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SOME EFFECTS OF GENOTYPE
ON THE CONVERSION OF PASTURE
TO MILK BY FRIESIAN COWS

A thesis presented in partial fulfilment
of the requirements for the degree of
Doctor of Philosophy in Animal
Science at Massey University

by

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1982

13320-88

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ABSTRACT

The New Zealand Dairy Industry has been aiming to bring about genetic improvement of dairy cattle by the use of genetically superior (progeny tested) bulls in the Artificial Breeding Scheme. There is evidence to show that there has been a genetic improvement in the level of milk fat production per cow, but little evidence to show the mechanisms by which the increase has been achieved.

At present the genetic merit of a New Zealand cow for milk or milk fat production is measured by her breeding index (BI). The main objective of the work was to determine the mechanisms whereby cows of high BI produce more milk than cows of low BI.

A total of 40 Friesian cows with high BI (approximately 125) or low BI (approximately 100 which is equivalent to the average cow in 1960) were identified and purchased from New Zealand dairy farmers. Experiments were carried out to determine the performance of high and low BI cows; when grazed as one group; when fed cut pasture individually in stalls at two levels of feeding; and when subjected to complete energy and nitrogen balances whilst lactating and non-lactating.

Over the whole lactation, high BI cows produced more milk fat and gained less liveweight than low BI cows. The difference between BI groups in milk fat production was in close agreement with the expected differences based on BI's. Differences in liveweight changes between genotypes were not measurable in the short term (approximately five weeks) feeding experiments.

One exception was in late lactation when high BI cows partitioned significantly ($P < 0.10$) more metabolisable energy to milk at the expense of body tissue than the low BI cows.

The two genotypes had similar intakes of fresh cut pasture offered ad libitum in stalls. However high BI cows ate, on average, 7% more pasture per unit metabolic liveweight than low BI cows, but the differences between genotypes in intake were significant only in two of the four indoor feeding experiments ($P < 0.05$, $P < 0.10$).

There were no significant differences between BI groups in their ability to metabolise feed energy and in their efficiency of use of metabolisable energy (as measured by heat production at a given energy intake). There was one anomalous result during restricted feeding in early lactation when high BI cows produced less heat ($P < 0.05$) at a common energy intake than low BI cows. Differences in nitrogen balance between genotypes were small and inconsistent.

The feed required to maintain body condition and to promote a gain of body condition during the dry period was similar for both genotypes.

The statistical methods developed in the course of analysing the experimental data were outlined in detail because it was considered that the analyses were more appropriate than those normally used.

It was concluded that high BI cows produced more milk fat because they ate more and partitioned a higher proportion of their metabolisable energy intake to the synthesis of milk rather than to liveweight gain, than the low BI cows.

The implications of the results were considered by making some preliminary predictions about the likely effect of genetic merit on farm productivity.

ACKNOWLEDGEMENTS

It is a pleasure to acknowledge the following people and organizations:

My supervisors, Dr. A.W.F. Davey, Dr. C.W. Holmes, and Dr. D.D.S. Mackenzie, who were always willing to discuss ideas and who assisted in the conduct of the experiments, and in the writing up of this thesis;

Dr. A.W.F. Davey who had initiated and set in motion the research programme outlined in this thesis, prior to the candidate's arrival at Massey University.

Professor R.J. Townsley who perceived that the normal methods of statistical analysis used in nutritional and energy balance experiments were inappropriate and who spent considerable time developing more appropriate methods of analysis;

Mr. A. Gilmour for providing technical help in analysis of the data;

Dr. C.W. Holmes who designed and supervised the construction of the hoods used in the calorimetry experiments, and with assistance from Mrs. Y. Moore, tested and operated the calorimeters during the calorimetric balance experiments.

To members of the Dairy Husbandry Department, Massey University who contributed ideas and practical assistance;

Mr. J. Raven, Mrs. Y. Moore, Mrs. J. Rumbal and Miss J. Frain for skilled assistance with laboratory analyses and analyses of milk samples.

Messrs B. Parlane and R. McLenaghan for harvesting the pasture, and for assisting with the handling and feeding of the stock;

Messrs A. Lowe, S. Harmer and G. Fowlds for milking and management of the cows;

Mr. J.W. Hughes of the Ruakura Agricultural Research Centre who developed the urine harnesses used in the calorimetric balance experiments.

The New Zealand Dairy Board for identifying the animals and to New Zealand dairy farmers for providing the animals;

Mrs. A. Bull and Mrs. C. Castle for typing this thesis;

The Massey Agricultural Research Foundation, the Town Milk Producers Federation, and the New Zealand University Grants Commission who provided financial assistance for the project;

The Victorian Department of Agriculture who granted me leave with pay, and the Dairy Research Committee who granted me a postgraduate education grant;

This thesis is dedicated to my wife Elizabeth in appreciation for her support and encouragement, and for the creditable way in which she has brought up our young family during the three years of this study.

CHAPTER ONE

A REVIEW OF THE RELATION BETWEEN GENETIC
MERIT OF COWS FOR VOLUME OF MILK PRODUCTION
AND THEIR GENETIC MERIT FOR EFFICIENCY OF
MILK PRODUCTION

1.1

INTRODUCTION

There is good evidence that genetic improvement of New Zealand dairy cattle has been brought about by the use of genetically superior (progeny tested) bulls in the Artificial Breeding Scheme of the Dairy Board Farm Production Division (Wickham et al. 1978).

The reasons for the resultant production advantages are largely unknown for dairy cattle grazed mainly on pasture, and Bryant (1978) has pointed out that quantifying the major components of cow efficiency is essential for the further exploitation of cows grazed on pasture.

The lack of experimental evidence on the question of genetic merit in relation to efficiency of milk production was also highlighted by the debate surrounding the high production of the Jersey herd at Ruakura No. 2 Dairy (Campbell et al. 1977; Karlovsky, 1977 and 1978; Jury, 1977; Campbell, 1978; Linton, 1978).

In this review the evidence of genetic improvement of dairy cattle grazing pasture is summarised, with the main emphasis on the effects of genetic selection on the efficiency of milk production. In the absence of critical evidence for the grazing cow, two approaches have been taken to determine the factors which might be important in explaining the increased levels of milk production due to genetic improvement.

Firstly, the characteristics of the high yielding cow are outlined, drawing upon evidence from calorimetric balance studies, and from nutritional studies involving different planes of nutrition.

The second approach is to examine the results of genetic selection experiments. The only data available were confined necessarily to experiments where cows were fed on concentrates and conserved roughages.

Interest is focused on energy, although protein requirements and the status of high yielding cows with regard to hormones and blood metabolites is also considered.

Finally an outline of proposed experiments is made.

1.2 EVIDENCE OF GENETIC IMPROVEMENT

Emphasis in this section is placed on the evidence of genetic improvement for cows grazing pasture as the main diet. Most of the evidence comes from New Zealand because of its well-documented history of genetic improvement. Genetic progress has been made in other countries, for example, Powell et al. (1977) outlined progress made in the United States.

An early analysis of the genetic structure of the New Zealand dairy herd indicated that genetic differences between herds were likely to be small (Stewart, 1952). Further evidence to support this fact was obtained by Brumby (1961). Twenty herds in which cows achieved high levels of production and twenty herds in which cows achieved low levels of production were identified within an 80 km radius of Ruakura Agricultural Research Station. Two heifer calves from cows of productive levels close to the herd average were taken, each year for three successive years from each of the 40 herds, reared and subsequently milked at Ruakura. The milk fat production of all the cows was similar indicating that the large difference in production between the herds, in production per cow, was mainly a result of management differences, not genetic differences.

One of the first indicators of genetic improvement was reported by Carter (1964). The No. 2 Dairy herd at Ruakura had been artificially bred since the inception of artificial breeding in New Zealand in the late 1940's, and could therefore be regarded to be of high genetic merit at the time. A selection of cows covering a wide range of production levels were brought to Ruakura, grazed at No. 2 Dairy for three years and

their production was compared with that of the Ruakura cows. The average milk fat productions over the three years were 172 and 148kg for Ruakura cows and the farmer's cows respectively. The measured difference of 24kg milk fat was similar to the expected difference of 20kg which was estimated from the pedigrees of the Ruakura cows, and which was a measure of their expected superiority in dairy merit above the "average" dairy cow.

Further evidence of genetic improvement was outlined by Wickham et al. (1978). By comparing the herd test production of cows of similar age which had been sired through either artificial breeding or natural mating the production differences attributable to the two sources of herd sires were estimated. The average milk fat yield of the artificially bred progeny minus the milk fat yield of the naturally bred progeny ranged from 7kg in 1970-71 to 11kg in 1975-76. The measured differences in milk fat yield between the two sources of herd sire were in agreement with the predicted differences based on the difference in genetic merit of the sires involved.

The measure of genetic value used in New Zealand is the breeding index (BI). For a female a BI is a weighted combination of her own production records and the BI's of her sire and dam (see Appendix 2.1 for method of calculation of breeding indexes). The BI for a male is a weighted combination of daughter productions and the BI's of his sire and dam. The New Zealand Dairy Board, in their Annual Farm Production Report, record the changes that have occurred in BI values for cows in the New Zealand dairy herd and proven bulls used in artificial breeding.

Selected data from the 1979/80 Report are presented below.

Season	Average BI of proven bulls	Average BI of cows:	
		Sired by proven bulls	All other cows
1953/54	107	100	100
1959/60	113	105	100
1969/70	124	110	104
1979/80	134	118	110

Clearly there has been an improvement in genetic merit (as assessed by BI) of the proven bulls used in artificial breeding since 1953/54, and consequently an increase in the genetic merit of the cows sired by proven bulls. The average BI of all other cows has increased also, but not as much as with the cows sired by proven bulls. How much of the increase in genetic merit of all other cows is because of the fact that artificial breeding has been used somewhere in their ancestry is not known.

Wickham (1979) provided additional evidence that the estimated breeding index value of a cow is a good measure of her genetic merit for milk fat production.

Of particular interest is the project currently being carried out in Poland comparing the productivity of cows sired by Friesian bulls from ten different countries (Stolzman, 1982; Jasiorowski, et al. 1982). The project commenced in 1974 through co-operation between the Food and Agriculture Organisation and breed organisations and research institutes in ten countries, namely Canada, Denmark, Federal Republic of Germany, Israel, Netherlands, New Zealand, Poland, Sweden, United Kingdom, and United States of America. Semen from about 40 bulls

from each country was used on a total of more than 30,000 Black and White cows in Poland. The young bulls from each country were selected at random from those bulls being tested within the Artificial Breeding schemes and from which the final selection of top bulls for widespread use would be made.

Production results for approximately 5,500 first-cross (F_1) heifers when fed mainly on conserved roughages show that the genetic merit in terms of milk fat and milk yield of New Zealand dairy cattle is among the highest in the world (Table 1.1); although the results cannot be used to show that there has been genetic improvement in New Zealand. Similar comparisons made in Poland under more intensive feeding conditions have produced similar results.

It can be concluded that selection applied to the dairy cattle population in New Zealand has produced a genetic increase in milk fat production. There is also evidence that the estimated breeding index of an animal is a good estimate of its genetic merit.

Table 1.1 Milk and milk fat yields in the first lactation of first-cross heifers sired by semen from bulls in various countries expressed as deviations from the average (from Stolzman, 1982).

Country of sire's origin	Milk fat yield * (kg)	Milk yield † (kg)
U.S.A.	+10.8	+347
New Zealand	+10.6	+180
Israel	+ 8.0	+258
Canada	+ 7.3	+216
Sweden	- 0.6	- 21
U.K.	- 1.7	- 45
Denmark	- 5.0	-154
Germany	- 6.0	-129
Netherlands	- 6.6	-215
Poland	-16.8	-437

* Average milk fat yield = 146kg

† Average milk yield = 3615kg

1.3 CHARACTERISTICS OF THE HIGH YIELDING COW

The productive ability of cows reviewed in this section is attributable both to genetic and environmental factors and it is not possible for individual cows to determine the proportion of each of the components. A measure of the average contribution to the productive ability of a group of cows attributable to genotype, can be made from genetic selection experiments which are reviewed in Section 1.4.

What is meant by a high yield?

Broster and Alderman (1977) discussed the problems of defining a 'high-yielding' cow, but considered that a high yielding cow could be reasonably regarded as producing yields of the order of 7,000kg/lactation under the feeding systems that prevail in Europe.

For grazing cows there is much less emphasis on production per cow. The pasture supply varies markedly in both quantity and quality throughout the year, resulting in wide fluctuations in the level of feeding of cows. In this situation even the cows with the highest yields could be described as "low-yielding" when compared with cows in the U.K. or U.S.A. That milk yields are relative to environmental conditions, was clearly demonstrated by Stichbury (1979), using evidence from the comparison of different strains of Friesian cattle in Poland. There was a wide range in the production of the dams of the bulls from which semen was taken and used on Friesian cows in Poland, the range

being as follows;

	Milk fat/lactation
U.S.A. dams (highest producers) (in U.S.A.)	400kg
N.Z. dams (lowest producers) (in N.Z.)	209kg
The production of the bull's daughters was (from Table 1.1)	
U.S.A. daughters (in Poland)	157kg
N.Z. daughters (in Poland)	157kg

Obviously the higher production of the American dams was not due to breeding, but to environmental differences. An appropriate definition of a 'high-yielding' cow might be therefore :

"The cow that can produce the most milk under a given set of environmental conditions."

1.3.1 Evidence from energy metabolism studies.

Before outlining characteristics of high yielding cows using data from energy balance studies, it is essential to consider the experimental methods used in studies of energy metabolism, and the possible limitations of the evidence derived from energy balance experiments.

The partition of energy within the animal is shown in Figure 1.1.

The major technique used to study energy metabolism is the calorimetric balance (ARC, 1980). In a calorimetric balance, direct measurements are made of the gross energy intake and the energy of faeces, urine, methane, and milk (for lactating animals).

Total heat production is usually measured indirectly from respiratory gas exchanges and includes heat resulting from body tissue catabolism, heat produced as a result of maintenance processes, heat produced during the conversion of dietary energy to milk, and also heat produced from rumen fermentation (Moe et al., 1971).

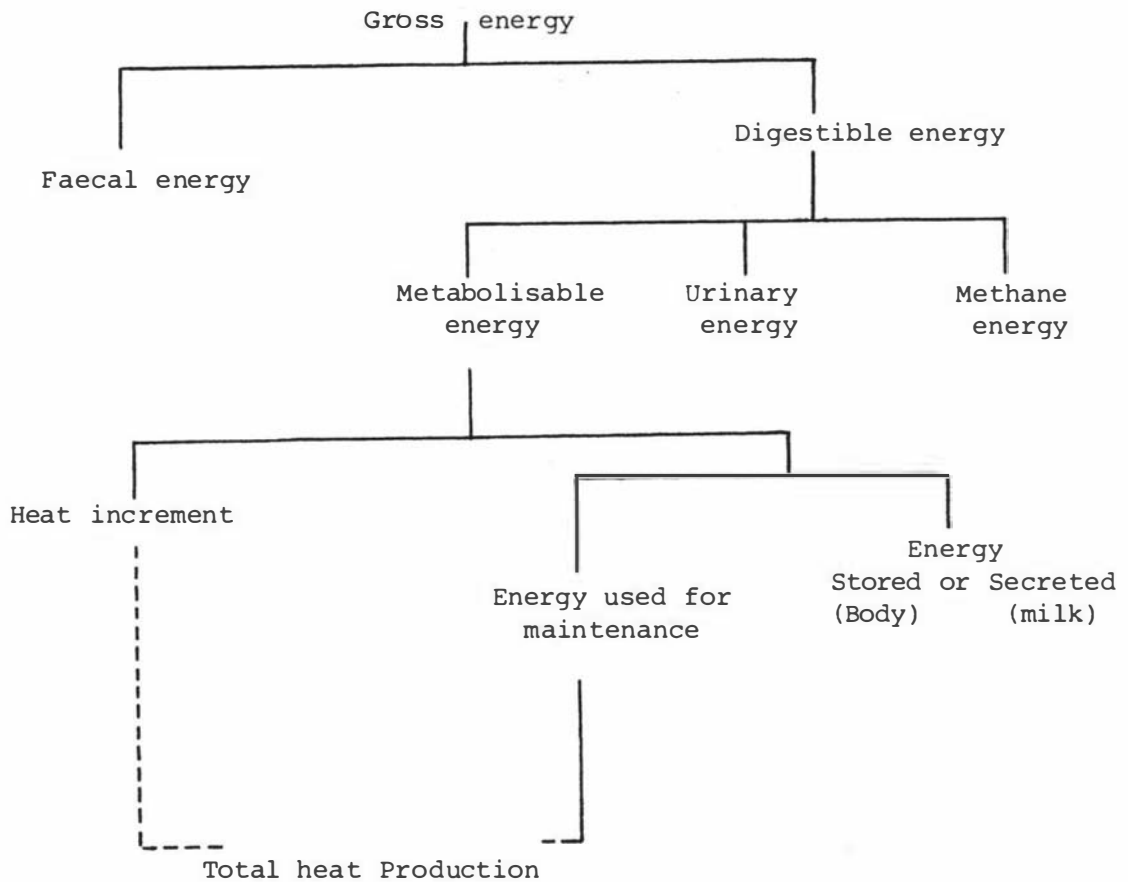


FIG.1.1: Partitioning of feed energy within the animal

Change in body tissue energy is not measured, but is estimated by difference; namely gross energy intake minus the sum of faecal, urinary, methane, heat, and milk energies. Tissue energy is subject therefore to

the cumulative errors of measurement of the other five energy outputs and is associated with the largest error of determination of any of the measurements (Moe et al. 1971).

There are also considerable problems associated with the statistical analysis of calorimetric balance experiments. For example, Moe et al. (1971) outlined the methods of obtaining partial regression coefficients which relate components of energy balance to metabolisable energy intake by multiple regression techniques. The following model was used by them to represent the relation between dietary energy intake and the use of energy by the lactating animal:-

$$ME = k_1 MBS + k_2 \text{milk} + k_3 TISP + k_4 TISN + a$$

where

- ME = metabolisable energy
- MBS = liveweight in kg raised to the 3/4 power
- TISP = body tissue energy gain
- TISN = body tissue energy loss
- milk = milk energy
- k = partial regression coefficient
- a = a constant term.

Moe (1981) stated : "With several physiological processes occurring simultaneously, however, estimates of partial efficiency for each process are clouded by experimental error and also by necessary assumptions. The most troublesome assumption concerns maintenance".

Some of the statistical problems associated with multiple regression analyses such as those outlined above

are:-

- there is co-linearity between the independent variables,
- some of the variables have large errors, namely ME and TISN and TISP; in addition; errors which are common to both independent and dependent variables,
- it is likely that there will be correlations between the errors of the variables.

Van Es (1976) pointed out that :

"The resulting regression equations are very well suited to predict the dependent variable, but for statistical reasons their regression coefficients lack physiological significance."

Obviously the nature of calorimetric experiments and the statistical methods used to provide many of the estimates of efficiency, make interpretation of energy balance data very complex.

Evidence from energy balance experiments which relate to high producing cows is considered in terms of:-

1. The ability of cows to metabolise feed energy.
2. The intake, efficiency of use and partitioning of metabolisable energy to milk and liveweight gain.

1.3.1.1 Metabolisability

The metabolisability of the gross energy of feed (q) is defined as the metabolisable energy of a diet divided by the gross energy (ARC, 1980). The metabolisable energy (ME) of a feed is its gross energy less the energy lost in the faeces, urine, and methane. The main determinant of metabolisability is digestibility.

The digestibility of a feed is generally estimated at the maintenance level of feeding using non-lactating, non-pregnant sheep or cattle. However the values for digestibility measured at four to five times maintenance intake with milking cows were lower than those values made at maintenance intake (see Tyrrell and Moe, 1975, for references). There was, on average, a depression of four % units of digestibility for each increase in intake equivalent to maintenance for forage/concentrate diets (Tyrrell and Moe, 1975).

One of the consequences of the decrease in ration digestibility with increasing intake was the need to increase the allowance of energy per kg milk with increasing production (NRC, 1966).

There is limited evidence available concerning the effect of increasing intake on the digestibility of pasture. Digestibility measurements made with lactating dairy cows (Trigg et al. 1980) and sheep (Blaxter, 1962; Rattray and Joyce, 1969; Grainger et al. 1982) fed on pasture at different intakes have confirmed the decline in digestibility with increasing intake.

Hutton (1963) compared the digestibility of pasture fed to lactating and non-lactating identical twin cows over a 36 week period, and recorded a within-set difference in digestibility averaging 0.7 percentage units in favour of the non-lactating group. The non-lactating twins, on average, consumed only 65% of the intake of their lactating co-twins. At a later date, Hutton (1971) commented:

"Because plane of nutrition has a much smaller effect on apparent digestibility of pasture herbage than with most rations available in the feedlot, this effect is much less significant in the New Zealand environment."

Whilst at higher feeding levels the losses in faecal energy increase, methane and urinary energy losses, expressed as a percentage of gross energy, or digestible energy, decrease (Blaxter, 1969).

Whether or not the metabolisable energy of a diet falls with increasing intake will depend on the extent to which the increase in faecal energy losses are balanced by the decrease in methane and urinary energy losses. Trigg et al. (1980) found that decreases of energy digestibility at higher levels of intake were compensated for by concomitant reduction of energy loss through urine and methane excretion. The metabolisability of the pasture in the experiments carried out by Trigg et al. (1980) ranged from 0.54 to 0.61. Van Es (1975) suggested that the decrease in methane and urine energy losses with increasing intake is partly due to an increased rate of passage of digesta resulting in lower methane percentages, and partly to the lower percentage of the food N excreted as urea at higher milk yields.

Blaxter (1969) combined the data relating metabolisability of the feed to feeding level and its effect on digestibility into a single equation.

$$q_L = q_m + (L-1) (0.20(q_m - 0.623))$$

where

q_L = metabolisability of gross energy at any feeding level,

q_m = metabolisability of gross energy at maintenance feeding level,

L = multiple of maintenance feeding level.

From the above equation, for diets with a metabolisability of 0.6 at maintenance intake, there is no change in metabolisability with increasing intake, but when the metabolisability at maintenance is less than 0.6, metabolisability decreases with increasing intake (Table 1.2).

Table 1.2 The effect of intake on metabolisability of different quality diets predicted from the equation of Blaxter, (1969).

Metabolisability at maintenance intake	Metabolisability at twice maintenance intake	Metabolisability at four times maintenance intake
0.60	0.60	0.59
0.55	0.54	0.51
0.50	0.48	0.43 *

* it may not be possible to have an intake of four times maintenance at this 'q' value.

A more direct analysis of 72 experiments in which level of feeding was varied, confirmed the occurrence of depressions of metabolisability with poorer diets (ARC, 1980).

In addition to the evidence presented above, which mainly concerned the effect of feeding level on metabolisability, Flatt (1980) commented, based on energy metabolism studies done at Beltsville, that there are only small differences between individual animals in their losses of energy in faeces, urine, and methane. Thus it appears reasonable to suggest that differences in the ability of high and low producing cows to metabolise feed energy are small when reasonable quality diets are offered. However there is no direct evidence to verify this suggestion.

1.3.1.2. The intake, efficiency of use, and partitioning of metabolisable energy.

Calorimetric balance studies have played an important role in establishing the nature of the relation between metabolisable energy intake (MEI) and the output of energy in milk.

It has been shown that there is a linear relation between total energy balance (milk plus tissue energy) and MEI (Moe and Tyrrell, 1975; Van Es and Van der Honig, 1979). However with increasing level of nutrition, for an individual animal, there is an increasing diversion of energy away from milk into body tissue, thus milk production does not increase linearly with MEI (Figure 1.2). Moe and Tyrrell (1975) provided evidence which confirmed these relations between MEI and the components of total energy balance. When milk production was plotted against MEI there was a wide scatter of points because the proportion of energy for body tissue was not considered. However, total energy balance (milk plus body tissue) was related closely and linearly to MEI.

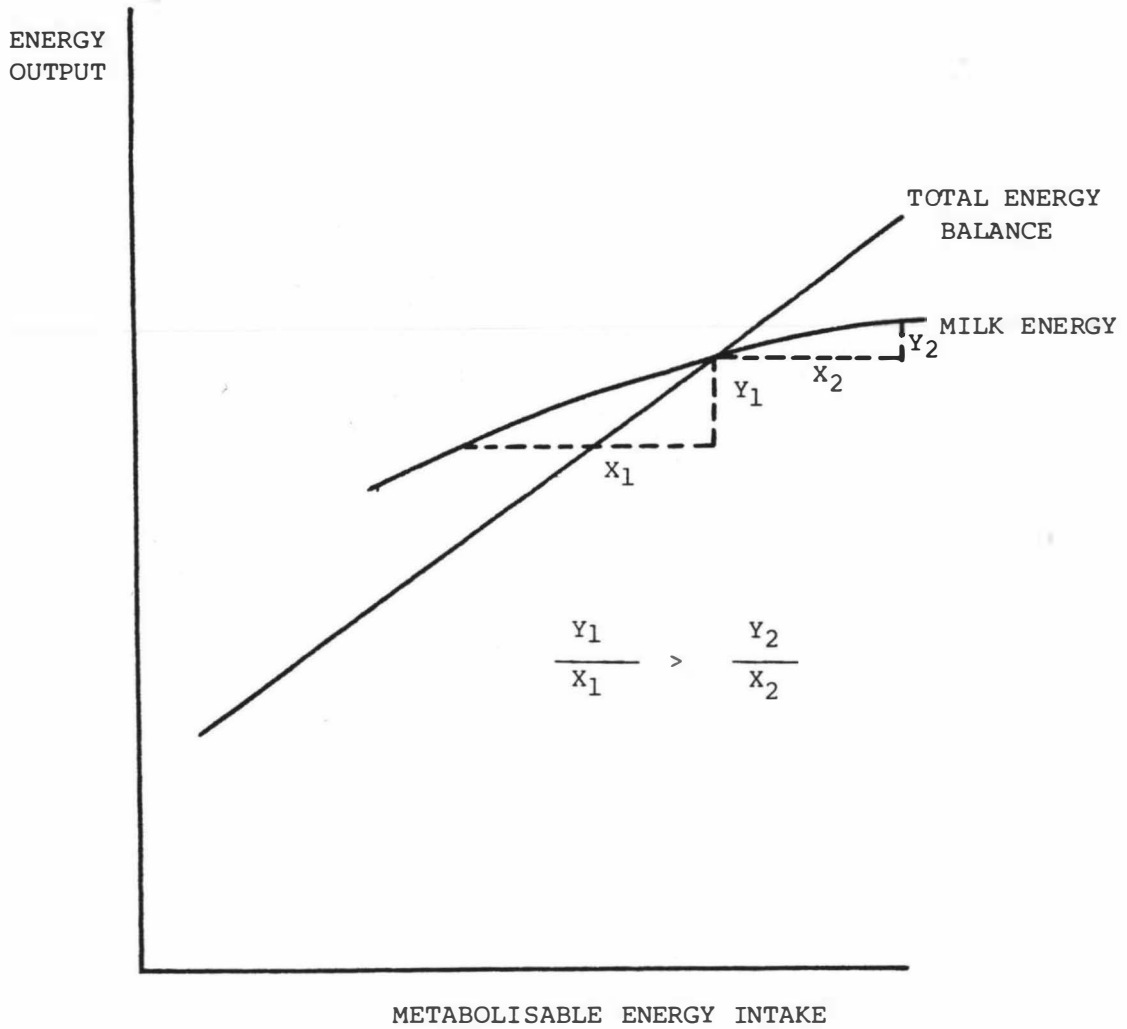


FIG. 1.2 In contrast to total energy balance, milk production is not linear with ME intake but is subject to diminishing returns in that an increasing amount of energy is diverted into body tissue energy at higher feed intakes (from Møe and Tyrrell, 1975).

Therefore it is clear that the relation between milk production and MEI is not a simple one because of concomitant changes in body tissue energy.

The difficulties in determining partial efficiencies, which relate components of energy balance to MEI, are considerable and were discussed earlier (see Section 1.3.1). For this reason it appears inappropriate to compare animals with regard to their partial efficiencies. Despite the limitations, there have nevertheless been several references in the literature to factors affecting partial efficiencies and Moe and Tyrrell(1975), reviewing energy balance measurements made at Beltsville, concluded that there is little evidence that the partial efficiency for milk synthesis (k_l) varies with level of milk yield. Furthermore, Moe et al. (1972) concluded that k_l is subject to little variation between cows since the metabolisable energy content of the diet is associated with 88% of the variation in k_l . Van Es (1976) reviewing approximately 1150 balance trials with lactating cows (including the Beltsville data) concluded that genetical variation in k_l between animals is small. Whether or not genetic differences between animals were expected to be evident in these balance trials is not clear.

Similarly Van Es (1972) reviewing data on maintenance requirements for energy concluded that within breeds there was no conclusive evidence that high or low maintenance requirements are heritable. There are, however, true breed differences in fasting metabolism (see Webster, 1978 for references).

Frisch and Vercoe (1980) showed that selection for growth rate in a harsh tropical environment, within a closed line

of crossbred Hereford x Shorthorn cattle, was associated with a decrease in fasting metabolism. In a favourable environment the reverse was the case (Frisch and Vercoe, 1977).

Heat production which is usually estimated indirectly in calorimetric balance studies, can provide a measure of the efficiency of use of metabolisable energy (ME). It is likely to be a more appropriate criterion on which to compare the efficiency of energy use by different animals fed on one level of MEI because it avoids many of the assumptions that are necessary to provide estimates of partial efficiencies of the components of energy balance.

Both Flatt et al. (1969) and Van Es and Van der Honig (1979) concluded that the efficiency of use of ME is the same for high and low producing cows, but the extent to which the differences in production are caused by genetic differences between cows is unknown.

To date energy balance experiments have not been specifically designed to examine whether or not high and low producing cows differ in the proportions of metabolisable energy partitioned between the synthesis of milk and body tissue. Flatt et al. (1969) demonstrated the capability of high producing cows to mobilise large quantities of body tissue during early lactation and to deposit it during late lactation. The amount of milk produced over the lactation in their study, ranged from 5000 to 8600 kg, hence it was possible to compare high and low producers. The energy balance data for individual cows were pooled over the whole lactation, but there was no clear relation between the amount of milk produced and the associated changes in body tissue. This may have been expected because the intake of metabolisable energy was not fixed; a high level of

milk production was associated with a high intake. Unfortunately data for individual cows within each stage of lactation were not presented. Additional evidence (Moe and Tyrrell, 1975) showed also that there was no relation between the productive ability of the cow and the associated change in body tissue; again intake was not fixed.

In conclusion, evidence from calorimetric studies shows that cows differing in productive ability do not differ in the efficiency with which they use metabolisable energy for total energy balance. Therefore it is likely that differences in production are related to the intake of dietary energy, the ability to metabolise feed energy and an increased partitioning of dietary energy to the synthesis of milk rather than body tissue. However insufficient evidence is available to determine the relative importance of these three factors.

None of the calorimetric balance studies reviewed, actually set out to compare cows which differed in their genetic ability to produce milk.

1.3.2 Evidence from nutritional studies.

1.3.2.1 Feed intake and potential for lactation.

One of the major factors which regulates food intake in the cow, is her physiological state (Bines, 1976a). Thus a cow with a high level of milk production may be expected to eat more than one giving a low milk yield. Pro rata feeding systems are indeed based on the assumption that milk yield increases linearly with increases in food intake; changes in liveweight of the animal are ignored. The limitations of this approach are well recognised and have been outlined by Broster (1976).

There is evidence that lactating cows will eat more than non-lactating cows (Hutton, 1963; Hodgson, 1977). Whether or not a causative relation exists between milk yield and level of feed intake is still in doubt (see Broster, 1972 for references). Differences between animals in liveweight change are likely to mask the relation. Another problem concerns the technique of multiple regression analysis which is used to partition the variance in energy intake between liveweight, liveweight change, and milk yield; other terms such as age and stage of lactation are sometimes included also (Wallace, 1956; Hutton, 1962; Curran and Holmes, 1970; Marsh et al., 1971; Bines et al., 1977).

Curran and Holmes (1970) discussed in some detail the problems of the use of multiple regression analysis to predict intake. The main problems are the correlations between the independent variables and the possibility that variables which explain a large part of the variation in intake have been left out of the model.

The estimated partial regression coefficients will be biased, hence do not enable the distribution of energy between performance functions to be determined accurately (N.B. a more complete discussion of this topic is given in Chapter 4).

The relative importance of various factors affecting intake was summarised by Broster et al. (1978) in a Table which is reproduced here (Table 1.3). From this Table it is clear that the two factors which explain most of the variation in intake, which can be accounted for, are liveweight and milk yield.

Table 1.3 Relative importance of various factors influencing intake in some published equations, based on an example of a 500kg cow giving 30 kg of milk per day, losing 0.5kg liveweight per day, and receiving a ration of 75% concentrates; 25% forage (dry matter basis; Table taken from Broster et al. 1978).

Reference	Dry Matter Intake (Kg)	Amount of dry matter intake partitioned to			
		Live-weight	Milk yield	LWC*	% conc.
Greenhalgh and McDonald (1978)	17.1	11.1	6.0		
McCullough (1974)	13.8	6.4	10.8	-2.4	-1.0
Bines, Napper and Johnson (1977)	#13.5	10.6	4.1	-1.2	
Ministry Agriculture and Fisheries Food (1975)	15.5	12.5	3.0		
Hyppola and Hasunen (1970)	19.2	17.3	1.9		
Conrad, Pratt and Hibbs (1964)	16.0	12.0	4.3		-0.3
Mean	14.7	10.6	4.9	-0.6	-0.2

* LWC = liveweight change

heifers (values for cows are about 24% higher)

Bines (1976b), based on limited evidence, commented that during the growth phase there are some consistent variations in appetite between animals of similar liveweight and that this may be useful to predict appetite of the mature cow.

It is concluded that there is no simple relation between milk yield and intake.

1.3.2.2 Milk production and liveweight change.

The issues considered here concern the partitioning of dietary energy between milk yield and liveweight change on fixed diets and the response of high and low producing cows to changes in their food intake.

Bines and Hart (1978) using data from Broster et al.(1969) demonstrated how the partitioning of energy can vary between heifers given equal rations (Table 1.4).

Table 1.4 Variation in food utilisation in two dairy heifers given equal rations for the first 67 days of lactation.

	Cow A (poor)	Cow B (good)
Initial liveweight (kg)	517	520
Total milk yield (kg)	837	1325
Liveweight change (kg)	+ 39	- 52

Further evidence (Broster et al. 1969; Broster et al. 1975) where data from individuals within groups on fixed diets were analysed by regressing liveweight change on milk production, showed that the high yielding animal produced extra milk at the expense of body reserves.

A number of experiments have shown that high yielding cows increase their milk yield more than low yielding cows when the feeding level is increased, either with a pro rata system of feeding (Mather et al. 1960; Brown et al. 1962; Griffiths, 1965; or by a fixed amount (Broster et al. 1969; Broster et al. 1975; Johnson, 1977). Broster et al. (1981) summarising some of the experimental estimates calculated a mean value of 0.008kg milk change in response to 1 MJ metabolisable energy/kg difference in initial milk yield. Therefore an additional 15kg initial yield (for example, 35kg/day versus 20kg/day) leads to a doubling of the mean response of 0.114kg milk/MJ metabolisable energy.

Not all experiments have found a positive relation between potential of the cow and her response in milk yield to an increase in intake (Jeffrey et al. 1976; Johnson, 1979; Ostergaard, 1979; Steen and Gordon, 1980).

Ostergaard (1979) and Steen and Gordon (1980) found that response was not related to potential of cows when high quality basal diets were offered ad libitum. Broster and Thomas (1981) provided an explanation for the apparent discrepancy in response related to cow potential. They proposed two yield response curves (Figure 1.3) which show that with low and fixed levels of feeding the response by the high yielder is greater than that of the low yielder (i.e. $r_1 > r_2$).

Cows given good quality forage ad libitum will have a high base level of feeding and furthermore high yielding cows will have a higher intake than lower yielders and thus the response to an additional amount of concentrate energy tends to be very similar (i.e. $r_3 = r_4$).

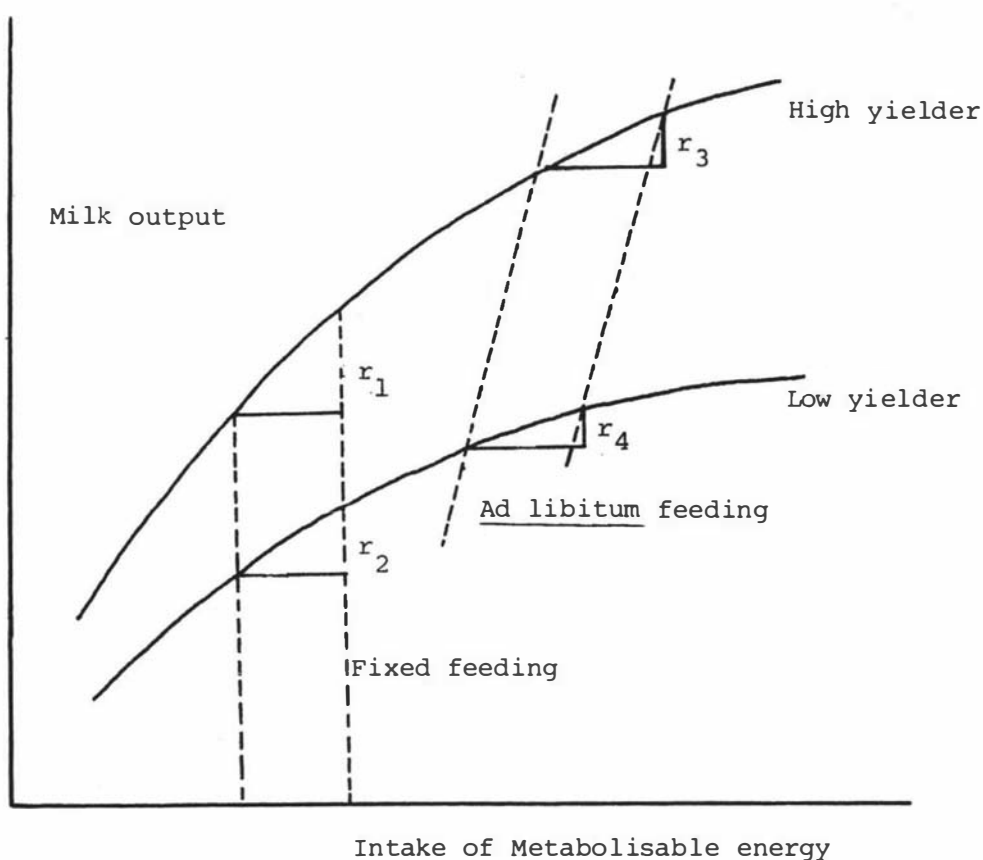


FIG.1.3 The response of low and high yielding cows to an additional input of energy. (Taken from Broster and Thomas, 1981).

The extent to which r_2 approaches r_4 will depend on the difference in cow potential and the coefficient relating intake to current yield. This effect may also be confounded by the small increase in total energy intake with additional concentrates consequent upon a high substitution rate.

Thus when differences in response caused by the effect of current yield and variation in substitution rate are taken into account, the apparent conflict in response observed with fixed and ad libitum feeding regimes can be resolved (Broster and Thomas, 1981).

The explanation by Broster and Thomas (1981) does not appear to explain why Johnson (1979) and Jeffrey et al. (1976) did not observe greater responses from high yielding cows. In the experiment by Johnson (1979) the basal level of feeding was not ad libitum and two fixed levels of concentrates, regardless of yield were fed above the basal ration. This implies that the feeding levels were fixed for cows of both yield potentials; namely mean daily peak yields (kg) of 35 and 29 for high and low yield cows. The different levels of concentrates were fed only during the period from week 9 to 20 of lactation and the differences in milk yield between the two levels of concentrates fed, were 120 and 115kg for high and low yielding cows respectively.

Jeffrey et al. (1976) in a series of experiments found that the response to grain supplementation for cows grazing on pasture was similar from both low and high producing cows and not affected by stage of lactation or age. The mean milk yields of the cows involved in these experiments ranged from 8 -14kg cow⁻¹, hence the overall level of feeding was apparently low, and a difference in response between high and low producing cows may have been expected, based on the evidence of Broster and Thomas (1981). Since individual intakes of pasture and grain were not measured, it was not possible to determine the degree of substitution of pasture by grain, or whether the rate of substitution differed between high and low producing cows.

The type of diet fed may also influence the response to extra feed. Broster and Thomas (1981) suggested a curvilinear relation between energy intake and milk yield for a forage : concentrate diet. Most data that are available for pasture fed cows indicate a linear response to increasing levels of feeding (Bryant, 1980; Stockdale et al. 1981; Grainger et al. 1982). Certainly no data are available to show the curvature in response in cows of high potential is significantly less than that of low producing cows as suggested by Broster (Figure 1.3). The effect of absolute plane of nutrition on difference in response by grazing cows of different potential therefore remains speculative.

Although in general it appears that high producing cows will show larger responses to extra food than low producing cows, this is not always so. The particular system of feeding and the absolute plane of nutrition can influence the response. Further research, particularly for cows grazing on pasture, where substitution effects can be important, is warranted.

1.3.2.3 Stocking rate experiments with high and low producing cows.

Evidence on the performance of high and low producing cows at different stocking rates is limited to two experiments which have however not been reported in any detail (Hutton, 1975; Gleeson, 1978).

Hutton (1975) reported an experiment done over three years where the production of identical twin cattle (presumably low producers) could be compared to that of high producing cows on farmlets which were irrigated and stocked at 5.6 cows per hectare. The high producing cows were selected for actual or potentially high milk production.

High producing cows consistently outproduced the low producing cows, but the relative advantage in production of the high producing cows decreased over the three years of the experiment (Table 1.5).

Table 1.5 The performance of high (H) and low (L) producing cows on irrigated farmlets stocked at 5.6 cows per hectare (from Hutton, 1975).

Item	Year	H	L	Relative Production H/L
Milk fat/cow (kg)	1972/73	151	134	1.13
	1973/74	126	113	1.12
	1974/75	115	108	1.06
Milk fat/ha	1972/73	839	746	-
	1973/74	701	627	-
	1974/75	639	601	-

The reasons for the decline in the relative performance of the high versus low producing cows are not clear since details on feed intakes, pasture parameters, and liveweight changes are not given.

Gleeson (1978) reported an experiment carried out for three years to determine the influence of stocking rate on dairy merit. Additional details have been given in various references (McFeely, 1976 and 1977; McFeely et al. 1977).

The four treatments were high quality cows stocked at 2.5 (HL) and 3.1 (HH) cows per hectare and lower quality cows similarly stocked (LL and LH). The cows were originally selected in the first year of the experiment within a pool of 150 cows and there were 18 cows in each of the four groups. All animals in any year were initially fed indoors for approximately six weeks and subsequently grazed from March to November. Concentrates were also fed, although it is not clear exactly when this was done. Of importance though is that the high merit cows were given more concentrates than the low merit cows, 430 versus 330kg.

High merit cows produced more than low merit cows at both stocking rates although the difference in milk yield between the two different merit groups was greater at the low versus high stocking rate, namely 1044 versus 895kg cow⁻¹ (Table 1.6).

Table 1.6 The performance of high quality cows stocked at 2.5 (HL) and 3.1 (HH) cows/ha and lower quality cows similarly stocked (LL and LH) over a three year period (from McFeely et al. 1977; McFeely, 1977).

Item	High merit		Low merit	
	HL	HH	LL	LH
Milk yield (kg cow ⁻¹)	4187	3692	3143	2797
Milk yield (kg ha ⁻¹)	10425	11435	7820	8665

Whether the interaction between stocking rate and genetic merit measured in terms of yield per cow is significant is not clear. Part of the observed difference in milk

yield between high and low merit cows could be attributable to the greater amount of concentrates received by the high cows.

Both experiments (Hutton, 1975; Gleeson, 1978) provide evidence that the productive merit of cows has an effect on farm productivity measured as output per hectare. Reasons for the increased performance are not apparent, mainly because of a lack of data presented. Further definition of the influence of cow quality on output per hectare is required.

1.3.3 Conclusions

Data reviewed indicate that the high yielding cow produces more milk because of an increased feed intake and/or a greater ability to partition dietary energy to milk yield at the expense of body reserves.

There is no evidence to suggest that high producing cows differ either in their ability to metabolise feed energy or in the efficiency with which they utilise metabolisable energy.

Based on limited data, productive ability appears to be an important factor in determining output per hectare, although further definition is required.

1.4 GENETIC SELECTION EXPERIMENTS

Response to selection applied to animal populations is a demonstration of genetic adaptation to environmental pressures. Although the final phenotypic trait, such as high efficiency of milk solids production in terms of food used may be clear enough, the combination(s) of biochemical and physiological and behavioural characteristics which determine phenotypic performance for that trait are not known. The same level of phenotypic performance may result from a number of different combinations of characteristics, these having different genetic bases and some being correlated genetically with others.

Freeman (1975) reviewed experimental data relating to genetic variation in nutrition of dairy cattle up to 1975 and concluded that although there is no doubt that when selection is applied, milk production is increased, there is little or no real knowledge as to the physiological changes caused by selection. He went on to say;

"The genetic information relating to feed use; i.e. heritabilities and genetic and environmental correlations, includes parameter estimates based on components of variance and regressions that have genetic interpretations. These parameter estimates are certainly useful and allow predictions of expected response to selection. They certainly give an idea of the nature of quantitative genetic control over gross measures of feed use, but selection experiments or actual experience that allows measurement of genetic changes is needed to help verify the nature and magnitude of these responses."

A summary of the main points from the review by Freeman will be made and more recent evidence will be considered in relation to the earlier work.

1.4.1 Summary of the main points of the review by Freeman (1975).

The experiments reviewed by Freeman (1975) all had one common feature, namely that pro rata feeding systems were used. Concerning pro rata feeding, Broster (1976) has stated:

"It should also be noted that experimentation based on rates of feeding per unit milk produced involves an element of confounding of food and milk which seriously limits interpretation in terms of efficiency of food conversion."

1.4.1.1 Gross energetic efficiency.

Many different measures of gross efficiency were used, but all were basically some ratio of the energy produced in milk to the total energy consumed by the animal. Heritability is the ratio of the additive genetic variance in the population to the total variance. The total variance includes both environmental and genetic portions of the variation. Heritability is the fraction of the selection differential, or the total selection practised that is expected to result in genetic improvement in the next generation.

The heritabilities for gross efficiency were all positive and relatively large indicating that genetic differences within a breed are large enough to allow genetic progress to be made.

1.4.1.2 Genetic differences in consumption.

One of the most comprehensive experiments was done by Miller et al. (1972), who found that the heritabilities of forage intake and total net energy intake in cows given forage ad libitum and concentrates according to production were 0.19 and 0.42. In general, repeatabilities (correlation between repeated measurements on the same cow) were high, ranging from 0.22 to 0.86 and the heritabilities were clearly greater than zero.

1.4.1.3 Correlated responses.

Correlated traits have genes in common which influence their expression. If two traits are correlated, then if selection for one of the traits is effective, an automatic change in the second trait can be expected without any selection being purposely applied to trait two. Phenotypic correlations simply measure the degree of association between two traits in the same animal that is caused by both genetic and environmental effects.

The phenotypic correlations between yield and consumption and between yield and efficiency are generally high, 0.60 - 0.95, except for forage intake with yield (Table 1.7).

Grain and forage intake were negatively correlated because cows were fed so that the difference between requirements and total consumption was met by varying the amount of grain offered. In other words the feeding regime induced this correlation. Liveweight gain was negatively associated with efficiency, yield and consumption.

Table 1.7 Phenotypic correlations among measures of consumption, efficiency, yield, and weight
(Table taken from Freeman, 1975)

Trait	Consumption			Efficiency			Weight			
	Grain	Forage	Total ENE	FCM/FU	FCM/100 FU Maintenance	FCM/ENE (Mcal)	TDN to Produce 100lbFCM ^a	Average Body Weight	Heart Girth	End of lactation
Yield										
FCM	0.83 ^b	0.10 ^b	0.72 ^b 0.68 ^c	0.82 ^d	0.95 ^e	0.82 ^c	-0.71 ^f	-0.11 ^d 0.44 ^g	0.02 ^e	-0.53 ^g
Milk	0.83 ^b	0.10 ^b	0.70 ^b	0.84 ⁱ				0.01 ^d 0.42 ^g		-0.53 ^g -0.17 ^h
Consumption										
Grain		-0.23 ^b								
Forage						0.74 ^b -0.32 ^b		-0.18 ^b 0.49 ^b		-0.48 ^b 0.08 ^b
Total ENE	0.58 ^b	0.66 ^b				0.17 ^b 0.14 ^c		0.41 ^b		-0.28 ^b 0.27 ^h
Total therms TDN Intake							-0.20 ^f	0.64 ^g		-0.33 ^g
Efficiency										
FCM/FU								-0.10 ^d		
FCM/100 FU Maintenance									-0.29 ^e	-0.41 ^h
FCM/ENE therms TDN to produce 100lb FCM								-0.04 ^g		-0.51 ^g
								0.08 ^f		

a Sign differs because of definition.
b Miller et al. (1972a).
g Hooven et al. (1968).

c Hooven et al. (1972).
d Gjelstad (1953).
h Miller et al. (1972b).

e Syrstad (1966).
f Stone et al. (1960).
i Mason et al. (1957).

Genetic correlations were generally similar in sign to the phenotypic correlations and larger in magnitude for traits not involving weight (Table 1.8). The genetic correlations involving yield, efficiency and consumption are large except for those including forage intake. The genetic correlations between liveweight gain during lactation with yield, consumption and efficiency are all large and negative, except forage consumption and liveweight gain, which was estimated as 0.26 by Miller et al. (1972).

Freeman concluded by adding a note of caution, mentioned earlier, that the projection of correlated responses are expected responses and have not been demonstrated experimentally. In addition, whether or not the heritabilities and correlated responses would be similar under different feeding regimes was not clear at the time.

1.4.2 More recent genetic selection studies.

Although the genetic selection experiment done by Hickman (1971) was reported before 1975, it was not included in the review by Freeman (1975).

The experiment reported by Hickman (1971), was started in 1954 by selecting dairy cows for high yield of total milk solids over the first 180 days of lactation, and to measure correlated genetic responses resulting from the selection pressure. Traits measured were feed efficiency, growth, and body size.

Feed consumption (total digestible nutrient consumption; TDN) for each heifer was estimated during the growth phase (180-240 days of age) and during the first lactation (60-120 days postpartum). Measurements of liveweights

Table 1.8 Genetic correlations among measures of consumption, efficiency, yield and weight
(Table taken from Freeman, 1975)

Parameter	Consumption				Efficiency				Weight		
	Grain	Forage	Total ENE	Therms	FCM/FU	FCM/100 FU Maintenance	FCM/ENE Mcal	FCM/ENE Therms	Average Body Weight	Heart Girth	Weight Gain
Yield											
FCM	1.0 ^a	0.32 ^a	0.82 ^a 0.86 ^b 0.83 ^c	0.89 ^d	0.94 ^e	0.88 ^f	0.93 ^b	0.88 ^c	0.29 ^e 0.28 ^d -0.11 ^c	-0.08 ^f	-0.77 ^c
Milk	1.0 ^a	0.20 ^a	0.77 ^a 0.76 ^c	0.86 ^d	0.95 ^g			0.86 ^c	0.19 ^e 0.30 ^d -0.12 ^c		
Consumption											
Grain		0.33 ^a							-0.21 ^a		-0.95 ^a
Forage									0.93 ^a		0.26 ^a
Total ENE	0.84 ^a	0.80 ^a						0.47 ^c	0.44 ^c		-0.43 ^a
Total therms								0.61 ^b	0.44 ^c 0.80 ^d		-0.39 ^c
Efficiency											
FCM/FU									0.13 ^e		
FCM/100 FU Maintenance										-0.55 ^f	
FCM/ENE Therms									-0.17 ^d -0.51 ^c		-0.88 ^c

a Miller et al. (1972a)
b Hooven et al. (1972)
c Miller (1972)

d Hooven et al. (1968)
e Gjelstad (1953)
f Syrstad (1966)

g Mason et al. (1957)

heart girths and wither heights were also made at these times. Pro rata feeding was used with the emphasis on high forage consumption for hay and silage which were fed ad libitum. Concentrates were fed to lactating animals at the approximate rate of 1kg/4kg of milk produced on a daily basis.

There was a closed herd requirement for the experiment which made possible the calculation of selection differentials on all data being collected and an estimation of the response to selection within the population. Both Holstein and Ayrshire cattle were used.

The response to selection for 180 day total solids yield indicated at least as much as theoretically expected from the selection differentials and heritabilities with very little change in milk composition.

The physiological changes associated with the selection for yield differed in kind and amount between the two breeds. Heifer growth rate increased in both breeds with increased feed efficiency (TDN/kg liveweight gain) for Holsteins, but increased feed consumption of the Ayrshires with no increase in efficiency. There was a decrease in heart girth of the Holsteins without a change in wither height, hence it is possible that the change in feed efficiency reflected the deposition of lean rather than fat by the Holsteins. Relation of heart girth to wither height in growing Ayrshires did not change.

The efficiency of energy utilisation for milk production (TDN/kg fat corrected milk) improved with selection for yield in Holsteins and Ayrshires, but the improvement could be shown to be significant ($P < 0.05$) only with the Holsteins. The increased efficiency of the Holsteins was accompanied by a significant decrease in weight gain during lactation and this could indicate an increase in the ability to utilise feed for production of milk rather than to gain liveweight during lactation. In addition there may have been some decrease in the maintenance requirements of the Holsteins because wither height and heart girth were significantly reduced as a result of selection for yield of milk solids.

All of the genetic selection experiments reviewed so far have involved feeding animals by pro rata feeding systems. Two more recent experiments (Grieve et al. 1976; Hind, 1979) provide an opportunity to examine the effects of selection under a different feeding regime. In both experiments complete diets of relatively constant energy concentration were fed during lactation.

In the study by Grieve et al. (1976) there were three objectives:-

- (i) To determine the relation between feed intake, yield and efficiency when complete diets were fed ad libitum throughout lactation,
- (ii) To investigate the relation between estimated transmitting ability (ETA; an estimate of productive ability based on pedigree information) and future feeding performance,

- (iii) To investigate the relation between ETA and ration digestibility.

Forty-nine Holstein heifers were individually fed on complete rations from 60 days prior to calving through the first 305 days of their first lactation. The heifers were selected to provide a wide range in ETA's for milk yield.

The correlation between ETA and dry matter intake (DMI) was 0.3 during mid-lactation and about half of that during early and late lactation. Early, mid, and late lactation were defined as 1-90, 91-180, and 181-305 days respectively. The correlation between DMI and solids-corrected milk yield (SCM) was high (0.8) in mid and late lactation and similar to those reported by Hooven et al. (1968). During early lactation feed intake was associated more closely with body weight than with other traits ($r = 0.44$).

Liveweight changes were negatively correlated with feed intake during lactation ($r = -0.25$) in agreement with a correlation ($r = -0.31$) observed by Hooven et al. (1968).

Feed intake was also highly correlated with gross efficiency over the 305 day lactation expressed as the ratio of SCM yield to dry matter intake ($r = 0.62$). Thus more efficient cows consumed more feed, produced more milk and did not gain so much liveweight. The correlation of 0.62 is much higher than those reported ranging from 0.11 to 0.18 (Hooven et al. 1968; Hooven et al. 1972; Miller et al. 1972). Hooven et al. (1968) suggested that efficiency depended more on milk yield than on food consumption. It is interesting that the correlation

between feed intake and feed efficiency of 0.62 ($r^2=0.38$) reported by Grieve et al. (1976) approaches the correlation 0.82 ($r^2 = 0.67$) between FCM yield and feed efficiency reported by Hooven et al. (1968). The former authors suggested that the different feeding regimes may explain the difference in correlations.

There was no association between ration digestibility and ETA; estimated on 24 of the 49 cows.

Hind (1979) presented details of a study designed to examine whether or not selection for high milk yield increased efficiency of milk production. Jersey and Friesian cattle were involved, but discussion here will be confined to the Friesians since some of the relevant data was not presented for Jerseys.

Eighty-eight foundation cows were reared on a standard complete pelleted diet fed ad libitum. Foundation cows were mated alternately to bulls with high and low breeding values (contemporary comparisons or CC) for milk yield.

The progeny of high bulls produced 700kg more milk than the progeny of low bulls during the first lactation which was in agreement with the difference of 930kg obtained in the national progeny test. Whilst the differences between progeny groups in milk yield were 27% (average of two lactations) there was a concomitant decrease in the fat and protein concentration of the milk of the high progeny group, hence the difference in total solids was only 19% (Table 1.9).

Table 1.9 Production and efficiency differences between Friesian progeny of high and low contemporary comparison bulls (Table taken from Hind, 1979).

Item	Production (kg)			Efficiency* (%kg/kg)		
	High -Low	s.e.#	% difference+	High -Low	s.e.	% difference
Milk yield	970	340	27	9.2	4.4	15
Fat corrected milk	810	350	23	7.0	4.6	12
Total solids	51	25	19	0.30	0.33	9
Food intake	660	260	11			

* ratio of production to consumption, expressed as a %

standard error of difference between two means

+ percentage by which high yielding progeny exceeded low yielding progeny.

High progeny animals consumed 11% more feed than low progeny animals. Differences between progeny groups for different measurements of efficiencies varied from 15 to 9% (Table 1.9).

Selection for increased milk yield resulted in increases in feed intake and efficiency of production. Liveweight changes were not reported.

1.4.3 Genotype by environment interaction.

Extensive reviews of this topic have recently been made by Freeman (1975) and Syrstad (1976). The purpose of the comment here is to highlight certain aspects of the topic.

The possibility of an interaction between genotype and feeding regime was examined in two similar studies where daughters of bulls from Jersey sires (Richardson et al. 1971) and Holstein sires (Lamb et al. 1977) were randomly allocated within progeny groups to an all forage or forage plus grain ration. In each experiment there was at least one New Zealand sire represented.

Although the overall conclusion from both studies was that there was no sire x ration interaction, it was interesting to note that in each experiment the daughters of the New Zealand sire performed at the top or near the top for the forage ration, but near the bottom for the concentrate ration.

A much more comprehensive study is currently underway in Poland (Stolzman, 1982; Jasiorowski et al. 1982); details were given earlier. First-cross heifers from New Zealand sires ranked near the top for milk fat yield and milk protein yield regardless of whether the diet was mainly forage or forage and concentrates. This suggests that interactions between genotype and feeding regime are of less importance than suggested by the earlier work done by Richardson et al. (1971) and Lamb et al. (1977). Whilst this has implications for sales of bull semen internationally, it must be remembered that almost all selection in dairy cattle populations is based on records of milk yield which are collected under the environmental conditions prevailing in commercial herds in the country

in which the semen will be used. Hence an interaction between sire and ration would be of little consequence for normal breeding work in practice within the same country and feeding system.

Of more importance is the observation, made on two occasions (Brumby, 1961; Carter, 1964), of an interaction between genotype and environment.

Brumby (1961) split 120 pairs of identical twins between 20 high producing and 20 low producing herds. He found that the correlation between the deviation in milk yield from the herd mean of an identical twin in a high herd with the deviation of its co-twin in a low herd was 0.11 ± 0.11 . The twin members did not rank in the same order at the different levels of production.

Brumby speculated that the interaction between genotype and herd environment was attributable to the fact that the milking technique in low producing herds was less adapted to the individual cow than in high producing herds, and that plane of nutrition did not contribute to the large interaction.

Carter (1964) compared the production of 'high' genetic merit cows (bred at Ruakura), to a selection of 'average' genetic merit cows from commercial herds; under controlled grazing and set stocking. Because of the use of herd testing and artificial insemination it could be reasonably assumed that the Ruakura cows were genetically superior at that time. Cows of 'high' and 'average' genetic merit were grazed together under each grazing management. The average production level of all cows over a 305 day lactation (averaged over three years) was 173 and 148kg milk fat under controlled grazing and set stocking respectively. The differences between the cows differing

in genetic merit were 28 and 22kg milk fat for controlled grazing and set stocking. The implication is that the differences in production between genotypes were greater at the higher levels of production. However the statistical significance of the interaction could not be tested because although there was replication of animals and replication in time there was no replication or randomization in space.

One aspect which has received no attention is whether or not genotype interacts with nutrition at different stages of the lactation cycle. For example, if high genetic merit cows utilise body reserves in early lactation to support their requirements for milk production to a greater extent than low genetic merit cows, provision of adequate body reserves is obviously more important for the high genetic merit cows.

1.4.4 Conclusions

There appears to be little doubt that when selection is applied, milk production can be increased. The physiological changes which accompany the increases in yield are not as well defined. Most emphasis so far has been placed on measuring gross efficiency and correlated genetic responses. That an improvement in gross efficiency has occurred with selection is not surprising since most experiments have used pro rata feeding systems. Correlated genetic responses which appear to be implicated in the improved performance are liveweight changes and feed intakes.

Other aspects, for example, digestive efficiency and utilisation of feed energy have received little or no attention.

The applicability of the results from selection studies where pro rata feeding is practised to other feeding regimes is not known. In particular there is a dearth

of information for the cow grazed on pasture.

Until the effects of genetic selection are more clearly defined it will be difficult to determine optimum feeding management strategies for these highly selected animals and whether or not changes in the selection procedures are necessary. Gowe and Fairfull (1982), in a discussion of poultry breeding studies, highlighted the value of understanding the physiological changes that accompanied a long-term breeding programme. This enabled them to more effectively pursue the specific objectives of their breeding programme without the adverse side effects which had previously occurred.

1.5 ADDITIONAL CONSIDERATIONS

Two other related aspects concern protein requirements and blood metabolites and hormones. It is not intended to review these aspects in any detail, but to consider them in relation to genetic selection.

Other aspects such as animal health, reproduction, milking characteristics, and grazing efficiency, have received very little attention in the literature in relation to genetic selection, and will not be considered.

1.5.1 Response to protein supplementation.

Comments on two aspects of the protein requirements of cows grazing on pasture are made here.

In recent years milk yield responses to

protein supplementation have been observed for cows fed pasture individually in stalls (Rogers et al. 1980; T.E. Trigg, unpublished data) and by cows grazed on pasture (Stobbs et al. 1977; Minson, 1981). Methods of protein supplementation used in these experiments were either by post-ruminal infusion or by feeding protein that was partially protected from degradation in the rumen.

Several explanations have been offered for the observed increases in milk yield due to protein supplementation and these have been summarised by Brookes (1982). Factors possibly implicated are; an increase in pasture intake; an increased mobilisation of body reserves; an increased supply of specific amino acids. One experiment not included in the summary by Brookes (1982) has shown, with the aid of indirect calorimetry, that milk yield responses to abomasal infusions of casein were largely a result of increased tissue mobilisation, and not to differences in utilisation of ingested feed (T.E. Trigg, unpublished data).

The second point concerns the effect of level of milk production on the response to protein supplementation. Clark (1975) in a review of lactational responses to post-ruminal administration of proteins and amino acids concluded that the greatest increases were from high yielding cows: increases were in excess of one kg of milk per cow per day for cows producing over 20kg milk per day, but cows producing less than 20kg per day seldom increased in milk yield more than one kg per day. Rogers et al. (1980) found that cows fed pasture in stalls responded to a protected casein supplement and that the response was greater by 0.5kg per kg increase in level of milk production.

Apparent protein deficiencies in pasture for milk production and the likelihood that the selection of cows for high milk yields will aggravate these deficiencies emphasise a growing need for further research into the protein requirements of grazing cows. The particular mode of action of protein supplements in promoting an increase in milk yield needs to be defined. For example if the response to protein supplementation was accompanied by an increase in the mobilisation of body reserves then there may be no long-term benefit in the efficiency of milk production (as these reserves would sooner or later need to be restored). On the other hand if the protein supplementation increased the efficiency of digestion or of intermediary metabolism then there may be a genuine increase in the efficiency of milk production.

1.5.2 Blood metabolites and hormones.

Discussion will be confined to outlining the major potential benefits of studies in which blood characteristics were measured.

Bauman and Currie (1980) drew attention to the considerable technical and interpretational difficulties involved in studies of blood metabolites and hormones. For example, studies relating hormone concentrations in serum to metabolic events do not account for possible changes in blood flow to a tissue or in alterations in numbers of hormone receptors in a target tissue. Plane of nutrition must be considered when comparing high and low producing cows, a point well made by comparing the work of Hart et al. (1975) to that of Bauman et al. (1979) and Vasilatos and Wangsness (1981).

Bearing these limitations in mind it is considered that the greatest potential benefits of studies, which attempt to understand the endocrine control of partitioning of nutrients and the utilisation of nutrients, are twofold. The first concerns the identification by measuring blood characteristics of the genetic merit of an individual animal early in its life. This would enable potential sires for use in artificial breeding to be screened as calves before entering a progeny testing scheme. In this way the accuracy of the selection procedure could be improved thereby enabling more rapid progress in the genetic improvement of milk production to be made.

Tilakaratne et al. (1980) found that calves with different potentials for milk production varied in aspects of energy and nitrogen metabolism and discussed the possibility of using the measured differences as criteria for genetic selection for milk production.

The second major benefit can be seen in the answer to the following question:

"Is direct selection for volume of milk production achieving the most rapid improvement in the efficiency of milk production?"

Freeman (1967) suggested that selection for milk production would achieve 75-90% of the progress of direct selection for efficiency of milk production given equal selection intensities for both traits. The selection intensity for milk production is likely to be larger because it is much easier in practice to measure milk production than efficiency. Thus greatest progress would be made by selecting for milk yield.

Baldwin et al. (1980) in a speculative paper suggested that identification of animals whose metabolic pathways approach theoretically maximum efficiency and subsequent genetic selection for "metabolic efficiency", should if the traits are heritable, lead to a genetic increase in the efficiency of milk production. However even if a similar selection intensity to that for milk yield could be practised, selection for increased metabolic efficiency may be at cross purposes with selection for efficiency of milk production. It is likely that the greatest impact of identifying animals with a more efficient metabolism will be when the underlying mechanisms responsible for their improved efficiency can be determined. Then it may be possible to manipulate through hormonal or other means the metabolism of all animals to achieve maximum metabolic efficiency, and hence to improve the average efficiency of animal production.

The approach suggested by Baldwin et al. (1980) is worthwhile, but is a long-term project. In the meantime therefore, the most practical means of improving the efficiency of milk production will be made by selecting for higher milk production as outlined by Freeman (1967).

1.6

CONCLUSIONS

Artificial breeding has been used in New Zealand for more than thirty years and there is no doubt that selection has increased the level of milk fat production per cow. Little is known of the physiological changes that have accompanied this improvement. An understanding of these physiological changes could lead to a more effective breeding programme. It could also then be possible to devise feeding management strategies so that potentially high producing animals could express their full potential.

Because of the lack of information for the grazing cow, the physiological changes that accompany selection have been gauged from the published results from studies where cows were fed forage: concentrate diets. Evidence from nutritional and genetic selection studies suggests that likely physiological changes are an increase in feed intake and a preferential partitioning of dietary energy to milk production rather than body gain. Changes in the ability of cows to metabolise feed energy and the efficiency of use of metabolisable energy as a result of selection, have received almost no attention, although limited evidence from energy balance studies with cows differing in milk production suggests that any such differences are small.

Other aspects such as grazing efficiency, animal health, reproductive ability, and milking characteristics have received little or no attention.

1.7 PURPOSE AND SCOPE OF THE INVESTIGATION

The main objective is to determine the mechanisms responsible for increases in milk fat production resulting from genetic selection in a grazing environment in New Zealand.

Upon realisation of the main objective there will be some discussion as to whether or not it is necessary to derive different management strategies for cows differing in breeding index. Specific experimental objectives are outlined at the beginning of Chapters two and three.

Initially two herds of cows differing in genetic merit (as measured by breeding index; see Appendix 2.1 for details) were set up by purchasing cows with high (average BI 126) or low (average BI 102) genetic merit from commercial dairy farmers in New Zealand. The low BI group represent the average genetic merit of cows in the base year (1960/61) when the BI scale was set at 100 and the high BI group are genetically 26% above that average. The best estimate of the average cow in New Zealand at present is approximately 114 (B Wickham, pers. comm.). Thus the cows involved in the experiments reported in this thesis probably represent the upper and lower extremes on a herd basis for the current New Zealand population.

The experiments reported in this thesis are summarised below:-

- four indoor feeding experiments where cows were fed on cut pasture for periods of up to five weeks; three of the experiments involved two levels of feeding (Chapter two);
- a grazing experiment in early lactation where high and low breeding index cows were grazed together and pasture intakes were estimated using an indigestible faecal marker (Chapter two);
- energy and nitrogen balance experiments, using indirect calorimetry and total collection of excreta, with lactating and non-lactating, pregnant cows (Chapter three).

In most of the experiments reported in this thesis cows were individually fed on pasture in stalls. This was necessary in order to be able to measure the expected differences in feed intake between genotypes. Differences in grazing efficiency between genotypes, although they may be important, have not been examined because it was considered that definition of the nutritional characteristics of the genotypes was a more logical starting point.

Several aspects relating to the analysis of the experimental results require comment. Firstly, because there was on average, a difference in size between genotypes of the cows purchased for the first year's experiments *, performance variables were expressed per unit metabolic liveweight before any statistical analysis was done.

* there was no difference in the average size of the genotypes of cows purchased in the second year.

The use of metabolic liveweight ($\text{kg}^{0.75}$) is debatable, but was based on the following reasons.

- the commonly accepted unit of size in energy metabolism studies is metabolic liveweight (Kleiber, 1965);
- reviewing the relation between size and feed intake ARC, (1980) concluded that metabolic liveweight was the preferred scaling factor for intake.

Evidence from the present experiments which also supports the use of 0.75 as the exponent for scaling performance variables is outlined on p.62.

The second point concerns the methods of statistical analysis used in this thesis.

During the course of the statistical analysis of the results of the energy balance data it became apparent that the commonly used methods of analysis were likely to give invalid tests of significance because of correlations between the errors of the components of an energy balance. Chapter four outlines the statistical methods for the analysis of data from energy balance and nutrition experiments which are considered to be more appropriate than those normally used.

A research programme with similar objectives to those outlined in this thesis, but using Jersey cattle, was commenced at Ruakura Agricultural Research Centre by Dr. Arnold Bryant during 1979 (the same time that the experiments reported in this thesis were commenced). Preliminary results of the work being carried out at Ruakura have been published (Bryant, 1981; Bryant and Trigg, 1981; Trigg and Parr, 1981) and will be considered in relation to the present work in Chapter five.

CHAPTER TWO

THE PERFORMANCE OF
HIGH AND LOW BREEDING INDEX COWS
UNDER STALL FEEDING AND GRAZING CONDITIONS

2.1 EXPERIMENTAL AIMS

The aims of the experiments were:-

To examine high (H) and low (L) breeding index (BI) cows in terms of

- milk production and milk composition
- changes in liveweight and condition score
- feed intake
- efficiencies of food conversion under stall feeding and grazing conditions.

2.2 MATERIALS AND METHODS

2.2.1 Environment

The first three experiments were done during 1979/80 and the next two experiments during 1980/81 at Massey University's No. 3 Dairy Research and Development Unit, Palmerston North, New Zealand. The pasture consisted predominantly of perennial rye grasses (Lolium perenne L., cvs. Grasslands Ruanui and Grasslands Nui; Lolium x hybridum Hausskn., cv. Grasslands Ariki) and white clover (Trifolium repens L., cv. Grasslands Huia) with red clover (Trifolium pratense L., cv. Grasslands Hamua) also being used in some of the experiments.

2.2.2 Animals and management

Friesian cows with high (approximately 125) or low (approximately 100) breeding indexes were identified by the Farm Production Division of the New Zealand Dairy Board and purchased from New Zealand dairy farmers. The animals when purchased were approaching their second calving, had identified ancestries for three generations, with at least two generations of male Friesian ancestry. Twenty cows (10 H and 10 L BI) in 1979, and nineteen cows in 1980 (10 H and 9 L BI) were purchased (for details of cows purchased and the

method of calculating breeding indexes, refer Appendix 2.1).

Prior to calving the animals grazed on pasture and were fed hay to ensure that body condition at calving was similar for both genotypes. After calving the cows were grazed on pasture, except during the stall feeding periods when pasture was harvested with a rotary mower, and fed to the animals twice daily at 0830 and 1600 hours. Details of animals involved in each experiment are given in Table 2.1.

Table 2.1 Numbers and ages of animals, feeding treatments and stage of lactation at which each experiment commenced for stall feeding (Experiments 1-4) and grazing experiments (Experiment 5).

Experiment No.	Year	No.cows H L		Age cows (year)	Feeding treatment	Stage lactation expt.started (days since calving)
<u>Stall feeding experiments</u>						
1	1979/80	10	10	3	<u>Ad libitum</u>	30
2	1979/80	10	10	3	<u>Ad libitum</u> ,	80
					70% <u>Ad libitum</u>	
3	1979/80	10	10	3	<u>Ad libitum</u> ,	150
					70% <u>Ad libitum</u>	
4	1980/81	10	8	3	<u>Ad libitum</u> ,	80
					70% <u>Ad libitum</u>	
<u>Grazing experiment</u>						
5	1980/81	16	16	3	Lax grazing	20
				(19 cows)		
				4		
				(13 cows)		

Cows were dried off, on average, after 249 days in 1979/80 and 218 days in 1980/81.

2.2.3 Treatments

The main treatment was genotype, high (H) or low (L) (approximately 125 or 100 as measured by breeding index) but the effect of genotype was examined at different feeding levels as described below. Stall feeding experiments were of five weeks duration with the first week of each period being used as a standardisation period, hence measurements during the first week were not used in the analysis of treatment effects.

Stall feeding experiments

Experiment 1 : Twenty cows (10 H and 10 L) were individually fed pasture ad libitum in stalls, commencing approximately four weeks after the mean calving date (Table 2.1).

Experiments 2, 3, and 4 : High and low BI cows were randomly allocated (within BI groups) to either ad libitum or 70% of previously determined (Experiment 1) ad libitum feeding at approximately 11 weeks after calving (Experiments 2 and 4) and 23 weeks after calving (Experiment 3) (Table 2.1).

In addition, for cows which were restricted, (70% ad libitum feeding) during each experiment, comparisons were made on a within-cow basis between milk fat yields, at the following times : mean milk fat yield during the last two weeks of the stall feeding period and at five weeks before and after this time, when cows were being grazed on pasture at higher levels of nutrition. This enabled differences between genotypes in milk fat yields to be assessed at different levels of feeding on a within-cow basis.

Grazing experiment

Experiment 5 : Thirty-two cows (16 H and 16 L) were laxly grazed on pasture as one group for four weeks commencing approximately three weeks after calving.

Faecal outputs were measured using an indigestible faecal marker; namely chromium sesquioxide (Cr_2O_3). Twelve cows (6 H and 6 L) were randomly chosen and dosed twice daily, after milking, with a gelatine capsule containing 10g Cr_2O_3 in oil (R.P. Scherer Pty. Ltd., Australia). Over the last 10 days of the chromium dosing period, samples of faeces were taken per rectum from each cow twice daily after milking and bulked over five day periods.

2.2.4 Measurements

Milk yield and composition

Milk yield was measured by the use of milk sampling meters (Tru-test Distributors Ltd.,) which sampled a proportion of the milk flow of each cow. Milk yields were recorded twice daily on four consecutive days each week during stall feeding experiments (Experiments 1-4), for three consecutive days in each week for the grazing experiment (Experiment 5), and at least once per week for the remainder of lactation. On each test day a composite sample of afternoon and morning milk was analysed for milk fat concentration (Milko-tester Mk III, Foss Electric, Denmark) and protein concentration (Pro-milk Mk II, Foss Electric, Denmark). The energy concentration of the milk for each cow was predicted using the following equation (estimated from 30 milk samples

from six Friesian cows involved in energy balance experiments reported in Experiment 1, Chapter 3).

$$E = 0.4817 + 0.3805 \text{ FPC} + 0.2837 \text{ PPC} \quad (R^2 = 0.96)$$

where E = energy concentration of the milk (MJ/kg)

FPC = milk fat percentage

PPC = milk protein percentage

Liveweight

Liveweights were recorded at 0800 hours after a 10 hour fast; on three consecutive days at the start (after the week of standardisation for cows in stalls) and end of the experimental periods and routinely once a month during lactation for all cows.

Liveweight decreased suddenly when the feeding level was changed from ad libitum to restricted feeding and this was almost certainly due to a reduction in gut contents.

Metabolic liveweight ($\text{kg}^{0.75}$) was therefore calculated using the average liveweight for each cow over the experimental period, and attempting to adjust liveweights to a common level of gut-fill as outlined below :-

- the liveweight of cows on the restricted level of feeding was used as the base,
- the % reduction in liveweight for each cow due to the restriction in feed intake was calculated for each cow as
$$\frac{\text{liveweight immediately before feed restriction} - \text{liveweight immediately after restriction}}{\text{liveweight immediately before feed restriction}} \times 100$$

liveweight immediately
after feed restriction

The % reductions were averaged for all underfed cows in any particular experiment and the average % reduction was then used to adjust the liveweights of cows on ad libitum feeding.

Reductions in liveweight following the restriction of feed ranged from 6-9%.

The body condition of each cow was scored (Earle, 1976) independently by three observers on two consecutive days at the start and end of each experimental period and monthly during lactation.

Chromium concentration of faeces

The faeces collected from cows grazing on pasture (experiment 5) were dried in an oven at 85degC for 7 days and subsequently ground (1mm sieve). The chromium concentration of the ground dry faeces was determined by the method of Fenton and Fenton, (1979).

Pasture

Stall feeding experiments

The intakes of pasture by individual cows were measured daily during experiments 1 to 4 by weighing the pasture offered to and refused by each cow.

The dry matter concentration of the pasture was determined by drying samples of pastures (done in triplicate) at 85degC for 48 hours. The in vivo, energy and N digestibility of the pasture fed during the stall feeding experiments was estimated using non-pregnant, non-lactating sheep fed to maintain weight (ARC, 1965).

Energy concentration of freeze-dried, ground pasture and sheep faeces was determined using an adiabatic bomb calorimeter and N concentration by the macro-kjeldahl method.

Metabolisable energy intake was estimated for each cow by multiplying the gross energy intake by the energy digestibility determined in vivo, then by the constant 0.82 (ARC, 1980) to convert digestible to metabolisable energy intake.

Grazing experiment

An in vivo estimate of the digestibility of dry matter and N was made during the chromium dosing period in Experiment 5. Sheep were fed on representative samples of pasture which had been cut with a lawn mower to grazing height before grazing by the cows. The faecal output of the cows divided by the in vivo indigestibility of the pasture gave an estimate of the pasture dry matter intake.

2.2.5 Statistical methods

The milk energy output, change in liveweight (Experiment 5) ad libitum intake (Experiments 1-4) were analysed by regression according to the following model:-

$y_{ij} = \mu + m_i + e_{ij}$ for j th cow in i th group
where

i = genetic level, H or L

j = j th cow

μ = mean for L cows

m_L = 0

m_H = difference in mean between genotypes

y = milk energy produced (MJ kg^{-0.75}* cow⁻¹ day⁻¹)

or change in liveweight (kg kg^{-0.75} cow⁻¹ per 28 days)

or ad libitum intake (MJ ME+kg^{-0.75} cow⁻¹ day⁻¹)

e = error

* kg^{-0.75} = per unit metabolic liveweight

+ ME = metabolisable energy

It was considered that the use of 0.75 as the exponent for scaling intake and performance variables was appropriate for two reasons. Firstly it enabled comparisons to be made readily with the results of many other workers (see p. 54). Secondly the actual exponents for H and L cows, calculated by regression analyses of the present experimental data, were 0.83 ± 0.17 (s.e.) and 0.53 ± 0.32 for the relation between \log_{10} liveweight and \log_{10} metabolisable energy intake. The two exponents could not be shown to be significantly different, therefore a pooled exponent (0.69 ± 0.16) was calculated, a value which was close to 0.75.

The milk energy produced and change in liveweight for Experiments 1-4 were analysed by stepwise regression according to the following model (Townesley et al. 1981)

$$y_{ij} = \mu + \alpha_i + m_i x_{ij} + e_{ij}$$

for jth cow in ith group

where

i = genetic level; H or L

j = jth cow

μ = intercept for L cows

α_L = 0

α_H = difference in intercept (between genotypes)

y = milk energy produced ($\text{MJ kg}^{-0.75} \text{ cow}^{-1} \text{ day}^{-1}$)
or change in liveweight ($\text{kg kg}^{-0.75} \text{ cow}^{-1}$ per
 28 days)

m = slope term

x = metabolisable energy intake ($\text{MJkg}^{-0.75} \text{ cow}^{-1}$
 day⁻¹)

e = between cow error

Differences between genotypes in slope were tested first followed by differences in intercept.

Faecal output (Experiment 5) was analysed as a split-plot (Snedecor and Cochran, 1967) with cows as whole plots, and with whole plot treatments (H and L BI) "allocated" to cows. Within cows (whole-plot) are split-plot treatments (periods; 0-5 days, 6-10 days faecal collection).

The model was :-

$$y_{ijk} = \mu + m_i + e_{ij} + T_k + (mT)_{jk} + \epsilon_{ijk}$$

where

y = faecal output ($\text{kg kg}^{-0.75} \text{ cow}^{-1} \text{ day}^{-1}$)

m = whole plot treatment

e_{ij} = whole plot error

T = sub-plot treatment

ϵ_{ijk} = sub-plot (within cow) error

i = genetic level (H,L)

j = j th cow

k = observation on j th cow ($k=1,2$)

Multivariate analysis was used to examine differences between genotypes in response to level of feeding on a within-cow basis (Experiments 2,3,4), and in milk fat yield in early, mid, and late lactation.

The analysis is analagous to a split-plot analysis where the repeated observations on the same cow correspond to the sub-plot factor. The model was:-

$$Y_{ijk} = \mu_k + m_{ik} + e_{ijk}$$

where

y = milk fat yield ($\text{kg cow}^{-1} \text{ day}^{-1}$)

μ = mean

m = treatment effect

e = error

i = genetic level (H,L)

j = j th cow

k = observation on j th cow ($k=1,2,3$)

The test statistic used in the multiple analysis of variance (MANOVA) is Wilks likelihood ratio test (see Bock, 1975 p152). Actual significance levels are presented in the results, and unless otherwise stated, a critical significance level of 5% has been taken.

2.3

RESULTS

2.3.1 Milk yield and liveweight changes over the whole lactation

High BI cows produced 28% and 18% more milk fat than L cows in 1979/80 and 1980/81 respectively (Table 2.2).

Table 2.2 Lactation performance (1979/80 and 1980/81) for high (H) and low (L) breeding index cows.

	1979/80		1980/81	
	H	L	H	L
No. of cows	10	10	19	17
Breeding index	127	100	126	102
Condition score at calving	4.7	4.7	4.7	4.9
Liveweight at calving (kg)	383	411	389	421
Lactation length (days)	249	249	238	233
Milk yield (l)	3270	2816	3500	3060
Milk fat yield (kg)	150	117	158	133
Milk protein yield (kg)	114	96	122	108
Milk fat %	4.58	4.22	4.46	4.33
Milk protein %	3.48	3.44	3.48	3.51
Change in condition score during lactation	-0.8	+0.1	-0.6	0
Change in liveweight during lactation (kg)	+5	+39	+22	+35

Although H and L cows calved in similar condition, H cows lost condition during lactation whereas L cows maintained condition (Table 2.2).

Daily milk fat yield and condition scores for 1979/80 (Figure 2.1) and 1980/81 (Figures 2.2 and 2.3) are shown graphically. Data for three and four year old cows are presented separately for 1980/81, because three year old cows were fed indoors for five weeks at different levels of feeding, whereas the four year old cows were grazed on pasture throughout lactation.

Differences in milk fat yield between H and L cows were also examined at different stages of lactation (Table 2.3).

Table 2.3 Milk fat yields(g cow ⁻¹ day ⁻¹) for high (H) and low (L) breeding index cows at different stages of lactation during 1979/80 and 1980/81.

	1979/80			1980/81		
	Stage of lactation (days)			Stage of lactation (days)		
	30	150	240	30	150	210
Milk fat yield H cows (g cow ⁻¹ day ⁻¹)	659	589	545	901	594	424
Difference in milk fat yield (H - L)	107	137	204	86	92	62
s.e. difference*	38	31	50	54	28	26
H/L	1.19	1.30	1.60	1.11	1.18	1.17

* the standard error (s.e.) of the difference in milk fat yield was derived from the multivariate analysis for which the analysis of variance is shown in Table 2.4

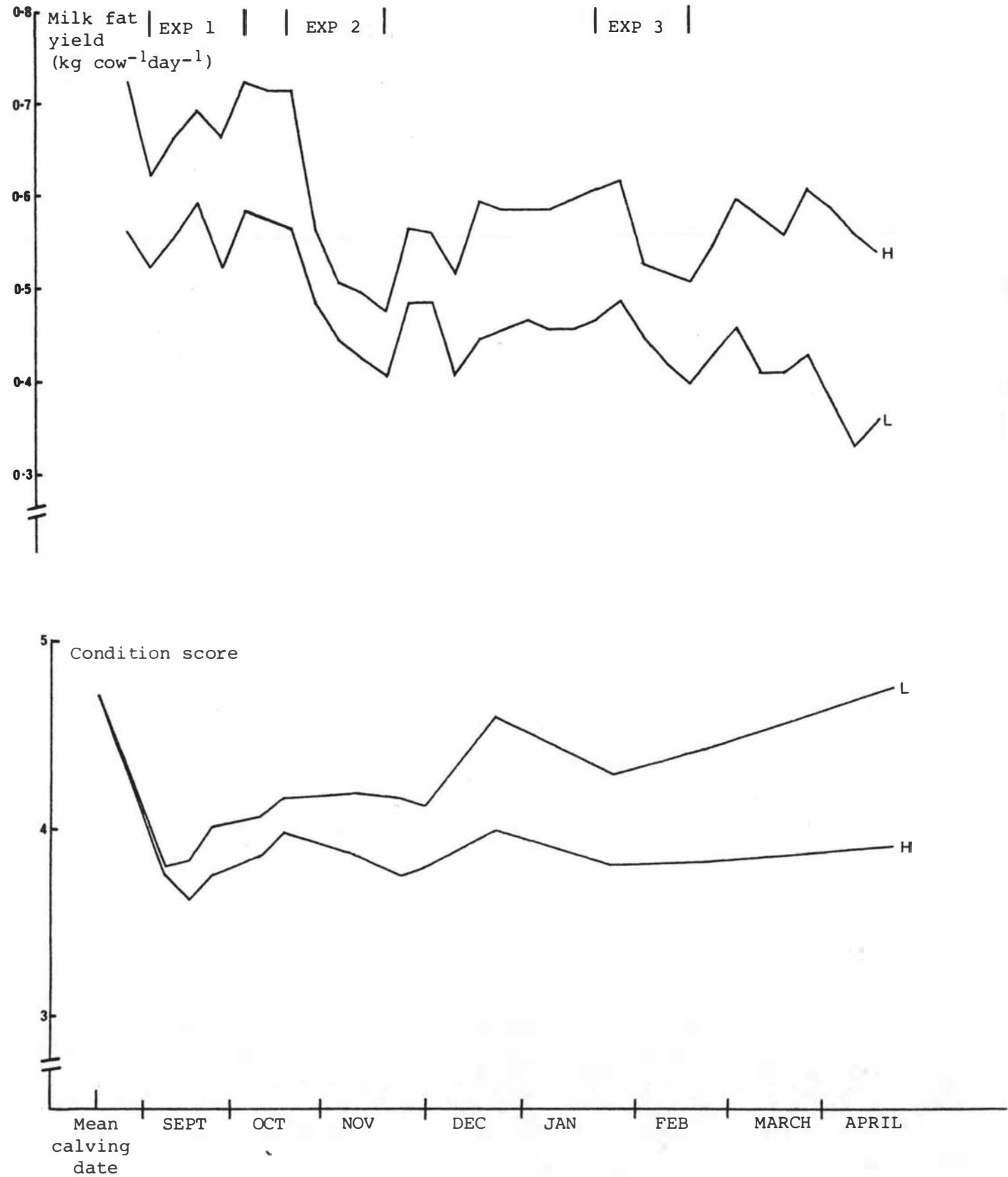


FIG.2.1 Mean daily milk fat yield and mean condition scores for ten high (H) and ten low (L) breeding index cows during 1979-80.

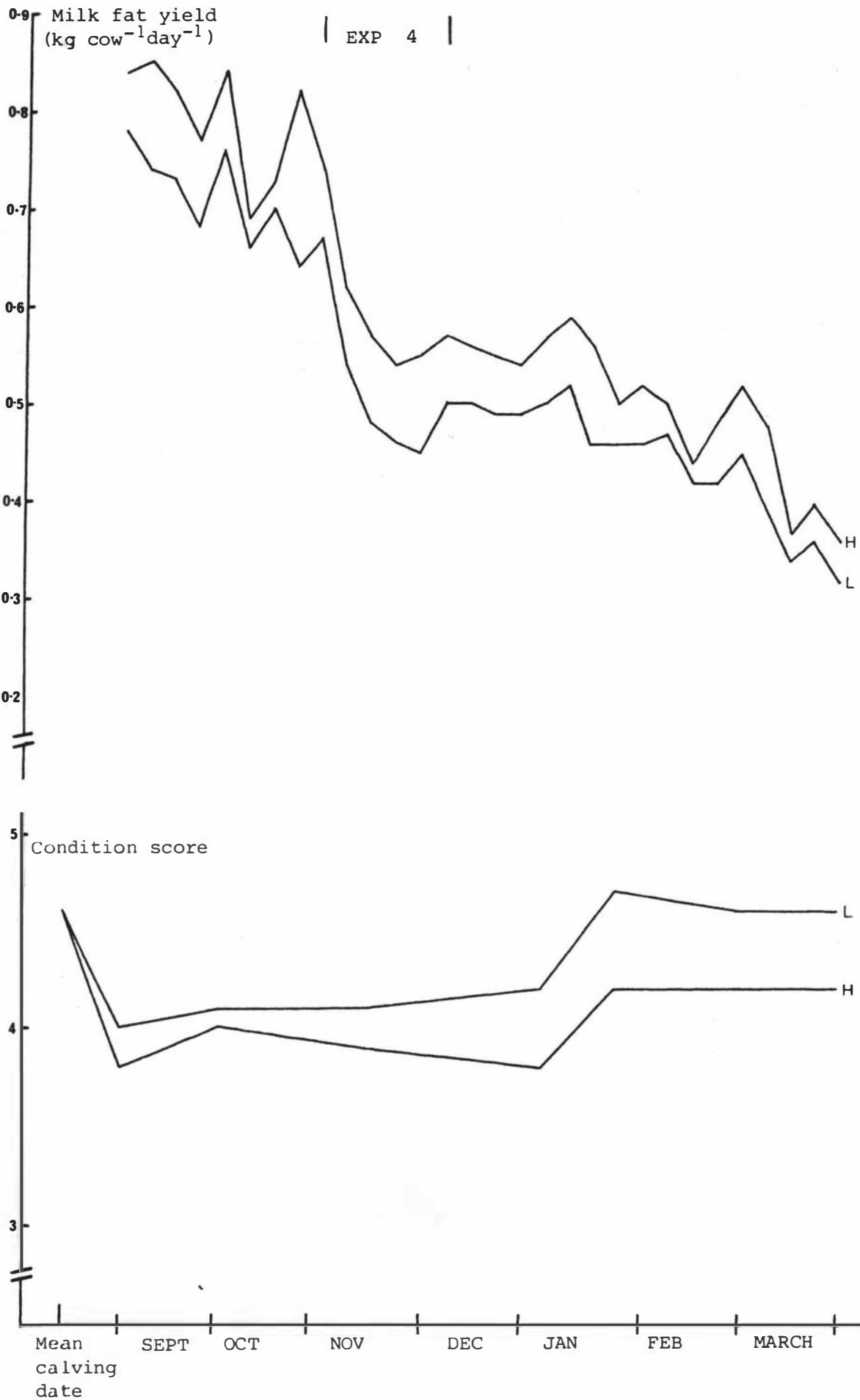


FIG.2.2 Mean daily milk fat yield and mean condition scores for ten high (H) and seven low (L) breeding index cows during 1980-81 (three yr. old cows).

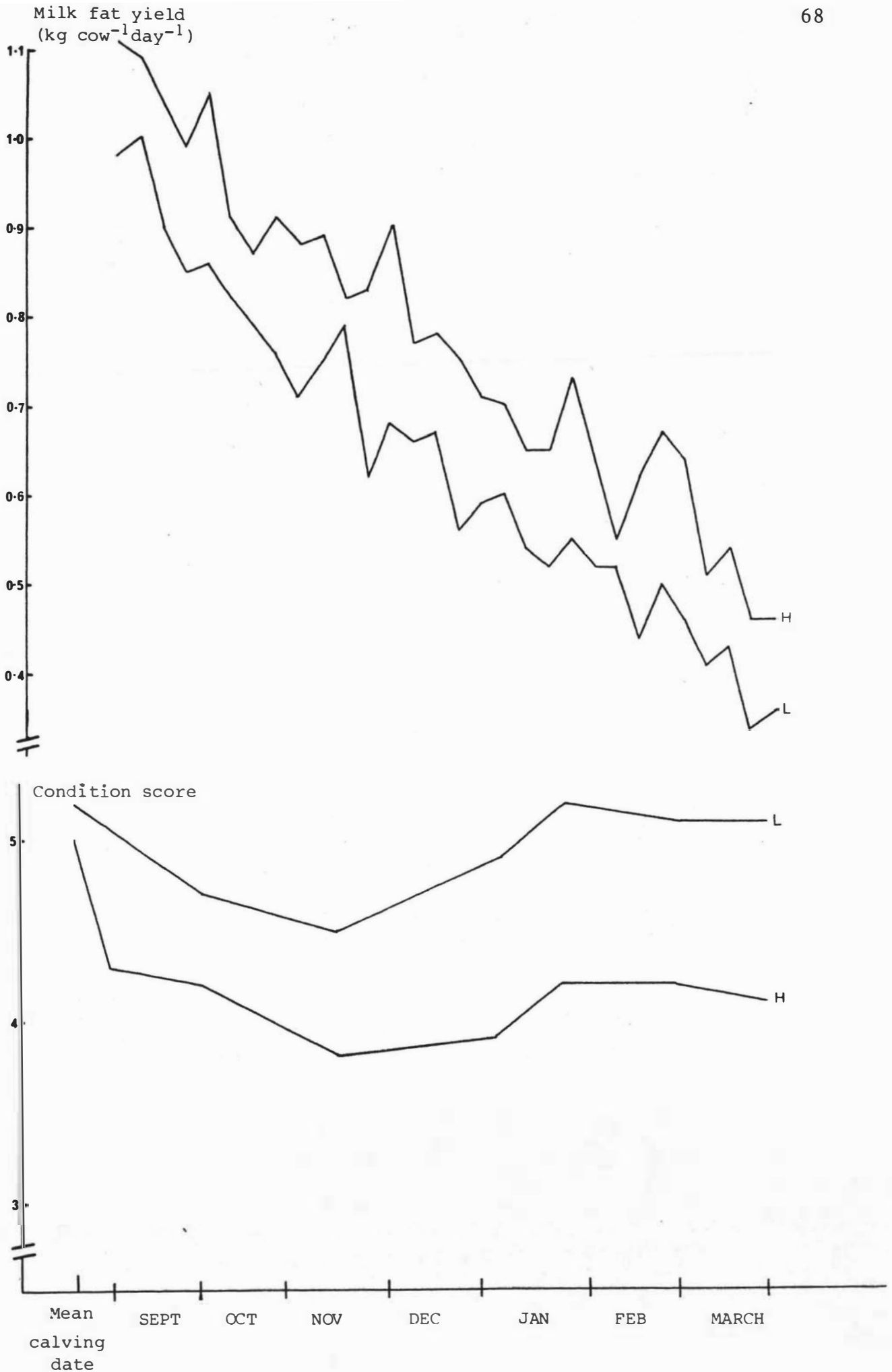


FIG. 2.3 Mean daily milk fat yield, and mean condition scores for six high (H) and seven low (L) breeding index cows during 1980-81 (4 yr. old cows).

The test of independence of errors (χ^2 3 d.f.) was significant ($P < 0.01$) for 1979/80 and 1980/81 justifying the use of multivariate analysis in examining differences in fat yield between genotypes at different stages of lactation.

High cows produced significantly more milk fat ($P < 0.01$ and $P = 0.02$ for 1979/80 and 1980/81 respectively) than L cows considered jointly at the three stages of lactation (Table 2.4).

In 1979/80 the differences in milk fat yield between genotypes increased as the lactation proceeded (Table 2.3), but the increased magnitude of the difference was not significant (Table 2.4). The results in 1980/81 were in marked contrast to those from 1979/80 because although the difference (H - L) in fat yield increased from early to mid lactation (Table 2.3), it declined from mid to late lactation. However considered jointly at the three 'times' during lactation there was no BI x times interaction in 1980/81 (Table 2.4)

The data were further examined by comparing the difference in milk fat yield between genotypes for early versus mid and early versus late lactation periods using contrasts, but the magnitude of the difference did not differ significantly, for the two contrasts studied (Table 2.4).

Table 2.4

Multivariate analysis of differences in milk fat yield between genotypes (BI) at early, mid, and late lactation ('times') on a within-cow basis for 1979/80 and 1980/81 (see text for further details).

Source of variation	df hypothesis	Generalised SS	Lambda	df Chi	χ^2	Probability
<u>1979/80</u>						
BI †	1	0.61086	0.4524	1	13.9	0.02
Contrast 1 ϕ	1	0.04905	0.9541	1	0.8	0.36
Contrast 2 ϕ	1	0.16059	0.8535	1	2.8	0.10
Times x BI ξ	1	0.00276	0.8282	2	3.2	0.20
<u>Error</u>						
BI	18	0.27635				
Contrast 1	18	0.04680				
Contrast 2	18	0.13706				
Times	18	0.00229				
<u>1980/81</u>						
BI	1	0.74256	0.8077	1	5.9	0.02
Contrast 1	1	0.19019	0.9994	1	0.0	0.90
Contrast 2	1	0.22817	0.9900	1	0.3	0.60
Times x BI	1	0.0093	0.9313	2	1.9	0.38
<u>Error</u>						
BI	28	0.59974				
Contrast 1	28	0.19009				
Contrast 2	28	0.22589				
Times	28	0.00864				

- † Test of average difference between genotypes at three stages of lactation('times').

- ϕ The contrasts test whether there is a difference in the 'difference in fat yield' between genotypes in early versus mid lactation (contrast 1) and early versus late lactation (contrast 2).

- ξ Times x BI jointly tests whether differences between genotypes in milk fat yield change at the three stages of lactation.

2.3.2 Feed intakes and feed quality

Measures of feed quality for the different experiments are presented in Table 2.5.

Table 2.5 Digestibility of energy and nitrogen, estimated metabolisable energy concentration and crude protein concentration of the pasture for Experiments 1 - 5.

Experiment	<u>Digestibility (%)</u>		<u>Composition of DM</u>	
	Energy	Nitrogen	Metabolisable energy (MJ/kg)	Crude Protein %
1	75.0	75.4	11.2	21.2
2	70.4	65.9	10.5	16.2
3	69.4	72.6	10.3	20.6
4	74.8	70.8	11.5	15.9
5*	69.4#	81.0	10.8	28.5

* Data presented refer to the chromium dosing period carried out over 10 days.

Dry matter (DM) digestibility so that DM intake can be calculated from faecal DM output.

Both genotypes had similar metabolisable energy intakes of fresh cut pasture offered ad libitum. However H cows had higher metabolisable energy intakes per unit metabolic liveweight than L cows, but these differences in intake were significant only in Experiments 1 ($P < 0.10$) and 3 ($P < 0.05$).

Table 2.6 Metabolisable energy intakes
(MJkg^{-0.75} cow⁻¹ day⁻¹) of fresh cut
pasture offered ad libitum in stalls to
high (H) and low (L) breeding index cows
(Experiments 1 - 4)

Experiment	No. cows		Intake H	H - L	s.e. diff	Probability
	H	L				
1	10	10	1.872 (147.8)*	0.094 (-1.5)	0.050	0.08
2	5	5	1.958 (159.8)	0.005 (-5.1)	0.065	0.94
3	5	5	2.050 (168.1)	0.212 (0.7)	0.048	< 0.01
4	5	4	2.062 (176.5)	0.013 (6.0)	0.076	0.86

* Figures contained in brackets are metabolisable energy intakes (MJcow⁻¹day⁻¹).

Estimates of faecal outputs and dry matter intakes for cows grazing pasture (Experiment 5) are presented in Table 2.7.

There were no significant differences in faecal output either between the two five day collection periods or between genotypes (refer Appendix 2.2.1 for statistical analysis). The least significant difference (P = 0.05) between genotypes in faecal output was 0.003 whereas the actual difference was 0.001 kg faecal DM kg^{-0.75} cow⁻¹ day⁻¹.

Table 2.7 Estimated average faecal outputs and dry matter intakes for six high (H) and six low (L) breeding index cows grazing pasture during consecutive five day periods (Experiment 5).

	Periods (days)		
	0-5	6-10	0-10
<u>Faecal output</u>			
(kg faecal DM kg ^{-0.75} cow ⁻¹ day ⁻¹)			
H	0.0464 (3.97)*	0.0451 (3.89)	0.0457 (3.93)
L	0.0459 (3.99)	0.0445 (3.84)	0.0452 (3.92)
<u>Estimated dry matter intake</u> ϕ			
(kg DM cow ⁻¹ day ⁻¹)			
H	13.0	12.7	12.8
L	13.0	12.5	12.8

* Figures enclosed in brackets are faecal outputs (kg faecal DM cow⁻¹ day⁻¹).

ϕ Calculated using a dry matter digestibility of 69.4% (refer Table 2.5).

2.3.3 Milk production and liveweight changes

Experiment 1

There was a 21% increase in milk fat yield of the H versus L cows caused by differences in both milk yield and milk fat concentration (Table 2.8).

Table 2.8 Dry matter intake, liveweight, condition score, milk production and milk composition of high (H) and low (L) breeding index cows fed pasture ad libitum in stalls (Experiment 1).

Item	H	L
Average liveweight † over experimental period (kg)	363	397
Average condition score at start of experimental period	3.7	3.8
Dry matter intake (kg DM cow ⁻¹ day ⁻¹)	13.0	13.1
Milk yield (L cow ⁻¹ day ⁻¹)	15.9	14.4
Milk fat yield (g cow ⁻¹ day ⁻¹)	676	559
Milk protein yield (g cow ⁻¹ day ⁻¹)	502	451
Milk fat %	4.27	3.80
Milk protein %	3.16	3.05
Change in liveweight (kg per 28 days)	+4.9	+11.1
Change in condition score (per 28 days)	+ 0.23	+ 0.37

† liveweights unadjusted for gut-fill.

High BI cows produced 23% more milk energy than L cows, reducing to 18% after covariance adjustment for differences in intake (Table 2.9). Low BI cows gained more liveweight than H cows during the experiment, but this difference was not significant (Table 2.9).

Table 2.9 Mean milk energy output ($\text{MJ kg}^{-0.75} \text{cow}^{-1} \text{day}^{-1}$) and liveweight changes ($\text{kg kg}^{-0.75} \text{cow}^{-1} \text{day}^{-1}$) for high (H) and low (L) breeding index cows before and after covariance adjustment for differences in intake (Experiment 1).

Item	H	H - L	s.e. diff	Probability
Milk energy	0.603	0.113	0.024	< 0.01
Milk energy after covariance adjustment	0.593	0.093	0.024	< 0.01
Liveweight change	0.066	-0.067	0.061	0.28
Liveweight change after covariance adjustment	0.088	-0.022	0.063	0.73

Experiments 2, 3, and 4

Treatment means for Experiments 2, 3, and 4, where cows were fed pasture in stalls at ad libitum or restricted levels, are presented in Tables 2.10, 2.11, and 2.12. High BI cows consistently produced more milk fat than L cows and this was associated with increases in milk yield and milk fat concentration.

Table 2.10 Treatment means for Experiment 2 for different genotypes (BI) at two levels of feeding (FL).

Item	High BI		Low BI	
	High FL	Low FL	High FL	Low FL
Dry matter intake (kg DM cow ⁻¹ day ⁻¹)	15.1	9.6	15.6	9.5
Average liveweight * over experimental period (kg)	372	353	404	396
Milk yield (l cow ⁻¹ day ⁻¹)	13.7	10.5	13.0	9.8
Milk fat yield (g cow ⁻¹ day ⁻¹)	578	438	480	400
Milk protein yield (g cow ⁻¹ day ⁻¹)	450	322	425	302
Milk fat %	4.23	4.17	3.70	4.10
Milk protein %	3.29	3.07	3.27	3.10
Change in liveweight (kg per 28 days)	+16	-2	+15	+5
Change in condition score (per 28 days)	-0.05	-0.10	+0.03	-0.15

* liveweights unadjusted for gut-fill.

Table 2.11 Treatment means for Experiment 3 for different genotypes (BI) at two levels of feeding (FL).

Item	High BI		Low BI	
	High FL	Low FL	High FL	Low FL
Dry matter intake (kg DM cow ⁻¹ day ⁻¹)	16.2	9.6	16.1	9.2
Average liveweight * over experimental period (kg)	396	377	442	424
Milk yield (ℓ cow ⁻¹ day ⁻¹)	13.3	10.1	11.1	8.7
Milk fat yield (g cow ⁻¹ day ⁻¹)	588	483	461	395
Milk protein yield (g cow ⁻¹ day ⁻¹)	446	341	372	294
Milk fat %	4.43	4.80	4.16	4.52
Milk protein %	3.36	3.39	3.36	3.37
Change in liveweight (kg per 28 days)	-1	+7	+3	+2
Change in condition score (per 28 days)	0	+0.05	+0.17	+0.08

* liveweights unadjusted for gut-fill.

Table 2.12 Treatment means for Experiment 4 for different genotypes (BI) at two levels of feeding (FL).

Item	High BI		Low BI	
	High FL	Low FL	High FL	Low FL
Dry matter intake (kg DM cow ⁻¹ day ⁻¹)	15.4	10.5	14.9	10.1
Average liveweight * over experimental period (kg)	398	367	384	382
Milk yield (g cow ⁻¹ day ⁻¹)	14.8	11.7	13.7	10.4
Milk fat yield (g cow ⁻¹ day ⁻¹)	660	484	567	439
Milk protein yield (g cow ⁻¹ day ⁻¹)	498	366	448	336
Milk fat %	4.46	4.14	4.14	4.22
Milk protein %	3.36	3.13	3.27	3.23
Change in liveweight (kg per 28 days)	+16	+3	+22	+3
Change in condition score (per 28 days)	+0.04	-0.08	+0.08	+0.10

* liveweights unadjusted for gut-fill.

The statistical analyses of the data have been confined to testing the significance of differences between genotypes for milk energy output and live weight changes.

Milk energy output is considered first.

Two measures of the efficiency with which the food eaten is converted to milk are marginal and gross efficiency. Marginal efficiency is represented by the regression coefficient estimated from the regression of milk energy output on energy intake, i.e. the increase in milk energy per unit increase in feed energy. Gross efficiency is the milk energy produced per unit of feed energy eaten

$$\left\{ \frac{\text{milk energy}}{\text{feed energy}} \right\}$$

Although in each experiment H cows had greater marginal efficiencies (represented by the regression coefficients) the differences between genotypes were not significant, hence a pooled coefficient was calculated (Table 2.13).

The significance levels for differences in coefficients between genotypes were $P = 0.42, 0.56$ and 0.20 for Experiments 2, 3, and 4 respectively (refer Appendix 2.2.2 for statistical analysis). On testing intercepts (C) between BI groups, H cows produced significantly more milk energy at a common metabolisable energy intake than L cows i.e. H cows had higher gross efficiencies of conversion of feed energy to milk energy than L cows (Table 2.13).

Table 2.13 Regression analyses relating milk energy output ($\text{MJ kg}^{-0.75} \text{cow}^{-1} \text{day}^{-1}$) and live weight change ($\text{kg kg}^{-0.75} \text{cow}^{-1} \text{day}^{-1}$) to metabolisable energy intake (MEI; $\text{MJ kg}^{-0.75} \text{cow}^{-1} \text{day}^{-1}$) for high (H) and low (L) breeding index cows for Experiments 2, 3, and 4. $Y = b\text{MEI} + C$

Experiment	Y	MEI ϕ (b-s.e.)	C* (H)	C (H-L)	s.e.†
2	Milk energy	0.155 \pm 0.021	0.192	0.056	0.016
	Liveweight change	0.211 \pm 0.049	-0.257	-0.040	0.038
3	Milk energy	0.150 \pm 0.028	0.193	0.088	0.020
	Liveweight change	-0.064 \pm 0.049	0.134	0.012	0.035
4	Milk energy	0.181 \pm 0.027	0.159	0.036	0.017
	Liveweight change	0.300 \pm 0.073	-0.406	-0.034	0.047

ϕ pooled regression coefficient

* intercept for H breeding index cows

† standard error of difference in intercept between genotypes.

There were no significant differences between genotypes for the change in liveweight per unit change in energy intake therefore a pooled coefficient was calculated. In Experiments 2, and 4, L cows gained more, or lost less liveweight than H cows at a fixed intake, but the reverse was the case for Experiment 3 although the differences between genotypes were not significant (Table 2.13). Both H and L cows gained significantly

more liveweight at the higher levels of intake during Experiments 2 and 4 but not during Experiment 3 (Table 2.13).

Response to level of feeding was also compared on a within-cow basis for cows which were fed on the lower level of feeding during Experiments 2, 3, and 4 (Table 2.14). The number of cows involved were five H and five L; five H and five L; and five H and four L cows for Experiments 2, 3, and 4 respectively. The significance levels for testing the independence of the errors of the repeated measurements $\chi^2 = 3$ df were $P = 0.07$, $P < 0.01$ and $P = 0.81$ for Experiments 2, 3, and 4 respectively (Appendix 2.2.3). Multivariate analysis was used for the three experiments. In Experiment 4 where the errors were independently distributed, the multivariate test of significance may have had slightly less power than the corresponding univariate tests.

For Experiments 2, and 3, but not for Experiment 4, H cows showed larger changes in milk fat yield in response to changes in feeding level than L cows, but these differences were not significant (refer Appendix 2.2.3 for details of analysis.)

Table 2.14 Milk fat yield and derived responses for high (H) and low (L) breeding index cows; five weeks before underfeeding (FY-5), during the last two weeks of underfeeding (FY0) in Experiments 2, 3, and 4, and five weeks after underfeeding (FY+5).

Item	Milk fat yield (g cow ⁻¹ day ⁻¹)			Response to level of feeding (g fat cow ⁻¹ 35 days ⁻¹)			
	H	L	H-L	FY-5 → FY0		FY0 → FY+5	
				H	L	H	L
<u>Expt 2</u>							
FY-5	718	600	118				
FY0	404	358	46	-314	-242	176	76
FY+5	580	434	146				
<u>Expt 3</u>							
FY-5	588	456	132				
FY0	438	348	90	-150	-108	122	62
FY+5	560	410	150				
<u>Expt 4</u>							
FY-5	724	665	59				
FY0	460	390	70	-264	-275	70	95
FY+5	530	485	45				

Experiment 5

High BI cows produced more milk fat than L cows but this was associated only with an increase in milk yield since milk fat concentration was similar for both genotypes (Table 2.15). High BI cows produced significantly ($P < 0.01$) more milk energy than L cows (0.774 v. 0.661 MJ kg^{-0.75} cow⁻¹ day⁻¹; s.e. difference = 0.035) and had similar weight gains to L cows (0.255 v. 0.231 kg kg^{-0.75}; s.e. difference = 0.047).

Table 2.15 Treatment means for high (H) and low (L) breeding index cows grazing on pasture as one group in early lactation (Experiment 5).

Item	H	L
Average liveweight * at start of experimental period (kg)	385	404
Average condition score at start of experimental period (kg)	4.0	4.3
Estimated dry matter intake ϕ (kg DM cow ⁻¹ day ⁻¹)	12.8	12.8
Milk yield (ℓ cow ⁻¹ day ⁻¹)	20.3	17.7
Milk fat yield (g cow ⁻¹ day ⁻¹)	930	805
Milk protein yield (g cow ⁻¹ day ⁻¹)	726	658
Milk fat %	4.64	4.60
Milk protein %	3.59	3.76
Change in liveweight (kg per 28 days)	+21	+20
Change in condition score (per 28 days)	+0.1	+0.1

* liveweights unadjusted for gut-fill

ϕ the pasture intake was estimated indirectly using chromium sesquioxide to estimate faecal output for 12 of the 32 cows involved over the last ten days of the four week experimental period.

2.4

DISCUSSION

In the 1979/80 lactation a difference of 27 BI units was associated with a difference in milk fat yield of 28% and in 1980/81 the difference in BI units of 24 was associated with a milk fat production difference of 18%. The agreement between the expected difference (based on breeding indexes) and the observed difference in milk fat yield was close.

In 1979/80, milk fat productions were low ($0.5 - 0.7 \text{ kg cow}^{-1} \text{ day}^{-1}$) in early lactation because of severe underfeeding during the first four weeks of lactation which was also associated with the loss of 0.9 units of body condition. In 1980/81 the level of feeding in early lactation was better and production levels were higher, $0.8 - 0.9 \text{ kg milk fat cow}^{-1} \text{ day}^{-1}$.

However, despite the higher production in early lactation, total production during 1980/81 was low, but this could be partly accounted for by a short lactation length (220 days) and a low availability of pasture in mid and late lactation.

During Experiment 1 cows were individually fed indoors whereas in Experiment 5 cows were grazed as one group. However the difference between genotypes in daily milk fat yield was similar in both experiments; 0.12 kg/cow . This suggests that competition and grazing components are not important factors accounting for differences between genotypes.

Both milk yield and the concentration of milk fat in the milk of the H cows was consistently higher than that of the L cows. This may indicate that selection for increased levels of milk fat production per cow has been associated with increases in milk yield and milk fat concentration.

Whilst it is clear that the H cows produced more milk fat than L cows, the reasons for the increased production are not as clear cut. Differences in intake and partitioning of energy between milk and liveweight appear to be implicated.

A consistent feature of the data was that in each experiment H cows had higher intakes per unit metabolic liveweight than L cows, although these differences in intake were significant only in Experiments 1 ($P < 0.10$) and 3 ($P < 0.05$).

High BI cows also partitioned more energy to milk rather than liveweight, than L cows for Experiments 1, 2, and 4, but not for Experiments 3 and 5. Estimates of liveweight changes in Experiment 3 appear to be suspect as, for example, the H cows lost weight on the high level of feeding, but gained weight on the low level of feeding. The reason for the inconsistent data on weight changes in Experiment 3 compared to the other experiments is not known.

For Experiment 5 the L cows had a slightly lower liveweight gain than the H cows but this was probably because of the slightly better condition of the L cows at the start of the experiment (4.3 v 4.0) since cows in higher condition partition less of their energy to liveweight gain (Grainger et al. 1982).

Over the whole lactation, differences in liveweight and condition score changes between H and L cows were more apparent (Table 2.2). These are differences in liveweight or condition that have occurred over approximately eight months. If these differences are expressed on a monthly basis then for example, in 1979/80 the L cows gained 4.2kg more liveweight per month than the H cows. This difference in weight gain is obviously very small and would be extremely difficult to detect. It is not surprising therefore that the liveweight changes measured over the short periods of Experiments 1 to 5 are not significantly different between genotypes.

To examine whether observed differences in intake and energy partitioning could explain the differences in milk energy output between genotypes, data for Experiments 1 to 3 were pooled (Table 2.16).

Table 2.16 Average metabolisable energy intake (MEI), milk energy output and change in tissue energy for Experiments 1 to 3.

Item	H	L	H-L	% difference HvL
Observed MEI (MJ kg ^{-0.75} cow ⁻¹ day ⁻¹)	1.723	1.612	0.111	7
Milk energy (MJ kg ^{-0.75} cow ⁻¹ day ⁻¹)	0.496	0.399	0.097	24
Change in tissue energy † (MJ kg ^{-0.75} cow ⁻¹ day ⁻¹)	0.043	0.064	-0.021	-33

† calculated by assuming 20MJ energy retained per kg liveweight (Holmes et al. 1981).

The question posed was : Could the measured difference in metabolisable energy intake between genotypes be explained in terms of the measured difference between genotypes in output of milk energy and changes in liveweight ?

The procedure followed was:-

- 1 The estimated differences (H-L) in energy retained as milk and body tissue were (Table 2.16) :-

$$\text{Difference in milk energy} = 0.097 \text{ MJ kg}^{-0.75} \text{ cow}^{-1} \text{ day}^{-1}$$

$$\text{Difference in tissue energy} = -0.021 \text{ MJ kg}^{-0.75} \text{ cow}^{-1} \text{ day}^{-1}$$

- 2 Assuming similar maintenance requirements for both genotypes k_l and k_g values of 0.65 (Holmes et al. 1981) can be used to convert the differences (H-L) in milk and tissue energy between genotypes to metabolisable energy. The equivalent MEI'^s are :-

$$\text{Differences in milk energy} = \frac{0.097}{0.65} = 0.149$$

$$\text{Difference in tissue energy} = \frac{-0.021}{0.65} = -0.032$$

Therefore the "expected" difference in MEI = 0.149 - 0.032 = 0.117 MJ kg^{-0.75} cow⁻¹ day⁻¹

- 3 Express the measured difference in MEI (0.111 MJ kg^{-0.75} cow⁻¹ day⁻¹, from Table 2.16) as a ratio of the "expected" difference in MEI (0.117).

$$\frac{\text{Measured difference in MEI}}{\text{"Expected" difference in MEI}} = \frac{0.111}{0.117} = 0.95$$

Thus although the differences in intake and partitioning of energy between milk and liveweight are small, they explain 95% of the observed difference.

Based on a summary of recent nutrition experiments (Holmes et al. 1981), the measured metabolisable energy intakes appear to be high for the observed energy retention in milk and tissue. It is suggested that the reason for this difference is related to soil contamination of the feed which was noticed during feeding in all

experiments, but particularly in Experiment 3. For this reason it is acknowledged that in the indoor experiments the ash concentration of the feed should have been monitored, and that estimates of intake based on chromium (Experiment 5) should have been expressed on an organic matter basis. However in the indoor feeding work it would have been difficult to obtain representative sub-samples of the soil-contaminated feed for two reasons. Firstly the soil was unevenly distributed throughout the feed. Secondly any handling of the feed, such as for determination of dry matter concentration, tended to shake free some of the soil in the feed. Hence it is suggested that in future experiments where the feed is harvested, an attempt should be made to minimize soil contamination of the feed.

An estimate of soil contamination was obtained in Experiment 3 when faecal outputs of the cows were estimated indirectly using chromium sesquioxide. The soil content of the faeces of the cows ranged from 19 to 29% being higher with cows on the restricted level of feeding but similar for both genotypes (Appendix 2.3). When the intakes were adjusted for soil contamination the ratio of observed/predicted MEI (based on measured values in milk and tissue energy) was 1.17, but using uncorrected intakes the ratio was 1.30, a much greater discrepancy (Appendix 2.3). The equivalent ratios calculated for Experiments 1 and 2 and 4, but uncorrected for possible soil contamination were 1.17, 1.18, and 1.15 respectively.

It seems likely, therefore, that the apparently high intakes were due mainly to soil ingestion, but the extent of this effect is known only for Experiment 3. The data cannot be readily compared with other experimental work

because the extent of the effect of soil contamination on the estimation of intake is not known.

Another problem, recently highlighted by Cowan et al. (1980) is the difficulty, particularly in the short-term, of obtaining reliable estimates of changes in liveweight. The measured liveweight changes may not provide an accurate measure of changes in body tissue energy and this may explain some of the discrepancy between observed and "expected" metabolisable energy intake.

Data on the feed requirements to maintain and gain body condition for non-lactating H and L cows are presented in Chapter 3.

2.5

CONCLUSIONS

The higher milk and milk fat production of high compared with low breeding index cows can be almost completely explained by their higher intakes and that they utilise a greater proportion of metabolisable energy intake for the synthesis of milk and a smaller proportion for the synthesis of body tissue. Differences in intake between genotypes were small but reasonably consistent. Differences in partitioning of energy between milk and body tissue by the different genotypes can be clearly seen over the whole lactation, but in the short-term, differences in partitioning of energy were much smaller and were more difficult to detect.

CHAPTER THREE

ENERGY AND NITROGEN BALANCE EXPERIMENTS WITH LACTATING AND NON-LACTATING HIGH AND LOW BREEDING INDEX COWS

3.1 EXPERIMENTAL AIMS

The aims were to measure, for high and low breeding index cows:-

(a) Whilst lactating

- the intake and utilisation of energy and nitrogen;
- the efficiency of use of metabolisable energy for total energy retention as milk and body tissue.

(b) Whilst dry

- the quantity of dietary energy required to maintain zero energy retention and zero change in body condition;
- the efficiency with which dietary energy is utilised for the retention of energy and for gain in body condition and liveweight.

3.2 MATERIALS AND METHODS

3.2.1 Experiment 1 : Energy balances with lactating cows in early and late lactation.

Six, four-year-old Friesian cows (three high and three low breeding index) were selected and accustomed to routines and equipment for excreta collection and calorimetry procedures. Average breeding indexes and calving dates were 127 and August 2nd; and 101 and August 5th; for the high breeding index (H) and low breeding index (L) cows respectively.

Cows were paired, one H and one L, according to calving date, and each pair (earliest calving pair first) received the following feeding sequence commencing approximately three weeks after calving;

ad libitum, 70% ad libitum, and ad libitum.

Approximately seven months after calving each pair were fed ad libitum, and then 70% ad libitum.

Each feeding level comprised a ten day preliminary, and a ten day collection period, with a total of 60 and 40 days in early and late lactation respectively (Table 3.1).

Table 3.1 Plan of Experiment 1, outlining the chronological sequence (days of experiment) by pairs of feeding treatments ad libitum, restricted, ad libitum in early lactation and ad libitum, restricted feeding in late lactation during 1980/81 lactation (see text for further details).

	<u>Ad libitum</u>		Restricted		<u>Ad libitum</u> *	
	Prelimin- ary	Balance	Prelimin- ary	Balance	Prelimin- ary	Balance
	(days)		(days)		(days)	
Pair 1	1-10	11-20	21-30	31-40	41-50	51-60
Pair 2	6-15	16-25	26-35	36-45	46-55	56-65
Pair 3	11-20	21-30	31-40	41-50	51-60	61-70

* In late lactation there were only two balance periods for each cow; ad libitum followed by restricted feeding.

During the last five days of each collection period, measurements of respiratory gas volumes were made.

Cows were fed at 0800 and 1500 hours on predominantly rye grass and white clover pastures, harvested daily. In early lactation the pasture was mown with a rotary mower, whereas in late lactation the pasture was cut

with a flail-type forage harvester (which lacerated and bruised the pasture resulting in a more rapid deterioration of the harvested feed).

Cows were weighed before and after the collection period, after a 15 hour fast on the restricted level of feeding, and the weights averaged. Metabolic liveweight was calculated using the exponent 0.75 (Kleiber, 1965) and this was applied to the restricted and ad libitum feeding periods.

3.2.2 Experiment 2 : Indoor feeding experiment including energy balances with non-lactating cows at two stages of pregnancy.

Twelve Friesian cows (six H and six L) approximately four and a half years old were selected, and eight of the twelve cows were accustomed to routines and equipment for excreta collection and calorimetry procedures.

Average breeding indexes and expected calving dates were 127 and August 20th; and 99 and August 17th, for H and L cows respectively.

3.2.2.1 Indoor feeding experiment using twelve cows.

The cows were fed daily at 0800 hours in stalls on fresh pasture (cut with a flail-type harvester) for 62 days, commencing at about 180 days of pregnancy. Six cows within the same genotype were randomly allocated to one of two feeding levels so that there were three cows of each breeding index at each feeding level. The feeding levels were intended to achieve maintenance of body condition and two x maintenance of body condition and were adjusted, according to stage of pregnancy (ARC, 1980).

3.2.2.2 Energy balances using eight cows at two stages of pregnancy during the indoor feeding experiment.

Eight of the twelve cows (two H and two L cows at each level of feeding) were each used for two energy balance periods at approximately 210 and 230 days of pregnancy (Table 3.2).

Table 3.2 Plan of Experiment 2, outlining the chronological sequence (days of experiment) of feeding treatments (estimated to be maintenance (M) and twice maintenance of body condition (2M)) by pairs for Periods 1 and 2 (approximately days 210 and 230 of pregnancy respectively).

		Period 1 (days)	Period 2 (days)
Pair 1	(2M)	1-10	21-30
Pair 2	(M)	6-15	26-35
Pair 3	(2M)	11-20	31-40
Pair 4	(M)	16-25	36-45

In order to measure liveweight changes which were unbiased by gut-fill over the stall feeding period, the twelve cows were initially grazed as one group (estimated intake 4-5kg pasture DM cow⁻¹ day⁻¹) to standardise gut-fill, followed by a 68 hour fast (with access to water).

For the final five days of stall feeding, all cows were fed at the maintenance level of feeding (to standardise gut-fill) before a further 68 hour fast. Liveweight changes were then calculated as the difference in weight (after a 68 hour fast) at the start and end of the stall feeding.

Metabolic liveweight, for use in energy balance calculations, was assessed by interpolating between the measured 44 hour fasted weight at the beginning and end of the stall feeding and using the exponent 0.75.

Condition score changes were calculated as the difference in condition score estimated on two consecutive days at the start and end of the experiment by three observers scoring independently.

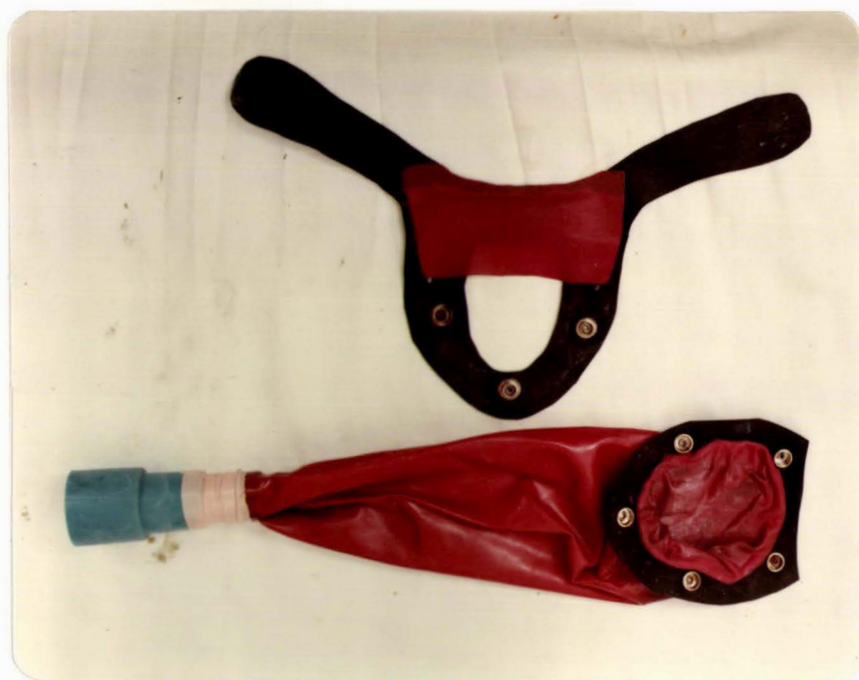
3.2.3 Collection Periods

Dry matter concentrations were measured in triplicate for feed offered to each pair of cows at each feeding and daily for feed refused by each cow. In addition, samples of feed offered and refused were bulked over the ten day collection periods and stored at -10degC. These were subsequently thawed and a representative sample taken, freeze-dried and ground for later analysis.

Faeces were collected in a large metal container using a collection harness (Figure 3.1) and urine in a plastic bucket located in a sump directly behind the cows. The urine harness is shown in Figure 3.1.

Faeces and urine were removed and weighed at 0800 hours daily, each was thoroughly mixed and an aliquot (3%) was taken, bulked over the ten day collection period and stored at -10degC in a plastic bucket with an airtight lid.

Later the bulked faeces and urine were thawed, well mixed and samples taken for nitrogen analysis, freeze-drying and subsequent analysis of energy concentration. For faeces, dry matter determinations were carried out in duplicate.



← Part A

← Part B

The urine harness developed by Mr. J.W. Hughes of Ruakura Agricultural Research Centre. Part A is glued to the cow and Part B is attached to Part A with press studs.



Cow harnessed for total collection and measurement of respiratory gas volumes.

FIG 3.1: The urine and faecal harness used for total collection of excreta (see also FIG 3.2).

Proportional aliquots of the milk were taken at each milking, bulked over five days, stored at 2degC and subsequently analysed for nitrogen and gross energy, In addition, two-day composite samples (5 composites for each collection period) were analysed for milk fat (Milko-tester MkIII, Foss Electric, Denmark), and protein concentration (Pro-milk MkII, Foss Electric, Denmark).

Finally, in early lactation only, the milk fat was extracted from at least two samples per cow for analysis of its fatty acid composition. Changes in the proportion (% by weight) of C₆₋₁₄ (C₆, C₈, C₁₀, C₁₂ and C₁₄) and C_{18:0-1} (C_{18:0} and C_{18:1}) give an indication of the changes in the amounts of milk fat production arising from dietary and body tissue sources respectively (Stobbs and Brett, 1974).

Somatic cell count (cells/ml) was measured twice for each cow during each balance period (Fossomatic, Foss Electric, Denmark).

3.2.4 Chemical Methods

All chemical analyses were done in duplicate, with discrepancies greater than 3% requiring a repeat analysis.

Nitrogen

The nitrogen content of feed, excreta, and milk, was determined by the macro-kjeldahl method (AOAC, 1965).

For feed and faeces about 1g of freeze-dried, ground material was weighed into a polythene bag, which had zero nitrogen content, and analysed for N. For urine and milk, weighed 10g wet samples were pipetted into small plastic bags and analysed for N. The accuracy of the method was assessed by determining the nitrogen content of L-lysine hydrochloride (15.34% N). The average of three estimations was 2.3% lower than the theoretical value, the range was from 1.6 to 2.9% lower.

Energy

Gross energy concentrations of feed, excreta, and milk were determined with an adiabatic bomb calorimeter (Gallenkamp and Co., U.K.).

Freeze-dried, ground samples of feed and faeces were made into pellets (approximately 1g) and gross energy determined. Milk samples were prepared by pipetting 3g milk onto weighed polythene film followed by freezing and freeze-drying. Urine samples were prepared by pipetting 50g of urine into a petri-dish, freezing the sample and then placing it on a weighed polythene film before freeze-drying. Gross energy determinations were carried out on the freeze-dried samples of urine and milk, corrections being made for the energy content of the polythene.

The accuracy of the bomb calorimeter was tested by determining the gross energy content of benzoic acid. The average of nine separate determinations was within 0.09% of the theoretical value, the range being from 2.3% below to 1.3% above the theoretical gross energy content.

Analysis of fatty acid composition of the milk fat

For extraction of the milk fat, samples of milk (from the two day composites used for fat and protein analysis) and sulphuric acid * (18ml of each) were placed into Babcock test bottles mixed by rotary action then spun in a centrifuge for five minutes.

* The sulphuric acid was made by adding 453g sulphuric acid (density 1.84) to 47 ml water.

Hot water was added to bring fat up to the neck of the bottle and the sample centrifuged for a further two minutes. The fat was siphoned off with a disposable pipette and the samples stored in glass bottles at -10degC.

Analysis of fatty acids by gas liquid chromatography

Fatty acids were analysed as their methyl esters using a Varian Aerograph 1200 Gas Chromatograph by a procedure similar to that used by Morrison (1976).

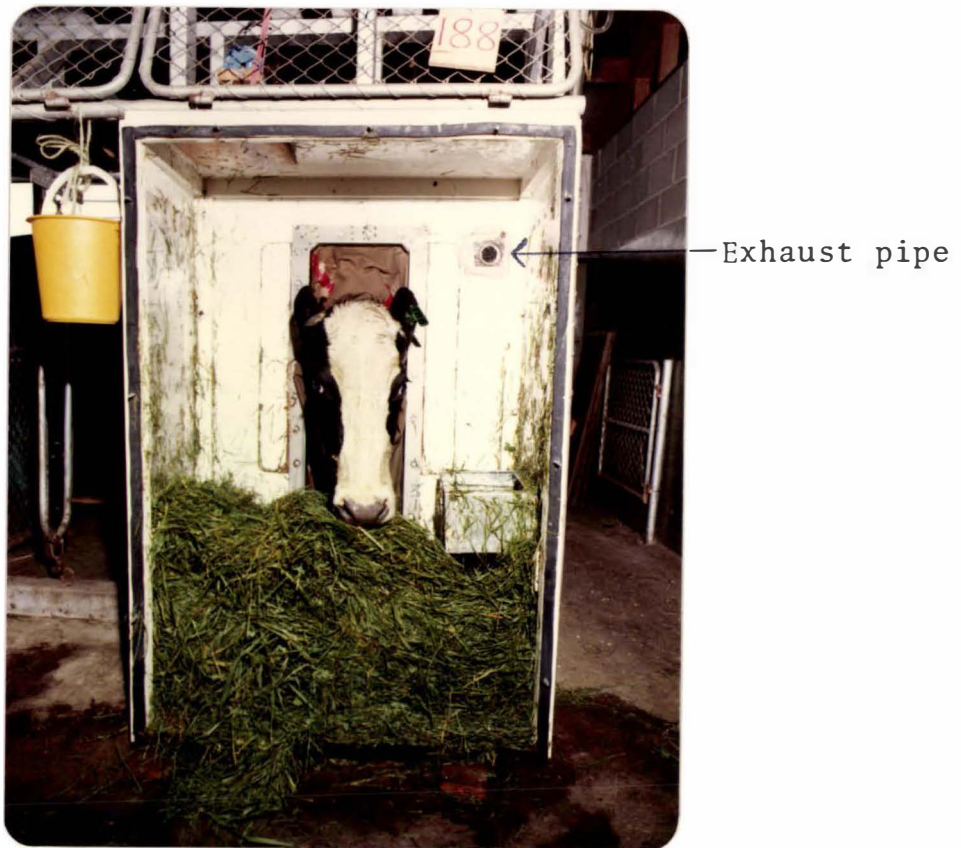
Proportions of the individual fatty acids were obtained by using a Varian Aerograph Digital Integrator 480. The full procedure for analysis of the fatty acids is contained in Appendix 3.1.

3.2.5 The Calorimeters and their Operation

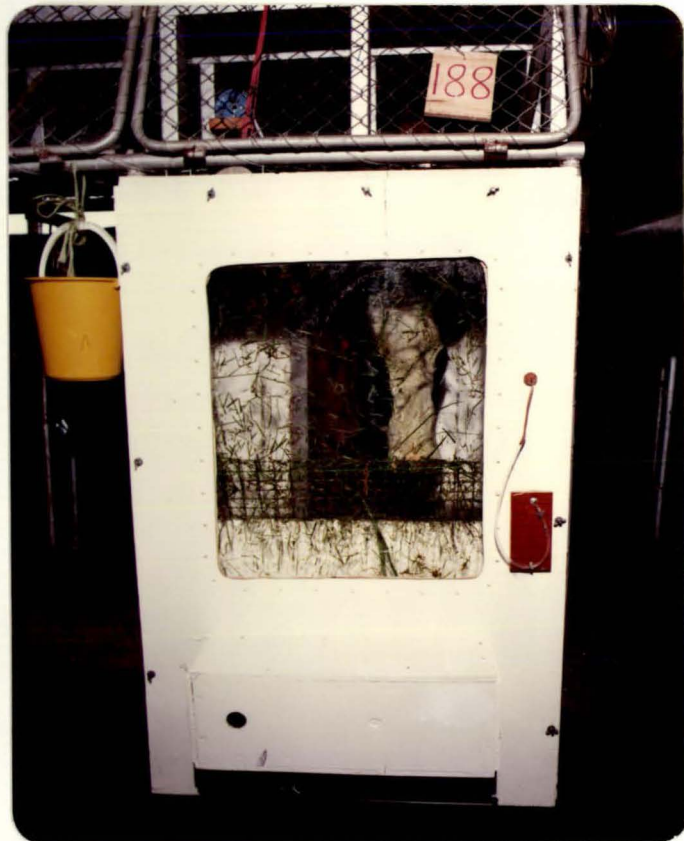
Measurements of respiratory gas volumes were made in two ventilated hoods in an open-circuit system which was similar to that described by Young et al. (1975). The hoods were constructed of marine plywood and had clear perspex panels in the front and on both sides. The dimensions of the hoods were

0.60m deep x 1.00m wide x 1.49m high (Figure 3.2).

A seal was made around the cow's neck using a close fitting flexible vinyl collar (attached to the hood) held firmly in position by elastic rubber cords.



(a) Hood with front removed for feeding the cow.



Air buffer showing
air inlet hole

(b) Hood with front in place during calorimetric measurements.

FIG 3.2: Hood used in calorimetry experiments
(see also FIG 3.1).

The cows were able to stand or lie down comfortably and had free access to water from a trough mounted on the inside of the hood. To feed the cows the front of the hood, which sealed onto a rubber gasket, was removed by undoing twelve wing nuts. Aliquot gas samples were not taken when the cows were being fed, which involved a total loss of approximately 30 minutes of gas sampling time per day.

Air was drawn into the hood through a hole in the air buffer (an arrangement to prevent outward movement of air through the inlet) at a controlled rate, between 250 and 500 litre/min depending on the level of feeding. This ensured that the exhaust air contained concentrations of oxygen, carbon dioxide and methane which were approximately 19.3 to 20.0, 0.7 to 1.2 and 0.05 to 0.16% respectively.

The exhaust air was cooled to between 0deg and 5degC in order to remove water vapour and the air was assumed to be saturated with water vapour at this temperature. The air was re-warmed to 28degC and drawn through dry gas meters. The air temperature was measured as it left the cooling device and again as it left the gas meters. Estimates of water vapour pressure together with barometric pressure readings made it possible to correct the measured volume of air to standard temperature and pressure, dry gas.

Aliquot samples of air were drawn into three spirometers fitted with mercury - o - ring sealed pistons; one for exhaust air from each hood and another for atmospheric air drawn from above the hoods.

The dried samples (using silica gel) were pumped through an automatic infra-red carbon dioxide and methane analyser (Grubb-Parson, U.K.; range 0-1.5% CO₂ and 0-0.2% CH₄) and an automatic paramagnetic oxygen analyser (Servomex, Co., Ltd., U.K.; range 19-21% O₂) connected in series. The outputs of these analysers were connected to a potentiometric chart recorder.

The gas analysers and the recorder were calibrated daily by pumping through them two compressed gas mixtures (containing O₂, CO₂, and CH₄) of known composition; the oxygen, carbon dioxide, and methane content of these gases were 19.308 and 20.798% O₂, and 1.177 and 0.669% CO₂, and 0.152 and 0.056% CH₄.

The reference gases had been calibrated at Ruakura Agricultural Research Centre against gas mixtures prepared by mixing pure gases (New Zealand Industrial Gases, Wellington, New Zealand) using precision gas mixing pumps connected in series (types M300 and NA27, Wosthoff, Bochum, W. Germany).

Heat production was calculated from the equation of Brouwer (1965):-

$$\text{HP} = (\text{O}_2 \times 16.18) + (\text{CO}_2 \times 5.02) - (\text{N} \times 5.99) - (\text{CH}_4 \times 2.17)$$

where HP = heat produced (KJ/24h)
 O₂ = oxygen consumed (ℓ/24h, STP)
 CO₂ = carbon dioxide produced (ℓ/24h, STP)
 N = urinary nitrogen excreted, averaged over the ten day collection period (gN/24h)
 CH₄ = methane produced, (ℓ/24h, STP)

The calorimetric procedures used did not include the carbon dioxide and methane excreted via the anus or in the urine, but neglect of these quantities could only affect heat production to 3 per cent (Murray et al. 1976; Graham, N. McC., personal communication). See Appendix 3.2 for calculation of heat production from raw data.

A systematic error (see Ekern et al., 1965) was incorporated in the estimate of heat production of the cows due to respiration of the pasture in the hood. There must have been some error for all cows, but greater for cows with residual feed on the ad libitum level of feeding. This would probably not affect the main comparison between breeding index groups, but would affect the absolute levels of heat production and hence the estimate of energy retention in the body and also any comparisons with other published calorimetry data. Corrections were therefore made by determining separately the oxygen consumption of different weighed quantities of grass placed in the hoods and adjusting the estimated volumes of O_2 and CO_2 respired by individual cows for the volumes of O_2 and CO_2 respired by the average quantity of grass refused by the cow over the five day gas measurement period. Corrections were small (1-3%) in early lactation with the mown grass, but much larger (18-25%) in late lactation with the flail-harvested grass. No corrections to estimated heat productions were made for Experiment 2 or when cows were on restricted levels of intake in Experiment 1 since there were no feed refusals left in hoods because feed was eaten within hours of being offered.

Test of the Calorimeters

The whole calorimetric system comprising the hoods, ventilation circuit, dry gas meter, gas analysers, recorder and calibration gases were tested by admitting dry nitrogen gas into the hood at rates determined volumetrically by a wet gas meter. The calorimetric equipment was then used to estimate the gas admission rate. In a series of seven tests, the measurements averaged 0.991 accuracy, ranging from 0.954 to 1.033 accuracy.

3.2.6 Statistical Analysis

Experiment 1

The data are multivariate in two senses. Firstly, measurements have been made on several variables (faeces, urine, methane, heat, and milk), and secondly, the variables have been measured at different times on individual animals. For example, with the energy balance data, the input of feed energy must be equal to the combined output of energy in faeces, urine, methane, heat, milk, and tissue, with tissue energy being calculated as the residual or difference energy. Correlations between errors (observed minus predicted values) associated with the various energy outputs are therefore expected. Hence a cow with a greater than expected output of energy in faeces will have a less than expected output of energy elsewhere in the system (eg, urine). Ignoring these correlations if they are present, would invalidate any tests of significance performed (Kramer, 1978).

In the present experiment there were insufficient cows to analyse both 'times' and energy outputs multivariately, hence only energy outputs have been analysed multivariately. The analysis of the repeated measurements over time is similar to covariance analysis in split-plot experiments (Snedecor and Cochran, 1967). Cows are 'main plots', and main plot treatments (high and low breeding index), are allocated to cows. Within cows (main plot) sub-plot treatments (levels of feeding; ad libitum, 70% ad libitum) are located.

In early lactation, the analysis was done in two parts, because it was considered that the physiological response of the cow was likely to be different depending on whether the feeding sequence was ad libitum, followed by 70% ad libitum, or 70% ad libitum followed by ad libitum.

Firstly the ad libitum, 70% ad libitum sequence was analysed and then the 70% ad libitum, ad libitum sequence. It is acknowledged that the 70% ad libitum feeding period was common to both analyses.

The analysis was done in two parts:-

Step 1 : Sub-plot analysis testing differences between genotypes in the slope of the line joining the two levels of feeding.

The measured energy outputs (faeces, urine, methane, heat, and milk) were analysed multivariately. The residual (tissue energy) was analysed separately as a univariate case, since if it were included in the multivariate analysis, all of the variation would be explained.

Because of the small number of cows used, it was not possible to do a multivariate analysis on the five energy outputs, hence faecal, urinary, and methane energies were combined into a single variable (FUM).

The errors of the three energy outputs FUM, heat and milk were tested for independence using the model on the following page.

$$y_{ijk} = \alpha_{ij} + \beta_i x_{ijk} + e_{ijk}$$

where

- y = energy output in FUM, heat or milk
 (MJkg^{-0.75} cow⁻¹ day⁻¹)
 x = gross energy intake (MJkg^{-0.75} cow⁻¹ day⁻¹)
 i = genetic level (H,L)
 j = jth cow
 k = observation on jth cow (k=1,2)
 e = sub-plot (within-cow) error (σ_e^2)

If the errors are independently distributed then the difference in slope between genotypes for the three energy outputs are more appropriately tested univariately. Otherwise valid tests of significance are obtained using the multivariate test of significance. If there were differences in slopes between genotypes, intercepts (α_j 's) were predicted for each cow for different values of x in the range of the data studied. If there were no differences in slopes then a common slope was fitted and intercepts (α_j 's) predicted.

Step 2 : Main plot analysis testing differences in intercepts between BI groups.

Between cow differences are due to breeding index and error. The appropriate error for testing the significance of differences in BI groups is obtained from between cows within BI groups, the model being:-

$$\alpha_{ij} = \mu + \lambda_i + \epsilon_{ij}$$

where α = intercept

μ = mean

i = genetic level (H,L)

j = jth cow

ϵ = main plot (between-cow) error ($k\sigma_c^2 + \sigma_e^2$)

The independence of the errors was tested in a multivariate analysis. If the errors were correlated then the multivariate test of significance is the correct test, but if the errors are independently distributed, univariate tests are more appropriate.

A further point concerns the use of metabolisable energy intake as the independent variable. Because metabolisable energy is estimated by difference (gross energy intake minus faecal, urinary, and methane energies) then it contains measurement errors related to the measurement of faecal, urinary, and methane energy. Use of observed metabolisable energy intake values in regression analysis would give biased estimates of regression co-efficients - this is the problem of "errors in the variables" (Johnston, 1963). Where the variable of interest can be estimated in terms of controlled variables, the method of two-stage least squares can be used to obtain consistent estimates of regression co-efficients in the original model. Metabolisable energy intake was therefore predicted as gross energy intake minus predicted values for faecal, urinary and methane energy (obtained from the regressions with gross energy intake as the independent variable).

Experiment 2

The analysis of the calorimetry (energy and nitrogen) data for the dry, pregnant cows was simplified because there were no repeated measures on the same cow during each time period studied (approximately day 210 and 230 of pregnancy), however the multivariate nature of the data remained.

The model was:-

$$Y_{ij} = \mu + \alpha_i + \beta_i x_{ij} + e_{ij}$$

for jth cow in ith group

where

i = genetic level; H or L

j = 1...6 cows

μ = intercept for L cows

α_L = 0

α_H = difference in intercept between genotypes

y = energy output in faeces, urine, methane, or heat (MJ kg^{-0.75} cow⁻¹ day⁻¹)

x = gross energy intake (MJ kg^{-0.75} cow⁻¹ day⁻¹)

e = between cow error

A stepwise regression procedure was used to test differences in slopes between BI groups, then differences in intercepts.

Other data were analysed by simple linear regression.

Actual significance levels are presented in the results and unless otherwise stated a critical significance level of 5% has been taken.

3.3 RESULTS

3.3.1 Experiment 1

3.3.1.1 Animal Health

No animal health problems were observed during the experimental periods or during the remainder of lactation. There was no evidence of mastitis as indicated by cell counts, which in early lactation were

<100,000 counts/ml

and in late lactation were

<400,000 counts/ml.

The higher count in late lactation was expected, as the counts/ml normally increase in late lactation (Natzke et al., 1972).

One of the L cows was excluded from the late lactation analysis as she had failed to conceive.

3.3.1.2 Lactation Performance

The H cows produced 46 and 30% more milk fat than the L cows during 1979/80 and 1980/81 (the current lactation period) respectively (Table 3.3). The average difference between all the H and L cows, i.e. including those not in the present experiments, was 28 and 18% for the two lactations.

Over the two lactations, L cows gained or maintained condition in contrast to the H cows which lost condition in both years (Table 3.3).

Table 3.3 Milk fat yields, lactation length, condition score at calving and condition score change over two lactations (1979-80 and 1980-81) for high (H) and low (L) breeding index cows.

	1979-80		1980-81	
	H	L	H	L
No. Cows	3	3	3	3
Fat yield (kg)	155	106	172	132
Average lactation length (days)	253		228	
Condition score at calving	4.8	4.7	4.7	4.9
Condition score change over lactation	-0.8	+0.2	-0.4	0.0

3.3.1.3 Performance of Cows during Collection Periods

The dry matter intakes, milk production and composition, and metabolic live weight ($\text{kg}^{0.75}$) for collection periods in early and late lactation are presented in Table 3.4.

The H cows consistently produced more milk fat than L cows and this was associated only with an increase in the concentration of milk fat in the milk since milk yields were similar for both genotypes.

Table 3.4 Daily dry matter intakes (DMI), milk production and composition and metabolic liveweight ($\text{kg}^{0.75}$) for collection periods in early and late lactation for high (H) and low (L) breeding index cows.

	Milk yield kg cow^{-1} day	Fat yield g cow^{-1} day	Protein yield g cow^{-1} day	Milkfat %	Milk protein %	DMI kg DM^{-1} cow^{-1} day	$\text{kg}^{0.75}$
<u>Early Lactation</u>							
<u>Ad libitum</u>							
H	22.0	1097	761	5.00	3.47	14.7	
L	22.2	967	715	4.35	3.23	13.9	
<u>Restricted</u>							
H	16.0	782	505	4.89	3.16	10.2	85.5
L	15.9	751	471	4.73	2.97	9.9	94.6
<u>Ad Libitum</u>							
H	19.3	941	657	4.88	3.41	17.0	
L	19.8	806	617	4.09	3.13	15.6	
<u>Late Lactation</u>							
<u>Ad Libitum</u>							
H	9.2	511	354	5.57	3.83	15.1	
L	8.4	364	279	4.49	3.43	14.0	
<u>Restricted</u>							
H	6.4	420	265	6.59	4.16	10.4	91.4
L	6.2	286	225	4.81	3.82	9.6	101.3

Energy Balance

Treatment mean values for measured variables, namely gross energy intake, faecal, urinary, methane, heat, and milk energies for the five collection periods during lactation are presented in Appendix 3.3.1.

The gross energy intakes of the H cows during ad libitum feeding periods were significantly higher than the L cows in two out of three periods, varying from 15 to 28% higher (Table 3.5). These differences in intake between genotypes were large. In previous experiments (see Chapter 2) there was, on average, a difference of only 7%. However the large difference was partly expected because in the previous lactation when the same six cows were fed ad libitum in early lactation (Experiment 1, Chapter 2) the difference in intake was 11%, but only 5% for all of the H and L cows (10 H and 10 L). The metabolisability (metabolisable energy/gross energy) of the feed was 0.64 and 0.54 in early and late lactation respectively.

Table 3.5 Gross energy intakes (GEI) during ad libitum feeding periods for high (H) and low (L) breeding index cows in early and late lactation.

	GEI (MJ kg ^{-0.75} cow ⁻¹ day ⁻¹)			Probability
	H	L	H/L	
<u>Early Lactation</u>				
<u>Ad libitum</u> 1	3.246	2.813	1.15	0.12
<u>Ad libitum</u> 2	3.866	3.080	1.26	< 0.01
<u>Late Lactation</u>				
<u>Ad libitum</u>	3.076	2.411	1.28	0.04

The average milk energy output was consistently higher for the H cows varying from 15-97% higher (Table 3.6).

Table 3.6 Average milk energy output ($\text{MJ kg}^{-0.75}$ cow⁻¹ day⁻¹) for high (H) and low (L) breeding index cows during collection periods in early lactation; ad libitum (AL 1), restricted (R 1), ad libitum (AL 2) feeding and in late lactation; ad libitum (AL) and restricted (R) feeding.

	Early Lactation			Late Lactation	
	AL 1	R 1	AL 2	AL	R
H	.822	.605	.765	.380	.288
L	.661	.527	.622	.202	.146
H-L	.161	.078	.143	.178	.142
H/L	1.24	1.15	1.23	1.88	1.97

Step 1

Sub-plot analysis testing differences between genotypes in the slope of the line joining the two feeding levels.

A Gross energy intake as the independent variable.

A detailed example of the statistical analysis is outlined in Appendix 3.4.

(i) Multivariate Analysis

The variables included in the analysis were FUM (faeces, urine, and methane), heat and milk produced. The probability for tests of correlations between errors were:

			Probability
Early lactation	(A → R)	χ^2 3d.f.	0.94
Early lactation	(R → A)	χ^2 3d.f.	0.67
Late lactation	(A → R)	χ^2 3d.f.	0.85
† (A→R) <u>ad libitum</u> followed by restricted (70% <u>ad libitum</u>) feeding			
⊕ (R→A) restricted (70% <u>ad libitum</u>) followed by <u>ad libitum</u> feeding			

(ii) Univariate Analysis

Because the errors were independently distributed univariate analyses were more appropriate. There were no significant differences in slopes between BI groups for any of the dependent variables. The probabilities for differences in slopes between BI groups were:-

	Early lactation (A → R)	Early lactation (R → A)	Late lactation (R → A)
FUM*	0.16	0.96	0.91
Heat	0.06	0.10	0.89
Milk	0.68	0.62	0.55
Tissue	0.14	0.29	0.86
Energy retained as protein	0.11	0.87	0.41
Energy retained as fat	0.44	0.53	0.85

As there were no significant differences in slopes, common slopes were fitted and intercepts (α_j 's) predicted.

In early lactation (A→R) the probability for differences in slopes between BI groups for heat energy approached significance ($P = 0.06$). The data were further examined

*The Statistical analyses of the single components of FUM (faeces, urine and methane) are presented in Appendix 3.7.

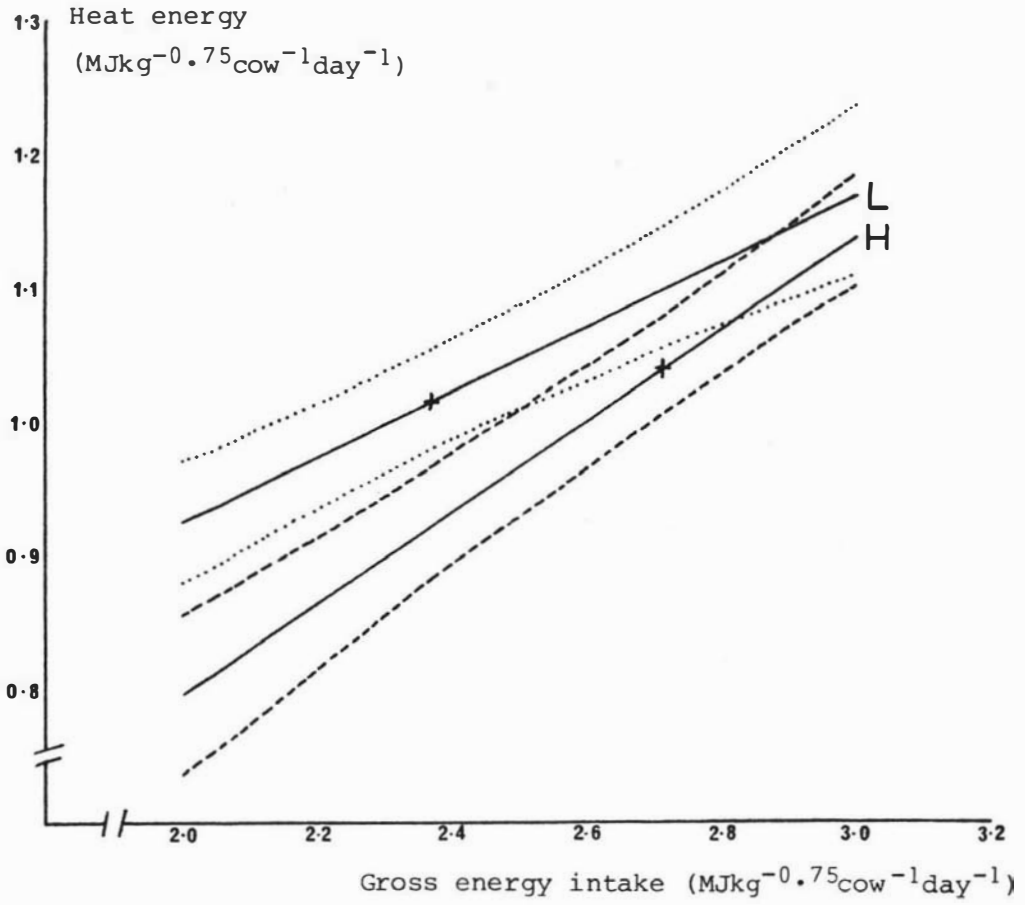


FIG.3.3: Heat energy versus gross energy intake (GEI) for high (H) and low (L) breeding index cows; regression lines (—) with 95% confidence limits for H (-----) and L (.....) cows.

by fitting a different slope for each BI group with the 95% confidence limits. (Figure 3.3) The confidence limits were calculated using the sub-plot (within-cow) error. From Figure 3.3 it can be seen that at the lower level of intake (approximately 2.0MJ gross energy $\text{kg}^{-0.75} \text{cow}^{-1} \text{day}^{-1}$) the two genotypes were significantly different, but the difference was not large enough to fit different slopes for each group when considering the ad libitum level of intake as well.

B Predicted metabolisable energy intake as the independent variable.

(i) Multivariate analysis

The dependent variables included in the analysis were heat and milk energies. The probabilities for tests of independence of errors were:-

			Probability
Early lactation	(A → R)	χ^2_2 1 d.f.	0.89
Early lactation	(R → A)	χ^2_2 1 d.f.	0.81
Late lactation	(A → R)	χ^2 1 d.f.	0.43

(ii) Univariate analyses were therefore appropriate.

There were no significant differences in slopes between BI groups for any of the dependent variables. The probabilities for differences in slopes between BI groups were:-

	Early lactation (A → R)	Early lactation (R → A)	Late lactation (A → R)
Heat	0.06	0.10	0.89
Milk	0.68	0.62	0.55
Tissue	0.22	0.13	0.79

As there were no significant differences in slopes, common slopes were fitted and intercepts (α_j 's) predicted.

Step 2

Whole plot analysis testing differences in intercepts between BI groups.

A Gross energy intake as the independent variable.

(i) Multivariate analysis

The probabilities for the tests of independence of the errors were:-

				Probability
Early lactation	(A → R)	χ^2	3 d.f.	0.93
Early lactation	(R → A)	χ^2	3 d.f.	0.38
Late lactation	(A → R)	χ	3 d.f.	0.34

(ii) Univariate analyses were therefore more appropriate.

	<u>Early lactation</u> (A → R)			
	Intercept H	H-L	s.e.diff	Prob-ability
FUM	-0.143	0.005	0.018	0.79
Heat	0.209	-0.079	0.040	0.12
Milk	0.249	0.059	0.052	0.32
Tissue	-0.315	0.016	0.072	0.84
Energy retained as protein	0.174	0.004	0.056	0.94
Energy retained as fat	-0.404	-0.005	0.061	0.94

Early lactation (R → A)				
	Intercept H	H-L	s.e.diff	Prob-ability
FUM	-0.084	-0.010	0.020	0.66
Heat	0.280	-0.049	0.027	0.14
Milk	0.409	0.062	0.034	0.14
Tissue	-0.604	-0.004	0.020	0.85
Energy retained as protein	-0.026	-0.007	0.020	0.75
Energy retained as fat	-0.536	-0.019	0.028	0.53

Late lactation (A → R)				
	Intercept H	H-L	s.e.diff	Prob-ability
FUM	0.103	0.009	0.042	0.84
Heat	0.379	0.009	0.038	0.83
Milk	0.099	0.107	0.019	0.01***
Tissue	-0.581	-0.125	0.062	0.14
Energy retained as protein	0.063	0.049	0.052	0.41
Energy retained as fat	-0.527	-0.083	0.055	0.24

In late lactation the H cows retained significantly more milk energy at a common energy intake than the L cows, otherwise differences in intercepts between groups were not significant.

B Predicted metabolisable energy as the independent variable.

(i) Multivariate analysis

The probabilities for the tests of independence of the errors were:-

			Probability
Early lactation	(A → R) χ^2_2	1 d.f.	0.76
Early lactation	(R → A) χ^2_2	1 d.f.	0.81
Late lactation	(A → R) χ^2	1 d.f.	0.45

(ii) Univariate analyses were therefore more appropriate.

	Early lactation (A → R)		s.e.diff	Prob-ability
	Intercept H	H-L		
Heat	0.133	-0.077	0.043	0.15
Milk	0.207	0.060	0.054	0.33
Tissue	-0.340	0.016	0.070	0.83

	Early lactation (R → A)		s.e.diff	Prob-ability
	Intercept	H-L		
Heat	0.241	-0.053	0.026	0.12
Milk	0.396	0.061	0.033	0.14
Tissue	-0.637	-0.008	0.017	0.68

	Late lactation (A → R)		s.e.diff	Prob-ability
	Intercept	H-L		
Heat	0.415	0.012	0.050	0.83
Milk	0.115	0.108	0.013	< 0.01***
Tissue	-0.530	-0.120	0.044	0.07

Again in late lactation the H cows retained significantly more milk energy at a given energy intake than the L cows, otherwise differences in intercepts were not significant.

Summaries of regression equations with gross energy and metabolisable energy as the independent variables for early lactation (A → R), early lactation (R → A), and late lactation (A → R) are presented in Tables 3.7, 3.8, and 3.9. Heat, milk and tissue energies with predicted metabolisable energy intake as the independent variable are presented graphically for the three periods during lactation, early lactation (A → R), early lactation (R → A), and late lactation (A → R) (Figures 3.4, 3.5, and 3.6).

Table 3.7 Summary of pooled regression equations with gross energy intake (GEI) and metabolisable energy intake (MÊI) as the independent variables for early lactation. (A → R).

X	Y	b(± s.e.)	Intercept
GEI (MJkg ^{-0.75} cow ⁻¹ day ⁻¹)	FUM	0.421 (±0.017)	-0.145
	Heat	0.307 (±0.028)	0.249
	Milk	0.171 (±0.041)	0.220
	Tissue	0.101 (±0.062)	-0.323
	Energy retained as protein	-0.057 (±0.013)	0.172
MÊI ^{^**} (MJkg ^{-0.75} cow ⁻¹ day ⁻¹)	Energy retained as fat	0.127 (±0.045)	-0.401
	Heat	0.530 (±0.048)	0.172
	Milk	0.295 (±0.071)	0.177
	Tissue	0.174 (±0.090)	-0.348
	E _{TOT} [*]	0.470 (±0.048)	-0.172

* Milk plus tissue energy

** "Predicted" metabolisable energy intake (refer statistical methods)

Table 3.8 Summary of pooled regression equations with gross energy intake (GEI) and metabolisable energy intake ($\hat{M}EI$) as the independent variables for early lactation (R \rightarrow A).

X	Y	b^{\pm} s.e.)	Intercept
GEI ($MJkg^{-0.75}$ $cow^{-1}day^{-1}$)	FUM	0.389 (± 0.023)	-0.080
	Heat	0.279 (± 0.030)	0.304
	Milk	0.091 (± 0.014)	0.378
	Tissue	0.241 (± 0.048)	-0.602
	Energy retained as protein	0.037 (± 0.024)	-0.022
	Energy retained as fat	0.190 (± 0.043)	-0.527
$\hat{M}EI$ † ($MJkg^{-0.75}$ $cow^{-1}day^{-1}$)	Heat	0.456 (± 0.050)	0.268
	Milk	0.150 (± 0.023)	0.366
	Tissue	0.394 (± 0.060)	-0.634
	E_{TOT} *	0.544 (± 0.050)	-0.268

* Milk plus tissue energy

† Predicted metabolisable energy intake
(refer statistical methods)

Table 3.9 Summary of regression equations with gross energy intake (GEI) and metabolisable energy intake ($\hat{M}EI$) as the independent variables for late lactation (A + R).

X	Y	b(⁺ s.e.)	Intercept***	
GEI (MJkg ^{-0.75} cow ⁻¹ day ⁻¹)	FUM	0.424 (⁺ 0.040)	0.098	
	Heat	0.199 (⁺ 0.046)	0.375	
	Milk	0.090 (⁺ 0.014)	0.099	H
			-0.008	L
	Tissue	0.287 (⁺ 0.057)	-0.518	
	Energy retained as protein	-0.012 (⁺ 0.044)	0.043	
	Energy retained as fat	0.254 (⁺ 0.041)	-0.494	
$\hat{M}EI$ ** (MJkg ^{-0.75} cow ⁻¹ day ⁻¹)	Heat	0.346 (⁺ 0.081)	0.409	
	Milk	0.157 (⁺ 0.025)	0.115	H
			0.007	L
	Tissue	0.497 (⁺ 0.097)	-0.482	
	E_{TOT} *	0.654 (⁺ 0.081)	-0.409	

* Milk plus tissue energy

** "Predicted" metabolisable energy intake (refer statistical methods)

*** In milk energy produced there was a significant difference in intercept between BI groups.

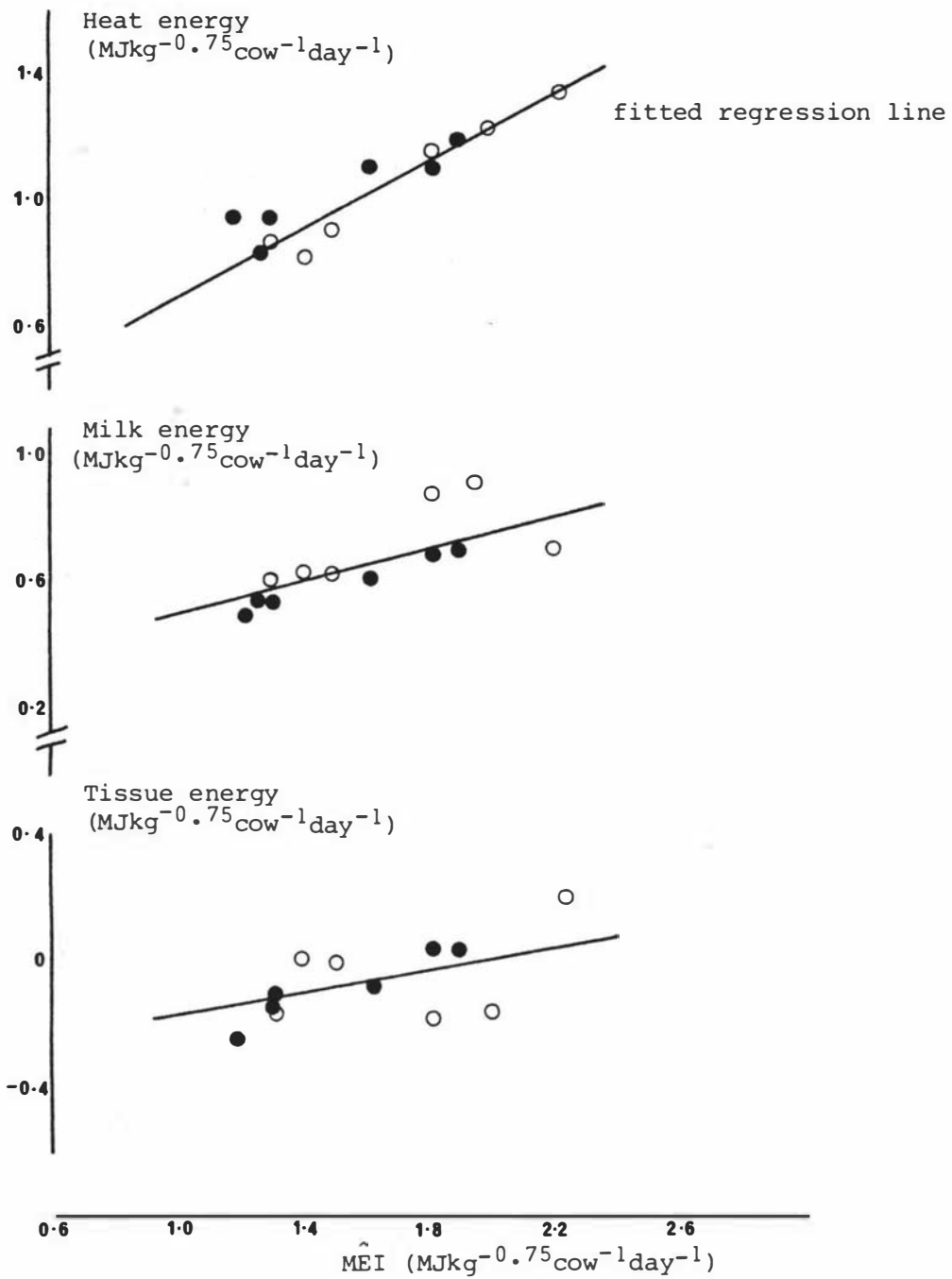


FIG. 3.4: Heat, milk and body tissue energy productions regressed against predicted metabolisable energy intake (\hat{MEI}) for high (○) and low (●) breeding index cows in early lactation when the feeding sequence was ad libitum followed by restricted feeding.

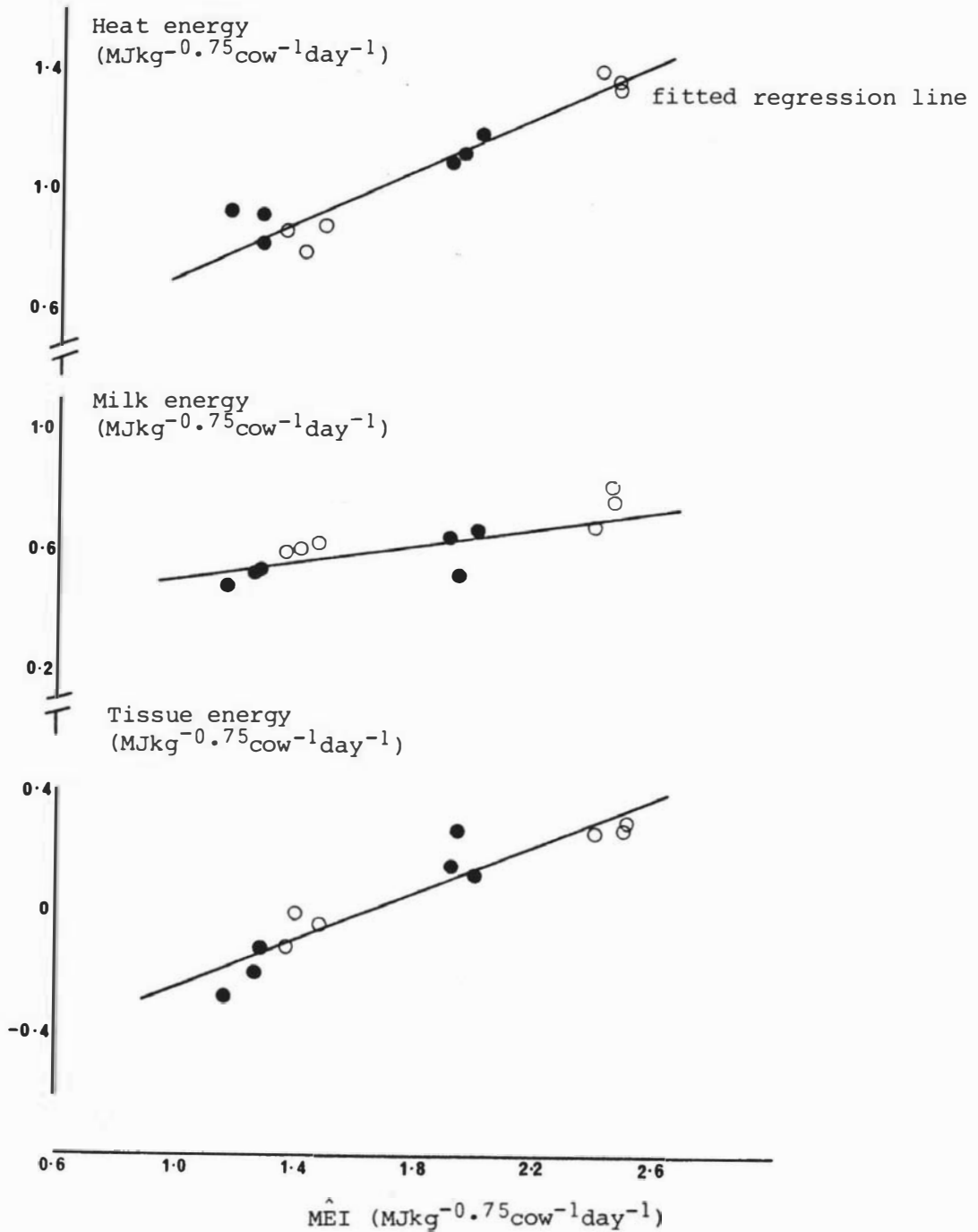


FIG.3.5: Heat, milk and body tissue energy productions regressed against predicted metabolisable energy intake (MEI) for high (○) and low (●) breeding index cows in early lactation when the feeding sequence was restricted followed by ad libitum feeding.

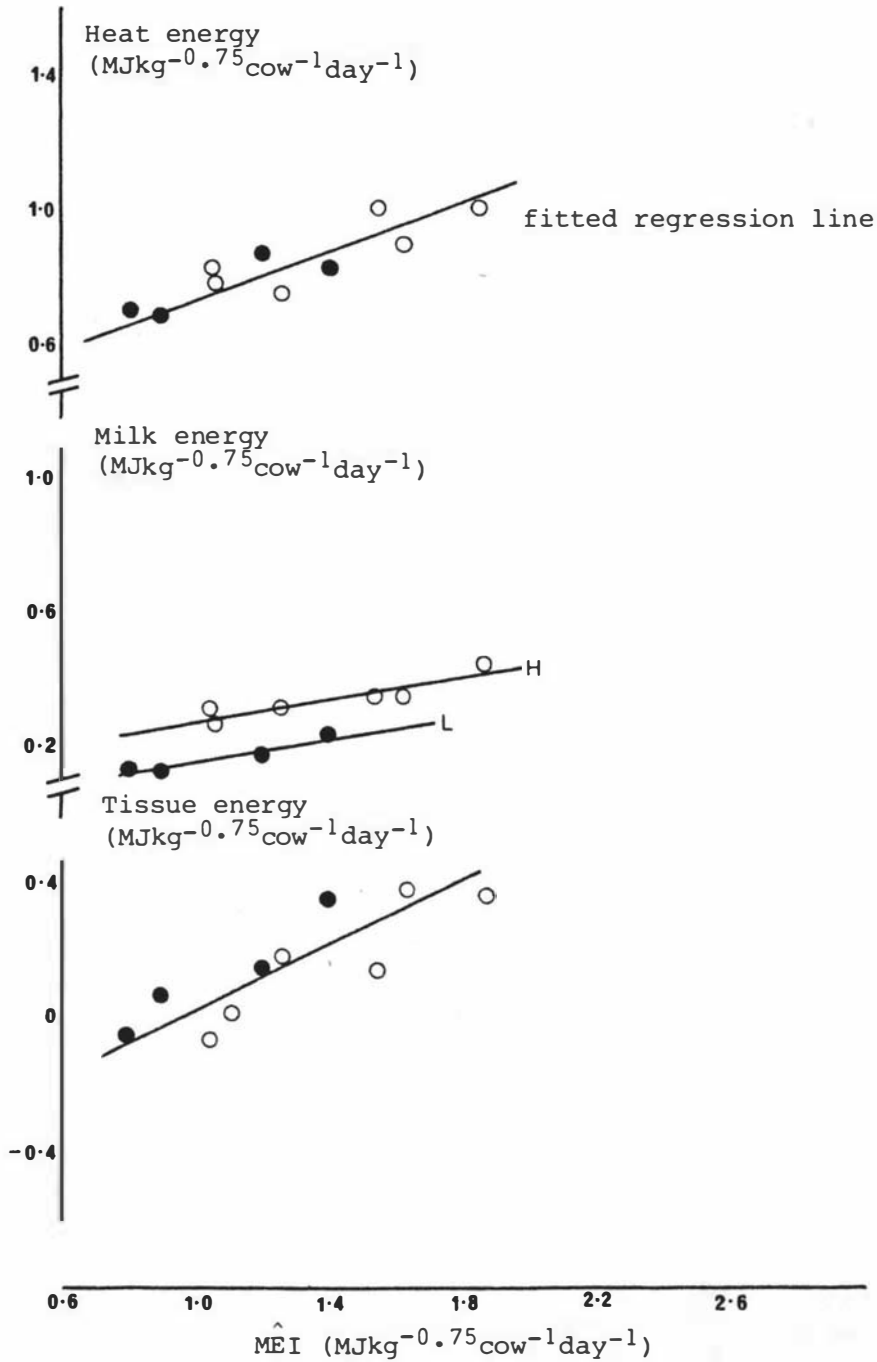


FIG.3.6: Heat milk and body tissue energy productions regressed against predicted metabolisable energy intake (\hat{MEI}) for high (○) and low (●) breeding index cows in late lactation when the feeding sequence was ad libitum followed by restricted feeding.

Nitrogen balance

Step 1 Sub-plot analysis testing differences in slopes between BI groups.

(i) Multivariate analysis

Dependent variables were faecal, urinary, and milk nitrogen with nitrogen intake as the independent variable. Tissue nitrogen (retained or lost) was analysed univariately. The probabilities for the tests of independence of errors were:-

			Probability
Early lactation	(A → R)	χ^2 3 d.f.	0.10
Early lactation	(R → A)	χ^2 3 d.f.	0.56
Late lactation	(A → R)	χ^2 3 d.f.	0.41

For testing the independence of errors a critical significance level of 10% has been taken. Therefore in early lactation (A → R) the errors are not independently distributed and the joint test of the differences in slopes between BI groups is the appropriate test. The joint test was a χ^2 test with 3 d.f. and the probability of the difference in slopes was 0.33, a common slope was therefore fitted and intercepts (α_j 's) predicted.

(ii) Univariate analysis

For early lactation (R → A) and late lactation (A → R) the errors were independently distributed and hence univariate analyses were done. The probabilities for differences in slopes between BI groups were:-

	Early lactation (A → R)	Early lactation (R → A)	Late lactation (A → R)
Faecal N	-	0.62	0.97
Urinary N	-	0.93	0.98
Milk N	-	0.93	0.38
Tissue N	0.32	0.75	0.43

As there were no significant differences in slopes, common slopes were fitted and intercepts (α_j 's) predicted.

Step 2

Whole-plot analysis testing differences in intercepts between BI groups.

(i) Multivariate analysis

The probabilities for the tests of independence of the errors were:-

			Probability
Early lactation	(A → R)	χ^2_3 3 d.f.	0.15
Early lactation	(R → A)	χ^2_3 3 d.f.	0.38
Late lactation	(A → R)	χ^2_3 3 d.f.	0.69

(ii) Univariate analyses were therefore more appropriate.

	<u>Early lactation (A → R)</u>			
	Intercept H	H-L	s.e.diff	Probability
Faecal N	-0.470	-0.011	0.130	0.94
Urinary N	-0.641	-0.020	0.219	0.93
Milk N	-0.073	0.008	0.137	0.96
Tissue N	1.184	0.023	0.444	0.96

Early lactation (R → A)

	Intercept H	H-L	s.e.diff	Prob- ability
Faecal N	-0.367	0.031	0.062	0.64
Urinary N	0.533	0.026	0.147	0.87
Milk N	0.360	0.035	0.055	0.56
Tissue N	-0.526	-0.091	0.128	0.51

Late lactation (A → R)

	Intercept H	H-L	s.e.diff	Prob- ability
Faecal N	-0.190	-0.131	0.115	0.43
Urinary N	-0.348	-0.246	0.200	0.31
Milk N	0.174	0.138	0.023	0.02 *
Tissue N	0.364	0.240	0.342	0.53

In late lactation the cows (H) retained significantly more nitrogen in their milk at a given nitrogen intake, than the L cows, otherwise differences in intercepts between groups were not significant. Summaries of regression equations relating to nitrogen balance are presented in Table 3.10.

Table 3.10 Summary of regression equations with nitrogen intake as the independent variable for early (A → R) and (R → A), and late lactation (A → R).

X	Y	b(⁺ s.e.)	Intercept	
<u>Early lactation (A → R)</u>				
Nitrogen intake (gm N LW ^{-0.75})	Faecal N	0.331 (⁺ 0.035)	-0.465	
	Urinary N	0.654 (⁺ 0.040)	-0.631	
	Milk N	0.219 (⁺ 0.053)	-0.076	
	Tissue N	-0.204 (⁺ 0.078)	1.172	
<u>Early lactation (R → A)</u>				
Nitrogen intake (gm N LW ^{-0.75})	Faecal N	0.314 (⁺ 0.056)	-0.383	
	Urinary N	0.382 (⁺ 0.034)	0.520	
	Milk N	0.111 (⁺ 0.016)	0.343	
	Tissue N	0.192 (⁺ 0.082)	-0.480	
<u>Late lactation (A → R)</u>				
Nitrogen intake (gm N LW ^{-0.75})	Faecal N	0.367 (⁺ 0.083)		
	Urinary N	0.610 (⁺ 0.090)		
	Milk N	0.085 (⁺ 0.015)	0.174	H BI
	Tissue N	-0.062 (⁺ 0.175)	0.036	L BI
			0.268	

Fatty acid composition of the milk

The fatty acid composition of the milk fat during collection periods in early lactation is presented in Table 3.11.

Table 3.11 Percentage by weight of C_{6-14} and $C_{18:0-1}$ fatty acids for high (H) and low (L) breeding index cows at the mean energy intakes during ad libitum, restricted, ad libitum feeding in early lactation.

	<u>Ad libitum</u>		Restricted		<u>Ad libitum</u>	
	H	L	H	L	H	L
C_{6-14} (% by weight)	19.1	19.0	19.9	13.0	21.2	25.7
$C_{18:0-1}$ (% by weight)	42.3	40.3	39.0	49.5	36.9	31.7

When L cows were underfed the proportion of $C_{18:0-1}$ in their milk fat increased and the proportion of C_{6-14} decreased, indicating greater use of body reserves to support milk production during restricted feeding. In contrast, for H cows the proportion of $C_{18:0-1}$ in the milk fat decreased slightly and the proportion of C_{6-14} remained relatively constant, indicating that there was no greater mobilisation of body reserves when

H cows were underfed. These implied changes in body tissue energy based on changes in fatty acid composition of the milk fat are in agreement with the observed changes in body tissue energy as estimated by calorimetric balance (Figure 3.7)

Upon re-feeding the proportion of $C_{18:0-1}$ in the milk fat decreased and the proportion of C_{6-14} increased for both H and L cows with more marked changes in the L cows. This suggests that the relative contribution of precursors from adipose tissue to milk fat synthesis was decreasing and that of dietary sources was increasing in agreement with the tissue changes estimated by calorimetry (Figure 3.7).

3.3.2 Experiment 2

3.3.2.1 Collection periods

(a) Energy balance

Each period was analysed separately, periods 1 and 2 being approximately day 210 and 230 of pregnancy respectively. The metabolisability of the feed was 0.59 and 0.64 for periods 1 and 2 respectively.

Treatment mean values for measured variables, namely gross energy intake, faecal, urinary, methane, and heat energies at the two stages of pregnancy are presented in Appendix 3.3.2.

A Gross energy intake as the independent variable

(i) Multivariate analysis

The variables included in the analysis were faeces, urine, methane and heat. The probabilities for tests of independence of errors were:-

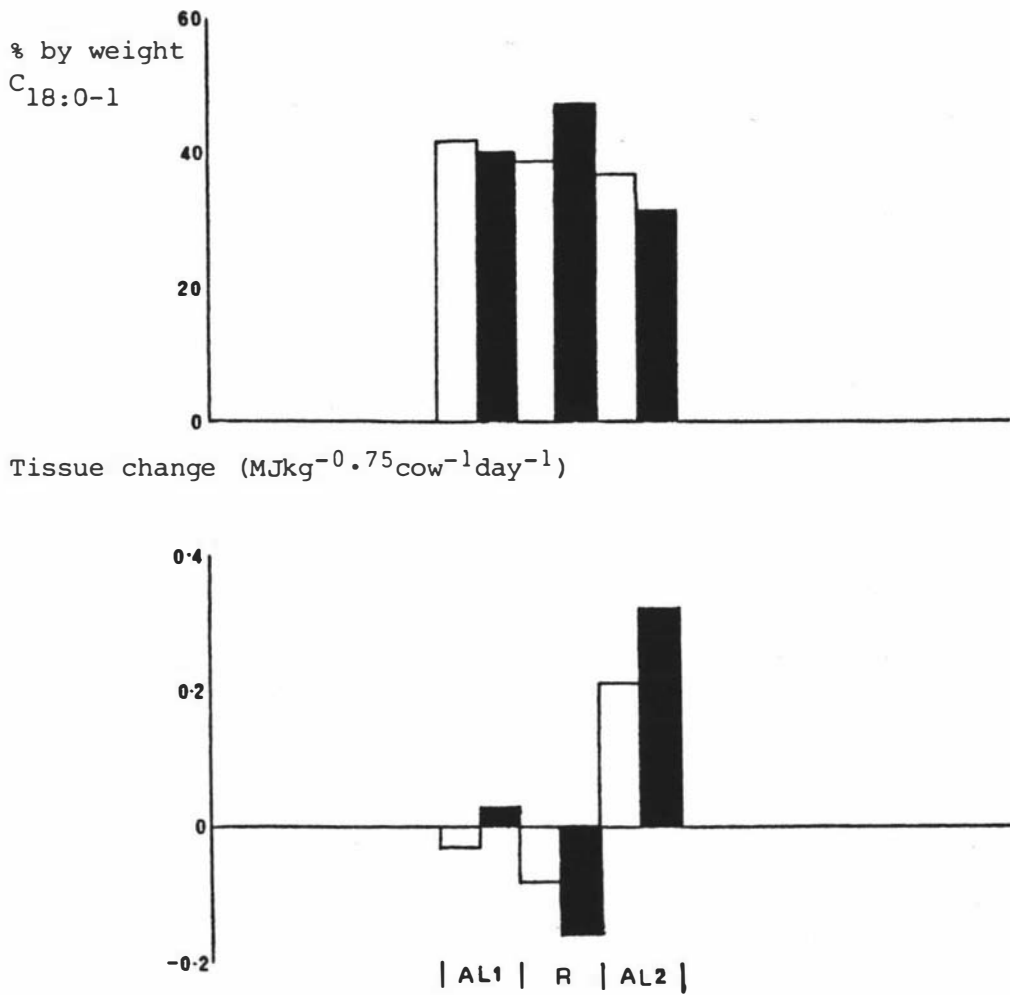


FIG. 3.7: Percentage by weight of C_{18:0-1} fatty acids, and tissue changes for H (□) and L (■) BI cows at the mean energy intakes during collection periods in the sequence ad libitum (AL1), restricted (R), ad libitum (AL2) feeding in early lactation.

			Probability
Period 1	χ^2	6 d.f.	0.02
Period 2	χ^2	6 d.f.	0.07

As the critical significance level was set at $P = 0.10$ then the errors are not independently distributed, hence tests of differences between BI groups in slopes and intercepts must be done jointly for the four energy outputs. The joint tests of differences in slopes between BI groups were:-

			Probability
Period 1	χ^2	4 d.f.	0.06
Period 2	χ^2	4 d.f.	0.10

As there were no significant differences in slopes between BI groups, common slopes were fitted and intercepts tested. The joint test of differences between BI groups in intercepts were:-

			Probability
Period 1	χ^2	4 d.f.	0.42
Period 2	χ^2	4 d.f.	0.65

As there were no differences in slopes or intercepts the simplest model with a common slope and common intercept was fitted.

(ii) Univariate analysis

The probabilities for differences in slopes between BI groups for tissue energy retention and tissue energy retention as fat and protein were:-

	Period 1	Period 2
Tissue energy	0.56	0.31
Energy retained as protein	0.47	0.07
Energy retained as fat	0.53	0.38

As there were no significant differences in slopes, common slopes were fitted. The probabilities for differences in intercepts between BI groups were:-

	Period 1	Period 2
Tissue energy	0.33	0.79
Energy retained as protein	0.70	0.91
Energy retained as fat	0.46	0.78

As there were no significant differences in slopes or intercepts between BI groups, common slopes and intercepts were fitted.

B Predicted metabolisable energy intake as the independent variable.

(i) Univariate analysis

Multivariate analysis was not appropriate in this case because only one output of energy (heat) was measured, the residual energy (tissue) being calculated by difference. The probabilities for testing differences in slopes were:-

	Period 1	Period 2
Heat	0.37	0.28
Tissue	0.37	0.28

As there were no significant differences in slopes, common slopes were fitted and intercepts tested.

Period 1

	Intercept H	H-L	s.e.diff	Prob-ability
Heat	0.412	0.071	0.041	0.15
Tissue	-0.412	-0.071	0.041	0.15

Period 2

	Intercept H	H-L	s.e.diff	Prob- ability
Heat	0.346	-0.002	0.040	0.96
Tissue	-0.346	0.002	0.040	0.96

As there were no significant differences in slopes or intercepts, common slopes and intercepts were fitted for the regression equations.

A summary of regression equations with gross energy and metabolisable energy intake as the independent variables are presented in Table 3.12, for Period 1 and 3.13 for Period 2.

Table 3.12 Summary of pooled regression equations with gross energy intake (GEI) and metabolisable energy intake ($\hat{M}EI$) as the independent variables for Period 1.

X	Y	b(\pm s.e.)	Intercept
GEI (MJkg ^{-0.75} cow ⁻¹ day ⁻¹)	Faeces	0.321 (\pm 0.032)	-0.032
	Urine	0.035 (\pm 0.003)	0.013
	Methane	0.054 (\pm 0.006)	0.015
	Heat	0.285 (\pm 0.052)	0.367
	Tissue	0.305 (\pm 0.068)	-0.363
	Energy retained as protein	0.114 (\pm 0.017)	-0.075
	Energy retained as fat	0.190 (\pm 0.079)	-0.286
	$\hat{M}EI$ (MJkg ^{-0.75} cow ⁻¹ day ⁻¹)	Heat	0.483 (\pm 0.087)
Tissue		0.517 (\pm 0.087)	-0.365

Table 3.13 Summary of pooled regression equations with gross energy intake (GEI) and metabolisable energy intake (MEI) as the independent variables for Period 2.

X	Y	b(⁺ s.e.)	Intercept
GEI (MJkg ^{-0.75} cow ⁻¹ day ⁻¹)	Faeces	0.260 (⁺ 0.055)	-0.009
	Urine	0.043 (⁺ 0.002)	0
	Methane	0.040 (⁺ 0.002)	0.044
	Heat	0.315 (⁺ 0.043)	0.331
	Tissue	0.341 (⁺ 0.095)	-0.366
	Energy retained as protein	0.024 (⁺ 0.015)	0.055
MEI (MJkg ^{-0.75} cow ⁻¹ day ⁻¹)	Energy retained as fat	0.317 (⁺ 0.084)	-0.420
	Heat	0.480 (⁺ 0.065)	0.348
	Tissue	0.520 (⁺ 0.065)	-0.348

Data relating to metabolisable energy intake and tissue energy retained for periods 1 and 2 is shown graphically (Figure 3.8)

(b) Nitrogen balance

(i) Multivariate analysis

Dependent variables were faecal, and urinary nitrogen with nitrogen intake as the independent variable.

The probabilities for the tests of independence of errors were:-

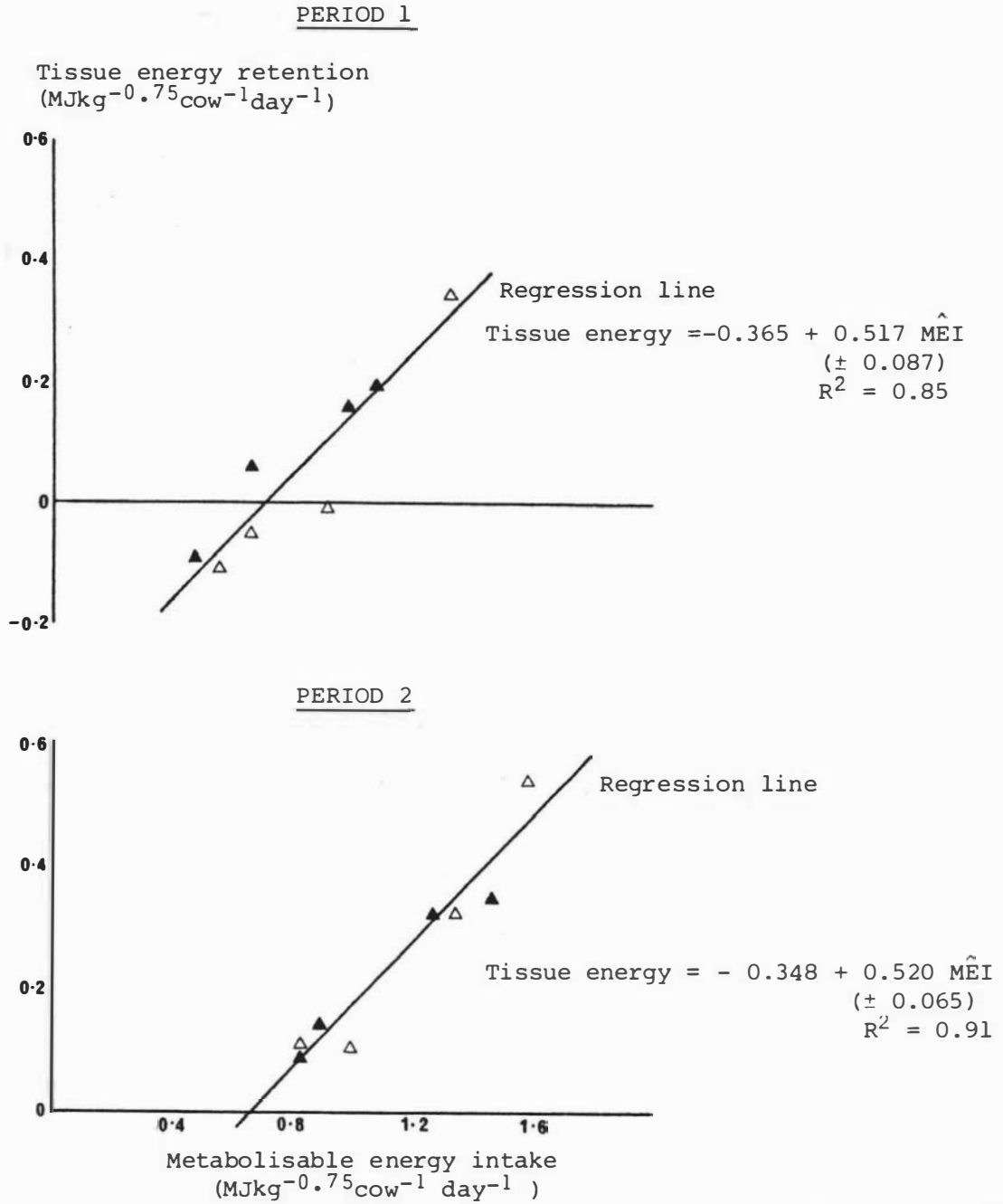


FIG. 3.8: Tissue energy retention versus metabolisable energy intake ($\hat{\text{MEI}}$) for H (Δ) and L (\blacktriangle) BI cows at approximately 210 (Period 1) and 230 (Period 2) days of pregnancy.

			Probability
Period 1	χ^2	1 d.f.	0.32
Period 2	χ^2	1 d.f.	0.02

As the errors were independently distributed in Period 1, univariate analyses were more appropriate, but in Period 2 the errors were correlated, hence joint tests of significance were done.

Period 2

The joint test of difference in slopes between BI groups for faecal and urinary N was χ^2 2 d.f. $P = 0.02$. Hence different slopes were fitted for each BI group for faecal and urinary N.

(ii) Univariate analysis

Period 1

The probabilities for the differences in slopes between BI groups were:-

	Probability
Faecal N	0.11
Urinary N	0.70
Tissue N	0.99

As there were no significant differences in slopes, common slopes were fitted and differences in intercepts tested, and the probabilities were:-

	Probability
Faecal N	0.59
Urinary N	0.81
Tissue N	0.65

As there were no significant differences in slopes or intercepts, common slopes and intercepts were fitted.

Period 2

Differences in slope between BI groups for tissue N were significant ($P = 0.04$), hence different slopes for each BI group were fitted.

In Period 2 at low N intakes ($2.7-3.5\text{gm Nkg}^{-0.75}$) H cows lost more N in urine and faeces and retained less in tissue than L cows, the reverse being the case at higher N intakes ($3.5-5.0\text{gm N kg}^{-0.75}$). This effect was not consistent with Period 1 when there was no difference between BI groups in partitioning of N.

A summary of regression equations relating to N balance in Periods 1 and 2 is given in Table 3.14.

Table 3.14 Summary of regression equations with nitrogen intake as the independent variable for periods 1 and 2 for high (H) and low (L) breeding index cows.

X	Y	b(⁺ s.e.)	Intercept	
	<u>Period 1</u>			
Nitrogen intake ($\text{gm N kg}^{-0.75}$ $\text{cow}^{-1} \text{day}^{-1}$)	Faecal N	0.267 (⁺ 0.016)	-0.115	
	Urine N	0.381 (⁺ 0.054)	0.698	
	Tissue N	0.353 (⁺ 0.048)	-0.583	
	<u>Period 2</u>			
	Faecal N	0.230 (⁺ 0.084)	0.045	H
		0.319 (⁺ 0.108)	-0.312	L
	Urine N	0.579 (⁺ 0.046)	0.002	H
		0.705 (⁺ 0.059)	-0.423	L
	Tissue N	0.190 (⁺ 0.043)	-0.005	H
		-0.024 (⁺ 0.055)	0.735	L

3.3.2.2 Indoor feeding experiment

As there were no significant differences between genotypes in energy balance, the data for the twelve cows over the 62 day indoor feeding period have been pooled across BI groups.

Dry matter intakes, liveweight, and condition score changes and average liveweight over the indoor feeding period are given in Table 3.15.

Table 3.15 Dry matter intakes, liveweight and condition score changes and average liveweight over the indoor feeding period (180-242 days of pregnancy) for cows fed at two levels; maintenance of body condition (M) and twice maintenance of body condition (2M).

Item	M	2M
No. cows	6	6
Dry matter intake (kg DM cow ⁻¹ day ⁻¹)	5.84	9.73
Condition score (start)	4.14	4.22
Condition score (end)	4.03	5.14
Change in condition score (62 Days)	-0.11	0.92
Liveweight * (start) (kg)	415	414
Liveweight * (end) (kg)	414	460
Change in liveweight * (62 days)	-1	+46
Average liveweight ** (kg) (during experiment)	426	447

* Liveweights are 68 hour fasted weights

**Liveweights are 44 hour fasted weights

The relation between change in liveweight (based on 68 hour fasted weights) and change in condition score over the 62 day indoor feeding period was:-

$$\text{DLW} = 5.3 + 43.8(^{\pm}7.6) \text{DCS} \quad R^2 = 0.77$$

where DCS = change in condition score
 DLW = change in liveweight (kg)

A change in one condition unit corresponded to an increase in liveweight of 43.8kg.

The relation between estimated metabolisable energy intake and change in condition score over the indoor feeding period (expressed per unit metabolic liveweight) was:-

$$\Delta\text{CSkg}^{-0.75} = -2.050 + 2.711(^{\pm}0.294) \text{MEIkg}^{-0.75} \quad R^2 = 0.89$$

where $\Delta\text{CSkg}^{-0.75}$ = change in condition score per (unit metabolic liveweight $\times 10^{-2}$) over the indoor feeding period
 $\text{MEIkg}^{-0.75}$ = metabolisable energy intake * per unit metabolic liveweight day^{-1} .

From this relationship it can be calculated that to improve the condition score of a 400kg cow by one unit, during the period 180-242 days of pregnancy, would require an additional 2288MJ of metabolisable energy, over and above the feed required to maintain the cow's current level of body condition (refer Appendix 3.6.1.2).

- * The metabolisable energy intake was estimated for each cow by multiplying the dry matter intake by the energy concentration (MJ metabolisable energy per kg DM) of the feed. The energy concentration of the feed was calculated as the average energy concentration of the feed over the two collection periods (total of 45 days).

To maintain body condition during the period 180-242 days of pregnancy, the feed requirement can be calculated (from the above equation) to be $0.76\text{MJ MEkg}^{-0.75}\text{cow}^{-1}\text{day}^{-1}$ (refer Appendix 3.6.1.1).

3.4 DISCUSSION

3.4.1 Energy balance

3.4.1.1 Experiment 1 : Lactating Cows

The H cows produced more milk energy in early and late lactation than the L cows (Table 3.6), and these differences were associated with a greater energy intake (Table 3.5) and differences in partitioning of nutrients between milk and body tissue.

Flatt et al. 1969) reached a similar conclusion for cows varying in productive ability and fed varying proportions of alfalfa:concentrates in the diet.

The energy losses in faeces, urine, and methane, were similar for H and L cows, hence the differences in the partitioning of energy related to differences in the partitioning of metabolisable energy. After adjustment for differences in intake between genotypes, only in late lactation were there significant differences between genotypes in partitioning of metabolisable energy. However trends in partitioning of metabolisable energy have been examined by fitting a different slope and a different intercept* for each BI group (refer statistical analysis). These equations were used to predict the outputs of energy (heat, milk and tissue) for each BI group at the mean energy intakes (average of H and L cows) during each of the five collection periods over lactation (Figure 3.9).

* The intercept for each BI group was calculated as the average of the intercepts of the individual cows in each group.

During four of the five collection periods, losses of energy as heat were similar for H and L cows, hence total energy (milk plus tissue) retained was also similar. However H cows partitioned more of the energy retained to milk and less to tissue than the L cows (Figure 3.9).

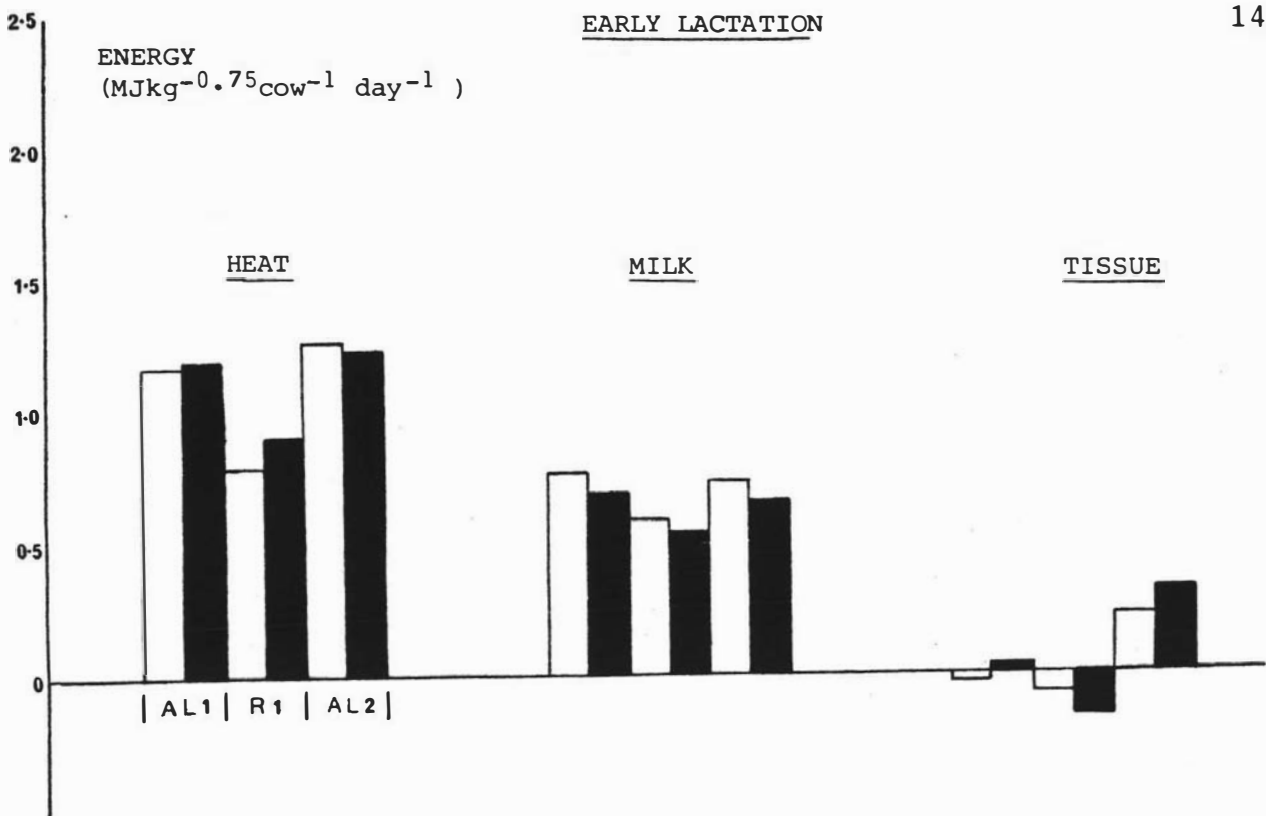
In the other collection period (restricted feeding in early lactation) H cows lost less energy as heat, retained only slightly (7%) more milk energy and mobilised less tissue energy than the L cows (Figure 3.9).

It is of interest, and reassuring that an independent estimate of relative tissue mobilisation, namely the proportion of long chain fatty acids in the milk fat, also indicated that the L cows mobilised more tissue reserves on the restricted level of feeding in early lactation than the H cows (Figure 3.7).

Causes of differences in milk energy output between genotypes can be clearly seen by examining the ratio (H : L) in milk energy output before and after adjusting for differences between genotypes in intake, and intake plus tissue changes (Figure 3.10). In early lactation differences in milk energy output were equally explained by differences between BI groups in intake and tissue changes.

In late lactation 60% of the observed difference was due to differences in tissue changes between BI groups, and only 40% to differences in intake. The exception was during restricted feeding in early lactation where there were differences in heat production.

EARLY LACTATION



LATE LACTATION

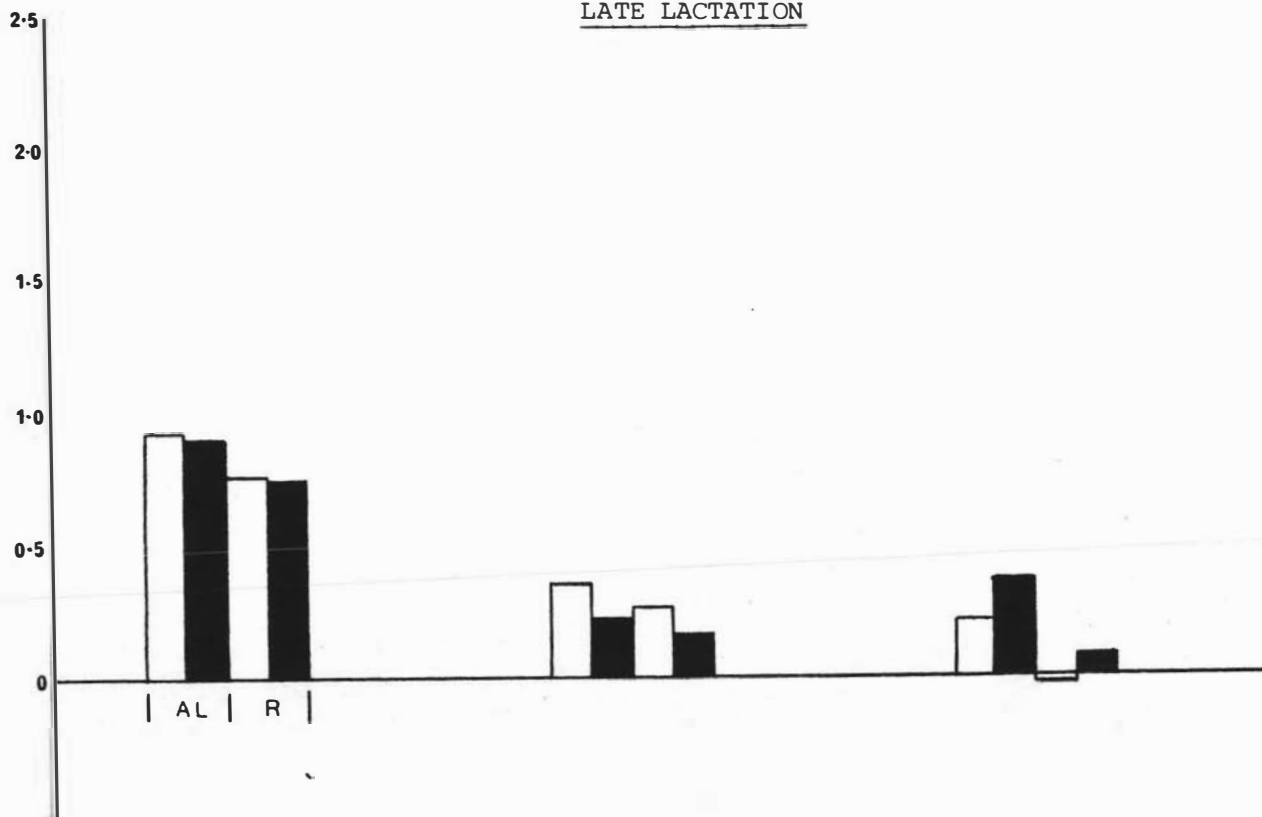


FIG. 3.9: Partitioning of metabolisable energy for H (□) and L (■) BI cows at the mean energy intakes during collection periods in the sequence ad libitum (AL1), restricted (R1), ad libitum (AL2) feeding in early lactation, and ad libitum (AL), restricted (R) feeding in late lactation (see text for further details)

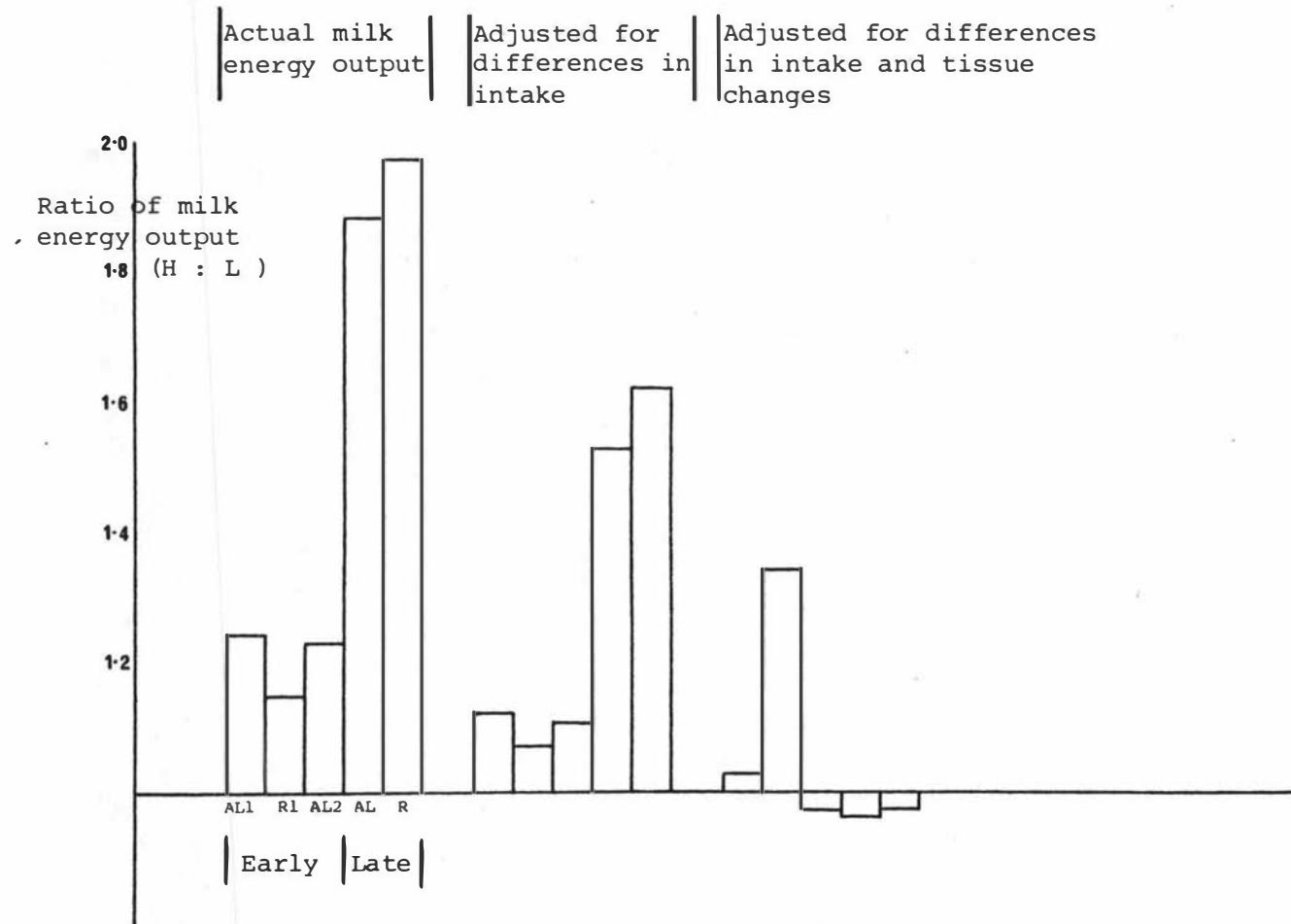


Fig.3.10: Ratio of milk energy output (H:L) for the five collection periods during lactation (AL1,R1,AL2,AL,R) in chronological order; unadjusted ratio, ratio after adjusting for differences in intake and tissue changes (see text for further details).

The measured heat productions in this study were partly due to heat produced by respiration of the pasture in the hoods. Although a correction was made to allow for the heat produced by the respiration of the pasture it is worthwhile examining how the absolute levels of heat production, after correction for herbage respiration, compare with other published calorimetry data where pasture forms the diet. When the regressions of energy retention (milk plus tissue energy) against metabolisable energy intake obtained from this study have been compared with other published data (Figure 3.11) there is reasonable agreement, giving more confidence to the corrected heat productions. Further details of the regression equations shown in Figure 3.11 are given in Appendix 3.5.

In the present experiment, during restricted feeding in early lactation, the H cows had a greater efficiency of milk production because of a lower heat production relative to the L cows, not to a greater mobilisation of body reserves. Growth hormone has been shown to increase the efficiency of milk production (measured as milk energy produced divided by gross energy) in dairy cows (Hutton, 1957; Machlin, 1973; Peel et al. 1980).

Whether the increased efficiency is due to a lowered heat production and/or a more favourable partitioning of nutrients to milk rather than tissue energy is not known, although current research at Cornell University (Peel, C.J. personal communication) should clarify the mode of action of growth hormone. Growth hormone concentrations were not measured in the current experiment, but have been observed to be higher for high versus low breeding index cows (Flux, D.S., Mackenzie, D.D.S., and Wilson, G.F., unpublished data) and are also at their highest concentration

Energy retention
 (MJkg^{-0.75} cow⁻¹day⁻¹)

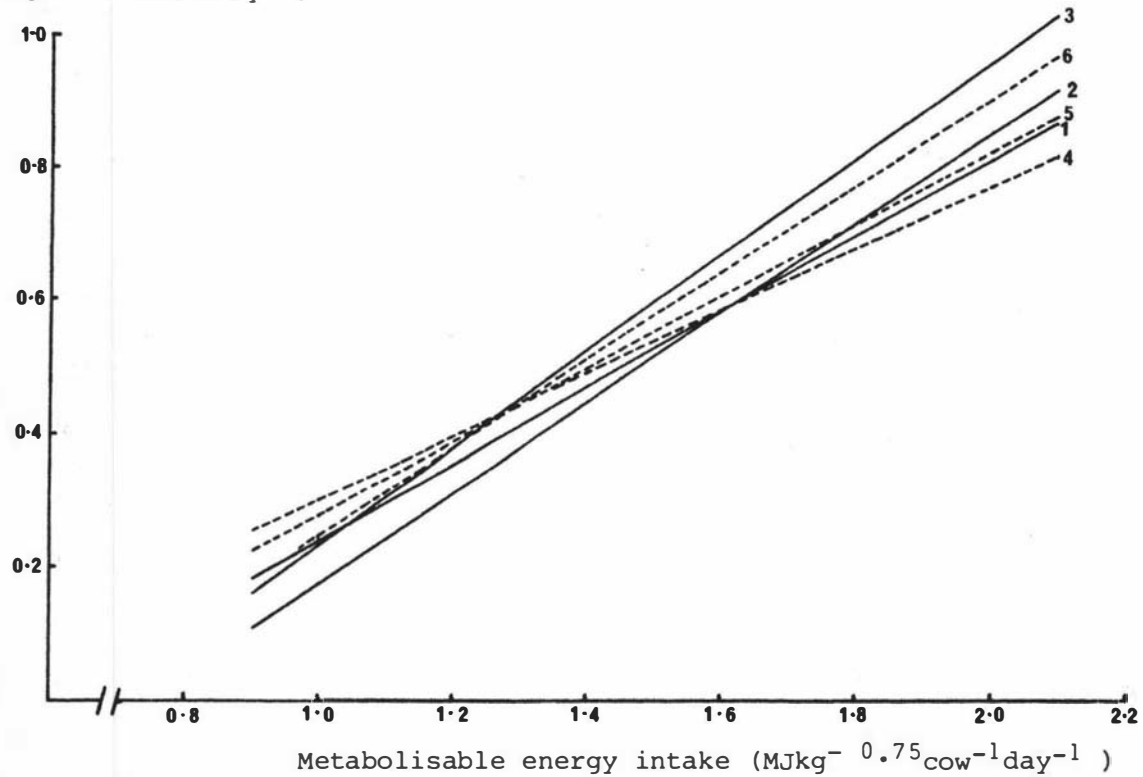


FIG.3.11: A comparison of the relation between energy retention (milk plus body tissue) and metabolisable energy intake obtained in the present study with other published data from experiments where cows were fed on pasture.

1.Trigg et al (1980)

2.Trigg and Parr (1981)

3.Van Es (1975)

4* Early lactation (ad libitum to restricted feeding)

5* Early lactation (restricted to ad libitum feeding)

6* Late lactation (ad libitum to restricted feeding)

* regressions from the present study

in early lactation (Vasilatos and Wangsness, 1981). Flux, Mackenzie and Wilson (unpublished data) observed that when intake was restricted in mid-lactation, growth hormone concentrations increased in high and low breeding index cows with a greater increase for the high breeding index cows.

It might be speculated therefore that changes in growth hormone metabolism may have played some role in the rather unexpected difference in heat production between the BI groups measured during restricted feeding in early lactation.

3.4.1.2 Experiment 2 : Non-lactating cows

There were only small, non-significant differences between genotypes in losses of energy in faeces, urine, and methane, and in the efficiency of utilisation of metabolisable energy. The data has therefore been pooled across genotypes and can be compared to other experimentally and theoretically derived estimates relating to feeding of dry, pregnant cows on pasture.

Maintenance of body condition

In the present experiment there were two estimates of the feed required to maintain body condition. Firstly using data from the 62 day indoor feeding period (12 cows) the feed required to maintain body condition was $0.76\text{MJkg}^{-0.75}\text{cow}^{-1}\text{day}^{-1}$ (see results) A second estimate can be made by calculating the metabolisable energy intake (MEI) required to maintain

zero maternal body energy retention. The calorimetric balance estimates the amount of MEI required for total zero energy retention in the dam and the foetus, however at this intake the dam will be in negative energy balance because a portion of the intake is utilised to maintain the growth of the foetus. Therefore to maintain zero maternal body energy retention, extra intake, equivalent to the MEI required to maintain the growth of the foetus must be added to the estimated MEI required to maintain total zero energy retention. Using this method the feed required to maintain zero maternal body energy retention for periods 1 and 2 (approximately day 210 and 230 of pregnancy) was 0.79 and 0.80 MJ MEkg^{-0.75}cow⁻¹day⁻¹ (see Appendix 3.3 for full calculations).

There is close agreement between the estimates of feed energy required to maintain body condition and zero maternal body energy retention, namely 0.76 and 0.80 (0.79 and 0.80) MJ MEkg^{-0.75}cow⁻¹day⁻¹ which gives some confidence in the visual assessment of changes in body condition.

Theoretical and recent experimental estimates of feed required to maintain body condition of dairy cows are summarised in Table 3.16. The agreement between theoretically derived estimates (ARC, 1980) to maintain zero maternal body energy retention and estimates for animals fed indoors in stalls (present experiment and Gray et al. 1981) is reasonable, the two estimates being about 30% higher. However the estimate for grazing cattle (Holmes and McClenaghan, 1980) is 70% higher than the theoretical estimate possibly indicating a greater requirement because of environmental factors such as

wind, cold, or to the greater activity of the grazing animal.

Table 3.16 Some experimentally measured and theoretically derived estimates of the metabolisable energy required by dry cows fed pasture.

Reference and experimental details	Estimation of changes in body condition	Stage of pregnancy (days)	Metabolisable energy required to maintain body condition ** (MJ kg ^{-0.75} day ⁻¹)
Present experiment Stall fed on pasture	Visual	180-242	0.76
Present experiment	Calorimetry*	210 (approx)	0.79
Present experiment	Calorimetry*	230 (approx)	0.80
<u>Gray et al (1981)</u>			
Stall fed on pasture plus silage	Slaughter*	180-230	0.71
<u>Holmes and McClenaghan (1980)</u>			
Grazed on pasture	Visual	195-237	1.02
ARC (1980)			
	Calorimetry	252	0.69
		224	0.60

* Calculated for zero change in body energy content plus pregnancy allowance.

** All values are expressed per fasted liveweight.

Gain in body condition

Holmes et al. (1981) drew attention to the fact that the few estimates of the quantity of metabolisable energy required to increase body condition of the non-lactating cow by one condition score are about twice as large as that predicted theoretically. In the present experiment the net energy retention for one unit of body condition (43.8 kg liveweight) was 1226 MJ (2366×0.518) or 28.0 MJ/kg liveweight gain. This is only 1.3 times greater than the most recent estimate by the Agricultural Research Council (1980) of 21.6 MJ/kg liveweight gain.

A summary of estimates of metabolisable energy required per gain in condition score are presented in Table 3.17.

Table 3.17 Some estimates of the feed required for increasing body condition in dry cows.

Reference	Metabolisable energy required to increase body condition * by one unit (MJ * cow ⁻¹)
Hutton and Bryant (1976)	1925
Holmes and McClenaghan (1980)	2310
Grainger <u>et al</u> (1978)	2900
Gray <u>et al</u> (1981)	1628
Present experiment	1828
ARC (1980)	1400

* One unit of condition assumed to be equal to 35kg liveweight.

* Metabolisable energy of the feed assumed to be 11MJ/kg DM.

Since the feed requirements to maintain and promote a gain of one unit of body condition are similar for H and L cows, the H cows will require extra feed during the dry period in order to calve in a similar condition to the L cows.

3.4.2 Nitrogen balance

3.4.2.1 Experiment 1

In early lactation differences between genotypes in partitioning of N were small and not significant. An interesting observation was that during the first ad libitum feeding period, the cows actually retained less nitrogen in the body than during the restricted feeding period, although in the second ad libitum feeding period the cows gained more nitrogen. The reasons for the difference in nitrogen retention in tissues between the two ad libitum feeding periods, were that the cows during the first ad libitum feeding period lost more N in faeces and retained more N in milk than during the second ad libitum feeding period. As a result the cows retained less N in the body during the first ad libitum period.

In late lactation the H cows retained a small but significantly greater amount of N in the milk at a given N intake than the L cows. This was because the L cows, at a given intake, lost more N in faeces and urine and although they were also in negative N balance (as opposed to the H cows in positive N balance) this was not sufficient to offset the greater losses of N in faeces and urine. Consequently the L cows retained less N in their milk.

In general differences between genotypes in N balance were small.

3.4.2.2 Experiment 2

During period 2 there were small but significant differences in N balance between genotypes. Low cows had greater losses of N in faeces and urine at high intakes and smaller losses at low intakes, hence L cows retained less N at high and more N at low intakes than the H cows. However the differences between genotypes in N balance were not consistent between periods since in Period 1 there were no differences in N balance between genotypes. The observed differences in N balance between genotypes in Period 2 are possibly a result of the small numbers of cows involved.

It is concluded that there are only small differences in N balance between genotypes.

3.5**CONCLUSION**

Losses of energy in faeces, urine, methane, and heat are similar for H and L cows. High BI cows produce more milk energy because of a higher intake and by partitioning more of the energy retained into milk at the expense of body tissue. The anomalous result, where H cows had a lower heat production than L cows during restricted feeding in early lactation, requires further investigation.

The feed required to maintain and promote a gain in body condition during the dry period is similar for H and L cows. High BI cows will require more feed over the dry period in order to attain a similar level of body condition at the next calving to the L cows, since H cows are in lighter condition at the end of lactation than L cows.

CHAPTER FOUR

THE IMPLICATIONS OF CORRELATED ERRORS
TO THE ANALYSIS AND INTERPRETATION OF
NUTRITION EXPERIMENTS

4.1 INTRODUCTION

One of the main objectives of the present thesis was to determine how high breeding index cows produce more milk than low breeding index cows. One of the methods used was to compare the energy balances for each genotype at different stages of lactation and during the dry period. (see Chapter 3).

In this chapter discussion will focus on possible correlations between errors of the components of an energy balance and on the subsequent statistical analysis and interpretation of the energy balance data.

Another method used in this thesis was to compare the performance of each genotype under stall feeding conditions where measurements are made of feed intakes, milk yields, liveweight changes, and liveweight for individual cows (see Chapter 2). Possible correlations between the errors of the measured variables, in particular milk yield and liveweight changes, are considered with particular reference to the correlations which might be 'expected', based on the preceding discussion relating to the complete energy balance.

Finally consideration is given to the effects of possible correlations between errors of measured variables on the statistical analysis and interpretation of stall feeding experiments.

4.2 ENERGY BALANCE EXPERIMENTS

4.2.1 Correlated errors and energy balance experiments

The major technique used to study energy metabolism is the calorimetric balance (ARC, 1980). A calorimetric balance equation for the cow can be written:

Energy input	Energy output
GEI + ϵ	FE + ϵ_1
	UE + ϵ_2
	CE + ϵ_3
	HE + ϵ_4
	ME + ϵ_5

	TE + ϵ_6

where GEI = gross energy intake
 FE = faecal energy
 UE = urinary energy
 CE = methane energy
 HE = heat energy
 ME = milk energy
 TE = tissue energy
 ϵ = error term

Tissue energy (TE) is not measured, but obtained by difference (TE = GEI - (FE + UE + CE + HE + ME)) hence it contains errors associated with the measurement of gross energy intake, faecal, urinary, methane, heat, and milk energies. Clearly TE may be a negative, zero, or positive value.

Metabolisable energy intake (MEI) and total energy retained (E_{TOT}) are also obtained by difference.

$$\begin{aligned} \text{MEI} &= \text{GEI} - (\text{FE} + \text{UE} + \text{CE}) \\ E_{TOT} &= \text{GEI} - (\text{FE} + \text{UE} + \text{CE} + \text{HE}) \\ &= \text{MEI} - \text{HE} = \text{ME} + \text{TE} \end{aligned}$$

The proportions of energy will change as a function of the values of several variables. Such variables include:

- metabolic size,
- stage of lactation,
- environment,
- amount of GEI,
- type and quality of diet,
- age,
- genetic merit,
- state of body reserves,
- breed.

The effects of metabolic size can be reduced by expressing energy values per unit metabolic liveweight. The most commonly accepted unit of size is liveweight raised to the power 0.75 (ARC, 1980). Other variables such as those listed above can be held constant in any particular experiment so as to study the effect of one variable such as genetic merit on the proportions of energy.

By expressing energy values per unit metabolic liveweight and making the necessary simplifying assumptions, the various outputs of energy (FE, UE, CE, HE, ME, and TE) can be related to the energy input (GEI) by a series of equations:-

$$FE = a_1 + b_1 GEI + e_1 \quad (1.1)$$

$$UE = a_2 + b_2 GEI + e_2 \quad (1.2)$$

$$CE = a_3 + b_3 GEI + e_3 \quad (1.3)$$

$$HE = a_4 + b_4 GEI + e_4 \quad (1.4)$$

$$ME = a_5 + b_5 GEI + e_5 \quad (1.5)$$

$$TE = a_6 + b_6 GEI + e_6 \quad (1.6)$$

Because $GEI = FE + UE + CE + HE + ME + TE$
it follows that:

$$\sum a_j = 0$$

$$\sum b_j = 1$$

In addition,

$$\sum e_j = 0$$

Correlations between errors (e_j) are therefore expected. Hence a cow with an output of energy in faeces which is greater than expected will have an output of energy which is less than expected elsewhere in the system (e.g. urine and methane).

If the equations do not explain a large part of the observed variation then this could be due to:

- (a) Measurement errors.
- (b) The functional form of the equations - the equations might be more adequately described with curvilinear rather than linear terms.
- (c) Missing variables - variables that determine a significant part of the observed variation may have been omitted unwittingly.

4.2.2 Implications of correlated errors to the statistical analysis and interpretation of energy balance experiments.

If the high and low breeding index cows were compared for the various energy outputs as though each energy output were fully independent, then it is likely that tests of significance would be invalid (Kramer, 1978).

A more valid appraisal of the differences between genotypes in their utilisation of energy should be made by comparing energy outputs jointly via simultaneous confidence intervals based on a multivariate distribution (Gill, 1981).

Whilst differences between genotypes in energy balance are the focus of attention in this thesis, the principles apply equally to other cases. For example, differences in energy balance between different diets or between groups of cows with different nutritional histories, could be examined.

The experimental design of energy balance experiments is generally one of two types. The first involves data obtained from cows fed at different levels, each cow being offered only one feeding level at any particular stage of lactation. The second type involves data obtained from each cow at two levels of feeding (for example, ad libitum and approximately 70% ad libitum), at each stage of lactation. The data in the second case involve repeated measurements on the same animal.

The statistical procedures developed in the course of analysing the calorimetry data in the present thesis are outlined in detail because the analysis is different from, but more appropriate than that used normally.

4.2.2.1 Energy balance with repeated measurements on the same cow (within-cow).

The data are multivariate in two senses. Firstly measurements have been made on several variables (faeces, urine, methane, heat, and milk) which are not fully independent of one another. Secondly the variables have been measured at different times on individual animals, hence involve repeated measurements.

The measured energy outputs (faeces, urine, methane, heat, and milk) are analysed multivariately. The residual (tissue energy) is analysed separately as a univariate case since if it were included in the multivariate analysis, all of the variation would be explained.

The analysis of the repeated measurements over time is similar to covariance analysis in split-plot experiments (Snedecor and Cochran, 1967). Cows are 'main-plots' and main-plot treatments (H and L BI) are 'allocated' to cows. Within cows (main-plot) sub-plot treatments (levels of feeding; ad libitum, 70% ad libitum) are located.

The sequence of levels of feeding can be either ad libitum followed by 70% ad libitum, or 70% ad libitum followed by ad libitum. It is likely that the physiological response of the cow will be different depending on the feeding sequence, consequently data for each feeding sequence should be analysed separately.

The analysis is done in two parts. Differences between genotypes in marginal efficiency (rate of change of energy output per unit increase in energy input) are compared first, followed by differences in gross efficiency

$$\left\{ \frac{\text{energy output}}{\text{energy input}} \right\}$$

Step 1 Sub-plot analysis testing differences in slopes (marginal efficiency) between BI groups.

(a) The errors of the five energy outputs (faecal, urinary, methane, heat, and milk) are tested for independence in a multivariate analysis using the following model (different intercept for each cow, one slope for each BI group) -

$$y_{ijk} = \alpha_{ij} + \beta_i x_{ijk} + e_{ijk}$$

where

y = faecal, urinary, methane, heat, milk energies (MJkg^{-0.75} cow⁻¹ day⁻¹)

x = gross energy intake (sub-plot treatment) (MJkg^{-0.75} cow⁻¹ day⁻¹)

α = intercept term

i = genetic level (H,L)

j = jth cow

k = observation on jth cow (k = 1,2)

e_{ijk} = sub-plot (within-cow) error (σ²e)

The appropriate error for sub-plot treatments is obtained from within-cow data (Figure 4.1). The main interest lies in whether or not there is any sub-plot treatment by BI interaction, i.e. whether the marginal efficiency differs between BI groups.

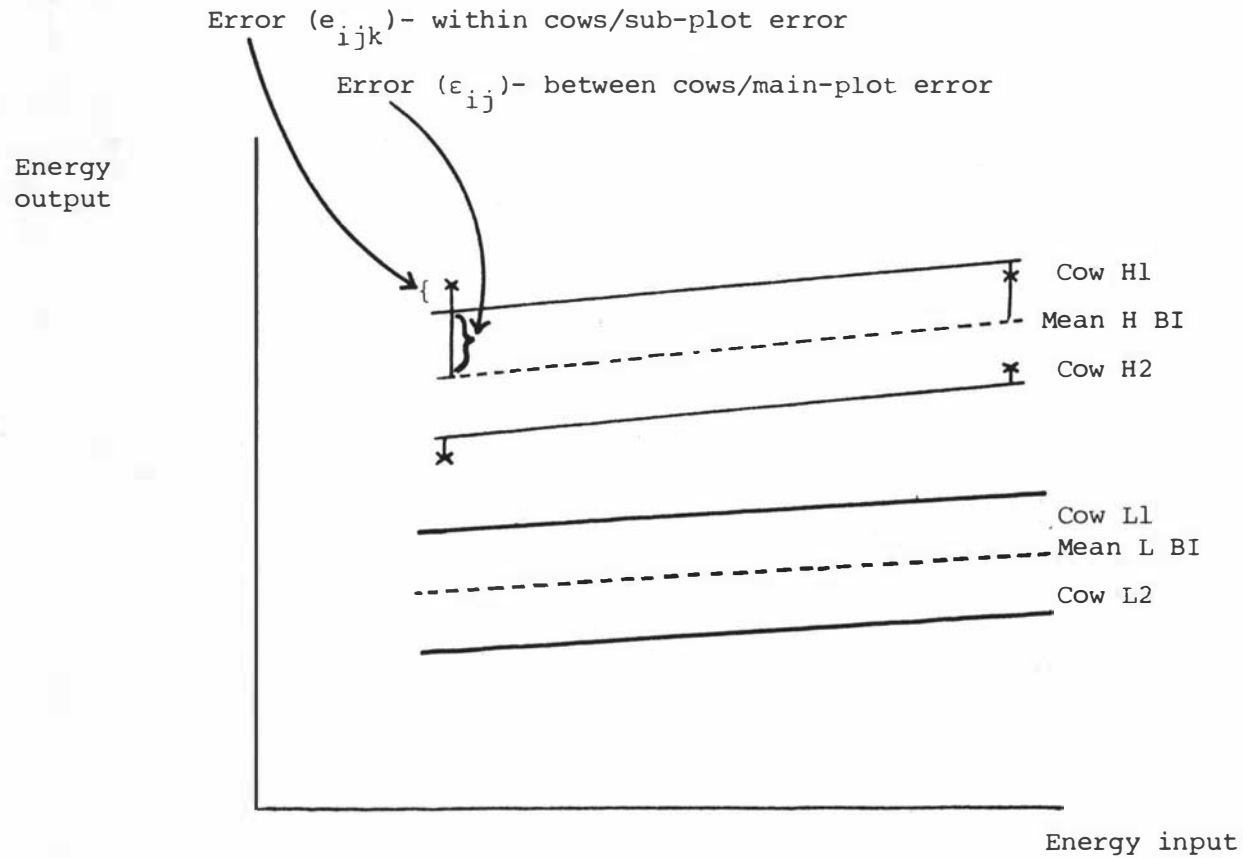


FIG. 4.1 : Schematic representation of split-plot error structure. The mean slope of the line for high (H) and low (L) breeding index (BI) cows and individual slopes for two H (H1 and H2) and two L (L1 and L2) BI cows are presented. The sub-plot and main-plot errors are labelled for cow H1 .

If the errors are independently distributed then the differences in slope between BI groups for the five energy outputs are more appropriately tested univariately. Otherwise valid tests of significance are obtained using the multivariate test of significance.

(b) (i) Univariate analysis

The null hypothesis is that the difference in slope between BI groups for each of the energy outputs, tested independently, is zero. This is exactly analogous to the first step in an analysis of covariance where it is tested to see whether the within group (usually treatment - in this case main-plot treatment groups) regressions have the same slope.

If the null hypothesis is accepted, a common slope is fitted and intercepts (α_j) predicted.

If the null hypothesis is rejected, then there are differences in slopes between BI groups. To examine the differences in slopes it is suggested that the slopes for each BI group should be plotted with their confidence intervals. When calculating the confidence intervals the appropriate error (within-cow error) must be used.

(b) (ii) Multivariate analysis

The null hypothesis is that the difference in slope between BI groups for each of the energy outputs, tested jointly, is zero. If the null hypothesis is accepted a common slope is fitted and intercepts (α_j 's) predicted. It is worth noting that the estimated regression coefficients calculated either multivariately or univariately are identical. Only the tests of significance are affected by the form of analysis.

Assuming the null hypothesis is rejected, then there are differences in slopes between BI groups. To characterise exactly what these differences are, it is necessary to perform a canonical variates analysis (CVA). CVA finds a linear function of the energy outputs which maximises the difference between the BI groups (Cooley and Lohnes, 1971).

Step 2 Main-plot analysis testing differences in intercepts (predicted α_j 's) between BI groups

Assuming that there were no sub-plot treatment by breeding index interactions, differences in intercepts can be tested. Between-cow differences are due to breeding index and error. The appropriate error for testing the significance of differences between BI groups is obtained from between cows within BI groups, the model being -

$$\alpha_{ij} = \mu + \lambda_i + \epsilon_{ij}$$

where

α = intercept

μ = mean

i = genetic level (H,L)

j = jth cow

ϵ_{ij} = main-plot (between-cow) error ($k\sigma_c^2 + \sigma_e^2$)

(a) The independence of errors are tested in a multi-variate analysis using the above model to determine whether the data should be analysed multivariately or univariately.

(b) (i) Univariate analysis

The null hypothesis is that the difference in intercept between BI groups for each of the energy outputs, tested independently, is zero.

(b) (ii) Multivariate analysis

The null hypothesis is that the difference in intercept between BI groups for each of the energy outputs, tested jointly, is zero. If the null hypothesis is rejected then differences between BI groups in intercept for the five energy outputs can be characterised by canonical variates analysis as previously outlined.

4.2.2.2 Energy balance with no repeated measurements on each cow

As there are no repeated measurements on each cow the analysis is simplified, but the multivariate nature of the data, with respect to the energy outputs, still remains. The procedure is to use stepwise regression analysis using the following model (different intercept for each BI group, different slope for each BI group)

$$y_{ij} = \mu + \alpha_i + \beta_i x_{ij} + e_{ij}$$

for jth cow in ith group

where

- i = genetic level; H or L
- j = 1...6 cows
- μ = intercept for L cows
- α_L = 0
- α_H = difference in intercept between genotypes
- Y = energy output in faeces, urine, methane or heat
(MJkg^{-0.75} cow⁻¹ day⁻¹)
- x = gross energy intake (MJkg^{-0.75} cow⁻¹ day⁻¹)
- e_{ij} = between-cow error

The order of fitting of the variables should be; gross energy intake, breeding index, gross energy intake by breeding index interaction. By using a stepwise procedure the error used in testing the significance of the main effects (gross energy intake and breeding index) and the interaction (gross energy intake X breeding index) is the error from the full model (model with a different intercept and slope for each BI group).

The decision to use either univariate or multivariate analysis depends on whether or not the errors are correlated.

4.2.3 Additional considerations

The system of equations described earlier which related the components of energy balance to gross energy intake can be used to illustrate two further points concerning

- (a) the estimation of the relation between tissue energy and gross energy intake,
- (b) the statistical procedure when metabolisable energy intake is used as the independent variable.

4.2.3.1 The estimation of the relation between tissue energy balance and gross energy intake

The relation

$$TE = a + bGEI$$

is estimated by predicting values for TE from

$$GEI - (FE + UE + CE + HE + ME).$$

Rearranging we obtain:

$$(FE + UE + CE + HE + ME) = -a + (1-b) GEI$$

As an example, the regression equations relating the energy outputs to GEI (using data from Chapter 3) are:

Energy output	Constant ('a' value)	GEI ('b' value)
FE	-0.175	0.306
UE	0.071	0.028
CE	0.025	0.055
HE	0.304	0.279
ME	0.378	0.091

From the above relations, the least squares estimate of $-a$ is given by the sum of the estimated constant terms, and the least squares estimate of $(1-b)$ is given by the sum of the estimated slopes.

Therefore, $\hat{TE} = -0.603 + 0.241 GEI$

4.2.3.2 The statistical procedure when metabolisable energy intake is used as the independent variable

Metabolisable energy allows for differences between feeds in their metabolisability.

It is common practice to relate milk and tissue energy or heat energy to metabolisable energy intake (MEI) by regression with MEI as the independent variable.

Because MEI values are estimated by difference ($GEI - FE - UE - CE$) they contain errors related to the measurement of gross energy intake, faecal, urinary, and methane energies. Use of observed metabolisable energy intake values in regression analysis would give biased estimates of regression coefficients - this is the problem of "errors in the variables" (Johnston, 1963).

Van Es (1972) outlining the estimation of maintenance requirements by means of regression methods has recognised that: "In any kind of regression the reliability of the regression coefficients is highest when the independent variable (in this case MEI) has no measurement error, when there is a great variation of this variable in the material used and when the variation of the dependent variable is caused mainly by the variation of the independent variable (s). All errors of measurement of the variables, or errors due to the use of an incomplete or incorrect model, result in a lower reliability of these coefficients."

Where the variable of interest can be estimated in terms of controlled variables, the method of two-stage least squares can be used to obtain consistent estimates of regression coefficients in the original model. Therefore MEI should be predicted as observed GEI minus predicted values for faecal, urinary, and methane energy (obtained from the regression equations with GEI as the independent variable). The method is demonstrated below.

Considering as before, the equations relating the various components of energy balance to GEI, \hat{MEI} can be estimated -

$$\hat{MEI} = GEI - (-0.079 + 0.389 GEI)$$

where the term in brackets is the sum of the three regression equations (FE, UE, and CE).

Therefore

$$\hat{MEI} = 0.079 + 0.611 GEI.$$

The second step is to use the predicted values for MEI to estimate, for example, the relation for milk energy (ME) -

$$ME = a + b \hat{MEI} + \text{error}.$$

We could obtain the (two-stage) least squares estimates for a and b either directly by regression of ME on \hat{MEI} , or indirectly, as follows:-

Substituting for \hat{MEI} from above, we obtain:

$$\begin{aligned} ME &= a + b (0.079 + 0.611 GEI) \\ &= a + b (0.079) + b (0.611) GEI \\ &= \alpha + \beta GEI \end{aligned}$$

The least squares estimate for a and $b(0.079)$ will equate with the least squares estimate for α , and similarly for $b(0.611)$ and β . Since the least squares estimates for α and β have already been calculated (see above), the least squares estimates for a and b can be obtained -

$$\begin{aligned}\hat{b} &= \hat{\beta} (0.611)^{-1} = 0.091 (0.611)^{-1} \\ &= 0.149 \\ \hat{a} &= \hat{\alpha} - \hat{b}(0.079) = 0.366.\end{aligned}$$

Thus

$$\hat{ME} = 0.366 + 0.149 \hat{MEI}$$

That the estimated relations are different, depending on whether MEI or \hat{MEI} is used as the independent variable, is shown in Table 4.1.

Table 4.1 Comparison of regression coefficients (b) and intercept terms (a) using predicted metabolisable energy intake (\hat{MEI}) and metabolisable energy intake (MEI) as the independent variables and heat, milk, and tissue energies ($MJkg^{-0.75} \text{ cow}^{-1} \text{ day}^{-1}$) as the dependent variables (see text for details).

Y	X	a	b(±s.e.)
Heat	\hat{MEI}	0.268	0.456 ± 0.050
Heat	MEI	0.280	0.449 ± 0.058
Milk	\hat{MEI}	0.366	0.149 ± 0.023
Milk	MEI	0.366	0.150 ± 0.022
Tissue	\hat{MEI}	-0.634	0.394 ± 0.060
Tissue	MEI	-0.646	0.401 ± 0.069

4.3 NUTRITION EXPERIMENTS

4.3.1 Correlated errors and nutrition experiments

In stall-feeding experiments, energy balance data are not available, but measurements are generally made on milk yield and milk composition, liveweight change and feed intake for individual cows. In this case, the balance is not complete, i.e. it is not expected that the sum of the parts: maintenance and milk and tissue balance will equal the whole. Missing parts include some of the components of total heat production of the cow and also losses of energy in faeces, urine, and methane.

Thus, some fraction of intake is partitioned between maintenance, milk yield, and liveweight change. Main interest is focussed on the partition of energy between the two components of total energy balance (E_{TOT}), milk yield and liveweight change.

Since it is generally accepted that there are only small differences between animals in losses of energy as faeces, urine, methane, and heat (Flatt, 1969) it might be expected that a cow which produces an output of energy in milk which is greater than expected, will have an output of energy in body tissue which will be less than expected. Support for this hypothesis stems from the observations that for individual cows within groups on constant diets there are negative regressions of liveweight change on milk yield (Broster et al. 1969; Broster et al. 1975). It should be noted that correlations between the errors of measured variables and correlations between the measured variables are equivalent when the feeding level is fixed.

Further examples of correlations between the errors of milk yield and liveweight change are now considered.

Moe and Tyrrell (1975) termed the uncertainty regarding distribution of energy between milk and body tissue as the "partition problem". More recently Townsley et al. (1981) outlined a modelling approach to the "partition problem" and their approach is outlined briefly.

The model comprises three equations:

$$\text{FCM} = f_1(W^{0.75}, \text{DMI}) + e_1 \quad (1.1)$$

$$\Delta W = f_2(W^{0.75}, \text{DMI}) + e_2 \quad (1.2)$$

$$e_1 * = f_3(e_2) \quad (1.3)$$

where

FCM = fat corrected milk

$W^{0.75}$ = metabolic liveweight

DMI = dry matter intake

ΔW = body tissue change

e = error term.

Equation (1.3) links equations (1.1) and (1.2) and may in part capture differences in genetic ability between animals and in part the effect of differences in nutritional history between animals.

* Since there is no a priori reason to consider one of the errors as dependent on the values of the other the relation may be more appropriately written as:

$$f_3(e_1 e_2) = 0$$

The estimated equations* presented in the paper for the analysis of a nutrition experiment were:-

$$FY_{0-20} = 65.11 - 0.62DMI + 1.49CS + 0.67CS*DMI \quad (1.1)$$

$$R^2 = 0.40$$

$$CS_{0-20} = 2.27 + 0.10DMI - 0.48CS - 0.02CS*DMI \quad (1.2)$$

$$R^2 = 0.63$$

where

FY_{0-20} = cumulative milk fat yield (kg)
0-20 weeks of lactation

CS_{0-20} = change in condition score,
0-20 weeks of lactation

CS = condition score at calving

DMI = dry matter intake (kgDM cow⁻¹ day⁻¹),
0-5 weeks of lactation.

* It is likely that additional variation would be explained if a term for liveweight was included in the model.

The third equation (1.3) was not presented, but is given below:

$$\hat{e}_1 = - 7.968(\pm 1.627) \hat{e}_2 \quad (1.3)$$

$$R^2 = 0.11$$

The estimate of equation (1.3) indicates a negative correlation between the errors of equations (1.1) and (1.2) in support of the observed negative regressions of liveweight change on milk yield for individual cows

within groups on constant diets

(Broster et al. 1969; and Broster et al. 1975).

To test the hypothesis further that there is a negative correlation between the estimated errors of liveweight change and milk yield, and to examine the possible influence of genetic merit on this relation, data from Experiments 2 and 4 (Chapter 2) were analysed according to the model proposed by Townsley et al. (1981). Experiment 3 was excluded as there were no significant relations between weight change and intake and/or breeding index. Only correlations between the errors of equations 1.1 and 1.2 are given below because the equations (1.1 and 1.2) were given in Chapter 2.

$$\text{Experiment 2 } e_1 = -0.030 (\overset{+}{-}0.099) e_2 \quad R^2 = 0.01$$

$$\text{Experiment 4 } e_1 = 0.126 (\overset{+}{-}0.087) e_2 \quad R^2 = 0.12$$

A negative correlation between the errors was observed for Experiment 2, but a positive correlation for Experiment 4, although neither correlation was significant ($P > 0.05$).

From discussion of the calorimetric balance equations, considered earlier in this chapter, it is known that the expected value of the sum of all the errors ($\sum e_j$) is equal to zero. There will be some positive and some negative correlations between the errors of the six energy outputs (faeces, urine, methane, heat, milk, and tissue energies). For example, a cow with an output of energy in milk which is greater than expected may have an output of energy in heat or faeces which is smaller than expected, but not necessarily an output

of energy in body tissue which is smaller than expected. In any particular experiment, the correlations between errors are possibly unique to that experiment.

Measurement errors, particularly related to the measurement of changes in body tissue energy may be affecting the observed values for the correlations between the errors. In feeding experiments liveweight changes are 'the best' estimate of changes in body tissue energy, but it is well known that liveweight change does not provide accurate estimates of changes in body tissue, a point recently highlighted by Cowan et al. (1980). In the calorimetric balance, tissue energy is estimated by difference therefore contains measurement errors associated with the measurement of GEI, FE, UE, CE, HE, and ME.

It is clear from the above discussion that in any particular feeding experiment the correlations between the errors of liveweight change and milk yield may be zero, positive or negative.

4.3.2 Implications of correlated errors to the statistical analysis and interpretation of nutrition experiments

There are two points to be made.

The first point was raised by Townsley et al. (1981) and it concerns the estimation of the 'partial' balance equation in a feeding experiment as a single equation, for example in the form:

$$\text{DMI} = a + bW^{0.75} + c\text{FCM} + d\Delta W + e$$

This model would be useful if the experimenter wished to estimate the dry matter intake associated with

different observed levels of the performance variables. Direct estimation of the equation, as outlined, leads to biased estimates of the partial regression coefficients, hence does not enable the experimenter to predict the distribution of energy between performance functions.

When the objective of a particular feeding experiment is to analyse the effects of alternative feeding strategies, a modelling approach (equations 1.1, 1.2) can be used. This has the advantage that unbiased estimates of the partial regression coefficients are made and enables the experimenter to estimate performance response to different feeding levels at different stages of lactation, hence is useful from a management viewpoint.

The second point concerns tests of significance. Since the various measurements on each individual cow are often correlated, it is inappropriate to apply univariate analyses separately to each of the response variables. Thus equations 1.1 and 1.2 should be analysed jointly in a multivariate analysis in order to obtain valid tests of significance.

Kramer (1978) stated : "Univariate analysis is rather a simplification open to the experimenter who happens to be measuring only one characteristic of his experimental material". It must be rare indeed for animal nutritionists to measure only one characteristic of their experimental material, but how often are multivariate statistical methods used to analyse the experimental data?

4.4

CONCLUSION

It is apparent that correlations between the errors of measured variables have implications to the statistical analysis and interpretation of nutrition experiments. The statistical methods developed in the course of analysing the experimental data in the present thesis have been outlined because it is considered that the analyses are more appropriate than those normally used.

The statistical methods outlined in this chapter are not new, but have been rarely used by animal nutritionists. In contrast, scientists in the Social Sciences have been aware for some time, of for example, the methods of and the advantages that multivariate analysis offers when several measurements are made on the one individual (see texts by Cooley and Lohnes, 1971; and Bock, 1975).

CHAPTER FIVE

GENERAL DISCUSSION

5.1

INTRODUCTION

The main objective of the work reported in this thesis was to determine the factors associated with the increases in milk fat production resulting from genetic selection in a grazing environment. Some aspects of the experimental results have been discussed already in the preceding chapters. The purposes of this chapter are:-

- to summarise and synthesise the results of the present experiments, which have compared the performance of high and low breeding index cows when fed individually in stalls on cut pasture, and when grazed as one group,
- to compare the results with other work reviewed in Chapter one and also with the work being done concurrently at Ruakura, with Jersey cows,
- to make some preliminary predictions about the effect of genotype on farm productivity,
- to identify the gaps that exist in the knowledge about genotype in relation to farm productivity and to outline the direction of future work.

The implications of correlated errors on the analysis and interpretation of nutrition experiments were covered in detail in Chapter four, and therefore will not be discussed here.

5.2

SUMMARY AND SYNTHESIS OF THE EXPERIMENTAL RESULTS

Although the main results have been discussed and summarised earlier in the thesis, no attempt has been made to synthesise the results of the stall feeding and grazing experiments reported in Chapter two, and the energy and nitrogen balance

experiments reported in Chapter three. A brief summary of the main results in each Chapter are given and the results are discussed jointly.

5.2.1 Chapter two : Performance of H and L cows when grazed and fed individually in stalls on cut pasture.

- (a) High cows consistently produced more milk fat than the L cows in the short-term experiments, and over the whole lactation; there was close agreement between the expected differences (based on breeding indexes) and the measured differences in milk fat yield.
- (b) The two genotypes had similar intakes of fresh cut pasture offered ad libitum in stalls. However H cows ate, on average, 7% more pasture per unit metabolic liveweight than low BI cows, but the differences between genotypes in intake were significant only in two of the four indoor feeding experiments ($P < 0.05$, $P < 0.10$).
- (c) Although differences between genotypes in their liveweight changes could not be shown to be significant in the short-term feeding experiments, over the whole lactation L cows appeared to gain more liveweight than H cows.
- (d) The differences between genotypes in either intake or the partitioning of energy between milk production and liveweight change were small. They did, however, explain 95% of the observed difference in milk energy output between the genotypes. This was assuming that maintenance energy requirements and net efficiencies of milk production and tissue deposition were the same for both genotypes.

- (e) The difference between genotypes in size was not regarded as significant because although there was a difference between genotypes in size of the animals purchased in the first year, in the second year the genotypes were similar in size.

5.2.2 Chapter three : Energy and nitrogen balance experiments with lactating and non-lactating H and L breeding index cows.

- (a) Differences between genotypes in their ability to metabolise feed energy were small and non-significant.
- (b) High BI cows had higher metabolisable energy intakes per unit metabolic liveweight than L cows.
- (c) Differences in heat production between genotypes were small and non-significant except in one case when H cows had a lower heat production than L cows during restricted feeding in early lactation. This anomalous result was discussed in Section 3.4.1.1. and will not be discussed further.
- (d) High BI cows partitioned a greater proportion of their metabolised energy to milk and a lesser proportion to body tissue than the L cows; the differences between genotypes were significant ($P < 0.05$, $P < 0.10$) in late lactation, but not in early lactation ($P > 0.10$, $P > 0.10$). There was an anomalous result during restricted feeding in early lactation when H cows lost less energy as heat, retained slightly (7%) more milk energy and mobilised less tissue energy than the L cows.
- (e) The feed required to maintain body condition and promote a unit of gain of body condition during the dry period was similar for both H and L cows.

- (f) Differences in N balance between genotypes were small and inconsistent.

5.2.3 Overall conclusion.

Considering the results of all the experiments together, there is no doubt that the use of artificial breeding and genetic selection has increased the level of milk fat production per cow. The results from the different experiments are in agreement with each other in that the two main factors associated with the increases in milk fat production were:-

- (a) High BI cows ate more than L cows,
- (b) High BI cows partitioned a greater proportion of their metabolisable energy intake to milk, rather than to liveweight gain, than the L cows.

5.3 COMPARISON OF EXPERIMENTAL RESULTS WITH OTHER PUBLISHED WORK

5.3.1. Energy balance experiments.

Before the current series of experiments were initiated at Massey University and Ruakura Agricultural Research Centre, there had been no attempt to compare the energy balances of cows which were genetically high and low producers. A preliminary report on the calorimetry work done at Ruakura, which compared Jersey cows differing in breeding indexes, was given by Trigg and Parr (1981). Of interest in the Ruakura work, was the observation that H cows partitioned a small but significantly ($P < 0.05$) higher proportion of gross energy to digested energy than the L cows in early lactation, but no such effect was found in mid lactation. There was no effect of genotype on energy digestibility in the Massey studies (the experiments reported in this thesis), or in the experiment by Grieve et al. (1976).

There was general agreement between the Ruakura and Massey experiments, that the intake of metabolisable energy per unit metabolic liveweight was higher for the H versus L cows and that the efficiency of use of metabolisable energy for milk plus tissue (as measured by heat production) was similar for both genotypes.

Not as well defined is the effect of genotype on the partitioning of energy between milk and liveweight change. In the Massey experiments, at a fixed intake, H cows partitioned more of their metabolisable energy to the synthesis of milk and less to liveweight gain than the L cows. The exception was during restricted feeding in early lactation (see Section 3.4.1.1. for discussion) when L cows mobilised more tissue reserves, but produced less milk than H cows.

Before discussing the work done at Ruakura, it is necessary to point out two errors in the paper of Trigg and Parr (1981). Firstly in their Table 2, a negative sign (-) has been omitted for the difference in intercepts between genotypes for tissue energy (Trigg T.E., personal communication). It is suggested that the table should have been as shown on the following page (considering only that part of their Table 2 where ME i.e. metabolisable energy, was the independent variable).

Table 2 : Regression analysis of energy partitioning in early lactation $y = b x + C^1$
(remade from Trigg and Parr, 1981).

X	Y	b(⁺ se)	C _(H) ³	C _(H-L) ⁴	s.e.
ME	Heat E	0.324 ⁺ 0.057	502	3	20
	Bal ² E	0.675 ⁺ 0.058	-500	-3	20
	Milk E	0.193 ⁺ 0.119	436	42	40
	Tissue E	0.482 ⁺ 0.153	-936	-44 ⁵	42

1 KJ kg^{-0.75}

2 Milk and tissue

3 Intercept for high breeding index cows

4 Difference in intercepts between genotypes

5 This value was incorrectly labelled as +44 in the original paper (N.B. Milk E + Tissue E must equal Bal E).

The second error appears to be one of interpretation. Trigg and Parr showed in their paper that ME intake per unit metabolic liveweight was higher for H than L cows in early lactation. They also presented data relating to changes in tissue fat of cows fed ad libitum in early lactation (their Table 3), which showed that H cows lost less tissue fat (123g fat) than L cows (257g fat). Trigg and Parr stated: "Relative to LBI cows, the smaller tissue fat loss by HBI cows in early lactation does not agree with overseas data (Broster et al. 1969)."

However Broster fed his cows at fixed levels of feeding, whereas in Trigg's work the difference in tissue fat loss between H and L cows is confounded with the difference in intake. Table 2 (p.185), indicates that if ME intake was the same for both genotypes, then H cows lose more tissue fat than the L cows, in agreement with Broster's work.

It is worth reiterating (see Section 2.4) that the differences in liveweight change between genotypes, based on the measured changes over the whole lactation, would be very small over short periods of time, and extremely difficult to detect. Accepting this, the calorimetric balance studies at Massey and Ruakura indicate, at a common intake, that the H cows partition a greater proportion of metabolised energy to the synthesis of milk and a lesser proportion to liveweight gain than the L cows.

5.3.2. Experiments in which animals were fed individually in stalls or grazed.

5.3.2.1 Milk fat production and changes in liveweight and condition score over the lactation.

Results from Ruakura (Bryant, 1981; Bryant and Trigg, 1981) and Massey are in agreement in showing that H cows produce more milk fat and gain less liveweight and condition over the lactation than L cows. It is interesting to note that a similar conclusion was reached from genetic selection experiments in which the animals were fed according to yield (Hooven et al. 1968; Miller et al. 1972).

However this is not surprising when it is considered that under both systems of feeding the animals of high genetic merit ate more than animals of low genetic merit.

The magnitude of the difference between genotypes in milk fat yield (kg/day) as lactation progressed has been observed to increase (Bryant and Trigg, 1981; 1979/80 lactation, this thesis), decrease (1980/81 lactation, this thesis) or remain constant (Carter, 1964). However none of the researchers have been able to detect significant changes in the magnitude of the difference in milk fat yield as lactation progresses. Therefore until evidence is produced to the contrary it must be assumed that the difference between genotypes in milk fat yield remains constant throughout lactation.

5.3.2.2. Marginal and gross efficiency of milk fat production.

Marginal efficiency i.e. the increase in milk production per unit increase in feed energy is considered first.

Cows which are grazed are invariably underfed at some stage of their lactation because of seasonal and day to day fluctuations in pasture growth. When the supply of pasture improves it is important that animals respond by increasing their milk yields. Many experiments (see Section 1.3.2.2. for references) have shown that when the feeding level is increased, high producing cows increase their milk yield to a greater extent than do low producing cows, but critical evidence for the grazing animal is sparse.

In the present thesis, H cows had greater marginal efficiencies (estimated both within and between cows) than the L cows, but the differences between genotypes were not significant ($P > 0.05$). Bryant (1981) also found that marginal efficiencies did not differ between genotypes when marginal efficiencies were estimated on a within cow basis where each cow was subjected to two levels of feeding.

Therefore at this stage it is concluded that when the feeding level is increased the extra milk fat produced will be similar for cows differing in genotype. However experiments to date have been confined to animals fed cut pasture in individual stalls and more evidence is required for the grazing animal.

Gross efficiency is defined as the milk produced per unit of food eaten. The average gross efficiencies of the H cows were almost invariably higher than those of the L cows in both the Massey and Ruakura experiments.

Another measure of gross efficiency is to compare, at a given intake, the amount of milk fat produced per unit of intake. Gross efficiency measured in this way was significantly greater ($P < 0.05$) for the H cows than for the L cows for the five experiments reported in Chapter two, and in the late lactation phase of the calorimetric balance experiment reported in Chapter three. The reason for the higher gross efficiency of the H versus L cows, at a fixed intake, appears to be associated with the fact that the L cows partition more of their metabolisable energy to liveweight gain than to milk production, whereas with the H cows the situation is reversed. This explanation was supported by calculations done in Section 2.4, which showed that the measured differences in intake and energy partitioning could explain 95% of the differences in milk energy output between genotypes.

A similar conclusion can be reached by examining the energy balance data for H and L cows in late lactation (Chapter three). If the critical significance level is set at $P = 0.10$ then difference between genotypes in

intercepts for both milk energy and tissue energy are significant. The estimated regression equations are:-

High BI

$$\begin{aligned} \text{Milk energy} &= 0.115 + 0.157 \text{ MEI} \\ (\text{MJ kg}^{-0.75} \text{ cow}^{-1} \text{ day}^{-1}) & \\ \text{Tissue energy} &= -0.530 + 0.497 \text{ MEI} \\ (\text{MJ kg}^{-0.75} \text{ cow}^{-1} \text{ day}^{-1}) & \end{aligned}$$

Low BI

$$\begin{aligned} \text{Milk energy} &= 0.007 + 0.157 \text{ MEI} \\ \text{Tissue energy} &= -0.409 + 0.497 \text{ MEI} \end{aligned}$$

where MEI = predicted metabolisable energy intake.

From the above equations the following data can be derived for H and L cows with a metabolisable energy intake of $1.4 \text{ MJ kg}^{-0.75} \text{ cow}^{-1} \text{ day}^{-1}$:-

	H	L
Milk energy	0.335 *	0.227
Tissue energy	0.166.	0.287
Milk + tissue energy (Total energy balance)	<u>0.501</u>	<u>0.514</u>

* units are $\text{MJ kg}^{-0.75} \text{ cow}^{-1} \text{ day}^{-1}$

The data show that although the total energy balance (milk + tissue energy) is similar for both genotypes, the gross efficiency of the H cows

$$\left(\frac{0.335}{1.4} = 0.239 \text{ MJ milk per MJ ME} \right)$$

is higher than that of the L cows

$$\left(\frac{0.226}{1.4} = 0.161 \text{ MJ milk per MJ ME} \right)$$

and this is because the L cows partitioned less of their

metabolisable energy to milk and more to liveweight gain than the H cows.

5.4 MECHANISMS OF INCREASED PRODUCTION.

The primary objective of this thesis was to determine how H cows produce more milk than L cows. The experiments carried out made it possible to compare genotypes with respect to the following items:-

- pasture intake
- the ability to metabolise feed energy
- the efficiency of use of metabolisable energy
- the proportions of metabolisable energy partitioned to the synthesis of milk and to liveweight gain.

The evidence in this thesis which is also supported by the results obtained at Ruakura, suggests that differences between genotypes in their ability to metabolise feed energy and the efficiency of use of metabolisable energy are small. The higher milk production of the H cows can be explained partly by their higher intake and partly by their ability to partition a greater proportion of their metabolisable energy to the synthesis of milk.

Why the H cows eat more and partition their metabolisable energy more favourably to milk production than the L cows is less clear? Forbes (1980) reiterated the concept that food is eaten in order to preserve an equilibrium between energy flow into and out of the body. In support of this theory Bryant (1981) noted that differences between genotypes in intake that occurred during lactation were not present during the dry period. Factors associated with lactation may therefore be responsible for the differences between

genotypes in intake. Whether these factors arise from the mammary gland itself, digestion end-products or hormonal status is unclear at this stage.

One aspect of the current work at Massey University is the measurement of the concentrations of blood metabolites and hormones of cows differing in breeding index. It is apparent from measurements made so far that during lactation H cows have a higher concentration of growth hormone in their blood than L cows. This is particularly interesting because Peel (1982) has shown that cows receiving daily injections (51 IU/day) of bovine growth hormone increase their milk fat yield by 20 to 40% with a small, non-significant decrease in feed intake. In addition energy balance measurements made by indirect calorimetry indicated that the extra milk energy produced had come from the mobilisation of body tissue. Therefore it is possible that differences between genotypes in growth hormone metabolism could explain why H cows have a greater ability to direct nutrients to the synthesis of milk rather than body tissue. Bauman and Currie (1980) have pointed out that variation in regulation of nutrient partitioning could involve inherited differences in circulating hormones, in numbers of hormone receptors in a target tissue and in the synthesis/degradation of regulatory enzymes.

5.5 IMPLICATIONS OF RESULTS ON FARM PRODUCTIVITY

The evidence in this thesis can be used to make some preliminary predictions about the likely effect of genetic merit on farm productivity. The calculations outlined below ignore confidence limits and assume that all figures are precise, hence can only be regarded as speculative.

5.5.1 Efficiency of feed conversion to milk fat during lactation.

During the 1979/80 lactation the H and L cows were fed indoors at three stages of lactation and pasture intakes were measured for a total of 12 weeks. Using the gross efficiencies of feed conversion (kg DM/kg milk fat) measured at these three times, the average gross efficiencies over the whole lactation were estimated to be 23.2 and 28.7 kg DM/kg milk fat for H and L cows respectively (see Appendix 5.1 for details).

The average per cow lactation (1979/80) productions of 150 and 117 kg for the H and L cows were below the five year average lactation milk fat production of 152 kg for all cows that were herd-tested in New Zealand during the period from 1975/76 to 1979/80 (New Zealand Dairy Board, 1979). There were several reasons why the productions were lower than might have been expected. Firstly the cows involved were only three-year-old animals. The average milk fat production of all pedigree Friesian cows was 12 kg higher than that of three-year-old cows (New Zealand Dairy Board, 1979). If such an age correction was applied then the adjusted production of the H cows (162 kg) would be greater than the average milk fat production of 152 kg for all cows herd-tested in New Zealand between 1975/76 and 1979/80. However no corrections to per cow productions have been made because as will be shown later any such corrections make little difference either to the main comparison between genotypes or to the absolute production per hectare.

Secondly the H and L cows were subjected to severe underfeeding during the first four weeks of lactation and in addition half of the cows of each genotype were deliberately underfed for a total of 10 weeks during experimental periods. Since the gross efficiencies of feed conversion to be used in subsequent calculations were based on cows fed ad libitum in stalls it was necessary to adjust the productions of those cows that were underfed in stalls. The production added to that of cows underfed in stalls was calculated as the sum of the production foregone during underfeeding and subsequently until daily production recovered to that of those cows previously fed ad libitum indoors. The total loss in production from underfeeding in stalls was 5 and 3 kg milk fat per cow for Experiments 2 and 3 respectively. Therefore the lactation yields adjusted for the underfeeding in stalls were 154 and 121 kg per cow for H and L cows respectively.

The feed required over the whole lactation is calculated as follows:-

	H	L
Milk fat produced (kg)	154	121
Kg DM/kg milk fat	23.2	28.7
Total feed required (kg DM)	3573	3473

The average lactation length was 249 days which is similar to the average lactation length of 251 days for all cows herd-tested in New Zealand between 1975/76 and 1979/80.

The estimated feed intakes for the whole lactation indicate that H cows ate approximately 2.9% more feed per cow than the L cows. This difference in intake although small was

unexpected because over the short-term feeding experiments there was little difference between genotypes in intake.

5.5.2 Efficiency of feed conversion to milk fat over 12 months.

Measurements made during May and June 1980 indicated that it required an average of 5.6 kg DM cow⁻¹day⁻¹ to maintain body condition (Carruthers, 1980). The feed required to promote a gain of one unit of condition can be estimated, from data given in Section 3.4.1.2, to be 2288 MJ metabolisable energy (approximately 200 kg DM; assuming a metabolisable energy concentration of the feed of 11.3 MJ ME/kg DM).

The body condition of the cows, which was on average 4.7 for both genotypes at calving, differed markedly by the end of lactation (H = 3.9, L = 4.8; see Table 2.2). Unless the H cows were fed at a higher level than the L cows during the dry period, it is likely that their production during the subsequent lactation would suffer. To avoid this, extra feed must be given to the H cows. The feed required to bring the body condition of both groups to a condition score of five over the 116 day dry period can be calculated as follows:-

	<u>Feed Requirements</u>	
	(kg DM)	
	H	L
Feed for maintenance of body condition		
Early pregnancy	280 (50 days x 5.6)	280
Late pregnancy*	442 (66 days x 6.7)	442
Feed required for condition score gain	220 (1.1 x 200)	40 (0.2 x 200)
Total Feed required	942	762

*The increase in feed requirements allows for calf development.

The feed intakes over the 12 months are the sum of the feed intakes over lactation and the dry period, namely 4515 (3573 + 942) and 4235 kg DM (3473 + 762) for the H and L cows respectively.

5.5.3 Predicted performance of high and low breeding index cows on a hypothetical farm.

On the assumption that 16,000 kg pasture DM are harvested annually the production per hectare can be estimated as:-

	H	L
Total feed eaten (kg DM)	16000	16000
Feed (kg DM cow ⁻¹)	4515	4235
Stocking rate (cows ha ⁻¹)	3.54	3.78
Milk fat (kg cow ⁻¹)	154	121
Milk fat (kg ha ⁻¹)	545	457

The calculations indicate that the H cows produce 19% more milk fat per hectare than L cows.

Therefore based on a difference of 27 breeding index units the observed difference in per cow milk fat production was 28%, however the actual advantage to the H cows in terms of milk fat per hectare was reduced to a difference of 19%. If the productions were adjusted for age as outlined previously, then the per cow productions for H and L cows would be 166 and 133 kg milk fat and milk fat per hectare could be calculated to be 552 and 464 kg, a 19% increase, indicating that a correction for age would make little difference to the calculation.

It is emphasized that the effect of breeding index on production per hectare is only speculative at this stage, but the preceding calculations give some indication of the likely effects. Similar calculations with Jersey cows differing in breeding index indicated that for a difference of 21 breeding index units there would be a difference in terms of milk fat per hectare of 15% (Bryant, 1981).

5.6 FUTURE RESEARCH

The major contribution of the work reported in this thesis has been to confirm that the differences measured in milk fat production agree with estimates from BI, and to identify the mechanisms whereby cows of high genetic merit produce more milk fat than cows of low genetic merit. However, the present work can be regarded as only a first step in the process of determining the effects of improving the genetic merit of cows on farm productivity. Obviously there are many questions that remain to be answered, but if the primary objective is to determine the effect of genotype on farm productivity, then what is the best way of achieving this objective?

Outlined below is what is considered to be an appropriate method of achieving the objective outlined above.

There are three specific gaps that exist in the knowledge at present which require clarification. The response of cows differing in BI, to different levels of condition at calving needs to be defined. This is particularly important because H cows are lighter in condition than L cows at the end of lactation, and it is not known at present what the consequences are, of calving H cows in lighter condition than L cows.

Most of the feeding experiments that have been done so far have been confined to early and mid lactation. More precise definition of the responses of cows differing in BI to different levels of feeding in late lactation are required.

Of importance also is the grazing efficiency of cows differing in breeding index. Grazing efficiency or degree of defoliation is defined as the herbage consumed at each defoliation expressed as a proportion of the herbage mass originally present (Hodgson, 1979). Because H cows achieve a proportion of their extra milk fat by eating more than L cows, it is necessary to identify the situations which reduce the difference between genotypes in their intakes and as a result reduce the advantage in milk fat production of the H cows. Limited evidence obtained so far (see p.85) suggests that competition and grazing components are not important factors accounting for differences between genotypes. However the responses of H and L cows to a range of herbage allowances when grazed together and separately needs to be defined.

These then are the three specific gaps that exist in the knowledge at present. Research is currently under way at both Ruakura and Massey which aims to provide information on these aspects.

The next step in achieving the stated objective is to attempt some form of "systems approach". But before going on it is appropriate to ask:

"Can the objective be achieved simply by running farmlet trials?"

A major criticism of farmlet trials is that the results are only applicable to the environment and years in which the work was carried out. For example, the farmlet trials of Gray and Matthews (1982) and Bryant and Cook (1980), examining different wintering systems for dairy cows, have produced opposite results.

There are no clear reasons as to why the results differ. Will the same confusion exist after farmlet work to define the effect of genotype on farm productivity has been carried out?

A systems approach is defined as the application of the scientific method to the study of complex systems (Wright, 1980). It is not intended to go into any detail about systems research, except to point out that the major features are; problem orientation, an interdisciplinary approach and the use of mathematical methods.

Whilst the benefits of a systems approach remain largely unproven as yet, it is suggested that the next step in achieving the stated objective is to develop a mathematical model of the system based on the information available at present, and to attempt to validate and where necessary, refine the model using data from farmlet experiments run concurrently. In the short term the model would probably be of most use in identifying the most sensitive

components of the system, but in the longer term, the refined model may have much wider applicability than the results of farmlet experiments run on their own.

Studies of blood metabolites and hormones can and are being run concurrently with the nutrition experiments. The major potential benefits of these detailed studies were outlined in Section 1.5. Also of importance, but secondary to the primary objective, is the adequate definition of the response of cows differing in breeding index to protein supplements (see Section 1.5.1). Other aspects such as the milking characteristics of high and low breeding index cows are currently being studied at Massey and Ruakura.

The two genotypes studies in this thesis have provided a useful framework to gain a greater understanding of the nutritional and physiological characteristics of the dairy cow.

APPENDICES

APPENDIX 2.1 DETAILS OF THE COWS PURCHASED FOR
THE EXPERIMENTS REPORTED IN THIS
THESIS, AND THE METHOD OF
CALCULATING BREEDING INDEXES.

Appendix 2.1.1 Details of cows purchased for use
in experiments.

Suitable cows were initially identified by the Farm Production Division of the New Zealand Dairy Board. The selection criteria were:-

- each cow to have fully identified ancestry information for the three preceding generations,
- the cows were to be Friesian, with no more than 1/8 Jersey blood,
- the cows, when purchased, had to be approaching their second calving with an expected calving date in August or early September,
- breeding indexes had to be high (> 120) or low (< 110),

Potentially useful experimental animals, as determined by the above criteria, were inspected for overall soundness (including pregnancy), and, if found suitable, were purchased.

Details of the cows purchased in 1979 and 1980 are given below. The values for breeding and production indexes presented are those calculated by the New Zealand Dairy Board after the completion of the first lactation.

Cows purchased in 1979

Cow	Breeding index	Production index	Birth Date
<u>High BI</u>			
154	126	123	6.8.76
156	125	125	22.7.76
159	124	108	4.9.76
168 *	121	101	8.8.76
176	133	136	17.8.76
178	125	124	1.9.76
180	131	149	13.8.76
182	127	133	21.7.76
191	126	127	11.8.76
193	133	127	26.7.76
195	124	107	1.10.76
<u>Low BI</u>			
151	101	100	12.9.76
153	100	84	27.7.76
158	97	78	22.4.76
160	101	91	23.4.76
166	99	75	3.8.76
181	103	96	30.6.76
186	104	110	17.7.76
187	99	93	5.8.76
188	101	83	31.7.76
189	96	85	12.8.76

* Not used in experiments during 1979/80.

Cows purchased in 1980

Cow	Breeding index	Production index	Birth Date
<u>High BI</u>			
149	124	114	9.8.77
150	128	123	25.7.77
163	123	117	17.8.77
169	134	138	28.7.77
171	127	118	12.8.77
183	121	116	15.9.77
190	123	118	5.8.77
196	125	121	20.8.77
200	126	128	3.8.77
207	126	133	29.7.77
<u>Low BI</u>			
170	104	114	14.6.77
201	100	105	10.8.77
202	107	105	7.8.77
203	106	98	24.7.77
204	102	98	9.7.77
205	107	107	20.8.77
206	102	112	24.8.77
209	99	106	26.7.77
210	104	113	14.8.77

Appendix 2.1.2 Calculation of breeding indexes.

The method of calculating breeding indexes has been outlined in full in a paper presented at an International Symposium on Sire and Cow Evaluation held in Warsaw, Poland, 1980 (Wickham and Stichbury, 1980).

The formula for calculating breeding indexes of cows is:-

$$BI = BI_{ANC} + \beta (LPI - BI_{ANC})$$

where BI = breeding index,
 BI_{ANC} = breeding index based on ancestry calculated as the average of the indexes for its sire and dam,
 β = regression coefficient (explained in more detail below),
 LPI = cow's lifetime production index (see below for further detail).

The regression coefficient (β) is estimated from the regression of a cow's BI (non-ancestry) on the difference between her actual LPI and her expected LPI, based on ancestry BI. The formula for β is:-

$$\beta = \frac{25N}{25N + \frac{35N + 40}{1 - R}}$$

Where N = number of lactations included in LPI
 R = reliability of the ancestry information ($\frac{1}{4}$ of the sire's reliability plus $\frac{1}{4}$ of the dam's reliability)

The assumptions inherent in the formula for β given above, are that the heritability (h^2) of milk fat production is 0.25 and that the repeatability of lactation records by the same animal is 0.6.

The LPI is the average production index (PI) of all lactations up to and including the ninth lactation. The PI is based on herd test results and provides an estimate of the productive merit of a cow within a herd, after taking into account the age and stage of lactation of the cow.

An example of the calculation of a breeding index is given below.

Example of BI calculation

Information for cow 182

BI_{sire} = 131 ; reliability = 0.76

BI_{dam} = 121 ; reliability = 0.53

LPI = 133 (based on one lactation, therefore N = 1)

The formula is:-

$$\begin{aligned}
 BI &= BI_{ANC} + \beta (LPI - BI_{ANC}) \\
 &= \frac{1}{2}(131 + 121) + \left[\frac{25}{25 + \frac{(35+40)}{1-0.25(0.76+0.53)}} \right] [(133-126)] \\
 &= 126 + 0.184 \times 7 \\
 &= 126 + 1 \\
 &= \underline{\underline{127}}
 \end{aligned}$$

APPENDIX 2.2 EXAMPLES OF THE STATISTICAL
ANALYSES USED IN CHAPTER 2

Appendix 2.2.1 Split-plot analysis examining
differences in faecal outputs
between genotypes and periods
(0-5 and 6-10 days of collection).

The split-plot analysis has been done since it uses the appropriate errors for testing differences in faecal output between periods and the BI x period interaction (within-cow error) and genotypes (between-cow error). The analysis of variance table is given below:-

Source of variation	df.	SS	MS	F
<u>Main plot</u>				
Breeding index	1	0.016017	0.016017	0.04
Error	10	3.622132	0.362213	
<u>Sub-plot</u>				
Period	1	0.106667	0.106667	0.85
Breeding index x				
Period	1	0.000267	0.000267	0.00
Error	10	1.249166	0.124917	

Appendix 2.2.2 Stepwise regression analysis testing differences between genotypes in the relation between metabolisable energy intake and output of energy in milk.

The order of fitting of variables was metabolisable energy intake (MEI) followed by genotypes (BI), then the interaction BI X MEI is fitted which tests whether there is a difference in slopes between BI groups. The analysis of variance table is presented below.

Source of variation	df.	SS	MS	F	Prob-ability
MEI	1	0.078784	0.078784	59.9	< 0.01
BI	1	0.015517	0.015517	11.8	< 0.01
BI X MEI	1	0.000920	0.000920	0.7	0.42
Error	16	0.021055	0.001316		
Total	19	0.116275			

All the regression equations presented in Table 2.13 have been analysed in the same way as outlined above.

Appendix 2.2.3 Multivariate analysis of differences in milk fat yield between genotypes, before, during, and after underfeeding on a within-cow basis.

The multivariate analysis is appropriate because repeated measurements have been made on the same cow at different 'times', hence it is likely that there is a correlation between the errors of the repeated measurements. Multivariate analysis takes into account the correlation between the errors hence the tests of significance are valid.

Notes for the analysis of variance table shown overleaf are presented below.

- † Test of average difference between genotypes before, during, and after underfeeding (considered as three 'times').
- φ The contrasts test whether the difference in fat yield between genotypes changes; five weeks before, versus during underfeeding (contrast 1) and during underfeeding versus five weeks later (contrast 2).
- * Times X BI jointly tests whether differences in fat yield between genotypes changes at the three 'times'.

Source of variation		df.	Generalised SS	Lambda	df. Chi	χ^2	Prob-ability
<u>Experiment 2</u>							
BI	†	1	16.667	0.5195	1	4.91	0.03
Contrast 1	ϕ	1	2.158	0.6997	1	2.68	0.10
Contrast 2	ϕ	1	3.562	0.6491	1	3.24	0.07
Times X BI	*	1	395.803	0.6135	2	3.42	0.18
<u>Error</u>							
BI		8	8.659				
Contrast 1		8	1.510				
Contrast 2		8	2.312				
Times		8	242.816				
<u>Experiment 3</u>							
BI		1	26.493	0.5647	1	4.29	0.04
Contrast 1		1	0.964	0.7714	1	1.95	0.16
Contrast 2		1	3.808	0.8818	1	0.94	0.33
Times X BI		1	213.274	0.7694	2	1.84	0.40
<u>Error</u>							
BI		8	14.961				
Contrast 1		8	0.744				
Contrast 2		8	3.358				
Times		8	164.088				
<u>Experiment 4</u>							
BI		1	7.100	0.6841	1	2.47	0.12
Contrast 1		1	5.684	0.9976	1	0.02	0.90
Contrast 2		1	0.624	0.8888	1	0.77	0.38
Times X BI		1	308.646	0.8829	2	0.75	0.69
<u>Error</u>							
BI		7	4.857				
Contrast 1		7	5.671				
Contrast 2		7	0.555				
Times		7	272.490				

APPENDIX 2.3 CALCULATION OF THE EFFECT OF SOIL
INGESTION ON ESTIMATION OF INTAKE
FOR EXPERIMENT 3.

During the last 20 days of Experiment 3, all cows (20 in total) were dosed twice daily with 10g chromium sesquioxide. Over the last ten days of the chromium dosing period, faeces were randomly sampled from the faeces immediately behind each cow in the feed barn, twice daily at 0800 and 1600 hours, and bulked over the 10 day period. Chromium concentration of the faeces was determined as described previously (see Materials and Methods, Chapter 2).

To estimate the recovery of chromium in the faeces, total collection of faeces was done for two cows and the predicted chromium concentration in the faeces, based on the measured faecal output, was compared to the measured chromium concentration of the faeces. When the measured concentration of chromium was divided by predicted chromium concentration in faeces, the agreement was 0.8834. As recovery of chromium was incomplete the faecal outputs of the remaining cows were adjusted on the basis of this ratio.

To estimate soil content of the faeces the following procedure was used, based on the method of Healey and Ludwig, (1965).

The faecal samples were dried at 100degC, weighed and ashed at 550degC for four hours. The ash was placed on a weighed filter paper and washed with 3N HCl, followed by distilled water. The filter paper plus residue was oven dried and then weighed to determine the amount of soil present. The soil percentage was

then calculated as a percentage of the original dried sample of faeces. Alternatively the soil content of the faeces could have been estimated from the measured ash content of the faeces according to the equation of Healey (1968):-

$$Y = 1.83 X - 33.6$$

Where Y = soil content of faeces

X = ash content of faeces.

The average ash content of the faeces of the 20 cows was 30.3%. Use of the equation above gave a soil content in the faeces of 21.8% and this is similar to the value of 23.0% estimated by using the procedure described above.

On ignition soil undergoes a loss in weight owing to oxidation of organic matter and removal of bound water. The LOI (loss on ignition) in the present experiment would have been only 3-4%, based on the data of Healey (1969), hence has been ignored.

A sample calculation of the adjustment of intake for soil ingestion is given below.

Data for cow 151

Dry matter intake (uncorrected)	=	11.20 kgDM cow ⁻¹ day ⁻¹
Faecal output		
(based on 88.34% recovery)	=	4.30 kgDM cow ⁻¹ day ⁻¹
Soil content of faeces	=	26.18%

Calculation

The soil (kg) present in faeces	=	4.30 x 26.18%
	=	1.13kg

therefore the corrected intake and faecal output are 10.07kg pasture DM and 3.17kg faecal DM.

Intakes for all the cows were adjusted in this way and the average metabolisable energy intake was re-calculated to be

1.439 MJMEkg^{-0.75} cow⁻¹ day⁻¹ (the uncorrected value was 1.602 MJMEkg^{-0.75} cow⁻¹ day⁻¹).

The observed energy retention (average of all cows) was 0.410 , comprising 0.389 milk energy and 0.021 tissue energy. Assuming a k value of 0.65 and a maintenance value of 0.6 MJkg^{-0.75} (Holmes et al. 1981) the estimated MEI would be 1.231 MJkg^{-0.75} cow⁻¹ day⁻¹. The ratio of measured to predicted MEI would be

$\frac{1.439}{1.231} = 1.17$ The ratio, using intakes uncorrected for soil contamination was

$\frac{1.602}{1.231} = 1.30.$

The intakes corrected for soil ingestion agree more closely with predicted MEI based on theoretical estimates (Holmes et al. 1981).

The soil content of the faeces was similar for H and L cows, 22.7 and 23.2% respectively, and greater, as expected, on the low level of feeding; 20.8 and 25.2% for high and low levels of feeding.

APPENDIX 3.1 GAS LIQUID CHROMATOGRAPHY OF FATTY ACIDS
OF MILK FAT

Appendix 3.1.1 Analysis of fatty acids.

Fatty acids were analysed as their methyl esters using an Aerograph Varian 1200 Gas Chromatograph fitted with a hydrogen flame ionisation detector. The stainless steel column (200cm x 0.2cm i.d.) was packed with 15.86% diethylene glycol succinate polyester (DEG's) on Chromosorb WHP (60-80 mesh). The "furnel coating method" (McNair and Bonelli, 1968) was used for preparation of the column packing. Column conditioning was carried out from 50-200degC over a period of five days with N₂ flow of 10ml/min.

Samples were injected on to the column at an initial column temperature of 50degC, then the oven temperature was programmed to increase at a rate of 6degC/min to a final temperature of 200degC, at which temperature the oven was held until the last of the sample had passed through the column. The mobile gas phase was N₂, flowing at 30ml/min. The detector was operated at 240degC using H₂ and air flows of 30 and 200ml/min respectively. The injector was held at 240degC.

Appendix 3.1.2 Preparation of fatty acid methyl esters.

25 μ l of transesterifying reagent (1.5ml 0.5M sodium methoxide in methanol, 6ml hexane and 2.5ml diethyl ether) was added to a 1-4mg sample of lipid in a 0.3ml reaction vial (Kontes Glass Co., U.S.A.) and immediately closed with a vapour-tight rubber seal. The vial was rotated gently for two minutes to mix the contents, 25 μ l hexane was added (by injection through the rubber seal) and the vial rotated gently for a further two minutes (Shehata et al. 1970). 1-8 μ l of the solvent mixture was used for analysis by g.l.c.

The transesterifying reagent was prepared fresh every two or three days from a stock solution of 0.5M sodium methoxide in methanol (prepared by dissolving 2.7gm sodium metal in 100ml absolute methanol) which was stored at 5degC in tubes fitted with teflon-lined screw caps.

Quantitative measurement of fatty acids by g.l.c.

Methyl esters of fatty acids were identified by comparison with standard methyl esters (from the Biochemistry Department, Massey University) chromatographed under identical conditions.

Proportions of the individual fatty acids were obtained by using a Varian Aerograph Digital Integrator 480.

APPENDIX 3.2 CALCULATION OF HEAT PRODUCTION FROM
RAW DATA

An example

Cow : 181 Restricted feeding

Date: 22/23 September, 1980

Ventilation rate

$$\begin{aligned} \text{Measured rate} &= 278.13 \text{ litres/min} \\ &= 278.13 \times 1440 \text{ (min/day)} \\ &= 400512.69 \text{ litres/day} \end{aligned}$$

Correction to STP

Air pressure 750.8mm

Dew temperature 13.6degC corresponds to 19.7mm vapour
pressure (measured at air cooler)

Temperature of air in gas meter room 21degC

$$\text{Correction factor} = \frac{750.8 - 19.7}{760} \times \frac{273}{273+21}$$

$$= 0.893$$

$$\begin{aligned} \text{Corrected ventilation rate} &= 400512.69 \times 0.893 \\ &= 357657.83 \text{ litres/day} \\ &\text{STP (dry gas)} \end{aligned}$$

Oxygen consumption

Reference gas	Known %	Recorder Chart divisions
A	19.308	80.1
B	20.798	18.3
Difference	1.490	61.8

$$\begin{aligned} \text{O}_2 \text{ per division} &= \frac{1.490}{61.8} \\ &= 0.0241\% \end{aligned}$$

Difference between exhaust gas and air in
for cow 181 (22-23/9) chart reading was 38.5 divisions.

$$\begin{aligned} \text{Therefore \% O}_2 &= 38.5 \times 0.0241\% \\ &= 0.928\% \text{ O}_2 \end{aligned}$$

$$\begin{aligned} \text{Volume O}_2 \text{ produced} &= 0.928\% \times 357657.83 \\ &= 3319.91 \text{ litres/day} \end{aligned}$$

CO₂ produced

Reference gas	Known %	Chart divisions
A	1.177	7.0
B	0.669	27.3
Fresh air (IN)		62.3

A - IN = 55.3 divisions B - IN = 35.0 divisions

Because the relation between the concentration of CO₂ and the electrical output of the analyser reading (no. of chart divisions) is not linear as in the case of O₂% and analyser, the % CO₂ from the exhaust gas must be calculated by interpolating from the regression of reference gases versus chart divisions (Figure A3.1). The 47.3 chart divisions difference for exhaust gas minus air in for cow 181 was equivalent to 0.975% CO₂.

$$\begin{aligned} \text{Volume CO}_2 \text{ produced} &= 0.975\% \times 357657.83 \\ &= \underline{3487.16 \text{ litres/day}} \end{aligned}$$

CH₄ produced

Reference gas	Known %	Chart divisions
A	0.152	31.8
B	0.056	61.0
Fresh air (IN)		77.3

A - IN = 45.5 divisions B - IN = 16.3 divisions

Interpolating from graph (Figure A 3.1) the 22.0 chart divisions difference between exhaust gas and air in
= 0.075% CH₄

$$\begin{aligned} \text{Volume CH}_4 \text{ produced} &= 0.075\% \times 357657.83 \\ &= \underline{268.24 \text{ litres/day}} \end{aligned}$$

Urinary nitrogen

Urinary nitrogen excreted = 191.60g/day

Heat production (Brouwer, 1965)

$$\begin{aligned} \text{HP} &= (\text{O}_2 \times 16.18) + (\text{CO}_2 \times 5.02) - (\text{CH}_4 \times 2.17) - (\text{N} \times 5.99) \\ &= (3319.91 \times 16.18) + (3487.16 \times 5.02) - (268.24 \times 2.17) - \\ &\quad (191.60 \times 5.99) \\ &= \underline{69.492 \text{ MJ/day}} \end{aligned}$$

* The value of 191.60g nitrogen is the average daily N excreted over the ten day collection period.

No. chart divisions

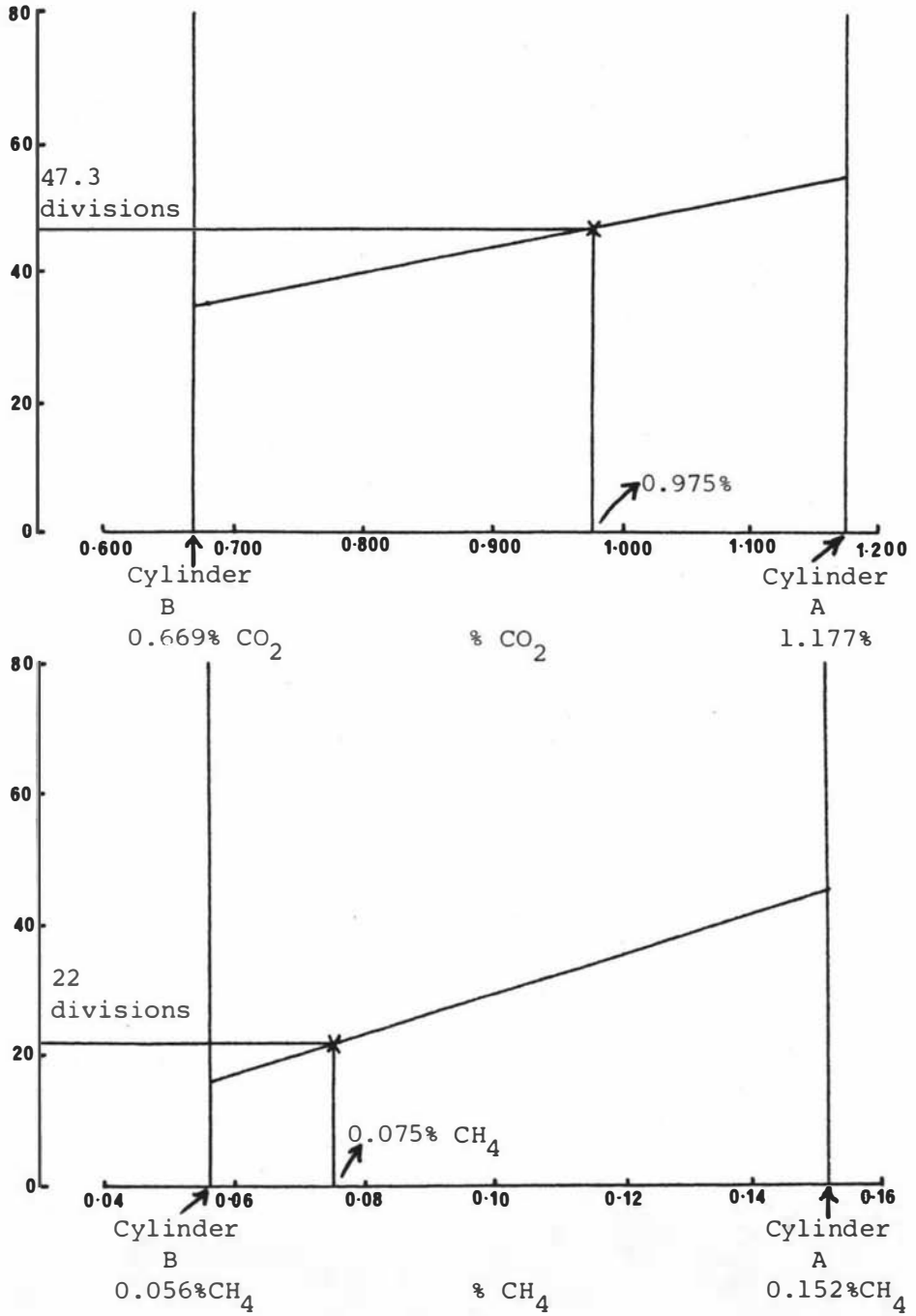


FIG.A3.1: An example of the method used to calculate the difference in CO₂% or CH₄% between the air in and the exhaust air from the calorimeter by interpolating from the relation of reference gases to the electrical output of the analyser reading (number of chart divisions).

APPENDIX 3.3 MEAN VALUES FOR GROSS ENERGY INTAKE, FAECAL, URINARY, METHANE, HEAT AND MILK ENERGIES FOR COLLECTION PERIODS DURING LACTATION AND THE DRY PERIOD FOR HIGH AND LOW BREEDING INDEX COWS.

Appendix 3.3.1 Collection periods during lactation

(a) Early lactation (energy units are MJcow⁻¹day⁻¹)

	<u>Level of Feeding</u>					
	<u>Ad libitum</u>		Restricted		<u>Ad libitum</u>	
	H	L	H	L	H	L
Gross energy intake	277.9	265.9	186.6	180.2	330.6	290.8
Faecal energy	70.6	62.7	41.5	38.8	85.9	73.0
Urinary energy	17.9	16.7	12.0	10.9	15.4	14.7
Methane energy	17.0	17.2	11.8	13.2	20.3	18.4
Heat energy	105.1	106.6	73.5	85.7	118.7	107.9
Milk energy	70.0	62.5	51.8	49.8	65.3	58.7
Liveweight (kg)	-	-	377	432	-	-
(measured during restricted feeding)						

(b) Late lactation (energy units are MJ cow⁻¹ day⁻¹)

	<u>Level of feeding</u>			
	<u>Ad libitum</u>		<u>Restricted</u>	
	H	L	H	L
Gross energy intake	281.5	261.2	194.5	176.1
Faecal energy	101.4	93.0	71.6	64.4
Urinary energy	10.4	11.2	7.7	7.5
Methane energy	16.8	16.8	12.3	12.6
Heat energy	91.2	91.9	73.0	75.5
Milk energy	34.8	22.0	26.3	15.8
Liveweight (kg) (measured during restricted feeding)	-	-	412	516

Appendix 3.3.2 Collection periods at two stages of pregnancy; approximately 210 and 230 days of pregnancy.

Appendix 3.3.2.1 At 210 days of pregnancy (energy units are MJ cow⁻¹ day⁻¹)

	<u>Level of feeding</u>			
	<u>2 x Maintenance</u>		<u>Maintenance</u>	
	H	L	H	L
Gross energy intake	173.1	178.1	93.0	91.6
Faecal energy	53.9	55.1	25.8	25.2
Urinary energy	7.3	7.4	4.3	4.6
Methane energy	10.6	10.9	6.3	6.9
Heat energy	87.9	87.1	62.8	56.3
Liveweight (kg)	427	476	410	450

Appendix 3.3.2.2 At 230 days of pregnancy (energy units are MJ cow⁻¹ day⁻¹)

	<u>Level of feeding</u>			
	<u>2 x Maintenance</u>		<u>Maintenance</u>	
	H	L	H	L
Gross energy intake	218.5	217.8	130.7	132.7
Faecal energy	55.9	55.5	36.0	31.2
Urinary energy	9.3	9.2	5.7	5.4
Methane energy	13.0	13.5	9.3	9.8
Heat energy	99.2	104.8	72.9	72.2
Liveweight (kg)	442	484	406	447

APPENDIX 3.4 AN EXAMPLE OF THE METHOD OF STATISTICAL ANALYSIS USED IN ANALYSING THE ENERGY BALANCE DATA.

The example is for early lactation with the feeding sequence ad libitum followed by restricted feeding. The independent variable is gross energy intake and the dependent variable is milk energy output.

Statistical analysis

Before proceeding with univariate analysis, possible correlations between errors of the measured variables must be examined.

1. Multivariate analysis: The dependent variables are FUM (faeces + urine + methane), heat, and milk, The independent variable is gross energy intake.

The test of independence of the errors was $\chi^2 = 0.399$ with 3 degrees of freedom. The probability of this χ^2 value is 0.94, hence the errors of the three variables are not correlated and univariate analysis is appropriate.

2. A different slope is fitted for each BI group and a different intercept for each cow. Interest here lies in whether there is a difference in slope between BI groups. This is the sub-plot part of the analysis and the appropriate error comes from within cows. The analysis of variance table is as follows.

Source of variation	df.	SS *	MS	F	Probability
GEI	1	0.027278	0.027278	4.54	< 0.01
BI X GEI	1	0.001129	0.001129	0.19	0.10
Cows	6	0.069648	0.011608	1.93	0.68
Error	4	0.024032	0.006008		
Total	12				

where GEI = gross energy intake
 BI = breeding index
 BI X GEI = term for fitting a different slope
 to each BI group
 cows = intercept terms for the six cows.

The probability that there is a different slope between BI groups is rejected as the BI X GEI term is only significant at the 0.10 level of probability. The data with a different slope fitted for each BI group is shown in Figure A3.4.1.

*sums of squares are adjusted for all other terms.

3. Since there is no significant difference in slopes between BI groups, a common or pooled slope is fitted (Figure A3.4.1) and intercepts predicted for each of the six cows.
4. A multivariate analysis must be done to check for correlations between errors of the predicted intercepts. The terms included are FUM, heat, and milk. The test of independence of the errors was a $\chi^2 = 0.467$ with three d.f. The probability of this χ^2 value is 0.93, hence the errors of the three variables are not correlated and univariate analysis is appropriate.

Milk energy
(MJkg^{-0.75}cow⁻¹day⁻¹)

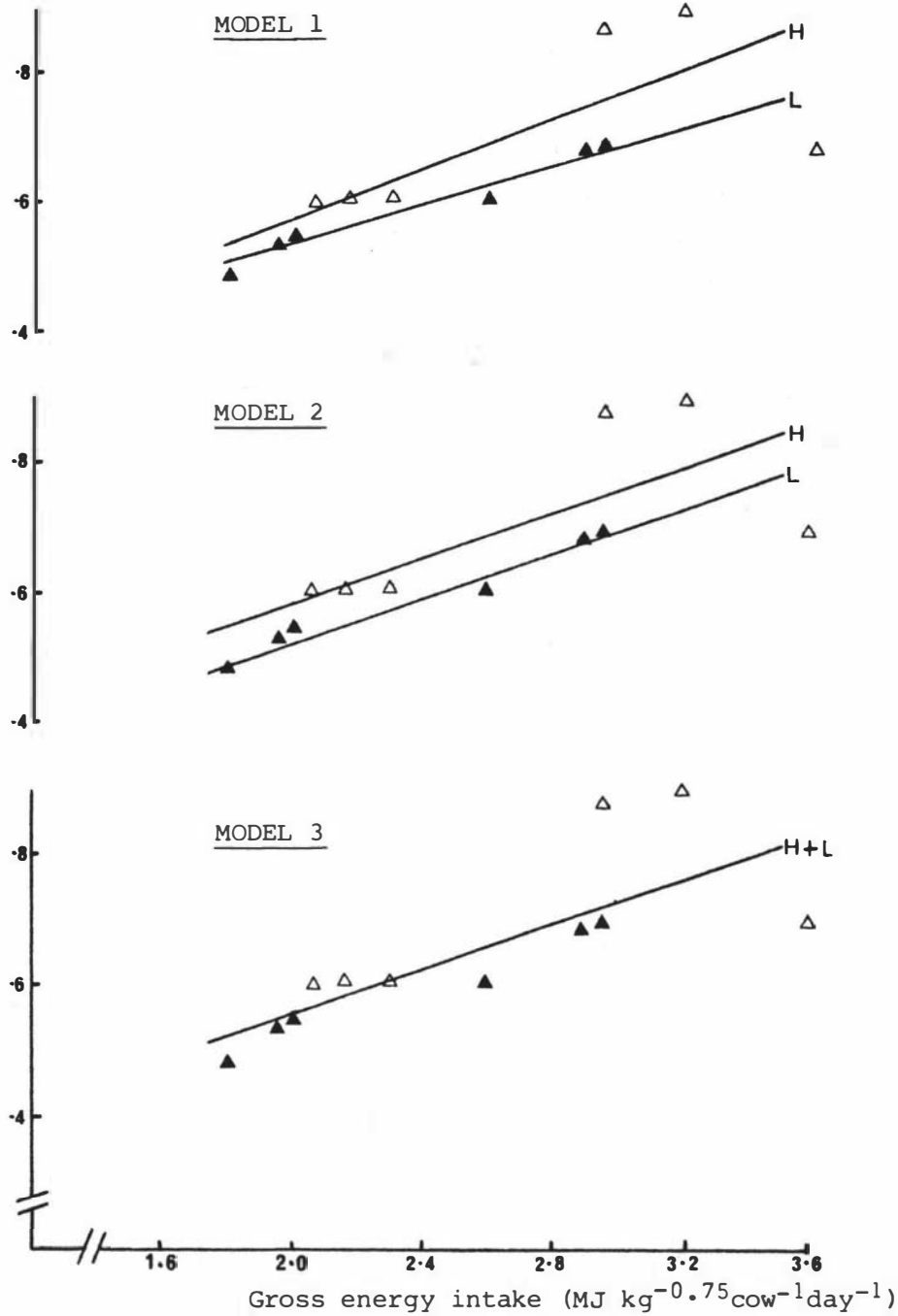


FIG.A3.4.1: Milk energy output versus gross energy intake for high (Δ) and low (▲) breeding index cows: Different slope for each BI group (Model 1), common slope, but different intercept for each BI group (Model 2), common slope and intercept for each BI group (Model 3).

5. The analysis of variance table for testing whether there are significant differences in intercepts between BI groups is given below.

Source of variation	df	SS	MS	F	Probability
BI	1	0.005197	0.005197	1.3	0.32
Error	4	0.016248	0.004062		
Total	5	0.021444			

The probability of differences in intercepts between BI groups is significant at the 0.32 level of probability.

6. Hence, as there are no differences between BI groups in either slopes or intercepts a pooled regression is fitted (Figure A 3.4.1).

The statistical method is perhaps most clearly summarised in Figure A 3.4.1, however the three main stages are summarised below.

Model 1: Different slope for each BI group.

$$\text{H Milk energy} = 0.204 + 0.187 \text{ GEI}$$

$$\text{L Milk energy} = 0.244 + 0.148 \text{ GEI}$$

Model 2: Common slope for each BI group, different intercept * for each BI group.

$$\text{H Milk energy} = 0.249 + 0.171 \text{ GEI}$$

$$\text{L Milk energy} = 0.190 + 0.171 \text{ GEI}$$

Model 3: Common slope and intercept for each BI group.

$$\text{H and L Milk energy} = 0.219 + 0.171 \text{ GEI}$$

* The intercept for each BI group is calculated as the average of the intercepts of the cows in each group.

APPENDIX 3.5 A COMPARISON OF THE RELATION BETWEEN ENERGY RETENTION (MILK PLUS BODY TISSUE) AND METABOLISABLE ENERGY INTAKE OBTAINED IN THE PRESENT STUDY WITH OTHER PUBLISHED DATA FROM EXPERIMENTS WHERE COWS WERE FED ON PASTURE.

The equations referred to in Chapter 3 relating total energy retention to metabolisable energy intake (MEI) are presented below with the predicted energy retentions at MEI's of 1.0 and 2.0 $\text{MJkg}^{-0.75} \text{cow}^{-1} \text{day}^{-1}$ for each equation.

Reference	Intercept (a)	Slope (k)	Energy retention at:	
			MEI=1.0	MEI=2.0
1. Trigg <i>et al.</i> (1980)	-0.335	0.571	0.236	0.807
2. Trigg and Parr (1981)				
	-0.502	0.675	0.173	0.848
3. Van Es (1975)	-0.485	0.720	0.235	0.955
4. Early lactation (<u>Ad libitum</u> → restricted)				
	-0.172	0.470	0.298	0.768
5. Early lactation (Restricted → <u>ad libitum</u>)				
	-0.268	0.544	0.276	0.820
6. Late lactation (<u>Ad libitum</u> → restricted)				
	-0.409	0.654	0.245	0.899

The predicted energy retentions at metabolisable energy intakes of 1.0 and 2.0 show considerable variations, but there is no consistent pattern in the data to suggest that one estimate is particularly different from the other estimates.

APPENDIX 3.6 DETAILS OF THE CALCULATIONS RELATING TO THE ENERGY REQUIRED TO MAINTAIN AND GAIN BODY CONDITION FOR DRY, PREGNANT, DAIRY COWS.

Appendix 3.6.1 Estimation of energy required to maintain and gain body condition for the 62 day indoor feeding period (12 cows).

Appendix 3.6.1.1 Maintenance of body condition.

Using the estimated equation:-

$$\Delta\text{CS kg}^{-0.75} = -2.050 + 2.711 \text{ MJMEkg}^{-0.75}$$

where

$$\Delta\text{CS kg}^{-0.75} = \text{change in condition score per (unit metabolic liveweight} \times 10^{-2}) \text{ over the 62 day feeding period,}$$

$$\text{MJMEkg}^{-0.75} = \text{metabolisable energy intake per unit metabolic liveweight day}^{-1}.$$

Assumptions

$$\text{change in condition score } (\Delta\text{CS}) = 0$$

$$\text{metabolic liveweight} = 89.44(400^{0.75})$$

$$0 = -2.050 + 2.711\text{MJMEkg}^{-0.75}$$

therefore

$$2.05 \times 89.44 = 2.711\text{MJME}$$

therefore

$$\text{MJME} = 67.6\text{MJ.}$$

Therefore to maintain constant body condition it required 67.6MJ metabolisable energy or $0.76\text{MJMEkg}^{-0.75}$ during the period 180-242 days of pregnancy.

Appendix 3.6.1.2 Gain in body condition.

$$\begin{aligned}
 \text{if } \Delta\text{CS} &= 1 \\
 \text{then } 1/0.894 &= -2.050 + 2.711\text{MJME}/89.44 \\
 1.119 + 2.050 &= 2.711 \text{ MJME}/89.44 \\
 \text{MJME} &= 104.5\text{MJ} \\
 \text{therefore extra feed required per day} & \\
 &= 104.5 - 67.6 \\
 &= 36.9\text{MJME day}^{-1} \\
 &= 2288\text{MJME for 62 days}
 \end{aligned}$$

Therefore to gain one body condition score an additional 2288MJ metabolisable energy is required over and above the feed required to maintain the body condition of the cow.

Appendix 3.6.2 Estimation of energy required to maintain zero maternal energy retention at two stages of pregnancy (approximately 210 and 230 days of pregnancy) by indirect calorimetry.

Appendix 3.6.2.1 Period 1 (approximately day 210 of pregnancy).

From the estimated equation:-

$$\text{ERkg}^{-0.75} = -0.364 + 0.517\text{MJMEkg}^{-0.75}$$

where

$$\text{ERkg}^{-0.75} = \text{energy retention (MJday}^{-1}\text{) per unit metabolic liveweight}$$

$$\text{MJMEkg}^{-0.75} = \text{predicted metabolisable energy intake (MJday}^{-1}\text{) per unit metabolic liveweight}$$

$$\text{ME}_m = 0.70 \text{ MJkg}^{-0.75}\text{day}^{-1} \left(\frac{0.364}{0.517} \right)$$

where ME_m = metabolisable energy intake required for zero energy retention.

To obtain an estimate of metabolisable energy intake required for maintenance of zero maternal energy retention an allowance for pregnancy must be made. The equation of Moe and Tyrrell(1972) has been used to estimate the energy requirement for pregnancy.

$$Y = 100.8 + 0.567 e^{0.0174t}$$

where

$$Y = \text{kcal MEkg}^{-0.75} \text{ day}^{-1}$$

$$t = \text{number of days pregnant}$$

$$e = \text{natural logarithm}$$

when $t = 210$

$$Y = 100.8 + 0.567 e^{3.654}$$

$$= 100.8 + 0.567 \times 38.6289$$

$$= 100.8 + 21.9$$

The constant term ($100.8 \text{kcal MEkg}^{-0.75}$) is the energy required to maintain the non-pregnant animal. The extra energy requirement of pregnancy is 21.9kcal or $0.09 \text{MJMEkg}^{-0.75} \text{ day}^{-1}$.

The energy required to maintain body condition is:-

$$\begin{aligned} \text{ME}_{\text{mp}} &= 0.70(\text{ME}_m) + 0.09(\text{ME}_p) \\ &= 0.79 \text{MJMEkg}^{-0.75} \text{ day}^{-1} \end{aligned}$$

Appendix 3.6.2.2 Period 2: (approximately day 230 of pregnancy).

From the equation

$$\text{ERkg}^{-0.75} = -0.348 + 0.520 \text{MJMEkg}^{-0.75}$$

$$\text{ME}_m = 0.67 \text{MJkg}^{-0.75} \text{ day}^{-1}$$

$$\text{ME}_p = 0.13 \text{MJkg}^{-0.75} \text{ day}^{-1} \quad (\text{Moe and Tyrrell, 1972})$$

$$\text{ME}_{\text{mp}} = 0.80 \text{MJMEkg}^{-0.75} \text{ day}^{-1}$$

The estimates of energy required for maintenance of zero maternal energy retention obtained from indirect calorimetry were 0.79 and $0.80 \text{MJMEkg}^{-0.75} \text{ day}^{-1}$ for periods 1 and 2 respectively.

Appendix 3.6.3 Theoretical estimates of the metabolisable energy required to maintain and gain body condition (ARC, 1980).

From P 109 Table 3.23 in ARC (1980).

and P 113 Table 3.30 in ARC (1980).

Appendix 3.6.3.1 Maintenance of zero maternal energy retention.

Assume that calf birthweight is 32kg
 cow weight is 400kg
 q (metabolisability of the feed)= 0.6

$$ME_m = 42\text{MJ day}^{-1} \quad (\text{Table 3.23})$$

$$ME_p = 11.4\text{MJ day}^{-1} \quad (8 \text{ weeks before parturition})$$

$$= 19.7\text{MJ day}^{-1} \quad (4 \text{ weeks before parturition})$$

(Table 3.30)

$$ME_{mp} = 53.4\text{MJ day}^{-1} \quad \left. \vphantom{ME_{mp}} \right\} 8 \text{ weeks before parturition}$$

or $0.60\text{MJkg}^{-0.75} \text{day}^{-1}$

$$ME_{mp} = 61.7\text{MJ day}^{-1} \quad \left. \vphantom{ME_{mp}} \right\} 4 \text{ weeks before parturition}$$

or $0.69\text{MJkg}^{-0.75} \text{day}^{-1}$

Appendix 3.6.3.2 Gain in body condition.

(Table 3.23, P 109, ARC, 1980).

Assume: $q = 0.6$
 cow weight * = 440kg
 liveweight gain * = $0.75\text{kg cow}^{-1}\text{ day}^{-1}$

The pregnancy requirement has been left out of the present calculation as it is assumed that it is the same for cows whether they are maintaining or gaining body condition.

ME_m (440kg cow^{-1}) 44.4 MJ day^{-1}

ME_{mg} 72.4 MJ day^{-1}

ME_g 30 MJ day^{-1}

ME_{mg} is the energy requirement for a cow weighing on average 440kg to gain 0.75kg day^{-1} .

ME_g is the extra energy required over and above maintenance to gain 0.75kg day^{-1} .

To gain 35kg liveweight it will require 46.7 days
 ($35/0.75$). The energy requirement is $46.7\text{ days} \times 30\text{MJ day}^{-1}$
 = 1400MJME.

The theoretical energy requirement, for a cow weighing on average 440kg and gaining at a rate of 0.75kg day^{-1} , to gain 35kg liveweight (one condition score) is 1400MJ metabolisable energy.

* average liveweight and liveweight gain from present experiment.

APPENDIX 3.7 STATISTICAL ANALYSES OF THE FAECES, URINE AND METHANE COMPONENTS OF ENERGY BALANCES CARRIED OUT DURING LACTATION.

Regression analyses of faeces, urine and methane energies for high (H) and low (L) breeding index cows are presented below.

$$Y = bX + C^*$$

Y	X	b ** [†] s.e.	C _H	C _{H-L}	s.e.***
<u>Early lactation</u> (<u>ad libitum</u> to restricted feeding)					
Faeces	Gross energy	0.301 [†] -0.016	-0.161	0.012	0.018
Urine	intake	0.068 [†] -0.009	-0.011	0.004	0.011
Methane		0.051 [†] -0.007	0.029	-0.011	0.007
<u>Early lactation</u> (restricted to <u>ad libitum</u> feeding)					
Faeces	Gross energy	0.306 [†] -0.021	-0.180	-0.010	0.025
Urine	intake	0.028 [†] -0.007	0.075	0.009	0.008
Methane		0.055 [†] -0.003	0.021	-0.009	0.004
<u>Late lactation</u> (<u>ad libitum</u> to restricted feeding)					
Faeces	Gross energy	0.338 [†] -0.034	0.066	0.023	0.037
Urine	intake	0.036 [†] -0.006	0.005	-0.008	0.007
Methane		0.050 [†] -0.007	0.030	-0.005	0.013

* Units are MJ kg^{-0.75} cow⁻¹ day⁻¹

** There were no significant differences between genotypes in the regression coefficient hence only the pooled coefficient has been presented.

*** Standard error of the difference between genotypes in the intercept.

There were no significant differences between genotypes in the regression coefficients or the intercepts hence pooled regressions could be calculated.

APPENDIX 5.1 CALCULATION OF KG DM EATEN PER KG MILK FAT
PRODUCED FOR THE 1979/80 LACTATION.

The data have been taken from Tables 2.8, 2.10 and 2.11. Gross efficiencies (kg DM/kg fat) have only been calculated for cows fed ad libitum during experimental periods because the milk fat production for these cows were similar before, during and after the experimental periods.

Expt.	Breeding Index	DMI* (kg cow ⁻¹ day ⁻¹)	Milk fat yield (g cow ⁻¹ day ⁻¹)	kg DM/kg fat
1	H	12.9	676	19.1
	L	13.0	559	23.3
2	H	14.2	578	24.6
	L	14.7	480	30.6
3	H	15.2	588	25.8
	L	15.1	461	32.8

* The dry matter intakes (DMI) have been standardised to a common DM digestibility of 75% (the DM digestibilities were 74.4, 70.6 and 70.4 for Experiments 1, 2 and 3 respectively). In addition the dry matter intakes for Experiment 3 have been corrected for soil contamination (see Appendix 2.3 for details).

The average lactation length for H and L cows was 249 days thus to obtain the average gross efficiency of feed conversion over the lactation, feed efficiencies were needed for the following times:-

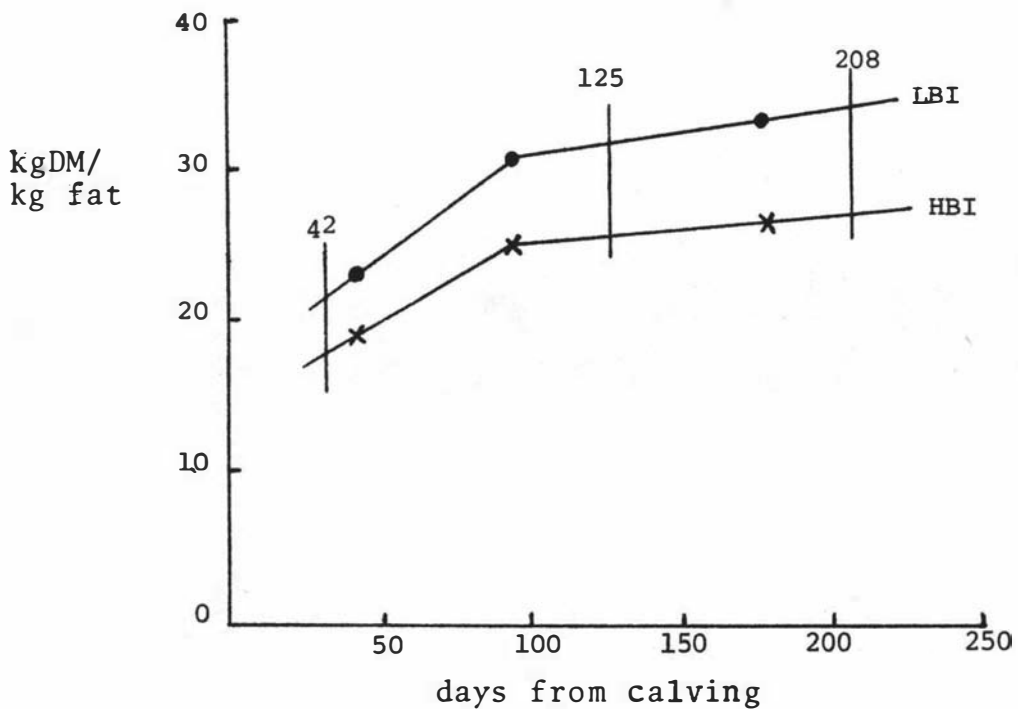
- 42 days
- 42 + 83 = 125 days
- 42 + 83 + 83 = 208 days

(N.B. 249 days = 3 x 83 days)

Gross efficiencies were measured during lactation at the following times:-

Expt.	Days from calving		
	Start	Start to mid-point	Total
1	30	18	48
2	80	18	98
3	160	18	178

The gross efficiencies at 42, 125 and 208 days from calving can be estimated from the diagram below.



The estimated gross efficiencies were:-

<u>Days from calving</u>	<u>Predicted efficiencies</u> (kg DM/kg fat)	
	<u>H</u>	<u>L</u>
42	18.0	22.0
125	25.0	31.0
208	26.5	33.0

From the above data the average gross efficiencies of feed conversion can be estimated to be 23.2 and 28.7 kg DM/kg milk fat for H and L cows respectively.

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