2 to 7. Lactating rats are sensitive to changes in the composition of their feed and are not as able to compensate for adverse changes in feed composition as are non-lactating rats (Musten et al, 1974; Peterson and Baumgardt, 1971). In particular, feeds of low protein, low carbohydrate content caused lactational failure in these rats. This work showed the important influence of feed composition on the lactational performance of rats.

Feeds, with a nutrient balance markedly different from the required nutrient balance, necessitate increases in metabolism to redress the balance and to dispose of surplus nutrients. During lactation, the level of production and the associated metabolic rate is high. Consequently the difference between the lactational level of performance and the animals maximum possible level of performance is small, relative to the non-lactating state. The capacity of lactating animals to respond to adverse feed compositions is therefore reduced (Kronfeld, 1976).

The above rationale was used to explain the effects of feed composition observed on lactating rats, in terms of heat production. Brody et al. (1938) measured the heat production of lactating rats to be twice that of non-lactating rats, similar to the heat loss by rats kept in desert conditions (42°C; Kirmiz, 1962). The following hypothesis was proposed to explain the observed results:

Food intake and consequently lactational performance was constrained by the dams capacity to dispose of heat.

Brody and Nisbet (1938) reported (with a characteristic combination of detailed results and sketchy methods) a comparison between lactating rats and cows. They found that in terms of energetic efficiency lactating rats and cows were similar. It might therefore be expected that the above hypothesis, if applicable to rats, is relevant to lactating cows. Indeed, ruminants have an additional source of heat to dispose of, the heat of rumen fermentation. Leng (1989) has recently proposed that cattle under tropical conditions

on poor quality forages are constrained by their ability to lose heat. He found that supplementation of poor quality forages with protein resulted in large increases in intake. The effect of the supplemental protein was to redress the balance between the protein and energy yields from rumen fermentation. This would have reduced the heat production necessary to accommodate the surplus energy of the original feed permitting an increased intake. Both for rats and for cattle there is therefore circumstantial evidence in support of this hypothesis. This chapter describes an experiment designed to allow the development of this study, from lactating rats to dairy cows.

There is an extensive literature on dairy cows which indicates that there are important effects of different feed types on the milk production (see Thomas and Martin, 1988; Sutton and Morant, 1989). However, there is a major difficulty in working with ruminants, namely how to describe the feed in terms which can be directly related to lactational performance (figure 129). In rats, feed composition can be related directly to lactational performance, but in ruminants this cannot be done because of the intermediate conversion in the rumen. This additional step, of rumen fermentation, in the conversion of food into milk may result in end-products of a substantially different composition from that of the feed. Investigation of the effects of changing feed composition in ruminants is further complicated because feeds completely composed of purified ingredients cannot be used, without concern for the physiological relevance of so doing. Thus simple substitutions of one feed component for another are precluded.

The fermentation process converts the feed into seven major absorbable products (figure 130), and numerous minor products. Ignoring the minor products, it is possible in theory to consider the effects of changing the feed composition of a seven component "feed" on lactational performance. A graphical representation of such a feed (analogous to the two dimensional triangle for a three component feed) is a *six* dimensional polyhedron. By this representation the complexity of such a system becomes apparent. Clearly, it is desirable to group some of the fermentation products so as to reduce the number of feed "components" if this is possible without unacceptable loss of precision.

Fig. 129: A representation of the conversion of food into milk by ruminants; showing the importance of rumen fermentation.
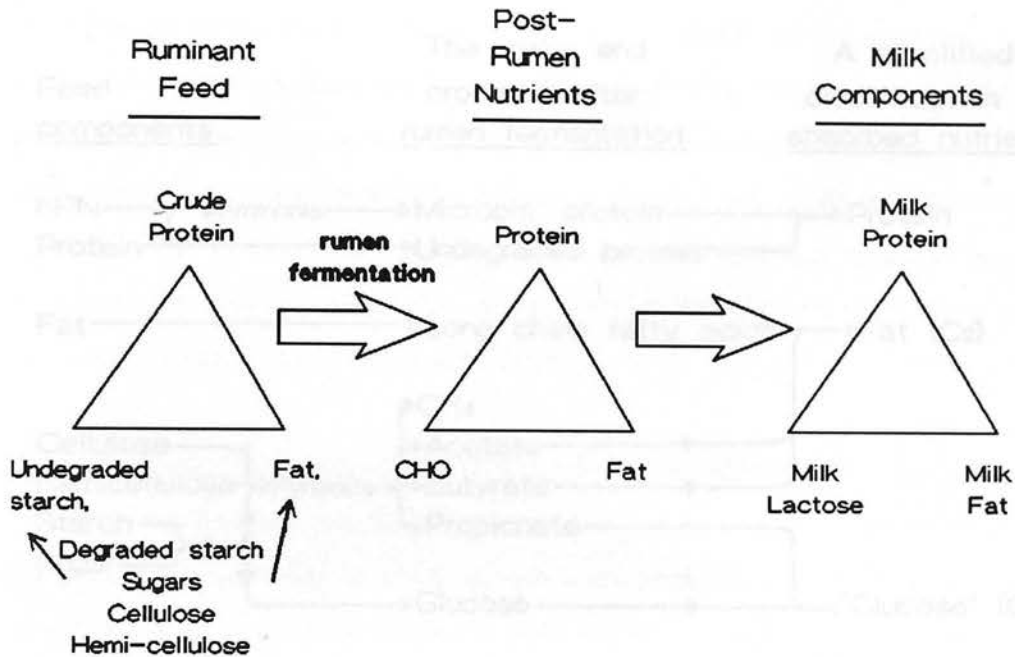
Combining rumen degradable protein (converted to microbial protein) with undegraded protein in one protein category may be acceptable, provided that the protein quality of the feed is not being investigated and the quality does not vary greatly. In the ruminant, carbohydrates and fats are metabolised via two general pathways according to the number of carbon atoms in the molecule. Molecules whose number of carbon atoms is greater than 10 (i.e. long chain fatty acids) or whose number of carbon atoms is divisible by 2 but not by 3 (mainly acetate and butyrate) are metabolised as fats. Molecules whose number of carbon atoms is less than 10 and is divisible by 3 (mainly propionate) are metabolised as glucose. Molecules not conforming to either of these classes can be split into C_3 and C_2 units (e.g. a C_5 molecule such as valerate would yield a C_2 and a C_3 unit) (see Annison and Armstrong, 1970; Lindsay, 1970; Van Soest, 1982). Thus the end-products of fermentation can be classified in a manner analogous to the feed classification used with non-ruminants (figure 130). Whether the effects of changing ruminant feed composition can be adequately described by three components remains to be investigated.

Figure 130: A simplified classification of feeds, fermentation products, and absorbed nutrients.

Feed components	The major end products after rumen fermentation	A simplified classification of absorbed nutrients

NPN ——————→ Ammonia ————→ Microbial protein ——————→ Protein
Protein ——————↗ Undegraded protein

Fat ——————————————————→ Long chain fatty acids ——→ Fat (C_2)

Cellulose ——↘
Hemicellulose ——→ Pyruvate ——→ CH_4
Starch ——↗ Acetate ————→
Sugar ——↗ Butyrate ————→
Propionate ————→
Glucose ——————→ "Glucose" (C_3)
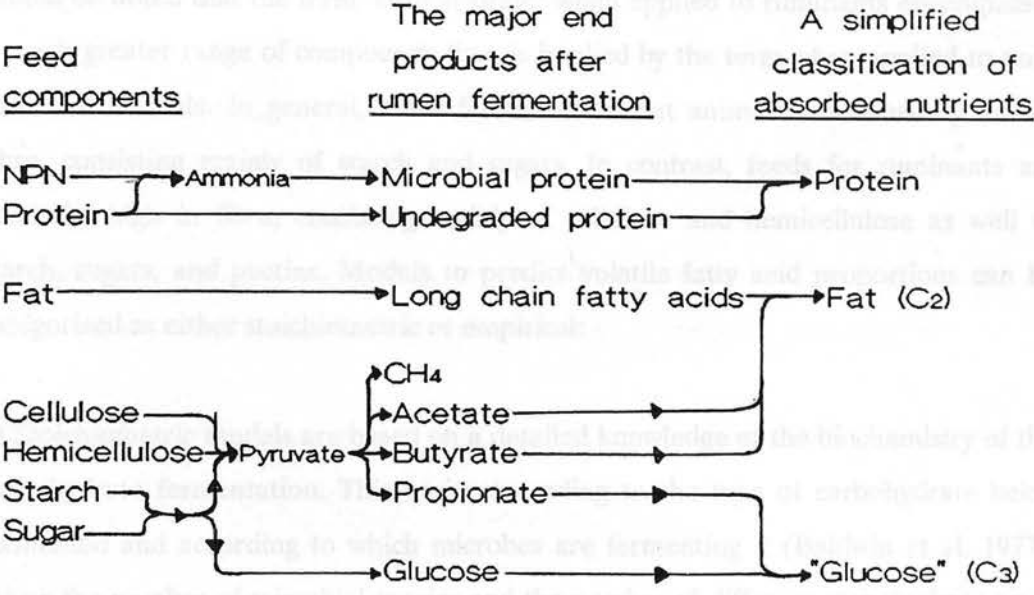
Even if this classification can be used, it still requires a knowledge of the relationship between feed composition and fermentation end-product proportions. Measurement of volatile fatty acid production requires the use of sophisticated techniques carried out on rumen fistulated animals (Sutton, 1985). These techniques are not suitable for routine use nor are they beyond reproach (Sutton, 1985). An adequate method for predicting how the feed is apportioned into the end-products of rumen fermentation is therefore required. The experiment described in this chapter attempted to address this problem.

Approaches to Prediction of Fermentation End-Products

A number of different approaches have been taken to enable prediction of the end-products of rumen fermentation from the chemistry of the feed. This subject in itself constitutes a huge body of literature which has been reviewed elsewhere (Hungate, 1966; Baldwin and Bywater, 1984; Hobson, 1988). The fermentation of the

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carbohydrate fraction of the feed is particularly difficult to predict precisely. There is a common intermediate (pyruvate) in the rumen degradation of all the feed carbohydrates to their end-products, the volatile fatty acids, CO_2, and methane. It should be noted that the term "carbohydrate" when applied to ruminants encompasses a much greater range of compounds than is implied by the term when applied to non-ruminant animals. In general, feeds for non-ruminant animals are relatively low in fibre, consisting mainly of starch and sugars. In contrast, feeds for ruminants are generally high in fibre, consisting mainly of cellulose and hemicellulose as well as starch, sugars, and pectins. Models to predict volatile fatty acid proportions can be categorised as either stoichiometric or empirical:

1) Stoichiometric models are based on a detailed knowledge of the biochemistry of the carbohydrate fermentation. This varies according to the type of carbohydrate being fermented and according to which microbes are fermenting it (Baldwin et al, 1977). Given the number of microbial species and the number of different carbohydrate types which may be present in the rumen, these models are complex. Due to their fundamental basis such models, if they are comprehensive, are not limited by the data from which they were derived. At present there is insufficient information to make them complete, necessitating some degree of reliance on simplifying empirical equations (Murphy et al, 1982; Baldwin and Argyle, 1988).

2) Empirical models are based on regression of volatile fatty acid proportions on the chemistry of the feed which gave rise to them. These models are simple, however they are limited to the range of feeds from which they were derived. There have been few systematic attempts to relate rumen volatile fatty acid proportions to controlled inputs of specific feed ingredients (Sutton, 1968, 1969). A large range of feeds has not been evaluated within one trial. It has therefore been necessary to pool data from different experiments, and the between experiment variation in results is not small (Murphy et al, 1982). Thus a relatively large degree of imprecision is incorporated into such models.

At present, neither type of model provides a wholly satisfactory method of predicting

volatile fatty acid proportions. In order to link the work done here on rats with that using cows it was important to link fermentation end-products to feed chemistry. Whilst recognising the limitations of regression based predictions, it was decided to adopt this approach to prediction of volatile fatty acid proportions.

In order to construct an empirical model, an experiment was designed with the following objectives:

- To provide a wide ranging data set in order to derive an empirical model for predicting the volatile fatty acid proportions arising from a given feed chemistry.

- To ensure that the data were not biased by changes in the rumen environment other than the changes directly attributable to the chemistry of the feed.

Experimental Design

The major influence on rumen fermentation which is not necessarily related to feed composition is the physical form of the feed. It has been shown, that the grinding of forages which reduces particle size, i.e. modifying physical form without altering chemical composition, has a marked effect on volatile fatty acid proportions (Woods and Luther, 1962; Jorgensen and Schultz, 1963). In order to measure the volatile fatty acid proportions arising from different test feeds, with the minimum alteration in the physical form of the feed, it was decided to use one common basal forage. A uniform grass silage was chosen, to which addition of test feeds would be made.

Possible modifications to the rumen environment caused by the addition of different test feeds to the silage which were not attributable to the chemistry of the feed, were classified as follows:

1) Changes in the overall level of feeding and consequently in the rate of passage through the rumen.

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2) Changes in the ratio of test feed to silage, altering the relative contributions of each feed to the physical form of the total ration.

3) Enhancement in the fermentation of the silage caused by the test feed alleviating a possible constraint in the nutritional supply to the rumen microbes.

In order to create a wide ranging data set, this experiment used sixteen test feeds. To test for the above effects, a minimum of three treatments would be required per effect, assuming that each of these effects had a linear relationship to the amount of test feed added. This would require 144 (3x3x16) treatments. By using co-incident treatments as shown in figure 134, only six treatments per test feed are needed to achieve the same result. By choosing the lowest level of test feed inclusion to be zero (figure 134), three of the six treatments were silage only treatments and therefore did not need replicating for each test feed. Thus only three treatments per test feed were required, provided that enough silage only treatments were included in the design to allow evaluation of between sheep and period variation. Silage only treatments were assigned at random to the design (table 135), subject to:

1) All three silage only treatments being represented in each period of the trial.

2) Each sheep receiving all three silage only treatments during the course of the trial.

Test feed treatments were allocated to sheep in blocks such that each sheep replicate received the three test feed treatments in different combinations (tables 135 and 136a). Test feeds have not been named but are referred to as numbers in table 136a. The reason for this is discussed below.

Availability of Data

This trial was sponsored by a commercial company. The data arising from this trial are subject to a confidentiality agreement but will subsequently become available after a period not exceeding 3 years from the publication date of the thesis. The results will be published as scientific papers.

Fig. 134: The treatments for each test feed expressed as test feed/silage in g DM. Treatment numbers are boxed.

[3]
$\frac{O}{800}$

[2] ↗ ↓ [4]
$\frac{O}{600}$ $\frac{200}{600}$

[1] ↗ [6] ↓ [5]
$\frac{O}{400}$ ---→ $\frac{200}{400}$ ---→ $\frac{400}{400}$

----→ Increasing total feed intake at constant test feed : silage ratio

——→ Increasing the ratio of test feed to silage at constant total feed intake.

-----→ Supplementing constant silage with test feed, increasing both total feed intake and test feed : silage ratio.
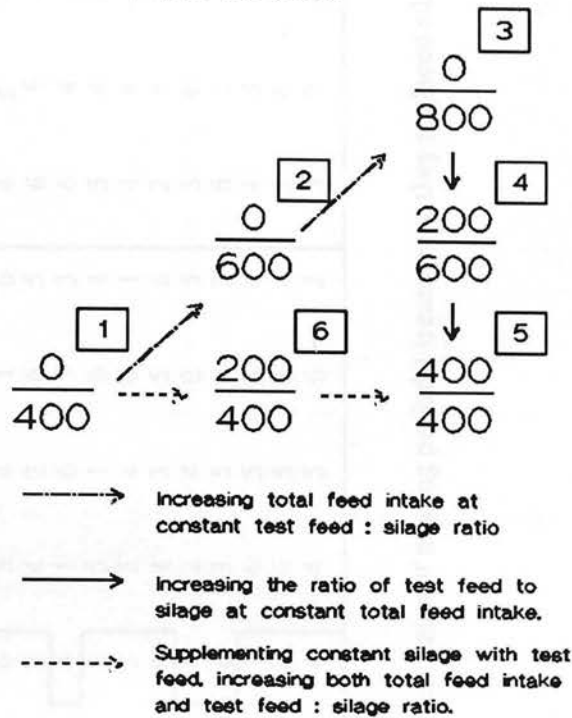
Methods

The trial consisted of 17 two week periods using eleven rumen fistulated sheep. The sheep were housed in individual pens (mesh floored; 1.5m x 1.5m). The test feed was mixed with the silage and offered as one daily feed, at 9am, water was freely available. Any feed refusals were collected daily and bulked by period for subsequent chemical analysis.

Sixteen test feeds and the basal silage were evaluated. The range in chemical composition of the feeds is given in table 136b. The feeds ranged from straw to molasses to rape seed meal and were chosen to have a high proportion of one of the following:

Rapidly fermented carbohydrate
Cellulose
Hemicellulose
Protein

Table 135: Allocation of feeding treatments and test feeds to sheep and periods

Block	1					2				3				4			
Period	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
Sheep																	
1	-	3	6	1	4	4	2	5	6	5	4	1	6	4	5*	6	5
2	-	5	3	4	1	6	4	5	3	4	2	6	5	6	5	4	6
3	2	6	5	4	5	4	1	6	4	1	6	5	3	6	4	5	2
4	6	4	5	3	4	3	5	6	2	5	1	6	4	5	4	1	6
5	5	2	6	4	2	6	3	4	5	5	4	6	1	4	6	5	3
6	-	6	1	6	5	2	6	5	4	4	3	5	6	2	5	6	4
7	1	4	5	6	3	5	6	4	6	2	4	2	5	3	6	5	4
8	5	1	2	6	4	1	4	5	6	3	5	4	2	6	1	4	5
9	4	5	5	2	6	5	6	6	1	6	5	3	4	4	5	4	6
10	-	6	4	5	1	4	5	5	3	6	5	2	4	5	6	4	1
11	-	5	4	6	3	6	5	1	4	4	5	6	2	5	2	6	4

Treatment numbers are as in figure 134.
* bold typed treatments were using block 1 feeds not block 4, i.e these are replicates of missing period 1 treatments, they replaced silage only treatments.

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Table 136a: Allocation of test feeds (1-16) to blocks.

Block	1	2	3	4
Sheep				
1	5	9	3	14
2	5	9	3	16
3	8	12	2	7
4	8	1	16	9
5	15	1	7	11
6	15	1	7	11
7	4	13	14	6
8	4	12	14	6
9	12	13	16	10
10	5	8	13	2
11	15	4	2	6

Table 136b: The range in composition covered by the test feeds collectively and the composition of the silage (g/kg DM).

Component	test feed min.	test feed max.	silage
Crude protein (CP)	28	540	119
Acidified ether extract (AEE)	0	129	43
Ash	0	111	70
Acid detergent lignin (ADL)	0	103	23
Acid detergent fibre (ADF)	0	565	360
Neutral detergent fibre (NDF)	0	712	570
Starch	0	759	4
Sugar	0	630	39
NDF insoluble protein	0	177	20

The composition of test feeds and silage was measured in samples taken every week. The silage was all of one cut, taken from the same field, and ensiled in one pit. At the start of the trial, all the required silage was cut into blocks and stored frozen until it was to be fed. Consequently silage composition (table 136b) was uniform throughout the trial.

To verify that two weeks was a sufficient time for the rumen to adapt to changes in feeding, the first period was of three weeks. There was no evidence that volatile fatty acid proportions altered in the third week of that period; this is in agreement with Sutton and Johnson (1969). Subsequent periods were of two weeks length.

During the last two days of each period, samples of rumen fluid were taken from an in situ sampler (figure 137) at 2, 4, 6, 8, 11, 14, 18, and 23 hours post feeding. Immediately after sampling, the sample was filtered through double muslin and pH was measured. Further microbial action was arrested by addition of saturated mercuric chloride solution to the sample (1 ml per 50 ml sample), and samples were frozen for analysis of volatile fatty acids at a later date. Samples were deproteinized with metaphosphoric acid (25% in 5N H_2SO_4) and volatile fatty acid concentrations were determined by gas-liquid chromatography (Pye Unicam series 304 chromatograph). The resulting curves were integrated using a Jones JCL 6000 integrator.

The trial had originally been designed to use 12 sheep, however one sheep was removed from the trial in the first period necessitating an adjustment to the design. As a consequence of this the design was statistically unbalanced and required a specialised analysis of variance. The data were analysed using partial regression techniques and restricted maximum likelihood analysis. This accounted for variation due to sheep, periods, and treatments (table 138). The treatment means derived from this analysis (adjusted for sheep and period effects) were used to derive regressions of volatile fatty acid proportions on feed chemistry.

Figure 137: The in situ sampler, and apparatus used to take rumen fluid samples.

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During the last two days of each period, samples of rumen fluid were taken from an in situ sampler (figure 137) at 2, 4, 6, 8, 11, 14, 18, and 23 hours post feeding. Immediately after sampling, the sample was filtered through double muslin and pH was measured. Further microbial action was arrested by addition of saturated mercuric chloride solution to the sample (1 ml per 50 ml sample), and samples were frozen for analysis of volatile fatty acids at a later date. Samples were deproteinized with metaphosphoric acid (25% in 5N H_2SO_4) and volatile fatty acid concentrations were determined by gas-liquid chromatography (Pye Unicam series 304 chromatograph). The resulting curves were integrated using a Jones JCL 6000 integrator.

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Figure 137: The in situ sampler, and apparatus used to take rumen fluid samples.

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Results

Changes in Level of Feeding

Analysis of the three silage only treatments (1, 2, 3; figure 134) indicated, that there was no significant effect of level of feeding on the volatile fatty acid proportions.

Because the effect of level of feeding was investigated only with the silage, any interaction with the test feed type was not directly measured. However, if there were any interaction, it would have become apparent when comparing measured with predicted volatile fatty acid proportions for treatment number 6 (figure 134). This test for interactions between feed type and level of feeding is described in the results section entitled "Enhancement of Silage Fermentation".

Changes in the Test Feed:Silage Ratio

Results from the three treatments which differed only in test feed:silage ratio (3, 4, 5; figure 134) showed a significant effect ($p < 0.01$) of test feed:silage ratio on volatile fatty acid proportions, within test feed type. The slope of the relationship between volatile fatty acid proportion and test feed:silage ratio was found by linear regression, for each feed. There was a highly significant effect ($p < 0.001$) of test feed type on the slope of the relationship between test feed:silage ratio and volatile fatty acid proportions. The interaction between test feed type and test feed:silage ratio is discussed in the results section entitled "Regression of Feed Chemistry on Volatile Fatty Acid Proportions".

Table 138: Percentage of the total variation accounted for by the restricted maximum likelihood analysis for the major volatile fatty acids.

	Acetate	Propionate	Butyrate
Sheep	19.6	15.2	0.6
Periods	13.2	12.5	4.1
Treatments	46.7	29.6	72.9

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Enhancement of Silage Fermentation

The results from treatments 6, 5, and 1 (figure 134) are a measure of the volatile fatty acid proportions arising from a fixed increment in test feed amount to a constant silage ration. This represents the cumulative effect of changing the level of feeding, the test feed:silage ratio, and includes any enhancement of silage fermentation. There was no effect of feeding level on volatile fatty acid proportions when measured with the silage only. Assuming that the same applies when any test feed is present in the total meal, and that there was no enhancement of silage fermentation, the volatile fatty acid proportions measured for treatment 6 should be predictable from the relationship between test feed:silage ratio and volatile fatty acid proportions (derived from treatments 3, 4, and 5). Predicted volatile fatty acid proportions plotted against measured values for feed 6 are presented in figures 140a, 140b, and 141. There was no significant difference between measured volatile fatty acid proportions and those predicted from the test feed:silage ratio. This shows that there was no enhancement of silage fermentation other than that which can be explained by the test feed:silage ratio. The improbable situation in which a possible enhancement of silage fermentation is cancelled out by the concomitant increase in level of feeding is precluded since the two effects would be additive. Further, there were no test feed types which deviated significantly from the relationship between predicted and measured values. This indicates that there was no interaction between level of feeding and feed type, nor between enhancement of silage fermentation and feed type on volatile fatty acid proportions.

Regression of Volatile Fatty Acid Proportions on Feed Chemistry

Daily mean volatile fatty acid proportions were regressed on the feed chemistry of the total feed (test feed + silage), expressed as 12 chemical fractions. The following objectives were used to derive the best regressions:

- To minimize the residual standard deviation.
- To minimize the number of predictors.
- To achieve a consistent set of terms between regressions.

Fig. 140a: Measured vs predicted acetate % from
regression on the test feed : silage ratio
(feeds 3, 4, and 5).
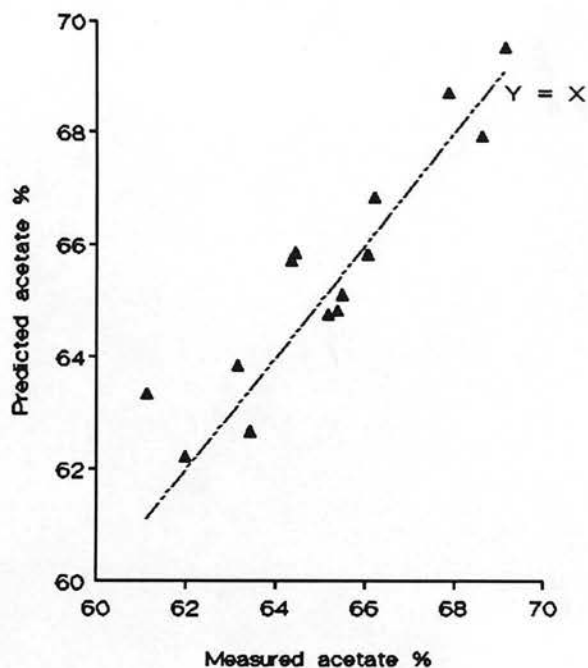
Fig. 140b: Measured vs predicted propionate % from
regression on the test feed : silage ratio
(feeds 3, 4, and 5).
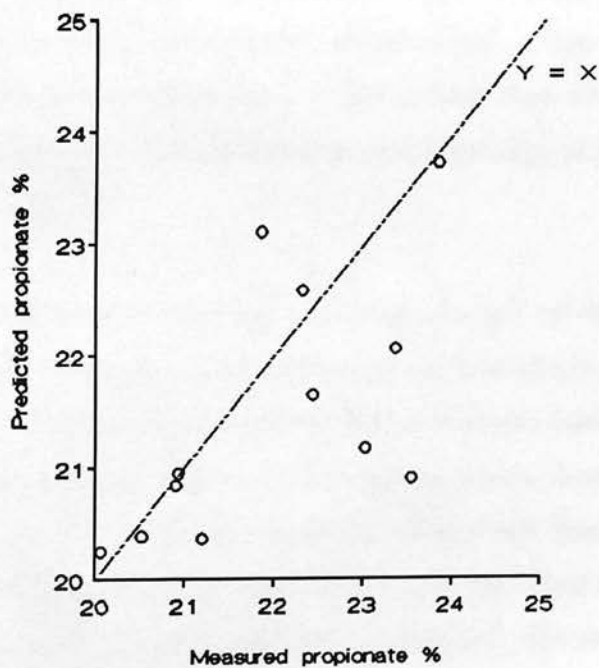
Fig. 141: Measured vs predicted butyrate % from regression on the test feed : silage ratio (feeds 3, 4, and 5).
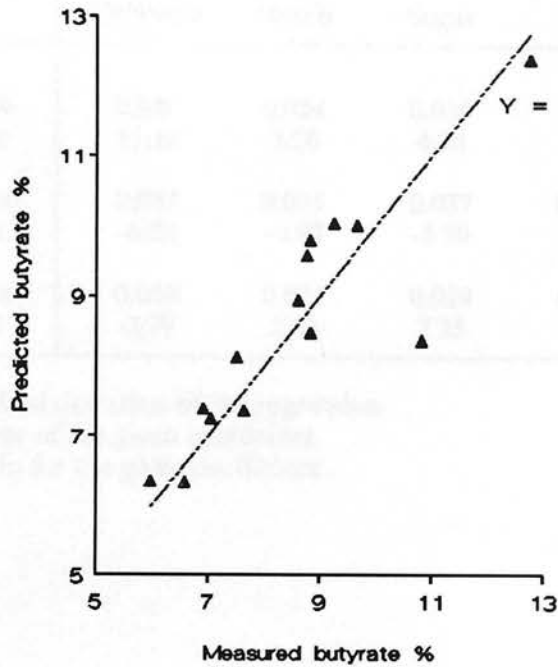
The standard errors of the coefficients for three major predictors, and the measures of precision for the regressions are given in table 142. Graphs of measured values plotted against predicted values from the regression are presented in figures 142, 143a, and 143b for acetate, propionate and butyrate. It can be seen that, for the feeds tested, volatile fatty acid proportions can be predicted from a knowledge of the feed chemistry with relatively high precision.

Inclusion of a predictor expressing the test feed:silage ratio had no significant effect on the regressions. A more rigorous test for evidence of physical effects not accounted for by the feed chemistry was to predict the volatile fatty acid proportions for the test feeds alone, by two different methods. The first was to predict volatile fatty acid proportions for each test feed from a chemical description of the test feed. This used the regressions derived from the feed chemistry of the total feed (test feed + silage) to predict the volatile fatty acid proportions for the test feed only. The second method was to predict volatile fatty acid proportions for each test feed from a non-chemical

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Table 142: The variation and significance associated with the multiple linear regressions and with the three major predictors of volatile fatty acid proportions.

Regression		Cellulose	Starch	Sugar	s	R^2adj
Acetate	se	0.040	0.024	0.036	1.099	76.1
	t	11.10	3.28	4.26		
Propionate	se	0.037	0.024	0.037	0.835	73.5
	t	-6.22	-6.87	-5.70		
Butyrate	se	0.030	0.021	0.029	0.741	82.1
	t	-2.79	5.63	7.35		

s = the residual standard deviation of the regression.
se = the standard error of the given coefficient.
t = the Students t ratio for the given coefficient.

Fig. 142: Measured vs predicted acetate % from the multiple linear regression on the feed chemistry of the total feed (all feeds).
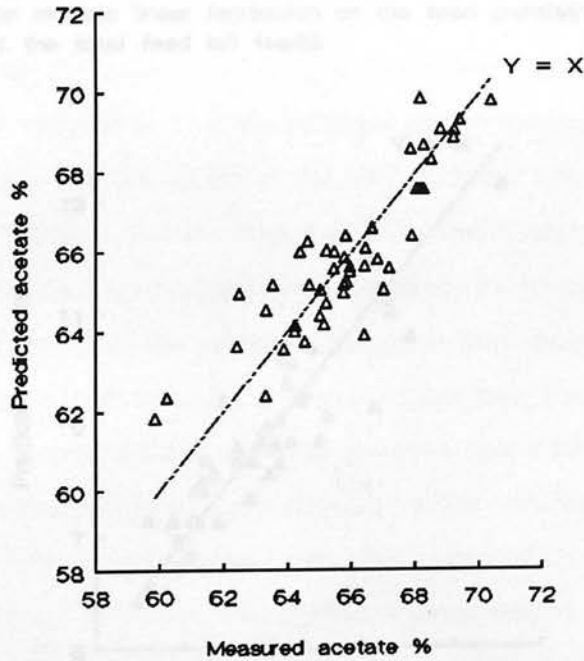
Fig. 143a: Measured vs predicted propionate % from the multiple linear regression on the feed chemistry of the total feed (all feeds).
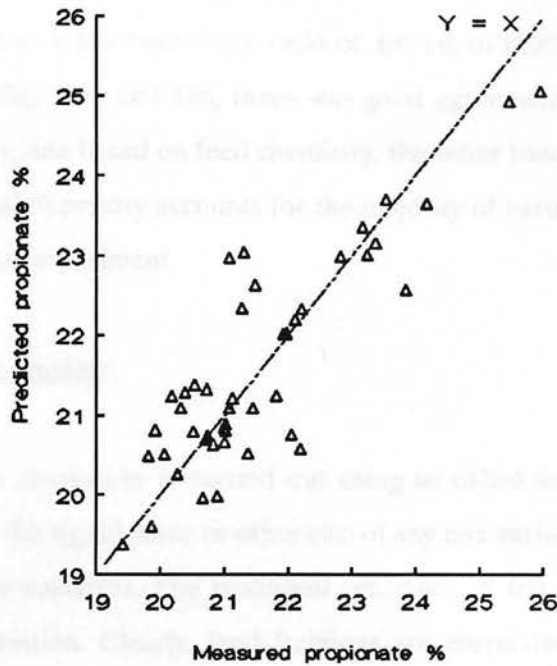
Fig. 143b: Measured vs predicted butyrate % from the multiple linear regression on the feed chemistry of the total feed (all feeds).
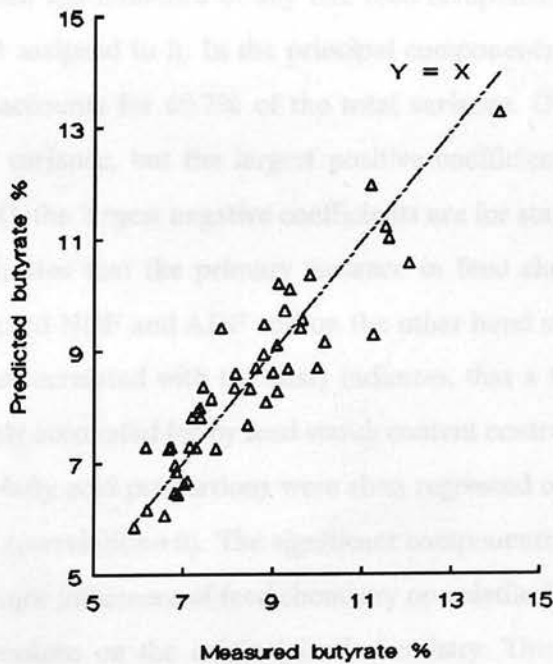
description of the test feed which incorporated any physical characteristics of that feed. This was to extrapolate the relationship between the test feed:silage ratio and volatile fatty acid proportions to a test feed:silage ratio of 1:0, i.e. to 100% test feed. As can be seen from figures 145a, 145b and 146, there was good agreement between these two methods of prediction; one based on feed chemistry, the other based on feed type. This indicates, that the feed chemistry accounts for the majority of variation in volatile fatty acid proportions in this experiment.

Principal Components Analysis

When multiple linear regression is carried out using so called independent variables which are correlated, the significance or otherwise of any one variable may be due to its correlation with other variables. The biological relevance of the chosen predictors is then brought into question. Clearly, feed fractions are correlated, not least because they sum to unity. Principal components analysis apportions the variance in a data set into variates which are orthogonal and therefore not correlated (see Rook et al, 1990a,b,c). The results of this analysis for feed fractions is presented in table 146. Within each component the influence of any one feed component is expressed by the size of the coefficient assigned to it. In the principal components analysis (table 146), the first component accounts for 60.7% of the total variance. Obviously all the feed fractions exert some variance, but the largest positive coefficients are for NDF and ADF (0.672 and 0.484), the largest negative coefficients are for starch and sugar (-0.536 and -0.129). This indicates that the primary variance in feed chemistry is a contrast between on the one hand NDF and ADF and on the other hand starch and sugar. The second component (uncorrelated with the first) indicates, that a further 23.7% of the total variation is mainly accounted for by feed starch content contrasting with sugar and protein. The volatile fatty acid proportions were then regressed on these components which are orthogonal (correlation=0). The significant components in these regressions represent the same major influences of feed chemistry on volatile fatty acid proportions as found in the regressions on the original feed chemistry. Thus, the results of the original regressions are validated.

Y = X

Acetate % from test feed : silage ratio

Acetate % from feed chemistry

Fig. 145b: Propionate % for the test feeds
alone, predicted by two methods.

Y = X

Propionate % from test feed:silage ratio

Propionate % from feed chemistry

Table 14?: The results of principle component analysis based on the chemistry of the test feeds only, the first five components (95% of 11) are shown. (The analysis is based on a correlation matrix.)

Component	1	2	3	4	5
Variation accounted for:					
Proportion	0.627	0.237	0.??	0.213	0.007
Cumulative	0.6??	0.844	0.9??	0.982	0.989
Coefficients for feed loadings:					
CP	-0.1?9	-0.307	0.?45	-0.231	0.240
ADF	-0.1??	0.0??	0.404	0.376	0.0?16
Ash	0.0??	-0.?30	0.069	-0.0??	-0.306
ADL	0.0??	-0.?06	0.072	-0.271	0.521
ADF	0.0??	0.185	-0.0?5	-0.0??	0.086
NDF	0.0??	0.353	-0.012	0.0??	0.213
Starch	0.?5?			-0.??	0.208
Sugar	-0.1??				0.?91
NDIP	0.02?	-0.0??	0.1??	0.4?0	0.5?9

(CP is crude protein; ADF is acid insoluble ether extract; ADL is acid detergent lignin; ADF is acid detergent fibre; NDF is neutral detergent fibre; NDIP is neutral detergent fibre insoluble protein.

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fig. 146: Butyrate % for the test feeds alone, predicted by two methods.

The principal components analysis on the volatile fatty acid proportions is presented in table 147. This shows that the primary variation in volatile fatty acids (64.8% of the total) is between acetate (0.607) and propionate (+0.125) plus butyrate (+0.322). The contrast between acetate and butyrate is greater than between acetate and propionate.

Conclusion

The objectives of this trial were achieved. A set of empirical regressions relating volatile fatty acid proportions to feed chemistry have been derived, which apply to a relatively wide range of feed composition. The precision of the regressions is high enough to make them useful estimates of volatile fatty acid proportions. Three possible influences on volatile fatty acid proportions which are not usually directly related to the feed chemistry, namely the level of feeding, the ratio of test feed:silage and enhancement of silage fermentation, have been accounted for in the derivation of these regressions.

Table 146: The results of principal components analysis on the chemistry of the test feeds; only the first five components (out of 11) are shown. (The analysis is based on the covariance matrix.)

Component	1	2	3	4	5
Variation accounted for:					
Proportion	0.607	0.237	0.124	0.013	0.007
Cumulative	0.607	0.844	0.969	0.982	0.989
Coefficients for feed fractions:					
CP	-0.069	-0.367	0.743	-0.283	0.240
AEE	-0.008	0.022	0.064	0.376	0.016
Ash	0.040	-0.020	0.069	-0.023	-0.306
ADL	0.057	-0.008	0.073	-0.073	0.521
ADF	0.484	0.185	-0.045	-0.673	0.086
NDF	0.672	0.333	-0.012	0.266	0.213
Starch	-0.536	0.731	0.021	-0.189	0.268
Sugar	-0.129	-0.427	-0.647	-0.195	0.391
NDFIP	0.020	-0.055	0.106	0.418	0.534

CP is crude protein; AEE is acid hydrolysed ether extract; ADL is acid detergent lignin; ADF is acid detergent fibre; NDF is neutral detergent fibre; NDFIP is neutral detergent fibre insoluble protein.

The principal components analysis for the volatile fatty acid proportions is presented in table 147. This shows that the primary variation in volatile fatty acids (64.8% of the total) is between acetate (0.844) and propionate (-0.325) plus butyrate (-0.422). The contrast between acetate and butyrate is greater than between acetate and propionate.

Discussion

The objectives of this trial were achieved. A set of empirical regressions relating volatile fatty acid proportions to feed chemistry have been derived, which apply to a relatively wide range in feed composition. The precision of the regressions is high enough to make them useful predictors of volatile fatty acid proportions. Three possible influences on volatile fatty acid proportions which are not usually directly related to the feed chemistry, namely the level of feeding, the ratio of test feed:silage, and enhancement of silage fermentation, have been accounted for in the derivation of these regressions.

Table 147: The results of principal components analysis on the volatile fatty acid proportions; only the first five components (out of 7) are shown. (The analysis is based on the covariance matrix.)

Component	1	2	3	4	5
Variance accounted for:					
Proportion	0.648	0.289	0.049	0.010	0.002
Cumulative	0.648	0.937	0.986	0.996	0.998
Coefficients for VFA proportions:					
Acetate	0.844	-0.061	-0.326	0.114	0.150
Propionate	-0.325	0.723	-0.412	0.207	0.113
Butyrate	-0.422	-0.685	-0.399	0.163	0.152
iso-Butyrate	-0.018	0.015	0.288	0.210	-0.383
Valerate	-0.064	0.048	0.175	-0.791	0.408
iso-Valerate	-0.017	-0.012	0.672	0.397	0.293
Hexanoate	0.003	-0.029	0.003	-0.301	-0.736

Within the confines of the confidentiality agreement under which this work was carried out, i.e. without presentation of quantitative results, it is difficult to discuss these regressions in relation to other published predictors of volatile fatty acid proportions.

The limitations of these regressions should be recognised and they should be used with caution until they have been experimentally tested. In particular, two points of weakness can be identified:

The Use of Only One Forage

All the results are derived from feeds based on one uniform silage. This represented at least half of the dry matter of the total feed offered. In fresh weight terms the silage comprised at least 80% of the feed, so any changes in the physical form of the test feed were always greatly diluted as changes in the physical form of the total feed. This is a desirable characteristic for comparison between test feeds. It does however highlight the limited range of physical forms within which the regressions are valid. Incorporating into the regressions data from a similar trial, comparing different forages would strengthen the model.

The Minimalist Design for Within Feed Effects

The experiment was designed to account for the effects of changes in the level of feeding, silage fermentation, and test feed:silage ratio. This was satisfactorily accomplished using only three treatments per effect, i.e. assuming that these effects could be accounted for by linear adjustment. The experiment was not designed, and did not permit exploration of these factors as potential influences on volatile fatty acid proportions. As such the predictive equations derived from these data, whilst accounting for these influences, are limited.

Application of the Regressions

The regressions derived from this data set were used to predict the volatile fatty acid

proportions arising from the different feeds used in the dairy cow trial, which is presented in the next chapter. The dairy cow trial was initially started using silage based feeds and Calan gate feeders. However there were considerable problems of poaching through the Calan gates and the trial was abandoned. The trial was subsequently repeated using individually stall housed cows, but at this time silage was not available and hay based feeds were used instead. Consequently the physical form of these feeds was outside the range to which the regressions apply.

The feeds in that trial were formulated using either unmolassed sugar beet pulp (S) or a mixture of ground maize and barley (C). The predicted differences between S and C in acetate, propionate, and butyrate are given in table 149. Sutton and co-workers (1987) measured volatile fatty acid proportions in a similar hay based trial and found the effect of decreasing fibre content on propionate to be the inverse of the prediction from these regressions (table 149). An important difference, which may account for the discrepancy between the current predicted values and those of Sutton et al. (1987) is that the predictions, as well as being derived from sheep, were derived from silage based feeds. It has been found that silage based feeds resulted in higher butyrate and lower propionate proportions than hay based feeds (Bath and Rook, 1965; McCullough and Smart, 1968). The difference between hay based and silage based feeds may be the result of differences in the rumen protozoal populations (Eadie and Mann, 1970).

Table 149: Prediction and measurement of the effect on volatile fatty acid proportions of replacing a fibrous concentrate by a starchy concentrate in a hay based feed.

	Cow experiment (chap. 9) predicted	Sutton et al. (1987) predicted	measured
Difference (S - C) in:			
NDF (g/kg)	153	66	66
ADF (g/kg)	85	38	38
Acetate	-2.5	-1.1	-2.9
Propionate	-0.5	-1.3	+1.2
Butyrate	+2.0	+1.2	+0.7

The forage:concentrate ratio was 40:60; differences in feed composition are in g/kg DM, differences in volatile fatty acid proportions are in molar %.

The discrepancy between the measured volatile fatty acid proportions arising from hay based feeds and the volatile fatty acid proportions predicted for those feeds using regressions derived from silage based data serve to emphasis the limitations of these regressions. Further, it is clear that these regressions could not be used, as was hoped, to predict the proportions of rumen fermentation end-products in the dairy cow experiment reported subsequently. However this data set represents a valuable basis from which to develop, with further experimentation, a rumen model.

The Environmental Sensitivity of Variation in Level of Feed Intake and to Source of Non-Protein Energy

This trial was conducted at the Hannah Research Institute. I am very grateful to David Chamberlain and to Pat Garnsworthy for making the resources.

The research team which collected the data in this trial bears testimony to the efforts of the farm manager at the Hannah, and to Susan Roberts, who then assisted in data processing.
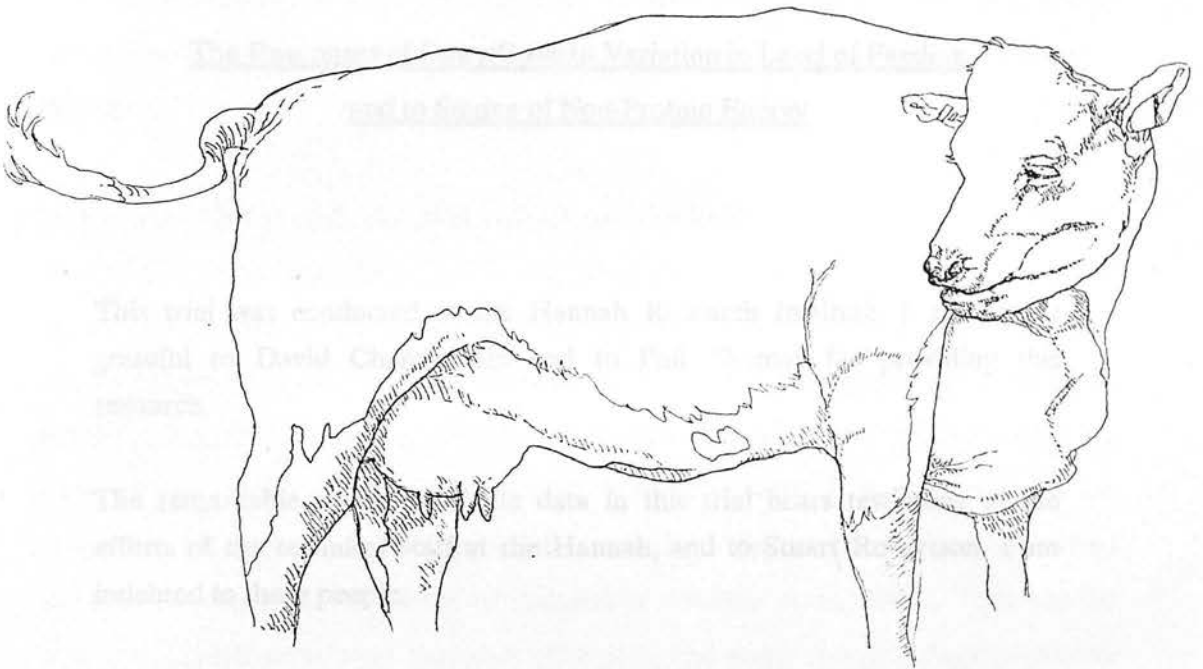
I would like to thank Neil Scott's hands, the outcome in condition scoring.

The gift of the unmolassed sugar beet pulp by Trident Feeds is gratefully acknowledged.

Note: All tables and figures are numbered according to the page on which they are found. As such table and figure numbers are not consecutive (see list of tables and figures, p. iv).

CHAPTER 9

The Responses of Dairy Cows to Variation in Level of Feeding and to Source of Non-Protein Energy

Note: All tables and figures are numbered according to the page on which they are found. As such table and figure numbers are not consecutive (see list of tables and figures, p. ix).

Introduction

The experiments described earlier in this thesis (chapters 4 to 7) have shown that the type of energy yielding nutrients in the feed can profoundly affect lactational performance in rats. The experiment presented in this chapter was designed to investigate this effect in dairy cows.

There is a massive body of literature concerned with the feeding of dairy cows. In this introduction I have made no attempt to present an extensive literature review, since such reviews already exist (e.g. Broster and Thomas, 1981; Garnsworthy, 1988; Sutton and Morant, 1989); rather I have described a few key experiments which are central to this work.

In the ruminant animal the proportions of carbohydrate and fat in the feed are markedly different from the proportions of glucogenic and lipogenic products absorbed by the animal after rumen fermentation (see chapter 8). There is scope for varying the "fat/carbohydrate" content of absorbed nutrients by altering the type of carbohydrate in the feed. Rapidly fermentable carbohydrates, especially when fed in large quantities, increase the acidity of the rumen environment. The result is a depression in the proportion of acetate produced by the fermentation. The compensating proportional increase is usually in the form of propionate (Sutton et al, 1988). Thus rapidly fermented carbohydrates are primarily glucogenic and slowly fermented carbohydrates are primarily lipogenic. Altering the proportions of these will affect the balance of absorbed nutrients.

It is clear that the form of carbohydrate in the feed of the dairy cow affects her lactational performance. Feeds containing a high proportion of concentrates (above 60%) usually cause a depression in milk fat content (Powell, 1938, Broster et al, 1985). This effect is more pronounced if the concentrate is of a particularly high starch content (Sutton et al, 1987). In this context *concentrates* represent *rapidly fermentable carbohydrate* and *forages* represent *slowly fermentable carbohydrate*. It is recognised that these definitions are not rigorous.

When interpreting the effects on milk production of changing the forage:concentrate ratio, a major complication is that the energy content of the feed is also altered. Consequently at equal feed intakes, feeds of different forage:concentrate ratio result in differences in energy intake. The results in table 154 show, that increasing the energy intake independent of the forage:concentrate ratio, and increasing the proportion of concentrate in the feed independently of the energy intake, both caused a depression in milk fat content (Broster et al, 1985). There was also a significant interaction between these two effects. Clearly, when comparing different sources of energy yielding nutrients for lactational performance, the interaction with level of intake should be considered.

Lactational responses to changes in the feeding level of dairy cows are generally described as conforming to a law of diminishing response; that is, the response in milk production per unit increment of feed decreases with increasing feed intake (Jensen et al, 1942; Burt, 1957; Blaxter, 1966). However, there are a number of possible confounding factors in the above studies which make the unconditional acceptance of this model inappropriate. An alternative, simpler model is the "*broken stick*" (Fisher et al, 1973). This model assumes a constant response per unit feed up to a maximum, followed by no further response in milk production with increasing feed. Both models are presented in figure 155.

Curnow (1973) has shown, that for a population of animals, who are individually following the *broken stick* model, the average response curve for the population will conform to the diminishing response model. It is therefore important to test between these two types of model by looking at data on individuals, or data which has been adjusted for differences between individuals. The work by Jensen et al. (1942) and Burt (1957) do not satisfy this criterion. The results described by Blaxter (1966) arose from a meticulous trial which involved daily individual records taken over a two year period (Blaxter and Ruben, 1953, 1954a,b). However, these results were never formally published and it is not clear how the final results were derived. Changes in level of feeding in the present trial were achieved by varying the amount of concentrate

Table 154: The effect of level of feeding and feed concentrate proportion on the milk fat content (g/kg) of British Friesian cows (parity >1) in weeks 9-16 of lactation. Data from Broster et al. (1985).

Level of Intake (MJ DE/day)	156	187	200 (ad lib)
Concentrate %			
60	41.0	35.6	34.5
75	31.5	31.3	21.9
90	24.1	21.9	21.3

offered, so both the level of feed intake and the forage:concentrate ratio were altered. This may well have affected the milk production response per unit feed. There are also studies which have reported linear responses to increments in feed intake (van Es and van der Honing, 1979, Broster et al, 1985) but it is not clear whether these were found in relation to individuals or to groups of cows.

The present experiment was therefore designed with the following objectives:

- To compare a glucogenic feed with a lipogenic feed at constant energy intake, protein intake, and forage:concentrate ratio.
- To compare different feeding levels using the above feeds so as to test between the two models of response described above.

In order to provide an adequate test of the two response models it was important, that the cows were individually rationed to be below the level of feeding that corresponded to the point of inflection of the broken stick model (figure 155). If this condition was not met it would not be possible to distinguish between the curve and the broken stick. Since milk production declines as lactation progresses it was important that feeding levels were scaled according to maximum milk production at the **end** of the trial. A forage:concentrate ratio which would not result in rumen acidosis whilst allowing a large difference in feed composition was also important.

Fig 155: Two alternative models of milk response to change in level of feeding.

Yield of milk component 'i'

Intake of feed component 'i'

——— broken stick

––– diminishing response
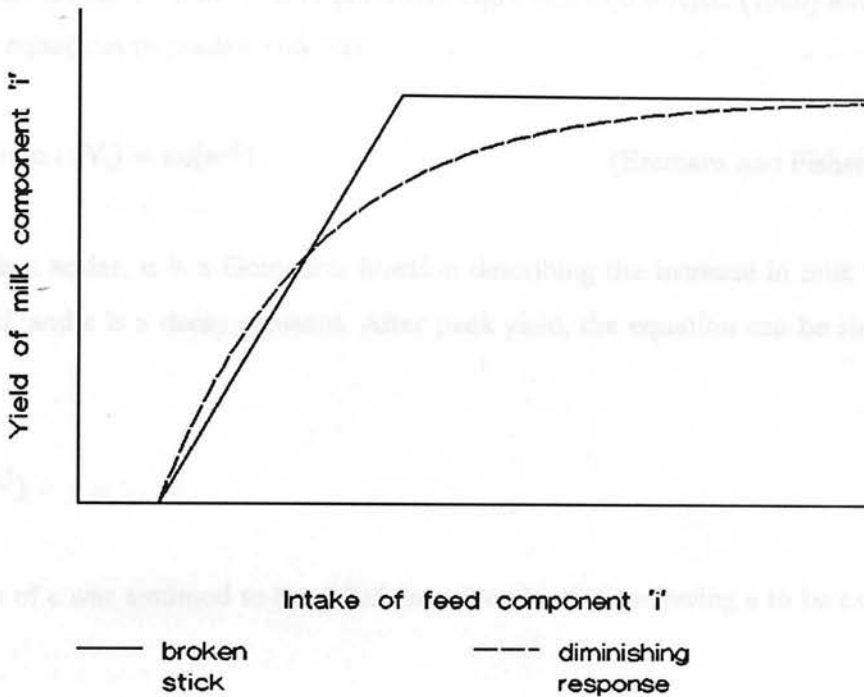
Given that these conditions were met, the following expectations were held:

1) That the glucogenic feed would increase milk production and depress milk fat content, as compared to the lipogenic feed.

2) That changes in level of feeding would alter milk production, eliciting a constant response per unit feed.

Materials and Method

18 Friesian cows (mean 17 weeks post partum), housed in individual stalls, were allocated in groups of six to one of three feeds (S, CS, C). The six cows on each feed were assigned sequentially for three periods, each of four weeks, to three feeding levels (L, M, H), using a replicated 3 x 3 Latin square design. Each cow was individually rationed according to the dry matter intake calculated to meet her predicted ME needs

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for maintenance plus milk yield at the **end** of the experiment, called DMRQ. This calculation was based on the ME requirement equations of the ARC (1980) and on the following equations to predict milk yield:

$$\text{Yield at time } t \ (Y_t) = au(e^{-ct}) \qquad \text{(Emmans and Fisher, 1986)}$$

where **a** is a scalar, **u** is a Gompertz function describing the increase in milk yield to peak yield, and **c** is a decay constant. After peak yield, the equation can be simplified to:

$$Y_t = a(e^{-ct})$$

The value of **c** was assumed to be -0.035 (see Wood 1979) allowing **a** to be estimated as:

$$a = Y_0/e^{-0.035 t_0}$$

where t_0 is the time in days from calving to the start of the trial, and Y_0 is the milk yield on that day. Given values of **a** and **c**, the milk yield at the end of the trial was predicted. Full data for this calculation are in appendix 6.

Calculation of the ME requirement (ARC, 1980) to achieve the milk yield at the end of the trial assumed no body state change during the trial. Feeding levels were then:

> H = DMRQ
>
> M = DMRQ - 1.5 kg DM
>
> L = DMRQ - 3.0 kg DM

The ME content of all the feeds was 10.8 MJ/kg and the crude protein content was 147 g/kg DM (table 157). Feeds consisted of hay and a pelleted concentrate in the ratio 40:60 (on a DM basis). The feeds differed in their fibre and rapidly digested carbohydrate content, achieved by substituting unmolassed sugar beet pulp (in feed S)

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Table 157: Feed ingredients and measured feed composition (g/kg DM unless otherwise stated).

	S	Feed C/S	C	Hay
Composition of total feed:				
ME (MJ)	10.77	10.72	10.68	-
Crude protein	147	148	147	89
NDF	540	453	387	734
ADF	281	235	196	401
Ether Extract[a]	15	17	18	14
Reducing Sugar	28	30	25	24
Ash	69	66	57	66
Concentrate ingredients:				
Barley	-	282	564	
Ground maize	-	100	200	
Unmolassed beet pulp	745	373	-	
Palmers S67 fish/meat-bone meal	58	56	54	
Soya bean meal	173	167	162	
"Megalac" protected fat[b]	5	2	-	
Dicalcium phosphate	15	8	1	
Limestone	-	6	11	
Calcined magnesite	1	2	2	
Salt	1	2	3	
CVT 01SI[c]	3	3	3	

a: Calculated from standard values.
b: Volac Ltd. Royston.
c: Trace element supplement. Nutrikem Ltd.

for cereals, barley and maize (feed C). The third feed (CS) was an equal mixture of feeds C and S. Feed ingredients and composition are given in table 157. Food was offered twice daily after milking, with ad libitum access to water. The following measures were made:

Food intake	daily
Food composition	weekly
Milk yield	daily
Milk composition	twice weekly, am/pm weighted composite samples
Liveweight	weekly
Condition score	fortnightly (ESCA, 1976)
Backfat thickness	fortnightly, by ultrasonic Vetscan

The data were analysed by analysis of variance using two sets of covariates; one to test for a carry over effect of feeding level in the previous period and one to test for an interaction between any carry over effect and feed type (appendix 1). To accommodate differences between cows, covariates for pre-experimental performance were also included in the analysis. These affected only the comparison between feed types, i.e. between Latin squares. Comparison between feeding levels was within squares and therefore variation due to differences between cows was already accounted for by the design. This design therefore fulfilled the criteria for comparison of differences between feeding levels (Curnow, 1973).

Results

There were no refusals of feed and no health problems at any time during the experiment. The average food intake for the whole trial was 10.4 kg DM/day (i.e. the average of level M). Transient changes in milk production due to the introduction of a new level of feeding had stabilized by day 7 of the period (figure 159a). Thus the first weeks data for each period was excluded from the mean value for the period. Two cows (nos 8 and 14) were excluded from the analysis in the last period, the open squares in figure 159b, as they had started to dry off (confirmed by post experimental data).

Source of Metabolisable Energy: There was no significant effect of feed type on milk production or liveweight and condition. Mean values for the three feeds are presented in table 160.

Level of Feeding: Milk yield and milk component yields were significantly increased (p<0.001) by increasing feed level (table 161). Milk composition was affected as follows: fat content was significantly decreased (p<0.05) and lactose content significantly increased (p<0.001) by increasing feed level (table 161). Milk protein content was not significantly affected by feeding level except when the covariate for carry over effects was included in the analysis. With the carry over effect adjusted for,

Fig. 159a: Daily milk yields for two cows
on feeding sequences MLH and MHL.

first day of
period 3

milk yield (kg/d)

L111

GL6

days of experiment
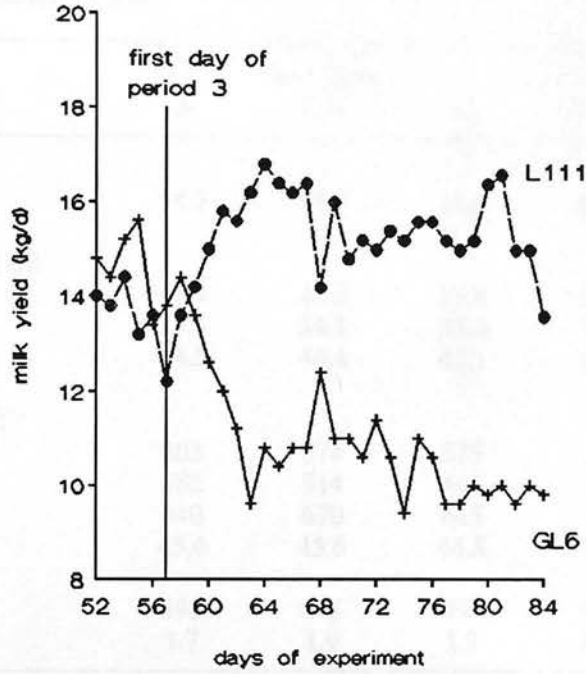
cow L111 fed low in period 2 and high in period 3
cow GL6 fed high in period 2 and low in period 3

Fig. 159b: Individual milk yields vs
level of feeding: all feed types.

Milk Yield (kg/d)

Feed intake (kg DM/d)

period   1   2   3   values
                                     excluded
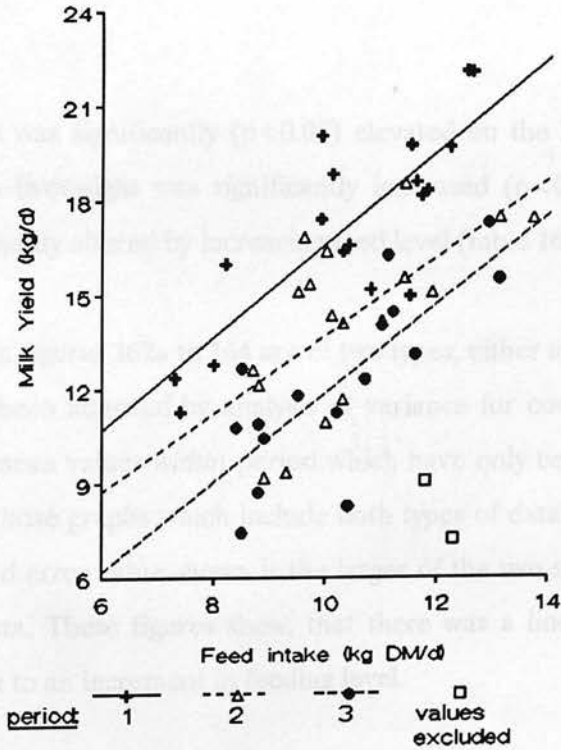
Table 160: Effect of feed type on milk production and liveweight; means from the last three weeks of each period, adjusted by covariance analysis using pre-experimental performance. Two missing values.

		feed type			
	S	C/S	C	sed	covar.[a]
Milk (kg/d)	14.3	14.9	14.5	0.85	MY
Milk content (g/kg) of:					
Fat	41.9	40.5	39.8	1.7	MFC
Protein	35.1	34.1	35.6	1.1	MPC
Lactose	44.5	44.4	45.1	0.8	MLC
Milk yields (g/d) of:					
Fat	603	574	575	53	MY
Protein	482	514	496	27	MY
Lactose	640	670	645	49	MY
Energy (MJ/d)	45.4	45.6	44.8	3.2	MY
Liveweight (kg)	495	495	499	5.2	LWT
Condition score[b]	1.7	1.9	1.7	0.2	-

a: Initial covariates are: MY = milk yield, MFC = milk fat content, MPC = milk protein content, MLC = milk lactose content, LWT = liveweight; all covariates were significant ($p < 0.05$).
b: condition score was measured in eighths of a unit (scale range = 5 units).

milk protein content was significantly ($p < 0.05$) elevated on the high level of feeding (figure 162a). Mean liveweight was significantly increased ($p < 0.001$), but condition score was not significantly altered by increasing feed level (table 161).

The data presented in figures 162a to 164 are of two types, either mean values *across* all periods which have been adjusted by analysis of variance for cow, feed, and period effects, or they are mean values *within* period which have only been adjusted for cow and feed effects. In those graphs which include both types of data (figures 162b, 163b, and 164) the standard error value shown is the larger of the two standard errors from the within period data. These figures show, that there was a linear increment in all measures in response to an increment in feeding level.

Table 161: The effect of feeding level on milk production and liveweight; means from the last three weeks of each period, adjusted for cow and period effects. Two values missing.

| | Feeding level | | | | |
	L	M	H	sed	p<
Milk (kg/d)	13.1	14.7	15.9	0.21	0.001
Milk content (g/kg) of:					
Fat	41.8	40.8	39.6	0.8	0.05
Protein	34.6	35.0	35.2	0.4	n.s.[a]
Lactose	44.0	44.9	45.1	0.3	0.001
Milk yields (g/d) of:					
Fat	539	595	618	13	0.001
Protein	441	502	548	8	0.001
Lactose	575	663	717	10	0.001
Energy (MJ/d)	40.9	46.0	48.9	0.7	0.001
Liveweight (kg)	488	496	505	1.5	0.001
Condition score[b]	1.69	1.72	1.77	0.03	n.s.
Decline[c] (ln kg/d)	-0.0090	-0.0075	-0.0061	0.0007	0.01

a: non significant in the model without carry over effects, significant when adjusted for carry over effects.
b: condition score was measured in eighths of a unit (scale range = 5 units).
c: Decline in daily milk yield

Deviations from linearity were tested for by inclusion of a covariate in the analysis to compare the difference between the effect of feeding level in periods 1 or 2 and the effect of feeding level in period 3 (appendix 1). There were no significant deviations from linearity. The difference between period means gives, independent of the absolute feeding level, the effect of decreasing feed level by one [H - M and M - L] or two steps [H - L] and of increasing feed level by one [M - H and L - M] or two steps [L - H]. It can be seen from figures 165a and 165b that the response to a change in feeding level was not affected by the absolute level of feeding or by the direction of change in feeding level, within the range measured.

Interaction between Feed Type and Level of Feeding: There was no significant interaction between feed type and feed level on any of the variables measured.

Fig. 162a: Mean milk composition versus level of feeding.

component: fat   fat c.o.   protein   protein c.o.   lactose
c.o. = adjusted for carry over effect (see text)
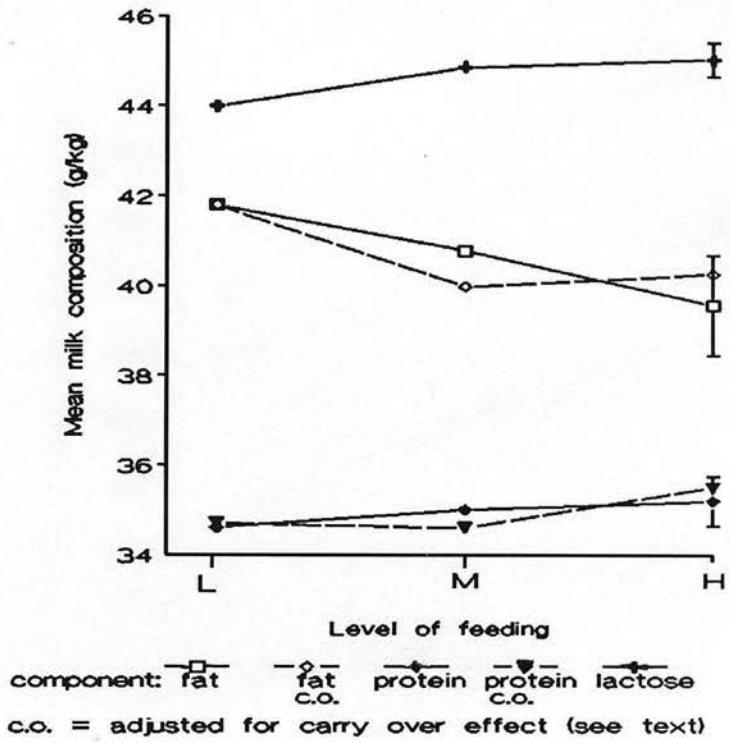
Fig. 162b: Mean milk yield vs feeding level; for each period and overall.

Period: All   1   2   3   se

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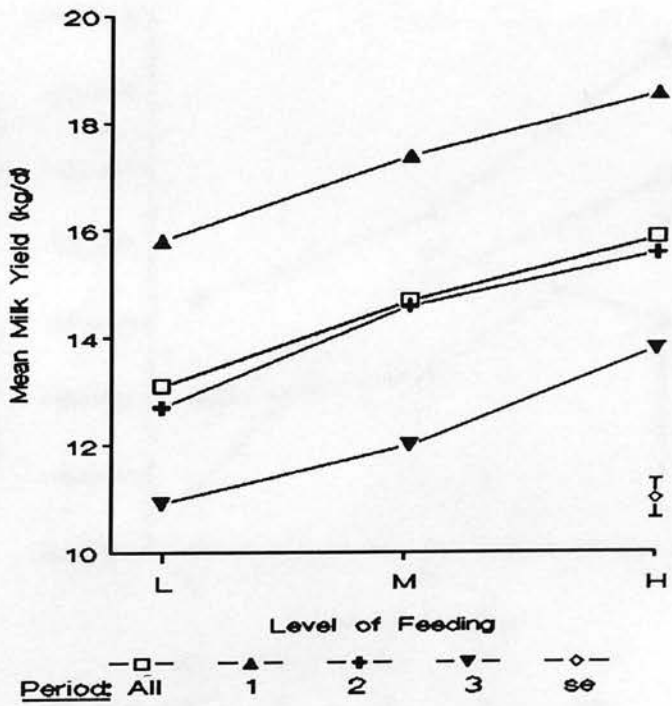
Fig. 163a: Mean milk component yields vs level of feeding.
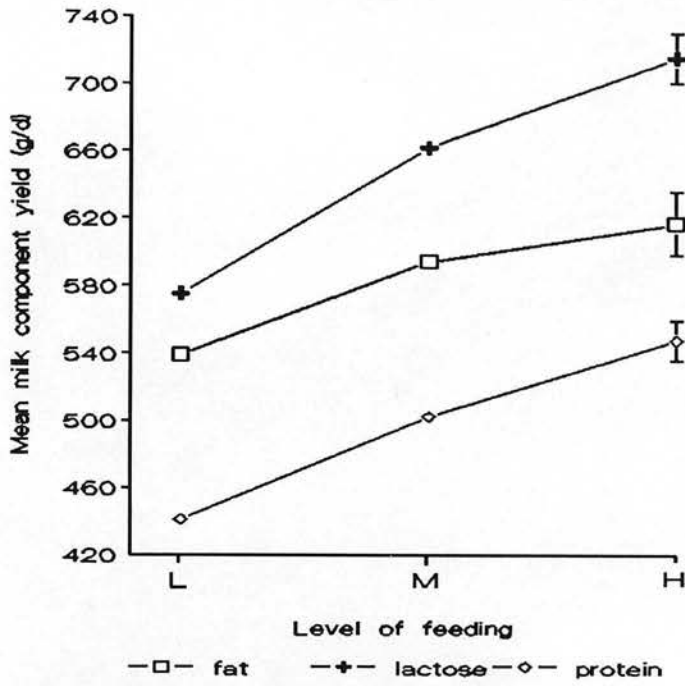
Fig. 163b: The slope of decline in milk yield (log kg/d) vs feeding level.

163
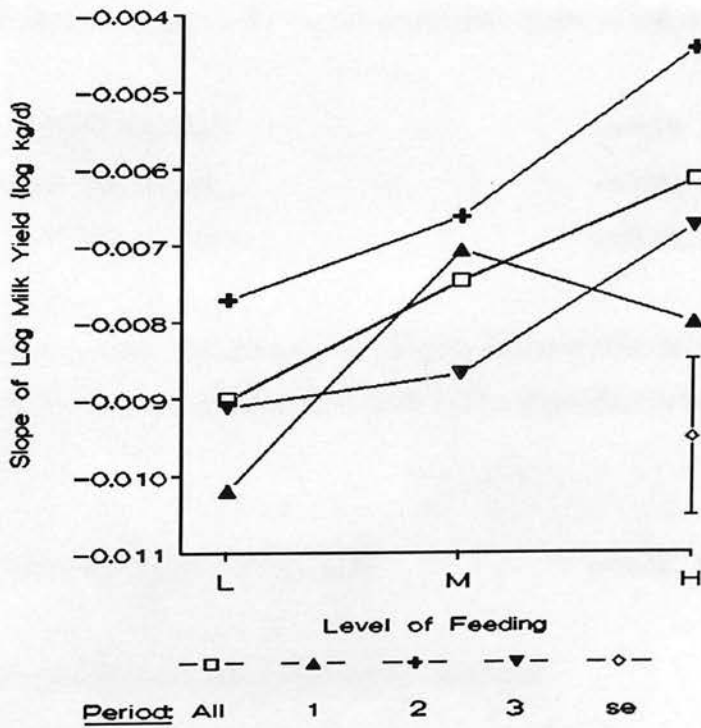
Fig. 164: Mean live weight vs feeding level: for each period and overall.
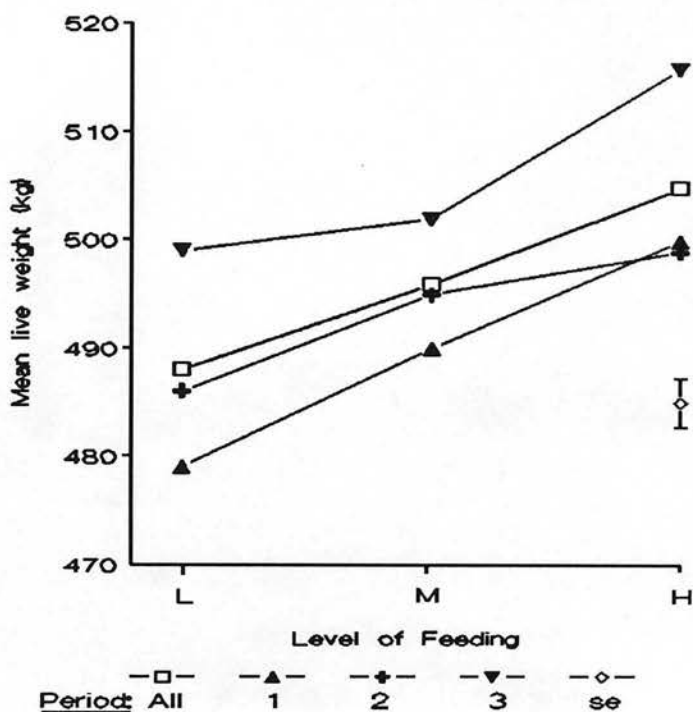
Time Trends: From all the above graphs it is clear that there was a significant effect of period (p<0.001) on all variables measured. For example, regressions of mean milk yield on metabolizable energy intake for the three periods are as follows:

$$Yield_1 = 7.5 + 0.93(ME\ intake_1) \qquad s=0.16;\ R^2(adj)=98.6\%$$
$$Yield_2 = 4.1 + 0.98(ME\ intake_2) \qquad s=0.36;\ R^2(adj)=94.3\%$$
$$Yield_3 = 2.4 + 0.95(ME\ intake_3) \qquad s=0.28;\ R^2(adj)=96.2\%$$

The slopes of these regressions are not significantly different from each other, but the intercepts are. The effect of period on milk yield can be quantified by including it in the regression, thus:

$$Yield = 9.7 + 0.95(ME\ intake) - 2.5(period) \qquad s=0.35;\ R^2(adj)=98.1\%$$

There was no interaction between period and feeding level.

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Changes in feeding level resulted ... ... increase in milk yield within each period, as reflected in the slope ... ... net change in milk yield in the first week of the period. The size of this net response to the slope of regression between logₑ milk yield and time ... significantly (p < 0.01) related to the level of feeding (figure 165a).

There was no significant carry-over effect of level of feeding on the response ... milk production as the production of ... milk fat content (p > 0.05) and milk protein content (p > 0.05). The slopes ... ... allowing for the carry-over effect can be seen ... figure 165b. Injected food ... ... 'treatment' effects ... ... feeding on milk fat content was established by carry-over effect whilst the effect of ... feeding on milk protein content was ... significant ... significant for the carry-over effect. There was no significant interaction between food type and carry-over effect.

Fig. 165a: Response in milk yield to changes in feeding level.

slopes = 1.41
R² = 97.8

Change in feeding level

+ period 2 minus 1   --△-- period 3 minus 2

Discussion

Average Manipulable Energy

Despite using feeds which contained a relatively low proportion of concentrate (60% DM), it was expected that there would be an effect of MH source on milk produced as ... ... de Groot, 1981; Thomas et al, 1980) Sutton et al. (1987) suggested a dietary effect. Fibrous concentrate at 60% inclusion in a hay based ration, and found only slight increase in milk fat content on the fibrous feed. The NDF and ADF contents of the feeds from the trial by Sutton et al. (1987) and from the present trial are presented in table 167. Since the difference in concentrate composition, in those terms, was more than ... related to that in the work of Sutton et al. (1987) an effect was expected. The absence of any effect instead ... ... probably due to the degree of ... ... to these present ... ... A ... ... study of the effect of feeding level possible. There is evidence which suggests that response to a change in feeding are greater in high yielding than in low yielding cows (Johnson, 1977; Strickland and Broster, 1981). Therefore at low (maintenance) levels of feeding fermentation differences between feed type are smaller than at high (lactating) levels.

Fig. 165b: Response in liveweight to changes in feeding level.

slope = 8.7 8.4
R² = 99.3 85.9

Change in feeding level

+ period 2 minus 1   △ period 3 minus 2

Contrary to expectation, there was a constant rate of decline in milk yield within each period, in addition to the rapid transient change in milk yield in the first week of the period. This rate of decline, expressed as the slope of a regression between $\log_e$ milk yield and time, was significantly ($p < 0.01$) related to the level of feeding (figure 163b).

Carry Over Effects.   There were no significant carry over effects of level of feeding in the previous period on milk production except for milk fat content ($p < 0.01$) and milk protein content ($p < 0.05$). The effect of adjusting for the carry over effect can be seen in figure 162a (dotted lines). The significant effect of level of feeding on milk fat content was not altered by the carry over effect whilst the effect of level of feeding on milk protein content was only significant after adjustment for the carry over effect. There were no significant interactions between feed type and carry over effects.

<u>Discussion</u>

Source of Metabolisable Energy

Despite using feeds which contained a relatively low proportion of concentrate (600 g/kg DM), it was expected that there would be an effect of ME source on milk production (de Visser and de Groot, 1981; Thomas et al, 1984). Sutton et al. (1987) compared a starchy and a fibrous concentrate at 60% inclusion in a hay based ration, and found only a slight increase in milk fat content on the fibrous feed. The NDF and ADF contents of the feeds from the trial by Sutton et al. (1987) and from the present trial are presented in table 167. Since the difference in concentrate composition, in these terms, was more than twice that in the work of Sutton et al. (1987) an effect was expected. The absence of any effect of feed type on milk production was probably due to the degree of rationing applied to these cows in order to make the study of the effect of feeding level possible. There is evidence which suggests, that responses to a change in feeding are greater in high yielding cows than in low yielding cows (Johnson, 1977; Strickland and Broster, 1981). Further, at low (maintenance) levels of feeding fermentation differences between feed types are smaller than at high (lactating) levels

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of feeding (Sutton, 1985; Sutton et al, 1988). In the last period and particularly on the high level of feeding the cows were least severely rationed in relation to their predicted level of performance. Even in this period there was no effect of feed type on milk fat content, though it must be recognised that this is statistically a much weaker comparison than that employing the whole design.

Interaction between Feed Type and Level of Feeding

Given that the feeds were of identical protein contents, and that no effect of feed type on milk production was found, it was not surprising that there was no significant interaction between feed type and level.

The highly significant linear response to increments in feed level (figures 162a,b, 163a, and 164) seems to provide evidence against the diminishing response model. However, the results also make the broken stick model difficult to adopt since the linear slope of the response is, in energy terms, 0.25 (table 169). If the broken stick model applies to the response of milk energy to feed energy, then by implication all the milk energy should have been produced with an efficiency of 0.25. Back calculation shows, that this was only possible if body reserves were being mobilised (table 169). Two possible explanations are presented below; the first favours the diminishing response model, the second favours the broken stick model; neither is adequately refuted by the results.

Table 167: The NDF and ADF contents of the total feed and the average milk fat contents in the current trial and in the trial of Sutton et al. (1987).

	Current experiment			Sutton et al. (1987)		
	(S) (fibrous)	(C) (starchy)		fibrous	starchy	
Feed content of						
NDF	540	387		454	388	
ADF	281	196		231	193	
			sed			sed
Milk fat content	41.9	39.8	1.7	39.5	36.9	1.1

Since these cows were gaining liveweight, it is not likely that they were mobilising body energy reserves (though it is possible, if body protein reserves were being simultaneously increased). It follows that not all the milk energy was produced with a constant efficiency of 0.25, and that in this experiment milk energy has been produced with decreasing efficiency as feed energy increased. Thus the linear slope observed is actually part of a diminishing response curve which, at the levels of feeding used in this experiment, is of such shallow curvature as to make it indistinguishable from a straight line. The efficiency of milk protein production from metabolisable protein supply was calculated as 0.37 (table 169; calculated according to Oldham, 1987). This estimate of the efficiency of milk protein production is low (ARC, 1984; Oldham, 1987). In order to produce all the milk protein with this efficiency body protein mobilisation would have been necessary, a condition not supported by the liveweight data.

Assuming that the responses in milk energy and milk protein conform to the diminishing response model, curves of diminishing response can be constructed. Two types of curve were constructed, one based on the observed mean yields and the other based on the smallest deviation which was significantly different from linearity. Using the standard error of the difference (sed) between feeding levels for a given milk product and the "t" value for p=0.05 (with d.f.=6); the minimum deviation from linearity which is significantly different from a straight line was calculated as:

$$\text{deviation} = t \times sed \times (3^{0.5}) = 4.238 sed$$

Adjusting the value of the milk product output for the middle level of feeding by the deviation (see figure 170) created the data for the second curve. Non-linear regression was used to fit curves to the data with the following equation:

$$\text{Yield} = b(1 - e^{(-c(\text{intake}-a))})$$

where   a is the maintenance requirement
           b is the maximum yield
           c describes the degree of curvature
           e is the exponent

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Table 169: Calculation of possible efficiencies of milk production assuming a linear response to increment in intake and no body state change.

	Metabolisable Energy[a] (MJ/d)	Protein (g/d)
Yield at feed level H	48.9	548
Yield at feed level L	40.9	441
Yield difference [H - L]	8	107
DMI[b] difference [H - L]	3 kg/d	3 kg/d
Feed content	10.7 MJ/kg	96 g/kg
Intake difference [H - L]	32.1	288
Measured efficiency	8/32.1 = 0.25	107/288 = 0.37
DMI at level H	11.9 kg/d	11.9 kg/d
Supply at level H	127	1142
Maintenance	51	272
Requirement for milk	48.9/0.25 = 196	548/0.37 = 1481
Total requirement	247	1753
Required minus supply	120	611

a: Energy in MJ/d and protein in g/d unless other units are indicated in the table.
b: DMI = dry matter intake

Feeding levels of energy and protein for maintenance and maximum milk energy and protein yields thus calculated are presented in table 171. These curves (described by 3 coefficients) have been constructed from only three data points, each point being the average value for 18 cows. However, as a result of the Latin square design and the meticulous efforts of the technical staff, the residual variations about these points, expressed as the sed value, was extremely low. To create significant curvature, the data were adjusted by the sed value; since this value was low, the resulting increase in curvature was therefore small (table 171), and yet resulted in marked differences in predicted performance. The differences between the values in table 171 derived from the original data and from the adjusted data indicate, that a *significant* curvature cannot be assumed for these data. This substantiates the linear model over the range measured.

Fig. 170: Milk energy response to intake of energy: fitted line and curve.

Mean milk energy yield (MJ/d)

55
50
45
40
35

L   M   H

80 90 100 110 120 130 140

Energy intake (MJ ME/d)
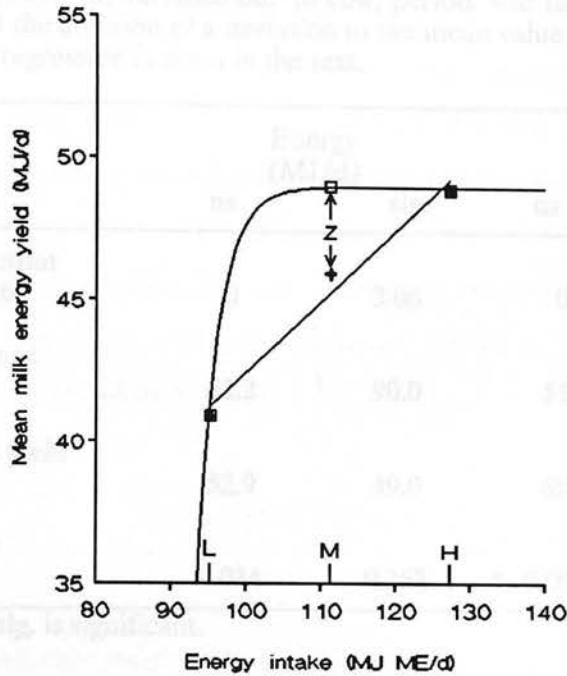
z = least significant deviation from linearity

That some milk components have a diminishing response to increments of feed does not necessarily invalidate the broken stick model. This is conditional on the production of different milk components being to some extent interdependent; there is substantial evidence that this is the case (Davies et al, 1983; Pearson et al, 1990).

If a cow was offered a feed which was limiting in its protein content, then milk production would respond primarily to the increase in protein intake from increments of feed. Since milk production was limited by protein supply, successive increments of feed would increment the energy intake at a rate faster than the rate of milk energy yield. Thus with successive increments of feed the milk energy yield divided by the energy intake would decrease. The efficiency of milk energy production would diminish, regardless of whether the response to feed protein was linear or not.

It is clear that both milk energy and milk protein showed a diminishing response to increments of feed, and therefore neither was the first limiting nutrient. It is unlikely

Table 171: The prediction of maintenance requirements and of maximum milk production by non-linear regression of yields on average intakes. The dependant data were mean yields adjusted for variance due to cow, period, and feed type (from table 161) with and without the addition of a deviation to the mean value for feeding level M. The equation for the regression is given in the text.

Curvature	Energy (MJ/d)		Protein (g/d)	
	ns	sig.	ns	sig.
Deviation in milk output from observed results	0	3.06	0	32.9
Predicted maintenance (coefficient a)	51.2	90.0	516	1132
Predicted maximum yield (coefficient b)	52.9	49.0	688	550
Degree of curvature (coefficient c)	0.034	0.363	0.0013	0.0091

ns is non-significant; **sig.** is significant.

that carbohydrate was the first limiting nutrient in the feed given the differences in carbohydrate content of the feeds offered. The measures taken in this trial are inadequate for evaluating other nutrients such as specific amino acids, which have been shown to affect milk production (Schwab et al, 1976) and may therefore have been limiting.

Time Trends

There was a significant time trend in the data (figure 163b) which was contrary to expectations. It had been expected that cows producing milk at a rate lower than their predicted rate on ad libitum feeding would have a milk yield limited by their feed intake and not by the productive capacity of their mammary glands. Therefore they would achieve a plateau milk production dependent on their level of rationing (Blaxter and Ruben, 1954a). Whilst there was a highly significant effect of feeding level on milk production, milk yield did not plateau within each period. There was a rapid response to change in feeding level which was complete by the seventh day on the new feeding level (figure 159a). For the remaining three weeks of the period there was a slow but

consistent decline in milk yield (figure 159a). This decline was expressed as the slope of $\log_e$(milk yield). It can be seen (figure 163b) that this slope was influenced by the level of feeding. This effect was not the direct result of previous feeding levels, as there were no carry over effects on milk yield or slope of yield decline. The decline in milk yield as lactation progresses is primarily due to a decline in the number of mammary gland cells (Wilde and Knight, 1989). Thus it would appear that the level of feeding not only influences current milk production, but also affects the rate at which the mammary gland atrophies. By this effect previous nutritional history may affect *capacity* to produce milk rather than *actual* milk production.

Conclusions

i) There was no significant effect of feed composition on milk production, at these levels of feeding.

ii) Milk production and liveweight were significantly elevated by increasing food intake. The response to increments of feed was linear.

iii) There was no interaction between feed composition and feeding level.

iv) Milk yield within each period did not plateau, but declined at a consistent rate which was significantly influenced by level of feeding.

v) There were no carry over effects of previous feeding level on yield of milk and milk components. Milk fat and protein contents were subject to a significant carry over effect.

CHAPTER 10

General Discussion

Note: All tables and figures are numbered according to the page on which they are found. As such table and figure numbers are not consecutive (see list of tables and figures, p. ix).

<u>Introduction</u>

The work presented in this thesis is at first sight rather diverse, encompassing three different species. However, there is a common theme which links the different aspects of this work. This theme is encapsulated in the questions:

1) How do we describe feeds in a manner which is biologically relevant?

2) How does lactation affect the ability of animals to accommodate their environment?

These questions are interdependent since a biologically relevant description of a feed depends upon the nature of the animal to which it is being offered; and the major aspect of a lactating animals' environment is its feed. For ease of discussion these two questions will be dealt with separately.

<u>How do we Describe Feeds in a Manner Which is Biologically Relevant?</u>

The original aim of this work was based on the notion that the conventional description of feeds, i.e. in terms of protein and energy, is inadequate for lactating animals. The production of milk lactose during lactation represents the only major net production of carbohydrate by mammals. It was argued that the carbohydrate content of the feed would be an important descriptor of feeds for lactational performance. This would apply particularly to ruminants, who sacrifice the ready availability of glucose in order to be able to digest cellulose; requiring them to synthesize the majority of their glucose by gluconeogenesis (Smith and Taylor, 1977). The work with lactating rats was designed to investigate the importance of feed carbohydrate in lactation.

The first rat experiment (chapter 4) showed that rats were able to lactate successfully on feeds containing as little as 6.7% carbohydrate. This was in agreement with several reported observations (Follis and Straight, 1943; Steingrimsdottir et al, 1980), however these reports did not furnish full data. The first rat experiment also showed that there were no significant differences in lactational performance between low and high

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carbohydrate feeds (67, 533 g/kg OM), except that the litters of dams on the low carbohydrate feed were significantly fatter (p<0.05) than those on the high carbohydrate feed. In common with the cited work, this experiment was carried out using high protein (400 g/kg OM) feeds. As such, the dams on the low carbohydrate feed were probably ingesting surplus protein which they could convert to glucose. Thus the protein content of the feed must be considered when investigating the effects of feed carbohydrate content on lactational performance.

This highlighted the necessity of considering all the components of the feed when comparing different feeds. This is a point which has often been overlooked when attributing nutritional causes to observed effects of feeding. As a means of keeping this concept in mind, a graphical representation of the feed as a triangular space was adopted, this is described fully in chapter 2. The second rat experiment (chapter 5) compared feeds of differing fat/carbohydrate content at two levels of protein inclusion (300 and 150 g/kg OM). The feeds of low protein content caused a highly significant (p<0.001) depression in lactational performance as compared to the high protein feeds. This was expected (Naismith et al, 1982). There were also a significant effect (p<0.05) of changing the feed fat/carbohydrate content at both levels of protein inclusion. The effect of changing the fat/carbohydrate content of the feed was subject to a massive interaction with protein content (p<0.001), see for instance figure 51a. Whilst this was most evident with feeds of low protein, low carbohydrate content, the effect was present across the whole range explored. This experiment demonstrated unequivocally the need for a feed description in terms of protein, carbohydrate, and fat in order to explain the observed lactational performance. A hypothesis to explain this interaction is discussed in the other section of this discussion (p. 178).

The milk of the rat contains a lower proportion of lactose to total milk solids (0.2) than does cows milk (0.4). The rat therefore has a lower glucose requirement per unit milk solids produced. The supply of glucose available to a rat from a unit of feed will be much greater than that available to a cow. Thus having shown the need to quantify the feed in terms of protein, carbohydrate, and fat for the lactating rat it was expected that an equivalent description of feeds for dairy cows would be needed to explain the effects

of different feeds on the lactational performance of dairy cows. A trial with dairy cows was carried out, one aim of which was to investigate the effect of glucogenic supply on milk production.

In order to investigate this it was necessary to find a satisfactory description of ruminant feeds (see Webster et al, 1988). There is good evidence that ruminants utilise their *absorbed* nutrients in broadly the same manner as non-ruminants utilise protein, carbohydrate, and fat (Mepham, 1983)). Assuming that absorbed nutrients could be classified as three proportions; aminogenic, glucogenic, and lipogenic (see chapter 8, p. 129), there remained the problem of describing the effects of rumen fermentation on feed constituents. An experiment (chapter 8) was designed to relate the end products of carbohydrate fermentation, the volatile fatty acids, to the chemical description of the feed, so that the feed could be described in terms of the proportions of the feed as "perceived" by the ruminant.

In vivo measurements in sheep of the volatile fatty acid proportions arising from 16 different feeds were used to construct regression equations relating the feed chemistry to the volatile fatty acid proportions. The resulting regressions accounted for 75% of the total variation in volatile fatty acid proportions. The average residual standard deviation of the regressions, expressed as a percentage of the mean value of the dependant variable was 4.8%. The limitations of these regressions are outlined in the discussion of chapter 8. It is, however, clear that within the range of feeds tested these regressions successfully predicted volatile fatty acid proportions. No test of the regressions by direct measurement of volatile fatty acids was carried out. The intended use of these regressions to allow a descriptions of the feeds used in the cow trial was precluded.

The dairy cow trial used three feeds whose composition were designed to cover a wide range of glucogenic/lipogenic content (chapter 9). There were no significant effects of feed type and hence feed composition on lactational performance. This was unexpected given the extremes in feed composition and seemingly contrary to other work (de Visser and de Groot, 1981; Thomas et al, 1984). The cows on this trial were severely

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rationed in order to investigate the lactational response to changes in level of feeding, (this aspect of the trial is discussed in the next section) and this may explain the absence of a feed composition effect.

The expectations, based on current descriptions of feeds, held in both the rat experiments and the cow trial were found to be wanting. Thus the work with rats and with ruminants has shown, in different ways, that feeds need to be defined in more biologically relevant terms. Using a three component description of the feed has permitted a clear understanding of the effects of feed composition on lactational performance, but has also emphasized the paucity of our current understanding.

How Does Lactation Affect the Ability of Animals to Accommodate Their Environment?

It is suggested above that the absence of an effect of feed composition on milk production in the dairy cow experiment was related to the absolute level of performance of the cows. Rationed cows have their milk production limited by their feed intake to a level below their potential milk production. The total daily requirement for glucose (and protein) is thus low compared to ad libitum fed lactating cows. This implies that a biologically relevant description of food should take account of the animals performance.

In addition to being the only time when mammals produce large amounts of carbohydrate, lactation is also the only time when the mammal is consistently functioning at such a high level of performance. It is not unusual for lactating mammals to have a food intake three times greater than their maintenance intake (Slonaker, 1925). As eloquently discussed by Kronfeld (1976) the level of production of an animal is inversely related to its ability to accommodate extremes of environment. In the context of feed composition this implies, that the closer an animal is to its maximum performance, the smaller the range of feed compositions which can support that performance. Thus, the biologically relevant description of feeds as "adequate" or "inadequate" depends upon the state of the animal.

It was clear from the second rat experiment (R2) that lactating rats respond differently from non-lactating rats to changes in feed composition. Dams offered the low protein feeds (150 g/kg OM) ate less than those offered the high protein feeds (300 g/kg OM), even when the latter feeds were of a low fat content (figure 54a). This is in agreement with other work (Naismith et al, 1982; Grigor et al, 1986), but it is contrary to expectations derived from non-lactating rats. Non-lactating rats offered a feed of inadequate protein content, attempt to alleviate the deficit by increasing their intake (Musten et al, 1974). In choice feeding experiments it has been shown that lactating rats increase their intake of protein with the onset of lactation (Richter and Barelare, 1938). The failure of the rats in this experiment (R2) to increase their protein intake suggests that they are subject to a constraint.

The results of this experiment (R2; chapter 5) indicate, that the constraint was not due to an excess or deficiency of any single nutrient. The energy intake of the dams was not constrained given the variation in observed energy intakes and given the extensive mobilisation of maternal body fat observed. Mobilisation of body fat would exacerbate any excess energy intake. The observation that on the high protein feeds, the energy intake of the dams increased as the carbohydrate content decreased suggested that the maternal heat production was constrained. At constant protein content, the heat increment per unit feed energy decreases as the carbohydrate content of that feed decreases (Forbes et al, 1946a,b,c). A drop in the heat increment would allow a greater energy intake to achieve the same heat production. The implication of this is that maternal heat production was maximal. Brody et al. (1938) measured the heat production of lactating rats and found it could reach twice the level of non-lactating heat production. Andik et al. (1963) showed that growing rats on low protein feeds were being constrained by their capacity to dispose of heat at 22°C. Given these observations it was proposed that:

Food intake and consequently the performance of lactating rats is constrained by their capacity to lose heat.

This hypothesis accounts for the increase in energy intakes, at constant feed protein content, observed when feed fat replaced carbohydrate as discussed above. The mobilisation of body fat is also in accordance with the hypothesis, since the efficiency of conversion of fat into milk fat is higher than the efficiency of conversion of food carbohydrate into fat (Chudy and Schiemann, 1969). As such, body fat represents a supply of energy available to the dam at a low cost in terms of heat production.

The lactational failure observed on the low protein, low carbohydrate feeds, can also be explained as a consequence of a constrained heat production as follows. The volume of milk produced is a function of the amount of lactose produced, and there is also a strong relationship between milk lactose and milk protein (Davies et al, 1983). If it is assumed that the low protein, low carbohydrate feeds resulted in a shortage of either protein or carbohydrate, then the milk production would consequently drop. A decline in milk production can also be seen as a decrease in the size of one outlet for the disposal of feed energy. The heat production associated with the ingestion of food would therefore be in part dependant on the milk production achieved using that food. Feeds that were of limiting protein or carbohydrate content which caused a decline in milk production, would exacerbate the problem of heat disposal, forcing the dam to lower her intake with the effect of decreasing milk production further still. Thus it is conceivable that food intake would spiral downwards. This explanation suggests, that the constraint on food intake could be related to the ratio of the first limiting nutrient:energy in the feed.

The third rat experiment was designed to explore the relationship between nutrient:energy ratios and lactational performance, focusing on the threshold of feed composition for lactation. The results of the third rat experiment indicated, that even when the nutrient:energy ratio was constant there was still a large difference in lactational performance between feeds. Clearly, the effects of feed composition on lactational performance cannot be explained by simple relationships between feed components. Heat production is influenced by the different productive processes which occur at varying rates in the animal. The effect on heat production of simple changes in feed composition is therefore difficult to predict.

In order to predict heat production, and to subsequently explore the hypothesis that a constraint on heat production was controlling lactational performance, a model was constructed (chapter 7). This model was however inadequate to either refute or support the hypothesis. Whilst the effects of feed composition on lactational performance can be described statistically by an interaction between the protein, carbohydrate, and fat contents of the feed, the reason for the interaction was not defined.

The importance of the animal in the definition of feeds for lactation was seen in the third rat experiment. The feed which resulted in the poorest performance in the third rat experiment was feed 2. This feed permitted two of the rats to achieve an adequate lactation with relatively small losses of body reserves. Two other rats on that feed suffered lactational failure despite massive mobilisation of body reserves, resulting in litters half the weight of the two successful rats (table 181). The remaining rat on that feed achieved an adequate lactation, but only by mobilising body reserves to the extent of the two rats who failed in their lactation. This demonstrates another aspect of the animals ability to accommodate extremes of feed composition. The intake of these five dams was correlated to their body protein losses, showing the importance of maternal body reserves. The use of a marginal feed amplified the difference between individuals. However, the body protein losses of these dams bore little relationship to estimates of their initial body reserves. None of the measures of the initial characteristics of these dams could be used to predict their ability to mobilise body reserves in response to an adverse environment. It would appear that not only the description of feeds for lactation but also our description of the animals, such as it exists, is inadequate. This also applies to dairy cows as personified by the often quoted differences between two "similar" cows (Broster, 1969) in lactational performance (table 181).

In dairy cows there is also evidence of the need to consider performance parameters in any biologically relevant description of the feed. The effect of different feed types on lactational performance is subject to an interaction with the level at which those feeds

Table 181: The lactational performance of individual animals offered identical feeds. The cow data are those of Broster et al. (1969). The rat data are from the fourth rat experiment.

	Rat 12	Rat 19	Cow 1	Cow 2
Initial maternal liveweight	359	358.5 g	517	519 kg
Intake	48.7	244.1 g/12d	rationed	rationed
Maternal liveweight gain	-98.2	-38.6 g/12d	+39.1	-51.6 kg/67d
Litter liveweight gain	32.9	179.8 g/12d		
Milk yield			824	1762 kg/67d

are given (Sutton et al, 1988). Therefore level of feeding would have to be a factor in any valid experiment investigating the effects of feed composition on lactational performance. As described in chapter 9, various models have been proposed to explain the effect of changes in level of feeding. The model incorporated into current predictions of the nutritional requirements of lactating ruminants (ARC, 1980) is a diminishing response curve, proposed by Blaxter (1966). This model is open to question and an alternative linear model has been proposed, the "broken stick" (Fischer et al, 1973; see figure 155). One purpose of the cow experiment (chapter 9) was to distinguish between the two models.

The results of this trial showed a clear linear response to level of feeding (see figure 162). However, the slope of the response resulted in an efficiency of milk energy production which was too low to sustain the total milk production. This can be explained in two ways. Firstly, that the response to level of feeding is a diminishing curve and that at these levels of feeding it is sufficiently shallow so as to be indistinguishable from linearity. Secondly, that the response to level of feeding is linear for the first limiting nutrient, consequently other nutrients and, in this case, energy intake are in excess of requirements for milk. In this situation the observed efficiency of milk production would be lowered. The cows showed significant increases in body condition with increasing feed level.

As in the previous section, it has been shown that the feeds of rats and dairy cows require a more biologically relevant description if successful prediction of lactational performance is to be made. It is clear that lactation limits the ability of the animal to respond to extremes of feed composition. Therefore the level of lactational performance needs to be considered when describing feeds for lactating animals (see also Leng, 1989). In lactation, the characteristics of the animal become very much more important. Even under the controlled conditions of rationed feeding, our understanding of the manner in which lactating animals respond to changes in their environment, in this case feeding level, is far from complete.

Conclusions

1) The description of feeds for lactating animals in terms of protein and energy alone is inadequate.

2) There is a highly significant interaction between feed protein, carbohydrate, and fat on lactational performance.

3) The assumption, that the level of heat production of the lactating dams is maximal, can explain the effects of feed composition on lactational performance.

4) The characteristics of the mother may have an important influence on the lactation, especially with extreme feeds. The important characteristics remain to be defined.

5) Dairy cows show a linear response to changes in level of feeding, which challenge the accepted model of diminishing response to changes in feeding level.

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The rat lactation model was written as a minitab program (in TR file), to calculate the model solutions from columns of data (referred to as c).

Column identities
Model parameters as defined in chapter 7 are in bold text.

c1 = treatment
c2 = proportion of protein in the feed, x (g/g)
c3 = proportion of carbohydrate in the feed, z (g/g)
c4 = proportion of fat in the feed, Y (g/g)
c5 = maternal intake; I ...
c6 = maternal ...
c7 = maternal ...

Appendix 1

The listing of the rat lactation model.
The analysis of variance programs used in the rat experiments.
The analysis of variance programs used in the cow experiment.

c8 = litter body protein gain; BP g/12 days
c9 = litter body fat gain; BF g/12 days
c10 = average maternal liveweight; kg
c11 = environmental temperature; °C
c12 = final maternal body protein; FINBP g
c13 = final litter body protein; FINBP g
c14 = litter maintenance protein requirement; PMA g/12 days
c15 = litter maintenance energy requirement; EMA g/12 days
c16 = milk protein yield, derived from litter growth data; PMK g/12 days
c17 = milk carbohydrate yield, derived from litter growth data; CMK g/12 days
c18 = milk fat yield, derived from litter growth data; FMK g/12 days
c19 = maternal maintenance protein requirement; PMA g/12 days
c20 = maternal maintenance energy requirement; EMA g/12 days
c21 = the predicted maternal capacity to lose heat; HCAP kJ/12 days
c22 = correction for body protein; BPCOR
c23 = correction for body fat; BFCOR
c24 = maternal heat production calculated using litter growth derived milk production; HEAT kJ/12 days
c25 = milk fat yield derived from maternal data only; FMK g/12 days
c26 = milk protein yield derived from maternal data only; PMK g/12 days
c27 = milk carbohydrate yield derived from maternal data only; CMK g/12 days

The rat lactation model was written as a minitab program (MTB file), it calculates the model solutions from columns of data (refered to as c).

<u>Column identities</u>

Model parameters as defined in chapter 7 are in bold text.

c_1 = treatment

c_2 = proportion of protein in the feed; **p** (g/g)

c_3 = proportion of carbohydrate in the feed; **c** (g/g)

c_4 = proportion of fat in the feed; **f** (g/g)

c_5 = maternal intake; **I** gDM/unit time

c_6 = maternal body protein gain; **BP** g/12 days

c_7 = maternal body fat gain; **BF** g/12 days

c_8 = litter body protein gain; **BP** g/12 days

c_9 = litter body fat gain; **BF** g/12 days

c_{10} = average maternal liveweight; kg

c_{11} = environmental temperature; ^{o}C

c_{12} = final maternal body protein; d14BP g

c_{13} = final litter body protein; d14BP g

c_{14} = litter maintenance protein requirement; **PMA** g/12 days

c_{15} = litter maintenance energy requirement; **EMA** g/12 days

c_{16} = milk protein yield, derived from litter growth data; **PMK** g/12 days

c_{17} = milk carbohydrate yield, derived from litter growth data; **CMK** g/12 days

c_{18} = milk fat yield, derived from litter growth data; **FMK** g/12 days

c_{19} = maternal maintenance protein requirement; **PMA** g/12 days

c_{20} = maternal maintenance energy requirement; **EMA** g/12 days

c_{21} = the predicted maternal capacity to lose heat; **HCAP** kJ/12 days

c_{22} = correction for body protein; **BPCOR**

c_{23} = correction for body fat; **BFCOR**

c_{24} = maternal heat production calculated using litter growth derived milk production;

<div align="right">

HEAT kJ/12 days

</div>

c_{25} = milk fat yield derived from maternal data only; **FMK** g/12 days

c_{26} = milk protein yield derived from maternal data only; **PMK** g/12 days

c_{27} = milk carbohydrate yield derived from maternal data only; **CMK** g/12 days

<u>The model program</u>

```
let c14 = 0.123*(2*c13-c8)
let c15 = 20.05*(2*c13-c8)
let c16 = (c8+c14)/(0.85*0.95)
let c17 = c16/2.4
let c18 = -0.886*c16-0.408*c17+1.814*c8+1.146*c9+0.693*c14+0.026*c15
let c19 = 0.123*(2*c12-c6)
let c20 = 20.05*(2*c12-c6)
let c21 = 12*(c10**0.75)*(1479.7-28*c11)
let c22 = (((c6**2)**0.5)+c6)/2
let c23 = (((c7**2)**0.5)+c7)/2
let c24 = c5*(9.718*c2+15.543*c3+6.343*c4)-10.229*c6+17.801*c22-16.024*c7
let c24 = c24+2.620*c23-2.668*c16-6.153*c17-13.404*c18+0.595*c20
let c25 = c5*(1.199*c2+0.599*c4+1.468*c3)-0.155*c21-1.262*c6+3.489*c22
let c25 = c25-1.514*c7+0.514*c23+0.117*c20
let c26=c5*(0.688*c2+0.628*c4+1.540*c3)-0.724*c6-1.587*(c7+c25)-0.040*c24
let c27 = c26/2.4
end
```

The analysis of variance program from R1 (in Genstat version 4).

```
'refe' 30890
'unit' $ 25
'fact' state $ 3
  :  feed $ 2
'input' 2
'read/p' rat,feed,state,Ilwt,Idwt,Iash,Ifat,Ipro
  :   lwtg,ewtg,dwtg,ashg,fatg,prog
  :   Pint,kjint,Fint,Cint,kjexp,H2Oint
'input' 1
'print/p' rat,feed,state,Ilwt,Pint,kjint,Fint,Cint $ 3(3),5(8.1)
  :    rat,lwtg,ewtg,dwtg,ashg,fatg,prog,H2Oint,kjexp $ 3,8(8.1)
'calc' DMI = Pint/0.384
'for' dum = 1,2,3
'restrict' feed,Ilwt,Idwt,Iash,Ifat,Ipro,lwtg,ewtg,dwtg,ashg,fatg,prog,
         DMI,Pint,kjint,Fint,Cint,kjexp,H2Oint $ state = dum
'treat' feed
'anova' lwtg,dwtg,ashg,fatg,prog,DMI,Pint,kjint,Fint,Cint,kjexp,H2Oint
'covar' Ilwt
'anova' lwtg,dwtg,ashg,fatg,prog
'covar'
'repeat'
'run'
'close'
'stop'
```

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The analysis of variance program from the second rat experiment (in Genstat 5)

```
unit [nval = 42]
fact [leve = !(15,30)] prot
fact [leve = !(10,25,40,55)] fat
fact [leve = 8] treat
read [chan = 2] rat,treat,prot,fat,d2mlwt,d2llwt,dmi,pi,fi,choi,gei,mlwtg,mdwtg,\
    mcpg,meeg,mashg,mgeg,llwtg,ldwtg,lcpg,leeg,lashg,lgeg
outp [width = 132] 1
prin rat,treat,d2mlwt,d2llwt,dmi,pi,fi,choi,gei ; deci = 2(0),7(1)
prin rat,treat,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg ; deci = 2(0),6(1)
prin rat,treat,llwtg,ldwtg,lcpg,leeg,lashg,lgeg ; deci = 2(0),6(1)
model dmi,pi,fi,choi,gei,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg,llwtg,ldwtg,lcpg,\
    leeg,lashg,lgeg,d2mlwt,d2llwt
fit [prin = m,s,e,a] prot + fat + prot.fat
predict prot,fat
model dmi,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg,llwtg
fit [prin = m,s,e,a] d2mlwt + prot + fat + prot.fat
predict d2mlwt,prot,fat; lev = 374.92,!(15,30),!(10,25,40,55)
treat treat
anova dmi,pi,fi,choi,gei,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg,llwtg,ldwtg,lcpg,\
    leeg,lashg,lgeg,d2mlwt,d2llwt
cova d2mlwt
anova dmi,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg,llwtg
cova d2llwt
anova llwtg,ldwtg,lcpg,leeg,lashg,lgeg
cova
rest fat,d2mlwt,dmi,pi,fi,choi,gei,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg,llwtg,\
    ldwtg,lcpg,leeg,lashg,lgeg ; prot.eq.30
treat fat
anova dmi,pi,fi,choi,gei,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg,llwtg,ldwtg,lcpg,\
    leeg,lashg,lgeg
cova d2mlwt
anova dmi,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg
cova d2llwt
anova llwtg,ldwtg,lcpg,leeg,lashg,lgeg
```

```
rest fat,d2mlwt,dmi,pi,fi,choi,gei,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg,llwtg, \
     ldwtg,lcpg,leeg,lashg,lgeg
rest fat,d2mlwt,dmi,pi,fi,choi,gei,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg,llwtg, \
     ldwtg,lcpg,leeg,lashg,lgeg ; prot.eq.15
treat fat
cova
anova dmi,pi,fi,choi,gei,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg,llwtg,ldwtg,lcpg,\
     leeg,lashg,lgeg
cova d2mlwt
anova dmi,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg
cova d2llwt
anova llwtg,ldwtg,lcpg,leeg,lashg,lgeg
close
stop
```

```
rest fat,d2mlwt,dmi,pi,fi,choi,gei,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg,llwtg, \
     ldwtg,lcpg,leeg,lashg,lgeg
rest fat,d2mlwt,dmi,pi,fi,choi,gei,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg,llwtg, \
     ldwtg,lcpg,leeg,lashg,lgeg ; prot.eq.15
treat fat
cova
anova dmi,pi,fi,choi,gei,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg,llwtg,ldwtg,lcpg,\
     leeg,lashg,lgeg
cova d2mlwt
anova dmi,mlwtg,mdwtg,mcpg,meeg,mashg,mgeg
cova d2llwt
anova llwtg,ldwtg,lcpg,leeg,lashg,lgeg
close
stop
```

The analysis of variance of the third rat experiment did not require a program and was analysed in minitab using the ONEWay analysis of variance command.

The analysis of variance program from R4 (in Genstat version 4).

```
'refe' r2anova
'unit' $ 29
'factor' diet $ 6
'input' 2
'read/p' rat,state,diet,DMI,mlwtg,llwtg,mcpg,meeg
    :   mashg,mgeg,lcpg,leeg,lashg,lgeg,d2mlwt,d2llwt
'input' 1
'prin/p' rat,state,diet,DMI,mlwtg,llwtg,mcpg,meeg $ (5)3,(8.2)
    :   mashg,mgeg,lcpg,leeg,lashg,lgeg,d2mlwt,d2llwt $ (8.2)8
'matrix' m $3,6=1,-1,0,0,1,-1,1,1,0,0,-1,-1,1,-1,0,0,-1,1
    :   n $ 3,6=0,1,1,-1,-1,0,0,1,-1,1,-1,0,0,1,-1,-1,1,0
'treat' reg(diet,3,m)
'anova' DMI,mlwtg,llwtg,mcpg,meeg,mashg,mgeg,lcpg,leeg,lashg,lgeg
'cova' d2mlwt
'anova' mlwtg,mcpg,meeg,mashg,mgeg
'cova' d2llwt
'anova' llwtg,lcpg,leeg,lashg,lgeg
'cova'
'treat' reg(diet,3,n)
'anova' DMI,mlwtg,llwtg,mcpg,meeg,mashg,mgeg,lcpg,leeg,lashg,lgeg
'cova' d2mlwt
'anova' mlwtg,mcpg,meeg,mashg,mgeg
'cova' d2llwt
'anova' llwtg,lcpg,leeg,lashg,lgeg
'run'
'close'
'stop'
```

The analysis of variance of the dairy cow trial used three programs (written in Genstat 4). **The first** analyses the latin square in three stages:

1) With no carry-over effect.
2) With a carry-over effect due to the level of feeding in the previous period.
3) The same carry-over effect as in 2), but allowing for an interaction between carry-over effect and feed type

The second program performs the same analysis but with covariates for initial cow performance. **The third** program incorporates a test to see if the response to level of feeding is linear.

The first program

```
'REFE' N327
'UNIT'DAT$ 54 = 1...54
'FACTOR' COW $ 18
    :   FEED $ 3
    :   LEVEL $ 3
    :   PERIOD $ 3
'INPUT' 2
'READ/P' COW,FEED,LEVEL,PERIOD,MY,Yo,k,lnYo,lnk,LWT
    :   MFC,MPC,MLC,MFY,MPY,MLY,MEnY
'INPUT' 1
'CALC'Z = LEVEL
'EQUATE'Y$18X,36 = Z$36,18X
'RESTRICT'Y$PERIOD = 1
'CALC'Y = 0
'RESTRICT'Y
'CALC'C1 = (Y.EQ.3)-(Y.EQ.1)
    :C2 = 2*(Y.EQ.2)-(Y.EQ.1)-(Y.EQ.3)
    :C11,C21,C31,C12,C22,C32 = 0
'FOR'I = 1,2,3;CC = C11,C21,C31;CCC = C12,C22,C32
'RESTRICT'CC,CCC$FEED = I
'CALC'CC = C1
    :CCC = C2
```

'REPEAT'
'RESTRICT'C11,C21,C31,C12,C22,C32
'PRINT/P' COW ,MY,Yo,k,lnYo,lnk,LWT $9.5
    :   COW,MFC,MPC,MLC,MFY,MPY,MLY,MEnY $9.5
'PRINT/FORM = P'COW,FEED,LEVEL,PERIOD,Y,C1,C2,C11,C21,C31,C12,C22,C3
2$6
'BLOCK'COW*PERIOD
'TREAT' FEED*LEVEL
'ANOVA' MY,Yo,k,lnYo,lnk,LWT,MFC,MPC,MLC,MFY,MPY,MLY,MEnY
'COVAR' C1,C2
'ANOVA' MY,Yo,k,lnYo,lnk,LWT,MFC,MPC,MLC,MFY,MPY,MLY,MEnY
'COVAR'C11,C21,C31,C12,C22,C32
'ANOVA/PRX = 0,PRYU = 0'MY,Yo,k,lnYo,lnk,LWT,MFC,MPC,MLC,MFY,MPY,M
LY,MEnY
'RUN'
'CLOSE'
'STOP'

The second program

```
'REFE' N327
'UNIT'DAT$ 54 = 1...54
'FACTOR' COW $ 18
    :   FEED $ 3
    :   LEVEL $ 3
    :   PERIOD $ 3
'INPUT' 2
'READ/P' COW,FEED,LEVEL,PERIOD,MY,Yo,k,lnYo,lnk,LWT
    :   MFC,MPC,MLC,MFY,MPY,MLY,MEnY
    :   IMY,IMFC,IMPC,IMLC,IMFY,IMPY,IMLY,IMEnY,ILWT
'INPUT' 1
'CALC'Z = LEVEL
'EQUATE'Y$18X,36 = Z$36,18X
'RESTRICT'Y$PERIOD = 1
'CALC'Y = 0
'RESTRICT'Y
'CALC'C1 = (Y.EQ.3)-(Y.EQ.1)
    :C2 = 2*(Y.EQ.2)-(Y.EQ.1)-(Y.EQ.3)
    :C11,C21,C31,C12,C22,C32 = 0
'FOR'I = 1,2,3;CC = C11,C21,C31;CCC = C12,C22,C32
'RESTRICT'CC,CCC$FEED = I
'CALC'CC = C1
    :CCC = C2
'REPEAT'
'RESTRICT'C11,C21,C31,C12,C22,C32
'PRINT/P' COW ,MY,Yo,k,lnYo,lnk,LWT $9.5
    :    COW,MFC,MPC,MLC,MFY,MPY,MLY,MEnY $9.5
    :   IMY,IMFC,IMPC,IMLC,IMFY,IMPY,IMLY,IMEnY,ILWT $9.5
'PRINT/FORM = P'COW,FEED,LEVEL,PERIOD,Y,C1,C2,C11,C21,C31,C12,C22,C3
2$6
'BLOCK'COW*PERIOD
'TREAT' FEED*LEVEL
'COVAR' IMY
'ANOVA' MY,Yo,k,lnYo,lnk,LWT,MFC,MPC,MLC,MFY,MPY,MLY,MEnY
'COVAR' C1,C2,IMY,
```

'ANOVA' MY,Yo,k,lnYo,lnk,LWT,MFC,MPC,MLC,MFY,MPY,MLY,MEnY
'COVAR'C11,C21,C31,C12,C22,C32,IMY
'ANOVA/PRX=0,PRYU=0'MY,Yo,k,lnYo,lnk,LWT,MFC,MPC,MLC,MFY,MPY,M
LY,MEnY
'COVAR' IMFC
'ANOVA' MFC,MFY,MEnY
'COVAR' C1,C2,IMFC
'ANOVA' MFC,MFY,MEnY
'COVAR'C11,C21,C31,C12,C22,C32,IMFC
'ANOVA/PRX=0,PRYU=0'MFC,MFY,MEnY
'COVAR' IMPC
'ANOVA' MPC,MPY,MEnY
'COVAR' C1,C2,IMPC
'ANOVA' MPC,MPY,MEnY
'COVAR'C11,C21,C31,C12,C22,C32,IMPC
'ANOVA/PRX=0,PRYU=0'MPC,MPY,MEnY
'COVAR' IMLC
'ANOVA' MLC,MLY,MEnY
'COVAR' C1,C2,IMLC
'ANOVA' MLC,MLY,MEnY
'COVAR'C11,C21,C31,C12,C22,C32,IMLC
'ANOVA/PRX=0,PRYU=0'MLC,MLY,MEnY
'COVAR' IMFY
'ANOVA' MFY,MEnY
'COVAR' C1,C2,IMFY
'ANOVA' MFY,MEnY
'COVAR'C11,C21,C31,C12,C22,C32,IMFY
'ANOVA/PRX=0,PRYU=0'MFY,MEnY
'COVAR' IMPY
'ANOVA' MPY,MEnY
'COVAR' C1,C2,IMPY
'ANOVA' MPY,MEnY
'COVAR'C11,C21,C31,C12,C22,C32,IMPY
'ANOVA/PRX=0,PRYU=0'MPY,MEnY
'COVAR' IMLY
'ANOVA' MLY,MEnY
'COVAR' C1,C2,IMLY

```
'ANOVA' MLY,MEnY
'COVAR'C11,C21,C31,C12,C22,C32,IMLY
'ANOVA/PRX=0,PRYU=0'MLY,MEnY
'COVAR' IMEnY
'ANOVA' MEnY
'COVAR' C1,C2,IMEnY
'ANOVA' MEnY
'COVAR'C11,C21,C31,C12,C22,C32,IMEnY
'ANOVA/PRX=0,PRYU=0' MEnY
'COVAR' ILWT
'ANOVA' MY,LWT,MFY,MPY,MLY,MEnY
'COVAR' C1,C2,ILWT
'ANOVA' MY,LWT,MFY,MPY,MLY,MEnY
'COVAR'C11,C21,C31,C12,C22,C32,ILWT
'ANOVA/PRX=0,PRYU=0'MY,LWT,MFY,MPY,MLY,MEnY
'RUN'
'CLOSE'
'STOP'
```

The third program

```
'REFE' N327
'UNIT' $ 54
'FACTOR'  COW $ 18
    :    FEED $ 3
    :    LEVEL $ 3
    :    PERIOD $ 3
'INPUT'2
'READ/P' COW,FEED,LEVEL,PERIOD,MY,Yo,k,lnYo,lnk,LWT
    :    MFC,MPC,MLC,MFY,MPY,MLY,MEnY
'INPUT' 1
'INTEGER'I = 1
'CALC'CLEVEL = 0
'RESTRICT'CLEVEL$PERIOD,LEVEL = 3,1
'CALC'CLEVEL = -1
'RESTRICT'CLEVEL$PERIOD,LEVEL = 3,3
'CALC'CLEVEL = 1
'RESTRICT'CLEVEL$PERIOD,LEVEL = I,1
'CALC'CLEVEL = 1
'RESTRICT'CLEVEL$PERIOD,LEVEL = I,3
'CALC'CLEVEL = -1
'RESTRICT'CLEVEL
'PRINT/FORM = P'COW,MY,Yo,k,lnYo,lnk,LWT$9.4
    :       COW,MFC,MPC,MLC,MFY,MPY,MLY,MEnY$9.4
    :       COW,FEED,LEVEL,PERIOD,CLEVEL$8
'BLOCK'COW*PERIOD
'TREAT' FEED*LEVEL
'COVAR'CLEVEL
'ANOVA/PRX=0,PRYU=0'MY,Yo,k,lnYo,lnk,LWT,MFC,MPC,MLC,MFY,MPY,M
LY,MEnY
'RUN'
'CLOSE'
'STOP'
```

Appendix 2

The data from the first rat experiment (R1)

Unless otherwise indicated, rats are identified by a two letter code which gives their feeding treatment and state and by a rat number, as follows:

L = low fat feed, H = high fat feed.
P = lactating,   N = non-pregnant non-lactating.

Food intake data (gDM)

<table>
<tr><th>ROW
HP41</th><th>day no</th><th>LP12</th><th>HP13</th><th>HN14</th><th>LN15</th><th>LP21</th><th>LP22</th><th>LN23</th><th>HP25</th><th>HP31</th><th>LN32</th><th>HN35</th><th>HP41</th></tr>
<tr><td>1</td><td>0</td><td>8.4</td><td>*</td><td>*</td><td>*</td><td>13.4</td><td>13.5</td><td>*</td><td>11.7</td><td>*</td><td>*</td><td>*</td><td>*</td></tr>
<tr><td>2</td><td>1</td><td>*</td><td>*</td><td>13.3</td><td>14.5</td><td>15.3</td><td>11.1</td><td>2.0</td><td>14.4</td><td>14.1</td><td>7.6</td><td>11.5</td><td>*</td></tr>
<tr><td>3</td><td>2</td><td>*</td><td>10.6</td><td>6.0</td><td>6.3</td><td>*</td><td>*</td><td>11.6</td><td>15.5</td><td>18.1</td><td>*</td><td>4.5</td><td>10.9</td></tr>
<tr><td>4</td><td>3</td><td>25.7</td><td>*</td><td>13.9</td><td>14.4</td><td>9.8</td><td>27.4</td><td>13.8</td><td>21.6</td><td>19.7</td><td>11.7</td><td>8.5</td><td>14.5</td></tr>
<tr><td>5</td><td>4</td><td>27.1</td><td>7.3</td><td>11.5</td><td>13.7</td><td>18.1</td><td>27.6</td><td>15.2</td><td>24.7</td><td>20.7</td><td>11.6</td><td>9.8</td><td>12.8</td></tr>
<tr><td>6</td><td>5</td><td>35.3</td><td>13.6</td><td>13.2</td><td>18.5</td><td>42.8</td><td>42.6</td><td>21.1</td><td>28.7</td><td>17.9</td><td>14.4</td><td>8.1</td><td>16.0</td></tr>
<tr><td>7</td><td>6</td><td>38.8</td><td>16.8</td><td>17.6</td><td>13.7</td><td>*</td><td>36.1</td><td>16.9</td><td>30.6</td><td>25.5</td><td>11.3</td><td>10.4</td><td>22.4</td></tr>
<tr><td>8</td><td>7</td><td>19.4</td><td>17.2</td><td>11.3</td><td>32.5</td><td>37.7</td><td>44.3</td><td>16.2</td><td>*</td><td>26.6</td><td>8.9</td><td>9.4</td><td>24.1</td></tr>
<tr><td>9</td><td>8</td><td>41.6</td><td>17.6</td><td>14.8</td><td>6.6</td><td>32.2</td><td>47.1</td><td>15.5</td><td>31.8</td><td>28.8</td><td>11.2</td><td>12.9</td><td>21.0</td></tr>
<tr><td>10</td><td>9</td><td>32.4</td><td>21.4</td><td>14.1</td><td>9.6</td><td>47.4</td><td>47.7</td><td>9.3</td><td>30.8</td><td>26.0</td><td>9.8</td><td>10.6</td><td>28.7</td></tr>
<tr><td>11</td><td>10</td><td>44.0</td><td>19.9</td><td>11.4</td><td>18.1</td><td>44.1</td><td>30.4</td><td>12.5</td><td>36.9</td><td>17.3</td><td>10.7</td><td>11.1</td><td>27.9</td></tr>
<tr><td>12</td><td>11</td><td>45.8</td><td>27.5</td><td>11.8</td><td>10.6</td><td>49.3</td><td>41.2</td><td>*</td><td>37.4</td><td>32.5</td><td>11.2</td><td>11.7</td><td>43.3</td></tr>
<tr><td>13</td><td>12</td><td>34.7</td><td>23.0</td><td>15.8</td><td>10.0</td><td>46.6</td><td>35.6</td><td>11.6</td><td>28.8</td><td>23.0</td><td>11.8</td><td>12.8</td><td>26.1</td></tr>
<tr><td>14</td><td>13</td><td>45.1</td><td>20.4</td><td>14.8</td><td>12.4</td><td>49.3</td><td>44.9</td><td>13.4</td><td>32.7</td><td>26.4</td><td>14.7</td><td>9.8</td><td>33.1</td></tr>
<tr><td>15</td><td>14</td><td>48.1</td><td>31.4</td><td>13.5</td><td>12.0</td><td>53.5</td><td>47.5</td><td>12.2</td><td>40.2</td><td>27.6</td><td>12.8</td><td>8.1</td><td>34.3</td></tr>
<tr><td>16</td><td>15</td><td>48.8</td><td>36.6</td><td>10.1</td><td>11.7</td><td>49.1</td><td>43.3</td><td>9.7</td><td>27.3</td><td>20.6</td><td>15.2</td><td>10.3</td><td>36.5</td></tr>
<tr><td>17</td><td>16</td><td>*</td><td>40.5</td><td>*</td><td>*</td><td>*</td><td>*</td><td>*</td><td>*</td><td>*</td><td>*</td><td>*</td><td>42.6</td></tr>
<tr><td>18</td><td>17</td><td>*</td><td>32.1</td><td>*</td><td>*</td><td>*</td><td>*</td><td>*</td><td>*</td><td>*</td><td>*</td><td>*</td><td>31.5</td></tr>
</table>

<table>
<tr><th>ROW</th><th>HN42</th><th>LN43</th><th>LN52</th><th>HN53</th><th>HN54</th><th>LN55</th></tr>
<tr><td>1</td><td>*</td><td>*</td><td>*</td><td>*</td><td>*</td><td>*</td></tr>
<tr><td>2</td><td>7.2</td><td>14.9</td><td>14.6</td><td>9.5</td><td>11.1</td><td>19.5</td></tr>
<tr><td>3</td><td>6.7</td><td>*</td><td>*</td><td>5.4</td><td>3.3</td><td>*</td></tr>
<tr><td>4</td><td>7.8</td><td>10.5</td><td>14.1</td><td>8.4</td><td>7.0</td><td>8.8</td></tr>
<tr><td>5</td><td>5.8</td><td>14.8</td><td>13.8</td><td>11.1</td><td>9.0</td><td>16.2</td></tr>
<tr><td>6</td><td>7.3</td><td>17.6</td><td>15.5</td><td>8.0</td><td>11.1</td><td>18.5</td></tr>
<tr><td>7</td><td>11.8</td><td>16.6</td><td>14.2</td><td>11.4</td><td>9.8</td><td>22.6</td></tr>
<tr><td>8</td><td>10.0</td><td>13.4</td><td>11.6</td><td>10.7</td><td>6.5</td><td>10.9</td></tr>
<tr><td>9</td><td>10.1</td><td>16.2</td><td>19.3</td><td>12.1</td><td>13.5</td><td>18.3</td></tr>
<tr><td>10</td><td>10.7</td><td>14.6</td><td>15.8</td><td>8.7</td><td>11.7</td><td>17.0</td></tr>
</table>

11	9.8	15.2	16.5	10.2	10.1	18.3
12	8.9	15.3	*	13.3	7.1	19.4
13	9.5	12.7	15.0	13.1	10.3	16.7
14	10.1	15.1	17.3	11.3	11.5	16.3
15	11.5	15.1	15.3	11.3	10.3	16.2
16	9.4	15.9	14.0	14.7	9.5	15.5
17	*	*	*	*	*	*
18	*	*	*	*	*	*

Maternal liveweight data (g)

day is days from birth

ROW	Day no	LP12	HP13	HN14	LN15	LP21	LP22	LN23	HP25	HP31	LN32
18	-10	*	261.0	*	*	*	*	*	*	*	*
19	-9	291.0	*	206.0	265.0	310.0	323.0	237.0	357.0	254.0	240.5
20	-8	*	*	*	*	*	*	*	*	*	*
21	-7	301.5	275.0	209.0	263.0	324.0	356.0	234.0	383.5	275.0	235.0
22	-6	*	*	*	*	*	*	*	*	*	*
23	-5	321.5	289.0	211.0	262.0	345.0	394.0	241.0	407.0	292.0	225.0
24	-4	*	*	*	*	*	*	*	*	*	*
25	-3	339.5	314.0	211.0	269.0	368.5	423.0	260.0	445.5	316.0	228.5
26	-2	*	*	*	*	*	*	*	*	*	*
27	-1	*	341.0	*	*	*	*	*	*	*	*
28	0	259.0	*	*	*	296.0	349.0	267.0	335.0	*	*
29	1	260.0	*	204.0	269.0	289.0	345.0	263.0	329.0	226.0	237.0
30	2	254.0	270.0	216.0	276.0	283.0	345.0	266.0	337.0	229.0	238.0
31	3	239.0	257.0	211.0	272.0	270.0	331.0	267.0	327.0	233.5	234.0
32	4	251.0	258.0	220.0	276.0	269.0	331.0	265.0	329.0	232.0	240.0
33	5	257.0	246.5	224.0	281.0	274.0	329.0	266.0	340.0	246.0	240.0
34	6	*	244.0	*	*	*	*	*	*	*	*
35	7	275.5	247.0	231.0	282.0	279.0	337.5	271.0	341.0	239.0	234.0
36	8	248.0	*	236.0	283.0	275.0	345.0	276.0	337.0	244.0	236.0
37	9	270.0	238.0	236.0	283.0	265.0	343.0	273.0	341.0	253.0	237.0
38	10	286.0	237.0	237.0	281.0	280.0	342.0	264.5	338.0	246.0	233.0
39	11	273.0	236.0	241.0	280.0	291.5	338.0	268.0	349.0	232.0	233.0
40	12	277.0	241.5	241.0	282.0	286.0	333.0	268.0	343.0	250.5	236.0
41	13	284.0	244.0	249.0	286.0	301.0	331.0	272.0	343.0	250.0	242.0
42	14	272.0	231.0	253.5	288.0	294.0	337.0	273.5	337.0	247.5	247.5
43	15	260.0	252.0	252.5	282.5	279.0	324.0	274.0	338.5	246.0	248.0
44	16	261.5	259.0	259.9	290.7	304.2	336.6	280.5	338.5	250.3	257.4
45	17	*	265.0	*	*	*	*	*	*	*	*
46	18	*	266.2	*	*	*	*	*	*	*	*

ROW	HN35	HP41	HN42	LN43	LN52	HN53	HN54	LN55
18	*	283.0	*	*	*	*	*	*
19	270.5	*	221.0	250.0	241.5	230.0	245.0	234.0
20	*	*	*	*	*	*	*	*
21	272.0	297.5	228.0	258.0	252.0	241.0	244.0	241.5
22	*	*	*	*	*	*	*	*
23	271.0	322.0	228.0	261.5	262.5	250.0	260.5	245.0
24	*	*	*	*	*	*	*	*
25	282.0	344.0	223.0	265.0	267.5	260.0	280.0	249.0
26	*	*	*	*	*	*	*	*
27	*	390.0	*	*	*	*	*	*
28	*	*	*	*	*	*	287.0	*
29	277.0	*	239.0	274.0	274.0	254.0	284.0	250.0
30	281.0	311.0	239.0	271.0	272.0	253.0	285.0	252.0
31	278.0	295.0	236.0	261.0	269.0	249.0	271.0	238.0
32	276.5	287.0	240.0	263.5	269.0	253.5	275.5	237.0
33	276.5	290.0	238.0	267.0	272.0	255.5	277.5	246.0

34	*	285.5	*	*	*	*	*	*
35	280.0	295.0	243.0	270.0	272.0	254.0	279.0	254.0
36	280.5	*	243.5	266.0	271.0	255.0	279.5	249.0
37	281.5	290.0	246.0	269.0	274.0	255.0	286.5	252.0
38	281.0	292.0	244.0	268.0	273.0	251.0	285.0	249.0
39	281.0	298.0	247.0	269.0	275.0	255.0	288.0	253.0
40	283.0	312.5	248.5	269.0	279.0	256.5	286.5	257.5
41	291.0	300.0	253.0	271.5	282.0	261.0	291.0	264.0
42	291.0	300.0	257.0	276.0	284.5	260.0	292.0	264.5
43	286.0	302.0	259.0	275.0	281.5	263.0	290.0	262.0
44	296.1	305.0	268.1	282.5	291.1	272.7	299.7	271.7
45	*	302.0	*	*	*	*	*	*
46	* -	319.8	*	*	*	*	*	*

Litter liveweights (g)

ROW	day	LP12	HP13	LP21	LP22	HP25	HP31	HP41
1	0	67.6	*	78.8	64.6	54.7	*	*
2	1	79.6	*	93.5	77.0	71.3	50.8	*
3	2	84.5	71.5	97.9	89.4	68.0	60.6	65.4
4	3	89.0	86.9	103.5	102.7	82.0	67.8	76.5
5	4	101.6	94.2	113.1	120.3	99.7	83.2	90.4
6	5	113.3	112.1	126.3	134.4	112.0	91.4	92.2
7	6	*	132.4	*	*	*	*	109.0
8	7	154.1	147.5	160.0	175.0	157.2	123.1	123.2
9	8	164.3	*	180.0	196.6	184.8	*	*
10	9	179.2	188.5	199.9	218.1	210.8	164.7	157.9
11	10	189.7	208.6	221.7	241.6	229.9	188.1	*
12	11	209.5	225.4	243.6	256.5	257.4	198.5	198.5
13	12	226.9	248.4	267.4	273.7	283.5	218.7	219.5
14	13	241.9	263.1	282.5	287.5	298.4	238.8	238.8
15	14	259.5	282.3	305.7	301.4	323.8	255.4	259.6
16	15	276.7	304.3	330.9	320.2	346.0	270.6	280.7
17	16	297.8	325.8	348.5	334.6	360.4	280.0	299.7
18	17	*	352.4	*	*	*	*	320.7
19	18	*	375.1	*	*	*	*	336.3

Carcass composition data

rat no   negative numbers are the litters of the dam with the same positive number
feed: 1 = control culls; 2 = low fat;   3 = high fat
state: 1 = NPNL; 2 = lactating; 3 = litters
I.Lwt = liveweight on day 2 of lactation (g)
F.Ewt = liveweight minus the weight of gut contents (g) on day 16 of lactation
DM% = dry matter of the carcass
Ash%DM = ash % in the dry matter
Fat%DM = Fat % in the dry matter
Pro%DM = protein % in the dry matter

ROW	rat no	feed	state	I.Lwt	F.Ewt	DM%	Ash%DM	Fat%DM	Pro%DM
1	45	1	1	231.7	222.2	44.079	8.729	45.20	42.0000
2	34	1	1	289.9	263.9	40.219	10.029	38.56	48.0250
3	51	1	1	213.0	205.3	38.874	10.833	34.80	51.5375
4	32	2	1	237.0	248.3	37.344	11.288	31.16	53.5000
5	15	2	1	269.0	279.5	41.403	9.928	39.12	48.3437
6	23	2	1	263.0	272.8	43.615	8.844	46.88	41.5562
7	55	2	1	250.0	261.2	40.560	11.248	30.16	53.1125
8	43	2	1	274.0	270.0	39.352	10.636	34.40	51.3312
9	52	2	1	274.0	278.3	42.231	9.410	41.92	45.6312
10	35	3	1	277.0	287.1	45.228	9.073	38.72	44.2562
11	42	3	1	239.0	256.8	37.232	12.083	28.36	56.4250
12	53	3	1	254.0	261.3	41.844	8.921	42.00	45.8562
13	54	3	1	284.0	293.8	38.005	11.970	30.56	51.5187
14	14	3	1	204.0	247.7	42.277	9.305	43.40	43.3562
15	44	1	2	221.5	213.9	37.036	9.695	43.12	43.7062
16	33	1	2	299.0	275.7	41.277	9.299	41.24	46.6625
17	24	1	2	278.3	278.3	40.788	10.112	40.88	46.1125
18	22	2	2	349.0	301.6	32.481	10.497	19.12	65.3812
19	21	2	2	296.0	270.1	33.411	13.268	17.48	59.5375
20	12	2	2	259.0	250.5	36.579	11.181	21.16	60.5875
21	31	3	2	219.0	228.8	32.000	13.078	17.88	64.1062
22	13	3	2	270.0	242.1	36.223	10.291	26.84	57.8375
23	41	3	2	311.0	291.9	37.021	9.184	33.40	53.2062
24	25	3	2	335.0	311.6	33.796	10.249	22.08	62.5812
25	-44	1	3	48.2	48.2	18.908	10.708	16.05	62.3562
26	-24	1	3	56.6	56.6	21.139	10.367	19.66	58.8750
27	-22	2	3	58.1	325.9	29.899	9.783	25.64	59.4062
28	-21	2	3	69.4	339.0	30.828	9.633	28.92	56.0000
29	-12	2	3	67.6	288.0	28.147	10.828	19.92	61.4562
30	-31	3	3	33.5	265.7	31.777	8.658	39.68	47.2000
31	-41	3	3	56.4	328.3	34.593	8.403	36.52	48.0687
32	-13	3	3	71.5	366.0	37.119	7.285	45.88	41.2062
33	-25	3	3	47.6	337.1	32.163	8.462	43.08	44.6437

Water intake data (g/day)

ROW	day no	LP12	HP13	HN14	LN15	LP21	LP22	LN23	HP25	HP31	day
1	0	26.8	*	*	*	30.5	25.7	*	33.8	*	0
2	1	29.5	*	35.8	13.3	19.9	17.0	12.9	42.3	43.1	1
3	2	28.7	32.3	24.7	16.4	26.1	20.4	11.3	36.9	42.8	2
4	3	30.8	49.6	29.5	9.5	30.3	27.5	12.8	58.3	40.5	3
5	4	36.8	25.0	25.9	8.6	33.3	20.6	9.7	66.2	57.1	4
6	5	35.2	47.6	30.5	10.7	44.1	38.4	13.1	26.7	52.5	5
7	6	23.0	42.2	30.6	11.2	38.7	35.2	12.7	62.6	60.9	6
8	7	16.6	53.4	27.4	13.3	41.6	47.9	17.7	84.9	71.7	7
9	8	30.4	56.0	30.8	15.3	33.5	44.4	12.0	65.7	68.9	8
10	9	30.8	68.0	35.8	14.7	39.7	53.0	15.7	*	*	9
11	10	29.1	43.2	26.4	8.5	32.0	24.7	12.4	70.9	31.2	10
12	11	48.4	77.5	24.7	14.2	44.5	41.6	13.3	94.6	72.0	11
13	12	40.6	43.0	26.8	12.0	41.2	39.6	9.9	60.2	52.2	12
14	13	70.4	75.1	32.1	12.8	53.9	69.8	15.5	73.3	54.2	13
15	14	57.9	70.3	29.3	11.9	57.6	84.4	19.7	107.6	61.4	14
16	15	62.9	82.5	21.4	11.7	57.7	75.3	18.7	66.4	48.5	15
17	16	*	106.0	*	*	*	*	*	*	*	16
18	17	*	88.1	*	*	*	*	*	*	*	17

ROW	LN32	HN35	HP41	HN42	LN43	LN52	HN53	HN54
1	*	*	*	*	*	*	*	*
2	12.0	32.1	*	21.7	10.3	8.5	13.9	21.1
3	11.7	19.9	22.9	14.8	15.7	14.6	13.9	16.0
4	11.1	26.8	39.8	22.8	27.7	11.3	20.5	16.6
5	6.9	20.4	32.5	14.6	14.7	13.5	18.2	21.2
6	14.0	23.3	40.5	24.8	17.3	11.4	18.9	20.3
7	12.5	26.8	57.6	21.9	16.2	9.7	22.9	15.1
8	18.6	31.2	62.1	32.1	20.6	21.0	21.6	23.8
9	12.1	19.5	54.1	23.0	19.1	12.3	19.9	22.2
10	12.7	27.0	77.2	27.1	22.5	16.9	21.8	26.4
11	8.5	19.4	55.5	25.6	15.0	12.9	21.4	18.8
12	13.7	22.7	95.6	25.3	18.6	14.4	26.1	22.5
13	14.9	26.7	41.0	29.9	16.6	11.9	23.2	17.2
14	14.1	24.5	92.3	32.8	21.5	9.5	20.3	20.4
15	13.4	19.0	63.8	29.1	17.7	8.7	26.0	17.4
16	13.1	25.3	88.9	27.1	21.2	10.7	23.3	21.4
17	*	*	86.3	*	*	*	*	*
18	*	*	83.3	*	*	*	*	*

Appendix 3

The data from the second rat experiment (R2)

The data for the regressions of body composition on initial liveweight

The allocation of rats to treatments is given with the litter liveweight data

Maternal Intake

DMI = dry matter intake (g); d = day

ROW	rat no	DMI d2	DMI d3	DMI d4	DMI d5	DMI d6	DMI d7	DMI d8	DMI d9
1	3	21.4862	16.5519	22.6105	27.4824	22.7979	36.2268	39.1000	*
2	5	21.9235	27.2950	25.9834	27.2950	30.2306	34.4155	33.6659	45.4709
3	6	23.3168	28.1580	27.2194	37.6428	35.1234	42.5828	41.6442	52.6604
4	8	14.4217	16.2810	23.5672	21.5572	32.0595	35.8283	36.8835	41.3558
5	9	18.5201	29.4828	34.9875	27.6168	32.3285	39.3260	43.2912	49.7756
6	12	8.8193	9.0577	4.8268	6.1974	5.7206	5.0056	5.2439	2.1452
7	14	22.2608	28.7430	31.1048	33.4162	35.4765	43.2653	37.8885	40.8533
8	15	33.7096	28.4298	*	40.6140	8.6000	3.6860	*	11.3740
9	19	25.2484	8.5289	20.8485	32.5589	34.7927	33.1004	32.3558	29.1067
10	20	*	*	*	*	*	*	*	*
11	22	8.7900	19.2983	10.7066	16.1260	11.8301	8.2612	6.0803	12.1606
12	25	18.5270	16.9171	18.4378	28.2269	27.7042	30.3178	29.9376	30.4128
13	26	*	*	*	*	*	*	*	*
14	28	17.1834	14.5398	15.1346	7.0055	3.6349	3.3706	2.3131	6.3446
15	29	27.1701	32.1669	42.6602	34.0407	37.7883	41.0362	36.9763	31.2925
16	31	7.6275	9.6536	2.6816	1.5493	1.8473	1.1918	1.4302	0.9534
17	33	24.0578	29.0966	21.5878	37.6428	29.4918	37.7910	45.2504	55.8714
18	34	17.7500	18.9605	21.4315	23.1422	31.4107	35.2123	33.1690	38.5387
19	35	23.6982	26.0307	31.3955	28.9230	35.2208	35.6406	44.5508	37.4599
20	36	*	*	*	*	*	*	*	*
21	37	20.5101	20.9162	14.7564	23.8946	*	*	12.6580	*
22	38	25.9834	34.9776	34.4155	23.6099	38.2255	*	13.1560	16.8910
23	39	22.4276	24.6506	29.9858	28.4050	33.5920	33.5426	38.3344	32.6040
24	40	26.3302	24.9470	31.2702	37.9886	41.1996	40.4586	39.6682	40.8044
25	42	13.2290	4.7076	4.4693	3.3370	0.8343	3.2775	1.4302	2.0856
26	43	23.1727	29.9183	36.9139	32.6666	31.7297	42.9725	36.9763	36.6016
27	44	*	*	*	*	*	*	*	*
28	45	17.8670	6.6243	11.5692	5.0849	*	20.1995	*	22.4853
29	47	*	*	*	*	*	*	*	*
30	48	20.0790	9.1865	3.1062	3.3706	2.9741	1.9166	2.4453	1.6523
31	49	11.1012	11.3042	9.0705	7.8520	9.0705	25.9930	22.7438	26.7376
32	50	*	*	*	*	*	*	*	*
33	51	5.4823	7.5679	5.4227	3.2179	4.7076	2.3240	1.4898	1.7281
34	56	20.0497	29.4465	26.4817	33.2655	39.0443	38.9940	41.3055	40.1497
35	57	30.6774	36.6054	32.6534	37.6428	38.5320	51.7218	51.5736	53.3520
36	58	13.9286	24.6092	29.1064	21.6736	20.9866	19.4251	4.5596	11.2428
37	61	19.2060	7.9969	4.6263	6.8073	3.1062	2.9741	2.9080	3.7671
38	63	9.5344	12.7523	2.6816	0.7151	1.3706	2.5624	1.3110	1.1918
39	64	17.8675	24.3302	28.3219	28.0843	31.8384	25.7083	22.1918	33.6917
40	65	*	*	*	*	*	*	*	*
41	66	22.3377	25.3161	33.4389	26.4668	32.2881	36.4172	46.9769	37.9741
42	67	13.4824	11.5658	4.3619	4.2958	1.7183	1.3218	2.1149	1.0574
43	68	21.3190	27.8034	29.7627	28.5498	32.1885	32.5617	37.8798	31.4421
44	69	*	18.6930	32.9137	35.7278	31.1048	39.2453	38.7930	36.6825
45	70	12.6927	5.0056	3.4562	0.4171	1.1918	0.8939	2.0261	0.8939
46	71	16.6795	23.3798	29.3674	29.1773	29.0347	30.5078	34.0243	23.0472
47	72	6.9379	17.2022	18.2952	27.0389	26.4211	27.0864	29.5574	31.3157
48	73	23.1384	28.6431	31.3955	29.9027	36.0605	35.9672	36.6203	41.3786
49	74	*	26.0338	31.3690	35.5680	44.3118	44.0648	49.6470	47.6216

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ROW	DMI d10	DMI d11	DMI d12	DMI d13	Cumulative DMI
1	*	51.6410	46.9699	53.0430	337.910
2	29.6685	39.2873	44.7214	42.4728	402.430
3	52.7592	48.9554	44.3118	54.9822	489.356
4	45.7275	46.2803	47.8883	49.0942	410.944
5	52.2480	48.8425	54.1606	57.4728	488.052
6	2.7411	3.6946	*	*	53.452
7	43.7175	50.1998	42.5618	49.7475	459.235
8	22.3320	40.2850	43.9300	47.0130	279.973
9	29.0020	*	61.3271	35.1700	342.039
10	*	*	*	*	214.1
11	13.1519	14.6059	14.0111	20.8183	155.840
12	32.2661	34.7846	40.3445	38.6338	346.510
13	*	*	*	*	225.9
14	4.3619	4.8246	*	*	78.713
15	35.4773	35.9145	50.0929	47.1573	451.773
16	1.0726	*	*	*	28.007
17	57.6004	53.8954	56.5630	65.6032	514.452
18	28.9872	25.3282	36.1627	46.4746	356.568
19	42.6381	38.2997	41.6585	42.3582	427.874
20	*	*	*	*	196.1
21	7.5810	23.6070	26.3780	15.5040	165.805
22	12.5790	41.7857	46.0900	60.0320	347.746
23	35.6668	38.1862	46.2384	41.4960	405.129
24	48.1156	44.4106	47.4240	54.6858	477.303
25	0.7151	1.1322	1.4898	*	37.899
26	39.3498	39.1000	57.4632	54.2153	461.080
27	*	*	*	*	*
28	*	36.7136	38.0664	48.8892	207.499
29	*	*	*	*	156.0
30	*	*	*	*	44.731
31	27.5498	28.0237	35.6726	35.9434	251.062
32	*	*	*	*	153.0
33	*	*	*	*	31.940
34	45.7778	44.5717	51.3052	52.5615	462.953
35	49.2024	56.7606	46.9794	51.8206	537.521
36	6.4334	5.1842	18.5506	26.9203	202.620
37	*	*	*	*	51.392
38	1.6089	1.7877	*	*	35.516
39	31.7434	28.9397	28.3694	45.5717	346.658
40	*	*	*	*	225.0
41	44.4046	42.7801	44.7431	48.1276	441.271
42	*	*	*	*	39.918
43	36.3870	31.3955	37.5532	38.0664	384.909
44	38.5920	43.8683	40.6020	41.9085	398.131
45	1.4898	0.8343	0.8343	*	29.735
46	38.0160	35.3549	34.6421	27.8942	351.125
47	26.0410	31.3157	31.9334	38.2061	311.351
48	43.9910	43.8510	39.1860	47.6763	437.810
49	52.6110	54.2906	49.7458	62.9356	498.199

Maternal liveweight (g)

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ROW	rat no	d1	d2	d3	d4	d5	d6	d7	d8	d9	d10
1	3	*	349.5	345.2	335.0	336.2	330.2	320.0	325.6	321.8	321.0
2	5	*	360.5	353.5	347.0	358.0	349.0	350.0	343.0	343.3	341.5
3	6	389.4	391.0	388.6	385.5	377.0	381.0	374.5	384.2	369.2	376.8
4	8	371.0	367.0	369.0	362.0	363.0	356.0	358.6	363.5	371.6	372.3
5	9	*	415.5	407.0	412.4	411.6	391.2	382.0	386.5	385.6	402.0
6	12	310.6	311.1	300.0	289.2	280.0	272.0	260.2	252.8	242.0	230.5
7	14	381.0	381.8	375.0	380.7	378.5	376.5	374.0	380.9	368.8	360.8
8	15	425.0	439.4	431.0	411.4	400.0	379.5	361.0	353.8	364.2	335.5
9	19 -	325.2	344.0	341.0	316.4	314.8	314.3	316.7	318.8	313.5	304.1
10	20	375.9	371.7	*	*	*	*	*	*	*	*
11	22	396.0	390.4	373.5	366.5	350.9	350.1	333.0	312.0	296.4	292.9
12	25	415.0	415.3	404.5	398.4	396.0	406.7	404.2	408.6	401.8	399.4
13	26	333.2	334.3	*	*	*	*	*	*	*	*
14	28	360.0	358.5	354.0	343.0	336.9	327.8	315.7	298.0	284.6	276.7
15	29	360.0	356.5	358.2	354.7	360.5	355.8	357.3	357.8	346.7	339.4
16	31	363.2	361.7	346.3	336.0	318.7	307.5	290.4	278.3	266.2	256.0
17	33	391.0	387.7	388.6	385.0	375.4	372.7	360.7	361.7	362.0	360.0
18	34	407.7	397.9	390.5	390.1	380.5	376.0	383.0	401.3	390.0	397.3
19	35	397.2	400.2	404.2	396.8	398.1	392.8	396.7	387.7	*	388.3
20	36	295.9	277.8	*	*	*	*	*	*	*	*
21	37	379.5	374.1	373.5	358.5	345.9	330.6	315.8	308.7	310.6	289.9
22	38	325.0	335.0	327.0	325.6	322.5	312.5	273.9	272.2	266.9	252.2
23	39	362.2	356.0	356.2	357.2	357.2	356.0	356.8	352.3	358.4	346.3
24	40	294.0	294.0	289.1	295.5	305.0	303.2	307.3	306.4	307.6	308.8
25	42	376.4	374.3	366.0	348.8	332.2	322.5	307.8	297.0	286.6	274.0
26	43	404.5	403.3	397.8	386.3	388.4	385.0	381.9	393.1	382.0	381.4
27	44	352.5	344.9	*	*	*	*	*	*	*	*
28	45	417.5	410.0	407.4	380.0	368.0	351.4	336.5	337.8	324.9	311.4
29	47	*	397.7	*	*	*	*	*	*	*	*
30	48	425.0	421.0	409.6	390.2	372.4	359.0	345.8	331.6	320.9	308.2
31	49	376.0	372.7	358.3	351.5	333.0	323.5	311.8	320.1	310.5	314.3
32	50	412.0	411.3	*	*	*	*	*	*	*	*
33	51	342.0	342.0	324.2	312.0	294.9	280.0	271.3	259.3	247.6	239.9
34	56	432.6	435.7	441.2	437.8	434.4	434.3	434.3	430.0	436.2	429.8
35	57	*	344.8	359.1	352.2	337.3	331.5	329.0	336.2	351.5	345.0
36	58	*	367.8	363.5	356.4	347.9	337.0	332.0	325.2	295.5	291.0
37	61	391.0	391.3	380.5	353.1	341.9	330.4	316.8	307.1	293.6	284.3
38	63	426.5	424.0	404.5	391.5	370.9	357.4	337.0	323.0	311.6	295.3
39	64	388.0	403.6	395.5	393.8	395.6	394.9	397.3	393.2	389.8	384.5
40	65	394.5	382.7	*	*	*	*	*	*	*	*
41	66	356.0	361.2	353.3	350.0	353.0	345.8	345.8	347.1	343.6	343.0
42	67	344.5	346.5	340.4	328.6	315.9	307.6	291.2	275.0	267.5	261.7
43	68	337.8	340.6	342.0	351.0	347.5	346.7	349.5	346.7	349.8	345.8
44	69	324.0	322.0	323.2	319.2	324.9	330.5	327.7	330.3	326.8	331.7
45	70	381.5	378.0	364.5	342.6	330.1	310.6	302.1	288.9	275.3	264.3
46	71	354.3	342.8	341.5	339.1	351.1	350.9	348.2	350.4	361.8	350.0
47	72	350.5	349.5	339.9	340.5	339.0	343.5	344.0	343.0	347.2	353.5
48	73	393.5	389.3	390.0	397.2	395.7	395.1	404.5	401.8	395.0	403.7
49	74	431.0	439.0	436.3	437.9	435.7	438.9	445.3	438.7	439.2	435.9

ROW	d11	d12	d13	d14	totgain

1	336.2	331.8	329.0	332.5	-17.000	
2	327.2	327.0	323.7	326.7	-33.800	
3	381.9	378.5	367.3	346.4	-44.600	
4	369.3	370.0	365.9	364.5	-2.500	
5	410.0	402.5	401.5	394.8	-20.700	
6	219.9	216.4	*	*	-101.600	
7	364.9	364.6	352.0	350.9	-30.900	
8	328.0	334.8	337.0	339.8	-99.600	
9	307.0	300.0	298.6	289.5	-54.500	
10	*	*	*	*	*	
11	290.5	284.2	269.2	271.8	-118.600	
12	400.4	408.2	409.0	398.4	-16.900	
13	*	*	*	*	*	
14	272.5	259.3	*	*	-99.200	
15	339.6	336.9	344.0	335.8	-20.700	
16	246.5	*	*	*	-115.200	
17	371.0	363.0	355.3	348.1	-39.600	
18	390.5	378.7	374.7	376.3	-21.600	
19	395.3	387.2	388.3	387.5	-13.300	
20	*	*	*	*	*	
21	278.1	320.8	331.9	319.4	-54.700	
22	251.4	259.3	270.5	290.1	-44.900	
23	339.7	337.8	340.0	334.0	-22.000	
24	301.4	309.6	302.1	304.5	10.500	
25	263.0	252.6	255.8	*	-118.500	
26	383.5	376.6	383.5	385.9	-17.400	
27	*	*	*	*	*	
28	344.5	341.2	336.5	345.1	-64.900	
29	*	*	*	*	*	
30	*	*	*	*	-112.800	
31	306.9	305.9	312.4	314.0	-58.700	
32	*	*	*	*	*	
33	*	*	*	*	-102.100	
34	437.6	431.1	431.5	423.2	-12.500	
35	336.8	340.0	326.1	330.7	-14.100	
36	280.4	265.0	273.0	269.8	-98.000	
37	*	*	*	*	-107.000	
38	282.5	277.0	*	*	-147.000	
39	381.9	387.1	383.0	386.2	-17.400	
40	*	*	*	*	*	
41	343.1	336.5	339.8	334.5	-26.700	
42	*	*	*	*	-84.800	
43	345.5	335.6	340.2	340.7	0.100	
44	328.0	329.3	319.6	328.3	6.300	
45	251.9	244.0	238.3	*	-139.700	
46	364.7	362.0	365.1	357.4	14.600	
47	345.8	350.5	344.0	347.3	-2.200	
48	404.5	412.1	403.1	402.7	13.400	
49	435.5	435.5	418.2	427.3	-11.700	

Litter livewight (g)

treat = feeds, except '*' = initial cull; d = day

ROW	rat no	treat	d2	d3	d4	d5	d6	d7	d8	d9	d10
1	3	1	90.6	107.8	125.2	135.5	152.1	163.6	179.7	198.6	219.2
2	5	1	97.4	111.5	129.1	141.4	160.4	174.7	199.6	215.1	234.0
3	6	2	114.6	136.2	158.9	181.8	207.7	232.2	258.2	294.9	326.0
4	8	4	82.7	91.3	111.8	132.3	151.0	179.8	204.6	236.2	265.2
5	9	6	121.8	142.5	167.7	203.1	232.8	269.3	308.1	342.4	381.5
6	12	5	118.8	124.0	133.5	133.5	139.6	143.9	147.4	149.8	149.8
7	14	4	118.0	138.8	159.9	189.1	214.8	246.7	278.2	308.4	342.2
8	15	3	121.1	142.5	162.9	175.4	188.0	197.2	201.3	216.7	222.3
9	19	3	91.7	105.4	113.9	125.7	143.2	160.4	177.5	196.0	206.8
10	20	*	51.0	*	*	*	*	*	*	*	*
11	22	7	138.4	145.3	164.3	177.3	185.4	199.3	204.7	206.8	213.6
12	25	8	108.1	127.8	144.4	166.1	190.4	216.3	242.6	275.2	302.7
13	26	*	*	*	*	*	*	*	*	*	*
14	28	7	95.0	107.9	123.5	137.3	141.0	147.4	152.6	153.0	158.7
15	29	1	105.6	116.8	132.2	150.9	169.9	192.0	212.3	237.6	253.0
16	31	5	107.8	117.9	130.6	133.6	132.4	135.0	132.5	131.9	128.5
17	33	2	118.2	137.8	162.5	182.0	212.4	234.3	264.6	294.6	330.0
18	34	8	105.3	121.4	142.9	168.6	194.0	224.3	258.9	288.8	318.3
19	35	6	111.5	130.4	158.9	184.7	211.3	243.1	277.4	306.8	343.5
20	36	*	81.9	*	*	*	*	*	*	*	*
21	37	3	125.9	137.2	154.2	167.5	172.1	180.2	185.7	193.2	201.1
22	38	1	98.9	118.4	134.2	154.0	168.4	184.8	189.1	195.0	199.9
23	39	2	116.5	130.0	144.4	166.7	183.3	206.1	229.3	250.1	274.9
24	40	2	102.9	118.0	133.4	152.1	179.1	204.0	231.9	254.8	278.8
25	42	5	101.1	112.6	118.1	124.1	130.0	128.2	133.7	133.9	134.6
26	43	1	112.5	113.4	129.5	145.8	161.1	179.4	198.9	217.7	234.0
27	44	*	89.3	*	*	*	*	*	*	*	*
28	45	6	113.0	137.7	142.7	159.4	165.0	161.5	172.1	193.6	203.1
29	47	*	*	*	*	*	*	*	*	*	*
30	48	7	123.2	142.4	156.5	161.0	167.6	172.0	171.7	168.3	167.5
31	49	3	120.1	124.5	132.2	141.0	148.0	154.8	165.7	181.3	189.1
32	50	*	34.6	*	*	*	*	*	*	*	*
33	51	5	119.0	126.2	138.1	144.4	146.0	148.1	145.5	140.4	133.6
34	56	4	114.7	127.9	151.3	175.7	205.5	235.8	272.0	302.3	332.8
35	57	2	114.0	128.0	154.9	179.2	207.1	236.8	272.4	300.8	340.0
36	58	1	111.1	122.1	138.9	158.6	172.4	185.2	197.7	201.4	209.7
37	61	7	104.4	124.5	132.3	139.4	147.4	149.9	148.3	144.8	145.2
38	63	5	118.9	131.1	146.8	156.5	156.5	154.6	156.8	150.5	146.2
39	64	8	114.3	128.8	154.9	181.5	212.1	239.5	267.2	289.4	324.9
40	65	*	77.1	*	*	*	*	*	*	*	*
41	66	3	120.7	130.8	145.1	163.3	180.8	199.2	215.4	236.0	254.1
42	67	7	108.7	120.6	133.4	133.5	134.7	136.0	134.7	134.3	130.8
43	68	6	115.7	131.9	150.4	175.8	202.9	231.0	259.5	289.9	313.0
44	69	4	109.2	128.8	146.2	168.1	190.4	217.2	244.2	275.1	298.6
45	70	5	114.6	127.8	138.7	139.4	140.8	139.9	141.7	142.2	139.4
46	71	8	101.7	114.8	136.0	157.2	185.3	212.4	238.9	261.8	288.2
47	72	8	97.4	109.0	127.9	147.0	170.9	194.8	221.3	246.8	268.2
48	73	6	111.2	123.1	139.4	162.7	185.9	209.7	238.8	268.4	295.0
49	74	2	113.2	129.2	151.3	180.1	207.4	239.3	271.9	310.0	336.5

ROW	d11	d12	d13	d14	totgain
1	237.9	262.1	282.2	309.6	219.0

ROW					
2	251.8	270.7	288.1	310.7	213.3
3	355.7	388.2	411.6	442.6	328.0
4	304.7	340.5	374.5	401.8	319.1
5	424.1	460.9	504.2	543.1	421.3
6	142.6	142.3	*	*	23.5
7	370.9	405.5	435.4	472.1	354.1
8	238.8	257.4	270.9	293.3	172.2
9	220.2	236.3	255.3	273.7	182.0
10	*	*	*	*	*
11	224.2	232.6	239.7	247.2	108.8
12	330.2	355.1	390.9	427.9	319.8
13	*	*	*	*	*
14	154.5	153.7	*	*	58.7
15	269.7	284.8	306.6	326.5	220.9
16	125.7	*	*	*	17.9
17	362.5	395.4	430.3	463.8	345.6
18	348.8	367.6	404.4	443.5	338.2
19	374.3	403.7	438.1	466.9	355.4
20	*	*	*	*	*
21	201.2	202.4	204.5	195.0	69.1
22	205.0	218.2	237.2	256.6	157.7
23	296.9	320.9	350.1	372.7	256.2
24	308.3	325.3	356.9	383.8	280.9
25	133.0	126.4	121.1	*	30.6
26	256.6	275.6	297.2	315.7	203.2
27	*	*	*	*	*
28	226.3	263.8	304.9	332.3	219.3
29	*	*	*	*	*
30	*	*	*	*	44.3
31	207.6	225.1	240.6	258.9	138.8
32	*	*	*	*	*
33	*	*	*	*	14.6
34	366.7	399.8	434.6	471.5	356.8
35	368.5	402.1	431.5	454.4	340.4
36	211.3	209.8	217.7	232.9	121.8
37	*	*	*	*	40.8
38	143.7	147.0	*	*	28.1
39	357.2	371.9	399.5	441.7	327.4
40	*	*	*	*	*
41	274.3	299.3	318.9	338.5	217.8
42	*	*	*	*	22.1
43	341.2	372.2	398.4	427.3	311.6
44	325.6	356.7	385.5	415.1	305.9
45	133.3	130.2	126.0	*	11.4
46	315.0	343.7	367.3	387.1	285.4
47	290.4	317.3	349.5	380.9	283.5
48	327.9	354.8	386.2	422.3	311.1
49	365.5	397.9	429.5	462.3	349.1

Carcass composition

MDM = maternal dry matter; MCP = maternal protein (%DM); MEE = maternal ether extract (%DM);
Mash = maternal ash (%DM); MGE = maternal gross energy (kJ/gDM). LDM = litter dry matter....

ROW	rat.	treat	MDM	MCP	MEE	Mash	MGE	LDM	LCP	LEE

1	3	1	0.357318	57.56	24.68	11.27	23.31	0.270940	52.02	32.68
2	5	1	0.373634	54.13	29.72	10.43	24.09	0.274558	50.20	34.62
3	6	2	0.349985	59.91	24.96	11.30	23.42	0.283154	54.43	30.36
4	8	4	0.336344	62.05	23.26	10.69	23.26	0.302229	47.23	38.28
5	9	6	0.385482	50.70	35.22	8.73	25.21	0.345984	40.64	36.84
6	12	5	0.324521	68.31	7.42	15.43	20.09	0.248911	61.93	20.59
7	14	4	0.338399	61.63	20.54	11.12	22.21	0.319789	46.94	37.66
8	15	3	0.382839	51.01	31.91	10.30	23.97	0.284079	49.47	34.62
9	19	3	0.374736	53.75	28.44	11.20	23.80	0.295208	49.28	31.56
10	20	*	0.444638	40.19	*	8.29	26.80	0.221068	50.52	34.86
11	22	7	0.359962	59.45	23.94	11.95	23.07	0.265808	55.87	27.70
12	25	8	0.407979	47.91	33.83	8.78	25.91	0.318420	44.12	34.48
13	26	*	0.439606	40.46	41.85	8.12	26.67	*	*	*
14	28	7	0.370636	55.94	*	11.77	23.52	0.256408	57.64	24.44
15	29	1	0.372882	54.92	27.39	11.17	23.37	0.279050	50.75	34.79
16	31	5	0.385300	54.83	28.97	11.99	23.19	0.228557	68.85	10.86
17	33	2	0.336720	62.82	22.40	10.89	22.86	0.292456	52.64	*
18	34	8	0.346128	58.74	25.31	10.56	23.55	0.317051	44.22	*
19	35	6	0.342589	60.20	*	9.79	23.72	0.328889	42.64	*
20	36	*	0.423731	46.73	39.35	10.11	25.45	0.169709	69.21	13.03
21	37	3	0.384737	52.17	*	9.87	24.66	0.244725	64.86	16.85
22	38	1	0.346327	55.75	24.22	12.11	22.63	0.277913	53.63	25.93
23	39	2	0.361218	56.62	28.13	10.70	23.65	0.259687	59.38	23.54
24	40	2	0.318354	67.40	15.91	11.27	22.23	0.263808	58.17	25.76
25	42	5	0.430844	43.95	41.88	10.81	25.78	0.248750	65.25	16.74
26	43	1	0.379215	51.10	31.59	9.53	24.83	0.274160	53.14	*
27	44	*	0.360435	51.47	30.13	11.10	24.02	0.162179	67.91	12.34
28	45	6	0.389392	51.65	31.68	9.99	24.41	0.304935	46.32	33.41
29	47	*	0.410453	43.51	43.11	8.80	26.66	0.175769	63.43	18.92
30	48	7	0.431627	47.26	37.45	10.71	25.75	0.272356	52.35	31.10
31	49	3	0.384484	57.77	23.84	11.68	23.05	0.278389	52.27	30.90
32	50	*	0.460961	38.73	*	7.64	27.05	0.185422	59.19	23.37
33	51	5	0.397483	52.62	24.75	12.23	23.19	0.272419	56.08	26.78
34	56	4	0.373373	54.43	32.23	9.41	24.90	0.310932	46.31	40.13
35	57	2	0.330389	63.27	16.89	11.15	22.32	0.312586	49.87	35.90
36	58	1	0.334268	63.94	17.33	13.50	21.64	0.271071	56.33	22.60
37	61	7	0.374587	55.58	21.95	13.44	22.53	0.257279	56.11	25.60
38	63	5	0.414049	46.23	41.33	10.81	25.31	0.302870	54.95	23.01
39	64	8	0.398174	48.82	*	8.95	25.27	0.328317	41.88	*
40	65	*	0.423656	46.85	38.27	9.57	25.40	0.171222	66.77	16.69
41	66	3	0.368885	55.38	27.47	10.13	24.02	0.290524	47.80	37.17
42	67	7	0.449395	41.96	*	9.54	25.95	0.228749	65.15	16.03
43	68	6	0.361740	55.17	28.87	10.09	22.79	0.327692	42.62	38.55
44	69	4	0.356639	59.58	19.24	10.77	23.41	0.315566	46.70	35.19
45	70	5	0.375305	55.57	28.89	12.24	23.53	0.248627	67.52	15.01
46	71	8	0.368007	52.97	31.56	10.12	24.31	0.316443	44.99	*
47	72	8	0.396265	49.26	35.62	8.48	25.60	0.309919	46.22	35.71
48	73	6	0.377008	49.28	34.45	8.83	25.16	0.313697	43.94	*
49	74	2	0.379545	52.53	32.10	8.69	25.28	0.285852	53.31	31.62

FMat lwt = Final maternal liveweight; FMat Ewt = Final maternal weight minus gut contents;
FLit lwt = Final litter liveweight; FLit Ewt = Final litter weight minus gut contents;
IMat lwt = Initial maternal lwt.; ILit lwt = Initial litter lwt.; natlitt = littersize at birth

ROW	LAsh	LGE	FMat lwt	FMat Ewt	FLit lwt	FLit Ewt	IMat lwt	ILit lwt	natlitt
1	9.34	24.66	332.5	293.5	309.4	302.2	349.5	90.6	13

2	9.32	25.10	326.7	302.0	310.7	303.5	360.5	97.4	14
3	8.83	24.49	346.4	328.7	442.6	434.2	391.0	114.6	13
4	8.46	25.65	364.5	344.6	401.8	395.1	367.0	82.7	16
5	7.46	26.34	394.8	369.3	543.1	531.2	415.5	121.8	13
6	11.73	22.52	216.4	209.5	142.3	137.7	311.1	118.8	9
7	8.40	25.82	350.9	328.3	472.1	462.5	381.8	118.0	13
8	10.11	24.61	339.8	315.9	293.3	284.1	439.4	121.1	15
9	9.27	25.26	289.5	271.1	273.7	267.5	344.0	91.7	10
10	8.58	24.94	371.7	360.0	51.0	48.0	371.7	51.0	4
11	11.51	23.28	271.8	257.2	247.2	247.2	390.4	138.4	10
12	7.88	25.46	398.4	371.5	427.9	422.2	415.3	108.1	15
13	*	*	334.3	326.5	*	*	334.3	*	5
14	11.08	23.17	259.3	250.0	153.7	153.7	358.5	95.0	12
15	9.21	24.77	335.8	320.8	326.5	321.2	356.5	105.6	11
16	13.18	20.69	246.5	239.9	125.2	125.2	361.7	107.8	13
17	8.92	24.80	348.1	332.9	463.8	455.8	387.7	118.2	14
18	8.54	25.55	376.3	350.5	443.5	434.4	397.9	105.3	17
19	7.84	26.16	387.5	365.3	466.9	460.7	400.2	111.5	17
20	12.08	20.98	277.8	269.3	81.9	80.8	277.8	81.9	16
21	12.65	21.71	319.4	310.6	195.0	192.1	374.1	125.9	12
22	10.41	24.13	290.1	261.6	256.6	251.0	335.0	98.9	16
23	10.65	22.94	334.0	315.8	372.7	364.0	356.0	116.5	13
24	10.29	23.33	304.5	282.1	383.8	374.4	294.0	102.9	14
25	12.84	21.81	255.8	247.3	121.1	121.1	374.3	101.1	13
26	10.48	24.10	385.9	356.8	319.8	315.7	403.3	112.5	13
27	12.23	20.93	344.9	331.6	89.3	83.6	344.9	89.3	14
28	8.83	25.94	345.1	325.3	332.3	323.0	410.0	113.0	13
29	10.82	22.37	397.7	385.6	*	*	397.7	*	9
30	10.10	24.30	308.2	299.9	167.5	167.5	421.0	123.2	11
31	10.16	24.01	314.0	291.6	258.9	252.9	372.7	120.1	12
32	10.92	23.08	411.3	402.3	34.6	32.0	411.3	34.6	9
33	11.17	23.32	239.9	232.0	133.6	133.5	342.0	119.0	13
34	8.07	25.99	423.2	405.2	471.5	460.3	435.7	114.7	10
35	8.68	25.47	330.7	309.3	454.4	447.5	344.8	114.0	16
36	11.22	23.17	269.8	254.2	232.9	228.1	367.8	111.1	13
37	11.51	23.23	284.3	277.2	145.2	145.2	391.3	104.4	15
38	10.68	23.67	277.0	271.1	147.0	146.2	424.0	118.9	13
39	7.63	26.03	386.2	351.3	441.7	431.7	403.6	114.3	12
40	11.67	21.43	382.7	373.5	77.1	71.7	382.7	77.1	15
41	9.26	25.33	334.5	309.3	338.5	333.6	361.2	120.7	11
42	11.79	21.66	261.7	258.0	130.8	130.8	346.5	108.7	10
43	7.59	26.30	340.7	318.6	427.3	421.3	340.6	115.7	11
44	8.27	25.92	328.3	307.2	415.1	407.2	322.0	109.2	12
45	12.79	21.04	238.3	232.7	126.0	126.0	378.0	114.6	14
46	8.27	25.34	357.4	328.6	387.1	381.8	342.8	101.7	14
47	8.02	25.50	347.3	321.8	380.9	370.8	349.5	97.4	13
48	8.10	25.74	402.7	382.6	422.3	412.7	389.3	111.2	10
49	9.10	24.41	427.3	406.8	462.3	449.5	439.0	113.2	12

Appendix 4

The data from the third rat experiment (R3)

Maternal food intake (gDM)

d = day

ROW	rat	treat	d 2	d 3	d 4	d 5	d 6	d 7	d 8
1	1	3	16.0406	21.7892	26.6106	33.2401	33.6574	33.5646	36.5317
2	2	2	9.7204	9.4302	1.8377	3.3852	1.4024	1.3541	1.9828
3	3	0	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
4	4	3	33.7037	31.4321	36.2535	43.2539	46.3600	42.8366	41.9558
5	5	1	12.9501	26.5896	33.4832	34.7634	26.8850	18.9574	13.2456
6	7	1	34.5172	29.0516	33.9264	39.0966	39.0966	42.6911	42.0017
7	8	2	20.8432	19.8760	15.3301	16.3940	18.2317	20.0210	23.4062
8	10	1	11.5222	20.6316	24.4230	23.8322	24.5215	30.5780	31.0704
9	11	1	16.5939	16.3477	25.0632	33.3847	35.6990	33.3355	34.8127
10	12	2	2.7082	9.1400	6.8671	6.0450	2.2246	3.3852	3.2885
11	14	3	14.6034	15.3915	21.2329	24.6635	29.2995	36.6244	32.9620
12	15	0	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
13	16	0	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
14	17	0	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
15	18	3	19.6103	24.6172	20.2593	28.4650	33.7037	33.2865	23.3654
16	19	2	16.6842	22.7776	13.1056	19.4891	17.7481	9.1400	12.9605
17	20	3	15.9942	24.8490	25.8689	27.3524	37.2734	36.9489	38.6179
18	21	2	16.2490	19.3924	22.5358	25.2439	19.4891	13.7342	7.9794
19	22	1	22.3057	15.8060	17.4310	12.3592	27.4759	34.6157	34.5172
20	24	0	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000

ROW	d 9	d 10	d 11	d 13	d 14	cumulative
1	39.2669	39.7769	42.1876	39.6842	41.2604	403.610
2	1.9828	1.9828	1.9828	1.9828	1.9828	39.027
3	0.0000	0.0000	0.0000	0.0000	0.0000	0.000
4	46.6382	56.9764	50.9496	51.3205	53.1749	534.855
5	7.4845	5.1702	1.0340	1.2310	1.4280	183.222
6	39.5397	48.8461	36.8315	46.1871	43.5282	475.314
7	16.2006	25.7759	28.0972	23.1644	16.7326	244.073
8	32.2522	32.4492	41.6078	38.3580	39.9336	351.180
9	36.0929	32.3507	32.8923	41.2631	35.3051	373.141
10	2.9983	2.9983	2.9983	2.9983	2.9983	48.650
11	37.2271	33.7964	47.3799	35.9754	44.2738	373.430
12	0.0000	0.0000	0.0000	0.0000	0.0000	0.000
13	0.0000	0.0000	0.0000	0.0000	0.0000	0.000
14	0.0000	0.0000	0.0000	0.0000	0.0000	0.000
15	21.5110	37.5052	43.5320	48.8634	49.5588	384.278
16	23.3579	18.1350	24.4218	31.1922	36.1249	245.137
17	46.4991	43.1148	46.5454	44.1347	49.9761	437.175
18	15.1367	5.7065	13.2023	21.6169	21.3751	201.661
19	39.0473	38.5549	54.1640	44.7099	55.3950	396.382
20	0.0000	0.0000	0.0000	0.0000	0.0000	0.000

Maternal liveweight (g)

d = day

ROW	rat	treat	d1	d2	d3	d4	d5	d6	d7	C8	C9
1	1	3	*	439.9	440.0	437.3	434.7	438.1	436.0	440.2	432.0
2	2	2	422.9	414.1	399.9	383.7	363.0	346.9	332.4	314.5	*
3	3	0	396.5	397.2	*	*	*	*	*	*	*
4	4	3	378.7	376.4	373.4	372.6	374.2	387.8	385.0	385.4	386.8
5	5	1	510.0	514.5	497.2	500.2	503.0	494.8	464.4	451.1	472.2
6	7	1	412.0	402.5	400.5	404.1	400.5	401.5	399.5	394.2	393.0
7	8	2	316.5	327.2	324.4	322.2	312.4	311.1	281.1	295.5	296.5
8	10	1	346.5	360.0	335.2	336.4	334.2	334.7	325.0	325.5	327.5
9	11	1	374.0	367.5	360.0	350.1	347.0	349.3	352.0	352.8	352.0
10	12	2	362.6	359.0	332.6	330.7	313.0	309.7	291.4	285.1	270.4
11	14	3	436.6	427.5	425.4	418.1	413.5	414.6	414.2	401.9	404.7
12	15	0	382.8	382.2	*	*	*	*	*	*	*
13	16	0	439.0	437.1	*	*	*	*	*	*	*
14	17	0	*	339.3	*	*	*	*	*	*	*
15	18	3	419.1	417.3	418.4	417.6	408.5	411.0	417.6	407.9	392.0
16	19	2	361.1	358.5	346.4	352.2	341.0	338.4	332.7	318.2	318.5
17	20	3	352.3	351.7	347.5	347.3	350.5	349.5	352.9	358.6	357.5
18	21	2	453.0	459.8	456.8	446.0	440.0	437.6	422.0	412.0	390.8
19	22	1	380.5	375.2	374.5	358.8	351.9	341.7	345.2	346.6	340.9
20	24	0	362.2	*	*	*	*	*	*	*	*

ROW	d10	d11	d12	d13	d14	Gain (g/12d)
1	442.5	442.0	434.3	429.2	420.0	-19.900
2	*	*	*	*	*	-99.600
3	*	*	*	*	*	*
4	374.5	387.9	390.7	376.7	380.3	3.900
5	468.4	433.9	414.6	392.2	381.4	-133.100
6	382.9	399.0	383.9	387.8	*	-17.800
7	284.8	288.5	294.4	293.6	280.2	-47.000
8	322.5	315.6	320.0	317.4	311.5	-29.300
9	344.2	342.0	335.7	344.5	332.9	-34.600
10	260.8	*	*	*	*	-98.200
11	412.5	400.8	404.8	394.1	399.7	-27.800
12	*	*	*	*	*	*
13	*	*	*	*	*	*
14	*	*	*	*	*	*
15	401.3	399.8	400.7	401.0	406.7	-10.600
16	317.8	312.1	305.4	309.6	319.9	-38.600
17	371.6	364.8	365.9	362.0	363.4	11.700
18	387.2	369.0	364.3	367.2	362.2	-97.600
19	341.3	344.3	360.0	349.1	348.8	-26.400
20	*	*	*	*	*	*

Litter liveweight (g)

d = day

ROW	rat	treat	d2	d3	d4	d5	d6	d7	d8	d9	d10
1	1	3	96.3	110.7	130.0	154.1	180.9	212.3	240.9	276.0	304.7
2	2	2	115.0	132.4	143.1	142.9	148.2	147.1	146.1	*	*
3	3	0	95.7	*	*	*	*	*	*	*	*
4	4	3	116.4	135.9	164.4	190.8	227.4	264.7	306.9	338.6	384.5
5	5	1	108.6	127.5	147.3	172.3	205.0	224.2	231.7	222.4	238.1
6	7	1	105.9	123.9	153.9	177.0	208.3	237.8	274.6	305.2	336.5
7	8	2	102.9	120.0	135.4	149.4	162.5	170.2	182.9	198.2	211.9
8	10	1	82.6	87.1	102.7	121.4	141.4	165.3	189.7	211.5	240.4
9	11	1	105.4	122.1	134.8	156.9	184.8	212.3	238.3	266.3	295.3
10	12	2	98.9	101.8	110.4	119.6	123.5	126.9	129.6	132.9	131.8
11	14	3	99.5	115.3	133.9	153.0	179.0	205.0	232.7	258.7	285.8
12	15	0	*	*	*	*	*	*	*	*	*
13	16	0	20.0	*	*	*	*	*	*	*	*
14	17	0	109.5	*	*	*	*	*	*	*	*
15	18	3	106.8	123.3	142.6	163.5	188.6	213.8	249.5	270.5	306.2
16	19	2	105.6	117.6	133.6	149.6	165.0	178.2	186.6	192.3	210.4
17	20	3	91.7	106.2	123.5	139.7	159.3	190.4	220.4	250.3	280.6
18	21	2	113.2	129.8	155.0	176.4	201.3	223.8	236.0	245.3	251.5
19	22	1	99.0	116.8	135.8	157.2	170.2	195.6	220.0	248.7	280.9
20	24	0	110.2	*	*	*	*	*	*	*	*

ROW	d11	d12	d13	d14	Gain (g/12d)
1	335.5	374.9	407.5	440.0	343.7
2	*	*	*	*	31.1
3	*	*	*	*	*
4	421.1	455.2	494.0	528.6	412.2
5	257.7	251.9	245.3	236.0	127.4
6	361.1	391.3	423.7	455.7	349.8
7	228.1	247.0	262.6	275.3	172.4
8	264.8	293.1	321.6	353.8	271.2
9	317.9	344.3	366.5	403.2	297.8
10	*	*	*	*	32.9
11	317.3	349.7	378.7	407.6	308.1
12	*	*	*	*	*
13	*	*	*	*	*
14	*	*	*	*	*
15	336.9	370.5	408.1	438.0	331.2
16	223.7	241.0	263.1	285.4	179.8
17	317.7	351.3	385.9	421.0	329.3
18	254.2	260.7	268.1	281.9	168.7
19	310.6	338.3	377.6	416.9	317.9
20	*	*	*	*	*

Carcass composition (%DM or kJ/gDM as appropriate)

M = maternal; L = litter; d2mlwt = maternal livweight on day 2; d2llwt = litter liveweight on day 2; fm lwt = final maternal liveweight; fm ewt = final maternal weight minus gut contents; fl lwt = final litter liveweight; fl ewt = final litter weight minus gut contents; natlit = littersize at birth

ROW	rat	treat	M DM	M CP	M EE	M ash	M GE	L DM	L CP	L EE	L ash
1	1	3	42.412	43.21	41.68	7.74	26.50	33.154	42.12	36.49	7.37
2	2	2	38.615	45.20	34.74	10.20	25.42	43.381	42.78	*	8.49
3	3	0	44.394	40.93	44.64	9.22	26.47	*	58.37	23.92	10.08
4	4	3	36.616	54.45	29.64	10.09	24.12	34.641	41.46	37.07	7.46
5	5	1	43.586	41.71	45.91	8.50	26.76	30.493	49.82	33.79	9.20
6	7	1	37.741	52.33	32.28	9.36	24.79	34.854	38.26	37.28	6.96
7	8	2	33.254	63.12	17.92	14.08	21.28	29.433	48.86	34.29	9.18
8	10	1	36.413	55.92	*	5.47	23.51	31.930	42.89	37.14	7.47
9	11	1	38.194	51.99	*	10.26	24.11	32.071	43.06	39.09	7.72
10	12	2	36.311	56.29	26.70	12.40	23.19	21.438	68.22	13.18	12.33
11	14	3	37.732	51.37	31.92	9.43	24.65	31.266	45.66	39.86	8.09
12	15	0	23.952	57.18	26.60	10.39	23.55	*	*	*	*
13	16	0	49.158	33.81	50.31	7.52	27.60	20.392	56.37	10.53	9.38
14	17	0	42.467	43.78	39.01	8.30	26.17	18.800	56.58	25.57	10.24
15	18	3	38.772	49.92	34.29	9.22	25.05	31.532	45.65	35.75	8.85
16	19	2	34.095	59.11	21.42	11.60	22.90	29.382	47.66	32.13	9.11
17	20	3	36.622	53.68	29.39	10.58	24.14	31.988	43.47	36.26	7.31
18	21	2	41.014	46.76	39.08	9.23	25.44	29.747	47.71	33.54	9.39
19	22	1	33.369	63.21	19.81	11.92	22.43	33.812	38.11	38.43	6.95
20	24	0	42.600	44.86	38.71	9.18	25.38	19.051	55.27	29.31	9.51

ROW	L GE	L sum	d2mlwt	d2llwt	fm lwt	fm ewt	fl lwt	fl ewt	natlit
1	26.52	85.98	439.9	96.3	420.0	398.8	440.0	434.8	11
2	26.33	*	414.1	115.0	314.5	306.1	146.1	146.1	11
3	23.11	92.37	397.2	95.7	397.2	375.6	95.7	94.0	11
4	26.33	85.99	376.4	116.4	380.3	356.4	528.6	515.8	12
5	24.75	92.81	514.5	108.6	381.4	373.3	236.0	235.3	11
6	26.76	82.50	402.5	105.9	*	350.6	455.7	449.5	15
7	24.82	92.33	327.2	102.9	280.2	241.4	275.3	274.7	13
8	26.16	87.50	360.0	82.6	311.5	292.8	353.8	347.0	15
9	26.01	89.87	367.5	105.4	332.9	301.1	403.2	394.2	12
10	21.06	93.73	359.0	98.9	260.8	255.8	131.8	130.2	11
11	25.54	93.61	427.5	99.5	399.7	376.4	407.6	399.4	12
12	*	*	382.2	*	382.2	372.4	*	*	6
13	22.77	76.28	437.1	20.0	437.1	424.1	*	*	4
14	23.59	92.39	339.3	109.5	339.3	388.0	*	109.5	13
15	25.53	90.25	417.3	106.8	406.7	386.2	438.0	431.7	9
16	24.79	88.90	358.5	105.6	319.9	303.6	285.4	275.9	11
17	26.29	87.04	351.7	91.7	363.4	344.7	421.0	416.6	12
18	25.06	90.64	459.8	113.2	362.2	342.9	381.9	276.8	10
19	26.78	83.49	375.2	99.0	348.8	314.7	416.9	407.8	17
20	25.11	94.09	*	110.2	*	348.0	110.2	106.6	12

The data from the fourth rat experiment (R4)

rat	feed
1	6
2	4
5	6
6	1
8	3
9	6
10	4
11	4
16	2
18	6
19	1
22	3
25	3
26	5
27	1
30	2
31	6
33	5
35	3
42	5
44	1
47	3
48	2
50	1
51	5
52	4
53	2
54	4	removed from the experiment
55	5
58	2

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Food intake (gDM)

day	m1	m2	m5	m6	m8	m9	m10	m11	m16
2	14.5814	4.2306	3.5133	14.4388	17.8437	19.3198	17.7638	4.66780	6.30
3	2.9335	1.7510	2.8415	15.4104	33.6150	6.0348	19.0600	0.57720	6.47
4	5.1651	4.9682	3.4295	19.1937	28.8243	18.2650	19.8576	2.91660	5.80
5	3.5848	13.6710	15.3449	23.0395	31.0808	15.2546	23.6512	2.93400	0.90
6	10.1320	18.7588	5.2403	24.3535	32.9338	15.5512	22.6384	2.53560	19.71
7	4.3310	15.6536	3.2426	26.4702	40.0618	3.8475	26.5378	1.64960	6.43
8	5.3865	6.7496	1.3468	21.3356	43.1429	8.8491	12.3364	2.26000	8.53
9	24.3975	6.4566	5.0665	22.6305	43.1157	20.8512	5.1382	2.26580	9.44
10	26.9350	19.9596	16.6654	24.2903	43.7150	2.0506	7.5452	1.77000	7.05
11	28.8361	20.9578	6.6394	27.4466	48.5450	3.2498	16.3590	2.46920	0.66

day	m18	m19	m22	m25	m26	m27	m30	m31	m33
2	20.5504	16.8949	19.4130	25.6305	24.9413	20.7986	6.9142	1.80	25.00
3	17.7502	23.1109	27.5836	28.0562	26.3995	27.2932	4.2745	1.84	27.34
4	24.3483	22.3954	30.9318	30.8984	29.5022	24.7658	1.5874	2.09	33.34
5	26.0458	29.2841	31.6706	40.7363	34.8059	30.3888	2.0030	3.64	31.61
6	23.1441	30.0130	26.5420	48.4218	34.6965	25.7324	4.0855	5.55	22.81
7	24.2432	35.4045	12.0168	43.5004	33.0861	32.8363	5.3223	14.26	40.18
8	21.6447	32.6973	32.8372	47.7116	33.4834	37.4738	18.9382	14.85	38.02
9	27.3470	29.6370	36.8359	45.0599	39.7873	37.9716	16.5976	26.05	38.78
10	34.0327	36.9013	39.0802	47.5089	42.0742	42.8343	13.5626	30.14	50.58
11	43.5166	39.5873	43.0734	50.1076	51.9744	47.6228	9.0595	30.83	38.37

day	m35	m42	m44	m47	m48	m50	m51	m52	m53
2	19.5441	29.7151	21.4681	22.7148	5.8282	23.8721	21.0857	6.1678	6.8303
3	28.8637	50.5948	26.3642	25.9311	2.7557	26.4324	32.2009	5.0844	8.4356
4	32.8889	71.3787	23.4734	36.2461	0.6689	26.7087	34.5967	4.1944	12.4480
5	39.3077	20.7194	25.2288	37.7019	4.8770	27.0988	25.7050	3.4778	18.5647
6	48.9463	32.9569	28.0513	47.9375	15.3782	11.8122	35.5050	8.0796	14.0661
7	47.4123	30.6820	26.1209	53.1886	15.9755	33.4152	35.3912	11.0324	15.5648
8	52.4436	41.7650	37.4588	46.1886	13.5605	40.4750	39.5873	4.9412	6.6604
9	51.2708	36.6956	43.0797	48.6566	18.6650	36.5657	39.3829	7.5550	11.5633
10	51.9334	43.8965	33.5730	54.8811	15.7632	39.0556	38.0279	9.2122	14.2611
11	56.3559	42.7825	39.2613	57.1206	16.3086	38.9684	38.9815	15.4688	1.5113

day	m55	m58
2	35.0999	11.3206
3	35.3000	20.5854
4	37.1390	16.7374
5	35.4569	11.2389
6	41.3514	11.4493
7	38.8804	11.1658
8	40.4622	22.8658
9	45.4627	30.8564
10	49.7643	35.0504
11	27.5728	22.4517

Maternal liveweight (g)

day	m1	m2	m5	m6	m8	m9	m10	m11	m16	m18	m19
2	413.0	379.0	378.0	397.5	426.0	378.0	378.0	401.0	344.5	345.5	335.8
3	391.0	361.5	362.0	406.0	428.5	365.0	378.5	385.0	345.5	340.0	335.0
4	375.0	340.0	343.0	399.5	435.0	346.0	375.5	365.0	328.7	335.0	339.5
5	361.5	330.0	332.0	394.5	426.0	352.5	378.5	348.5	315.8	338.0	334.0
6	351.0	338.5	325.5	383.0	424.0	344.5	374.7	333.5	305.4	333.0	338.0
7	345.0	341.0	310.0	385.0	400.0	340.5	374.0	323.5	294.0	323.5	337.5
8	328.3	328.0	297.0	379.0	406.0	313.0	374.0	310.0	301.5	327.5	339.5
9	315.5	305.5	287.0	372.0	411.5	306.5	350.0	300.0	280.0	323.5	332.0
10	332.5	287.5	281.5	366.5	406.0	314.9	326.6	290.0	270.7	324.7	337.0
11	340.0	321.5	297.5	362.0	403.5	293.0	308.2	277.3	263.5	324.5	326.5
12	345.7	318.4	284.6	347.8	406.1	280.1	318.6	275.9	259.6	329.8	327.3
13	*	*	*	*	*	*	*	*	*	*	*

day	m22	m25	m26	m27	m30	m31	m33	m35	m42	m44	m47
2	359.0	371.0	365.5	373.0	319.5	332.5	347.0	265.0	428.0	363.0	352.5
3	362.0	381.0	364.0	376.5	297.0	317.0	364.0	274.5	439.5	363.0	364.5
4	363.5	379.5	356.5	379.5	279.0	305.1	358.5	283.0	435.3	360.5	370.0
5	364.5	376.5	359.5	368.5	265.5	289.1	352.0	283.5	426.5	353.0	367.0
6	367.3	386.5	365.0	372.5	257.0	281.5	351.0	298.0	430.0	356.0	364.0
7	355.5	393.0	358.5	369.0	246.5	273.0	341.5	303.0	426.0	355.0	373.5
8	335.5	397.0	357.0	366.0	238.8	276.0	352.0	307.4	421.0	342.5	380.0
9	334.5	385.5	359.0	346.5	249.0	273.0	349.0	302.0	425.0	353.0	381.5
10	334.5	392.0	352.5	346.0	247.0	291.0	347.0	307.5	430.0	353.0	379.0
11	334.0	385.7	358.5	347.5	238.0	278.5	357.5	305.2	426.0	347.0	381.0
12	332.2	389.1	366.1	343.0	223.7	295.5	344.9	308.9	416.3	358.2	380.5
13	*	*	*	*	*	*	*	*	*	*	*

day	m48	m50	m51	m52	m53	m55	m58
2	380.00	351.0	344.5	377.0	381.000	388.5	317.5
3	361.50	373.0	342.5	346.0	368.500	411.5	322.0
4	342.00	364.5	353.0	329.5	357.354	420.0	310.5
5	324.00	360.0	355.7	317.0	362.500	427.0	292.5
6	320.00	358.0	338.0	307.0	359.500	422.0	292.7
7	327.50	348.0	343.2	296.5	354.500	416.0	281.5
8	327.70	346.5	339.4	291.5	334.000	404.0	285.0
9	320.50	341.5	340.2	280.0	329.000	400.5	276.7
10	318.00	347.5	343.7	278.0	329.000	397.0	268.2
11	311.00	349.5	338.0	271.0	301.900	406.5	283.0
12	312.35	352.1	336.3	285.1	*	383.0	282.0
13	*	*	*	*	*	*	278.3

Litter liveweight (g)

ROW	day	p1	p2	p5	p6	p7	p8	p9	p10	p11	p14
1	2	120.9	106.8	99.8	96.9	120.4	116.0	*	93.5	116.4	110.4
2	3	136.7	113.8	102.7	112.2	138.7	132.7	123.7	105.4	126.0	113.3
3	4	143.8	116.2	98.8	135.1	150.0	156.4	134.8	122.7	130.4	121.0
4	5	135.9	118.7	102.1	157.6	172.2	169.2	152.0	141.6	135.3	128.0
5	6	137.5	122.6	109.0	186.0	194.1	196.0	168.2	162.7	137.5	130.3
6	7	140.7	138.4	118.7	208.0	219.5	226.1	179.2	176.6	137.3	133.8
7	8	149.8	162.2	123.6	236.6	242.3	254.9	187.8	202.0	134.5	133.2
8	9	155.4	163.4	124.4	261.8	272.3	282.9	190.6	219.5	132.1	129.7
9	10	169.9	164.0	126.1	287.7	302.5	316.0	196.6	220.2	130.0	127.1
10	11	185.4	171.2	134.3	313.3	330.5	346.0	212.7	225.0	128.7	123.4
11	12	210.7	198.5	147.1	343.4	432.8	374.6	213.2	220.5	127.1	121.5
12	13	*	*	*	*	*	*	*	*	*	*

ROW	p16	p18	p19	p20	p22	p25	p26	p27	p30	p31	p33
1	96.5	88.9	78.4	94.1	92.6	108.9	110.1	106.7	114.8	85.3	83.2
2	99.8	102.3	92.1	93.4	108.8	121.7	128.5	125.5	125.8	88.1	96.4
3	108.4	117.4	106.9	102.2	127.6	140.9	151.2	149.1	129.4	91.0	117.2
4	114.2	132.1	125.5	106.3	148.0	158.8	174.9	171.5	122.5	92.2	145.1
5	119.1	150.4	143.8	113.9	168.3	181.0	198.8	198.6	122.0	91.8	156.1
6	118.6	168.0	170.0	120.8	191.0	209.2	224.6	223.7	121.5	93.9	179.1
7	136.6	183.2	192.2	123.7	200.9	232.8	250.8	254.5	122.9	102.6	206.6
8	150.9	197.2	218.1	130.6	225.3	267.0	275.5	286.0	133.1	109.7	237.4
9	158.1	216.1	239.7	130.9	248.8	288.8	307.4	318.3	155.4	121.0	270.2
10	166.4	245.8	268.1	132.5	273.8	315.5	334.9	353.1	174.0	138.2	301.8
11	169.9	273.5	297.4	125.4	302.8	337.4	366.8	386.8	178.2	148.2	337.7
12	*	*	*	*	*	*	*	*	*	*	*

ROW	p35	p41	p42	p44	p47	p48	p50	p51	p52	p53	p55	p58
1	65.6	84.6	89.9	115.5	90.6	82.6	128.4	104.3	115.9	85.0	88.4	107.8
2	81.5	90.6	103.9	131.8	104.3	88.0	153.3	120.9	120.3	90.9	113.6	119.8
3	96.0	99.0	118.0	153.5	120.2	91.4	183.6	140.1	121.4	99.0	140.2	137.5
4	114.9	111.7	141.2	158.0	144.2	92.9	210.8	166.2	112.9	110.6	150.0	136.5
5	132.4	129.3	163.5	178.8	165.4	97.6	236.3	190.4	113.5	123.9	183.3	144.2
6	154.1	151.0	191.0	198.6	192.2	112.9	270.2	217.8	116.6	141.1	215.2	152.8
7	177.0	172.7	221.9	222.6	221.3	130.0	299.8	246.7	119.8	156.0	254.8	169.4
8	208.3	191.7	250.7	246.6	248.8	138.6	334.2	276.0	119.7	162.9	288.9	184.8
9	233.0	209.8	278.4	276.7	280.9	162.9	367.6	302.1	118.2	172.1	327.5	200.5
10	263.2	234.8	312.7	302.4	312.4	178.7	401.7	331.4	119.4	184.8	358.8	209.5
11	290.8	243.9	348.4	329.5	346.2	190.8	361.6	363.1	128.3	194.7	385.4	229.3
12	*	*	*	*	*	*	*	*	*	*	*	236.7

Carcass composition

treat = feed number, except 7 = initial cull
MDM = maternal dry matter content
MCP = maternal crude protein (g/kg)
MGE =   "   gross energy (g/kg)
MEE =   "   ether extract (g/kg)
MAsh =   "   ash (g/kg)
LDM = litter dry matter content
L.. = litter...
Imlwt = maternal liveweight on day 2 of lactation
Fmlwt = maternal liveweight on day 12 of lactation
Fmewt = maternal liveweight minus gut contents
Illwt = litter liveweight on day 2 of lactation
Fllwt = litter liveweight on day 14 of lactation
Flewt = litter liveweight minus gut contents
natlit = the natural littersize

rat	treat	MDM	MCP	MGE	MEE	MAsh	LDM	LCP	LGE	LEE	LAsh
1	6	0.3966	490.0	25.89	340.0	100.0	0.580717	419	26.76	445	83
2	4	0.3407	600.0	22.88	215.0	129.0	0.272656	514	25.25	318	88
5	6	0.3957	514.3	25.66	321.6	115.4	0.278845	518	24.53	309	100
6	1	0.3958	491.8	26.58	395.8	94.3	0.556605	465	26.59	386	78
7	7	0.3653	557.8	25.39	288.8	101.0	0.336276	441	26.86	399	77
8	3	0.4175	444.0	27.26	419.0	83.0	0.313975	461	26.74	396	76
9	6	0.2917	473.0	25.34	381.0	104.0	0.285600	504	24.94	341	93
10	4	0.3718	560.0	25.01	288.0	112.0	0.340294	432	27.03	436	76
11	4	0.4252	452.5	27.17	405.2	113.6	0.526071	524	24.69	319	98
14	7	0.3871	487.1	25.13	356.0	113.3	0.500432	574	23.26	250	107
16	2	0.3507	656.0	22.16	155.0	144.0	0.554101	470	25.69	389	85
18	6	0.3645	546.2	24.32	285.6	101.0	0.326433	408	27.17	455	76
19	1	0.3637	541.0	25.35	295.6	106.1	0.284696	*	*	*	*
20	7	0.3678	552.8	24.37	278.8	113.6	0.244017	647	21.05	151	128
22	3	0.4004	498.0	26.09	371.0	92.0	0.270500	545	25.06	300	93
25	1	0.3621	556.0	24.94	301.0	94.0	0.278300	539	25.12	310	95
26	5	0.3007	618.0	22.92	204.0	110.0	0.576003	428	26.65	446	78
27	1	0.3139	645.0	23.23	198.0	115.0	0.555313	468	26.94	380	82
30	2	0.3381	689.0	21.42	140.0	156.0	0.290160	*	*	*	*
31	6	0.3519	602.0	22.44	217.0	134.0	0.245168	599	22.56	197	116
33	5	0.3579	569.0	24.08	265.0	108.0	0.606524	370	28.19	520	67
35	3	0.3443	608.0	23.48	232.0	110.0	0.551983	474	26.31	391	88
41	7	0.3235	708.0	21.03	99.0	144.0	0.244778	599	21.84	216	106
42	5	0.3158	550.0	24.18	286.0	99.0	0.319300	417	27.26	452	73
44	1	0.3734	522.0	25.25	330.0	94.0	0.298618	496	26.33	348	87
47	3	0.3552	586.0	23.67	262.0	109.0	0.510316	554	24.45	297	96
48	2	0.3464	604.0	22.76	210.0	128.0	0.284143	482	25.76	365	86
50	1	0.3620	555.0	23.95	279.0	113.0	0.299865	486	24.87	348	83
51	5	0.3996	465.0	26.33	380.0	88.0	0.580168	420	26.90	459	74
52	4	0.3787	494.0	25.02	357.0	115.0	0.284594	536	23.76	293	98
53	2	0.4713	653.0	22.15	166.0	120.0	0.283400	486	25.85	365	85
55	5	0.3529	593.0	23.56	259.0	105.0	0.342373	387	27.47	492	67
58	2	0.3344	702.0	20.68	109.0	143.0	0.536522	504	25.18	322	87

Appendix 6

The data from the dairy cow trial

The cows are identified by a code consisting of:

The feed type they were on; G = glucogenic (refered to as C in chapter 9)
L = lipogenic (refered to as S in chapter 9)
GL = a 50/50 mixture of G and L (C/S in chapter 9)

a three figure cow number.

a three letter code indicating the feeding level in periods 1, 2, and 3 respectively;
L = low, M = medium, and H = high level of feeding.

The data from which the levels of feeding were calculated (described on p. 156)

MY_0 = milk yield at time t_0 (kg/d)
t_0 = days post calving at the start of the trial.
a = peak yield. (kg)
t_{end} = days post calving at the end of the trial.
MY_{end} = predicted milk yield at the end of the trial = $a*exp^{(-0.035tend)}$ (kg/d)
Lwt = liveweight at the start of the trial. (kg)
DMRQ = the dry matter requirement to meet MY_{end} assuming no body state change (ARC 1980). (kg/d)
Milk fat and protein contents were taken to be 39 and 30 g/kg respectively.
Feed ME content = 10.7 MJ/kg.

Cow	MY_0	t_0	a	t_{end}	MY_{end}	Lwt	DMRQ
L177HML	21.8	128	34.12	238	14.8	552	11.8
L111MLH	26.6	82	35.44	195	17.9	512	13.1
L024LHM	20.0	97	28.08	207	13.6	522	11.0
L369HLM	22.4	98	31.57	208	15.2	504	11.7
L032MHL	26.8	88	36.47	195	18.4	580	13.7
L016LMH	19.0	99	26.87	209	12.9	480	10.4
G157HML	21.0	104	30.22	214	14.3	550	11.5
G052MLH	24.0	97	33.70	207	16.3	524	12.3
G170LHM	18.0	97	25.28	207	12.2	514	10.3
G228HLM	25.4	97	35.67	207	17.3	490	12.6
G006MHL	23.4	97	32.86	207	15.9	485	11.9
G087LMH	25.2	94	35.02	201	17.3	529	12.9
GL251HML	21.1	99	29.84	206	14.5	526	11.5
GL340MLH	20.9	98	29.45	205	14.3	605	11.8
GL249LHM	26.3	99	37.19	206	18.1	512	13.1
GL088HLM	25.2	86	34.05	196	17.1	481	12.5
GL353MHL	21.0	123	32.30	234	142	540	11.4
GL117LMH	21.6	113	32.08	223	14.7	460	11.2

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Milk yield (kg/d)

ROW	Day	L177HML	L111MLH	L024LHM	L369HLM	L032MHL	L016LMH	G157HML	G052MLH	G170LHM
1	1	20.2	20.8	16.8	22.8	22.0	17.2	18.0	16.2	15.8
2	2	21.2	21.4	15.6	21.0	23.2	16.8	17.6	19.2	15.6
3	3	20.2	21.6	15.4	21.0	22.6	15.6	15.6	16.4	15.6
4	4	19.8	20.2	14.8	20.6	20.0	14.2	17.0	15.4	15.4
5	5	20.2	21.6	*	20.0	23.0	14.0	17.0	17.6	14.8
6	6	19.8	19.8	13.0	19.4	22.0	14.4	16.0	14.4	13.6
7	7	18.8	19.2	14.0	18.0	22.0	13.2	16.0	17.0	13.0
8	8	20.0	19.4	13.0	19.8	*	13.4	18.0	16.2	14.2
9	9	19.0	19.6	13.6	20.0	21.8	13.8	17.0	15.2	13.2
10	10	18.6	19.8	13.4	18.8	21.0	11.2	16.4	17.2	13.4
11	11	19.4	19.8	14.8	17.6	21.4	12.4	16.2	*	13.8
12	12	19.6	20.4	13.8	18.6	21.4	13.2	15.2	*	14.4
13	13	19.0	19.6	13.6	17.4	20.0	11.0	15.2	14.2	13.0
14	14	18.6	18.0	12.0	17.6	18.2	10.6	13.6	15.8	13.0
15	15	*	18.8	12.4	*	20.4	12.6	15.2	18.0	12.6
16	16	18.4	19.0	12.8	*	20.8	10.8	16.4	18.4	13.2
17	17	18.2	20.4	12.4	18.6	18.4	12.8	16.6	17.0	13.6
18	18	19.2	19.8	12.4	18.0	18.8	11.8	15.4	16.0	11.4
19	19	18.0	18.8	13.0	19.8	18.6	10.6	16.0	15.4	12.0
20	20	18.6	18.2	12.4	17.6	18.4	10.4	15.0	16.4	11.8
21	21	17.4	17.0	13.2	18.4	20.8	10.0	14.4	15.2	11.6
22	22	18.8	17.6	12.6	18.8	18.8	11.4	13.2	15.6	11.4
23	23	17.6	18.2	11.8	18.2	18.8	10.0	13.8	13.0	11.4
24	24	17.2	18.2	13.0	17.8	19.4	10.0	13.4	13.4	10.4
25	25	16.8	17.6	12.4	17.8	19.2	11.2	13.4	13.4	11.6
26	26	17.4	17.0	12.4	16.2	19.4	10.6	14.0	13.4	12.0
27	27	18.8	18.8	12.0	18.8	20.0	10.0	13.4	10.6	10.6
28	28	17.0	15.4	11.6	16.0	19.6	9.0	13.2	14.4	11.2
29	29	16.4	16.6	14.2	16.0	19.0	9.4	14.2	13.2	10.6
30	30	15.6	15.4	11.8	16.6	19.6	9.8	13.8	15.8	12.4
31	31	14.8	16.0	12.8	15.0	19.8	10.6	12.8	*	11.8
32	32	14.4	15.4	13.4	15.0	17.8	11.0	11.0	16.4	12.6
33	33	15.6	17.0	13.6	14.0	20.4	10.4	13.6	11.0	12.4
34	34	14.8	16.2	14.0	13.6	19.4	11.4	12.8	15.2	11.6
35	35	13.6	14.8	12.6	11.6	16.4	11.2	12.0	11.0	12.2
36	36	14.6	14.8	14.6	13.8	19.0	9.4	13.0	11.2	11.4
37	37	15.4	13.8	13.2	13.8	18.8	9.2	10.6	10.8	11.8
38	38	14.4	15.4	13.0	13.6	17.8	9.4	11.6	12.2	11.8
39	39	15.2	16.0	15.0	14.2	18.6	9.6	12.6	9.4	11.6
40	40	15.4	15.4	15.0	14.0	17.6	9.8	12.0	10.2	13.0
41	41	13.0	13.6	14.4	13.2	17.0	9.2	11.8	7.6	13.4
42	42	14.8	15.0	15.6	13.2	16.6	10.0	10.4	10.0	12.0
43	43	14.2	13.8	14.0	12.8	16.6	8.2	11.0	7.0	11.2
44	44	14.2	14.6	14.8	12.0	18.8	11.0	11.4	10.0	11.6
45	45	14.4	14.6	14.6	13.0	20.2	10.8	10.8	9.0	12.6
46	46	12.4	15.4	14.2	12.4	19.0	10.4	10.6	9.8	11.8
47	47	14.2	14.6	14.8	13.0	15.8	8.4	12.0	11.6	11.2
48	48	13.6	14.4	14.8	12.2	17.4	8.8	10.4	7.8	12.0
49	49	14.0	14.0	13.6	12.0	16.6	9.2	10.0	7.4	11.4
50	50	13.4	13.8	13.8	12.0	18.6	9.0	11.4	10.0	11.0
51	51	13.8	13.4	13.8	12.2	16.4	8.6	8.6	7.8	11.4

52	52	12.8	14.0	14.6	12.0	16.6	8.8	9.6	9.2	10.6
53	53	14.0	13.8	14.0	*	17.2	8.8	10.2	7.4	11.6
54	54	15.0	14.4	13.6	12.2	17.4	8.4	10.4	9.2	11.4
55	55	13.8	13.2	13.6	11.0	16.0	7.8	*	*	11.6
56	56	14.2	13.6	13.4	10.2	15.8	8.0	*	*	11.0
57	57	13.6	*	12.8	11.6	16.0	9.2	10.2	8.4	11.2
58	58	13.4	13.6	14.8	11.8	17.4	8.4	8.8	8.0	10.2
59	59	13.6	14.2	14.2	11.8	17.8	8.8	9.4	9.0	9.8
60	60	12.4	15.0	13.2	12.6	15.4	8.2	8.8	9.2	10.4
61	61	11.6	15.8	13.0	11.4	13.8	10.2	8.0	9.0	9.6
62	62	11.0	15.6	12.0	12.2	13.2	8.6	9.0	9.2	11.2
63	63	10.0	16.2	*	11.6	12.2	7.6	7.6	8.4	8.8
64	64	11.8	16.8	11.8	12.6	12.8	8.4	7.0	10.6	9.6
65	65	11.6	16.4	12.0	13.0	13.4	9.4	7.4	8.8	9.2
66	66	11.4	16.2	12.0	11.4	14.0	8.4	8.6	8.0	10.0
67	67	11.6	16.4	12.2	12.4	13.2	8.2	7.4	8.0	9.8
68	68	11.4	14.2	12.4	12.2	14.2	8.2	7.8	7.4	9.0
69	69	11.0	16.0	11.8	11.0	13.4	9.4	8.8	8.4	9.4
70	70	11.6	14.8	11.8	12.4	13.8	7.6	9.0	5.8	9.0
71	71	11.8	15.2	12.0	10.0	12.8	8.6	7.8	7.2	8.8
72	72	11.0	15.0	13.2	12.0	10.8	8.2	7.0	7.0	8.4
73	73	10.4	15.4	12.0	11.4	12.6	8.6	7.4	5.8	8.6
74	74	10.0	15.2	11.0	11.4	12.2	7.4	7.4	6.0	9.0
75	75	11.6	15.6	12.4	10.8	12.4	8.0	7.4	6.8	8.0
76	76	10.6	15.6	11.6	11.2	12.4	8.6	8.0	6.0	8.2
77	77	10.2	15.2	11.6	10.0	11.8	7.8	7.4	6.6	8.4
78	78	10.6	15.0	11.6	10.8	11.4	8.0	6.8	7.6	8.2
79	79	10.8	15.2	12.2	10.8	12.0	8.2	7.2	7.4	8.8
80	80	10.4	16.4	12.2	10.6	12.0	7.8	7.6	6.6	9.0
81	81	10.2	16.6	12.0	11.2	11.0	8.6	6.6	6.8	8.0
82	82	11.2	15.0	11.2	11.4	10.6	7.8	6.6	7.0	8.0
83	83	10.6	15.0	10.6	9.0	11.2	8.0	6.2	7.6	8.0
84	84	9.0	*	10.4	10.8	11.2	*	6.4	6.0	7.6

ROW	G228HLM	G006MHL	G087LMH	GL251HML	GL340MLH	GL249LHM	GL088HLM	GL353MHL
1	24.6	22.0	23.0	20.8	19.6	24.4	22.8	20.0
2	24.6	19.8	22.4	18.8	18.0	23.8	23.2	19.6
3	23.6	18.8	22.2	20.8	17.8	23.8	24.4	20.0
4	24.0	19.6	22.2	20.0	18.2	22.2	23.4	17.0
5	24.2	20.0	*	19.4	18.0	22.6	24.0	17.0
6	23.8	19.0	21.4	20.6	16.6	20.8	22.6	17.4
7	23.6	18.0	21.2	19.0	17.4	20.4	21.6	18.6
8	25.0	18.4	21.4	21.6	17.8	20.2	24.2	17.8
9	24.0	17.8	21.4	20.2	17.8	19.4	23.8	19.0
10	23.6	17.6	21.6	20.8	15.6	19.6	22.2	17.2
11	23.0	17.6	21.0	20.8	17.8	20.2	22.4	18.0
12	22.4	17.4	21.0	20.6	17.4	20.0	23.6	17.6
13	23.8	17.6	20.6	21.6	16.2	19.4	23.0	19.0
14	21.4	17.2	20.4	19.8	16.2	20.0	22.4	18.4
15	22.4	18.0	20.0	19.6	16.8	19.6	23.4	19.2
16	22.6	16.2	20.2	20.2	*	20.0	24.0	18.0
17	23.0	17.0	21.6	21.0	*	19.6	23.4	18.4
18	20.4	17.0	18.8	20.0	*	18.8	22.4	18.0
19	21.6	16.8	20.0	20.0	14.8	19.4	21.4	17.8

20	21.6	16.4	*	18.4	16.0	18.4	21.4	17.0
21	21.6	15.8	18.6	17.2	15.8	17.8	21.4	17.0
22	21.4	16.0	19.8	18.4	16.2	18.4	20.4	15.6
23	21.2	15.4	18.8	19.8	17.2	19.0	21.4	16.6
24	21.0	16.0	19.4	19.2	15.6	18.0	19.0	16.4
25	20.6	15.8	19.8	18.8	17.0	17.0	21.6	16.0
26	*	15.6	19.2	18.4	16.6	17.8	21.0	16.6
27	21.6	14.2	18.2	*	14.6	16.8	22.0	16.0
28	20.8	14.8	*	*	15.8	16.8	21.4	16.6
29	*	14.4	19.2	18.6	16.2	16.6	21.0	16.4
30	21.0	15.2	18.0	16.6	12.8	16.8	20.4	15.8
31	19.0	15.6	20.2	18.0	12.4	18.0	19.2	17.0
32	17.4	15.4	19.8	16.6	15.4	17.4	18.4	16.0
33	18.4	15.2	20.2	17.0	13.4	18.2	18.0	*
34	16.8	14.6	19.8	17.4	13.0	18.4	17.6	*
35	17.0	14.6	20.0	17.4	14.4	17.8	16.0	*
36	16.4	15.8	18.2	16.6	12.0	18.2	*	15.8
37	*	15.6	19.4	17.0	12.6	17.4	15.4	16.4
38	17.6	15.4	*	17.4	13.8	17.6	15.4	16.4
39	17.4	16.0	19.6	17.4	13.0	19.4	16.2	16.8
40	16.4	15.2	*	17.2	*	19.0	16.2	15.8
41	16.0	15.4	19.8	17.0	*	17.4	15.0	16.4
42	17.0	15.2	18.8	16.4	12.6	18.2	15.2	16.6
43	16.6	15.2	*	17.0	12.6	17.8	16.2	15.8
44	17.6	15.8	19.4	17.0	14.2	18.0	15.2	14.4
45	17.2	16.0	19.8	16.6	12.0	17.6	15.8	15.4
46	16.8	14.4	18.0	16.8	13.2	17.0	15.8	15.0
47	15.6	14.6	19.2	16.2	11.6	17.6	15.2	15.4
48	17.2	*	20.2	14.8	11.6	17.8	15.0	15.4
49	17.6	15.4	18.0	15.6	12.4	18.2	15.2	15.2
50	17.6	14.4	17.2	16.4	12.4	17.0	15.4	14.8
51	16.8	14.8	17.4	15.8	11.2	17.2	15.6	14.6
52	16.4	14.8	18.4	16.6	12.0	16.0	14.6	16.2
53	16.4	14.4	19.2	16.4	10.0	17.2	13.8	15.0
54	17.8	15.2	17.2	*	11.2	16.8	14.2	16.6
55	16.6	15.6	18.4	15.4	11.4	16.4	14.4	13.8
56	16.2	13.4	16.6	14.8	11.0	16.6	12.6	15.6
57	17.0	13.8	16.4	16.4	10.2	16.0	13.2	15.8
58	16.2	14.4	18.0	15.6	11.4	16.2	12.8	16.4
59	17.2	13.6	18.8	15.2	*	15.6	15.4	14.4
60	15.8	12.6	18.8	15.8	*	14.6	14.6	14.2
61	16.2	12.0	18.0	13.0	*	14.8	*	13.6
62	15.8	11.2	17.4	13.8	11.0	14.6	15.4	12.8
63	16.0	9.6	17.8	12.8	11.6	14.6	13.6	11.8
64	16.6	10.8	17.0	13.2	9.4	14.2	14.6	12.6
65	15.6	10.4	18.4	13.0	10.6	13.0	14.6	12.6
66	16.4	10.8	*	12.6	11.4	13.6	14.6	12.0
67	17.2	10.8	*	13.6	11.0	13.8	14.0	11.8
68	16.6	12.4	18.2	13.4	8.6	14.0	14.2	10.4
69	16.0	11.0	18.6	15.0	11.2	14.2	15.0	11.0
70	16.8	11.0	17.0	*	10.2	13.6	15.2	12.6
71	16.6	10.6	17.8	14.4	9.6	13.8	14.4	10.4
72	17.4	11.4	17.8	12.8	9.2	13.6	14.8	10.8
73	17.0	10.6	17.6	13.8	10.0	13.6	14.8	9.4
74	17.6	9.4	18.0	12.6	9.4	13.4	14.4	10.8
75	15.6	11.0	18.6	13.4	9.4	12.8	14.4	10.0

76	17.0	10.6	17.4	13.0	8.8	13.2	13.6	10.2
77	17.6	9.6	17.0	13.0	9.0	13.0	14.6	11.8
78	16.0	9.6	17.0	13.0	8.0	12.4	13.2	9.4
79	16.8	10.0	17.8	11.4	7.8	12.4	13.6	8.6
80	15.2	9.8	16.6	11.4	8.8	12.0	14.6	10.4
81	15.4	10.0	16.4	11.2	8.2	*	11.8	10.4
82	14.4	9.6	15.2	10.4	8.0	*	13.6	10.4
83	15.6	10.0	17.8	10.2	6.6	12.0	13.2	10.4
84	14.6	9.8	15.4	12.0	5.4	10.8	12.0	10.4

Milk protein content (%)

ROW	day	L177HML	L111MLH	L024LHM	L369HLM	L032MHL	L016LMH	G157HML	G052MLH
1	0	3.14700	2.79991	3.17268	3.07917	3.18500	3.07490	3.29568	3.37580
2	0	3.16535	2.77634	2.89784	2.98223	3.27480	3.12141	3.31153	3.29727
3	0	3.17406	2.71632	2.83600	3.02576	3.24330	3.16714	3.33986	3.24528
4	0	3.26144	2.80000	2.99959	3.11011	3.30393	3.35258	3.43568	3.52488
5	0	3.24265	2.75700	3.00639	3.10375	3.36958	3.41333	3.46513	3.49000
6	0	3.28792	2.90271	3.16091	3.38178	3.34782	3.36425	3.39610	3.54973
7	0	3.46614	3.00133	3.20048	3.29057	3.29478	3.30917	3.46571	3.61794
8	0	3.57786	3.04625	3.28919	3.38348	3.53448	3.60179	3.64552	3.68164
9	0	3.51775	2.80927	3.24881	3.55250	3.50474	3.65304	3.65714	3.74500
10	0	3.31653	3.07623	3.42254	3.16720	3.54798	3.61200	3.60000	3.68122
11	0	3.46913	3.00473	3.47242	3.33000	3.57034	3.56579	3.62473	3.67830
12	0	3.55421	3.01750	3.53507	3.36056	3.64731	3.57833	3.50857	3.74426
13	0	3.69889	3.10699	3.51278	3.49111	3.73884	3.78522	3.63071	3.89468
14	0	3.66935	3.08403	3.47972	3.43581	3.71600	3.46615	3.67887	3.71327
15	0	3.58449	3.10913	3.54859	3.35556	3.68989	3.68957	3.75778	3.78267
16	0	3.59286	3.12536	3.52543	3.57600	3.73407	3.74545	3.72333	3.80108
17	0	3.67441	3.19000	3.45269	3.60259	3.72386	3.84609	3.75340	3.79067
18	0	3.58161	3.27373	3.44864	3.68688	3.80390	3.78756	3.74227	3.85217
19	0	3.60288	3.43471	3.40690	3.76032	3.90767	3.93571	3.86105	4.13930
20	0	3.65552	3.33317	3.45639	3.74516	3.89485	3.82098	3.80514	3.83625
21	0	3.77293	3.28986	3.52842	3.78667	3.63857	3.79429	3.78067	3.82310
22	0	3.72000	3.29263	3.49200	3.74175	3.99213	3.82513	3.87622	3.80733
23	0	3.70941	3.28375	3.56759	3.96082	3.92426	3.90214	3.81711	3.72000
24	0	3.70863	3.27482	3.53467	3.74357	3.99255	3.88721	3.79000	3.82000
25	0	3.67115	3.22429	3.47759	3.63269	3.93296	3.80333	3.77000	3.67207

ROW	G170LHM	G228HLM	G006MHL	G087LMH	GL251HML	GL340MLH	GL249LHM	GL088HLM
1	3.27687	2.80050	3.06152	2.99061	3.38282	3.21557	2.93374	3.20694
2	3.23896	2.89150	2.94173	2.87757	3.28000	3.04044	2.89342	3.26513
3	3.19275	2.79096	2.97652	2.78660	3.27951	3.07822	2.94155	3.20385
4	3.34594	2.92983	3.04182	2.88571	3.33125	2.94393	3.02386	3.33536
5	3.45697	3.00865	3.18709	2.90424	3.29052	3.14000	3.11859	3.39235
6	3.50632	3.05059	3.13556	2.91274	3.29890	3.08869	3.08234	3.38600
7	3.50333	3.12057	3.24714	3.09429	3.29176	3.19986	3.13867	3.36471
8	3.64052	3.28932	3.53038	3.01909	3.44553	3.18729	3.35400	3.40565
9	3.50789	3.19600	3.44395	3.05929	3.45384	3.23831	3.29195	3.20330
10	3.76429	3.07034	3.51403	2.85182	3.23687	3.05714	3.43517	3.46696
11	3.77102	3.08767	3.52364	2.98653	3.13940	3.06209	3.49370	3.42650
12	3.65793	3.20069	3.53750	3.11980	3.33793	3.07462	3.49021	3.62556
13	3.84228	3.43429	3.61026	3.07440	3.32195	3.12603	3.60047	3.62733
14	3.73881	3.28286	3.65500	3.15500	3.33476	3.14318	3.62435	3.56785
15	3.87339	3.27444	3.66200	3.19308	3.46138	3.22738	3.69989	3.65025
16	3.89931	3.26951	3.63250	3.24688	3.29341	3.08200	3.61244	3.87174
17	3.99536	3.26361	3.70735	3.35610	3.26333	3.15576	3.68402	3.99758
18	3.98885	3.48810	3.70000	3.31745	3.24253	3.22875	3.69918	4.10548
19	4.04667	3.45121	4.01280	3.53768	3.35697	3.38091	3.80278	4.10941
20	4.06714	3.49465	4.02389	3.50286	3.37235	3.34364	3.81217	4.06171
21	4.15767	3.50659	3.94107	3.53414	3.50000	3.36611	3.88127	4.12671
22	4.11200	3.43955	4.11936	3.52611	3.44349	3.29638	3.90060	3.92625

23	4.20372	3.48706	4.18250	3.57605	3.50455	3.34762	4.02984	4.08274
24	4.20850	3.47909	4.14377	3.59488	3.47643	3.25293	3.80743	4.06932
25	4.28923	3.48410	4.09653	3.49805	3.67746	3.17714	4.04333	4.23310

ROW	day	GL353MHL	GL117LMH
1	0	3.33727	2.99510
2	0	3.26624	2.84191
3	0	3.22753	2.79677
4	0	3.25667	2.75111
5	0	3.35979	2.93000
6	0	3.37867	2.85256
7	0	3.34171	3.01973
8	0	3.50100	3.07838
9	0	3.50805	3.02480
10	0	3.53375	3.04093
11	0	3.59000	3.08628
12	0	3.61321	3.12375
13	0	3.63321	3.11286
14	0	3.65680	3.15375
15	0	3.74368	3.12231
16	0	3.64973	3.19368
17	0	3.62947	3.14143
18	0	3.62000	3.45164
19	0	3.85635	3.40000
20	0	3.98339	3.34671
21	0	4.06102	3.37372
22	0	4.12648	3.36375
23	0	4.22875	3.37403
24	0	3.84615	3.23087
25	0	3.88400	3.38314

Milk lactose content (%)

ROW	day	L177HML	L111MLH	L024LHM	L369HLM	L032MHL	L016LMH	G157HML	G052MLH
1	0	4.88370	4.92252	4.74171	4.86376	4.99679	4.65286	4.79409	4.54268
2	0	4.88535	4.82247	4.76568	4.82223	4.94150	4.69141	4.69141	4.43636
3	0	4.86031	4.87263	4.60000	4.85435	4.85489	4.42714	4.73151	4.36640
4	0	4.73763	4.75273	4.53432	4.70187	4.87607	4.51645	4.69568	4.69046
5	0	4.81157	4.75433	4.71492	4.69875	4.82021	4.41833	4.65658	4.48927
6	0	4.68479	4.73177	4.65000	4.80000	4.83013	4.34137	4.63156	4.59757
7	0	4.71364	4.77193	4.51381	4.54455	4.69804	4.18583	4.64214	4.55079
8	0	4.79940	4.76875	4.48226	4.63292	4.78094	4.30464	4.62313	4.31343
9	0	4.71762	4.82000	4.53209	4.74375	4.71842	4.12913	4.54343	4.20000
10	0	4.60875	4.69442	4.40030	4.44200	4.70517	4.14400	4.54600	4.15341
11	0	4.55130	4.58176	4.39879	4.37667	4.67607	4.02754	4.41636	4.23245
12	0	4.50158	4.58875	4.52893	4.47916	4.71505	3.90167	4.24429	3.98043
13	0	4.49750	4.58082	4.45278	4.51833	4.65899	4.02696	4.30893	3.90745
14	0	4.45032	4.48052	4.44479	4.41581	4.67000	3.89231	4.33887	3.58694
15	0	4.38536	4.52870	4.44718	4.14556	4.66068	4.04553	4.27352	3.54822
16	0	4.41214	4.56739	4.47471	4.37800	4.61558	3.79136	4.14000	3.72541
17	0	4.48529	4.43571	4.49239	4.22167	4.59614	3.75174	4.18702	3.92533
18	0	4.45000	4.55267	4.38136	4.36875	4.59390	4.02293	4.22545	4.05174
19	0	4.47404	4.63635	4.46586	4.41113	4.54167	4.11881	4.06737	3.86186
20	0	4.44345	4.56439	4.26361	4.36742	4.52848	3.83659	4.10730	3.59400
21	0	4.47103	4.61740	4.36544	4.39842	4.35529	3.94571	4.09933	3.36207
22	0	4.43600	4.47263	4.46400	4.31175	4.50656	3.89568	3.97946	3.35700
23	0	4.44529	4.60500	4.44828	4.26367	4.48459	3.98548	3.97711	3.42800
24	0	4.39333	4.61879	4.40833	4.13429	4.43582	3.99419	3.93000	3.78000
25	0	4.39865	4.60929	4.38241	4.27077	4.46111	3.97000	3.67419	4.03345

ROW	G170LHM	G228HLM	G006MHL	G087LMH	GL251HML	GL340MLH	GL249LHM	GL088HLM
1	4.79494	4.90350	4.74533	4.75403	5.03117	4.92557	4.64496	4.64141
2	4.68831	4.95192	4.77755	4.67595	4.89740	4.87648	4.58018	4.78359
3	4.63870	4.88744	4.77370	4.70113	4.92631	4.76178	4.45165	4.64308
4	4.67377	4.93496	4.72773	4.70629	4.91000	4.75607	4.44257	4.50214
5	4.65576	4.91694	4.71314	4.74337	4.86093	4.69753	4.44990	4.63878
6	4.64632	4.91784	4.77556	4.66581	4.81080	4.75384	4.46000	4.70067
7	4.47807	4.79514	4.68831	4.54143	4.82094	4.71972	4.36378	4.67784
8	4.52017	4.84534	4.64354	4.61788	4.77000	4.62271	4.40671	4.67398
9	4.63263	4.84073	4.63026	4.58786	4.81163	4.61205	4.36000	4.73651
10	4.44238	4.75621	4.59506	4.53909	4.81072	4.37662	4.26621	4.60304
11	4.49966	4.63256	4.52818	4.45947	4.62518	4.27313	4.23565	4.48450
12	4.44828	4.65483	4.44750	4.47612	4.62724	4.18308	4.16278	4.42519
13	4.49965	4.60000	4.48195	4.47899	4.62463	4.13365	4.21639	4.37373
14	4.30390	4.68798	4.43000	4.44444	4.62917	4.09091	4.30388	4.30684
15	4.54411	4.68833	4.45000	4.40426	4.65552	4.08311	4.30000	4.37190
16	4.49172	4.63122	4.44000	4.39594	4.58878	3.73800	4.20000	4.35609
17	4.47464	4.55771	4.40382	4.48415	4.56333	3.56356	4.20134	4.26606
18	4.52423	4.62241	4.35213	4.45085	4.59646	3.65484	4.22836	4.29384
19	4.50467	4.64916	4.17480	4.52878	4.53303	3.68909	4.21278	4.34368
20	4.47061	4.69372	4.17556	4.37061	4.44647	3.50636	4.11174	4.32000
21	4.47907	4.76871	4.25357	4.50379	4.65099	3.52333	4.17000	4.44329
22	4.50400	4.73602	4.17702	4.49278	4.56905	3.49681	4.18224	4.37250

23	4.44651	4.66235	4.19125	4.54628	4.56455	3.47905	4.14516	4.38603
24	4.44025	4.64403	4.22264	4.48537	4.46250	3.44415	4.12429	4.29644
25	4.44436	4.63359	4.16143	4.46069	4.34220	3.13143	4.08056	4.21448

ROW	day	GL353MHL	GL117LMH
1	0	5.10545	4.79694
2	0	5.09247	4.64876
3	0	4.95000	4.57258
4	0	5.14667	4.35679
5	0	5.00628	4.54481
6	0	4.95167	4.58513
7	0	4.85732	4.58622
8	0	4.97913	4.59838
9	0	4.90244	4.48360
10	0	4.89500	4.41987
11	0	4.81963	4.33166
12	0	4.84107	4.44500
13	0	4.71786	4.39000
14	0	4.79720	4.47625
15	0	4.78412	4.36308
16	0	4.67560	4.35316
17	0	4.71316	4.15143
18	0	4.81211	4.39820
19	0	4.63429	4.42667
20	0	4.66712	4.35532
21	0	4.58163	4.38397
22	0	4.60537	4.38625
23	0	4.64813	4.38224
24	0	4.64692	4.15696
25	0	4.67200	4.44343

Milk fat content (%)

ROW	day	L177HML	L111MLH	L024LHM	L369HLM	L032MHL	L016LMH	G157HML	G052MLH
1	0	4.21150	3.58738	4.36878	4.00495	4.92214	3.22265	4.11136	4.24482
2	4	4.17515	3.61465	4.34973	4.00806	4.45840	2.57014	3.49776	3.13455
3	7	4.38344	3.59053	4.24600	3.62380	4.89205	3.64857	3.01068	5.47551
4	11	4.31021	3.46455	3.87473	3.75418	4.27495	2.79419	3.46667	1.05488
5	14	4.30084	3.56500	4.46590	4.85625	4.84611	3.57917	4.75566	3.11073
6	18	3.37198	3.53776	4.39091	2.62522	4.21205	4.21685	3.77312	3.79297
7	21	4.93068	4.12096	4.84714	3.90250	5.42076	4.55250	4.24000	3.20206
8	25	4.25190	3.79000	4.92387	5.50270	5.35010	3.96536	4.17299	3.86134
9	28	4.64362	3.83805	3.88881	2.31000	4.39053	3.80261	4.01486	3.38500
10	32	3.82333	3.90494	4.54149	5.04360	4.24607	4.09400	3.06400	5.82293
11	35	4.32493	2.96608	4.53424	3.40000	4.00337	4.17140	2.89582	2.56830
12	39	4.45579	3.78000	2.91880	3.84479	3.64043	3.95583	3.60286	3.04511
13	42	4.85972	4.21178	4.70833	3.80111	4.84188	4.71217	4.61536	3.97830
14	46	4.01839	3.96026	4.63028	4.28290	4.57200	3.91385	3.91226	4.15918
15	49	4.54333	3.89609	4.19915	8.18444	4.74841	4.08128	4.12019	3.05622
16	53	4.49714	3.07826	4.87800	4.15800	4.84907	4.28955	3.89000	2.59568
17	56	4.48059	4.44143	4.41657	4.67722	4.42976	4.47870	3.76957	4.68467
18	60	4.80613	3.11720	4.32000	4.04000	5.28701	3.35244	3.61455	2.84565
19	63	4.28923	4.15482	4.48069	4.14839	5.24650	3.96548	3.47211	3.67093
20	67	3.00345	4.07585	4.52377	3.68790	4.69091	3.93439	4.13541	2.72000
21	70	4.03000	4.43973	4.71211	4.69316	7.04314	3.83143	4.58956	2.25759
22	74	4.69600	4.26684	5.04400	4.71754	5.03164	3.82270	4.59000	2.75733
23	77	5.19706	4.04250	4.02655	3.81571	5.02049	4.06476	3.62395	2.10200
24	81	4.45294	3.40253	4.71667	3.27036	4.57545	4.77698	3.55273	2.47000
25	84	5.04192	4.59857	5.00741	4.66173	4.93519	3.56333	4.98806	3.09931

ROW	G170LHM	G228HLM	G006MHL	G087LMH	GL251HML	GL340MLH	GL249LHM	GL088HLM
1	4.37602	3.10600	3.81286	4.23026	3.42621	3.61536	3.66683	3.73041
2	4.36558	2.95292	3.76245	4.60027	3.88900	4.61835	3.94739	3.43000
3	4.46942	2.84400	3.66283	4.55642	3.90282	3.49211	3.88418	4.19308
4	4.14275	3.06409	3.35591	4.92971	2.99875	4.27865	3.15376	3.65321
5	4.25364	3.02486	3.67070	3.44478	3.35361	3.39847	3.59111	3.78870
6	4.29579	3.21510	3.75667	4.25468	3.58020	3.69606	3.81660	3.68167
7	5.03403	3.58314	4.27390	5.37429	3.79341	3.62875	4.36156	3.87118
8	5.28190	3.39340	4.31443	4.74879	4.09851	4.12094	4.37753	3.71741
9	4.62737	3.24109	4.04579	3.65714	4.15314	3.95964	4.05341	1.46642
10	3.66556	3.37586	3.85078	4.61515	3.77530	3.32636	3.90103	4.00913
11	4.37356	3.28465	4.28182	4.51347	3.66964	3.81134	4.02000	4.41500
12	4.12086	3.54586	4.36750	4.88429	3.45828	4.13308	3.83443	4.33407
13	4.37456	3.55238	4.67364	4.86697	4.12756	3.62810	4.40791	4.06613
14	4.86203	3.88286	4.34333	4.58778	3.92810	3.23864	4.29153	4.61253
15	4.31607	3.23444	4.04200	4.55862	3.24241	3.72836	3.54443	4.31139
16	4.17793	3.18829	4.20500	4.98292	3.92073	3.17800	3.96686	3.72783
17	4.29071	3.57759	4.44353	3.51146	3.81333	3.48881	4.36390	4.58303
18	4.16038	3.81671	4.49000	4.16340	3.77734	3.38953	4.40521	4.63397
19	3.91333	3.47024	4.60320	4.52622	3.91636	3.86818	4.37222	3.84794
20	4.51959	3.41465	4.12056	4.73714	3.99941	2.63364	4.45522	4.48971
21	4.72954	3.76635	4.54036	4.13678	4.01563	2.86611	4.63535	3.75934
22	4.52400	3.85205	4.87808	4.74222	4.21095	3.04936	4.64373	4.50750

23	4.86558	3.46412	4.82500	3.96756	4.06242	3.49048	4.58563	5.12343
24	4.49025	2.57649	4.63000	5.70878	3.94857	3.17415	4.72857	4.06153
25	5.20949	3.73346	5.00469	5.03598	3.60915	2.90429	4.74370	4.87586

ROW	day	GL353MHL	GL117LMH
1	0	3.92364	3.50102
2	4	3.99259	3.48371
3	7	3.22742	3.24065
4	11	4.73944	5.19951
5	14	3.58011	3.55704
6	18	3.93367	3.53359
7	21	4.31415	3.89946
8	25	4.19175	3.78074
9	28	3.72415	3.54413
10	32	3.72500	3.32600
11	35	3.71741	3.07331
12	39	3.75107	3.26125
13	42	3.89714	3.41260
14	46	3.91320	3.35250
15	49	4.17309	3.44692
16	53	3.43533	3.65105
17	56	4.40263	3.21214
18	60	4.17380	2.23443
19	63	4.66810	3.46000
20	67	4.45712	3.53506
21	70	4.71408	3.70500
22	74	4.83870	3.75125
23	77	4.94688	3.17821
24	81	4.52615	1.72652
25	84	4.73600	3.60157

Liveweight (kg)

ROW	Day	L177HML	L111MLH	L024LHM	L369HLM	L032MHL	L016LMH	G157HML	G052MLH	G170LHM
1	0	530	486	501	499	547	467	560	509	504
2	1	533	492	496	501	548	464	565	511	498
3	7	526	485	478	491	540	440	559	500	484
4	8	528	486	483	496	540	444	562	499	483
5	14	531	485	466	497	554	435	559	506	468
6	15	529	486	462	500	548	439	561	507	472
7	21	524	489	468	495	552	438	564	508	474
8	22	530	490	470	494	547	438	566	505	473
9	28	535	498	472	511	548	441	564	519	472
10	29	530	504	471	510	547	441	565	523	466
11	35	532	481	485	498	556	456	560	512	480
12	36	524	488	490	500	561	457	559	520	490
13	42	522	467	488	487	551	452	558	516	483
14	43	520	468	484	492	552	450	556	521	490
15	49	522	470	490	490	550	455	557	519	485
16	50	520	471	485	486	552	454	559	518	484
17	56	527	474	492	498	570	462	561	507	498
18	57	526	477	497	500	563	464	560	514	496
19	63	521	524	493	501	555	479	558	548	502
20	64	524	519	493	501	560	481	564	546	500
21	70	530	509	482	504	554	484	566	555	496
22	71	525	508	484	500	555	482	563	550	500
23	77	528	510	486	506	556	483	564	542	500
24	78	526	509	482	502	555	484	564	540	502
25	84	526	506	487	495	558	486	570	550	499
26	85	525	502	490	500	552	485	570	546	502

ROW	G228HLM	G006MHL	G087LMH	GL251HML	GL340MLH	GL249LHM	GL088HLM	GL353MHL	GL117LMH
1	423	444	494	491	580	479	457	530	448
2	430	444	493	499	589	486	465	533	449
3	412	437	480	483	575	475	459	531	421
4	414	443	476	482	581	469	464	528	428
5	433	445	476	488	571	464	467	522	421
6	430	441	475	485	572	463	466	527	421
7	430	450	474	481	573	466	464	527	421
8	426	449	473	485	575	465	466	524	420
9	438	445	475	484	583	471	475	527	428
10	440	452	473	485	583	470	469	524	432
11	419	468	481	488	568	498	450	528	441
12	422	470	485	490	568	494	449	538	441
13	410	457	479	475	554	491	444	530	431
14	405	454	477	473	561	496	444	530	435
15	403	460	480	459	562	492	444	538	439
16	410	469	484	476	564	493	444	534	438
17	414	479	491	488	572	505	448	517	445
18	418	481	496	490	574	506	455	527	453
19	416	459	495	482	580	500	460	530	460
20	424	466	498	480	600	502	460	540	454
21	428	468	507	479	600	509	468	532	462

22	428	468	505	472	602	500	462	530	456
23	431	469	505	476	597	497	465	522	461
24	426	470	508	476	598	498	460	520	456
25	431	476	520	487	602	514	471	527	462
26	430	476	513	481	599	500	466	530	460

Condition Score

These values are condition score multiplied by 16, to give whole numbers.

ROW	Day	L177HML	L111MLH	L024LHM	L369HLM	L032MHL	L016LMH	G157HML	G052MLH	G170LHM
1	3	16	14	12	12	16	12	14	12	18
2	20	16	12	10	14	16	11	14	12	17
3	31	16	12	13	13	16	10	12	10	18
4	44	16	13	12	13	16	12	14	13	18
5	59	16	12	10	12	14	11	14	15	20
6	72	16	13	12	13	16	13	14	15	19
7	87	16	13	12	13	14	15	14	16	17

ROW	G228HLM	G006MHL	G087LMH	GL251HML	GL340MLH	GL249LHM	GL088HLM	GL353MHL	GL117LMH
1	9	9	16	14	16	13	14	16	14
2	11	12	15	13	18	11	13	17	12
3	9	10	15	14	20	12	13	16	14
4	10	12	15	13	18	13	14	16	13
5	9	10	15	13	20	13	13	15	14
6	10	9	16	13	20	11	14	17	15
7	9	10	15	14	18	13	14	20	14